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CLINICAL REPORT

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Case report of adrenocortical carcinoma associated with double germline mutations in *MSH2* and *RET*

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ABSTRACT

Adrenocortical carcinoma (ACC) is a rare aggressive malignancy that originates in the outer layer of the adrenal gland. Most ACCs are sporadic, but a small percentage of cases are due to hereditary cancer syndromes such as Li-Fraumeni syndrome (LFS), Lynch syndrome (LS), and familial adenomatous polyposis (FAP). Multiple endocrine neoplasia type 2A (MEN2A) is an inherited disorder that predisposes to medullary thyroid cancer, pheochromocytoma, and parathyroid hyperplasia. We present here a case of ACC with both LS and MEN2A; the family and medical history were consistent with Lynch. This is, to our knowledge, the first report of a patient with ACC associated with germline mutations in *RET* and *MSH2*, and no phenotypical characteristics of MEN2A.

KEYWORDS
adrenocortical cancer, Lynch syndrome, multiple endocrine neoplasia type 2A

1 | INTRODUCTION

The association of adrenocortical carcinoma (ACC), Lynch syndrome (LS), and multiple endocrine neoplasia type 2A (MEN2A) in the same patient is a novel occurrence. The incidence of ACC alone is less than 0.7–1.5 per 1 million people per year (Kebebew et al., 2006). Most often, ACC occurs sporadically with 5% of cases due to hereditary cancer syndromes such as Li-Fraumeni syndrome (LFS), familial adenomatous polyposis (FAP), and LS (Else et al., 2013; Mazzuco et al., 2012). Despite poor overall survival statistics, recent studies have identified a few long-term survivors, suggesting a more heterogenous prognosis of ACC (Baudin, 2015). Known prognostic factors include stage at diagnosis, age, hormone-related symptoms, proliferative index rating (Ki67), Weiss score, and tumor differentiation (Mazzuco et al., 2012).

LS is a heritable cancer syndrome caused by heterozygous germline mutations in mismatch repair genes (*MSH2*, *EpCAM*, *MLH1*, *MSH6*, and *PMS2*). LS predisposes to colorectal, endometrial, ovarian, gastric, hepatobiliary, and urinary cancers along with tumors of the small bowel and central nervous system. Some genotype–phenotype correlations have been noted; for example, patients with *MSH2* mutations carry a higher risk of extracolonic cancers or multiple malignancies (Cohen & Leininger, 2014; Lynch et al., 2009).

MEN2A is caused by mutations in the *RET* proto-oncogene and is characterized by predisposition to medullary thyroid cancer (MTC), pheochromocytoma, and parathyroid hyperplasia. There are distinct genotype–phenotype correlation patterns associated with the location of the mutations which lead to different degrees of penetrance. The diagnosis of MEN2 is suspected in a patient with a germline RET mutation even in the absence of any clinical features (Kloos et al., 2009). The patient described here had the p.Val804Met (Figure 1; low to moderate risk for MTC) mutation in the *RET* gene.

2 | CASE REPORT

The patient is a G1P1 47-year-old female diagnosed with ACC at 44 years of age. She carried a deleterious germline mutation in the *MSH2* gene (c.211+1G>T splice variant; Figure 2) and had a family history of LS. Her clinical presentation and family history were more characteristic of LS at diagnosis; the patient did not have any manifestations of MEN2A. She was initially tested for the familial *MSH2* variant at 43 years of age, and upon confirmation of positive status, she underwent a preventive complete hysterectomy. Five years after the LS diagnosis, a 12-cm adrenal mass was found on CT scan. The tumor was further subclassified as an ACC with microscopic tumor invasion and lymphovascular extension; the patient was started on mitotane chemotherapy. She has since undergone numerous surgeries for recurrent ACC including right partial hepatectomy, omentectomy, partial nephrectomy with excision of tumor, and cytoreductive surgery with hyperthermic intraperitoneal chemotherapy (HIPEC).

