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Expanding the Clinical Phenotype of Hereditary *BAP1* Cancer Predisposition Syndrome, Reporting Three New Cases

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Abstract

The clinical phenotype of *BAP1* hereditary cancer predisposition syndrome (MIM 614327) includes uveal melanoma (UM), cutaneous melanoma (CM), renal cell carcinoma (RCC), and mesothelioma. However, the frequency of the syndrome in patients with UM and the association with other cancers are still not clear. In this study, we screened 46 previously untested, unrelated UM patients with high risk for hereditary cancer for germline mutation in *BAP1*. We also studied four additional patients with a personal or family history suggestive of *BAP1* hereditary cancer syndrome. We identified three patients with germline pathogenic mutations (c.2050 C>T, p.Gln684*; c.1182C>G, p.Tyr394*, and c.1882_1885delTCAC, p.Ser628Profs*8) in *BAP1*. Two of these three patients presented with UM and the third with a metastatic adenocarcinoma likely from a hepatic cholangiocarcinoma. Reported family histories included UM, mesothelioma, RCC, CM, and several other internal malignancies. The results of this study confirm the association between germline *BAP1* mutation and predisposition to UM, mesothelioma, CM and RCC. However, other cancers, such as cholangiocarcinoma and breast carcinoma may be part of the phenotype of this hereditary cancer predisposition syndrome. In addition, the results support the existence of other candidate genes in addition to *BAP1* contributing to hereditary predisposition to UM.

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INTRODUCTION

Germline mutations in the *BAP1* gene have been identified in a small number of families with hereditary cancers (Abdel-Rahman et al., 2011; Popova et al., 2013; Testa et al., 2011; Wiesner et al., 2011). The clinical phenotype of *BAP1* hereditary cancer predisposition syndrome (MIM 614327) includes uveal melanoma (UM), mesothelioma, cutaneous melanoma (CM), renal cell carcinoma (RCC), and melanocytic *BAP1*-mutated atypical intradermal tumors (MBAITs) also known as atypical Spitz tumors/nevi (Abdel-Rahman et al., 2011; Carbone et al., 2012; Murali et al., 2013; Popova et al., 2013; Testa et al., 2011; Wiesner et al., 2011). MBAITs share some histological characteristics with atypical Spitz tumors, but they are significantly different histologically and molecularly to justify the separate name of MBAITs (Carbone et al., 2012). Several other tumors were also reported in these families, including meningioma, lung adenocarcinoma, ovarian, pancreatic and breast cancers (Abdel-Rahman et al., 2011; Carbone et al., 2012; Njauw et al., 2012). However, it is still not clear whether these tumors are part of the phenotype of this syndrome. Proper characterization of the phenotype is crucial to define diagnostic criteria and design management and follow-up protocols for patients with germline *BAP1* mutations.

In the following study, we report three additional patients with germline *BAP1* mutations, including one presenting with metastatic adenocarcinoma likely from a cholangiocarcinoma. Our study supports that UM, CM, RCC, and mesothelioma are part of the clinical phenotype of *BAP1* hereditary cancer predisposition syndrome. However, it also indicates that other cancers, such as cholangiocarcinoma and breast carcinoma could be part of the phenotype.

MATERIALS AND METHODS

Patient Selection

This work was done under a research protocol approved by the Institutional Review Board of The Ohio State University. We evaluated a total of 50 patients, average age 46 years (range 15–85), 34 women and 16 men, not included in our previous study (Abdel-Rahman et al., 2011). Forty-six of those presented with UM, including nine patients with family history of UM, one patient with bilateral UM, one patient with two separate primary UM, and 35 UM patients with one or more of the following: (1) 30 years or younger at time of diagnosis; (2) personal history of a separate primary cancer; and (3) strong family history of cancer (Abdel-Rahman et al., 2010). Figure 1A represents a summary of cancer histories of the 46 UM patients included in the study. In addition, we included four patients with personal or family history highly suggestive of *BAP1* hereditary cancer predisposition including a male patient diagnosed with metastatic adenocarcinoma and a family history of CM, mesothelioma, pancreatic, and ovarian cancers; a female patient diagnosed with lung adenocarcinoma and a family history of UM, uveal nevi, lung, and colon cancers; a female patient diagnosed with bilateral breast cancer with family history of UM and breast cancers; and a female patient diagnosed with MBAITs (Carbone et al., 2012; Wiesner et al., 2011). Peripheral blood was obtained from all patients for DNA extraction.

