

Ovarian Clear Cell Carcinoma in Cowden Syndrome

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ABSTRACT

Cowden syndrome (CS) is an autosomal dominant mendelian disease related to germline pathogenic variants affecting the *PTEN* gene. CS is characterized by macrocephaly, mucocutaneous lesions, and an increased risk of breast and thyroid cancers. Rare ovarian cancer cases (mostly embryonic tumors) associated with *PTEN* have been described in the literature, but no current CS guidelines are available for ovarian cancer risk management. We report on a woman diagnosed with ovarian clear cell carcinoma (OCCC) at 28 years of age. The patient displayed macrocephaly, trichilemmomas, oral papillomatosis, and acral keratosis. A family history of multiple cancer cases within the *PTEN*-related tumor spectrum was identified. In addition, PET scan and fine-needle biopsy results led to a diagnosis of thyroid follicular neoplasia. *PTEN* sequencing revealed that she carried a germline inherited pathogenic variant in exon 5 c.388C>T, p.(Arg130*) (NM_000314). Somatic mismatch repair immunohistochemistry analysis showed normal expression, and germline *BRCA1/2* sequencing did not reveal pathogenic or likely pathogenic variants. An ovarian cell immunohistochemistry analysis reported total loss of *PTEN* expression, which strongly suggested the role of *PTEN* in the oncogenesis of this cancer. Hence, a total thyroid resection was performed instead of thyroid lobectomy and a risk-reducing bilateral mastectomy was discussed. Co-occurrence of this pathogenic germline mutation in *PTEN* in this patient, early development of OCCC at age 28 years, and total loss of *PTEN* expression in the tumor might support the involvement of *PTEN* in the carcinogenesis of her ovarian cancer. We describe a new ovarian cancer case with an atypical histologic type—clear cell carcinoma—in CS. This observation might be the first indication of the need to expand the *PTEN*-related tumor spectrum to incorporate OCCC. The CS diagnosis significantly changed the therapeutic outcome of this patient.

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Case Summary

This report presents the case of a 28-year-old woman diagnosed with a left adnexal mass in the context of pelvic pain that appeared 2 months earlier. First ultrasonography revealed a 4-cm pelvic mass suspected to be a uterine fibroid. Pelvic MRI showed a 10-cm left adnexal mass with a polylobed aspect, associated with a cystic portion that had a mucinous borderline epithelial part. The patient's CA 125 level was 237 UI/mL and carcinoembryonic antigen level was 37.6 ng/mL.

The patient first underwent laparoscopic left adnexectomy and left iliac adenopathy resection surgery. Histologic analysis revealed a 15-cm ovarian clear cell carcinoma (OCCC) with ruptured capsule associated with a 7-cm satellite nodule inside the mesosalpinx. Analysis of 5 iliac lymph nodes was normal. A multidisciplinary discussion resulted in a contralateral adnexectomy with hysterectomy, pelvic and para-aortic node dissection, appendectomy, omentectomy, and peritoneal biopsy decision. Histologic analysis confirmed that it was an OCCC stage IIB based on the FIGO 2014 classification.¹ After surgery, 6 cycles of carboplatin and paclitaxel were started. Because of her young age at diagnosis, the patient was referred to an oncogeneticist. Germline *BRCA1/2*-targeted sequencing was first performed² and showed no pathogenic or likely pathogenic variant. The ovarian tumor was microsatellite stable and mismatch repair protein immunostaining showed normal expression of *MLH1*, *MSH2*, *MSH6*, and *PMS2*.

The patient had no noteworthy personal medical history. Her mother underwent total thyroid resection and died at age 33 years of a metastatic left breast cancer (invasive ductal carcinoma). Sister III.2 (Figure 1) had abdominal subcutaneous nodular lesions (probably lipoma) and was diagnosed with an endometrial adenocarcinoma at age 28 years, and also underwent total thyroidectomy because of thyroid nodules with microcalcifications. Sister II.2 had a learning disability, sister II.5 had left lobe thyroid cysts, and 2 sisters (III.2 and III.5) and 1 niece (IV.1) had macrocephaly. Clinical examination of the patient highlighted macrocephaly (63 cm; +5 SD), ear and back

