Localization of Hidden Insulinomas with ^{68}Ga-DOTA-Exendin-4 PET/CT: A Pilot Study

Kwadwo Antwi[1], Melpomeni Fani[1], Guillaume Nicolas[1], Christof Rottenburger[1], Tobias Heye[1], Jean Claude Reubi[2], Beat Gloor[3], Emanuel Christ*[4], and Damian Wild*[1]

[1]Clinic of Radiology and Nuclear Medicine, University of Basel Hospital, Basel, Switzerland; [2]Division of Cell Biology and Experimental Cancer Research, Institute of Pathology, University of Bern, Bern, Switzerland; [3]Department of Visceral Surgery, University Hospital of Bern, Bern, Switzerland; and [4]Division of Endocrinology, Diabetology, and Clinical Nutrition, University Hospital of Bern, Bern Switzerland

^{111}In-DOTA-exendin-4 SPECT/CT has been shown to be highly efficient in the detection of insulinomas. We aimed at determining whether novel PET/CT imaging with [Nle^{14},Lys^{40}(Ahx-DOTA-^{68}Ga)NH_2]exendin-4 (^{68}Ga-DOTA-exendin-4) is feasible and sensitive in detecting benign insulinomas. **Methods:** ^{68}Ga-DOTA-exendin-4 PET/CT and ^{111}In-DOTA-exendin-4 SPECT/CT were performed in a randomized cross-over order on 5 patients with endogenous hyperinsulinemic hypoglycemia. The gold standard for comparison was the histologic diagnosis after surgery. **Results:** In 4 patients histologic diagnosis confirmed a benign insulinoma, whereas one patient refused surgery despite a positive ^{68}Ga-DOTA-exendin-4 PET/CT scan. In 4 of 5 patients, previously performed conventional imaging (CT or MR imaging) was not able to localize the insulinoma. ^{68}Ga-DOTA-exendin-4 PET/CT correctly identified the insulinoma in 4 of 4 patients, whereas ^{111}In-DOTA-exendin-4 SPECT/CT correctly identified the insulinoma in only 2 of 4 patients. **Conclusion:** These preliminary data suggest that the use of ^{68}Ga-DOTA-exendin-4 PET/CT in detecting hidden insulinomas is feasible.

Key Words: glucagon-like peptide-1 receptor targeting; exendin-4; insulinoma; ^{68}Ga

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The most common cause of endogenous hyperinsulinemic hypoglycemia in adults is an insulinoma. Endogenous hyperinsulinemic hypoglycemia is biochemically diagnosed by a prolonged supervised fasting test in an inpatient setting (*1*). Approximately 5%–10% of insulinomas are multiple, mainly in the context of multiple endocrine neoplasia type 1, less than 10% are malignant, and most are benign single insulinomas (*1*). The only curative treatment of an insulinoma is its surgical removal. Therefore, exact preoperative localization of the insulinoma is critical to planning the surgical intervention (*1*). MR imaging, CT, or endoscopic ultrasound is normally used to localize insulinomas (*1*). However, the small size of the tumors (often <1 cm) limits the sensitivity of these methods (*1*). Sensitivity can be increased by including methods such as angiography with selective arterial calcium stimulation and hepatic venous sampling. However, this procedure is invasive, with a concomitant risk of complications. The sensitivity of somatostatin receptor scintigraphy and SPECT/CT is usually low (33%–50%) and inconsistent for ^{18}F-DOPA PET (90% in a prospective study and 20% in a retrospective study) (*2*).

It has been shown that targeting the glucagonlike peptide-1 receptors (GLP-1R) using the specific ligand [Lys^{40}(Ahx-DOTA-^{111}In)NH_2]exendin-4, [Lys^{40}(Ahx-DTPA-^{111}In)NH_2]exendin-4, or [Lys^{40}(Ahx-HYNIC-^{99m}Tc/EDDA)NH_2]exendin-4 is a very sensitive (≥95% sensitivity), noninvasive method to localize benign insulinomas with SPECT (*2–4*). In comparison to SPECT, PET has a higher spatial resolution and sensitivity and accurately quantifies tracer uptake (*5*). Recently, it has been shown that ^{68}Ga-DO3A-VS-Cys^{40}-exendin-4 PET/CT can detect malignant insulinomas (*6*).

