

The added value of [¹⁸F]fluoro-L-DOPA PET in the diagnosis of hyperinsulinism of infancy: a retrospective study involving 49 children

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Abstract

Purpose Neuroendocrine diseases are a heterogeneous group of entities with the ability to take up amine precursors, such as L-DOPA, and convert them into biogenic amines, such as dopamine. Congenital hyperinsu-

linism of infancy (HI) is a neuroendocrine disease secondary to either focal adenomatous hyperplasia or a diffuse abnormal pancreatic insulin secretion. While focal hyperinsulinism may be reversed by selective surgical resection, diffuse forms require near-total pancreatectomy when resistant to medical treatment. Here, we report the diagnostic value of PET with [¹⁸F]fluoro-L-DOPA in distinguishing focal from diffuse HI.

Methods Forty-nine children were studied with [¹⁸F]fluoro-L-DOPA. A thoraco-abdominal scan was acquired 45–65 min after the injection of 4.2±1.0 MBq/kg of [¹⁸F]fluoro-L-DOPA. Additionally, 12 of the 49 children were submitted to pancreatic venous catheterisation for blood samples (PVS) and 31 were also investigated using MRI.

Results We identified abnormal focal pancreatic uptake of [¹⁸F]fluoro-L-DOPA in 15 children, whereas diffuse radiotracer uptake was observed in the pancreatic area in the other 34 patients. In children studied with both PET and PVS, the results were concordant in 11/12 cases. All patients with focal radiotracer uptake and nine of the patients with diffuse pancreatic radiotracer accumulation, unresponsive to medical treatment, were submitted to surgery. In 21 of these 24 patients, the histopathological results confirmed the PET findings. In focal forms, selective surgery was followed by clinical remission without carbohydrate intolerance.

Conclusion These data demonstrate that PET with [¹⁸F]fluoro-L-DOPA is an accurate non-invasive technique allowing differential diagnosis between focal and diffuse forms of HI.

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Introduction

Congenital hyperinsulinism (HI) with a permanent increase in insulin secretion is the most important cause of recurrent and profound hypoglycaemia in early infancy [1–3]. Prompt recognition and correction of hypoglycaemia are necessary to prevent the high risk of permanent neurological complications [4]. Control of HI commonly involves medical treatment with diazoxide, possibly in association with octreotide and/or nifedipine [5–7], which inhibits insulin secretion. In addition, frequent feedings with a carbohydrate-enriched diet are also requested. However, frequently this treatment is not effective, and pancreatectomy is the only option for patients resistant to this treatment [8, 9]. Despite an indistinguishable clinical presentation, at least two different histopathological forms of HI—focal and diffuse—have been recognised based on different molecular entities [1, 2, 10–14]. Focal HI is characterised by pathological pancreatic β -cells gathered in focal adenomatous hyperplasia usually 2.5–7.5 mm in diameter. Diffuse HI corresponds to abnormal insulin secretion by all the pancreatic β -cells, with disseminated β -cells showing enlarged abnormal nuclei [3]. Finally, about 10% of HI cases are clinically atypical and cannot be classified; these cases have an unknown molecular basis and histopathological form [15]. The distinction between the two major forms of HI is crucial since their surgical treatment and the outcome differ considerably. Focal HI is cured by selective resection of the adenomatous hyperplasia, whereas severe diffuse forms require a near-total pancreatectomy with an accompanying high risk of iatrogenic diabetes [16, 17].

The two histopathological forms correspond to two distinct molecular entities in which mainly the SUR1 and KIR6.2 genes are implicated. Different underlying genetic defects have been described previously [18, 21]. Focal HI is associated with a paternally inherited mutation in the SUR1 or KIR6.2 gene, in both cases located in the 11p15.1 region, loss of the maternal 11p15 allele restricted to the pancreatic lesion and a somatic reduction to homozygosity [12]. The diffuse form of HI is more heterogeneous and its genetic basis has been recognised in only 50% of the cases. Diffuse HI is mostly related to mutations in the genes coding for the sulphonylurea receptor. Unfortunately, the results of mutational analyses are usually unavailable when important clinical decisions regarding surgical treatment have to be taken.

