CASE REPORT

RJME Romanian Journal of Morphology & Embryology http://www.rjme.ro/

Incidental Walthard cell nests in the intestine. Report of two cases and review of the literature

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Abstract

Walthard cell nest (WCN) is a rare incidental finding in the intestine including the vermiform appendix and the mesocolon. This article describes a case of WNCs in the vermiform appendix and the mesocolon. The location in the mesocolon is reported for the first time. A review of the seven published intestinal cases including our report is presented. The lesion is seen in children, adults, and both sexes. The number of present cell nests varied between one and three per case. The diagnosis rests on histopathological examination. A potential source for histopathological diagnostic error includes a NET. The coexistence of WCNs with an appendicular neuroendocrine tumor (NET) may complicate the diagnosis. WCNs show the immunohistochemical profile of urothelial differentiation. To establish a diagnosis of WCN the absence of significant atypia, significant mitotic activity, atypical mitoses, necrosis, desmoplastic stroma, and tissue invasion must be confirmed. The lesion is a benign proliferation and once the appendectomy or colectomy has been performed, no further treatment is required.

Keywords: Walthard cell nest, urothelial cell rest, vermiform appendix, mesocolon, incidental urothelial rest.

Introduction

Walthard cell nests (WCNs) also known as urothelial cell rests are islands of benign, small, cuboidal to polyhedral epithelial cells. They are incidental findings commonly encountered in the gynecological tract. These are most frequently seen in the subserosa of the fallopian tubes, mesosalpinx, mesovarium, and ovarian hilus. However, WCNs are comparatively infrequent in the male genitourinary tract including the epididymis, *tunica albuginea*, testis, and spermatic cord [1]. These cell nests are rare in the vermiform appendix and mesocolon. Thus, as far as we know, only five cases of this lesion have been previously described in the appendix [2–6] and none in the mesocolon. WCNs have been reported in children and adults.

Awareness of this lesion is relevant as it may simulate a neuroendocrine tumor (NET), metastasis, peritoneal implants from a urothelial cell carcinoma of the urinary or gynecological tract, or even miliary tuberculosis in laparoscopy.

Aim

Herein, we describe three incidental WCNs in the subserosa of a case of appendectomy for appendicitis and two WCNs in the mesocolon. The rarity and the potential for misdiagnosis of the lesion justify the report of two new cases. In addition, a revision of the literature on the subject is included.

Case presentations

Case No. 1

A 49-year-old woman, with no significant past medical history, presented to the Emergency Department with a feeling of abdominal distension and moderate epigastric pain for approximately 16 hours. The pain subsequently progressed to the right iliac fossa and hypogastrium, where it persisted with progressively increasing intensity. No urinary or bowel rhythm changes were observed. An abdominopelvic echography showed a vermiform appendix measuring 5 cm in length and 1 cm in diameter with increased echogenicity and erasure of surrounding fat, in keeping with acute appendicitis. A laparoscopic appendectomy with an attached mesoappendix was performed.

Histopathological findings

A macroscopic examination of the appendix showed that the serosa was dulled with fibrinopurulent plaques. The entire appendix and mesoappendix were fixed with 10% neutral buffered formalin. Representative tissue samples were embedded in paraffin. For routine microscopy, 5-µm-thick sections were stained with Hematoxylin–Eosin (HE). Immunohistochemical (IHC) staining was performed using the EnVision FLEX+ Visualization System (Dako, Agilent Technologies, SL, Las Rozas, Madrid, Spain). The IHC reaction was performed using appropriate tissue control for the antibodies utilized. Automatic staining was accomplished

on a Dako Omnis stainer (Agilent Technologies, SL). Antibodies used are detailed in Table 1.

