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Clinical Case Seminar A2(1-8)

Non Obvious Diagnosis of an Occult ACTH Dependent Cushing Syndrome

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Abstract

A 54 years old man was admitted to our hospital due to progressive signs and symptoms of Cushing syndrome. Once a biochemical diagnosis of Adrenocorticotropic Hormone (ACTH)-dependent hypercortisolism was established, high dose 8 mg overnight Dexamethasone Suppression Test (HDDST), 1-deamino-8-D-arginine vasopressin (DDAVP) stimulation test and a Magnetic Resonance Imaging (MRI) led to conflicting results and an ectopic ACTH syndrome was diagnosed following a Bilateral Inferior Petrosal Sinus Sampling (BIPPS).The localization of the source of ectopic ACTH secretion turned out to be a challenging task because most of imaging exams gave a negative result. After a prolonged follow-up, a chest Computed Tomography (CT) scan gave a morphological confirmation of a small focus in the right lung previously detected by a ^{68}Gallium-DOTATOC- Positron Emission Tomography (PET). A right lower lobectomy of the lung was performed and an ACTH-positive typical pulmonary carcinoid was diagnosed. Before surgery, a good management of hypercortisolism was obtained with somatostatin analog lanreotide for years, and only after a likely escape phenomenon was successfully prescribedoff-labelpasireotide. In this patient with occult ectopic ACTH syndrome (EAS),a watchful waiting approach, based on imaging re-evaluation,representeda valuable option, provided that a good management of hypercortisolism and its end-organ complications wasobtained.

Key-Words: Cushing; Ectopic; Covert; Occult

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Introduction

The evaluation of hypercortisolism may be difficult due to the lack of specific signs and symptoms and tothe need to excludea pseudo-Cushing syndrome by multiple laboratory tests [1].The EAS is considered among the most challenging endocrine diseases [2-3] because of pitfallsin both diagnostic and therapeutic options [2, 3-5].The localization of an occult tumor with imaging techniques is often difficultand about 20% of ectopic ACTH-secreting tumorsare not localized[4, 6-7].Overexpression of somatostatin receptors inNeuroEndocrine Tumors (NETs) cells and in many ectopic ACTH secreting tumors has important implications for imaging and therapeutic options[8].When a clearevidence is not available by imaging,Cushing's comorbidities management anda long-term follow-up is mandatory[2, 4-7, 9].

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Case Report

A 54 years old man was admitted to our hospital with long-standing signs and symptoms of Cushing syndrome.His previous medical history was characterized by a weight gain of approximately 20 kg in the last years, a five-years hypertension and dyslipidemia that have been treated with irbesartan, amlodipine and a statin. During a previoushospitalization for the evaluation of a peptic ulcer, he was diagnosed with multiple painless rib fractures and moderate bilateral adrenal hyperplasiaonCTscan, whilelaboratory tests revealed the presence of lymphocytopenia and potassium 3.5 mEq/L. He complained fatigue combined with easy bruising, poor skin healing and muscle weakness and he showed a facial plethora at physical examination.Once the patient was admitted to our hospital, endocrinological laboratory evaluation [Table 1] revealed elevated 24 hours urinary free cortisol (24hCLU) levels (760 µg/24 h), absence of suppression of serum cortisol at Low dose 1 mg overnight Dexamethasone Suppression Test (LDDST) and elevated midnight serum cortisol levels, sothe diagnosis of Cushing syndrome was confirmed. Asubsequent evaluation,showed ACTH levels (240 pg/ml) consistent with an ACTH-dependent cause of Cushing Syndrome. Concerning HDDST,a >50% cortisol suppression was observed and the gadolinium-enhanced pituitary MRI revealed no abnormalities. Chromogranin A (CgA) was normal and other hormonal abnormalities were excluded [Table 4]. Because of the inconclusive result of the previous tests, the patient was sent to an experienced referral center (San Raffaele Hospital, Milano) to perform a BIPSS. During the hospitalization,the patient underwentDDAVP stimulation test,which showed a positive ACTH and cortisol response [Table 2a]; BIPSS with Corticotropin Releasing Hormone (CRH) stimulation showed an ACTH petrosal sinus/peripheral ratio consistent with an ectopic cause of the ACTH secretion [Table 2b]. An acute octreotide challenge test (OctCT) was performed and showed a good response [Table 3]. Chest and abdomen CT were negative and the somatostatin receptor scintigraphy with indium 111-pentetreotide (Octreoscan, OCT) did notdisplayany pathological uptake.Due to the biochemical and imaging studies suggestingan occult Ectopic ACTH Syndrome and the lackof life-threatening hypercortisolism in the short term,the disease was managed by medical therapy and a watchful waiting approach was adopted. During the follow-up,imaging techniques were repeatedregularly [Table 1]. Three68Gallium-DOTATOC-PETs were performed in the following years and they always showed a small area in the right lung,which was not confirmed by CT scan. Another gadolinium-enhanced MRI of the pituitary revealed no abnormalities. The absence of a morphological confirmationof the lesion by CT scan, prevented a surgical approach until the last CT identified a focal lesion in the right lung concordant with the focus showed by 68Gallium-