For several reasons the distinction between the two forms of HI before surgery is difficult: (a) in contrast to adult pancreatic adenoma, focal lesions in infants are impossible to identify macroscopically by ultrasound (US), magnetic resonance imaging (MRI) or computed tomography (CT), as they are lobulated like the normal pancreatic parenchyma; (b) until recently, the localisation of

insulin hypersecretion before surgery was only possible through pancreatic venous catheterisation in order to collect venous blood samples (PVS), with possible additional intravenous stimulation of insulin secretion by calcium and tolbutamide to allow a pancreatic map of the glucose, insulin and C-peptide concentrations [22–26]. PVS is an invasive method, is technically difficult to perform and requires general anaesthesia. The plasma glucose concentration must be carefully maintained between 2 and 3 mmol/l before and during PVS. Moreover, all medical treatments have to be stopped 5 days beforehand. Therefore, the development of a less invasive examination that permits differential diagnosis between focal and diffuse HI would be of major interest.

L-Dihydroxyphenylalanine (L-DOPA), a precursor of catecholamines, is converted to dopamine by the aromatic amino acid decarboxylase (DDC) enzyme. The ability to take up and decarboxylate amine precursors such as L-DOPA or 5-hydroxytryptophan and store their biogenic amines (dopamine and serotonin) in secretory granules in the cytoplasm is characteristic of neuroendocrine cells [27, 28]. Pancreatic islets have been shown to take up L-DOPA and convert it to dopamine through the aromatic amino acid dopa decarboxylase [29–31].

In a preliminary study, we assessed the use of positron emission tomography (PET) imaging, performed with [18 F] fluoro-L-dihydroxyphenylalanine ([18 F]fluoro-L-DOPA), in a group of 15 patients, to detect the functional pancreatic islet tissue [32]. In this retrospective study, we enrolled 49 children (including the 15 patients previously studied) with HI in order to evaluate the accuracy of [18 F]fluoro-L-DOPA PET in differentiating the two major histopathological forms of HI and its potential to replace PVS.

Materials and methods

Patients

Forty-nine children (30 boys and 19 girls; age 1–18 months, median 4.1 months) with HI were studied with [18 F]fluoro-L-DOPA PET (Table 1). HI diagnosis was based upon persistent hypoglycaemia with low plasma ketone bodies and free fatty acids, together with measurable circulating insulin levels during hypoglycaemic episodes.

The patients fasted for at least 6 h prior to the PET study. Treatment with octreotide was stopped for at least 1 day and treatment with diazoxide for at least 2 days before the PET study, with the exception of one case in which the PET study was performed under glucagon administration (patient 26). Five patients had two PET studies: (a) one patient had a PET study before and after treatment with an inhibitor of DDC, L- α -hydrazino- α -methyl- β -(3,4-dihydroxyphenyl)