Table 1 – ***Immunohistochemical antibodies used in this study***

Antibody	Source	Clone	Dilution	Retrieval solution pH (Dako)
CD68	Dako	PG-M1	FLEX RTU	High
CK7	Dako	OUVTL12/30	FLEX RTU	High
Synaptophysin	Dako	SY38	FLEX RTU	High
Chromogranin A	Dako	DAK-A3	1:100	High
Ki67	Dako	MIB1	FLEX RTU	Low
GATA3	Gennova Scientific	150-823	1:25	High
p63	Dako	DAK-p63	FLEX RTU	High

CD68: Cluster of differentiation 68; CK7: Cytokeratin 7; Dako (Agilent Technologies, SL, Las Rozas, Madrid, Spain); GATA3: GATA binding protein 3; Gennova Scientific, SL, San José de la Rinconada, Sevilla, Spain; RTU: Ready-to-use.

Histological appendiceal examination revealed ulceration of the mucosa, acute transmural phlegmonous inflammation with serositis, distal fibrofatty obliteration of the lumen, and three well-defined subserous nests of small cells close to the vicinity tip of the appendix (Figure 1). Each nest consisted of closely packed small uniform cells (Figure 2). These were cuboidal to polyhedral with moderate clear to eosinophilic cytoplasm. A nest showed early peripheral liquefaction with initial cavitation (Figure 3). The nuclei were round to oval with irregular borders and small nucleoli (Figure 4). Some elliptical nuclei showed longitudinal grooves producing a "coffee bean" pattern (Figure 5). No atypia, mitoses, *stratum granulosum*, keratinization, genuine prickles, ciliated cells, calcification, or mucinous changes were observed. Immunohistochemically, the nests showed nuclear positivity for GATA binding protein 3 (GATA3) (Figure 6) and negativity for chromogranin A, synaptophysin, cluster of differentiation (CD)68, and cytokeratin (CK)7. The proliferation fraction on immunolabelling for Ki-67 was less than 1%. The exhaustion of the paraffin block did not allow staining for p63.

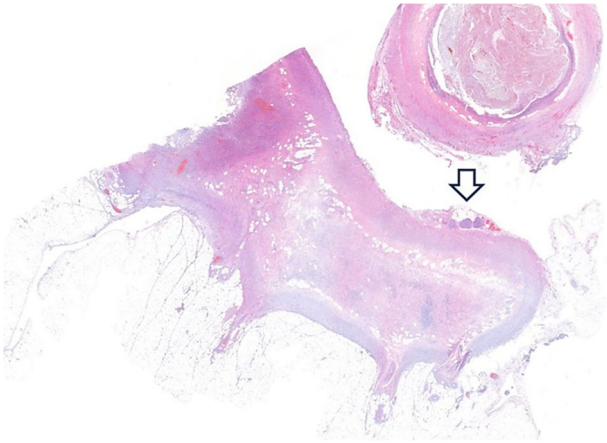

Figure 1 – ***Acute transmural inflammation and distal fibrofatty obliteration of the vermiform appendix. Three well-delimited nests of small cells are close to the vicinity tip of the appendix (arrow). Hematoxylin–Eosin (HE) staining, ob. ×0.8.***

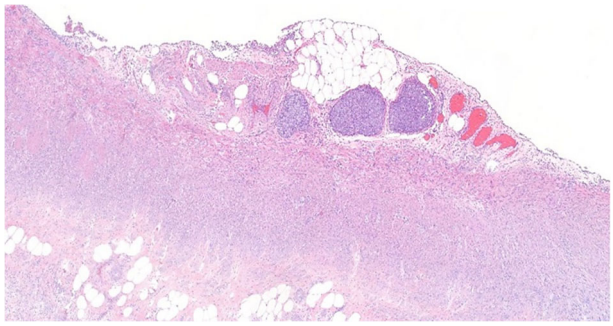

Figure 2 – ***The subserous nests consisted of closely packed small uniform cells. HE staining, ob. ×4.0.***

