

CASE REPORT

A rare case of clear-cell carcinoid of appendix

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Abstract

The carcinoids are the most frequent type of tumors arising from the appendix. In the majority of cases, these tumors are asymptomatic and usually are discovered after appendectomy. Definitive diagnosis relies on pathological examination of the resected appendix, size of the tumor being critical for the further management. Clear-cell change in neuroendocrine tumors (NETs) has rarely been described in the appendix. We choose to present a clear-cell carcinoid subtype of appendiceal NET to raise awareness on this potentially curable and rare condition that can be overlooked. We highlight the importance of the pathological exam and the morphological and immunohistochemical behavior of the tumor in confirming the diagnosis and aiding in the treatment decision making. Also, important entities should be considered in the process of differential diagnosis such as goblet-cell carcinoid or renal-cell/ovarian carcinoma.

Keywords: appendiceal NETs, carcinoid, neuroendocrine tumor, immunohistochemistry, clear-cell carcinoid.

Introduction

Neuroendocrine tumors (NETs) are a type of well-differentiated carcinomas with slow-growing pattern, originating in the cells of neuroendocrine system. Appendiceal NETs represent one of the most common types of malignant lesion of the appendix, being present in almost 0.3–0.9% of all appendectomies [1–3].

According to the last *World Health Organization* (WHO) Classification, NETs of the appendix include well-differentiated (G1) and moderately differentiated (G2) neoplasms [4]. Carcinoids are a G1 type and they account for 50–70% of all appendiceal neoplasms with an incidence of 0.075 cases/100 000/year [5]. They are slow-growing tumors with an excellent prognosis in the majority of cases due to its extremely infrequent metastases to lymph nodes or other locations. Typical morphological features include solid nests architecture composed of cells with classical neuroendocrine characteristics: small and uniform with finely granulated eosinophilic cytoplasm and centered nuclei with coarse chromatin. Apart from this classical appearance, there have been described in the literature rare variants of clear-cell carcinoids.

We are presenting a case of clear-cell carcinoid with further discussions on microscopic findings, differential diagnosis and prognosis.

Case presentation

We present the case of a 27-year-old Caucasian female patient, complaining of rapid onset epigastric tenderness, right lower quadrant pain, nausea and a low-grade fever, with symptoms occurring in the last six hours. This

patient lives in urban environment and has no significant prior medical history. The date hospital evaluation was November 7, 2016. Initial laboratory findings revealed leukocytosis with neutrophilia. The results of the abdominal ultrasound evaluation were within normal limits. The patient was admitted at the 2nd Department of Surgery (Emergency University Hospital, Bucharest, Romania) with diagnosis of acute appendicitis (admission Chart No. 102088) and emergency appendectomy was performed. Intraoperative, besides the positive diagnosis of acute appendicitis, it was discovered a 19 mm tumor at the tip of the appendix, with no perforation or invasion of the body, base, or the appendix mesentery. Macroscopically, the tumor was a well-circumscribed mass located at the distal end of the appendix with firm consistency and yellow texture, suggestive for carcinoid tumor (Figure 1).



Figure 1 – Macroscopic aspect of surgical specimen with distal appendix tumor measuring 19/10 mm.

No infiltration in the mesoappendix was identified

and no other regional lymphadenopathies. Postoperative recovery was uneventful. The medical oncology evaluation recommended follow-up. Nine months after the hospital discharge, the patient is in good clinical condition, pending for the contrast-enhanced computed tomography (CT) of the abdomen and pelvis.

The histopathological examination was performed on paraffin-embedded tissue samples. Microscopically, the tumor exhibited an unexpected morphology. The clear cells appearance dominated the global aspect on low-power view at Hematoxylin–Eosin (HE) staining (Figure 2A).

Solid nests separated by smooth muscle fibers were spread into the entire muscular wall of the appendix (Figure 2B), with invasion into the serosa (Figure 2C). Integrity of the mucosa was fully preserved. On higher-power view, 100% of cells showed distinct boundaries, foamy microvesicular clear cytoplasm and centered nuclei with coarse chromatin (Figure 2D). There was no area of necrosis, any signet-ring cells or extravasated mucin. Sections from the base of the appendix were examined and described normal morphology without any invasion by tumor cells (Figure 3).

