Germline testing in hereditary cancer genes subsequent to the identification of mutations in tumor specimens (Abstract No:1527)

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BACKGROUND

- Molecular profiling of tumors by Next Generation Sequencing (NGS) is readily available and is proving to be crucial in identifying targeted therapies.
- Molecular profiling of tumors has the potential to reveal an underlying germline mutation leading to the diagnosis of a hereditary cancer predisposition.
- A clinical diagnostic laboratory has observed an increasing number of requests for germline genetic testing related to a mutation found on molecular tumor profiling.
- Presently, it can be difficult to accurately predict the likelihood that a tumor mutation in a known cancer predisposition gene will be in the germline.
- Allele frequency and paired tumor/germline testing may be helpful in discerning somatic versus germline origin, but this information is often unavailable.

STUDY AIMS

- We sought to determine the frequency and clinical significance of germline testing subsequent to tumor profiling and to investigate whether patients who underwent germline testing would have been appropriate candidates for hereditary gene testing based on National Comprehensive Cancer Network (NCCN) or other consortium testing criteria.

METHODS

- A laboratory database and test requisitions were reviewed retrospectively. Age at cancer diagnosis, family history and a description of the patient's tumor were recorded.
- Tumor sequencing results from outside laboratories were reviewed along with associated notes on test requisition forms for the following genes: *APC, ATM, BARD1, BRCA1, BRCA2, BRIP1, BMPR1A, CDH1, CDK4, CDKN2A, CHEK2, EPCAM, FH, FLCN, MAX, MET, MITF, MLH1, MRE11A, MSH2, MSH6, NBN, NF1, PALB2, PMS2, PTEN, RAD50, RAD51C, RAD51D, RET, SDHA, SDHAF2, SDHB, SDHC, SDHD, SMAD4, STK11, TMEM127, TP53, TSC1, TSC2*, and *VHL*.
- Germline targeted sequencing by single site analysis, single gene testing, or multi-gene panel testing was performed as ordered by referring clinicians using DNA isolated from blood or saliva.
- For cases involving mutations in genes with available clinical testing criteria, clinical histories were reviewed to determine whether testing criteria were met.

Table 1: GERMLINE MUTATIONS

Gene	# Mutations from Tumors	# Confirmed in Germline (% met testing criteria)
APC	3	0
ATM	2	0
BRCA1	4	3 (100%)
BRCA2	12	4 (25%)
CDH1	4	0
CDKN2A	3	0
CHEK2	1	1 (NC)
MEN1	6	0
MLH1	1	0
MSH2	2	0
MSH6	2	0
NF1	6	1 (100%)
PTEN	6	1 (100%)
RB1	2	0
STK11	1	0
TP53	18	0
VHL	1	0

NC – no criteria available

TABLE 2: *BRCA2* POSITIVE PATIENTS NOT MEETING TESTING CRITERIA

	Alterations Detected in Tumor**	Tissue/Tumor Tested	Age	Gender	Test Ordered	Family History	Germline Mutation	Other Germline Alteration
Case 1	*BRCA2* S1982fs*22; *TP53* R342Q	Uterus/abdominal desmoplastic small round cell (DSRCT)	10-20 yrs	Female	*BRCA2* SSA and *TP53* SSA	None reported	*BRCA2* c.5946delT	*TP53* p.R342Q VUS
Case 2	*BRCA2* T2746fs*17; loss of *PTEN* exon 2	Breast/unknown primary melanoma	30-40 yrs	Female	*BRCA2* SSA	Multiple BCC and Breast cancer at > 50	*BRCA2* c. 8237_8238delCA	N/A
Case 3	*BRCA2* S1982fs*22; TP53 S149fs*21 ; loss of *PTEN* gene	Rectum/adenocarcinoma	30-40 yrs	Male	*BRCA2* SSA	None reported	*BRCA2* c.5946delT	N/A

**Nomenclature used in reporting tumor mutations may differ from what is reported in the germline. However, the mutations here are equivalent.
SSA - Single site analysis; **BCC** – Basal cell carcinoma, **VUS** – variant of uncertain significance

RESULTS

- Ten of 74 (13%) tumor mutations were confirmed in the germline. The remaining mutations are presumed to be somatic. (Table 1)
- Germline mutations were identified in 5 of 17 genes with the majority of germline findings in *BRCA1* (3) and *BRCA2* (4).
- Although *TP53* alterations were most frequently submitted for germline analysis, none were confirmed in the germline, despite 3/18 patients meeting National Comprehensive Cancer Network (NCCN) criteria for *TP53* germline testing.
- Three patients who did not meet NCCN testing criteria, based on available clinical history, carried a *BRCA2* germline mutation. (Table 2)

TAKE-HOME POINTS

- Mutations identified in tumor tissue that occur in genes known to be associated with cancer predisposition such as *BRCA1/2,* may occur sporadically in the tumor or be carried in the germline.
- Tumor profiling may result in detection of germline mutations in patients who otherwise may not have been referred for hereditary gene testing, having a significant impact on medical management for the patient and family.
- *TP53* tumor mutations are rarely an indicator for germline *TP53* testing

REFERENCE

- National Comprehensive Cancer Network. NCCN Clinical Practice Guidelines in Oncology: Genetic/Familial High-Risk Assessment: Breast and ovarian Version 1.2015 Accessed 02/03/2015 at www.nccn.org

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