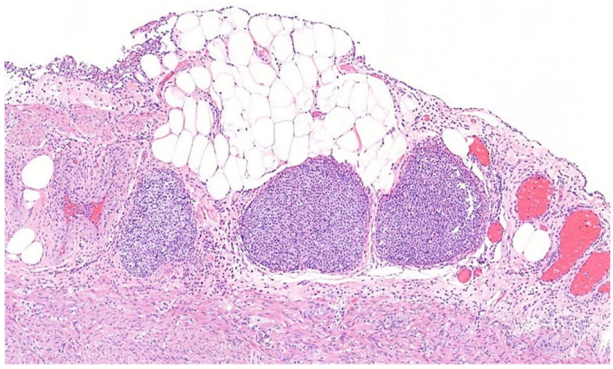

Figure 3 – ***The cell nests showed well-ordered cells. Early peripheral liquefaction with initial cavitation was observed in a nest. HE staining, ob. ×9.4.***

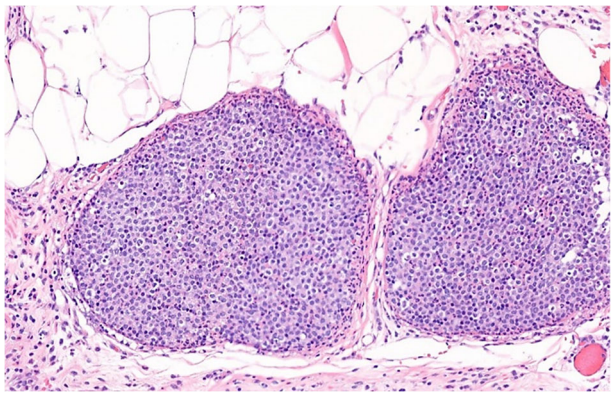

Figure 4 – ***The cells were cuboidal to polyhedral with moderate clear to eosinophilic cytoplasm. The nuclei were round to oval with fine chromatin and small nucleoli. HE staining, ob. ×22.4.***

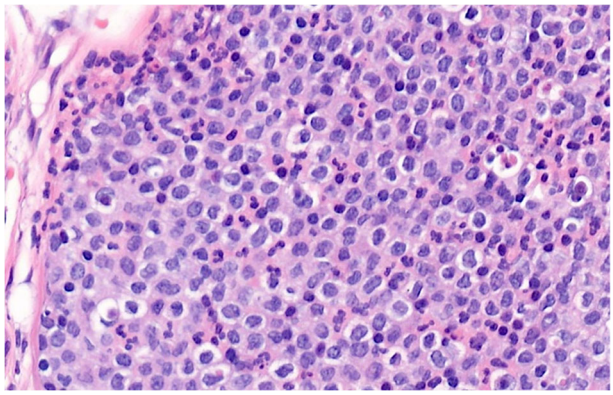

Figure 5 – ***Some nuclei presented longitudinal grooving producing a "coffee bean" pattern. HE staining, ob. ×44.9.***

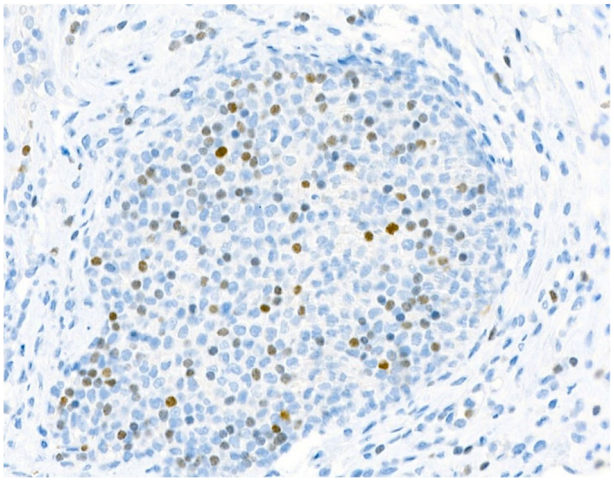

Figure 6 – ***Cells showed nuclear reactivity for GATA3, ob. ×32.5. GATA3: GATA binding protein 3.***

Case No. 2

A 79-year-old man with a history of multiple myeloma with chronic renal failure was found incidentally to have an ulcerated cecal mass during the follow-up of the myeloma. The patient underwent a right hemicolectomy.