She was referred to the National Institutes of Health (NIH) for participation on the Natural History and Biospecimen Acquisition for Children and Adults with Rare Solid Tumors (NCT03739827). Histopathologic examination from the initial surgery showed a high-grade, poorly differentiated ACC with Ki 67% (Figure 3). Clinical analysis of a somatic gene panel of 523 cancer genes (TruSight Oncology 500 (TSO500), Illumina) performed on the patient's tumor (estimated tumor content of 70%) revealed the presence of a pathogenic mutation in the *RET* gene (c.2410G>A; p.Val804Met) with a VAF of 90%. Due to the high allele frequency of this mutation and the implications of this finding for a possible hereditary cancer condition, confirmatory germline sequencing was performed on the patient's saliva (GeneDx, Bethesda, MD). The result confirmed that the *RET* p.Val804Met (c.2410G>T) was present in her germline (heterozygous). In addition, TSO 500 identified the *MSH2* c.211+1G>T splice site mutation with a VAF of 88% which is consistent with loss of heterozygosity (LOH). Microsatellite instability was indeterminate in this assay.

The family history was significant for LS on the paternal side (Figure 4). Her father (age of death was 52), paternal grandfather (unknown age of death), and two paternal uncles (unknown age of death) had been diagnosed with colon cancer. The patient's sister (39 years old) was diagnosed with colon cancer at 28 years of age; she was the initial proband found to have a deleterious germline mutation in the *MSH2* gene (c.211+1G>T, IVS1+1G>T). In addition, her paternal half-brother (60 years old), also found to have the familial MSH2 mutation, was diagnosed with renal (papillary grade II) cancer, bladder cancer, and primary urethra low-grade carcinoma. The family history was otherwise unremarkable for other cancers or conditions related to mutations in either *MSH2* or *RET*. The patient was counseled on the implications of the germline *RET* mutation and testing was offered to relatives. Results on her 11-year-old son (asymptomatic, disease free) were negative for both mutations, the patient's 52-year-old sister (asymptomatic, disease free), as well as all available relatives declined testing. Given the patient's ACC disease burden, a modified surveillance was recommended.

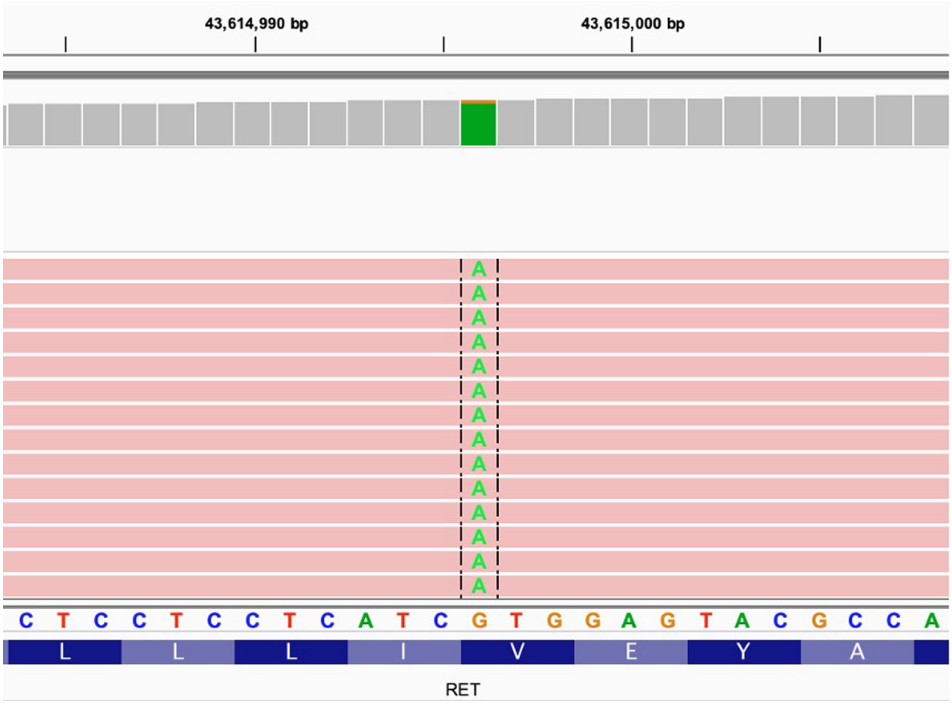

FIGURE 1 RET mutation identified in the proband (p.Val804Met) [Color figure can be viewed at wileyonlinelibrary.com]

