

Prevalence of Germline Mutations in Cancer Susceptibility Genes in Patients With Advanced Renal Cell Carcinoma

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IMPORTANCE Identification of patients with hereditary renal cell carcinoma (RCC) is important for cancer screening and, in patients with advanced disease, for guiding treatment. The prevalence of cancer-related germline mutations in patients with advanced RCC and the phenotypes associated with some rare mutations are unknown.

OBJECTIVES To examine the prevalence of germline mutations in both known RCC predisposition genes and other cancer-associated genes and to identify clinical and pathologic factors associated with germline mutations.

DESIGN, SETTING, AND PARTICIPANTS In this cohort study conducted from October 1, 2015, to July 31, 2017, 254 of 267 patients with advanced (American Joint Committee on Cancer stage III or IV) RCC who were seen in medical oncology or urology clinics agreed to germline sequencing and disclosure of results under an institutional protocol of matched tumor-germline DNA sequencing.

MAIN OUTCOMES AND MEASURES Mutation prevalence and spectrum in patients with advanced RCC were determined. Clinical characteristics were assessed by mutation status.

RESULTS Of the 254 patients (median age [range], 56 [13-79] years; 179 [70.5%] male; 211 [83.1%] non-Hispanic white), germline mutations were identified in 41 (16.1%); 14 (5.5%) had mutations in syndromic RCC-associated genes (7 in *FH*, 3 in *BAP1*, and 1 each in *VHL*, *MET*, *SDHA*, and *SDHB*). The most frequent mutations were *CHEK2* (n = 9) and *FH* (n = 7). Of genes not previously associated with RCC risk, *CHEK2* was overrepresented in patients compared with the general population, with an odds ratio of RCC of 3.0 (95% CI, 1.3-5.8; *P* = .003). Patients with non-clear cell RCC were significantly more likely to have an RCC-associated gene mutation (9 [11.7%] of 74 vs 3 [1.7%] of 177; *P* = .001), and 8 (10.0%) had a mutation in a gene that could guide therapy. Of patients with mutations in RCC-associated genes, 5 (35.7%) failed to meet current clinical guidelines for genetic testing.

CONCLUSIONS AND RELEVANCE Of patients with non-clear cell RCC, more than 20% had a germline mutation, of which half had the potential to direct systemic therapy. Current referral criteria for genetic testing did not identify a substantial portion of patients with mutations, supporting the role of a more inclusive sequencing approach.

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Renal cell carcinoma (RCC) is among the 10 most frequently diagnosed cancers in the United States, affecting approximately 64 000 patients per year.¹ Approximately 30% of patients are initially diagnosed with locoregional (stage III) or metastatic disease.^{2,3} Renal cell carcinoma comprises a heterogeneous group of cancers. Clear cell RCC (ccRCC), the most common subtype, is characterized by loss of function of the von Hippel-Lindau (VHL) protein.⁴ Additional histologic subtypes are collectively known as non-clear cell (nccRCC) and include papillary type I and II, chromophobe, microphthalmia transcription factor family translocation associated, collecting duct, medullary, and other rare subtypes.⁵ Tumors that do not meet criteria for any established subtypes are categorized as unclassified.⁶

Several autosomal dominant inherited cancer syndromes predispose patients to ccRCC and nccRCC.^{7,8} Among these are VHL, hereditary leiomyomatosis and RCC (HLRCC), hereditary papillary RCC, and Birt-Hogg-Dubé, caused by germline mutations in *VHL* (OMIM 608537), *FH* (OMIM 136850), *MET* (OMIM 164860), and *FLCN* (OMIM 607273), respectively.⁴ There is also an increased risk of RCC among patients with germline mutations in *BAP1* (OMIM 603089), *SDHB* (OMIM 185470), *SDHC* (OMIM 602413), *SDHD* (OMIM 602690), *TSC1* (OMIM 605284), *TSC2* (OMIM 191092), and *MITF* (OMIM 156845).⁹⁻¹³

Inherited RCC syndromes are thought to account for 5% of all cases; however, these estimates were derived from mostly early-stage RCC, and no studies have specifically looked at advanced disease.^{14,15} Recent studies¹⁶⁻¹⁸ suggest that germline mutations may be more frequent in patients with advanced cancer compared with those with early-stage disease. Identifying patients with inherited mutations has become particularly relevant because mutations in certain genes, such as *FH* and *MET*, can guide systemic therapy or clinical trial eligibility.¹⁹⁻²² Furthermore, identification of germline mutations could provide useful information for patients and family members for cancer risk stratification and early detection. Despite the potential benefits of identifying germline carriers, there is still uncertainty of who should be referred for genetic testing.