Histopathological findings

Pathology revealed a moderately differentiated infiltrating tubular adenocarcinoma invading the bowel wall stopping at the subserosa. The neoplasm had no vascular permeation or neural infiltration and spared the 16 mesocolic lymph nodes removed [pT3, pN0, *American Joint Committee on Cancer* (AJCC) stage IIA]. The surgical margins were free of neoplasia. The appendix showed a mucinous neoplasia of low grade.

Tissue samples were embedded in paraffin. For routine microscopy, 5-µm-thick sections were stained with HE. IHC staining was performed using the methods described in Case No. 1. Antibodies used are specified in Table 1.

In the sections of the mesocolon, just beneath the serosa, two well-delimited solid nests were incidentally observed (Figure 7). They comprised a collection of small densely packed cuboidal to polygonal cells (Figure 8) with dark-stained oval nuclei with irregular borders, and small nucleoli. Some oval nuclei showed longitudinal grooving. Occasionally, voluminous nuclei were observed. Cytoplasms were scanty and eosinophilic (Figure 9). No mitotic activity was present. IHC staining revealed positivity for CK7, GATA3, and p63 in the cell nests (Figure 10, A–C).

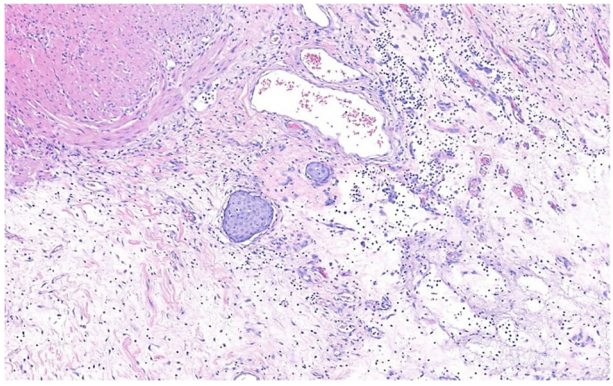

Figure 7 – ***Two well-delimited solid nests were incidentally observed in the subserosa of the mesocolon. Edema and inflammatory cell infiltration were present. HE staining, ob. ×9.7.***

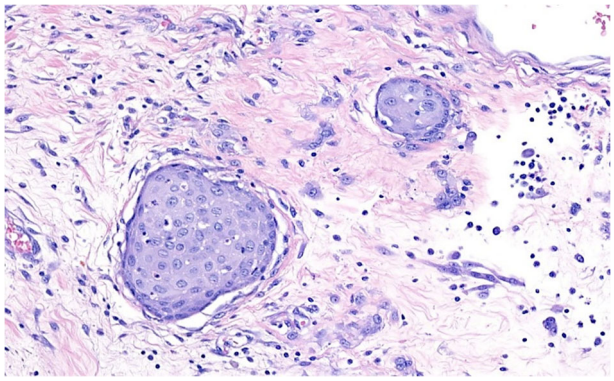

Figure 8 – ***The nests embraced a collection of small densely packed cuboidal to polygonal cells. Occasional nuclei showed an increase in volume. HE staining, ob. ×29.8.***

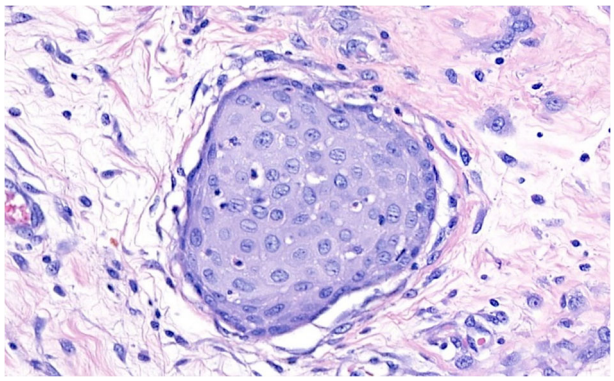

Figure 9 – ***Some oval nuclei showed longitudinal grooving. Cytoplasms were scanty and eosinophilic. HE staining, ob. ×49.6.***