DNA Extraction, Mutational Screening, and Genotyping

Germline DNA was extracted from mononuclear cells at the Human Cancer Genetics Sample Bank, The Ohio State University, according to the published protocol using a simple salting out procedure (Miller et al., 1988). Tumor DNA was extracted from archival material using Qiagen DNeasy kits (Qiagen, Valencia, CA). Mutational screening was carried out by direct sequencing according to previously published protocol (Abdel-Rahman et al., 2011). All identified sequence variations were confirmed at least once in an independent PCR experiment. Genotyping was carried out on tumor tissue from the index case of FUM103 (III.1) diagnosed with metastatic carcinoma and from individual FUM064 (III-12) diagnosed with peritoneal papillary tumor using previously reported microsatellite markers (Abdel-Rahman et al., 2011).

Immunohistochemistry

Immunohistochemistry was carried out on tumor tissues from FUM103 (III.1) and FUM064 (III-12). For BAP1, we used a mouse monoclonal antibody (Clone C4, SantaCruz biotechnology) at 1:100 dilution and the Dako EnVision+System HRP utilizing the manufacturer's protocol. Staining of the nontumor tissue was used as positive control and immunostaining without the primary antibody was used as negative control. Positive staining was assessed by a pathologist (MHA) using a Nikon Eclipse i50 brightfield microscope with Nikon digital sight DS-U1 5MP digital camera (Nikon, Japan). For the papillary peritoneal tumor immunohistochemistry for calretinin, MOC31 (Ruitenbeek et al., 1994), Ber-EP4 (Latza et al., 1990), and PA×8 (Tong et al., 2010) were carried out in a certified clinical laboratory.

RESULTS

Out of the 50 patients tested, we identified three with pathogenic mutations in *BAP1* and 4 with variants of uncertain significance: c.2057-4G>T (rs149499021) in two different patients, both c.2057-22A>C (rs144083199) and c.*45C>G (rs56898787) in a third patient and c. 932-58_59delTG in a fourth patient (Table 1). Splice site prediction of these four variants, utilizing both NetGene 2 version 2.42 (Hebsgaard et al., 1996) and NNSPLICE version 0.9 software (Reese et al., 1997), indicated that they are not potential splice sites, suggesting that they are likely not pathogenic.

Case Summaries

FUM064—A germline truncating mutation (c.2050 C>T, p.Gln684*) of *BAP1* was identified in the proband (IV.1), who presented with UM (age 41), an epithelial malignancy of unknown origin at the porta hepatis with distant metastasis (age 42) and an unclassified spindle cell proliferation in her thigh (age 42). The tumor at the porta hepatis was positive for pancytokeratin and negative for HMB45, MART-1, and S100 indicating that it is a second primary malignancy rather than a metastasis from her UM. The family history was striking for UM diagnoses in her father and paternal second cousin, as well as diagnoses of mesothelioma in three paternal relatives and multiple other cancers in paternal relatives, including renal cell, pancreatic, breast, and colorectal carcinomas (see Fig. 1B). A paternal cousin once-removed (III-12) presenting with peritoneal papillary tumor was also positive

for the same mutation; thus, making their parents and the proband's paternal grandmother obligate carriers of the same mutation. Cancer diagnoses reported in these obligate carriers included UM, an unspecified spinal tumor, and a "stomach" cancer.

The peritoneal tumor in individual III-12 was originally diagnosed as a papillary peritoneal serous adenocarcinoma. However, slide review and immunohistochemistry showed strong positive staining of the tumor cells for calretinin with focal positivity for MOC31 and Ber-EP4 and negative staining for PA×8 supporting its mesothelial origin (Supporting Information Fig. S1). Genotyping of the tumor tissue from individual III-12 showed retention of heterozygosity of microsatellite markers in close proximity to *BAP1* suggesting no somatic deletion. However, immunohistochemistry showed loss of BAP1 nuclear localization in tumor cells (Fig. 2B) with strong expression in nontumor tissue suggesting biallelic inactivation of BAP1 in the tumor tissue. No other tumor tissue was available from the family for testing.