The aim of our study was, first, to determine whether the use of [Nle^{14},Lys^{40}(Ahx-DOTA-^{68}Ga)NH_2]exendin-4 (^{68}Ga-DOTA-exendin-4) PET/CT in the detection of benign insulinomas is feasible and, second, to compare its detection rate with that of [Nle^{14}, Lys^{40}(Ahx-DOTA-^{111}In)NH_2]exendin-4 (^{111}In-DOTA-exendin-4) SPECT/CT in the same patients.

MATERIALS AND METHODS

Patients

Five consecutive patients were screened and accepted for our prospective pilot study. The patients were referred from 4 tertiary care centers in Switzerland. All patients fulfilled the following inclusion criteria: biochemically proven endogenous hyperinsulinemic hypoglycemia with neuroglycopenic symptoms, negative results on sulfonylurea screening (exclusion of hypoglycemia factitia), contrast-enhanced 3-T MR imaging performed no more than 2 mo previously, and age above 18 y. Patients with evidence of malignant insulinoma on conventional imaging were excluded, as well as pregnant women, patients with allergies to exendin-4, and patients with renal insufficiency (blood creatinine concentrations > 140 μmol/L).

The institutional review board approved this study, and all subjects gave written informed consent.

Procedures

^{111}In-DOTA-exendin-4 SPECT/CT and ^{68}Ga-DOTA-exendin-4 PET/CT were performed within 24–73 h in randomized crossover order. Synthesis and labeling of ^{68}Ga-DOTA-exendin-4 and ^{111}In-DOTA-exendin-4 have been described elsewhere (*7*).

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For correspondence contact: Damian Wild, Division of Nuclear Medicine, University of Basel Hospital, Petersgraben 4, CH-4031 Basel, Switzerland.
E-mail: damian.wild@usb.ch
*Contributed equally to this work.
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Total-body planar images and SPECT/CT of the abdomen were performed 4 and 72 h after intravenous injection of 79.2 ± 9.3 MBq (range, 66–90 MBq, 10.5–14.4 μg) of ^{111}In-DOTA-exendin-4. The SPECT/CT scanner (Symbia Intevo; Siemens Healthcare) was equipped with a medium-energy, parallel-hole collimator (window setting, 172 and 247 keV; width, 15%; rotation, 2 × 180°; matrix, 256 × 256; projections, 60; acquisition time per projection, 24 s). All patients underwent unenhanced low-dose CT (130 kVp, 40 mA) for attenuation correction and to provide an anatomic reference.

PET/CT was performed on a PET/16-detector CT scanner (Discovery ST; GE Healthcare). One bed position of the upper abdomen was acquired during 8 min, 2.5 h after intravenous injection of 79.8 ± 3.9 MBq (range, 76–97 MBq, 12.0–15.3 μg) of ^{68}Ga-DOTA-exendin-4. All patients underwent unenhanced low-dose CT for attenuation correction and to provide an anatomic reference (120 kVp, 30–100 mAs). Blood samples were taken 2, 5, 15, 30, 60, 120, and 180 min after injection of ^{68}Ga-DOTA-exendin-4 and ^{111}In-DOTA-exendin-4 to measure glucose levels and blood clearance. An additional blood sample was taken 300 min after injection of ^{111}In-DOTA-exendin-4. All conventional scans were independently interpreted by experienced radiologists at the referral centers. GLP-1R SPECT/CT and PET/CT scans were independently assessed by 2 board-certified nuclear medicine physicians. Both interpreters were masked to other imaging results and the patient's clinical history. In cases of discordant findings, a consensus was reached between the 2 interpreters. Tumor-to-background ratios were measured for ^{111}In-DOTA-exendin-4 (counts) and ^{68}Ga-DOTA-exendin-4 (maximal standardized uptake values).

