Cancer Genetics 209 (2016) 130–137

Cancer Genetics

ORIGINAL ARTICLE

Increased yield of actionable mutations using multi-gene panels to assess hereditary cancer susceptibility in an ethnically diverse clinical cohort

Charité Ricker [a,b,*,1], Julie O. Culver [a,1], Katrina Lowstuter [a], Duveen Sturgeon [a], Julia D. Sturgeon [a], Christopher R. Chanock [a], William J. Gauderman [c], Kevin J. McDonnell [a,b], Gregory E. Idos [a,b], Stephen B. Gruber [a,b,c]

[a] *USC Norris Comprehensive Cancer Center, University of Southern California, Los Angeles, CA 90033, USA;* [b] *Department of Medicine, University of Southern California, Los Angeles, CA 90033, USA;* [c] *Department of Preventive Medicine, University of Southern California, Los Angeles, CA 90033, USA*

This study aims to assess multi-gene panel testing in an ethnically diverse clinical cancer genetics practice. We conducted a retrospective study of individuals with a personal or family history of cancer undergoing clinically indicated multi-gene panel tests of 6–110 genes, from six commercial laboratories. The 475 patients in the study included 228 Hispanics (47.6%), 166 non-Hispanic Whites (35.4%), 55 Asians (11.6%), 19 Blacks (4.0%), and seven others (1.5%).

Panel testing found that 15.6% (74/475) of patients carried deleterious mutations for a total of 79 mutations identified. This included 7.4% (35/475) of patients who had a mutation identified that would not have been tested with a gene-by-gene approach. The identification of a panel-added mutation impacted clinical management for most of cases (69%, 24/35), and genetic testing was recommended for the first degree relatives of nearly all of them (91%, 32/35). Variants of uncertain significance (VUSs) were identified in a higher proportion of tests performed in ethnic minorities.

Multi-gene panel testing increases the yield of mutations detected and adds to the capability of providing individualized cancer risk assessment. VUSs represent an interpretive challenge due to less data available outside of White, non-Hispanic populations. Further studies are necessary to expand understanding of the implementation and utilization of panels across broad clinical settings and patient populations.

Keywords Hereditary cancer, cancer risk assessment, multi-gene panels, variants of uncertain significance, ethnic minorities

Over the last twenty years, hereditary cancer risk assessment has evaluated individuals using a syndrome-by-syndrome and gene-by-gene testing approach. After the formation of a differential diagnosis, the most probable condition has been routinely evaluated first with subsequent analyses performed sequentially, guided by clinical judgment and often by insurance coverage and patient motivation. However, the rapid integration of next-generation sequencing (NGS) technologies into the practice of clinical cancer genetics has allowed for simultaneous assessment of multiple syndromes and genes in one analysis.

An emerging body of literature has begun to report on findings from these multi-gene assays in the research, clinical, and laboratory settings. The cohorts studied have included mostly White, non-Hispanic populations and have varied from large laboratory based reports (1–4) to small highly-selected clinical cohorts (5–8). Additionally, registry based cohorts have looked at non-*BRCA* gene mutations in previously tested negative patients (3,9,10). While these studies have varied in their methodologies and assays, most have found that a panel

Received August 13, 2015; received in revised form November 13, 2015; accepted December 31, 2015.

* Corresponding author.
E-mail address: Charite.Ricker@med.usc.edu

[1] These authors contributed equally.

2210-7762/$ - see front matter © 2016 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.cancergen.2015.12.013

testing approach identifies mutations that would not have been identified with a syndrome-by-syndrome approach as well as many variants of uncertain significance (VUSs) (1–4,9–11).

The additional yield of mutations identified via multi-gene panel testing in a clinical setting has not yet been well defined, especially among under-represented minority populations. Despite the ethnic and racial diversity of the US population, to date, all of the published cohorts are predominately Caucasian, of non-Hispanic ancestry. We report on the largest multi-ethnic cohort to-date of individuals evaluated for mixed clinical indications by twenty different laboratory assays offered by six CLIA-approved commercial laboratories. In this retrospective study we assessed the additional yield of panel testing by cancer site as well as the clinical characteristics of mutation carriers in a diverse clinic population.

Materials and methods

Participants

Study participants were seen for clinical cancer genetic counseling at two University of Southern California (USC) Cancer Genetics sites: USC Norris Comprehensive Cancer Center and the Los Angeles County + USC Medical Center. A retrospective IRB-approved chart review was conducted of 475 cancer genetics clinic patients who underwent genetic testing with a multi-gene hereditary cancer panels. Individuals were eligible for inclusion if a multi-gene panel was ordered from any laboratory on or before 7/14/14. Genetic test results and clinicopathologic characteristics of each patient were reviewed. All patients received pre- and post-test genetic counseling from genetics professionals.