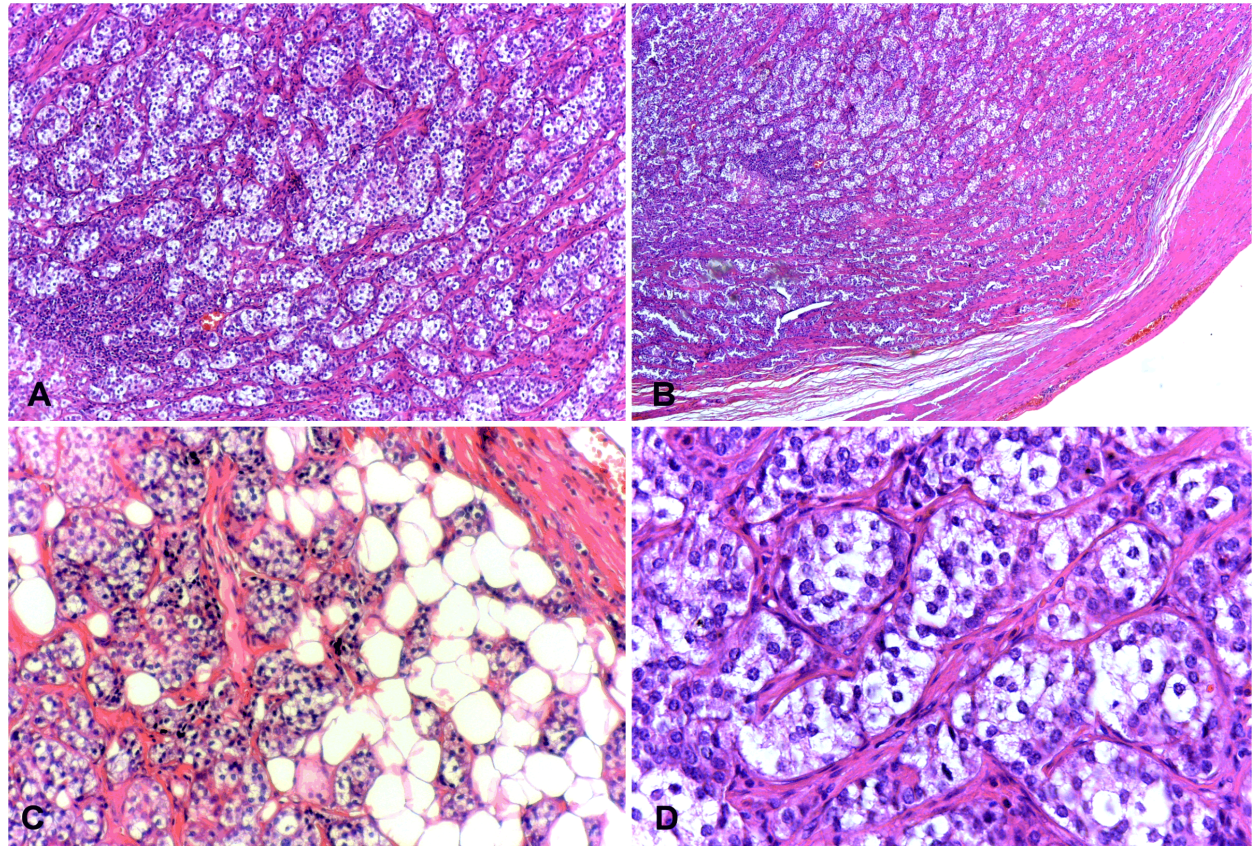


Figure 2 – Clear-cell carcinoid: (A) Nested architecture of clear cells separated by smooth muscle fibers septa; (B) Progressive invasion of muscular fibers by clear cells is clearly seen. On the surface of the muscular wall, capillaries can be spotted. The two areas seem separated due to artifact fixation; (C) Invasion into the serosa, where tumor cells are spotted between adipocytes. Also, smooth muscle fibers from the muscular wall are seen; (D) Details on clear cells. Foamy microvesicular cytoplasm and centered nuclei with coarse chromatin, as classical features of clear-cell carcinoid. Also, inside the nests there is a second type of cells with acidophilic cytoplasm, but in a smaller amount. Distinct boundaries between the cells are seen. HE staining: (A) $\times 100$; (B) $\times 40$; (C) $\times 200$; (D) $\times 400$.