Table 1 Clinical profile of the 49 patients studied

Patient/ sex	Age at diagnosis/PET (months)	Response to medication	MRI	PVS	PET			Surgery/histology (type of HI)	
				Type of HI ^a	Type of HI ^a	SUV _r ^b			
						Right	Middle		Left
1/M	Neonatal/5	Dzx-, Octr-	No	Yes (F)	F	1.5	0.8	0.8	Yes/F
2/M	Neonatal/3	Dzx-, Octr-	Yes	Yes (F)	F	1.2	0.9	0.9	Yes/F
3/F	Neonatal/16	Dzx-, Octr+	No	Yes (F)	F	1.3	0.8	0.8	Yes/A
4/F	Neonatal/2	Dzx-, Octr-	No	Yes (F)	F	1.4	0.9	0.8	Yes/F
5/M	Neonatal/15	Dzx-, Octr+	Yes	Yes (F)	D	1.1	0.8	1.1	Yes/A
6/F	2.5/13	Dzx+	No	Yes (D)	D	1.2	1.0	0.8	No
7/M	Neonatal/2	Dzx-, Octr-	Yes	Yes (D)	D	1.1	0.9	0.9	Yes/D
8/M	Neonatal/4	Dzx-, Octr-	Yes	Yes (D)	D	0.9	0.9	1.1	Yes/D
9/M	Neonatal/5	Dzx-, Octr+	No	No	D	1.0	1.0	1.0	No
10/M	Neonatal/4	Dzx-, Oct -	Yes	Yes (F)	F	0.8	0.8	1.4	Yes/F
11/F	Neonatal/8	Dzx-, Octr+	No	Yes (D)	D	1.1	1.0	0.9	No
12/M ^c	Neonatal/3 and 12	Dzx-, Octr+	Yes	No	D	1.1	1.0	1.0	No
				No	D	1.0	1.0	1.0	
13/F	Neonatal/5	Dzx-, Octr-	No	Yes (D)	D	0.9	1.1	1.1	Yes/D
14/M ^d	Neonatal/14 and 17	Dzx-, Octr-	No	No	D	1.0	1.0	1.0	
			No			ND	ND	ND	Yes/D
15/M	Neonatal/2	Dzx-, Octr+	No	No	D	1.1	1.0	1.0	No
16/M ^e	Neonatal/7 and 20	Dzx-, Octr+	Yes	No	F	1.8	0.6	0.6	Yes/F
					F	1.7	0.6	0.7	Yes/F
17/M	Neonatal/9	Dzx-, Octr+	Yes	No	D	1.1	0.8	1.1	No
18/F	Neonatal/15	Dzx-, Octr+	Yes	No	D	1.0	0.9	1.1	No
19/M	Neonatal/11	Dzx-, Octr+	Yes	No	D	1.1	1.0	0.9	No
20/F	Neonatal/2.5	Dzx-, Octr+	No	No	D	1.0	1.0	1.0	No
21/M	Neonatal/17	Dzx+	Yes	No	D	1.0	1.0	1.0	No
22/M	Neonatal/6	Dzx+	No	No	D	1.2	0.9	0.9	No
23/M	Neonatal/1	Dzx-, Octr+	Yes	No	F	0.6	0.6	1.3	Yes/F
24/F	Neonatal/2	Dzx-, Octr+	No	Yes (F)	F	1.2	0.9	0.9	Yes/F
25/M	Neonatal/2	Dzx-, Octr+	Yes	No	F	1.3	0.9	0.7	Yes/F
26/F ^f	Neonatal/2.5	Dzx-, Octr-, glucagon	No	No	D	ND	ND	ND	Yes/D
27/M	Neonatal/4	Dzx-, Octr+	Yes	No	D	1.0	1.0	1.0	No
28/M	Neonatal/4	Dzx-, Octr+	Yes	No	F	0.9	1.4	0.7	Yes/F
29/M	Neonatal/3	Dzx-, Octr+	Yes	No	F	1.7	0.7	0.6	Yes/F
30/F	Neonatal/4	Dzx-, Octr+	Yes	No	D	1.1	0.9	0.9	No
31/F	3/13	Dzx-, Octr+	No	No	F	0.8	1.5	0.8	Yes/F
32/M	3/19	Dzx+	No	No	D	1.1	0.9	1.0	No
33/M	Neonatal/1	Dzx-, Octr+	Yes	No	D	1.1	1.0	0.9	No
34/F	Neonatal/1.5	Dzx-, Octr-	Yes	No	D	1.1	1.0	0.9	Yes/D
35/M	Neonatal/3	Dzx-, Octr?	No	No	D	1.0	0.9	1.1	No
36/M	Neonatal/4.5	Dzx+, Octr+	Yes	No	F	1.0	0.6	1.5	Yes/F
37/M ^g	2.5/6 and 12	Dzx-, Octr-	Yes	No	F	1.5	0.8	0.8	Yes/F
					F	1.5	0.7	0.8	Yes/F
38/F	Neonatal/1	Dzx-, Octr+	Yes	No	D	1.2	0.9	0.9	No
39/F	Neonatal/3	Dzx+, Octr+	Yes	No	D	1.1	1.0	0.9	No
40/M	Neonatal/18	Dzx-, Octr+	Yes	No	D	1.1	0.9	1.0	No
41/M	Neonatal/8	Dzx+, Octr+	Yes	No	D	1.1	0.9	1.1	No
42/F ^g	Neonatal/2	Dzx-, Octr-, glucagon	Yes	No	D	1.1	1.0	1.0	No
43/F	Neonatal/3	Dzx-, Octr+	Yes	No	D	1.1	1.0	1.0	No
44/F	2.0/10	Dzx-, Octr-	No	No	D	1.0	1.0	1.0	Yes/A
45/M	Neonatal/1	Dzx-, Octr-	Yes	No	D	1.0	0.9	1.1	Yes/D
46/F	Neonatal/7	Dzx-, Octr-	No	No	D	1.1	1.0	1.0	No

Table 1 (continued)

Patient/ sex	Age at diagnosis/PET (months)	Response to medication	MRI	PVS	PET			Surgery/histology (type of HI)		
					Type of HI ^a	Type of HI ^a	SUV _r ^b			
							Right		Middle	Left
47/M ^c	Neonatal/1 and 2	Dzx-, Octr-	Yes	No	F	1.3	0.9	0.8	No	
						1.3	1.0	0.7	Yes/F	
48/M	Neonatal/8	Dzx-, Octr+	Yes	No	D	1.1	1.0	0.9	No	
49/F	Neonatal/11	Dzx-, Octr+	Yes	No	D	1.0	1.0	1.0	No	