We assessed the frequency of germline mutations in 76 cancer-associated genes in patients with advanced RCC unselected for inherited syndrome risk factors, such as age at onset, multifocal disease, or family history. Our aims were to examine the prevalence of germline mutations in known RCC predisposition genes and other cancer-associated genes and to identify clinical and pathologic factors associated with germline mutations.

Methods

Patient Selection

From October 1, 2015, through July 31, 2017, a total of 267 patients with advanced (AJCC stage III or IV) RCC seen in medical oncology or urology clinics at Memorial Sloan Kettering Cancer Center (MSKCC) were offered germline sequencing and disclosure of results under an institutional protocol of matched tumor-germline DNA sequencing. All patients with advanced nccRCC or advanced ccRCC who participated in clinical trials

Key Points

Question What is the prevalence of germline mutations in known renal cell carcinoma predisposition genes and other cancer-associated genes and what are the clinicopathologic factors associated with mutations?

Findings In this cohort study of 254 patients with advanced renal cell carcinoma unselected for inherited cancer risk factors, 5.5% had mutations in renal cell carcinoma-associated genes and 10.5% in other cancer-associated genes. Among patients with non-clear cell renal cell carcinoma, 20.0% had any germline mutation, and 9% had a mutation diagnostic of hereditary leiomyomatosis (eg, *FH* mutation).

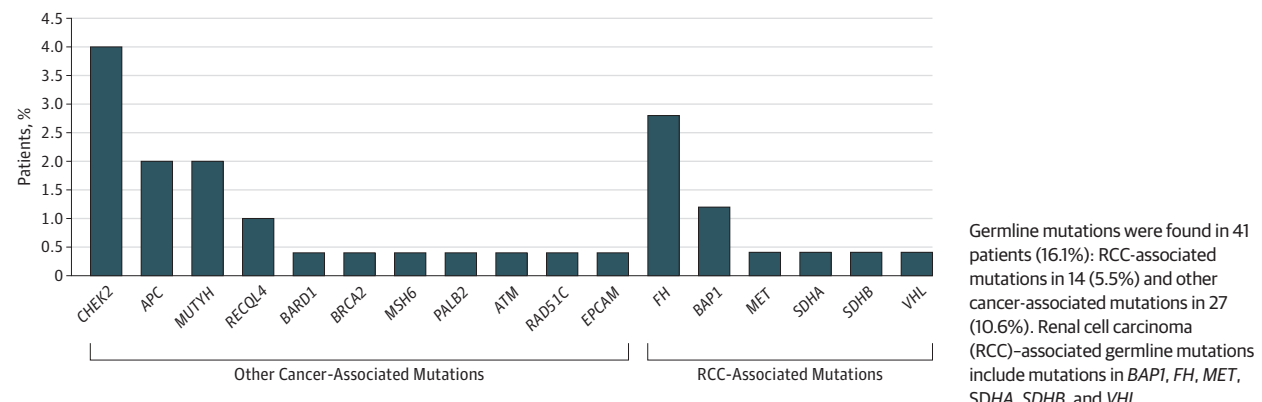
Meaning The results of this study suggest that germline mutations may be frequent in patients with advanced renal cell carcinoma; genetic testing should be considered, especially for patients with advanced non-clear cell renal cell carcinoma.

were approached. Patients were unselected for family history of cancer, age at onset, multifocal tumors, or personal history of multiple malignant tumors. All patients viewed a standard pretest education video and were offered pretest genetic counseling. If pathogenic or likely pathogenic variants were detected, patients discussed the results with a certified cancer genetic counselor (A.A.), who then coordinated family cascade testing, as appropriate. Clinical and family history data were obtained from medical records and self-administered questionnaires. Pathologic test results were reviewed by genitourinary pathologists (Y.-B.C.) at the institution. A total of 128 patients were previously described in a large series that covered multiple tumor types, including patients with localized disease who are not described here; clinical annotation, including RCC tumor subtypes, was also not reported.¹⁸ The study was approved by the MSKCC Institutional Review Board. All patients provided written informed consent. Data were deidentified except to the investigators of the study.