Histologic diagnosis was regarded as the standard for comparison. The pathologists were masked to the results of other diagnostic tests but were aware of the patient's clinical history. Finally, GLP-1R expression was evaluated in vitro by GLP-1R autoradiography as previously described (*8*).

RESULTS

The clinical characteristics are summarized in Table 1. In all 5 patients, the fasting plasma glucose level was measured. Symptoms of neuroglycopenia in association with low plasma glucose levels (mean, 2.1 mmol/L; range, 1.6–2.6 mmol/L) and inadequately increased insulin (mean, 12.1 mU/L; range, 3.9–21.9 mU/L) and C-peptide (mean, 0.714 nmol/L; range, 0.5–0.98 nmol/L) levels were documented in all patients after 12–52 h of fasting.

The labeling yield of ^{111}In- and ^{68}Ga-DOTA-exendin-4 was more than 95%, and the radiochemical purity was at least 95% for ^{111}In-DOTA-exendin-4 and at least 93% for ^{68}Ga-DOTA-exendin-4, at a specific activity of 30 MBq/nmol.

Blood sampling of ^{111}In-DOTA-exendin-4 revealed a biexponential blood clearance with half-times of 16 ± 2 min and 110 ± 19 min. ^{68}Ga-DOTA-exendin-4 also revealed a biexponential blood clearance, with half-times of 14 ± 3 min and 41 ± 4 min. Both compounds showed a plasma clearance of about 50% in the α phase (Supplemental Fig. 1; supplemental materials are available at http://jnm.snmjournals.org).

The imaging results are summarized in Table 2. In patient 4, focal uptake of ^{111}In-DOTA-exendin-4 and ^{68}Ga-DOTA-exendin-4 was highly suggestive of an insulinoma (Fig. 1); however, the patient has refused surgery so far.

All patients received an infusion of exogenous glucose (1,000 mL, 10%) for 5 h starting just before injection of the radiotracer. This step prevented the occurrence of any severe hypoglycemic episodes. Two patients experienced nausea—and 2 patients nausea and vomiting—after injection of ^{111}In-DOTA-exendin-4. One patient experienced nausea after injection of ^{68}Ga-DOTA-exendin-4. No other adverse effects were observed.

In all 4 patients who underwent surgery, histologic examination confirmed the diagnosis of a benign insulin-producing tumor (Table 2) and symptoms of hypoglycemia resolved immediately after surgery.

Histopathologic diagnosis was made at the local institution. In patient 5, imaging and additional in vitro GLP-1R autoradiography was performed (Figs. 2 and 3).

DISCUSSION

This report provides the proof of principle that ^{68}Ga-DOTA-exendin-4 PET/CT is feasible and sensitive in the preoperative detection of hidden insulinomas in patients. In all 4 of the patients who underwent surgery, ^{68}Ga-DOTA-exendin-4 PET/CT had already detected the insulinoma by 2.5 h after injection. GLP-1R imaging changed the clinical management of 3 of the 4 patients, whereas in the first patient only ^{68}Ga-DOTA-exendin-4 PET/CT correctly localized the small insulinoma, which was crucial for planning surgery. ^{68}Ga-DOTA-exendin-4 PET/CT revealed higher tumor-to-background ratios (2.5 h after injection) than ^{111}In-DOTA-exendin-4 SPECT/CT (4 and 72 h after injection) because of an advantageous partial-volume effect in PET (*9*) and faster blood clearance of ^{68}Ga-DOTA-exendin-4 than of ^{111}In-DOTA-exendin-4. Furthermore, PET has a higher spatial resolution than SPECT (*5*). This is relevant for the detection of insulinomas near the highly active kidneys. The better spatial resolution of PET together with the higher tumor-to-background ratio

