Unique Genomic Landscape of High-Grade Neuroendocrine Cervical Carcinoma: Implications for Rethinking Current Treatment Paradigms

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PURPOSE High-grade neuroendocrine cervical cancer (HGNECC) is an uncommon malignancy with limited therapeutic options; treatment is patterned after the histologically similar small-cell lung cancer (SCLC). To better understand HGNECC biology, we report its genomic landscape.

PATIENTS AND METHODS Ninety-seven patients with HGNECC underwent comprehensive genomic profiling (182-315 genes). These results were subsequently compared with a cohort of 1,800 SCLCs.

RESULTS The median age of patients with HGNECC was 40.5 years; 83 patients (85.6%) harbored high-risk human papillomavirus (HPV). Overall, 294 genomic alterations (GAs) were identified (median, 2 GAs/sample; average, 3.0 GAs/sample, range, 0-25 GAs/sample) in 109 distinct genes. The most frequently altered genes were PIK3CA (19.6% of cohort), MYC (15.5%), TP53 (15.5%), and PTEN (14.4%). RB1 GAs occurred in 4% versus 32% of HPV-positive versus HPV-negative tumors (P < .0001). GAs in HGNECC involved the following pathways: PI3K/AKT/mTOR (41.2%); RAS/MEK (11.3%); homologous recombination (9.3%); and ERBB (7.2%). Two tumors (2.1%) had high tumor mutational burden (TMB; both with MSH2 alterations); 16 (16.5%) had intermediate TMB. Seventy-one patients (73%) had ≥ 1 alteration that was theoretically druggable.

Comparing HGNECC with SCLC, significant differences in TMB, microsatellite instability, HPV-positive status, and in PIK3CA, MYC, PTEN, TP53, ARID1A, and RB1 alteration rates were found.

CONCLUSION This large cohort of patients with HGNECC demonstrated a genomic landscape distinct from SCLC, calling into question the biologic and therapeutic relevance of the histologic similarities between the entities. Furthermore, 73% of HGNECC tumors had potentially actionable alterations, suggesting novel treatment strategies for this aggressive malignancy.

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INTRODUCTION

The treatment of solid malignancies has evolved and is perhaps best exemplified by the approach to non–small-cell lung cancer, for which molecular characterization and use of targeted agents have emerged as standard therapeutic paradigms. Recently, The Cancer Genome Atlas (TCGA) completed and published the integrated genomic and molecular characterization of cervical cancer.1 In addition to data previously released for both ovarian (high-grade serous) and endometrial (endometrioid and serous) cancers, this publication completed the molecular and genomic evaluation of the most common gynecologic malignancies.2,3

Traditionally, cervical cancer clinical trials have excluded less common histologies such as high-grade neuroendocrine cervical carcinoma (HGNECC). Despite the low incidence of HGNECC (< 2% of all cervical cancers) the oncologic impact is significant because these tumors exhibit more aggressive clinical characteristics.4,5 Unfortunately, the 5-year overall survival rate for patients with early-stage disease is only 36%, and those with metastatic spread face an even more dismal prognosis. Given these poor outcomes, patients with HGNECC represent an area of unmet clinical need.

Developing therapeutic options for patients with rare tumors is challenging, relying on international collaboration, as well as small case series or retrospective reports rather than prospective clinical trials. The current therapeutic paradigm for the treatment of HGNECC was adopted from the more common, morphologically similar, small-cell lung cancer (SCLC) and includes surgical resection if feasible, followed by platinum plus etoposide-based combination chemotherapy, and possibly radiation.6,7 There are few studies informing treatment of recurrent disease, and there are no drugs
High-grade neuroendocrine cervical cancer appears molecularly distinct from the histologically similar small-cell lung cancer. Up to 73% of patients’ samples harbored potentially actionable alterations, informing novel treatment strategies.

Relevance
Continued understanding of the molecular underpinnings of high-grade neuroendocrine cervical carcinoma will be critical to driving drug discovery for this disease.

CONTEXT
Key Objective
To define the molecular landscape of high-grade neuroendocrine cervical cancer in a large cohort of patients.

Knowledge Generated
High-grade neuroendocrine cervical cancer appears molecularly distinct from the histologically similar small-cell lung cancer.

Relevance
Continued understanding of the molecular underpinnings of high-grade neuroendocrine cervical carcinoma will be critical to driving drug discovery for this disease.

PATIENTS AND METHODS
We evaluated a fully informative genomic profile of patients diagnosed with poorly differentiated (G3) neuroendocrine cervical carcinomas inclusive of both small- or large-cell subtypes (HGNECCs) whose cancers were submitted for hybrid capture–based next-generation sequencing (NGS) testing from March 2013 to December 2017 (N = 97). A cohort of 1,800 similarly tested cases of SCLC from the same period were subsequently evaluated to allow for comparison of genomic alterations (GAs). The submitting physicians provided specification of a poorly differentiated, neuroendocrine tumor type of cervical origin, which was then independently reviewed by a gynecologic pathologist (J.E.) to confirm high-grade neuroendocrine pathologic features in the pathology report and/or the representative sample of tumor submitted for sequencing (grade 3 cytomorphic features, some component of small-cell or large-cell carcinoma histology, and/or positivity for neuroendocrine markers). The database was de-identified with only the diagnosis available. NGS data were generated by FoundationOne (Foundation Medicine; Cambridge, MA). The study was performed in accordance with University of California, San Diego, Institutional Review Board guidelines for a de-identified database. Approval for this study, including a waiver of informed consent and a Health Insurance Portability and Accountability Act waiver of authorization, were also obtained from the Western Institutional Review Board (Protocol No. 20152817).

Tissue Samples and Mutational Analysis
Available tissue from diagnostic or therapeutic procedures was used to determine oncogenic molecular alterations. Sequencing information was collected on 97 patients with HGNECC and 1,800 with SCLC, whose formalin-fixed, paraffin-embedded tumor samples were submitted to Foundation Medicine for genomic profiling. The test sequences the entire coding region of 182 or, more recently, 236 or 315 cancer-related genes plus up to 47 introns of up to 19 genes often rearranged or altered in cancer to an average depth of coverage of > 500x. The pathologic diagnosis of each case was confirmed on routine hematoxylin- and eosin-stained slides and all samples forwarded for DNA extraction contained a minimum of 20% tumor nuclear area. Microsatellite instability (MSI) status was evaluable in 75 HGNECC and 1,573 SCLC cases.

The sequencing methods used for comprehensive genomic profiling have been validated and reported previously (Appendix). The optimized loci used to evaluate MSI status were selected from a total set of 1,897 that have adequate coverage on all versions of the assay. Each locus is intrinsic and has a reference repeat length of 10-20 bp, which allows for analysis with the read length used by FoundationOne testing. Principal components analysis is used to produce an NGS-based MSI score. There was no need to extend beyond the first principal component, because it explained approximately 50% of the total data variance, whereas none of the other principal components explained > 4% each. Ranges of the MSI score were assigned MSI-High (MSI-H), MSI ambiguous, or microsatellite stable (MSS). MSI-Low calls are not made because there was no gold-standard test set, but we presume such samples would significantly overlap with the MSI-ambiguous category reported here. For samples in which MSI-specific quality control criteria were not met (n = 22 HGNECC; n = 227 SCLC), a status of MSI unknown was assigned, and these cases were excluded from additional MSI analysis.