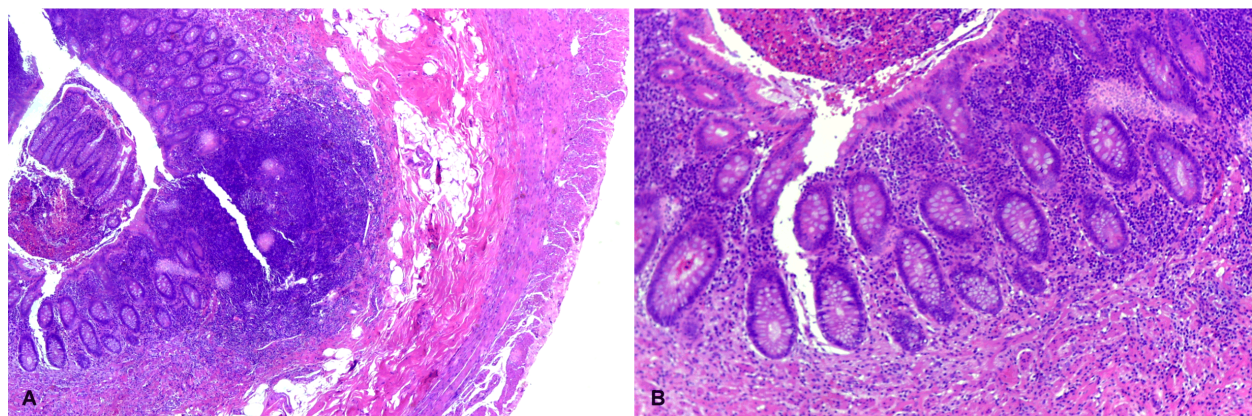


Figure 3 – Normal region of the appendix: (A) Note the characteristic lymphoid nodule and the surrounding specific crypts within the mucosa; (B) Note the single-layered surface epithelium and the underneath crypts with plenty of lymphocyte infiltrate between them. HE staining: (A) $\times 40$; (B) $\times 100$.

Due to unexpected morphology of the presumed diagnosis of carcinoid at the initial macroscopic view, immunohistochemistry was performed in order to prove the origin of the tumor cells and to exclude possibilities like metastases from a clear-cell carcinoma, such as renal cell or ovarian cancer. All tumor cells showed strong positivity to CD56 immunomarker and strong positivity to endocrine markers, such as chromogranin A and

synaptophysin. CDX2 positivity confirmed the digestive origin of the cells (Figure 4). The immunohistochemical staining also shows that tumor cells do not express cytokeratins CK7, CK20 or vimentin and therefore, carcinoma and mesenchymal tumor were excluded (Figure 5, A–C). Ki67 proliferation marker was 2% (Figure 5D). Thus, the final diagnosis was considered as well-differentiated NET, with clear-cell carcinoid subtype.