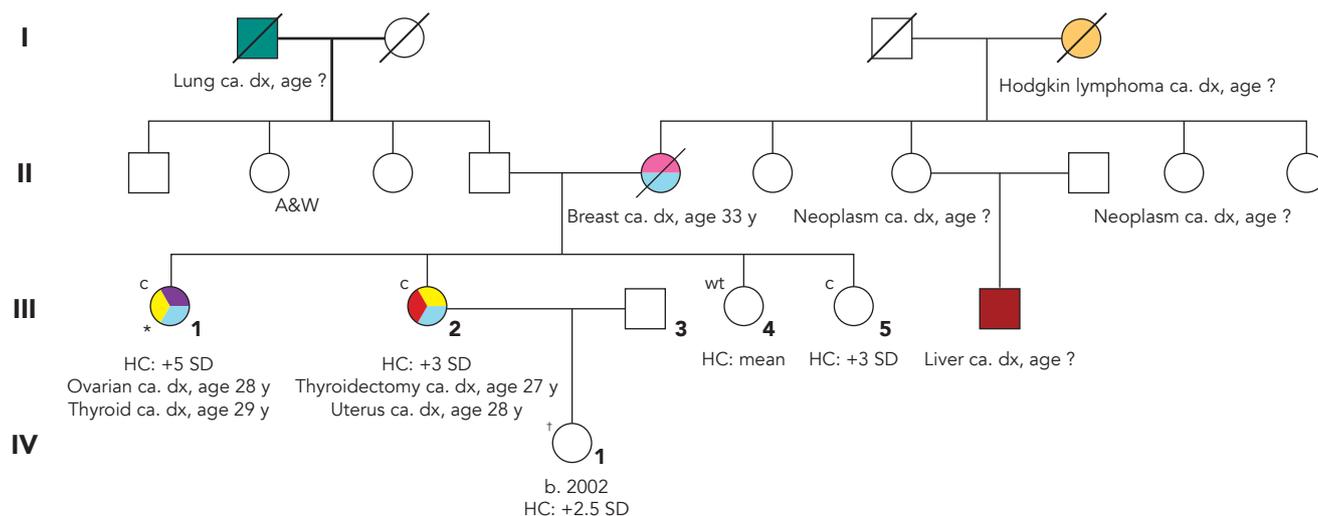


Figure 1. Family pedigree diagram (light blue, symptomatic PTHS; orange, suspected Hodgkin lymphoma; pink, breast cancer; red, uterine cancer; purple, ovarian cancer; yellow, thyroid cancer; turquoise, lung cancer; carmine red, suspected liver cancer).

Abbreviations: A&W, alive and well; b, born; c, *PTEN* variant carrier status; ca. dx, cancer diagnosis; HC, head circumference; PTHS, *PTEN* hamartoma tumor syndrome; SD, standard deviation; wt, *PTEN* wild-type.

*Patient reported in present article.

†Untested person.

trichilemmomas, oral papillomatosis, and acral keratosis (Figure 2). Further exploration of the family medical history revealed that a pathogenic *PTEN* variant may have already been identified in her maternal family, but ascertained that genetic data were not available. The association of specific cutaneous lesions, macrocephaly, and family history led to *PTEN* sequencing. This analysis revealed that the patient carried a germline pathogenic variant in exon 5 c.388C>T, p.(Arg130*) (NM_000314) responsible for Cowden syndrome (CS). Cascade family testing and ex post facto retrieval of genetic family data confirmed that this *PTEN* variant was maternally inherited. Ovarian cell immunohistochemistry analysis reported a total loss of *PTEN* expression, which supported *PTEN* involvement in this cancer (Figure 3).

Biopsy of a subcutaneous nodule revealed an epidermal cyst. Eight months after extensive second surgery, no evolutive tumor was found on a follow-up CT scan of the chest, abdomen, and pelvis. However, PET scan showed increased metabolic activity in the L3 vertebra and the right thyroid lobe, although MRI of the spine did not show any suspicious lesion. Thyroid ultrasound revealed a right lobe nodule, category 4B based on the Thyroid Imaging Reporting and Data System (TI-RADS). Fine-needle nodule puncture resulted in a diagnosis of follicular thyroid neoplasia (classified as Bethesda category IV). A total thyroid resection was recommended instead of thyroid lobectomy because of the *PTEN* mutation. In addition, a microvesicular adenoma and 2 mi-

cronodules with polymorphic vesicles were identified on the whole resected specimen.