Tumor Mutational Burden
The number of somatic mutations detected on NGS (interrogating up to 1.2 Mb of the genome) were quantified
and that value extrapolated to the whole exome, using a validated algorithm. Alterations likely or known to be germline polymorphisms or bona fide oncogenic drivers were excluded. Tumor mutational burden (TMB) was measured in mutations per megabase. TMB levels were grouped into 3 bins: TMB-low (TMB-L; 1-5 mutations/Mb), intermediate (TMB-I; 6-19 mutations/Mb), and high (TMB-H; ≥ 20 mutations/Mb). The cutoff of 20 coding mutations/Mb is approximately equal to 400 nonsynonymous mutations per exome. 

Human Papillomavirus Detection

In addition, the presence of high-risk human papilloma-virus (HPV) was examined in submitted specimens, as previously reported. Hybrid-capture reagents included baits designed to capture unique regions of select viral genomes including HPV-16 and -18. Sequence read pairs were aligned to the reference genome of the respective viral genomes, and the number of pairs mapping to each viral genome was counted. A total HPV-16/18 aligned read count of ≥ 5 reads per million was considered a positive HPV status, and < 5 reads per million was considered HPV not detected.

End Points and Statistical Methods

Descriptive statistics were used to summarize the baseline patient characteristics. Fisher exact test was used to determine the association between categorical variables in univariate analysis and the Z-test was used to assess population differences, where appropriate. All tests were 2 sided. All statistical tests were carried out using GraphPad Prism, version 6.0 (GraphPad Software, San Diego, CA).

RESULTS

Characterization of GAs in HGNECC

The median age of the cohort was 40.5 years (range, 25-77 years). Of the 97 patients, 83 were high-risk HPV positive (85.6%) and 14 were negative (14.4%). All samples were reflective of HGNECC, including both small-cell and large-cell HGNECC cases. Among the HGNECC cohort (N = 97), the most frequently identified GAs (discerned in > 10% of the cohort) involved PIK3CA (19.6% of patients), MYC (15.5%), TP53 (15.5%), and PTEN (14.4%) (Fig 1; a detailed list of all GAs can be found in Appendix Table A1). A total of 109 different genes were mutated in the 97 patient samples evaluated (variants of unknown significance were excluded from all analyses). The most frequently reported number of GAs per sample was 2, with a range of 0-25 (average, 3.0 GAs/sample; Fig 2). When evaluating TMB, 2 cases (2.1%) were TMB-H and 16 cases (16.5%) were TMB-I. Most patients’ tumors (n = 79; 81.4%) were TMB-L (Table 1). Nine patient samples had no known or likely GAs on comprehensive genomic profiling. Of the 88 patients who had an alteration, 72 had at least 1 alteration for which there currently existed an agent potentially targeting that alteration.
cases of HGNECC with evaluable microsatellite status (2.7%).

**Less Frequent GAs**

Additional genomic characterization was performed in which we specifically explored the homologous recombination deficiency (HRD), RAS, PI3K/AKT/mTOR, and ERBB pathways. Nine cases (9.3%) were had GAs in HRD-related genes, with the most frequent alterations noted in BRCA2 (n = 6 of 9; 66.7%).17,19 Three additional patient samples had BRCA1, ATM, and PALB2 mutations (n = 1 in each case) case (n = 3 of 9; 33.3%).

Eleven patient samples (11.3%) had alterations in the RAS pathway, with KRAS and BRAF mutations being the most frequent (72.7% [n = 8 of 11] and 27.3% [n = 3 of 11], respectively). Of the identified BRAF mutations, only 1 was a V600E alteration. Furthermore, a total of 40 patient samples (41.2%) harbored mutations in the PI3K/AKT/mTOR pathway; mutations in PIK3CA were identified in 47.5% of these samples (n = 19), and PTEN mutations were reported in 35% (n = 14). Last, 7 patients (7.2%) had mutations in the ERBB pathway, with ERBB2 mutations occurring in tumors of 4 individuals (57.1%).

**Comparison of HGNECC and SCLC**

Given the histologic similarity between SCLC and HGNECC, tumor samples from a cohort of 1,800 patients with SCLC were compared with the HGNECC samples (Table 2). The SCLC samples featured significantly lower frequencies of GAs in PIK3CA, MYC, and ARID1A. In contrast, the HGNECC samples featured significantly lower frequencies of GAs in TP53 and RB1. High-risk HPV was identified in much less than 1% of SCLC tumor samples compared with 85.6% of HGNECC tumor samples. There was a single MSI-H SCLC case (n = 1 of 1,449; 0.001%), whereas MSI-H status was found in 2 HGNECC cases (2.7%). Last, TMB was significantly higher in the SCLC samples compared with the HGNECC samples with respect to both intermediate and high TMB levels. The small-cell subset of HGNECC samples showed analogous gene mutation differences from SCLC.

**DISCUSSION**

Neuroendocrine carcinoma is an uncommon but aggressive variant accounting for approximately 1.5% of all newly diagnosed cervical cancers.20 The great majority of these lesions are high-grade large- or small-cell subtypes, with only rare reports of well-differentiated cervical carcinoid tumors.20 The treatment of patients with HGNECC remains an evolving field, with the identification and validation of therapeutic targets becoming increasingly important.

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**TABLE 1. Molecular Features of High-Grade Neuroendocrine Cervical Cancers**

| Altered Gene | Total HPV Positive (n = 83) | HPV Negative (n = 14) | P  |
|--------------|---------------------------|----------------------|----|
| PIK3CA       | 19.6                      | 17                   | 36 | .0037|
| MYC          | 15.5                      | 17                   | 7  | .0484|
| TP53         | 15.5                      | 11                   | 43 | .0001|
| PTEN         | 14.4                      | 8                    | 50 | .0001|
| ARID1A       | 9.3                       | 5                    | 36 | .0001|
| RB1          | 8.2                       | 4                    | 36 | .0001|

**NOTE.** The No. (%) of low (1-5 mutations/Mb), intermediate (6-19 mutations/Mb), and high (≥20 mutations/Mb) tumor mutational burdens were as follows: patients with ≥1 HPV-positive oncogenic alteration: 70 (84), 12 (14.4), and 1 (1.2), respectively; and for patients with ≥1 HPV-negative oncogenic alteration: 9 (63.3), 4 (28.6), and 1 (7.1), respectively (P = .13).