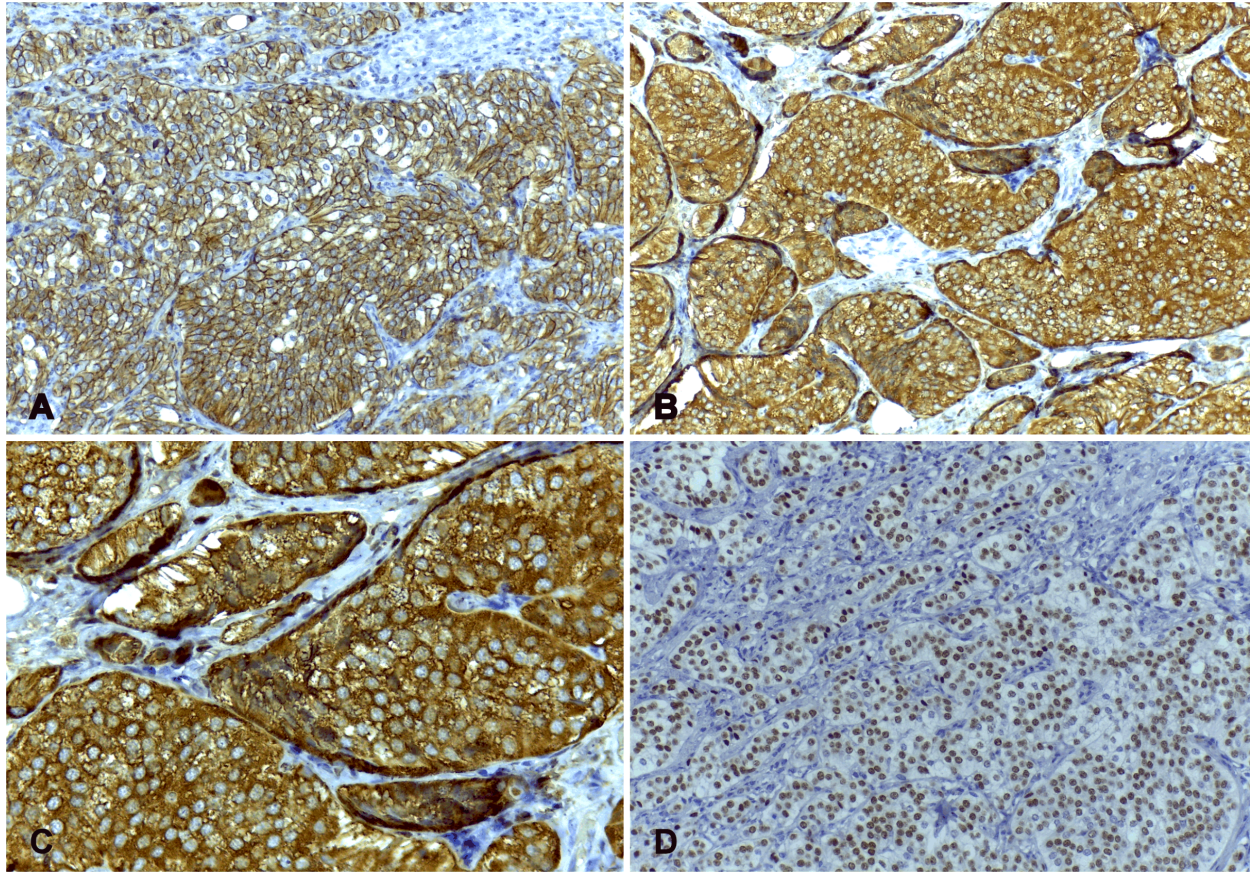


Figure 4 – Clear-cell carcinoid – immunostaining: (A) The entire population of clear-cells is positive to CD56 membrane immunomarker (Anti-CD56 antibody immunomarking, $\times 200$); (B) 100% of the clear cells show positive cytoplasm staining for chromogranin A, an immunomarker for carcinoid identification (Anti-chromogranin A antibody immunomarking, $\times 200$); (C) 100% of the clear cells within nests show cytoplasm positivity for synaptophysin, a protein marker of neuroendocrine cells. Cells from muscular septa are negative (blue staining) (Anti-synaptophysin antibody immunomarking, $\times 400$); (D) Clear cells arranged in round nests show 100% nuclear positivity for CDX2, a classic immunomarker for digestive origin, in comparison with cells in the surrounding muscular septa which are negative (Anti-CDX2 antibody immunomarking, $\times 200$).