Dzx- not responsive to diazoxide treatment, Dzx+ responsive to diazoxide treatment, Octr- not responsive to octreotide treatment, Octr+ responsive to octreotide treatment, ND not determined, F focal, D diffuse, A atypical

^a Form of HI identified by PVS and PET (visual inspection)

^b Quantitative analysis of [¹⁸F]fluoro-L-DOPA uptake PET (individual SUV_r values for the three pancreatic regions)

^c First PET study performed without medication and the second one under diazoxide and octreotide administration

^d Second PET study performed after carbidopa treatment

^e Both PET studies were done before each surgical intervention (focal forms)

^f PET study performed under glucagon infusion

^g Glucagon administration was stopped 5 days before the PET study

propionic acid or carbidopa (patient 14); (b) two patients had a PET study 72 h after drug withdrawal and another PET following administration of octreotide and diazoxide (patients 12 and 47); (c) finally, two patients were operated on twice within 1 year and were submitted to a PET study before each surgical resection (patients 16 and 37). During all PET studies, normoglycaemia was maintained through glucose infusion, which was carefully adjusted according to frequent blood glucose monitoring. Glucose infusion rates between 6.4 and 13.2 mg/kg/min were required. PET acquisition was performed under light sedation (pentobarbital associated or not with chloral hydrate). Twelve of the 49 children were also submitted to a PVS (Table 1) after drug withdrawal (the octreotide was stopped for at least 3 days and diazoxide for at least 5 days).

Fifteen patients in whom [¹⁸F]fluoro-L-DOPA uptake results strongly suggested focal HI and nine patients with diffuse HI resistant to medical treatment ($n=9/34$) had surgical resection. Pancreatic tissue obtained from resections was fixed in formalin and embedded in paraffin; serial sections were studied by immunohistochemistry after a water bath antigen retrieval step. The primary antibodies used were: anti-proinsulin (1/400 mouse monoclonal antibody, 1G4, Novocastra), anti-chromogranin A (1/200 mouse monoclonal antibody DAK-A3, DAKO), anti-synaptophysin (1/50 rabbit polyclonal antibody A0010, DAKO) and anti-DOPA decarboxylase or anti-DDC (1/100 rabbit polyclonal antibody, Chemicon international) [32, 33].

Magnetic resonance imaging

Thirty-one patients underwent MRI of the abdomen using a 1.5-T imager (Signa, GE). T1-weighted SPGR (spoiled gradient acquisition at the steady state) acquisition with inversion

recovery was performed to allow three-dimensional reconstruction of MR images. MRI was used to reveal potential signal abnormalities in the pancreas and to allow the co-registration between PET and MRI.

Positron emission tomography

The PET studies were performed using an ECAT EXACT HR+ scanner (Siemens Medical Solutions). Intravenous injection of 4.2 ± 1.0 MBq/kg [¹⁸F]fluoro-L-DOPA was performed 30–50 min before transmission acquisition. Then, a thoraco-abdominal emission scan (3D acquisition mode) was started between 45 and 65 min after radiotracer injection [32]. Depending on the height of the child, two to four bed positions were acquired. The emission sets were corrected for scatter using a model-based correction, allowing simulation of the map of single scatter events. The images were reconstructed using an attenuation-weighted ordered subset expectation maximisation iterative algorithm with four iterations and six subsets. The final spatial resolution in reconstructed images was approximately 6.0 mm.

For each patient, based on individual MRI when performed, but using PET images, three regions of interest (ROIs) were defined over the right, middle and left pancreas corresponding to the head, isthmus–body and body–tail, respectively. Regions were identified on one to four contiguous slices. The mean radioactivity concentration in each ROI (Bq/ml), measured about 60 min post injection, was divided by the injected dose of the radiotracer (in Bq and corrected for the radioactive decay between injection and scan start times) and the body weight (grams) to calculate standardised uptake values (SUVs). Then, ratios between each SUV region and the mean pancreatic SUV were calculated (SUV_r).

Based on visual analysis, patients were classified into two groups—focal and diffuse—and quantitative analysis was performed using SUV_r values. Comparisons between focal and diffuse SUV_r were performed using an ANOVA statistical test.