**TABLE 2. Comparison of Clinical and Molecular Features of HGNECC, Cervical Small-Cell Carcinoma, and SCLC**

| Feature          | HGNECC (N = 97) | Cervical Small Cell (n = 79)* | SCLC (n = 1,800) | Cervical Small Cell v SCLC, P | HGNECC v SCLC, P |
|------------------|-----------------|------------------------------|------------------|-----------------------------|------------------|
| Median age, years (range) | 40.5 (25-77) | 40.5 (24-73) | 64 (10-89) | .001 | .0001 |
| Genomic alterations/case | 3.0 | 3.0 | 4.6 | NS | NS |
| PIK3CA           | 19.6           | 24.0                        | 5.1              | .0001 | .0001 |
| MYC              | 15.5           | 12.7                        | 6.3              | .0001 | .0001 |
| TP53             | 15.5           | 12.7                        | 90.1             | .0001 | .0001 |
| PTEN             | 14.4           | 13.9                        | 8.9              | NS | NS |
| ARID1A           | 9.3            | 10.1                        | 4.2              | .012 | .01778 |
| RB1              | 8.2            | 6.3                         | 70.9             | .0001 | .0001 |
| MSI-Highb       | 2.7            | 3.1                         | 0.004            | .0001 | .0001 |
| TMB ≥ 6-19 mutations/Mb | 16.5      | 15.2                        | 62.3             | .0001 | .0001 |
| TMB ≥ 20 mutations/Mb | 2.1         | 2.5                         | 8.2              | .0767 | .030 |
| HPV-16/18 positive | 85.6         | 87.0                        | 0.01             | .0001 | .0001 |

**NOTE.** Data reported as % unless otherwise indicated.

Abbreviations: HGNECC, high-grade neuroendocrine cervical cancer; HPV, human papillomavirus; MSI, microsatellite instability; NS, not significant; SCLC, small-cell lung cancer; TMB, tumor mutational burden.

*The 79 patients with cervical small-cell cancers were a subset of the 97 patients with HGNECC.

**DISCUSSION**

Neuroendocrine carcinoma is an uncommon but aggressive variant accounting for approximately 1.5% of all newly diagnosed cervical cancers.20 The great majority of these lesions are high-grade large- or small-cell subtypes, with only rare reports of well-differentiated cervical carcinoid tumors.20 The treatment of patients with HGNECC remains an evolving field, with the identification and validation of therapeutic targets becoming increasingly important.
clinically challenging, with limited response rates to chemotherapy; however, anecdotal reports of exceptional responders have been described.9,10

The paradigm for management of HGNECC has been informed by the treatment of the more commonly diagnosed (and histologically similar) SCLC, which accounts for approximately 15% of all lung cancer cases. In prior studies, whole-genome sequencing of 110 SCLC specimens identified essentially ubiquitous *TP53* and *RB1* inactivating mutations, with biallelic losses of each gene respectively in 100% and 93% of cases without chromothripsis.21

In an effort to better define the molecular landscape of HGNECC, we evaluated the comprehensive genomic profiling of 97 patient samples. The most frequently identified GA was *PIK3CA* mutation, occurring in 19.6% of submitted samples (n = 19). At least 1 characterized alteration was identified in 88 patient samples (90.7%) and of these, 72 had a potentially pharmacologically tractable alteration.

Interestingly, the frequency and distribution of GAs identified in this cohort of patients are similar and distinct from mutational patterns described in the more common HPV-related cervical cancer histologies.1 As detailed in TCGA’s integrated genomic characterization of cervical cancer (ie, squamous, adenocarcinoma, and adenosquamous histologies), mutations in the *PIK3CA* gene were the most frequently identified aberration, occurring in 26% of samples, approximating the nearly 20% rate in our cohort. In addition, significantly mutated genes reported by the TCGA, identified in similar proportions in this patient cohort, included *ARID1A* (7% in TCGA and 9.3% in our cohort) and *KRAS* (6% in TCGA and 8.2% in our cohort). Conversely, the examined neuroendocrine cohort had a greater frequency of *PTEN* mutations (8% in TCGA v 14.4% in our cohort). These molecular differences may be reflective of the varying histologies or, potentially, the differential high-risk HPV detection rates (85.6% in our cohort v 95% in the TCGA).1

Importantly, the high-risk HPV rate in our cohort should be interpreted with caution because the assay used has not undergone formal concordance study with gold standard tests such as hybrid capture and can detect only HPV 16/18.

Our own, much larger cohort of SCLC samples (n = 1,800) recapitulates prior studies and had a strikingly different molecular portfolio when compared with HGNECC samples. The frequency of *TP53* and *RB1* alterations in the SCLC cohort significantly exceeded that seen in our HGNECC cohort (15.5% and 8.2%, respectively), the HPV16/18 positive subset (11% and 4%, respectively), and the subset where HPV16/18 was not detected (43% and 36%, respectively; Table 2). Furthermore, mutations affecting the *NOTCH* pathway were identified in 25% of the examined SCLC samples; the *NOTCH* pathway is hypothesized to function as a regulator of neuroendocrine differentiation. In our examined HGNECC cohort, only 7 patients (7.2%) had *NOTCH* alterations. Alterations in *PIK3CA*, *MYC*, and *PTEN* were significantly more common in HGNECC when compared with SCLC (Table 2). MSI-H status was also more common in the HGNECC cohort whereas TMB-H was more common in SCLC (despite the lack of MSI-H status). Finally, HPV positivity was discerned in 85.6% of our HGNECC samples, but in only 0.01% of our SCLC samples (P < .0001). No parallels in molecular alterations were identified when comparing our findings for HGNECC with those of prior SCLC studies, supporting our premise that the similarity between these entities is largely morphologic and the treatment approaches for HGNECC can likely be improved through improved molecular granularity.

Despite the infrequency of HGNECC, the identification of potentially actionable GAs may inform treatment of a subset of patients with historically limited therapeutic options.18,22–25

In this cohort of patients, alterations in the PI3K/AKT/mTOR pathway were commonly seen (*PIK3CA* [19.6%]; *PTEN* [14.4%]). The use of everolimus, or an alternate mTOR or *PIK3CA* inhibitor, may be considered in such circumstances, although the utility of a *PIK3CA* mutation in predicting response to single-agent everolimus in the presence of multiple GAs remains limited.26,27

Although less frequently identified, alterations in the HRD pathway were detected in 9.3% of patient samples, potentially supporting use of a poly-ADP ribose polymerase inhibitor. The identification of both TMB-H (n = 2) and GAs in mismatch repair genes (n = 3) may also inform the use of immune checkpoint inhibition.28 In May 2017, the FDA approved pembrolizumab for the treatment of mismatch repair–deficient or MSI-H solid tumors that progressed after prior therapy. This disease site–agnostic approval allows for a promising therapeutic option for patients with a previously unmet clinical need. More recently, the FDA accepted and granted priority review to a supplemental Biologics License Application for pembrolizumab for the treatment of adult and pediatric patients with unresectable or metastatic solid tumors with tissue TMB-H whose disease has progressed after prior treatment and who have no satisfactory alternative treatment options, supported by data from the phase II Keynote-158 trial. Notably, there are 2 published case reports of patients with recurrent, treatment-refractory HGNECC with exceptional and durable responses to checkpoint inhibition; 1 of these tumors was from our current HGNECC cohort and had a mismatch repair defect and the other lacked correlative genomic testing.9,10

Last, the identification of *ARID1A* (9.3%) and *SMARCA4* (4.1%) mutations may predict sensitivity to an alternate therapeutic strategy.29 Homeostasis requires balanced *ARID1A* and *EZH2* activity, facilitated via chromatin-mediated gene expression. Loss of *ARID1A* expression results in imbalanced *EZH2* activity, and use of an *EZH2* inhibitor such as tazemetostat may capitalize on this oncogene addiction. Importantly, 2 of the 4 SMARCA4
aberrations were identified in patients with MSI-H lesions, possibly reflecting that the SMARCA4 may be a passenger mutation resulting from the underlying MSI. Furthermore, of the 4 cases with SMARCA4 alterations, 1 was HPV-18 positive and another was p16 positive by immunohistochemical assessment.