Sequencing and Interpretation of Variants

Tumor and blood samples from patients were sequenced using the Memorial Sloan Kettering-Integrated Mutation Profiling of Actionable Cancer Targets (MSK-IMPACT), a next-generation sequencing assay that achieves hybridization capture with target-specific probe from exons of at least 341 cancer-associated genes in the first iteration and 468 in the most recent, as described previously (eTable 1 in the Supplement).²³ Germline variants in 76 genes associated with cancer predisposition included in the panel were further analyzed.²⁴ All variants with less than 1% frequency in the public database ExAC were interpreted. Only pathogenic or likely pathogenic variants (associated with disease causation and henceforth referred to as pathogenic variants) were considered to be deleterious and are included in this analysis. Pathogenicity was determined by a clinical molecular geneticist or molecular pathologist according to American College of Medical Genetics (ACMG) criteria and updated as of January 2018.²⁵ Variants of unknown significance were reviewed but are not reported in this analysis. Mutations were classified as high (relative risk [RR], >4), intermediate (RR, 2-4) or

Figure 1. Frequency and Distribution of Pathogenic Germline Mutations



low (RR, <2) penetrant, recessive, or of uncertain clinical actionability based on disease risks and prior modeling.²⁶⁻²⁸ The Fraction and Allele-Specific Copy Number Estimates From Tumor Sequencing (FACETS) algorithm was used to evaluate loss of heterozygosity (LOH) in the locus of the variants.²⁹

Comparison of Guidelines-Based Personal and Family History-Based Testing vs Agnostic Testing

Family history, race/ethnicity, and clinical features were abstracted from self-administered questionnaires and medical records. The ACMG guidelines were used to determine indicated genetic tests (eTable 2 in the Supplement).²⁷ For patients who met the guidelines, we assumed that a multigene panel test would be ordered and would include the following genes: *VHL*, *FH*, *FLCN*, *MET*, *SDHB*, *SDHC*, *SDHD*, *BAP1*, *TSC1*, *TSC2*, *TP53* (OMIM 191170), and *MITF*. A pathogenic variant would be considered as incremental if it was detected by sequencing but not by testing based on application of published guidelines.

Statistical Analysis

Demographic and clinical characteristics of the RCC cohort are presented using descriptive statistics. The prevalence of pathogenic or likely pathogenic germline mutations and RCC-associated gene mutations are reported in the RCC cohort and among patients with ccRCC and nccRCC. Clinical characteristics of patients with an RCC-associated gene mutation were compared with those of patients with no RCC-associated gene mutations using the Fisher exact test. Allele frequencies in the cohort were compared with allele frequencies in a non-cancer population obtained from the public database ExAC minus cases derived from The Cancer Genome Atlas. Additional details of ExAC are available at <http://exac.broadinstitute.org/faq>. Known pathogenic and likely pathogenic variants were collapsed by gene and burden tests performed among the RCC cohort vs the noncancer cohort from the ExAC database; odds ratios (ORs) were computed. Statistical analysis was performed using R, version 3.3.2 with RStudio, version 1.0.136 (R Foundation for Statistical Computing). A 2-sided Fisher exact test $P < .05$ was considered to be statistically significant.