The PET images were also co-registered to MRI slices. Owing to the low contrast observed between kidneys and surrounding organs on MR images, enhancement of the grey level intensity of these structures based on a manual segmentation of the kidneys was performed. Volume-based co-registration of PET and enhanced MR images was done using mutual information as the matching criterion [34]. The co-registration was evaluated visually using a fusion mode, taking into account the superimposition of the liver and the kidneys in both modalities.

Pancreatic venous samples

Twelve children were submitted to a PVS study. PVS allows collection of venous blood samples from the entire pancreas for measurements of plasma glucose, insulin and C-peptide levels. The PVS studies were performed under general anaesthesia [22, 23]. Normoglycaemia was maintained throughout glucose infusion, which was carefully adjusted on the basis of frequent blood glucose monitoring. Maximal glucose infusion rates between 6.4 and 20 mg/kg/min were needed.

Results

All pancreatic MRI studies were uninformative concerning the differentiation between focal and diffuse HI but confirmed the localisation of [^{18}F]fluoro-L-DOPA in the pancreas. PET images showed that most of the radioactivity accumulated in the kidneys and urinary bladder, the main elimination route of the radiotracer. Furthermore, in some patients, we observed a physiological radiotracer accumulation by the gall-bladder and the biliary duct, which could indicate an intestinal elimination, probably diet related. The

physiological accumulation was observed even though all patients fasted for at least 6 h before the PET examination and the image acquisition was started between 45 and 60 min after the radiotracer injection. Consequently, the high radioactivity in these organs, and particularly in the left kidney, might increase the difficulty in identifying focal forms localised in the tail of the pancreas when the PET images alone are interpreted. Co-registered MR and PET images help to identify whether the hot spot is localised in the head, the body or the tail of the pancreas. This anatomical localisation of the hot spot might appear rather coarse but practically accounts for misregistration error that may occur for the abdomen.

In 15 of the 49 patients, focal uptake (“hot spot”) of [^{18}F]fluoro-L-DOPA was observed in the pancreatic area (Fig. 1a). In these patients, the area of the suspected pancreatic abnormality, based on imaging diagnosis, was biopsied. In 14 children, the frozen section confirmed that the histological abnormality was confined to the focal lesions, and a limited pancreatic resection was performed. The immunohistochemical analysis corroborated the PET data, showing that the distribution of abnormal β -cells was restricted to the adenomatous hyperplasia in all but one patient. The abnormal β -cells identified by their overexpression of proinsulin, synaptophysin and chromogranin A also overexpressed DDC (Fig. 1b). In one patient (no. 3) no focal lesion was observed by the surgeon and the immunohistochemical analysis showed an atypical form of HI. In atypical forms, such as that presented by patient 3, active and inactive parts were coexisting in the same Langerhans islets and DDC was mainly positive in the β -cells recognised by their large size.

In the 14 children in whom a focal lesion was confirmed by immunohistochemical analysis, the limited pancreatic resection was followed by clinical remission without carbohydrate intolerance.

Concerning the accuracy of PET in the localisation of focal adenomatous hyperplasia, the PET findings were corroborated by surgery in 13 of the 14 children with focal lesions. The foci were localised in the right of the pancreas

Fig. 1 Patient with focal HI. **a** The abnormal hot spot is visualised in the middle region of the pancreas on coronal slices (yellow arrow). Physiological distribution of the radiotracer with higher accumulation in the urinary bladder and a lower accumulation in the liver is also observed. **b** The abnormal hot spot corresponds to an important agglomeration of proinsulin, synaptophysin, chromogranin A and DDC in the lesion