Despite the large sample size and robust genomic data, this study has limitations. The retrospective design and use of archival tumor tissues from various time points during therapy may make interpretation of GAs difficult. In addition, the lack of demographic and clinical data, as well as treatment history, precludes exploratory assessments of response to a selected targeted agent. Last, HPV status was determined using molecular surrogates that differ from the assays used in clinical practice. It remains unclear if HPV infection is a prerequisite for neuroendocrine cervical carcinoma, although recent publications suggest > 85% of neuroendocrine cervical carcinomas are HPV positive, with HPV-16 and HPV-18 accounting for > 95% of the identified high-risk HPV strains.30

This report highlights the potential therapeutic utility of genomic testing in patients with this uncommon disease.27 Of interest, despite the histologic similarity between HGNECC and SCLC, which has led to the latter being used as a model for treating the former, the molecular portfolio of these 2 entities is strikingly different. Therefore, it is plausible that patients with HGNECC may benefit from alternative therapeutic strategies.

It is not anticipated that traditional prospective trials will accrue sufficient patient numbers in this disease setting, and novel study designs, including umbrella, basket, and platform trials, should be considered given the presence of actionable targets. Interestingly, the first reported cohort of the DART trial (ClinicalTrials.gov identifier: NCT02834013)31 was the neuroendocrine cohort, with a 44% overall response rate in those with high-grade disease. Ultimately, comprehensive genomic characterization may catalyze the investigation and identification of effective therapies, allowing us to improve oncologic outcomes in this aggressive disease.

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**APPENDIX**

**Sequencing Methods Used for Comprehensive Genomic Profiling**

Sample processing and sequencing were performed in a Clinical Laboratory Improvement Amendments– and College of American Pathologists–accredited laboratory. Briefly, after pathologic review to confirm sufficient tumor nuclei (minimum, 20%) and mitigate pathologic inconsistencies, at least 50 ng of DNA was extracted from 40 microns of tumor samples provided as formalin-fixed, paraffin-embedded tissue blocks. The samples were assayed using adaptoligation and hybrid-capture next-generation sequencing (FoundationOne) for all coding exons from 182 (version 1), 287 (version 2), or 315 (version 3) cancer-related genes plus selected introns from 14 (version 1), 19 (version 2), or 28 (version 3) genes frequently rearranged in cancer.

Sequencing of captured libraries was performed using HiSeq2500/4000 (Illumina, San Diego, CA) to a mean exon coverage depth of > 500×, and resultant sequences were analyzed using both an algorithmic pipeline and manual curation for base substitutions, small insertions or deletions (indels), copy number alterations (focal amplifications and homozygous deletions), and selected gene fusions, as previously described. Clinically relevant genomic alterations were defined as alterations targetable by anticancer drugs currently available on the market or in registered clinical trials. Germline variants documented in the dbSNP database (dbSNP142; http://www.ncbi.nlm.nih.gov/SNP/), with ≥ 2 counts in the ExAC database (http://exac.broadinstitute.org/), or recurrent variants of unknown significance that were predicted by an internally developed algorithm to be germline were removed, with the exception of known driver germline events.

Known, confirmed somatic alterations deposited in the Catalog of Somatic Mutations in Cancer (version 62) were highlighted as biologically significant, as were inactivating events in tumor suppressor genes. To maximize mutation-detection accuracy (sensitivity and specificity) in impure clinical specimens, the test was previously optimized and validated to detect base substitutions at a ≥ 5% mutant allele frequency (MAF), indels with a ≥ 10% MAF with ≥ 99% accuracy, and fusions occurring within baited introns/exons with > 99% sensitivity. Each tumor sample was analyzed alongside an internally validated mixture of 10 heterozygous diploid HapMap control samples, which custom algorithms used to normalize the sequence coverage distribution across baited targets.
TABLE A1. Detailed Genomic Assessment of High Grade Neuroendocrine Cervical Cancer Samples

