Diagnostic pitfalls in digestive neuroendocrine tumours

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Abstract

Gastro-entero-pancreatic neuroendocrine tumours (GEP NET) represent a rare and highly heterogeneous entity that often is revealed by vague and non-specific symptoms, leading to a delayed diagnosis. Here we will review some of the most regularly observed false positive and false negative cases and provide clues to recognize and manage them properly. Particularly, the value of chromogranin-A as a serum tumour marker and Somatostatin receptor scintigraphy as an imaging test, are reviewed. Indeed, chromogranin-A and other hormones, such as gastrin, as well as urinary 5-hydroxy-indolic acetic acid (5-HIAA) are often tested to diagnose NET without appraising the clinical situation, leading to extensive work-up on false bases. On the other hand, some tests are performed in situations where they do not add additional information (e.g. 5-HIAA in pancreatic or rectal NET) because invariably negative. Somatostatin receptor scintigraphy is an expensive examination, still not reimbursed in Belgium, for which indications must be carefully assessed, knowing its specificity and sensitivity. (Acta gastroenterol. belg., **2009**, 72, **29-33**).

Key words : neuroendocrine tumours, chromogranin-A, Somatostatin Receptor Scintigraphy.

Introduction

NET form a group of heterogeneous and fascinating tumours. They are rare, although their incidence has more than tripled over the last 30 years in the US (1). Interestingly, because of their often indolent course, their prevalence is higher than that of gastric, oesophageal or pancreatic cancers in the US. Although they can manifest to the clinician with very characteristic symptoms such as flushing and secretory diarrhoea, they are much more often asymptomatic at early stage, and then show vague and misleading symptoms, that can compete with irritable bowel syndrome. Therefore sensitive and specific diagnostic tools are necessary to help doctors to "find a needle (i.e a small tumour) in a haystack (of symptoms)". The value of chromogranin-A and other hormones such as gastrin or serotonin metabolites that are frequently dosed when NET are suspected, can lead to frequent false positive cases and sometimes heavy decisions. It is thus important to recognize clinical situations responsible for the rise of serum or urinary markers, unrelated to tumours. Another quite unique feature of NET is the presence of somatostatin receptors (SSTRs) at the surface of tumour cells, which after binding to the natural somatostatins (SMS) or SMS analogues (SSA), inhibit the release of several hormones. The presence of SSTRs has led to the development of Somatostatin receptor scintigraphic methods for diagnosis and staging of tumours, the first one being the octreoscan®, which uses octreotide labelled with the gamma emitter 111-indium, and more recently octreotide analogs labelled with positron-emitting radionuclide. However, it is also important to bear in mind that physiological process can lead to false positive results of Somatostatin Receptor Scintigraphy, that must be recognized to avoid useless work-up and overtreatment.

Chromogranin-A

Chromogranin-A is a glycoprotein that is widely expressed by neuroendocrine cells and constitutes one of the most abundant components of secretory granules (2). Chromogranin-A is physiologically released by exocytosis and may be detected in the blood. If a tumour develops in an endocrine tissue, it becomes the main source of circulating Chromogranin-A (3).

Sensitivity and specificity of Chromogranin-A in NETs

The first study to assess accuracy of Chromogranin-A in peptide-producing endocrine tumours of all types showed 81% sensitivity and 100% specificity (3). Following that seminal study, various results have been found in terms of accuracy in small studies, with control groups not well defined. Recently, two studies, using high numbers of well-defined controls and NET patients

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have shown similar results, i.e. sensitivity of 84% and specificity of 85-96% (4,5). One of the 2 studies has also evaluated the value of Chromogranin-A in patients with tumours versus patients with chronic atrophic gastritis. They found that given a high specificity (set at 95%) due to a higher cut-off value, sensitivity dropped to 55%. These low values highlight the necessity of the carefull examination of the clinical status for a given patient. Especially, the use of Proton Pomp inhibitors (PPIs), renal failure, hypertension, which are frequent situations, need to be taken into account.