FUM103—A germline truncating mutation c.1182C>G, p.Tyr394* was identified in the proband (III.1) who presented with a metastatic adenocarcinoma to the rib and a hepatic focal lesion. Upper endoscopy and colonoscopy were negative for malignancies. The metastatic adenocarcinoma was positive for pancytokeratin, cytokeratin 7, cytokeratin 19, and Ber-EP while negative for cytokeratin 20, cytokeratin 17, CD30, α -feto protein, S100, calretinin, hepatocyte paraffin 1 (Hep Par 1), and carcinoembryonic antigen. Abdominal computerized tomography showed a hepatic focal lesion which was treated by stereotactic body radiation therapy and no tumor tissue available for evaluation. Family history included pancreatic carcinoma, CM, mesothelioma, and ovarian cancer (Figs. 1D and 2C). No tumor tissue was available from any of the relatives for further evaluation. Genotyping of the metastatic tumor from the proband showed retention of heterozygosity of markers surrounding the *BAP1* gene. Immunostaining for BAP1 showed strong cytoplasmic staining with loss of nuclear localization in tumor cells.

FUM104—This family was referred to our centre from an outside institute. The index case (III.1) was a deceased female, who had a personal history of three separate primary cancers: CM, UM (originating from ciliary body), and colorectal cancer. Family history was positive for RCC in multiple individuals, mesothelioma in multiple individuals, breast, pancreatic, ovarian, and liver cancers. The index case's son presented with RCC and was tested negative for *VHL* mutations. A germline frameshift mutation c.1882_1885delTCAC, p.Ser628Profs*8 in *BAP1* was identified in the index case, her son and a great maternal aunt, who presented with invasive breast cancer (Figs. 1C and 2B). The mutation leads to early truncation of the BAP1 protein at codon 636. No tumor tissue or germline DNA from additional individuals was available for further studies from this family.

DISCUSSION

BAP1 hereditary cancer predisposition syndrome is a recently identified familial cancer syndrome. The association of germline *BAP1* mutation with increased risks for UM, mesothelioma, CM, RCC, and MBAITs is now fairly well established (Abdel-Rahman et al., 2011; Popova et al., 2013; Testa et al., 2011; Wiesner et al., 2011). However, several other

cancers have been reported in these families, and it is still unclear whether these cancers are part of the syndrome (Carbone et al., 2012, 2013).

In the present study, we report three new families with germline pathogenic mutations in *BAP1*. One of the mutations (p.Q684*) has been previously reported in another hereditary mesothelioma/UM family (Testa et al., 2011). Discussion with the authors of that study suggests that the two families are unrelated, although we cannot rule out a founder mutation. The two other mutations (p.Tyr394* and p. Ser628Profs*8) have not been previously reported. Cancers reported in patients with germline *BAP1* mutation in our study included cancers associated with *BAP1* hereditary cancer predisposition syndrome, such as UM, CM, RCC and mesothelioma, as well as, other cancers such as hepatic cholangiocarcinoma and breast carcinoma. In addition, breast, pancreatic and ovarian cancers have been reported in first- and second-degree relatives of the index cases. However, germline DNA and tumor tissues were not available for other individuals to identify their mutational status.

The index case of family FUM103 presented with a metastatic adenocarcinoma in his rib with a hepatic focal lesion. The immunostaining pattern of the tumor suggested that the primary tumor is likely a hepatic cholangiocarcinoma. However, no tissue was available from the hepatic focal lesion for validation. Loss of nuclear expression of *BAP1* in the tumor tissue supports it being part of the *BAP1* cancer phenotype. Pancreatic and biliary cancers have been previously reported in few *BAP1* families (Njauw et al., 2012). Further epidemiological studies are needed to validate our findings.