| Case No.  | No. of Genes Assessed | Gene   | Age (years) | Functional Status | Alteration Type | Description                  |
|----------|------------------------|--------|-------------|-------------------|-----------------|------------------------------|
| HGNECC_1 | 236                    | AKT3   | 42          | Known             | CN              | Amplification                |
| HGNECC_1 | 236                    | KDM6A  | 42          | Known             | SV              | V607M                        |
| HGNECC_1 | 236                    | MCL1   | 42          | Known             | CN              | Amplification                |
| HGNECC_10| 236                    | ALK    | 40          | Known             | SV              | L560F                        |
| HGNECC_11| 315                    | FGFR2  | 44          | Known             | SV              | S252W                        |
| HGNECC_11| 315                    | MED12  | 44          | Known             | SV              | D23Y                         |
| HGNECC_11| 315                    | PTEN   | 44          | Known             | SV              | R130P                        |
| HGNECC_11| 315                    | RB1    | 44          | Known             | SV              | G442fs*15                     |
| HGNECC_11| 315                    | TP53   | 44          | Known             | SV              | R175H                        |
| HGNECC_12| 315                    | ARID1A | 66          | Likely            | RE              | Truncation                    |
| HGNECC_12| 315                    | TET2   | 66          | Known             | SV              | R1516*                       |
| HGNECC_13| 315                    | LRP1B  | 58          | Likely            | RE              | Deletion                      |
| HGNECC_13| 315                    | PTEN   | 58          | Known             | SV              | T319fs*1                     |
| HGNECC_14| 315                    | ARID1A | 59          | Likely            | SV              | N104fs*6                     |
| HGNECC_14| 315                    | ERBB2  | 59          | Known             | CN              | Amplification                 |
| HGNECC_14| 315                    | PIK3CA | 59          | Known             | SV              | E545A                        |
| HGNECC_14| 315                    | RB1    | 59          | Known             | SV              | R255*                        |
| HGNECC_14| 315                    | TOP2A  | 59          | Known             | CN              | Amplification                 |
| HGNECC_14| 315                    | TP53   | 59          | Known             | SV              | R158L                        |
| HGNECC_15| 315                    | BRCA1  | 58          | Likely            | SV              | Q780*                        |
| HGNECC_15| 315                    | IGF1   | 58          | Known             | CN              | Amplification                 |
| HGNECC_15| 315                    | MYC    | 58          | Known             | CN              | Amplification                 |
| HGNECC_15| 315                    | NOTCH2 | 58          | Known             | SV              | P66fs*27                     |
| HGNECC_15| 315                    | TP53   | 58          | Likely            | SV              | L330fs*15                    |
| HGNECC_16| 236                    | TP53   | 46          | Known             | SV              | D281N                        |
| HGNECC_17| 315                    | BRAF   | 40          | Known             | SV              | V600E                        |
| HGNECC_17| 315                    | CDK12  | 40          | Known             | SV              | G909R                        |
| HGNECC_17| 315                    | GRIN2A | 40          | Known             | SV              | R1318W                       |
| HGNECC_18| 315                    | MED12  | 34          | Known             | SV              | G44A                         |
| HGNECC_19| 315                    | CDH1   | 45          | Known             | SV              | W20*                         |
| HGNECC_19| 315                    | NFE2L2 | 45          | Known             | SV              | D29H                         |
| HGNECC_19| 315                    | NFE2L2 | 45          | Known             | SV              | R34Q                         |
| HGNECC_19| 315                    | PIK3CA | 45          | Known             | SV              | E545K                        |
| HGNECC_2  | 236                    | IRS2   | 35          | Known             | CN              | Amplification                 |
| HGNECC_2  | 236                    | MYCN   | 35          | Known             | CN              | Amplification                 |
| HGNECC_20 | 315                    | MYC    | 33          | Known             | CN              | Amplification                 |
| HGNECC_21 | 182                    | MYC    | 27          | Known             | CN              | Amplification                 |
| HGNECC_22 | 236                    | TP53   | 27          | Known             | SV              | R273C                        |
| HGNECC_22 | 236                    | MLL2   | 55          | Known             | SV              | R1702*                       |
| HGNECC_23 | 236                    | TP53   | 55          | Known             | SV              | R280T                        |
| HGNECC_23 | 236                    | BRCA2  | 61          | Known             | SV              | I2627F                       |
| HGNECC_23 | 236                    | PIK3R1 | 61          | Likely            | RE              | Truncation                    |
| HGNECC_23 | 236                    | PPP2R1A| 61          | Known             | SV              | P179R                        |
| HGNECC_23 | 236                    | PTEN   | 61          | Known             | CN              | Deletion                      |

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| Case No. | No. of Genes Assessed | Gene   | Age (years) | Functional Status | Alteration Type | Description  |
|----------|-----------------------|--------|-------------|------------------|-----------------|--------------|
| HGNECC_23 | 236 | RB1     | 61         | Known            | CN              | Deletion     |
| HGNECC_23 | 236 | TP53    | 61         | Known            | SV              | R248Q       |
| HGNECC_24 | 236 | ARID1A  | 38         | Likely           | SV              | A141fs*42    |
| HGNECC_24 | 236 | ARID1A  | 38         | Likely           | SV              | Q1397fs*46   |
| HGNECC_24 | 236 | CTNNB1  | 38         | Known            | SV              | S37C        |
| HGNECC_24 | 236 | PIK3CA  | 38         | Known            | SV              | H1047R      |
| HGNECC_24 | 236 | PTEN    | 38         | Known            | SV              | D92E        |
| HGNECC_24 | 236 | PTEN    | 38         | Known            | SV              | Y180*       |
| HGNECC_25 | 315 | MYC     | 25         | Known            | CN              | Amplification |
| HGNECC_26 | 315 | KRAS    | 25         | Known            | SV              | G13D        |
| HGNECC_26 | 315 | MYC     | 25         | Known            | CN              | Amplification |
| HGNECC_27 | 315 | ABL2    | 49         | Known            | SV              | P497fs*7     |
| HGNECC_27 | 315 | ATRX    | 49         | Likely           | SV              | D1940fs*15   |
| HGNECC_27 | 315 | BLM     | 49         | Known            | SV              | N515fs*16    |
| HGNECC_27 | 315 | ERBB3   | 49         | Known            | SV              | R475W       |
| HGNECC_27 | 315 | FBXW7   | 49         | Known            | SV              | R465H       |
| HGNECC_27 | 315 | FGFR1   | 49         | Known            | SV              | V127M       |
| HGNECC_27 | 315 | JAK1    | 49         | Known            | SV              | K886fs*16    |
| HGNECC_27 | 315 | JAK1    | 49         | Known            | SV              | P430fs*2     |
| HGNECC_27 | 315 | MEN1    | 49         | Likely           | SV              | R521fs*43    |
| HGNECC_27 | 315 | MLL2    | 49         | Likely           | SV              | P2302fs*20   |
| HGNECC_27 | 315 | MLL3    | 49         | Known            | SV              | K2797fs*26   |
| HGNECC_27 | 315 | MSH2    | 49         | Likely           | SV              | E48*        |
| HGNECC_27 | 315 | MSH2    | 49         | Likely           | SV              | Q324*       |
| HGNECC_27 | 315 | NOTCH1  | 49         | Known            | SV              | R1586H      |
| HGNECC_27 | 315 | PIK3CA  | 49         | Known            | SV              | E545D       |
| HGNECC_27 | 315 | PREX2   | 49         | Known            | SV              | S565fs*3     |
| HGNECC_27 | 315 | PTEN    | 49         | Known            | SV              | K267fs*9     |
| HGNECC_27 | 315 | PTEN    | 49         | Known            | SV              | R130Q       |
| HGNECC_27 | 315 | QKI     | 49         | Known            | SV              | A338T       |
| HGNECC_27 | 315 | SETD2   | 49         | Likely           | SV              | F636fs*6     |
| HGNECC_27 | 315 | SMARCA4 | 49         | Likely           | SV              | Q214*       |
| HGNECC_27 | 315 | SMARCA4 | 49         | Likely           | SV              | T296fs*7     |
| HGNECC_27 | 315 | STK11   | 49         | Likely           | SV              | W332*       |
| HGNECC_27 | 315 | TET2    | 49         | Known            | SV              | R550*       |
| HGNECC_27 | 315 | TET2    | 49         | Likely           | SV              | R1440fs*38   |
| HGNECC_28 | 315 | MLL2    | 60         | Likely           | SV              | L951fs*7     |
| HGNECC_28 | 315 | PTCH1   | 60         | Known            | SV              | G682C       |
| HGNECC_28 | 315 | PTEN - ANKRD22 | 60 | Likely | RE truncation |
| HGNECC_29 | 315 | CREBBP  | 38         | Likely           | SV              | S2377*      |
| HGNECC_29 | 315 | KRAS    | 38         | Known            | CN              | Amplification |
| HGNECC_29 | 315 | KRAS    | 38         | Known            | SV              | G12D        |
| HGNECC_3  | 315 | ARFRP1  | 29         | Known            | CN              | Amplification |
| HGNECC_3  | 315 | AURKA   | 29         | Known            | CN              | Amplification |