Results

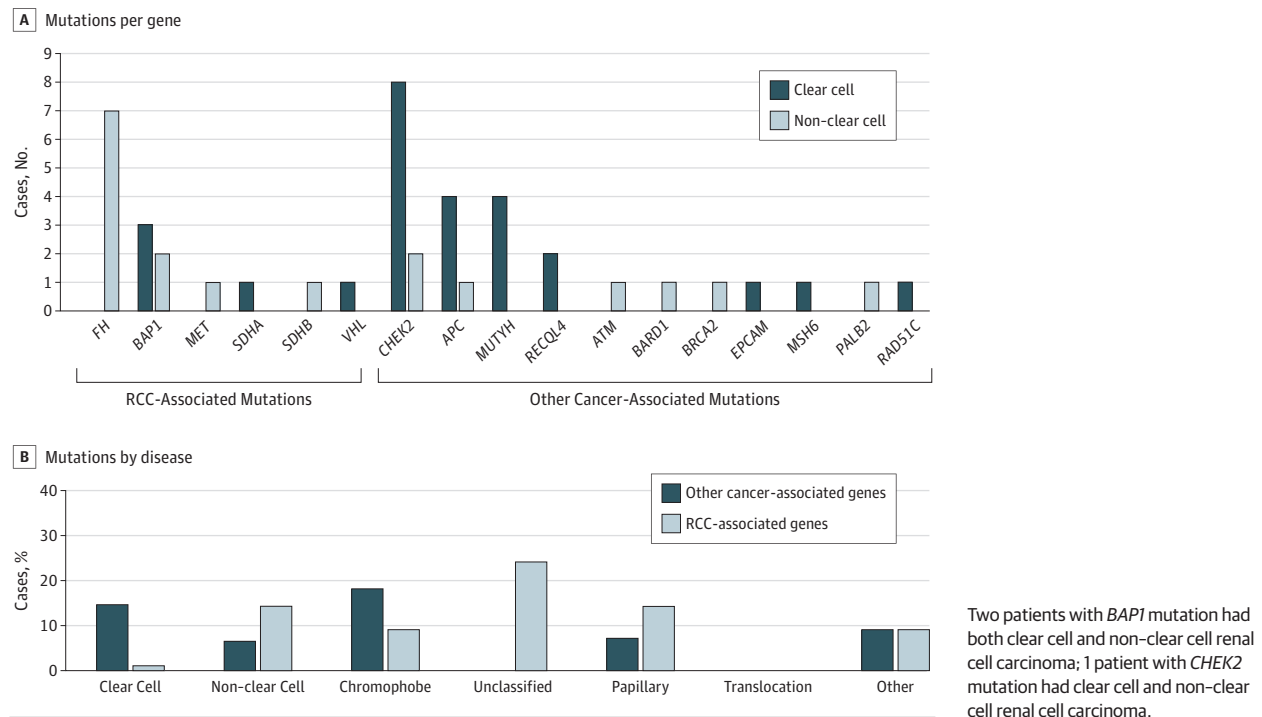
Patient Characteristics

During the study period, 267 individuals with advanced RCC consented to tumor-normal testing with MSK-IMPACT and were offered disclosure of germline results under a separate protocol. Of these, 254 (95.1%) (median [range] age, 56 [13-79] years; 179 [70.5%] male and 75 [29.5%] female; 211 [83.1%] non-Hispanic white) consented to receive the germline results. The primary reason for declining was potential cause of worry; other reasons are listed in eTable 3 in the Supplement. Demographic and clinical characteristics are given in eTable 4 in the Supplement. Among the 254 patients, 177 had ccRCC (69.7%), 74 had nccRCC (29.1%), and 3 (1.2%) had both. Overall, 33 patients (13.0%) had a history of a second malignant tumor, excluding nonmelanoma skin cancers. The most frequent secondary malignant tumors were prostate in 8 patients (3.1%), breast in 4 patients (1.6%), and melanoma in 3 (1.2%). Overall, 14 patients (5.5%) had bilateral or multifocal disease, and 24 (9.4%) reported a family history of RCC.

Frequency of Mutations and Comparison With the Population

A total of 41 patients (16.1%) carried pathogenic or likely pathogenic germline variants in 17 different cancer-predisposition genes (Figure 1). No individuals had more than 1 germline mutation. Fourteen patients (5.5%) carried mutations in RCC-associated genes (7 in *FH*, 3 in *BAP1*, and 1 each in *VHL*, *MET*, *SDHA*, and *SDHB*). Twenty-seven patients (10.5%) carried mutations in genes not clearly associated with RCC, including 9 (3.5%) in *CHEK2* (OMIM 604373); 2 of these were in the variant *CHEK2* p.Ile157Thr, which is a low-penetrance breast cancer risk allele of uncertain actionability.³⁰ Of the 41 patients, 17 patients (41.5%) carried mutations of high penetrance, 9 (22.0%) of moderate penetrance, 12 (29.3%) of low penetrance or uncertain clinical actionability, and 3 (7.3%) in genes linked to autosomal recessive syndromes (eTable 5 in the Supplement). Of the 177 patients with ccRCC, 25 (14.1%) had a germline mutation and 3 (1.7%) in an RCC-associated gene (Figure 2). Of the 74 patients with nccRCC, 13 (17.5%) had a germline mutation, with 9 (12.2%) in an RCC-associated gene.

Figure 2. Pathogenic Mutations by Histologic Subtype



Of the 3 patients with both ccRCC and nccRCC, all 3 had germline mutations, 2 in *BAP1* and 1 in *CHEK2*.

For genes not traditionally associated with RCC, we compared mutation frequencies with those in the general population. The odds of mutations significantly exceeded the odds in the population only for *CHEK2* (OR, 3.0; 95% CI, 1.3-5.8; $P = .003$) when all variants were considered (eTable 6 in the Supplement).