One individual (III-12) from family FUM064 presented with a well-differentiated papillary mesothelioma (WDPM). The tumor lesion was originally diagnosed as a low-grade papillary serous carcinoma of the peritoneum but slide review and immunostaining confirmed the mesothelial nature of the tumor. WDPM has been recently reported by another group in two siblings with germline mutation in *BAP1* (Ribeiro et al., 2013). It is a rare subtype of epithelioid mesothelioma most commonly involving the peritoneum of women and it is not related to asbestos exposure. Our study supports that it is part of the *BAP1* hereditary cancer syndrome phenotype.

An earlier study by our group suggested that the frequency of germline mutation in *BAP1* is low (1/53) in patients with UM, even in those with strong personal or family histories of cancer (Abdel-Rahman et al., 2011). Our current study confirms our earlier findings and suggests the existence of additional candidate genes predisposing to hereditary UM. In support of that a recent study by another group has shown that only 1/8 of familial UM cases had germline *BAP1* mutation (Popova et al., 2013).

Whether other cancers seen in other mutation carriers in these families are coincidental or due to the mutation has yet to be definitively established. Germline mutation in *BAP1* has been observed in one patient in our study as well as reported in a few high-risk breast cancer families suggesting that breast cancer could be part of the phenotype. Identification of additional affected families will further clarify the full tumor spectrum associated with this disorder. In addition, the limited current data do not allow any accurate estimation of either the lifetime risks or average age of diagnosis for each of these associated cancers.

Nevertheless, it appears clear that carrying a *BAP1* germline mutation puts an individual at significantly increased risk of cancer. Pending further clarification of the full phenotype for this condition, we propose the following management guidelines for mutation carriers:

1. Annual ophthalmological examination starting at age of 11 years (5-year younger than the earliest reported UM in *BAP1* families (Hoim et al., 2013) and referral of patients with any pigmented lesions to an ocular oncologist for follow-up or treatment.
2. Annual dermatological examination starting at age 22 years (5 years younger than the earliest reported CM in *BAP1* families (Abdel-Rahman et al., 2011).
3. Follow the American Cancer Society guidelines for screening of other cancers.
4. Follow-up with primary care physician for symptoms and signs of other cancers.

The high frequency of germline *BAP1* mutations in patients presenting with metastatic disease suggests that UM is more aggressive in these patients (Njauw et al., 2012). Early diagnosis and treatment could change the outcome in these patients.

In conclusion, germline *BAP1* mutations appear to predispose patients to an increasing spectrum of cancers including UM, CM, mesothelioma, and RCC. However, other cancers, including cholangiocarcinoma and breast carcinoma may be part of the phenotype. In addition to *BAP1*, other candidate gene(s) likely contribute to hereditary cancer predisposition in UM. Finally, the current evidence justifies establishment of surveillance protocols for early diagnosis of UM and CM in patients with germline mutation in *BAP1*. Further clarification of the full phenotype for this condition is still needed.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

- Abdel-Rahman MH, Pilarski R, Ezzat S, Sexton J, Davidorf FH. Cancer family history characterization in an unselected cohort of 121 patients with uveal melanoma. *Fam Cancer*. 2010; 9:431–438. [PubMed: 20157784]
- Abdel-Rahman MH, Pilarski R, Cebulla CM, Massengill JB, Christopher BN, Boru G, Hovland P, Davidorf FH. Germline *BAP1* mutation predisposes to uveal melanoma, lung adenocarcinoma, meningioma, and other cancers. *J Med Genet*. 2011; 48:856–859. [PubMed: 21941004]
- Carbone M, Korb Ferris L, Baumann F, Napolitano A, Lum CA, Flores EG, Gaudino G, Powers A, Bryant-Greenwood P, Krausz T, Hyjek E, Tate R, Friedberg J, Weigel T, Pass HI, Yang H. *BAP1* cancer syndrome: malignant mesothelioma, uveal and cutaneous melanoma, and MIBITs. *J Transl Med*. 2012; 10:179. [PubMed: 22935333]
- Carbone M, Yang H, Pass HI, Krausz T, Testa JR, Gaudino G. *BAP1* and cancer. *Nat Rev Cancer*. 2013; 13:153–159. [PubMed: 23550303]