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DOTATOC-PETs.

Table 1. First/second level diagnostic evaluation and imaging investigation. I and II level diagnostic tests were performed between november 2008 (Garibaldi Nesima hospital, Catania) and march 2009 (S. Raffaele hospital, Milano).

	Year	Results	Notes
C count	2008	WBC 13900/mL; LY 9.5%; NE 84%	
	2009	WBC 16400/mL; LY 19.4%; NE 74.6%	
Potassium	2008	4.6 mEq/L	
	2009	3.8 mEq/L	
I level			
24hCLU	2008	760 µg/24h (RV 32-243)	
LDDST	2008	36.3 µg/dL	Administration of 1 mg of dexametasone at 12 pm followed by cortisol measurement at 8 am
AMC	2008	23.7 and 20.1 µg/dL	Two cortisol determinations at 11 and 12 pm respectively
II level			
ACTH	2008	240 pg/mL (RV <60)	
HDDST	2008	Cortisol suppression 51.84%	Basal cortisol: 24.5 µg/dL Post-suppression test cortisol: 11.8 µg/dL
CRHST	\	\	Not Available
Pituitary MRI	2008	Negative	
DDAVPStimulation Test	2009	ACTH increment 227% Cortisol increment 69%	See table 2a for details
BIPSS with CRH stimulation	2009	ACTH ratio < 2 (basal) ACTH ratio < 3 (after CRH stimulation)	See table 2b for details
OctCT	2009	ACTH suppression after 4 h: 89.5% Cortisol suppression after 4 h: 63.4%	See table 3 for details
CgA	2008	46 ng/mL (RV <101.9)	
NSE	2009	21.6 µg/L (RV 0-12.5)	
Calcitonin	2009	2 pg/mL (RV 1-12)	
Imaging			
Total body CT	2008	Negative	
OCT	2009	Negative	
Total body CT	2010	Negative	
PET	2010	Small focus in the right lung	Computed tomographies performed between 2010-2014 are not currently available. They always showed a negative result
PET	2014	Small focus in the right lung	
Pituitary MRI	2014	Negative	
Total body CT	2015	Negative	
PET	2017	Moderate focus in the right lung	
Total body CT	2017	Focal lesion in the right lung	

AMC: Awake Midnight serum Cortisol, CRHST: CRH Stimulation Test, LY: Lymphocytes, NE: Neutrophils, NSE: Neuron Specific Enolase, RV: Reference Values, WBC: White Blood Cells

Table 2a. DDAVPStimulation Test. ACTH (pg/ml) and cortisol (µg/dL) measurement before and after administration of 1-deamino-8-D-arginine vasopressin. Information about desmopressin dosage is unavailable.

	ACTH (pg/mL)	ACTH rise (%)	Cortisol (µg/dL)	Cortisol rise (%)
Basal	44		26.4	
15'	144	227%	38.0	
30'	144		43.3	
45'	119		44.5	
60'	97		44.7	69%

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Table 2b. BIPSSwith CRH stimulation. ACTH measurement (pg/ml) before and after administration of 100 µg CRH iv. Information about the ovine or human nature of CRH is not available.