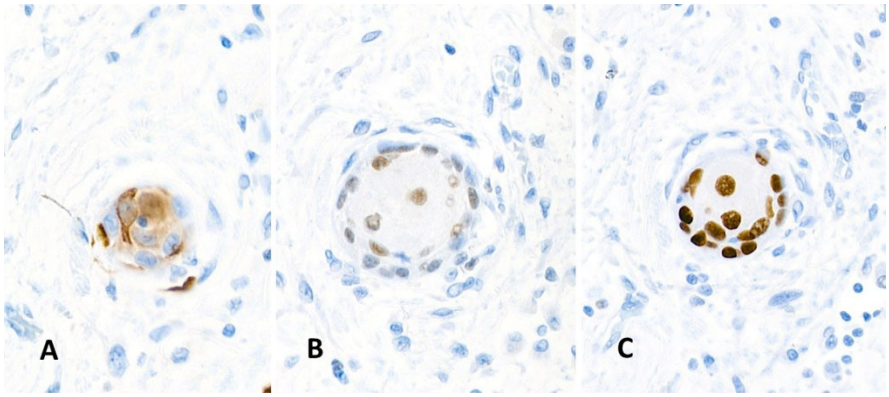

Figure 10 – ***Positivity of the cell nests for CK7 (A), GATA3 (B), and p63 (C). (A) ob. ×41.1, (B) ob. ×40.5, (C) ob. ×41.1. CK7: Cytokeratin 7.***

Discussions

WCNs are benign epithelial nests most often found in the female gynecological tract. Thus, Hunt & Lynn [7] observed these nests in the fallopian tube subserosa in 5.2% of specimens, in a study of 287 fallopian tube specimens. The lesion is uncommon in intratesticular and paratesticular structures [8, 9].

Incidental WCNs in cases of acute appendicitis are extremely rare. Gorter *et al.* [2] reported a case of WCNs in 241 cases of simple appendicitis (frequency 0.4%). WCNs have not been published previously in the mesocolon. They are reported here for the first time in that location. A review of the seven intestinal published cases, including our report, is presented in Table 2.

Table 2 – ***Walthard cell nests of the intestine. Cases reported***

Case #	Reference	Age [years]	Sex	No. of nests	Association	Location
1.	[2]	NR Pediatric patient	NR	NR	No	Vermiform appendix
2.	[3]	28	F	NR	No	Vermiform appendix
3.	[4]	NR Pediatric patient	M	1	No	Vermiform appendix
4.	[5]	33	M	"A few"	Neuroendocrine tumor	Vermiform appendix
5.	[6]	10	F	1	No	Vermiform appendix
6.	Present report, Case No. 1	49	F	3	No	Vermiform appendix
7.	Present report, Case No. 2	78	M	2	Multiple myeloma Cecal adenocarcinoma Low-grade mucinous appendiceal neoplasia	Mesocolon

F: Female; M: Male; NR: Not reported.

