Genomic Landscapes of Acral Melanomas in East Asia

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Abstract. Background/Aim: Acral melanomas (AM) represent a rare subgroup of melanomas with poor clinical outcomes and are enriched in Asian populations. Recent advances in next generation sequencing have provided opportunities to apply precision medicine to AM. Patients and Methods: Here, we present a series of 13 patients with melanomas from Taiwan and Singapore, including 8 patients with AM profiled using whole exome sequencing and summarize the recent studies on the genomic landscape of AM. Results: We identified mutually exclusive mutations in BRAF, NRAS, HRAS, NF1 and KIT in 6 AM cases. In addition, recurrent copy number gains in CCND1 and CDK4, as well as recurrent deletions in CDKN2A/CDKN2B, ATM and RAD51 were observed, supporting the potential use of CDK4/6 or PARP inhibitors in the treatment of these patients. Conclusion: The genomic landscape of AM provides an important resource for applying novel targeted therapies in this rare disease.

Acral melanomas (AM) are a subset of melanomas that arise from non-hair bearing glabrous skin on the palms and soles, or on the nail apparatus (1). Despite global rarity, AM is the commonest subtype of melanoma in Asian populations (2). Notably, patients with AM harbor worse prognosis as compared with cutaneous melanomas, and survival outcomes remain dismal despite modern advances in the therapeutic landscape of melanomas (3, 4).

Recently, next generation sequencing (NGS) technologies, involving whole exome (WES) (5-7) or genome sequencing (WGS) (8-14), have enhanced the molecular understanding of AM. At the molecular level, AM is a distinct disease as compared with cutaneous melanomas, defined by few point or indel mutations and high degrees of complex structural rearrangements and focal copy number alterations (8-10). Unlike cutaneous melanomas, the tumor mutation burden (TMB) is consequently lower and mutational signatures of ultraviolet damage are infrequent (11). At the individual gene level, hotspot mutations in BRAF and NRAS occur in over 50% of cutaneous melanomas, whereas their occurrence in AM is considerably lower (approximately 10-25%). On the other hand, mutations in NF1 and KIT, as well as oncogenic amplification of genes such as CCND1, CDK4, and TERT have been demonstrated to be common events in AM (5, 8). These unique genomic alterations harbor therapeutic implications - small molecule inhibitors of KIT and other tyrosine kinases, including imatinib, nilotinib and dasatinib, have demonstrated significant (albeit modest) efficacy against KIT-mutant AM (15-18). Similarly, CDK4/6 inhibitors have also shown promising activity in AM (19). Taken together, the unique genomic landscape of AM offers an opportunity for the application of precision medicine in this rare disease and warrants further investigation.

In this article, we present a series of patients with AM from Taiwan and Singapore profiled using whole exome sequencing and summarize the recent studies on the genomic landscape of AM using NGS, extending our current understanding of this Asian-prevalent subtype of melanoma.
Patients and Methods

Study design and participants. A total of 13 patients with histologically-proven melanoma from the National Cancer Centre Singapore (Singapore) and Chang Gung Memorial Hospital at Linkou (Taiwan, R.O.C.) were included in the study. Clinical information collected included sex, age, stage at diagnosis (20) and primary tumor location. Written informed consent was obtained in accordance with the Declaration of Helsinki. The study was approved by the Institutional Review Boards of all participating hospitals. All authors had access to the study data and had reviewed and approved the final manuscript.