Factors Associated With Germline Mutations

We analyzed prevalence of mutations among patients thought to be at higher risk for inherited syndromes, including those with family history of RCC, early onset (≤ 46 years of age), and multifocal disease at diagnosis. Patients with nccRCC or multifocal RCC were significantly more likely to have an RCC-associated mutation (eTable 7 in the Supplement). Seven patients (9.5% of the nccRCC cohort) had germline *FH* mutations diagnostic of the hereditary syndrome HLRCC; all patients had tumors of unclassified histologic type or identified as *FH* deficient. All but one tumor, for which tissue was limited, were identified by genitourinary pathologists (Y.-B.C.) as suggestive of features of HLRCC. Median age at diagnosis for *FH*-positive patients was 49 years (range, 23-58 years); 4 presented with metastatic disease, and 3 later developed metastatic disease (range, 10 months to 10 years). None had multifocal RCC or family history of RCC. Of the other benign tumors associated with HLRCC, uterine fibroids were present in all women, but cutaneous leiomyomas were identified in only 1 patient after the HLRCC diagnosis was known.

Three patients had *BAP1* mutations: a woman in her 60s with ccRCC, a man in his 40s with ccRCC and a chromophobe tumor, and a man in his 80s with ccRCC, multiple papillary tumors, and colon cancer (pedigrees in the eFigure in the Supplement).

Staining for *BAP1* protein showed lack of expression in the ccRCC tumors of all 3 patients and the chromophobe tumor (eFigure in the Supplement). The papillary tumors were not available for immunohistochemical analysis.

Application of Clinical Genetic Counseling Referral Criteria

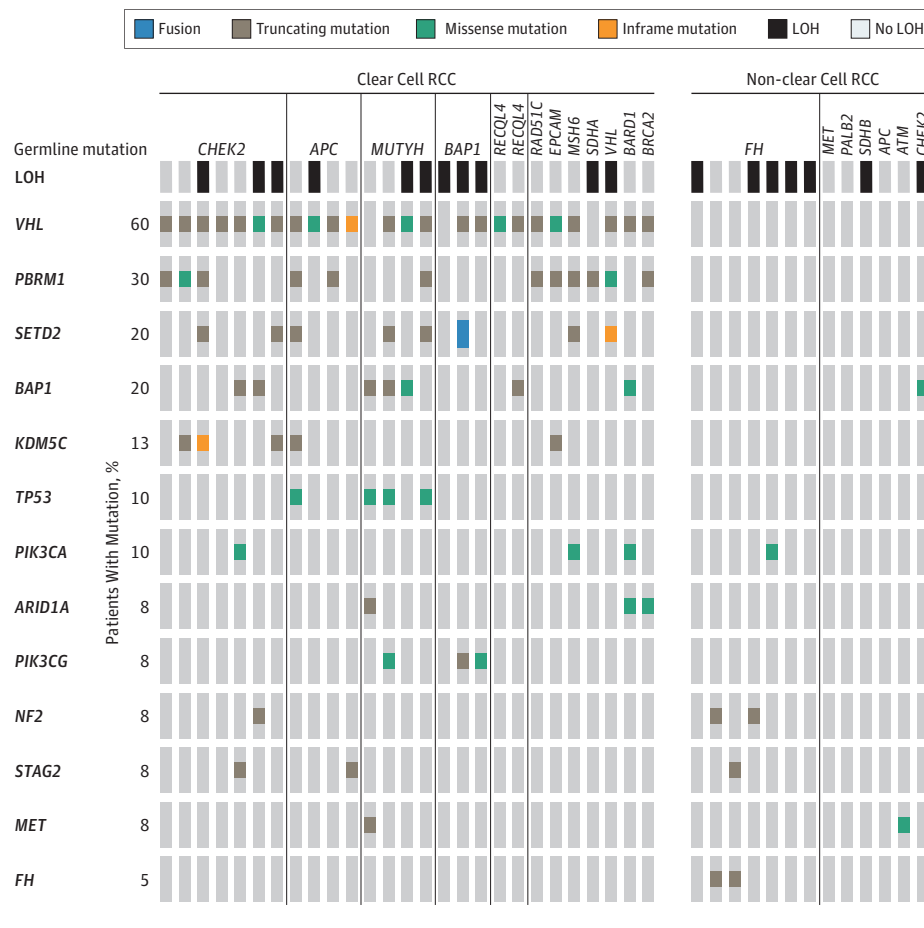
Overall, 99 patients (39.0%) would have met ACMG criteria for clinical genetics referral. Of 14 patients with RCC-associated mutations, 5 (35.7%) would not have met referral criteria, including 3 patients with *FH*, 1 with *VHL*, and 1 with *SDHA*. Of 12 patients with high or moderate penetrance mutations not related to RCC (*ATM* [OMIM 607585], *BRCA2* [OMIM 600185], *CHEK2* excluding variant I157T, *MSH6* [OMIM 600678], *PALB2* [OMIM 610355], *RAD51C* [OMIM 602774]), 7 patients would not have met referral criteria. At the time of analysis, at least 6 relatives of patients with mutations underwent germline testing through our clinic; several were also found to be carriers and referred for discussion of dedicated cancer screening.