The lesion is seen in children, adults, and both sexes. The number of present cell nests varied between one and three per case although a report described "a few" [5]. WCNs may present as solid nests, multilayered surface plaques, or cysts with eosinophilic luminal secretion [1]. In our Case No. 1, a solid cell nest showed the beginning of cystic transformation. In the laparoscopy, they may appear as small greyish nodules measuring up to 2 mm in size. In these cases, the differential diagnosis includes miliary tuberculosis and peritoneal implants or metastases from malignancies of the gastrointestinal (GI) tract or female genital tract. Microscopically, the WCNs can simulate a NET. However, negativity for chromogranin A and synaptophysin rule out this possibility. Curiously, both WCNs and a NET were found simultaneously in the appendix of a 33-year-old man [5]. We consider that both processes appeared coincidentally. CK7 may show negativity in the WNCs [10] as in our Case No. 1. CK7 is characteristically expressed in the bladder's urothelial (transitional) epithelium. Still, it is relatively nonspecific and of little diagnostic value except when used in a panel with a specific differential diagnosis [10]. On the other hand, GATA3 is a typical marker of WCNs [10]. Other valuable markers include CK5/6, CK MNF116, and p63 [5, 6, 9]. WNCs lack reactivity for paired box (PAX)8, CK20, and calretinin [9].

Michal [11] described five cases of adenomatoid epithelial structures located beneath the peritoneal surface of the vermiform appendix. The mature structures consisted of cystically dilated adenomatoid spaces lined by one or two layers of flattened cells without atypia or mitoses. In two cases, these structures showed focal squamous cell metaplasia. The epithelial structures were considered to be of peritoneal origin. They do not correspond to cystic WCNs.

Multiple theories have been proposed to explain the origin of WCNs, including Müllerian remnants [1, 4, 6], mesothelial metaplasia, squamous (epidermoid) metaplasia, and transitional (urothelial) metaplasia. However, WCNs lack staining for Müllerian tumor markers such as PAX8 and PAX2 [12]. Teoh [1] considered that WCNs arise from a metaplastic change in the serous cells. Initially, there is stratification of serous cells forming a minute plaque on the serosa. Then the plaque invaginates into the subserous tissue where it takes the shape of a nest, or a cyst detached from the surface. The metaplastic epithelium is not truly squamous (epidermoid). The cells do not show keratinization, keratohyalin granules, or genuine prickles [13–15]. On the other hand, cell nests in the subserosa showing the panel CK7+, GATA3+, P63+, PAX8-, CK20-, Wilms tumor 1 protein (WT1)-, and calretinin- [5] suggest metaplastic transitional (urothelial) differentiation. This phenotype is consistent with Willis's statement that the adult peritoneum has wider metaplastic potential than the pericardium or pleura [16]. This type of metaplasia would be induced by peritoneal irritation caused by inflammation, previous surgery, peritoneal dialysis, or other processes. This implies that the progenitors for this process are the subserous multipotent cells. Chang *et al.* [6] reported a case showing CD34 positivity around a cell nest. They interpreted this finding as an endothelial peripheral lining and suggested an intravascular location of the cell nest consistent with a vascular migratory origin. This could explain the presence of WCNs in diverse organs [6].

The lesion is benign and does not require further treatment. However, to confirm a diagnosis of WCN, the absence of significant atypia, atypical mitoses, necrosis, desmoplastic stroma, and tissue invasion must be established.

Conclusions

WCN of the intestine including the vermiform appendix or the mesocolon is a rare incidental lesion that can be observed in all age groups and both sexes. One to three WCNs per case were found. WCNs can simulate miliary tuberculosis and peritoneal implants from malignancies of the GI tract or female genital tract. Microscopically, the main differential diagnosis of WCN includes a NET. The coincidence of WCNs with an appendicular NET may

complicate the diagnosis. Accurate diagnosis of WCNs is basic for appropriate management. They show no significant atypia, atypical mitoses, necrosis, or tissue invasion. This lesion presents the IHC profile of urothelial differentiation. WCNs are benign proliferations. No further treatment is required after appendectomy or colectomy.

Conflict of interests

The authors declare no conflicts of interests.

Compliance with ethical standards

No Ethics Committee approval is required in our institution for a case report involving a single patient.

Patient consent statement

Written informed consent was obtained from the patients to publish this case report and all accompanying images. A copy of the written consent is available for review by the Editor-in-Chief of this Journal.

Funding

This study was not funded externally.

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Received: December 7, 2024

Accepted: January 29, 2025