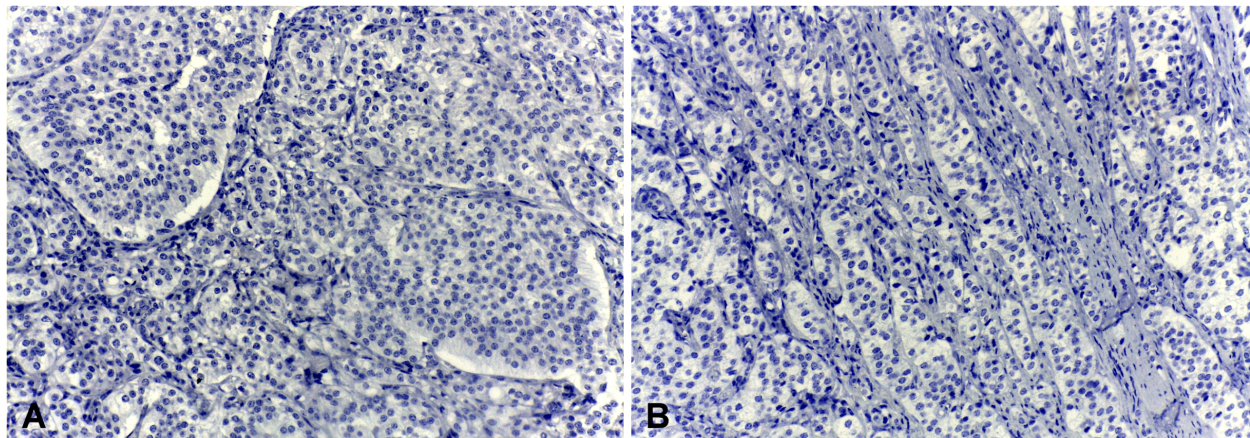


Figure 5 – Clear-cell carcinoid – immunostaining: (A) All cells including stromal cells and clear-cells are negative to CK7 immunomarker (Anti-CK7 antibody immunomarking, $\times 200$); (B) All cells show negativity to CK20 immunomarker (Anti-CK20 antibody immunomarking, $\times 200$).

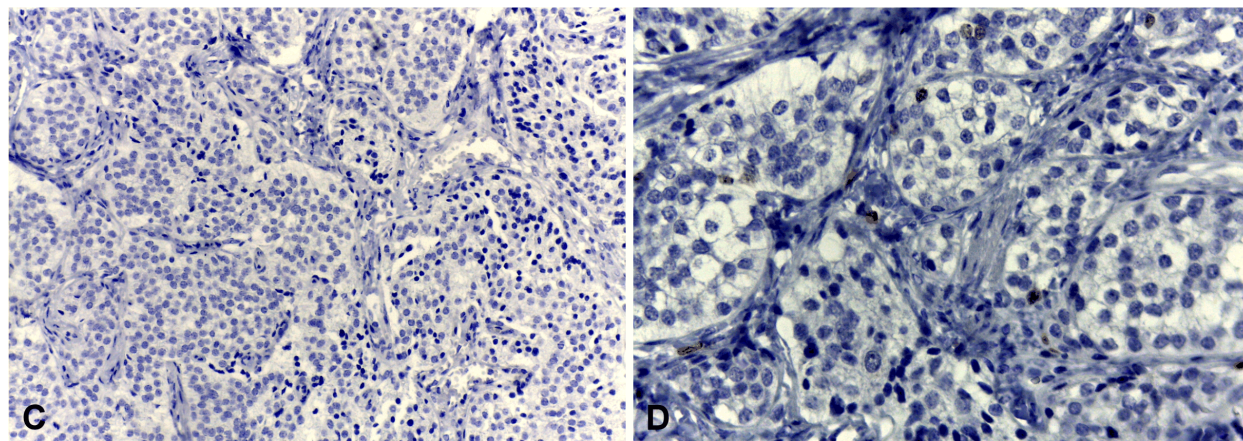


Figure 5 (continued) – Clear-cell carcinoid – immunostaining: (C) Both clear and stromal cells were negative to vimentin, a cytoplasmic marker for mesenchymal cells (Anti-vimentin antibody immunomarking, $\times 200$); (D) 2% of tumor cells show nuclear positivity to Ki67. The rest of clear-cells are negative, also the nuclei of smooth muscle cells (Anti-Ki67 antibody immunomarking, $\times 400$).

Discussion

Well-differentiated appendicular NETs occur with greatest frequency in the fourth to fifth decade of life, females being affected more frequent than males [6]. However, a recent paper including seven cases of clear cell carcinoid of appendix showed that five patients were male [7]. If the tumor is located at the tip of the appendix, the diagnosis is made incidentally during appendectomy or laparoscopy/laparotomy for other indications; if the tumor is situated at the base of the appendix, it can lead to lumen obstruction and appendicitis. Clinical picture with carcinoid syndrome is very rare in patients with primary NETs of the appendix [8].