- Hebsgaard SM, Korning PG, Tolstrup N, Engelbrecht J, Rouze P, Brunak S. Splice site prediction in *Arabidopsis thaliana* pre-mRNA by combining local and global sequence information. *Nucleic Acids Res.* 1996; 24:3439–3452. [PubMed: 8811101]
- Hoim V, Edsgard D, Helgadottir H, Eriksson H, All-Ericsson C, Tuominen R, Ivanova I, Lundeberg J, Emanuelsson O, Hansson J. Hereditary uveal melanoma: A report of a germline mutation in BAP1. *Genes Chromosomes Cancer.* 2013; 52:378–384. [PubMed: 23341325]
- Latz U, Niedobitek G, Schwarting R, Nekarda H, Stein H. Ber-EP4: new monoclonal antibody which distinguishes epithelia from mesothelial. *J Clin Pathol.* 1990; 43:213–219. [PubMed: 1692040]
- Miller SA, Dykes DD, Polesky HFrn. A simple salting out procedure for extracting DNA from human nucleated cells. *Nucleic Acids Res.* 1988; 16:1215. [PubMed: 3344216]
- Murali R, Wiesner T, Scolyer RA. Tumours associated with BAP1 mutations. *Pathology.* 2013; 45:116–126. [PubMed: 23277170]
- Njauw CN, Kim I, Piris A, Gabree M, Taylor M, Lane AM, DeAngelis MM, Gragoudas E, Duncan LM, Tsao H. Germline BAP1 inactivation is preferentially associated with metastatic ocular melanoma and cutaneous-ocular melanoma families. *PLoS One.* 2012; 7:e35295. [PubMed: 22545102]
- Popova T, Hebert L, Jacquemin V, Gad S, Caux-Moncoutier V, Dubois-d'Enghien C, Richaudeau B, Renaudin X, Sellers J, Nicolas A, Sastre-Garau X, Desjardins L, Gyapay G, Raynal V, Sinilnikova OM, Andrieu N, Manie E, de Pauw A, Gesta P, Bonadona V, Maugard CM, Penet C, Avril MF, Barillot E, Cabaret O, Delattre O, Richard S, Caron O, Benfodda M, Hu HH, Soufir N, Bressac-de Paillerets B, Stoppa-Lyonnet D, Stern MH. Germline BAP1 mutations predispose to renal cell carcinomas. *Am J Hum Genet.* 2013; 92:974–980. [PubMed: 23684012]
- Reese MG, Eeckman FH, Kulp D, Haussler D. Improved splice site detection in Genie. *J Comput Biol.* 1997; 4:311–323. [PubMed: 9278062]
- Ribeiro C, Campelos S, Moura CS, Machado JC, Justino A, Parente B. Well-differentiated papillary mesothelioma: clustering in a Portuguese family with a germline BAP1 mutation. *Ann Oncol.* 2013; 24:2147–2150. [PubMed: 23585512]
- Ruitenbeek T, Gouw AS, Poppema S. Immunocytology of body cavity fluids. MOC-31, a monoclonal antibody discriminating between mesothelial and epithelial cells. *Arch Pathol Lab Med.* 1994; 118:265–269. [PubMed: 8135629]
- Testa JR, Cheung M, Pei J, Below JE, Tan Y, Sementino E, Cox NJ, Dogan AU, Pass HI, Trusa S, Hesdorffer M, Nasu M, Powers A, Rivera Z, Comertpay S, Tanji M, Gaudino G, Yang H, Carbone M. Germline BAP1 mutations predispose to malignant mesothelioma. *Nat Genet.* 2011; 43:1022–1025. [PubMed: 21874000]
- Tong GX, Devaraj K, Hamele-Bena D, Yu WM, Turk A, Chen X, Wright JD, Greenebaum E. Pax8: a marker for carcinoma of Mullerian origin in serous effusions. *Diagn Cytopathol.* 2010; 39:567–574. [PubMed: 20607683]
- Wiesner T, Obenaus AC, Murali R, Fried I, Griewank KG, Ulz P, Windpassinger C, Wackernagel W, Loy S, Wolf I, Viale A, Lash AE, Pirun M, Socci ND, Rutten A, Palmedo G, Abramson D, Offit K, Ott A, Becker JC, Cerroni L, Kutzner H, Bastian BC, Speicher MR. Germline mutations in BAP1 predispose to melanocytic tumors. *Nat Genet.* 2011; 43:1018–1021. [PubMed: 21874003]

Figure 1.