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| Case No. | No. of Genes Assessed | Gene | Age (years) | Functional Status | Alteration Type         | Description       |
|----------|-----------------------|------|-------------|-------------------|-------------------------|-------------------|
| HGNECC_3 | 315                   | GNAS| 29          | Known             | CN                      | Amplification     |
| HGNECC_30| 315                   | PTEN| 24          | Likely            | SV                      | M205Fs*14         |
| HGNECC_32| 315                   | BRD4| 67          | Likely            | SV                      | F656Fs*4          |
| HGNECC_32| 315                   | ERBB3| 67          | Known             | SV                      | V104L             |
| HGNECC_32| 315                   | FBXW7| 67          | Known             | SV                      | R505C             |
| HGNECC_32| 315                   | KRAS| 67          | Known             | SV                      | G12V              |
| HGNECC_33| 315                   | MLL3| 42          | Known             | SV                      | R380C             |
| HGNECC_33| 315                   | PIK3CA| 42         | Known             | CN                      | Amplification     |
| HGNECC_33| 315                   | PIK3CA| 42         | Known             | SV                      | E545K             |
| HGNECC_33| 315                   | SOX2| 42          | Known             | CN                      | Amplification     |
| HGNECC_34| 315                   | ARID1A| 52        | Likely            | SV                      | D1850Fs*33        |
| HGNECC_34| 315                   | CIC | 52          | Known             | SV                      | D473N             |
| HGNECC_34| 315                   | CTCF| 52          | Likely            | SV                      | T204Fs*26         |
| HGNECC_34| 315                   | GRM3| 52          | Known             | SV                      | G621V             |
| HGNECC_34| 315                   | MSH2| 52          | Likely            | SV                      | C199Fs*15         |
| HGNECC_34| 315                   | MSH2| 52          | Likely            | SV                      | Q846*             |
| HGNECC_34| 315                   | PIK3CA| 52        | Known             | SV                      | Q546H             |
| HGNECC_34| 315                   | PIK3CA| 52        | Known             | SV                      | R38H              |
| HGNECC_34| 315                   | PTEN| 52          | Known             | SV                      | N323Fs*2          |
| HGNECC_34| 315                   | PTEN| 52          | Known             | SV                      | T468M             |
| HGNECC_34| 315                   | RANBP2| 52        | Known             | SV                      | L811R             |
| HGNECC_34| 315                   | RBM10| 52         | Likely            | SV                      | R98*              |
| HGNECC_34| 315                   | RNF43| 52          | Likely            | SV                      | G659Fs*41         |
| HGNECC_34| 315                   | SMARCA4| 52       | Likely            | SV                      | L1161Fs*3         |
| HGNECC_34| 315                   | SMARCA4| 52       | Likely            | SV                      | P305Fs*21         |
| HGNECC_34| 315                   | SPEN| 52          | Known             | SV                      | R75H              |
| HGNECC_35| 315                   | PPP2R1A| 77      | Known             | SV                      | P179R             |
| HGNECC_35| 315                   | PTEN| 77          | Known             | SV                      | D92E              |
| HGNECC_35| 315                   | PTEN| 77          | Known             | SV                      | L325R             |
| HGNECC_35| 315                   | RB1 | 77          | Likely            | SV                      | G89*              |
| HGNECC_35| 315                   | SMARCA4| 77       | Known             | CN                      | Deletion           |
| HGNECC_35| 315                   | TP53| 77          | Known             | SV                      | R306*             |
| HGNECC_37| 315                   | FGFI4| 50          | Known             | CN                      | Amplification     |
| HGNECC_37| 315                   | IRS2| 50          | Known             | CN                      | Amplification     |
| HGNECC_37| 315                   | KRAS| 50          | Known             | SV                      | K182_T183del      |
| HGNECC_37| 315                   | PBRM1| 50         | Likely            | SV                      | Q1346*            |
| HGNECC_38| 315                   | MSH2| 49          | Likely            | SV                      | R929*             |
| HGNECC_38| 315                   | MYC | 49          | Known             | CN                      | Amplification     |
| HGNECC_38| 315                   | PIK3R1| 49        | Known             | SV                      | T576del           |
| HGNECC_38| 315                   | PTEN| 49          | Known             | SV                      | A126T             |
| HGNECC_38| 315                   | RB1 | 49          | Known             | CN                      | Deletion           |
| HGNECC_38| 315                   | TP53| 49          | Known             | SV                      | K321Fs*24         |
| HGNECC_4 | 315                   | EP300| 26          | Likely            | SV                      | A1437Fs*65        |
| HGNECC_4 | 315                   | GNAS| 26          | Known             | SV                      | R201H             |
| Case No. | No. of Genes Assessed | Gene   | Age (years) | Functional Status | Alteration Type       | Description                  |
|----------|-----------------------|--------|-------------|-------------------|------------------------|------------------------------|
| HGNECC_4 | 315                   | MYC    | 26          | Known             | CN                     | Amplification                |
| HGNECC_4 | 315                   | PTPRD  | 26          | Likely            | SV                     | N1023fs*7                    |
| HGNECC_40| 315                   | AKT1   | 35          | Known             | SV                     | W80R                         |
| HGNECC_40| 315                   | ARID1A | 35          | Known             | SV                     | R1276*                       |
| HGNECC_40| 315                   | ARID1B | 35          | Likely            | SV                     | Q1331*                       |
| HGNECC_40| 315                   | BRAF   | 35          | Known             | SV                     | I326V                        |
| HGNECC_40| 315                   | CTNNB1 | 35          | Known             | SV                     | D32Y                         |
| HGNECC_40| 315                   | PIK3CA | 35          | Known             | SV                     | C420, P421del                |
| HGNECC_40| 315                   | PIK3R1 | 35          | Likely            | SV                     | Splice site 1746-2A>G        |
| HGNECC_41| 236                   | JAK2   | 26          | Known             | CN                     | Amplification                |
| HGNECC_42| 236                   | ARFRP1 | 36          | Known             | CN                     | Amplification                |
| HGNECC_42| 236                   | ERBB2  | 36          | Known             | SV                     | S310F                        |
| HGNECC_42| 236                   | SRC    | 36          | Known             | CN                     | Amplification                |
| HGNECC_42| 236                   | TOP1   | 36          | Known             | CN                     | Amplification                |
| HGNECC_42| 236                   | TP53   | 36          | Known             | SV                     | R248Q                        |
| HGNECC_42| 236                   | ZNF217 | 36          | Known             | CN                     | Amplification                |
| HGNECC_44| 315                   | FANCC  | 72          | Likely            | RE                     | Truncation                   |
| HGNECC_44| 315                   | KRAS   | 72          | Known             | SV                     | G12D                         |
| HGNECC_44| 315                   | NOTCH1 | 72          | Likely            | SV                     | A305fs*27                    |
| HGNECC_44| 315                   | NOTCH1 | 72          | Likely            | SV                     | Splice site 2354-1G>A        |
| HGNECC_44| 315                   | PIK3CA | 72          | Known             | SV                     | R88Q                         |
| HGNECC_44| 315                   | RICTOR | 72          | Known             | CN                     | Amplification                |
| HGNECC_45| 315                   | ARID2  | 33          | Likely            | SV                     | S1157*                       |
| HGNECC_45| 315                   | CCNE1  | 33          | Known             | CN                     | Amplification                |
| HGNECC_45| 315                   | EPHA5  | 33          | Known             | SV                     | S964Y                        |
| HGNECC_46| 315                   | CCNE1  | 41          | Known             | CN                     | Amplification                |
| HGNECC_46| 315                   | GRIN2A | 41          | Known             | SV                     | R19C                         |
| HGNECC_46| 315                   | NCO1   | 41          | Likely            | SV                     | Y1617*                       |
| HGNECC_47| 315                   | BCL2   | 51          | Known             | CN                     | Amplification                |
| HGNECC_47| 315                   | GATA6  | 51          | Known             | CN                     | Amplification                |
| HGNECC_47| 315                   | NX2-1  | 51          | Known             | CN                     | Amplification                |
| HGNECC_49| 315                   | ARID1A | 59          | Likely            | SV                     | Splice site 4923, 4993, 259del330 |
| HGNECC_49| 315                   | ARID1A-ARID1A | 59 | Likely | RE | Deletion |
| HGNECC_49| 315                   | PALB2  | 59          | Likely            | SV                     | Q141fs*27                    |
| HGNECC_5  | 315                   | RB1    | 40          | Likely            | SV                     | E629fs*12                    |
| HGNECC_5  | 315                   | KRAS   | 46          | Known             | SV                     | G13D                         |
| HGNECC_50 | 315                   | MAGI2  | 46          | Known             | SV                     | R564Q                        |
| HGNECC_51 | 315                   | BRCA2  | 38          | Likely            | RE                     | Truncation                   |
| HGNECC_52 | 315                   | NOTCH2 | 73          | Likely            | SV                     | S1270fs*11                   |
| HGNECC_52 | 315                   | PIK3R1 | 73          | Likely            | SV                     | D330fs*15                    |
| HGNECC_53 | 315                   | AKT1   | 54          | Known             | SV                     | E17K                         |
| HGNECC_53 | 315                   | ARID1A | 54          | Likely            | SV                     | Y1260fs*9                    |
| HGNECC_53 | 315                   | BRCA2  | 54          | Likely            | SV                     | N3124I                       |
| HGNECC_53 | 315                   | GNAS   | 54          | Known             | SV                     | R201C                        |
| Case No. | No. of Genes Assessed | Gene     | Age (years) | Functional Status | Alteration Type | Description |
|---------|-----------------------|----------|-------------|------------------|----------------|-------------|
| HGNECC_53 | 315                  | PIK3CA   | 54          | Known            | SV             | E542K       |
| HGNECC_53 | 315                  | RUNX1T1  | 54          | Known            | SV             | R395W       |
| HGNECC_54 | 315                  | PIK3CA   | 34          | Known            | CN             | Amplification |
| HGNECC_54 | 315                  | SOX2     | 34          | Known            | CN             | Amplification |
| HGNECC_55 | 315                  | EP300    | 34          | Known            | SV             | D1399N      |
| HGNECC_55 | 315                  | MYC      | 45          | Likely           | SV             | E1571fs*3   |
| HGNECC_56 | 315                  | MYC      | 45          | Known            | CN             | Amplification |
| HGNECC_56 | 315                  | NCOR1    | 45          | Known            | SV             | R1794Q      |
| HGNECC_56 | 315                  | PIK3CA   | 45          | Known            | SV             | C420R       |
| HGNECC_56 | 315                  | SOX2     | 45          | Likely           | SV             | S397*       |
| HGNECC_57 | 315                  | TP53     | 28          | Known            | CN             | R248W       |
| HGNECC_57 | 315                  | KRAS     | 28          | Known            | SV             | G13C        |
| HGNECC_57 | 315                  | MLL3     | 28          | Likely           | SV             | Q419*       |
| HGNECC_58 | 315                  | MYC      | 30          | Known            | SV             | E14K        |
| HGNECC_58 | 315                  | SMARCA4  | 30          | Likely           | SV             | Y820*       |
| HGNECC_59 | 315                  | CIC      | 35          | Likely           | SV             | Q427*       |
| HGNECC_59 | 315                  | MYC      | 35          | Known            | CN             | Amplification |
| HGNECC_6  | 315                  | CRLF2 - DHRSX | 44 | Likely | RE | Fusion |
| HGNECC_6  | 315                  | IRS2     | 44          | Known            | CN             | Amplification |
| HGNECC_60 | 315                  | NUP93    | 27          | Known            | SV             | E14K        |
| HGNECC_61 | 315                  | MYC      | 36          | Known            | CN             | Amplification |
| HGNECC_61 | 315                  | NOTCH2   | 36          | Known            | SV             | P6fs*27     |
| HGNECC_61 | 315                  | NOTCH4 - NOTCH4 | 36 | Likely | RE | Deletion |
| HGNECC_63 | 315                  | BRC2     | 38          | Known            | SV             | F1192C      |
| HGNECC_63 | 315                  | MYC      | 38          | Known            | CN             | Amplification |
| HGNECC_65 | 315                  | ARID1A   | 50          | Likely           | SV             | E1060*      |
| HGNECC_65 | 315                  | CTNNB1   | 50          | Known            | SV             | S33C        |
| HGNECC_65 | 315                  | MLL2     | 50          | Likely           | SV             | Y1514fs*2   |
| HGNECC_65 | 315                  | PIK3CA   | 50          | Known            | SV             | V344G       |
| HGNECC_65 | 315                  | PTEN     | 50          | Known            | SV             | N184fs*6    |
| HGNECC_65 | 315                  | PTEN     | 50          | Likely           | SV             | E285*       |
| HGNECC_66 | 315                  | CCND2    | 39          | Known            | CN             | Amplification |
| HGNECC_66 | 315                  | FGF23    | 39          | Known            | CN             | Amplification |
| HGNECC_66 | 315                  | FGF6     | 39          | Known            | CN             | Amplification |
| HGNECC_66 | 315                  | KDM5A    | 39          | Known            | CN             | Amplification |
| HGNECC_66 | 315                  | KRAS     | 39          | Known            | SV             | G12S        |
| HGNECC_66 | 315                  | LRP1B    | 39          | Likely           | SV             | M1882fs*22  |
| HGNECC_66 | 315                  | MYC      | 39          | Known            | CN             | Amplification |