TABLE 1
Clinical Characteristics

Characteristic	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
Age (y)	39	48	64	63	56
Sex	F	F	M	M	F
Presenting symptoms	Hypoglycemia, palpitation, hyperhidrosis	Hypoglycemia, dizziness, hyperhidrosis	Hypoglycemia, palpitation, hyperhidrosis	Hypoglycemia, palpitation, hyperhidrosis	Hypoglycemia, tachypnea, hyperhidrosis
Duration of symptoms (mo)	20	39	48	24	24

TABLE 2
Preoperative Imaging and Operative Procedures

Procedure	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
Conventional imaging					
CT	FN	NA	NA	NA	NA
ASVS	FN	FN	NA	NA	NA
Endoscopic ultrasound	FN	FN	FN	NA	NC
MR imaging	FN	FN	FN	−	TP
^{111}In-DOTA-exendin-4 SPECT/CT					
Detection of tumor 4 h after injection	FN	TP	TP	+	FN
Tumor-to-background ratio at 4 h	NA	1.04	3.87	1.96	NA
Detection of tumor 72 h after injection	FP	TP	TP	+	FP
Tumor-to-background ratio at 72 h	NA	1.12	3.93	2.04	NA
^{68}Ga-DOTA-exendin-4 PET/CT					
Detection of tumor	TP	TP	TP	+	TP
Tumor-to-background ratio	2.00	3.79	6.87	5.95	32.42
SUV_{max} of tumor	5.5	10.8	49.1	25	30.8
Surgery and histology					
Surgical procedure	Enucleation	Pancreas tail resection	Pancreas tail resection	Not operated	Enucleation
Localization	Body/tail transition	Tail	Tail	NA	Tail
Dimension (mm)	8	10	10	NA	18
Histopathology	Insulinoma	Insulinoma	Insulinoma	NA	Insulinoma

ASVS = selective arterial calcium stimulation and hepatic venous sampling; SUV_{max} = maximal standardized uptake value; NA = not applicable; NC = not conclusive; FN = false-negative; FP = false-positive; TP = true-positive; − = negative result; + = positive result.

of ^{68}Ga-DOTA-exendin-4 PET/CT brought about a higher insulinoma detection rate than was obtained with ^{111}In-DOTA-exendin-4 SPECT/CT.

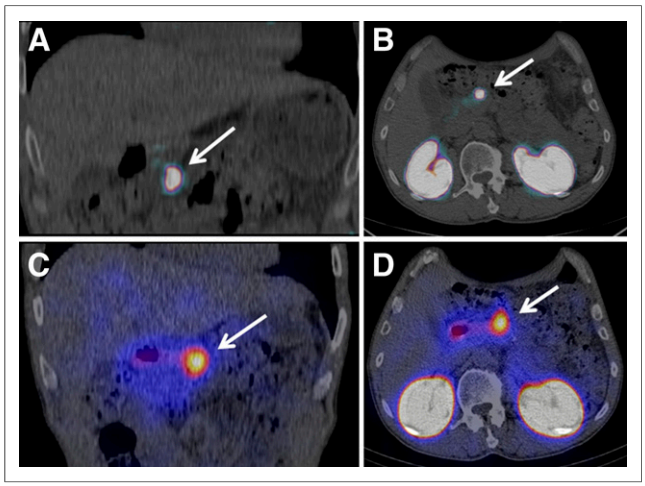

FIGURE 1. (A and B) Coronal (A) and transaxial (B) PET/CT images from patient 4 obtained 2.5 h after injection of 80 MBq of ^{68}Ga-DOTA-exendin-4. (C and D) Coronal (C) and transaxial (D) SPECT/CT images of same patient 72 h after injection of 90 MBq of ^{111}In-DOTA-exendin-4. Focal uptake of ^{111}In-DOTA-exendin-4 and ^{68}Ga-DOTA-exendin-4 is seen in pancreatic body (arrows); patient has refused surgery so far.

Late ^{111}In-DOTA-exendin-4 SPECT scans (72 h after injection) showed a slightly higher tumor-to-background ratio than early SPECT scans. This finding is consistent with our previously published work (*2*) in which we suggested late scans 3–7 d after injection in patients with negative findings on early scans. However, late scans may increase the risk of false-positive results because of the physiologic expression of GLP-1R in pancreatic islets or acini and the Brunner gland of the duodenum (*8*).