Whole exome sequencing. Genomic DNA isolated from formalin-fixed paraffin-embedded or snap frozen tissue with adequate tumor content, as well as from their paired normal tissue, were selected for whole exome sequencing. A qualified pathologist provided the initial microscopic evaluation and assessment of tumor content. Whole exome sequencing was performed with hybrid selection using the Human All Exon kit SureSelect Target Enrichment System (Agilent Technologies, Santa Clara, CA, USA) version 6 and sequenced on the Illumina HiSeq X platform (Illumina, San Diego, CA, USA) as paired-end 150-base pair reads. Read pairs were aligned to the human reference genome NCBI GRC Build 37 (hg19) using Burrows-Wheeler Aligner (BWA MEM) (Wellcome Genome Campus, Hinxton, Cambridge, UK) (21). Optical duplicates were marked with Picard followed by base score recalibration using GATK version 4.1.4 (Broad Institute, Cambridge, MA, USA) for post alignment data processing (22). Somatic variants from the resulting normal and tumor BAM files were identified using Mutect2, and subsequently annotated and prioritized using VEP (Wellcome Genome Campus, Hinxton, Cambridge, UK) (23). Tumor mutation burden was estimated based on the proportion of nonsynonymous variants over the region of interest (ROI) of the exome panel used. Mutational signature identification was performed using SigProfiler Biomathematics Tools (Wellcome Genome Campus, Hinxton, Cambridge, UK) (24). Biologically significant copy number changes were identified with GISTIC 2.0 (Broad Institute, Cambridge, MA, USA) and copy-number segmentations were processed with TitanCNA v1.17.1 (University of British Columbia, Vancouver, Canada) (25, 26).

Results

Patient demographics. A total of 13 patients were included in the study based on tissue availability (Singapore cases, n=9; Taiwan cases, n=4). The median age was 61 years (range=39-97 years) and there was a male predominance (n=9; 69.2%). Eight cases were acral melanomas, all arising from the feet. Three were mucosal melanomas of the nasopharynx, anus and vagina. Two cases of cutaneous melanomas arising from the chest wall and upper arm were also included for comparison. None of the patients had distant metastases at diagnosis. Table I summarizes the clinical characteristics of all patients in the study cohort.

| Patient ID | Subtype | Primary Site | Age at diagnosis | Gender | Country of origin | Stage at diagnosis |
|------------|---------|--------------|------------------|--------|-------------------|-------------------|
| Mel-01     | Acral, lentiginous | Left sole    | 85               | Male   | Taiwan            | IIC               |
| Mel-02     | Acral, lentiginous | Left big toe | 78               | Male   | Taiwan            | II                |
| Mel-03     | Acral, lentiginous | Left foot    | 65               | Male   | Taiwan            | II                |
| Mel-04     | Acral, lentiginous | Right sole   | 60               | Male   | Taiwan            | III               |
| Mel-05     | Acral, nodular pattern | Right big toe | 97          | Male   | Singapore        | IIC               |
| Mel-06     | Acral, lentiginous | Right heel   | 56               | Male   | Singapore        | III               |
| Mel-07     | Acral, unspecified pattern | Left foot   | 51               | Female | Singapore        | III               |
| Mel-08     | Acral, nodular pattern | Right sole   | 61               | Male   | Singapore        | III               |
| Mel-09     | Mucosal | Nasopharynx   | 41               | Female | Singapore        | II                |
| Mel-10     | Mucosal | Anus          | 75               | Female | Singapore        | III               |
| Mel-11     | Mucosal | Vagina        | 44               | Female | Singapore        | III               |
| Mel-12     | Cutaneous, unspecified pattern | Chest wall | 39               | Male   | Singapore        | IIIB              |
| Mel-13     | Cutaneous, nodular pattern | Left upper arm | 80          | Male   | Singapore        | IIC               |

Table I. Characteristics of the patients included in the study.
Figure 1. Somatic mutation landscape of acral, mucosal and cutaneous melanoma of East Asian origin. Variants of interest are represented in an oncoplot, including recurrent mutations in genes such as *BRAF*, *NRAS* and *NF1*. Three of 11 non-cutaneous melanomas were "triple wild-type" – one of which was characterized by a KIT exon 11 L576P mutation.
Figure 2. Mutational signatures of melanomas in the cohort. The proportions of mutations conferred by each inferred mutational signature in individual cases are as shown.