Chronic atrophic gastritis and use of Proton Pump inhibitors

The physiopathology behind atrophic gastritis and use of PPI is the same. The absence (atrophic gastritis) or inhibition (PPI) of acid production in the gastric fundus leads to a reaction of G-cells in the antrum, leading to gastrin production and secretion, in an attempt to stimulate acid secretion (6). This gastrin production is known to be a strong proliferative stimulus for the Enterochromaffin-like (ECL) cells of the gastric fundus (7). These ECL cells can develop into small, usually benign tumours called ECL-omas. ECL-omas have never been found in patients taking PPI, in contrast with rats chronically exposed to PPIs (8), suggesting that an additional factor is necessary to lead to tumour development in humans (9). Gastrin and chromogranin secretion are invariably found in patients having atrophic gastritis (10) or taking PPI, whether as acute or chronic treatment, up to 2-3 times the normal value compared to healthy controls (11). Duration of PPI exposure was associated with higher gastrin and Chromogranin-A values. Interestingly, in that same study, infection with Helicobacter Pylori (HP) was linked to a doubling of gastrin levels and a 30% increase of Chromogranin-A levels. Also, authors observed that use of H2-antagonists did not lead to an increase of Chromogranin-A. So, atrophic gastritis, long-term use of PPIs and HP infection represent frequent clinical situations that need to be recognized before embarking on an extensive work-up of the patient. Furthermore, a recent publication highlighted the fact that Chromogranin-A serum values should be assessed in a fasting state, because a doubling of its value after a meal was observed (12).

Renal failure

Early studies have clearly shown that renal failure increases Chromogranin-A levels (13). Authors demonstrated that even slightly elevated serum creatinine values (1.6 mg/dl) were associated with a threefold increase of Chromogranin-A values and that patients with end stage renal disease had values as high as 10 times normal. However no study assessed the diagnostic value of Chromogranin-A in the setting of renal failure, therefore no cut-off value can be proposed in patients with impaired renal function.

Essential hypertension

Patients with hypertension and normal renal function have higher Chromogranin-A values compared to normotensive matched control patients, with a 80% increase (14). This increase is appearing late in the course of the disease and is not influenced by anti-hypertensive treatments.

Gastrin

Gastrin is a hormone produced by G-cells of the stomach antrum in response to various stimuli. Particularly, physiologic (food) or pathologic (atrophic gastritis) situations related to acid secretion are a major cause of gastrin secretion. As gastrinomas are part of the spectrum of NETs and are the cause of symptoms such as diarrhea, GERD and more characteristically the so-called "Zollinger-Ellison" syndrome (ZES), it is important to assess the value of this specific hormone to avoid false positive cases.

Gastrin is highly influenced by food intake and should never be tested outside an overnight fast. In patients with HP infection, gastrin secretion is augmented, and meal leads to a further very important rise in gastrin level in these patients (15). Long standing achlorydria state, such as in atrophic gastritis or chronic use of PPIs are also major causes of non-tumoural elevation of gastrin values. PPI should be stopped for at least 7-14 days before testing again for gastrin. The measurement of gastric pH, the observation of atrophic fundic folds during endoscopy and the presence of anti-stomach antibodies are important features to search and recognize to differentiate atrophic gastritis from gastrinomas.

The diagnosis of the ZES can thus not rely only on fasting gastrin values. A provocation test should be performed, such as the secretin stimulation test, which showed the greater sensitivity and specificity in a large cohort of ZES patients (16).

NSE

Neurone-specific enolase (NSE), which is a marker of small cell lung cancer, has been tested in the setting of NET. This marker should definitely be abandoned, as a study showed already 10 years ago that it had a poor sensitivity (38%) and specificity (73%) in a large cohort of NET patients (17). This seems not to be the case in daily clinical practice, as shown by the Belgian survey of NET, the DNET registry, where NSE was found to be tested in as many as 74% of patients (18). In their paper, authors found NSE values to be exclusively elevated in poorly differentiated NET. If chromogranin-A is negative in such a patient, it can be suggested to test for NSE (17).