The shorter imaging procedure, higher tumor-to-background ratio, better spatial resolution (*5*), and lower radiation burden of ^{68}Ga-DOTA-exendin-4 PET/CT (*7*) favor this novel method over ^{111}In-DOTA-exendin-4 SPECT/CT. Furthermore, absolute quantification of tumor and background uptake, which might be useful to improve interobserver agreement and specificity, is better evaluated with PET/CT than with SPECT/CT. GLP-1R PET/CT is expected to have a high clinical impact on the management of patients with endogenous hyperinsulinemic hypoglycemia. Previous experience (*2*) suggests that insulinomas in the context of multiple endocrine neoplasia type 1 express GLP-1R. It is therefore likely that GLP-1R PET/CT will be a valuable tool to distinguish between insulin-secreting lesions and gastrin-secreting or nonsecreting pancreatic lesions in this genetic syndrome, thereby determining the surgical strategy. Whether GLP-1R PET/CT will be useful in diagnosing other conditions of endogenous hyperinsulinemic hypoglycemia, such as β-cell hyperplasia or hypertrophy (nesidioblastosis), in adults and children remains to be determined.

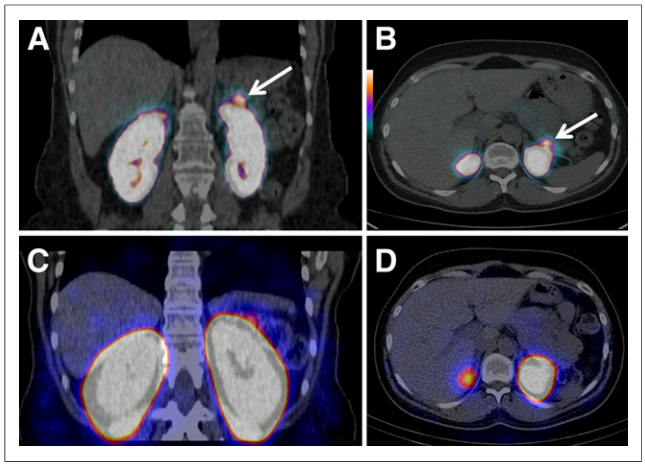

FIGURE 2. (A and B) Coronal (A) and transaxial (B) PET/CT images from patient 5 obtained 2.5 h after injection of 76 MBq of ^{68}Ga-DOTA-exendin-4. (C and D) Coronal (C) and transaxial (D) SPECT/CT images of same patient 72 h after injection of 66 MBq of ^{111}In-DOTA-exendin-4. Arrows show focal ^{68}Ga-DOTA-exendin-4 uptake in distal pancreatic tail consistent with surgically removed insulinoma. SPECT/CT did not show the insulinoma.

This study had limitations. First, conventional imaging could not be standardized because of differences in local availability. However, MR imaging was performed on all patients. Second, conventional imaging had a tendency to underperform compared with what has been reported in the published literature (*1,10*). This discrepancy might be explained by the fact that most patients were referred after having undergone conventional imaging with negative results and were therefore part of a negative selection. Finally, this study included only 5 patients. However, it was intended to be a pilot study. Because of the promising results, a larger study evaluating GLP-1R PET/CT has been started.