Somatic Mutations in Patients With Germline Mutations

Tumor sequencing data were available for all but 1 patient. For 3 patients with both ccRCC and nccRCC, only the ccRCC was sequenced. In patients with germline mutations, we show the distribution of somatic variants and LOH in the tumor at the loci of interest (Figure 3). Of 9 patients with germline *CHEK2* mutations, 4 (44.4%) had LOH. Of 7 patients with germline *FH* mutations, 5 (71.4%) had LOH and 2 (28.6%) had a somatic second hit.

We investigated the clinical utility of tumor-only testing for guiding therapy using previously described methods.³¹ No patients had a level 1 or level 2A standard of care somatic biomarker predictive of response in RCC. A total of 36 (14.2%) had

Figure 3. Somatic Mutations and Loss of Heterozygosity (LOH) in Patients With Germline Mutations



a level 2B or 3 somatic mutation that was a predictive biomarker in other cancers but not routinely used in RCC (eTable 8 in the Supplement).

Discussion

We report the prevalence of germline cancer-associated mutations in patients with advanced RCC unselected for suspicion of a hereditary syndrome. To date, approximately 5% of all RCC cases have been linked to high-penetrance cancer predisposition syndromes.⁴ Most studies^{14,15} on hereditary RCC, however, have been limited to selected, high-risk populations, and none to our knowledge have specifically studied patients with advanced disease. In this study, we found that among 254 patients with advanced RCC, 41 (16.1%) harbored pathogenic germline mutations, with 14 (5.5%) in RCC-associated genes and 27 (10.5%) in non-RCC-associated genes. The mutation rate was even higher among patients with uncommon RCC variants: more than 20% of patients with an nccRCC had germline mutations, of which half could help direct therapy according to previously published data from prospective clinical trials.^{19,20,32,33}

Previous data^{16,17} suggest that the prevalence and spectrum of germline mutations differ between early- and late-

stage disease in several cancers. In an analysis of 1040 patients with diverse cancers, a recent study¹⁸ found that the prevalence of germline, cancer-predisposing mutations was higher among patients with metastatic compared with localized disease (22.3% vs 8.3%; $P < .001$). In a separate study³⁴ of 1235 patients with RCC with all stages of disease who were referred for multigene germline testing, 6% had mutations, but the spectrum of mutations differed from that in our cohort. The most frequent mutations were in *FLCN* (1.8% vs 0% in our cohort). Despite the enrichment in that series for suspected hereditary syndromes, *FH* mutations were less prevalent (1.3% vs 3% in our cohort) and only 2 *BAP1* mutations were identified. *FH* and *BAP1* mutations may be associated with more aggressive RCC, which could account for their overrepresentation in the current cohort of patients with advanced cancer.^{35,36}

We found that 9% of all patients with advanced nccRCC had a germline *FH* mutation diagnostic of HLRCC, a higher percentage than previously reported.^{37,38} Hereditary leiomyomatosis and RCC is an autosomal dominant inherited syndrome associated with RCC and uterine and cutaneous leiomyomas. These patients were difficult to identify by clinical criteria. None had a family history of RCC, and typical cutaneous lesions were not identified by the treating oncologists. Although expert genitourinary pathologists identified histologic features suggestive of HLRCC, it is

unclear whether nonspecialist pathologists would be able to draw the same conclusions.^{39,40} Relatives who are also found to carry *FH* mutations should be considered for RCC screening. Early detection may increase the likelihood of cure and survivorship.^{38,41} We also found that 2 of 3 individuals with *BAP1* mutations had nccRCC tumors, which, to our knowledge, have never been reported in *BAP1* carriers.⁴² The available chromophobe tumor had loss of expression of *BAP1* on immunohistochemical analysis, which is rare in chromophobe tumors.⁴³ The possible association of *BAP1* mutations with nccRCC should be investigated in other cohorts.