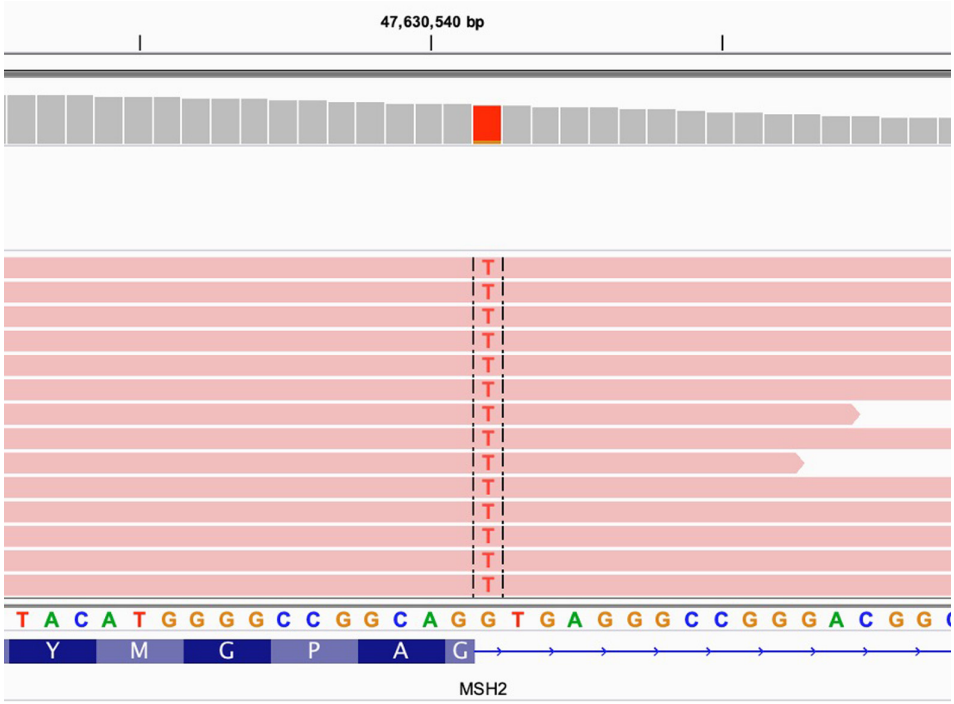

FIGURE 2 MSH2 mutation identified in the proband (c.211 +1G>T) [Color figure can be viewed at wileyonlinelibrary.com]

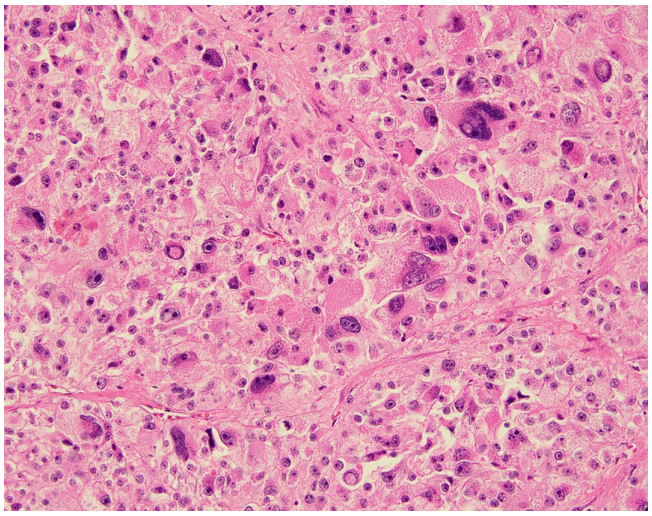

FIGURE 3 Adrenocortical carcinoma is composed of sheets of large epithelioid cells with pale to eosinophilic cytoplasm and nuclear pleomorphism (magnification 200×) [Color figure can be viewed at wileyonlinelibrary.com]

3 | DISCUSSION

We describe here the case of a 47-year-old female with metastatic ACC, and a deleterious germline mutation in two cancer predisposing genes (*RET* and *MSH2*); the patient was referred to the NIH without prior knowledge of her *RET* status.