	Peripheral venous blood	Right inferior petrosal sinus venous blood	Right PS/peripheral ratio	Left inferior petrosal sinus venous blood	Left PS/peripheral ratio
Basal	33	40	1.21	34	1.03
2'	32	51	1.59	40	1.25
5'	36	50	1.39	42	1.17
10'	36	50	1.39	46	1.28
15'	35	\	\	45	1.29

PS: Petrosal Sinus

Table 3. Octreotide Challenge Test: determination of ACTH and cortisol before and after administration of Octreotide, 1 mg sc

	ACTH (pg/ml)	Cortisol (µg/dL)
Basal	57	290
60'	14	196
120'	11	169
180'	13	147
240'	6	106

Table 4. Hormonal evaluation

	2008	RV
TSH	0.55 µU/mL	0.35-4.94
FT3	2.0 pg/mL	1.71-3.71
FT4	0.87 ng/dL	0.7-1.48
FSH	4.11 mIU/mL	1.37-13.58
LH	2.1 mIU/mL	0.57-12-07
Testosterone	4.4 ng/mL	1.42-9.23
Prolactin	22.14 ng/mL	2.58-18.12
PTH	43.1 pg/mL	15-68
Calcium	9.7 mg/dL	8.4-10.2
Phosphorus	2.8 mg/dL	2.3-4.7

FSH: Follicle Stimulating Hormone, FT3: Free Triiodothyronine, FT4: Free Thyroxine, LH: Luteinizing Hormone, PTH: Parathyroid Hormone, TSH: Thyroid Stimulating Hormone

A right lower lobectomy of the lung was therefore performed and,onhistopathological examination, an ACTH-positive typical pulmonary carcinoid was diagnosed [Table 5]. Post-operative laboratory evaluation revealed remission of hypercortisolism and cortone acetate was prescribed to manage a glucocorticoid withdrawal syndrome (37.5 mg daily).During the prolongedfollow-up before surgery, hypercortisolism was managed with a medical approach [Table 6]. The first drug prescribedwas ketoconazole (400 mg/die), even if it was discontinued few months later, due to the need to perform dynamic tests. Considering the responsiveness to a somatostatin analog with the octreotide challenge test, the patient was therefore treated with lanreotide at an initial dose of 60 mg im/28 days with clinical and biochemical normalization of cushingoid features. 24-h urinary free cortisol and ACTH levelswere around the normal range and a satisfying Cushing's features management during the follow up was obtained. During the subsequent follow-up,lanreotide dose was increased to 90 mg/28 days and later to 120 mg/28 days untilalaboratory evaluation revealed elevated

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24-h urinary free cortisol levels (309 and 348 mcg/24 h) andlanreotide was therefore discontinued.Then, the patient was treated with pasireotide 0,6 mg sc twice daily off-label,obtaining the normalization of 24-h urinary free cortisol levels in two months.

Table 5. Histopathological examination of the ACTH-secreting tumor

Diagnosis	Typical ACTH-secreting carcinoid
Size	7 mm
Proliferation index (Ki67)	2-3%
Mitotic index	0x10HPF
Immunophenotype	Synaptophysin (+), CgA (+), ACTH (+), CKpool (+), TTF1 (-), S100 (-), SOX10 (-), GATA3 (-), MelanA (-)

HPF: High Power Fields

Table 6. Main therapeutic steps. 24hCLU before and after therapeutic modifications are reported.

Therapy	Posology	Year of prescription	24hCLU (µg/24h) RV 10-110		Notes
Ketoconazole	400 mg/die	November 2008	\		Discontinued 2 weeks before performing march 2009 hormonal evaluation
Lanreotide			Before	After	
	60 mg im/28 days	April 2009	\	\	
	90 mg im/28 days	September 2010	\	2013: 75 2013: 69 2014: 10 2014: 13	
	120 mg im/28 days	June 2015	379	2010: 76 2010: 72	
Pasireotide			Before	After	Discontinued in june
	0.6 mg sc twice/daily	January 2017	309 348	76 45.5	2017, before performing right lower lobectomy of the lung. The patient showed a poor biochemical control of hypercortisolism in the last month before discontinuation.
Surgery		June 2017			Right lower lobectomy of the lung

Discussion

Our patient showed most of features that specifically characterize Cushing's syndrome (easy bruising, facial plethora, proximal muscle weakness[1]), and the first diagnostic steps were quite straightforward witha diagnosis of ACTH-dependent hypercortisolism. Although overlapping values are often observedbetween Cushing disease and ectopic ACTH syndrome [4-5],elevated ACTH levels suggested an ectopic ACTH-dependent hypercortisolism. HDDST and DDAVP