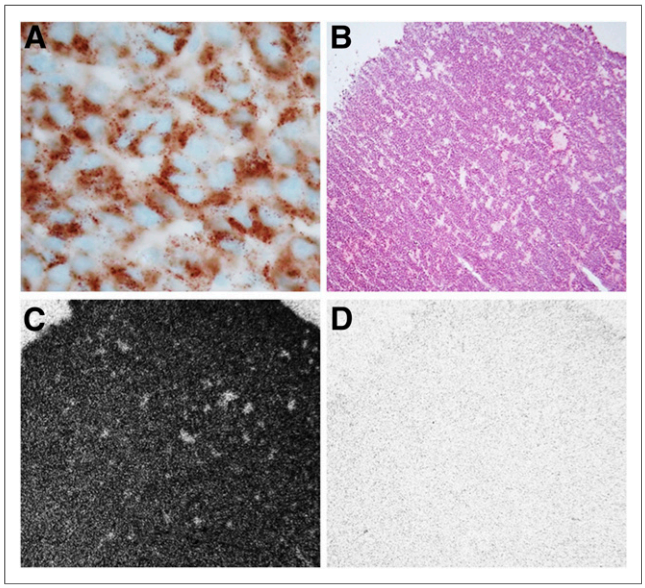

FIGURE 3. Hormone and receptor evaluation of resected insulinoma in patient 5. (A) Immunohistochemistry for insulin showing strongly labeled tumor cells. (B) Hematoxylin- and eosin-stained tumor tissue. (C) In vitro autoradiography revealing high GLP-1R density (mean, 5,766 dpm/mg of tissue). (D) Autoradiogram showing nonspecific binding of ^{125}I-GLP-1 (7–36) amide in presence of 100 nM GLP-1 (7–36) amide.

CONCLUSION

This study proved that ^{68}Ga-DOTA-exendin-4 PET/CT is a feasible and sensitive tool for the detection of insulinomas. The higher spatial resolution, the possibility of quantification, and the lower radiation burden favor ^{68}Ga-DOTA-exendin-4 PET/CT over ^{111}In-DOTA-exendin-4 SPECT/CT.

DISCLOSURE

The costs of publication of this article were defrayed in part by the payment of page charges. Therefore, and solely to indicate this fact, this article is hereby marked "advertisement" in accordance with 18 USC section 1734. This work was supported in part by the Swiss National Science Foundation (grant 320030_152938/1) and the Desirée and Niels Yde's Foundation (grant 389-12). No other potential conflict of interest relevant to this article was reported.

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REFERENCES

1. de Herder WW, Niederle B, Scoazec J-Y, et al. Well-differentiated pancreatic tumor/carcinoma: insulinoma. *Neuroendocrinology.* 2006;84:183–188.
2. Christ E, Wild D, Ederer S, et al. Glucagon-like peptide-1 receptor imaging for the localisation of insulinomas: a prospective multicentre imaging study. *Lancet Diabetes Endocrinol.* 2013;1:115–122.
3. Christ E, Wild D, Forrer F, et al. Glucagon-like peptide-1 receptor imaging for localization of insulinomas. *J Clin Endocrinol Metab.* 2009;94:4398–4405.
4. Sowa-Staszczak A, Pach D, Mikołajczak R, et al. Glucagon-like peptide-1 receptor imaging with [Lys40(Ahx-HYNIC-^{99m}Tc/EDDA)NH2]-exendin-4 for the detection of insulinoma. *Eur J Nucl Med Mol Imaging.* 2013;40:524–531.
5. Martin WH, Delbeke D, Patton JA, Sandler MP. Detection of malignancies with SPECT versus PET, with 2-[fluorine-18]fluoro-2-deoxy-D-glucose. *Radiology.* 1996;198:225–231.
6. Eriksson O, Velikyan I, Selvaraju RK, et al. Detection of metastatic insulinoma by positron emission tomography with [^{68}Ga]exendin-4: a case report. *J Clin Endocrinol Metab.* 2014;99:1519–1524.
7. Wild D, Wicki A, Mansi R, et al. Exendin-4-based radiopharmaceuticals for glucagonlike peptide-1 receptor PET/CT and SPECT/CT. *J Nucl Med.* 2010;51:1059–1067.
8. Körner M, Stöckli 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.
9. Soret M, Bacharach SL, Buvat I. Partial-volume effect in PET tumor imaging. *J Nucl Med.* 2007;48:932–945.
10. Druce MR, Muthuppalaniappan VM, O'Leary B, et al. Diagnosis and localisation of insulinoma: the value of modern magnetic resonance imaging in conjunction with calcium stimulation catheterisation. *Eur J Endocrinol.* 2010;162:971–978.