The association of ACC and LS was initially described by Raymond et al., 2013, who reported that 3.2% of LS patients had ACC (Raymond et al., 2013). In 2016 Challis et al. reported the first case of familial ACC in association with an *MSH2* pathogenic mutation (Challis et al., 2016). These studies advocate for the inclusion of ACC in the clinical diagnostic criteria of LS and for increased surveillance in LS patients at risk for ACC; an argument fortified by the present case which showed biallelic inactivation of MSH2 in an ACC tumor of a patient with Lynch.

The family history of this patient was significant for colorectal cancer in four paternal relatives. In addition, the patient's sister was diagnosed with colorectal cancer at 28 years of age, and her paternal half-brother developed kidney and bladder cancer. Both siblings were carriers of the familial *MSH2* mutation. Genotype–phenotype correlations in the c.211+1G>T mutation have not been identified to date. In general, MSH2 mutations are associated with 48% risk of colorectal cancer, 21% risk of endometrial cancer, and 24% ovarian cancer; the risk of any cancer with *MSH2* mutations range from 14.3% by age of 40 to 80.4% by age of 75 (Pérez-Cabornero et al., 2013). A four-generation pedigree (Figure 4) did not reveal any additional cancers, which is consistent with the incomplete penetrance and variable expression seen in *MSH2* pathogenic variants.

Upon enrollment in the NIH protocol (NCT03739827), the patient's tumor was found to be positive for the pathogenic *RET* mutation (c.2410G>A, p.Val804Met) with an allele fraction (VAF) of 90% (tumor content 70%). The high VAF was likely due to LOH at the *RET* locus as has been reported previously by several studies looking at the somatic mutation profile of the *RET* gene in MTC (Dvorakova et al., 2006; Quadro et al., 2001). Germline testing was performed; the results confirmed the presence of this mutation in her germline (heterozygous); the patient was counseled about the clinical predispositions associated with this finding.

In the past two decades, since the discovery of the *RET* gene, substantial evidence has emerged delineating genotype–phenotype correlations of this gene (e.g., calcitonin level variations with different

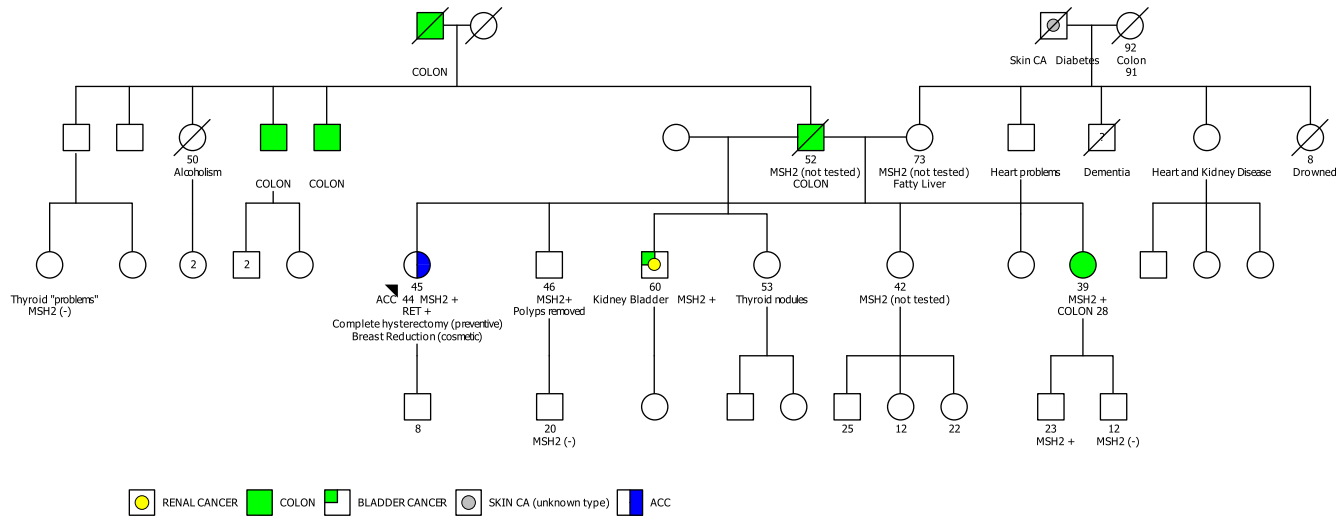

FIGURE 4 Four-generation family history pedigree [Color figure can be viewed at wileyonlinelibrary.com]