After thyroidectomy, levothyroxine substitution therapy and local estrogenic treatment were started. Follow-up to monitor for cancer risk was explained to the patient and then implemented based on the CS guidelines published by the French Cowden Disease Network³ and NCCN.⁴ Although annual mammography and breast MRI screening was a medically indicated option, the patient considered undergoing a prophylactic bilateral mastectomy a few months after diagnosis. This option was validated by a cancer multidisciplinary discussion group, especially considering the CS diagnosis and her family history. At the time of writing, bilateral mastectomy has not been performed yet. Meanwhile, annual breast MRI and mammography were performed. Colonoscopy every 3 to 5 years, renal ultrasonography every 2 years, and annual dermatologic examinations were implemented. Three years after the OCCC diagnosis, the patient was healthy and no additional tumor events had occurred.

Discussion

The *PTEN* gene (phosphatase and tensin homolog on chromosome TEN, chromosomal position 10q22) codes for the *PTEN* protein, which is involved as a negative regulator of the PI3K/AKT/mTOR pathway. Therefore, *PTEN* is a tumor suppressor gene. CS is an autosomal dominant mendelian disease that belongs to the group of *PTEN* hamartoma tumor syndrome (PTHS), caused by germline *PTEN* mutations. The

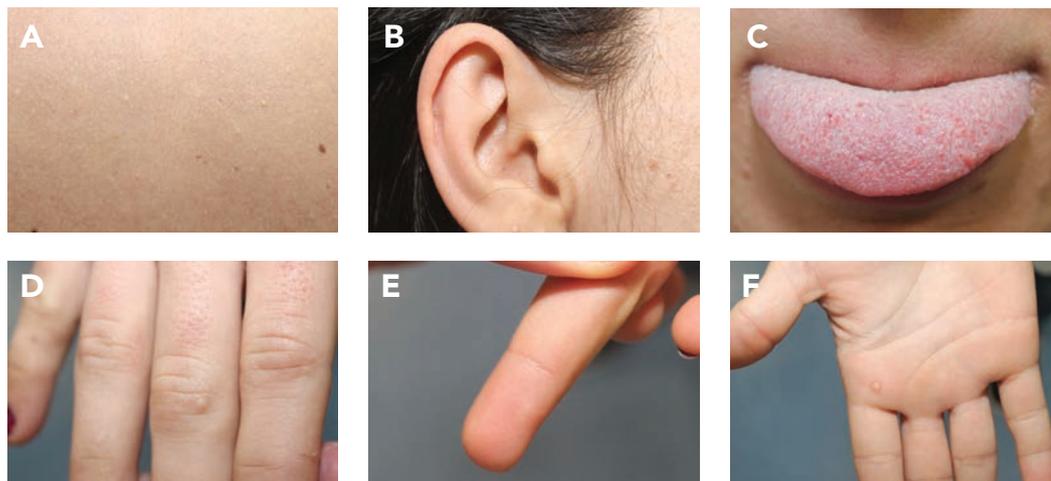


Figure 2. Typical cutaneous lesions of Cowden syndrome: (A, B) back and ear trichilemmomas, (C) oral papillomatosis, and (D–F) acral keratosis and multiple wart-like lesions.

clinical characteristic features of CS are macrocephaly; mucocutaneous lesions such as trichilemmomas, papillomatous papules, or acral keratosis; and an increased risk for benign and malignant tumors. Estimated lifetime risks for cancer are 85% for breast (95% CI, 71.4%–99.1%), 35% for thyroid (95% CI, 19.7%–50.7%), 34% for kidney (95% CI, 10.4%–56.9%), 28% for endometrium (95% CI, 17.1%–39.3%), and 9% for colon (95% CI, 3.8%–14.1%).^{3,5,6} Other CS-associated features include Lhermitte-Duclos disease; hamartomatous polyposis; and noncancer manifestations, such as autism and benign thyroid lesions (eg, multinodular goiter, fibrocystic mammary lesions, immune-related disorders).⁷ Complete diagnostic criteria for PHTS are described in the NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines) for Genetic/Familial High-Risk Assessment: Breast and Ovarian.⁴ At the pretesting cancer genetics consultation, the patient presented only 2 major criteria: macrocephaly and multiple mucocutaneous lesions, including facial trichilemmomas and acral keratosis, because ovarian cancer is not included in the PHTS testing criteria and spectrum. Only after *PTEN* sequencing did the patient present a third major criterion: follicular thyroid neoplasia. She did not strictly fulfill the individual PHTS testing criteria. However, her family medical history and the *PTEN* deleterious mutation supposedly identified in her maternal family led to a targeted analysis of *PTEN* that confirmed her carrier status for a (p.Arg130*) deleterious variant in the *PTEN* gene.