Novel biomarkers predictive of therapy response are needed in RCC.^{31,44} In this cohort, 10% of patients with nccRCC had a predictive germline biomarker, none of which would have been identified with somatic-only sequencing. In RCC, several germline mutations are prognostic indicators of response to therapies. In a phase 2 biomarker study²⁰ of a dual MET/VEGFR2 in patients with papillary RCC, the presence of a germline *MET* mutation was associated with response. Another phase 2 study³³ of bevacizumab plus erlotinib for patients with sporadic or hereditary papillary RCC showed significant activity with the combination, particularly in those with HLRCC. On the basis of this study,³³ the 2018 National Comprehensive Cancer Network guidelines added bevacizumab plus everolimus or erlotinib as options specifically for patients with germline *FH* mutations diagnostic of HLRCC.⁴⁵ Because of the high positive germline mutation rate among patients with nccRCC, the difficulty using clinical criteria to identify them, and the potential for therapeutic actionability, patients with advanced nccRCC should be referred for genetic counseling and appropriate testing.

Compared with the general population, patients with RCC had a significantly increased frequency of germline *CHEK2* mutations. Germline *CHEK2* mutations are associated with increased susceptibility to several cancers. Few studies^{46,47} have found an increased risk of RCC among carriers of the 1100deC truncating mutation and Polish founder mutations, with ORs ranging from 2.1 to 3.6. We found an OR of 3.0 of RCC when all variants of *CHEK2* were considered. A potential role of *CHEK2* in the pathogenicity of RCC is also supported by the LOH in the tumor in 4 of 9 germline *CHEK2* mutants. Although there are currently no RCC-specific screening recommendations for individuals with *CHEK2* mutations, there may be incremental screening for other cancers, justifying including this gene on RCC panel tests.²⁸

For patients with advanced RCC, current clinical guidelines for referral to genetic counseling miss patients who could

benefit from genetic counseling. The ACMG has developed practice guidelines that consider age at diagnosis, tumor multifocality, family history, and histologic findings.²⁷ We found that despite almost 40% of patients in this cohort meeting the broad guideline criteria, 36% of patients with high-penetrance RCC-associated mutations would have been missed. With potentially increasing numbers of mutations identified in patients and relatives, optimal cancer screening schedules need to be studied. For example, there are no consensus screening guidelines for several of the more rare RCC genetic syndromes, including *BAP1*-associated cancer syndrome, although some groups have suggested screening schedules.⁴⁸

Limitations

There are limitations to this study. Many patients in this cohort also participated in clinical trials. Therefore, patients who contributed to this study most likely were younger and had fewer comorbid conditions than the general cancer population. For example, the median age at diagnosis in this study was 56 years compared with 64 years in the United States.¹ Nevertheless, this limitation may underestimate the number with germline mutations because most trials exclude patients with a history of other malignant tumors. We acknowledge that some Ashkenazi Jewish founder mutations, such as *APC* (OMIM 611731) p.Ile1307Lys and *CHEK2* p.Ser428Phe, may be over-represented in our cohort. Although the association of *APC* and RCC risk is unclear, for *CHEK2*, we also performed a burden test excluding the p.Ser428Phe variant, and the association with RCC was still present (eTable 6 in Supplement). Finally, although we analyzed 254 patients, the sample size was still limited, and further studies should be conducted.

Conclusions

Our results suggest that germline mutations in cancer-associated genes in patients with advanced RCC may be prevalent, and many of these mutations can be used to guide therapy. Phenotype-directed or tumor-only testing would have failed to identify most patients with actionable mutations. A broader approach to tumor-normal sequencing of all patients with advanced RCC, especially those with nccRCC, might help identify individual patients for whom targeted therapies are indicated, as well as family members who may benefit from preventive interventions tailored to their increased cancer risk.

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Study concept and design: Carlo, Mukherjee, Vijai, Kemel, Cadoo, Lee, Robson, Motzer, Offit.

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Study supervision: Chen, Coleman, Cadoo, Lee, Feldman, Motzer, Offit.

Conflict of Interest Disclosures: Dr Carlo reported serving a consulting or advisory role for Pfizer.