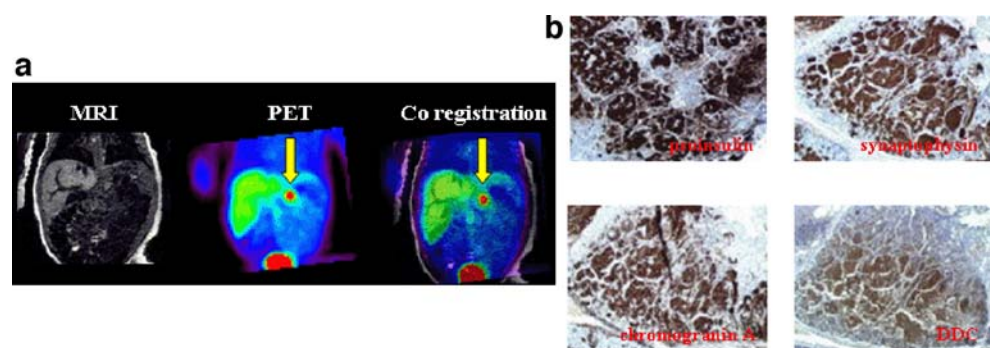
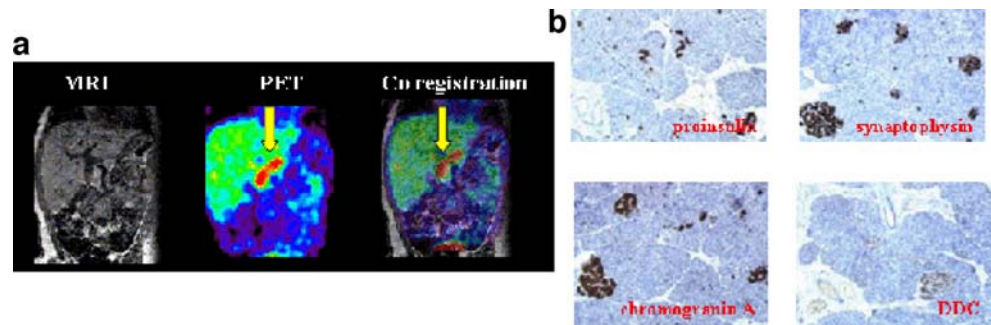


Fig. 2 Patient with diffuse HI. **a** Higher and diffuse uptake of the radiotracer is observed over the pancreatic area (coronal slices, yellow arrow). Lower hepatic accumulation corresponding to the physiological distribution of the radiotracer is also visualised. **b** Corresponding immunohistochemical results (proinsulin, synaptophysin, chromogranin A and DDC) obtained in the same patient after subtotal pancreatectomy



in nine cases, in the middle in two and in the left in three. In one child (patient 1), an abdominal median uptake hot spot was observed on PET images but the surgeon found adenomatous hyperplasia in the right pancreatic region corresponding to the head of the pancreas.

When diffuse accumulation of [^{18}F]fluoro-L-DOPA (Figs. 2a, 3a) was observed ($n=34$), diffuse HI was suspected. Nine of these 34 children were resistant to the medical treatment and were submitted to surgery. In seven cases a near-total pancreatectomy was performed while in two cases (patients 5 and 44) a hemi-pancreatectomy was carried out, after frozen section histological examination of multiple random pancreatic biopsies. The PET results were confirmed by histological data. In diffuse forms, the

abnormal pancreatic cells, identified by their expression of proinsulin, synaptophysin, chromogranin A and DDC, were gathered in small clusters scattered throughout the pancreas (Fig. 2b). In patients 5 and 44, the histological data were compatible with an atypical form (Fig. 3b).

In patients 12 and 47, no differences in [^{18}F]fluoro-L-DOPA uptake were observed between the two PET studies performed without and with octreotide plus diazoxide.

One child (patient 14) suspected of having the diffuse form of HI on the basis of PVS and PET, was treated with an inhibitor of dopa decarboxylase (carbidopa). The diffuse uptake found before treatment disappeared completely after carbidopa treatment. In one case (patient 26), the PET study was performed under glucagon infusion and showed no uptake of the radiotracer in the pancreas [33].

Twelve children were also submitted to a PVS. In 11 cases, the PET and PVS results were concordant: six focal and five diffuse HI forms suspected on PET images were corroborated by PVS findings. In one patient (no. 5) in whom diffuse HI was suspected on PET, the PVS suggested focal HI. This patient was submitted to a hemi-pancreatectomy and in all tissue slices, histological analysis identified only quiescent Langerhans islets.

For each subject, SUV_T values are given in Table 1. For diffuse forms, SUV_T values were not statistically different between the middle and the left pancreatic region (mean \pm SD 0.97 ± 0.07 and 0.99 ± 0.08 for the middle and the left pancreas respectively). For the right pancreatic region (corresponding to the head of the pancreas), the SUV_T (1.06 ± 0.07) was significantly increased by 8.7% ($p < 0.0001$) when compared with the other two regions.

For focal forms, the [^{18}F]fluoro-L-DOPA uptake was higher for the “hot spot” (1.44 ± 0.17 , $p < 0.0001$), independently of its localisation, while no differences were observed for the remaining pancreas (0.79 ± 0.02 and 0.78 ± 0.11). The aforementioned value corresponds to an increase of nearly 84%. Comparisons between the two groups showed significantly higher SUV_T values in the focal “hot spot” compared with SUV_T values measured in diffuse forms of HI ($p < 0.0001$).