Genetic testing multi-gene panels

Tests were selected on the basis of clinical indication and insurance coverage. Panel tests ordered in the participants were performed by Myriad Genetics (n = 354), Ambry Genetics (n = 100), Fulgent Diagnostics (n = 17), University of Washington Genetics Lab (n = 2), City of Hope Molecular Diagnostics Laboratory (n = 1), and Baylor Genetics Laboratory (n = 1). Ambry panels included CancerNext™ (n = 62), OvaNext™ (n = 6), BreastNext™ (n = 10), ColoNext™ (n = 11), PGLNext™ (n = 1), RenalNext™ (n = 3), and BRCAPlus™ (n = 7); from all other labs, only one hereditary cancer panel was ordered. Some panels had more than one version as laboratories added more genes to existing panels. Genes included on panels ordered are detailed in the Supplementary material. All laboratories utilized in this cohort report variants of uncertain significance.

Review of mutations and variants

Each case was discussed at the time of assessment in a multidisciplinary cancer genetics case conference, and a differential diagnosis was formed. For each deleterious mutation or suspected deleterious mutation identified on a panel, the mutation was then classified as being either as a “target-gene” or a “panel-added” mutation. If the gene was included in the differential diagnosis and would have been part of a syndrome-by-syndrome testing approach, then the mutation was categorized as being in a “target-gene.” If the gene would not have been tested using a syndrome-by-syndrome approach, then the mutation was categorized as “panel-added.” *MUTYH* mutations were considered “target-gene” mutations in colon cancer cases or families with colon cancer. *PALB2* was considered a target gene if there was a combination of breast and pancreatic cancer in the family. *TP53* was considered a target gene in breast cancer diagnosed under age 35. *CDH1* was considered a target gene if the hereditary diffuse gastric cancer testing criteria were met or in a patient with lobular breast cancer with any gastric cancer in the family. Mutations in genes that were not routinely targeted prior to the clinical availability of panels, such as *CHEK2*, *ATM*, *BARD1*, *RAD51C,* and others, were all considered “panel-added” mutations.

The number of variants of uncertain significance (VUSs) detected for each patient was counted. Any variants that were reported as “likely polymorphisms” were omitted from this analysis. If the reporting laboratory reclassified a variant before 12/31/14, the updated classification was used for this analysis.

The mutation and variants reported in our analysis are all described in accordance with the interpretation provided by lab report. Public databases were not routinely searched to further interpret the laboratory classification. However, the clinical interpretation was altered in two cases. One patient with a *MSH2* VUS was counseled mutation-positive due to personal and family phenotype of Lynch syndrome and an informative segregation analysis. One patient, known to have a 56 base pair deletion in *BRCA1*, underwent a panel test with a different laboratory due to suspicion of Lynch syndrome, but the NGS panel was unable to detect the previously identified *BRCA1* mutation.

Statistical analysis

Using STATA (*Stata Statistical Software: Release 13*, College Station, TX: StataCorp LP) the odds ratio of detecting a mutation by gender, race/ethnicity, and whether the proband had a diagnosis of cancer was determined. The odds ratio of detecting a VUS was also determined for each of these three predictors.

The correlation between the number of genes on a panel and the likelihood of identifying a variant of uncertain significance was measured using simple linear regression to estimate the slope and to measure R^2. Seven panel groups were created by collapsing the 14 panels with multiple versions from the same laboratory into the same group. The panel groups included, Ambry BRCAplus™ v1, Ambry ColoNext™ v1, Ambry BreastNext™ (3 versions), Ambry OvaNext™ (3 versions), Ambry CancerNext™ (3 versions), Myriad myRisk™ v1, and Fulgent (2 versions); an average number of genes tested was determined for each panel group. Ambry PGLNext,™ Ambry RenalNext™, the Baylor High Risk Colorectal Cancer Panel, the City of Hope Molecular Diagnostic Lab Breast Cancer Susceptibility Panel, and the two University of Washington BROCA™ panels were excluded because they were used infrequently and there was very little overlap of the composition of the panels. The proportion of panels with a mutation identified or with a variant of uncertain significance was determined for each panel group. Data were analyzed separately for mutations and for VUSs. This analysis was performed both