Dr Mukherjee reported being employed within the past 2 years by Regeneron Pharmaceuticals. Dr Hyman reported serving a consulting or advisory role for Atara Biotherapeutics, Chugai Pharma, CytomX Therapeutics, Boehringer Ingelheim, and AstraZeneca and receiving research funding from AstraZeneca, Puma Biotechnology, and Loxo. Dr Ladanyi reported serving a consulting or advisory role for Boehringer Ingelheim and AstraZeneca, receiving honoraria from Merck & Co, and receiving research funding from Loxo. Dr Cadoo reported receiving research funding from AstraZeneca. Dr Stadler reported serving a consulting or advisory role for Allergan, Genentech/Roche, Regeneron, Optos, and Adverum. Dr Lee reported serving a consulting or advisory role for Exelixis and receiving research funding from Pfizer, Eisai, and Bristol-Myers Squibb. Dr Feldman reported receiving research funding from Novartis and Seattle Genetics. Dr Voss reported serving a consulting or advisory role for Novartis, Calithera Biosciences, Natera, GlaxoSmithKline, Exelixis, Pfizer, and Alexion Pharmaceuticals and receiving travel and accommodation expenses from Novartis and Takeda, honoraria from Novartis, and research funding from Bristol-Myers Squibb and Genentech/Roche. Dr Robson reported serving a consulting or advisory role for AstraZeneca and McKesson and receiving travel and accommodation expenses from AstraZeneca, honoraria from AstraZeneca, and research funding from AstraZeneca, AbbVie, Medivation, Myriad Genetics, and InVita. Dr Motzer reported serving a consulting or advisory role for Novartis, Pfizer, Eisai, Exelixis, and Merck & Co and receiving research funding from Bristol-Myers Squibb, Genentech/Roche, Pfizer, GlaxoSmithKline, Eisai, and Novartis. No other disclosures were reported.

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Invited Commentary

Broadening the View of Germline Mutations in Kidney Cancer

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Next-generation sequencing has revolutionized precision oncology, with paired somatic and germline DNA variant analysis becoming more powerful and more widely accessible for clinical applications. The field of clinical cancer genetics previously relied primarily on a patient's personal and family medical history to delineate specific



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hereditary cancer syndromes that are associated with deleterious variants in specific genes. Tumor sequencing technology is currently used frequently in oncology with the goal of identifying deleterious gene variants that are actionable or druggable, resulting in the rapid proliferation of targeted anticancer therapies. Through this approach, a significant portion of patients have been incidentally found to have pathogenic or likely pathogenic germline mutations, and a previous study¹ found that patients with advanced cancers across tumor types are enriched for hereditary pathogenic variants in cancer-related genes. Of importance, many of these patients do not meet current clinical criteria for germline testing. Thus, mutations in genes that are potentially associated with a patient's cancer treatment and care, as well as cancer risk and prevention for the patient's family members, are going undiscovered.

Hereditary renal cell carcinoma (RCC) syndromes are thought to account for approximately 5% of kidney cancers and arise from pathogenic variants in a diverse array of genes. The histologic features of RCC, family history of RCC, multifocal disease, age at onset of 46 years or younger, and/or the presence of other characteristic tumor types contribute to the decision to refer a patient to a clinical cancer geneticist for germline testing using targeted panels of RCC-related genes.^{2,3} It is well known that RCC histologic subtypes have distinct molecular profiles and driver mutations, and thus the histologic subtype of kidney cancer can suggest the presence of certain

germline mutations.³ However, the prevalence of pathogenic germline variants in patients with locoregional and/or metastatic RCC unselected for family history or other hereditary features has not been well evaluated. In addition, the presence of pathogenic germline variants in a broader set of cancer-related genes outside canonical RCC-related genes has not been comprehensively described in this population.

In this issue of *JAMA Oncology*, Carlo et al⁴ provide a single-institution, retrospective analysis of paired tumor and normal DNA sequencing of 76 cancer-related genes in patients with advanced RCC (stage III and IV). In line with a recent study¹ in patients with advanced cancer across various tumor types, this patient population with advanced RCC is enriched for germline variants in cancer-associated genes at an overall prevalence of 16%. Most pathogenic variants (10.5%) were in genes not typically associated with RCC that mostly centered on the DNA damage response (DDR) and cell cycle checkpoint pathways, highlighting the concept that broader panel testing may be warranted in select patients with RCC. In addition, non-clear cell RCC (nccRCC) histologic subtypes were disproportionately enriched for germline variants (21%), with a significantly higher rate of variants in RCC-related genes compared with clear cell histologic subtypes, suggesting that all patients with advanced nccRCC warrant referral to genetic counseling to consider germline testing. Approximately one-third of the patients with germline variants in RCC-related genes did not meet current clinical criteria for genetic testing, highlighting the importance of these study findings.

With regard to clinical indicators of germline mutation carrier status, the study by Carlo et al⁴ revealed that histologic findings matter: patients with nccRCC and/or multifocal disease were significantly more likely to carry mutations. The most commonly mutated gene in nccRCC histologic subtypes was