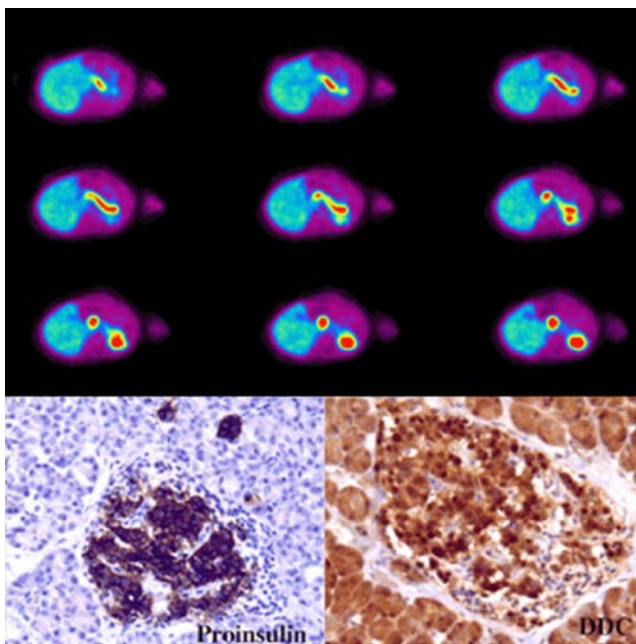


Fig. 3 **a** Axial PET images obtained for a patient with an atypical HI show high and diffuse uptake of the radiotracer over the pancreatic area. Physiological accumulation in the left kidney is also observed. **b** Histological data (proinsulin and DDC) revealed an atypical form of HI

Discussion

In this study including 49 patients with HI, we have corroborated our previous results showing that PET using [^{18}F]fluoro-L-DOPA can differentiate focal and diffuse HI [32]. When focal uptake of [^{18}F]fluoro-L-DOPA was detected, the immunohistochemical data obtained at surgical resection confirmed the diagnosis of focal HI (14/15). On the other hand, histological data from seven out of nine operated children with a diffuse pattern of [^{18}F]fluoro-L-DOPA uptake revealed wide dispersal of the pathological β -cells throughout the pancreas. The histological findings corroborated the PET results in 93% of the focal forms and in 85% of the operated patients with diffuse forms and illustrated pancreatic β -cell co-localisation of proinsulin and DDC in focal HI [33]. Finally, two of the three atypical HI forms were identified by PET as diffuse HI and one as focal HI. Our results are consistent with those published recently by Hardy et al. These authors estimated the accuracy of [^{18}F]fluoro-L-DOPA PET in diagnosing HI to be 96% [35]. Concerning the localisation of focal forms, we correctly localised 92%, while the Philadelphia group reported that PET correctly indicated the location of all the lesions (100%).

In the present study, lesion localisation was performed using PET images or fused PET and MR images when available. MRI was not acquired using the same bed as the PET study. As a consequence, the movements of the abdominal organs between the two imaging modalities were more pronounced than they would have been had a combined PET/CT been used for acquisition of functional and anatomical images. However, MRI has the advantage of reducing the dose exposure of the subject as compared with CT.

The 14 children with focal HI who underwent a limited pancreatectomy showed normal postoperative blood glucose levels. At present, these 14 children are in clinical remission.

Seven children with diffuse HI had a near-total pancreatectomy. These children had clinical follow-up, taking into account the amount of residual pancreatic tissue that was left after surgery. The surgery was followed by a series of hypoglycaemic episodes of variable severity and frequency, and almost all of these children developed diabetes mellitus.

The three operated atypical HI forms presented a diffuse histological pattern involving an area corresponding to half of the pancreas. For atypical forms, partial surgery (hemipancreatectomy) followed by close medical supervision is recommended. In fact, sometimes hypoglycaemia is not cured by partial pancreatectomy. Nevertheless, the hypoglycaemia can be medically controlled. One of the three patients identified as having an atypical form and submitted

to hemipancreatectomy is currently in clinical remission while the other two children are under clinical treatment.

The 25 children with a diffuse form of HI who did not undergo surgery are being maintained under medication.

Twelve children were also submitted to a PVS, and in 11 of these cases the PET and PVS agreed. Ten of these children were submitted to surgery and the histological data confirmed the results from both methods in nine of these cases. In the case in which PVS and PET findings diverged, histology showed an atypical form. Thus, the diagnosis provided by PET is as precise as that obtained by PVS, and PET is adequate to guide surgical resection in most cases.