Ovarian cancer is the fifth leading cause of cancer-related death in women.⁸ Among major histologic subtypes, OCCC, a distinct subtype of epithelial ovarian cancer, is the second most common ovarian carcinoma.^{9,10}

OCCC is rarely bilateral, and patients are usually diagnosed at an early stage.¹¹ Histologic features of OCCCs are characterized by solid, papillary, and tubulocystic patterns.¹² Most OCCCs are presumed to arise from endometriotic cysts,¹³ and key driver events involving *HN-FIB* overexpression and somatic *ARID1A* mutations have been identified.¹⁴

This study reports on the first case of OCCC associated with a pathogenic *PTEN* germline variant to our knowledge. Only few ovarian tumors have been associated with CS in the literature.¹⁵ A PubMed search using the terms “Cowden syndrome” or “PHTS” and “ovarian clear cell carcinoma” did not identify any other cases. Furthermore, among the 1,545 cases of primary ovarian cancer in The Cancer Genome Atlas (TCGA)¹⁶ database, a somatic mutation in *PTEN* was found in 10 cancer tissue samples and all were ovarian serous cystadenocarcinoma. In unpublished data from the French CS reference center (Institut Bergonié, Bordeaux, France), the histologic types of ovarian cancer described until now were mostly embryonal tumors. Indeed, 5 patients from unrelated families presented with ovarian cancers: 3 with germ cell tumors (vitelline tumor, choriocarcinoma, and teratoma) diagnosed at ages 4, 12, and 29 years, respectively; 1 with an ovarian serous cystadenocarcinoma diagnosed at age 24 years; and 1 displayed a heterogeneous tumor type (germ cells, sex cord tumor, or stromal tumor) diagnosed at age 2 years¹⁵ (Virginie Bubien, MD, PhD; written communication; January 2018).

Hereditary OCCCs are usually associated with Lynch syndrome^{17,18} or *BRCA1/2* germline mutations,¹⁹ but these hypotheses were explored and excluded. Somatic mutations of *PTEN* have been described in OCCC (5% somatic mutations),^{20,21} and loss of *PTEN* expression revealed by immunohistochemistry is frequently reported (up to

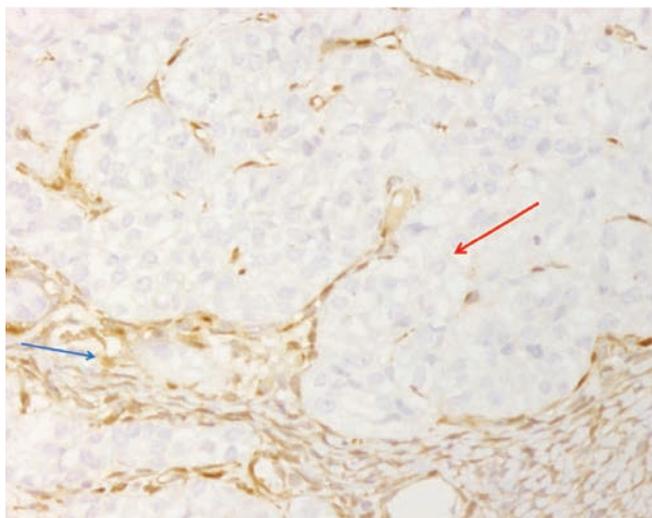


Figure 3. Ovarian cell PTEN immunohistochemistry (red arrow, cancerous cells without PTEN immunomarking; blue arrow, stromal cells with PTEN immunomarking). Immunohistochemical analysis was performed on a BenchMark ULTRA (Roche Diagnostics) using a PTEN primary antibody, clone 138G6 (Cell Signaling Technology), 1/100 dilution with a 40-minute incubation. Pretreatment was performed using CC1 standard for 64 minutes. Staining was performed using the ultraView Universal DAB Detection Kit (Roche Ventana).