Although appendicular NETs are relatively uncommon, they are the most frequent tumors arising from the appendix, and in the majority of cases, they are asymptomatic and are founded accidentally with occasion of appendectomy. Regarding the management of appendicular NETs, size, lymph node involvement and pathology features are of paramount importance. The complete resection for 2 cm lesions the appendectomy is sufficient, while for bigger lesions or more extensive disease is required re-exploration and right hemicolectomy [9–11]. In our case, the tumor was 19 mm and the patient was further scheduled for follow-up. Awareness should be raised on the presence of this entity of tumors, as cure is achievable in the majority of cases.

The most common locations of carcinoid tumors are the gastrointestinal tract and the lungs. Although it usually has a benign behavior, it can rarely be associated with carcinoid syndrome when there are widespread metastases, usually to the liver and retroperitoneum [4]. In our case, no symptoms of carcinoid syndrome were present, but classical signs of appendicitis. As far as clear-cell type is concerned, there have been reported before in the literature different locations: stomach, appendix, gallbladder, bile ducts and pancreas [5]. Also, extra-intestinal sites of carcinoid with clear-cell component have been described: thyroid, thymus, adrenal medulla, lung [5].

Most of the cases of clear-cell carcinoid were reported in the context of von Hippel–Lindau (VHL) disease, as it is well known the association of this disease with increased risk of clear-cell renal carcinoma or pancreatic

NETs [12]. In case of VHL disease, hypotheses of possible metastatic renal cell carcinoma have been proposed, but further investigations on this type of carcinoid should be made [13]. Few reports of clear cell NET in non-VHL cases were presented, as our paper does [14, 15].

Microscopic characteristics of a classical carcinoid include foamy microvesicular cytoplasm, centered nucleus with coarse chromatin, classic nested or trabecular neuroendocrine pattern. About the process of this cytoplasmic clearing, there have been many controversial attributions: glycogen, lipids or mucin [16].

One study, in 2010, analyzed 13 NETs with clear-cells (five of them exclusively composed of clear cells) and reported their clinical and morphological features. The location of all tumors was the appendix, as in our case, and electron microscopy and special stains together with immunohistochemistry were used proving the lipid structure of this clear cytoplasm. Therefore, the term of “lipid-rich” has been proposed for these types of carcinoids [5]. However, few cases reported composition of the clear cytoplasm other than glycogen or lipids. Gaffey *et al.* used ultrastructural examination of a pulmonary carcinoid and it revealed electron dense secretory granules with no glycogen or lipid structure [17]. Ordóñez *et al.* reported about one gastric clear cell carcinoid that ultrastructural analysis found dilated endoplasmic reticulum the corresponding for clear vacuoles [18]. Another gastric clear carcinoid described by Luk *et al.* proved to be a reminiscent of a gastric xanthelasma [19]. By now, the lipid content is the most acceptable possibility of the clear-cell composition.

In the differential diagnosis process, different entities should be considered. Goblet cell carcinoid (GCC) is a dual endocrine and exocrine tumor with a poor prognosis, so important distinction from carcinoid should be made for therapeutic purposes. Usually, the differences are easily observed, based on recognition of goblet cells with signet-ring features in HE- and Periodic Acid–Schiff (PAS)-stained section [16]. Metastases from renal cell carcinoma or ovarian carcinoma should be revealed by immunohistochemistry: negativity to endocrine markers (chromogranin A and synaptophysin) and positivity to CK7, vimentin and CD10. Also, malignant features such as nuclear atypia should be present.

✉ Conclusions

Distinguishing between a clear-cell carcinoid and goblet-cell carcinoids or metastases of a clear-cell adenocarcinoma is extremely important, as prognosis and treatment differs between these types of tumors. Clear-cell carcinoid has a good prognosis, as it is a well-differentiated type, as well as other types of carcinoids. The mechanism of cytoplasm clearing is not well known, but currently, lipids accumulation is the most accepted theory. Usually associated with von Hippel–Lindau disease, clear-cell carcinoids are also found outside this condition, as presented in this case.

Conflict of interests

The authors declare that they have no conflict of interests.

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