Quantitative analysis using SUV_T allows discrimination between the two forms of HI and completes the visual inspection. In the previous report, 15 children were included and we used only the SUV to compare the two HI forms [32]. Although the pancreatic SUV seemed higher in focal HI than in diffuse HI, the difference was not statistically significant. This result was most probably due to the small number of patients with focal HI studied. In the present work, 49 children were included and we used the SUV_T to distinguish between focal and diffuse HI. For all cases classified as diffuse HI, the SUV_T values were lower than 1.2. High values of this parameter were obtained for the right pancreas, corresponding mostly to the head of the organ, probably owing to a high concentration of β -cells in this region. In a recent study of five cases of focal HI, Otonkoski et al. showed that the SUV_T was higher than 1.5 [36], while in our work it ranged between 1.2 and 1.8.

In one patient, the diffuse [^{18}F]fluoro-L-DOPA uptake observed in the pancreas before treatment with carbidopa, a DDC inhibitor, was no longer detectable after its administration. This result demonstrates, *in vivo*, that pancreatic β -cells are able to take up L-DOPA, an amino acid precursor, and contain the enzyme DDC responsible for the conversion of [^{18}F]fluoro-L-DOPA into [^{18}F]fluoro-dopamine. [^{18}F]Fluoro-L-DOPA is transported across the cell membrane by the amino acid transporter. Then, it is decarboxylated into [^{18}F]fluoro-dopamine, which is stored in vesicles. When decarboxylation is prevented by a DDC inhibitor, such as carbidopa, [^{18}F]fluoro-L-DOPA could be released from the tissue. Thus, the tracer uptake shown by PET before treatment disappeared after carbidopa administration [33]. Carbidopa, usually indicated in the treatment of Parkinson's disease, induces cytotoxicity against human tumour lines, especially small cell lung carcinoma, and carcinoid [37, 38]. Both are neuroendocrine diseases, like HI. Carbidopa administration has been found to reduce the number of hypoglycaemias in children with a diffuse form resistant to diazoxide and octreotide. However, after a short period of relapsed hypoglycaemia, the treatment became ineffective and surgery was indicated [33].

The effect of the medications (octreotide and diazoxide) generally used for HI treatment on [^{18}F]fluoro-L-DOPA uptake was evaluated in two children: one with a diffuse form and one with a focal form. Both children had two PET studies: one without medication and one with medications. The uptake of [^{18}F]fluoro-L-DOPA remained unchanged between the two PET studies. Thus, in contrast to PVS, PET studies could be performed without stopping these medications. In contrast, glucagon (a potent anti-hypoglycaemia agent) administration interferes with the uptake of [^{18}F]fluoro-L-DOPA: in patient 26 this drug was not stopped before the PET study and no pancreatic tracer uptake was observed; in patient 42, the glucagon administration was stopped 5 days before the PET examination and no interference between this drug and [^{18}F]fluoro-L-DOPA uptake was observed. In a previous study, we demonstrated a correlation between insulin and DDC localisation [33]. This correlation is associated with the close anatomical relationship between pancreatic α - and β -cells, and might explain the absence of [^{18}F]fluoro-L-DOPA uptake under glucagon perfusion. In fact, the interaction between insulin and glucagon release within the islet of Langerhans is well known [39]. Glucagon, interfering with β -cell activity, will probably also interfere with DDC activity [40, 41].

Two patients (nos. 16 and 37) had two PET examinations within 1 year. In both patients, both PET images identified focal [^{18}F]fluoro-L-DOPA uptake on the right side of the pancreas corresponding to focal HI. Both patients were submitted to partial pancreatic resection after each PET study. In patient 16, the histological analysis concluded that two individual focal lesions were present, situated at the pancreatic head. However, owing to the PET image spatial resolution, we could not individualise the two lesions on the first study. In patient 37, the lesion was not found at the first surgery, most probably owing to its difficult access (posterior side of the pancreatic head).

Conclusion

The results of this report including 49 children confirm that PET with [^{18}F]fluoro-L-DOPA is valuable for the differential diagnosis between focal and diffuse HI using a qualitative analysis associated with quantitative measurements. In contrast to PVS, PET is non-invasive and atraumatic. Furthermore, PET studies may be performed without stopping medication (with the exception of glucagon and also carbidopa, which is not yet recommended for HI therapy) and under light sedation. PET results were concordant with the immunohistochemical analysis performed after partial (focal HI) or near-total pancreatectomy (diffuse HI resistant to medical treatment) for 21 out of the 24 children submitted to surgery.

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