37.5% of OCCC).²² The variant (p.Arg130*) that we report here is associated with ovarian *PTEN*-mutated cancers (regardless of their histology) in 6.2% of patients in the COSMIC database.²³ The cellular and molecular mechanisms linking a germline heterozygous mutation in *PTEN*, the somatic total loss of expression of *PTEN* in the tumor, and cancerogenesis are not fully understood. To our knowledge, no published sequencing data exist for *PTEN* germline exploration in OCCC. Despite available data about this patient's tumor, we were unable to confirm this assumption. However, this observation might be the first clue that a germline *PTEN* variant is possibly involved in the cancerogenesis of OCCC. This assumption is supported by co-occurrence of this pathogenic germline mutation in *PTEN*, early development OCCC at age 28 years, and total loss of *PTEN* expression in the tumor.

The cancer genetics management of this patient and subsequent molecular diagnosis of CS significantly changed her curative and preventive/risk-reducing care. Indeed, the CS diagnosis was of great clinical value in the decision to perform a total thyroid resection instead of a thyroid lobectomy after detection of a right lobe thyroid nodule, category 4B TI-RADS. In addition to her maternal family history and her mother's breast cancer–related early death, the information about the high risk of breast cancer in women with CS was also of great significance in the patient's choice to undergo a risk-reducing bilateral mastectomy. Furthermore, specifically in women with CS,

mild to severe fibrocystic mammary dysplasia frequently exacerbates radiologic intricacy of breast monitoring. Despite breast MRI accuracy, it often leads to iterative biopsies. As frequently observed in these patients, avoiding invasive and stressful repeated procedures was another key reason for the patient to consider the mastectomy option.

Based on the French³ and NCCN Guidelines⁴ for CS, the patient was immediately offered a comprehensive physical examination, including skin and thyroid clinical screening by specialists. Because the consideration of and medical approval process for a risk-reducing mastectomy took several months, and because the patient's mother died of breast cancer at age 33 years, a breast MRI and mammography were performed immediately. Notably, the French and NCCN follow-up recommendations are globally similar but slightly differ in some points, mostly regarding the age at which to begin cancer screening. The French guidelines recommend that screening for colon and renal cancer begin at age 30 years.³ Hence, although she was <35 years of age at diagnosis and no colon cancer occurred before age 40 years among her relatives, a colonoscopy was recommended immediately, as was renal ultrasound screening, although she was <40 years of age.

In both the French³ and NCCN guidelines⁴ for CS, there is no recommendation specifically dedicated to managing ovarian cancer risk. Indeed, CS is not associated with a high lifetime risk of ovarian cancer. Furthermore, there is no effective consensus screening method for ovarian cancer, neither in high-risk nor average-risk women. Risk-reducing ovarian surgical resection does not appear as a medically reasoned option.

Ovarian cancer is not included in CS testing criteria. However, this might be reassessed as genetic testing availability increases, especially considering that CS diagnosis in OCCC might lead to new personalized medicine options, such as molecularly targeted drugs. In vitro therapeutic studies on OCCC recently supported that the PI3K/mTOR inhibitor combination significantly and specifically decreased cell proliferation and focal adhesion in OCCC, and might be a promising treatment strategy for this cancer.^{24,25}

Offering family genetic testing to relatives of the proband and subsequently setting up personalized medical management is a critical component of cancer genetic counseling. Interestingly, the CS diagnosis in our patient's family supposedly had been identified previously, but the patient was not given clear and actionable information about it. This is consistent with the lack of referral for genetic counseling and testing reported in families with *BRCA1/2* and Lynch syndrome.²⁵ This emphasizes the need to reinforce health provider skills to improve efficient family communication about these genetic issues.

Conclusions

This case of a CS diagnosis in a woman with epithelial ovarian cancer (clear cell carcinoma subtype), which led to a significant change in care management, highlights the high medical relevance and clinical utility of the multigene sequencing panel strategy, including *PTEN* sequencing, for which many international scholarly organizations recently advocated.^{4,27} The increase in cancer gene panel analyses through next-generation sequencing will probably modify the knowledge about *PTEN* involvement in isolated ovarian cancer. It

may lead to the redefinition of the CS tumor spectrum and shed light on new therapeutic targets.

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