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| Case No. | No. of Genes Assessed | Gene         | Age (years) | Functional Status | Alteration Type | Description                  |
|---------|-----------------------|--------------|-------------|-------------------|-----------------|------------------------------|
| HGNECC_67 | 315                  | BRF - N/A    | 27          | Likely            | RE              | Rearrangement                |
| HGNECC_68 | 315                  | EGFR         | 60          | Likely            | SV              | T145M                        |
| HGNECC_69 | 315                  | BRD4         | 49          | Likely            | SV              | E1249*                      |
| HGNECC_69 | 315                  | PIK3CA       | 49          | Known             | SV              | R88Q                         |
| HGNECC_7  | 315                  | MYCN         | 47          | Known             | CN              | Amplification                |
| HGNECC_71 | 315                  | FGFR1        | 43          | Known             | CN              | Amplification                |
| HGNECC_71 | 315                  | NOTCH1       | 43          | Known             | SV              | V1575L                       |
| HGNECC_72 | 236                  | PTEN         | 35          | Known             | CN              | Deletion                     |
| HGNECC_72 | 236                  | TP53         | 35          | Known             | SV              | Splice site 375G>A           |
| HGNECC_73 | 315                  | MYC          | 28          | Known             | CN              | Amplification                |
| HGNECC_73 | 315                  | TP53         | 28          | Known             | SV              | R283C                        |
| HGNECC_74 | 315                  | ERBB2        | 65          | Known             | CN              | Amplification                |
| HGNECC_74 | 315                  | IGF2R        | 65          | Known             | SV              | R1325H                       |
| HGNECC_74 | 315                  | PTEN         | 65          | Known             | SV              | R130G                        |
| HGNECC_74 | 315                  | PTEN         | 65          | Likely            | SV              | C250fs*4                     |
| HGNECC_74 | 315                  | TP53         | 65          | Likely            | SV              | Splice site 783-1G>T         |
| HGNECC_76 | 315                  | ATRX         | 35          | Likely            | SV              | R1504*                       |
| HGNECC_78 | 315                  | NUP93        | 52          | Known             | SV              | E14K                         |
| HGNECC_78 | 315                  | SOX2         | 52          | Known             | CN              | Amplification                |
| HGNECC_79 | 315                  | MTR         | 38          | Known             | SV              | TI834_T1837del               |
| HGNECC_8  | 236                  | LRPIB        | 37          | Known             | SV              | D3472N                       |
| HGNECC_8  | 236                  | MCL1         | 37          | Known             | CN              | Amplification                |
| HGNECC_8  | 236                  | ATM          | 28          | Likely            | SV              | K468fs*18                    |
| HGNECC_8  | 315                  | BRD4 - KIAA0319 | 28          | Likely            | RE              | Fusion                       |
| HGNECC_8  | 315                  | MLL3         | 28          | Likely            | SV              | Y306*                        |
| HGNECC_82 | 236                  | ARID1A       | 48          | Likely            | SV              | G1848fs*6                    |
| HGNECC_82 | 236                  | CTNNB1       | 48          | Known             | SV              | G34V                         |
| HGNECC_82 | 236                  | PIK3CA       | 48          | Known             | SV              | H1047R                       |
| HGNECC_83 | 315                  | CCNE1        | 37          | Known             | CN              | Amplification                |
| HGNECC_83 | 315                  | IGF1R        | 37          | Known             | CN              | Amplification                |
| HGNECC_83 | 315                  | MYCN         | 37          | Known             | CN              | Amplification                |
| HGNECC_84 | 236                  | MCL1         | 40          | Known             | CN              | Amplification                |
| HGNECC_84 | 236                  | PIK3CA       | 40          | Known             | SV              | E545K                        |
| HGNECC_85 | 315                  | AKT1         | 45          | Known             | CN              | Amplification                |
| HGNECC_85 | 315                  | CDKN1B - N/A | 45          | Likely            | RE              | Rearrangement                |
| HGNECC_86 | 315                  | ERBB2       | 52          | Known             | SV              | S310Y                        |
| HGNECC_86 | 315                  | RB1          | 52          | Likely            | SV              | Splice site 2490-1G>A        |
| HGNECC_87 | 315                  | MLL3         | 38          | Likely            | SV              | Y1348*                       |
| HGNECC_87 | 315                  | PIK3CA       | 38          | Known             | CN              | Amplification                |
| HGNECC_87 | 315                  | PIK3CA       | 38          | Known             | SV              | E542K                        |
| HGNECC_87 | 315                  | SOX9         | 38          | Likely            | SV              | Y503*                        |
| HGNECC_88 | 315                  | BRCA2        | 54          | Likely            | SV              | E1608*                       |
| HGNECC_88 | 315                  | MAGI2        | 54          | Known             | SV              | R1220*                       |