Summary of the patients included in the study. (A) Venn diagram summarizing the cancer history of the 46 UM patients included in the study. CM: cutaneous melanoma; UM: uveal melanoma; RCC: renal cell carcinoma. Black plus sign indicates individuals with mutation identified by sequencing. Grey plus sign indicates obligate carriers. (B) FUM 064: Individuals IV.1 and III.12 were heterozygous for a truncating mutation in *BAP1* the c.2050C>T, p.Gln684*. Individuals II.1, III.1 and II.4 are obligate carriers. Mutations reported in the family were UM (III.1, IV.1, and IV.5), mesothelioma (II.3, III.3, III.11, and III.12), RCC (III.9), stomach (II.4), unknown primary (II.1, II.2), spindle cell malignancy (IV.1), pancreatic (III.5), papillary thyroid (III.4), colorectal (IV.3), and breast (III.2). (C) FUM 104: Individuals III.1, II.5, and IV.3 were heterozygous for a frame shift mutation (c.1882_1885delT-CAC, p. Ser628Profs*8) in *BAP1*. Cancers reported in the family were UM (III.1), RCC (III.3, III.4, III.5, and IV.3), mesothelioma (III.3, III.4), colon (III.1), lung (III.3, III.4), breast (II.5, III.6, IV.1, IV.5), hematological (IV.6), bladder (IV.4), and pancreatic (not listed). No other individuals were tested. (D) FUM103: Individual III.1 was heterozygous for a truncating mutation, c.1182C>G, p.Tyr394*. Patient presented with metastatic adenocarcinoma likely from a hepatic cholangiocarcinoma. Cancers reported in the family were pancreatic (II.2), CM (II.1), ovarian (II.5), mesothelioma (II.3), unknown (II.4, II.3), and nonmelanoma skin cancer (III.2). [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

Figure 2.

Mutations detected and BAP1 expression in tumors. (A) The identified mutations in the three families. Heterozygous mutations were detected in the germline of the three families and in the tumors from FUM064/III-12 and FUM103/III-1. PB: peripheral blood and T: tumor. All chromatograms are utilizing forward primer. (B) BAP1 immunostaining in the tumor of FUM064/III-12 show loss of nuclear staining of tumor cells (thick arrow) with positive staining of the nuclei in the stromal cells (thin arrows). (C) BAP1 immunostaining in the tumor of FUM103/III-1 show strong cytoplasmic expression of BAP1 with loss of nuclear localization in the tumor cells (thick arrow) with positive staining of the nuclei in the stromal cells (thin arrows). [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

TABLE 1

Germline *BAP1* Mutations and Variants Identified in the Study

FUM	cDNA location	Protein location	Location	MAF/MAF count ^a	dbSNP ID	Notes
FUM064	c.2050C>T	p.Gln684*	Exonic	Not reported	–	Reported as pathogenic mutation
FUM084	c.2057-4G>T		Intronic	A = 0.005/10	rs149499021	VUS ^b
FUM144	c.2057-4G>T		Intronic	A = 0.005/10	rs149499021	VUS
FUM077	c. 932-58_59delTG		Intronic	Not reported	–	VUS, not reported
FUM104	c.1882_1885delTCAC	p.Ser628Profs*8	Exonic	Not reported	–	Frameshift truncating mutation
FUM089	c.2057-22A>C		Intronic	G = 0.001/3	rs144083199	VUS
FUM089	c.*45C>G		Intronic	C = 0.030/65	rs56898787	VUS
FUM103	c.1182C>G	p.Tyr394*	Exonic	Not reported	–	Truncating mutation

^aMAF/MAF count: minor allele frequency/minor allele frequency count 1,000 genome.

^bVUS: variant of uncertain significance.