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Stimulation Test, performed as second line diagnostic evaluation,suggested a Cushing Disease. Most ectopic ACTH-secreting tumors responsive to HDDST are pulmonary carcinoids [4-6], and since MRI was negative, BIPSS was performed as gold standard technique to rule out the possibility of a Cushing Disease[3,4-5]. Despite BIPSSis characterized by high sensibility and specificity, >95% in most series [3,7], it is interesting to note that false positives have been reported in ACTH secreting bronchial carcinoids [5].A CRH Stimulation Test was not performed because not available at that time. However, in the case oftwo dynamic testssuggesting a pituitary ACTH-secreting adenoma and a negative MRIa BIPSS should have been performed to discriminate the source of the ACTH secretion.Once a biochemical diagnosis of EASis given,the diagnostic challenge is the localization of the ACTH-secreting tumor, followed by its surgical removal(as first-line option)[9].According to the currentliterature,approximately50% of ACTH secreting tumors are detected at the initial investigations ("overt"), 20% arenot ("occult") and 30% are localized at thesubsequent follow-up ("covert") [6-7]. In our patient,we were not able to localize the tumor for several years, even withthe useofmultiple imaging techniques and a long-term follow-up; only 9 years after the initial biochemical diagnosis of EAS, a typical pulmonary carcinoid was diagnosed (making our case a challenging "covert" EAS).

Most tumours underlyingectopic Cushing syndromearelocalized in the lung [6] and CT isconsidered the "most useful first examination", with a sensitivity >98% in overt cases. MRI, OCT, and PET are alsouseful imaging techniquesto confirm a previousimaging or to localize a still occult tumor. 68Gallium-SSTR (Somatostatin Receptor)-PET shows the greatest sensitivity in localizing covert cases, so it is considered a very useful second/third level examination and the best follow-up imaging technique[7]with an high impact on NETs clinical decision-making [11-12].

In our patient a CT total body and an OCT were performed as first-line examinations, while CT and 68Gallium-DOTATOC-PETrepresented the corner stone of the follow-up.

Since all available diagnostic techniquesmay provideboth false-positive and negative results, a double step approach is suitable. In this view, surgery may be performed only when bothfirst and second line exam are able to detect and localize tumor mass [7].In particular, the diagnosis of pulmonary carcinoids ischallenging because they are often small (average 1 cm) and, therefore, difficult to detect and localize [2,5-6].

While PET imaging has been described as "additive and synergistic to CT imaging" [10], CT imaging is mandatory in the diagnosis of NETs as underlined in the last Neuroendocrine tumors guidelines. Regarding PET imaging, four recent meta-analyses showed that 68Gallium-SSTR-PET hasaspecificity

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ranging 88-95% [8, 11-14].

The choice of a watchful waiting approach instead of surgery was carefully evaluatedalong with thesurgeon. Giventhe good controlof hypercortisolism by medical therapy, the functional information obtained by 68Gallium-SSTR-PET alone (without a CT image) was notconsidered specific enough to suggestsurgery.

Many treatment options are available for the medical management of the EAS [3, 9] and, among them, steroidogenesis inhibitors are often used for the treatment of an occult EAS. In our patient, one of the most currently used drugs (ketoconazole) was the first to be prescribed. A couple of months later it was discontinued because its inefficacy and targeted therapy with a somatostatin analog was initiated, on the basis of the positive result with octreotide challenge test [2, 5]. Escape to treatment,after a long-standing effective therapy,may be observed because the long-term use of somatostatin analogs may alter expression of surface receptors in tumor cells; in such cases, it may be helpful to increase the analog dose (until the highest recommended dose is reached) or to change the therapeutic approach [2, 3, 15]. Although few studies exist on the use of pasireotide for the management of neuroendocrine tumors [16-18], its off-label use was suggestedby the good biochemical control of hypercortisolism obtained with thesomatostatin analogfor several years. After a treatment with pasireotide 0,6 mg sc twice daily, a normalization of 24-h urinary free cortisol levels was obtained, before the surgical removal of the tumour. A study to evaluate the efficacy of pasireotide twice daily for normalizing 24-h urinary free cortisol in patients with EAS is ongoing and will be completed in December 2018 [19].

Conflicts of Interest: There is no potential conflict of interest, and the authors have nothing to disclose. This work was not supported by any grant.

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