Serotonin and metabolites

Serotonin (5-hydroxytryptamin, 5-HT) is one of the main neuromodulators of the central nervous system. It

is usually produced by midgut NET, rarely by foregut tumours and never by rectal NET. Serotonin dosage in the blood is unreliable and should not be performed anymore. The dosage of its main metabolite, 5-hydroxyindolic acetic acid (5-HIAA) on a 24 h urinary collection is the gold standard, and should be proposed at least once to patients with midgut tumours because the usual carcinoid syndrome, which occurs in less than 10% of patients with midgut tumours (19), can frequently be underestimated by patients. The collection has to be performed on chlorydric acid and specific nutrients have to be avoided : bananas, avocados, plums, eggplant, tomatoes, plantain, pineapples, and walnuts, because they contain serotonin. Respecting these recommendations leads to a 88% specificity (20).

Somatostatin receptor Scintigraphy (SRS)

As NET have high affinity somatostatin receptors, scintigraphic methods have been developed in the early nineties to allow *in vivo* imaging (21). SRS using ^{111}In-pentetreotide, better known as Somatostatin Receptor Scintigraphy, is the most widely used device. Although sensitive and very helpful for whole body staging compared to traditional imaging methods (22), this technique has pitfalls, one of them being the poor spatial resolution, leading to the development of PET-based imaging, the most promising being ^{68}Ga-DOTATOC (23). As a consequence, the precise staging for liver metastatic extent before surgery is clearly suboptimal by SRS, as clearly shown by a study comparing CT scan, MRI and Somatostatin Receptor Scintigraphy in a large cohort of patients having liver metastases from NET, where MRI showed twice more metastases than SRS (24).

It is important to be aware that other tumours than NET can express somatostatin receptors and show uptake at SRS, such as lymphoma (25), meningiomas, paragangliomas, pheochromocytomas (26), but also non tumoural diseases, e.g. sarcoidosis or rheumatoid arthritis (27). Among NET, insulinomas are poorly detected, not because they do not harbour SSTRs but mostly because of their small size, below the detection threshold of SRS. Mutiple organs do take up labeled octreotide, such as the spleen, the liver, the thyroid, but also the gallbladder. This can sometimes be misinterpreted for a NET of the liver (Fig. 1), if patients are fasting at the time of imaging. To overcome this, repeated scans after a meal allow the clearance of the activity. Also, the colon (depending on the use of laxatives) and the ureters can take up the radioligand, which could be mistaken for peritoneal deposits. These pitfalls may not be encountered with the new radioligands, such as the ^{68}Ga-labelled somatostatin analogue DOTA-D-Phe1-Tyr3-octreotide (DOTATOC), with gallium-68 being a positron emitter and DOTATOC showing 10 times more affinity than octreotide for the subtype 2 of sst, allowing PET-CT examination, leading to a very precise anatomical localization of small tumours (Fig. 2). Due to rapid imaging

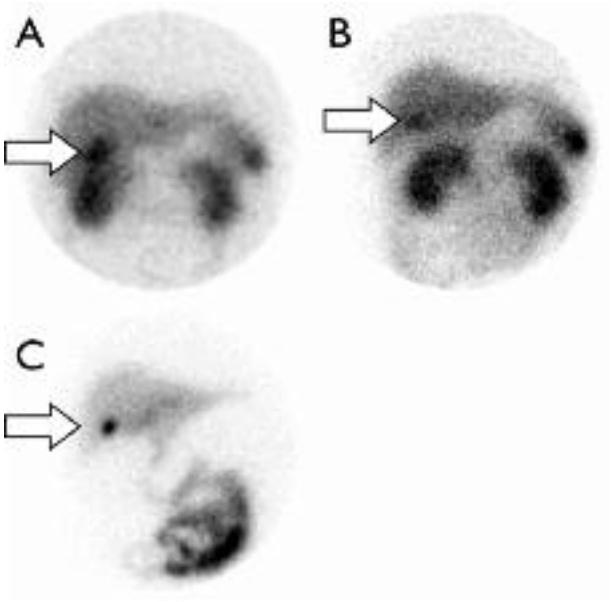

Fig. 1. — Somatostatin Receptor Scintigraphy showing uptake in the gallbladder, mimicking liver metastasis, 4 h (A) and 24 h (B) after injection, confirmed by 99Tc-HIDA scintigraphy (C).

bowel activity can be avoided, in contrast with ^{111}In-octreotide scintigraphy, where scanning peformed at 24 and/or 48 h leads to unavoidable bowel activity, even after use of laxatives. A small comparative study has shown superiority of this new imaging tool compared to Somatostatin Receptor Scintigraphy for the tumour detection rate, especially lung and bone lesions (23).