codons). The p.Val804Met mutation is one of the most common mutations in the *RET* gene; the American Thyroid Association (ATA) classifies this mutation as moderate risk in the guidelines for the management of MTC (Loveday et al., 2008; Møller et al., 2017; Romei et al., 2016). This variant has received significant attention because of its variable age-related penetrance and clinical heterogeneity (Feldman et al., 2000; Frohnauer & Decker, 2000; İmge Aydoğan et al., 2016; Learoyd et al., 2005; Rich et al., 2014; Wells et al., 2015). The manifestation of disease from carriers of the p.Val804Met mutation ranges from metastatic MTC at an early age to no evidence of disease by age 80 (Feldman et al., 2000; Wells et al., 2015) and includes several reports of pheochromocytoma (Høie et al., 2000; Nilsson et al., 1999), papillary thyroid carcinoma (Brauckhoff et al., 2002; Gibelin et al., 2004), and concomitant papillary and medullary carcinoma (Shifrin et al., 2009). The patient discussed here had no manifestation of *RET*-related conditions, as evidenced by normal levels of plasma metanephrines, calcitonin, and carcinoembryonic antigen (CEA) on evaluation. At the time of referral, she presented with metastatic lesions in the liver and retroperitoneal soft-tissue nodules. Therefore, the management of the complications associated with the presence of a germline mutation in the *RET* gene was modified to a less invasive protocol. Recommendations for her surveillance included yearly metanephrines and calcitonin labs with an annual thyroid ultrasound. The patient's 11-year-old son tested negative for both mutations. Given that no other family members agreed to germline genetic testing of the *RET* mutation, the possibility of a de novo mutation cannot be ruled out.

Tumor panel sequencing in cancer patients is now part of the standard of care in most clinical settings. Therefore, secondary germline findings from tumor profiling have become more common in recent years. Approximately 2.3%–12% adult cancer patients have actionable germline pathogenic variants detected from tumor panels (Mandelker & Zhang, 2018; Schrader et al., 2016; Seifert et al., 2016); similar findings have been reported in the pediatric population with numbers ranging from 8.5% to 10% (Meric-Bernstam et al., 2016). These findings are usually unexpected, given that the main focus of tumor profiling is identification of potential targets for therapy or further characterization of the tumor. However, germline pathogenic variants can be inferred from tumor-only testing (as opposed to tumor-normal testing) but must be confirmed with follow-up germline testing. The patient's clinical presentation and family history were consistent with LS only, and she did not meet criteria for MEN2A testing; thus, the possibility of a second hereditary cancer syndrome was extremely low. This case highlights the importance of performing confirmatory germline testing, even when the likelihood of a second underlying cancer predisposition syndrome is low.

Additionally, this case exemplifies the importance of including genetic counselors as part of a multidisciplinary team that manages patients undergoing tumor genomic testing. The confirmation of a secondary germline cancer-predisposing mutation generated significant anxiety in this patient, mostly due to concerns for her son. However, through discussion with a genetic counselor she understood the benefits of early testing in asymptomatic relatives, who can then undergo tailored cancer surveillance and choose from available preventative options.

4 | CONCLUSIONS

We report a rare case of the co-occurrence of two hereditary cancer syndromes, LS and MEN2A, in a metastatic ACC patient with a clinical presentation and family history suggestive only of LS. The presence of the second germline mutation was inferred from a tumor somatic panel and confirmed by germline testing; management recommendations were implemented for the patient, and additional testing was offered to relatives. The clinical consequences of these two

pathogenic germline mutations have not been reported before; at this time, it is too early to speculate on possible synergistic or modifier effects of the co-occurrence. Future studies will focus on the long-term outcomes of these family members.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

AUTHOR CONTRIBUTIONS

All authors participated in the conception and design of the study. All authors have given their approval of the final version of the manuscript and take responsibility for the manuscript's content and accuracy.

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