(Continued on following page)
### TABLE A1. Detailed Genomic Assessment of High Grade Neuroendocrine Cervical Cancer Samples (Continued)

| Case No. | No. of Genes Assessed | Gene | Age (years) | Functional Status | Alteration Type | Description |
|----------|------------------------|------|-------------|-------------------|----------------|-------------|
| HGNECC_89 | 315 | FGF10 | 29 | Known | CN | Amplification |
| HGNECC_89 | 315 | RICTOR | 29 | Known | CN | Amplification |
| HGNECC_9  | 236 | FGF10 | 27 | Known | CN | Amplification |
| HGNECC_9  | 236 | MYC | 27 | Known | CN | Amplification |
| HGNECC_9  | 236 | RICTOR | 27 | Known | CN | Amplification |
| HGNECC_90 | 315 | DNMT3A | 73 | Known | SV | R882H |
| HGNECC_90 | 315 | MLL2 | 73 | Likely | SV | Q5446* |
| HGNECC_90 | 315 | SOX2 | 73 | Known | CN | Amplification |
| HGNECC_91 | 315 | PIK3R1 | 59 | Known | SV | D560Y |
| HGNECC_92 | 315 | CDKN1B | 48 | Likely | SV | P137fs*8 |
| HGNECC_92 | 315 | GNAS | 48 | Known | SV | R201H |
| HGNECC_92 | 315 | KIT | 48 | Known | SV | D816V |
| HGNECC_92 | 315 | TP53 | 48 | Known | SV | R248Q |
| HGNECC_93 | 315 | DDR2 | 46 | Known | SV | T836M |
| HGNECC_93 | 315 | MUTYH | 46 | Known | SV | G382D |
| HGNECC_94 | 315 | MERTK | 35 | Known | SV | R865Q |
| HGNECC_94 | 315 | PIK3CA | 35 | Known | SV | N345K |
| HGNECC_95 | 315 | MUTYH | 58 | Known | SV | Y165C |
| HGNECC_95 | 315 | PIK3CA | 58 | Known | SV | E726K |
| HGNECC_96 | 315 | CDKN1B | 67 | Likely | SV | S27* |
| HGNECC_96 | 315 | CUL4B | 67 | Known | SV | S110F |
| HGNECC_96 | 315 | PIK3CA | 67 | Known | SV | D350G |
| HGNECC_96 | 315 | TP53BP1 | 67 | Known | SV | E1165K |
| HGNECC_97 | 315 | FGF10 | 63 | Known | CN | Amplification |
| HGNECC_97 | 315 | RICTOR | 63 | Known | CN | Amplification |

Abbreviations: CN, copy number; N/A, not applicable; RE, rearrangement; SV, structural variation.