These early results need to be confirmed in larger trials, also comparing with traditional imaging techniques that have shown high sensitivity, e.g. MRI for liver lesions.

Currently, SRS is mandatory in case of any decision of surgical treatment of a foregut or midgut tumour. It is not useful in case of insulinoma, except for the rare malignant insulinomas. For appendiceal and rectal tumours less than 1 cm, SRS is also useless. It is complementary of conventional imaging, with a better sensitivity on a whole body scale, but as previously described, less sensitivity for the liver. Clinicians should be aware to perform SRS at least 4 weeks after last long acting SMS analogue injection, because of the competition for the same SSTR, leading to a significant drop of sensitivity.

Conclusion and research agenda

Diagnosis of NET remains a difficult task for the clinician. As serum chomogranin-A as a tumour marker is very often tested, it is very important to spread the information of its limitations. Fasting, avoidance of PPI and eradication of Helicobacter pylori infection are the prerequisite of an efficient measurement. A cut-off value has been proposed in the setting of chronic atrophic gastritis, with a high specificity, suggesting a high negative

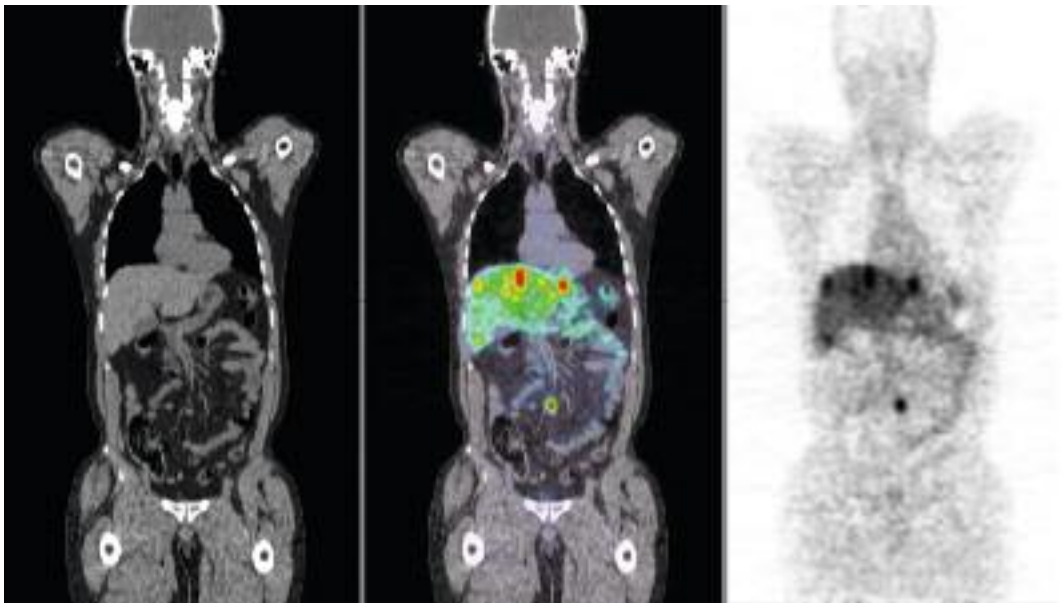

Fig. 2. — ^{68}Ga-DOTATOC PET-CT of a patient with carcinoid syndrome, diffuse liver metastases and a small unique mesenteric tumour.

predictive value. Questions remain wether the true value of an increased chromogranin-A in the setting of renal failure or a long history of essential hypertension. Also, question about its prognostic value remains unknown, although some recent literature sheds some light (28).

With regard to NET imaging, the future lies on the novel radiolabelled analogs, using positron emitting radioemitters, allowing PET imaging. These very promising tools need to be validated in the future before making them available to the medical community.

The Belgian registry launched by the Belgian Group of Digestive Oncology, the "DNET" registry, will help us assessing the value of these biochemichal markers and SRS imaging in Belgian patients with a proven diagnosis of NET.

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