Figure 3. Copy number alterations in the global melanoma cohort. Analysis of somatic copy number alterations identified 4 gained and 13 lost genomic regions. The involved regions and important cancer-related genes within are highlighted.
614 missense single nucleotide variants (SNVs), 37 nonsense SNVs, 25 indels and 243 silent mutants. Somatic nonsynonymous variants of interest are represented in an oncplot, including recurrent mutations in known melanoma-associated genes such as BRAF (31%), NRAS (31%), NF1 (15%) and HRAS (8%) (Figure 1). BRAF mutations were all missense and present in 1 of 8 AM cases (p.V600E), 1 of 3 mucosal melanomas (p.G534R), and both cutaneous melanomas (p.V600E and p.V600K). NRAS mutations were all missense in the p.Q61 hotspot for 2 of 8 AM cases and 2 of 3 mucosal melanoma cases. Notably, 3 of 11 (27.3%) non-cutaneous melanomas were “triple wild-type” – one of which was a KIT exon 11 L576P mutant.

The estimated proportions of mutations contributed by inferred mutational signatures in individual melanoma cases were examined. Signatures 1 and 5, which are related to aging and observed in most cancer types, were present in most of the cases. The signatures for ultraviolet DNA mutagenesis - Single Base Substitution (SBS) 7a and SBS 7b, as characterized by a majority of C>T mutations, were observed in 6 cases (2 cutaneous, 1 mucosal, and 3 acral melanomas), though the relative contribution per case was minor (Figure 2).

**Somatic copy number alterations.** Analysis of somatic copy number alterations identified 4 gained genomic regions (5p13.2, 11q13.3, 12q13.3, 22q13.2). We further identified 13 deleted regions (1p36.33, 5p15.33, 6p21.33, 6q25.1, 8p23.1, 9p21.3, 9q34.11, 11p11.2, 11q23.3, 14q32.33, 15q15.1, 16p11.2, 16q23.2) (Figure 3). Further introspection of individual cases revealed several important gained regions of interest, including chromosome 11q and 12q – containing oncogenes CCND1 and CDK4, respectively (Figure 4). Gene-level copy number analysis revealed recurrent copy number gains/amplifications in CCND1 (46%), CDK4 (31%), SKP2 (15%) and EP300 (15%), and recurrent deletions in CDKN2A/CDKN2B (54%), ATM (38%), RAD51 (38%) and FANCA (15%) (Figure 5).

**Recent genomic studies on acral melanoma.** A total of 92 articles were screened. After excluding review articles (n=9), meta-analyses (n=1), case reports (n=4), commentaries (n=5) and other studies (n=63), 10 articles remained and were included in the final analysis. The study design and main findings are summarized in Table II.

**Discussion**

Newell et al. have recently reported the largest series of AM profiled using whole genome sequencing (n=87). The authors observed several significantly mutated genes including BRAF, NRAS, NF1, NOTCH2, PTEN and TYRP1, as well as KIT alterations. Mutational signature analysis revealed a subset of tumors, mostly subungual, with an...
Figure 4. Copy number landscape of individual melanoma cases. Red arrows mark selected gained regions of interest, including chromosome 11q and 12q – containing oncogenes CCND1 and CDK4, respectively.
ultraviolet radiation signature. Recurrent complex rearrangements were observed on chromosomes 5, 6, 7, 11 and 12, associated with amplification of TERT, CDK4, MDM2, CCND1, PAK1 and GAB2. In keeping with previous reports (11, 12), structural alterations including whole genome duplication, aneuploidy and complex rearrangements (such as breakage-fusion-bridge and chromotripsis) are common in AM (8). A unique form of complex rearrangement involving amplified fold-back inversions, termed “Tyfonas”, were commonly observed in AM (40%)

Interestingly, our data revealed ATM deep deletions, with or without concurrent shallow deletions of RAD51 and FANCA in 3 of 8 (37.5%) AM patients, supporting the use of PARP inhibitors for their treatment (37). Recent real-world data suggests that the efficacy of immune checkpoint inhibitors is significantly lower in patients with AM as compared to cutaneous melanomas (38). While the lack of high TMB may in part explain this dismal result, genetic gains of CDK4 or CCND1, as well as deletions of CDKN2A, may indicate the potential utility of CDK4/6 inhibitors in AM (34-36). Interestingly 2 of the cases contained deep deletions of ATM. Altogether, the somatic alterations of AM may suggest potential avenues of therapeutic susceptibility.

Contemporary treatment options for advanced BRAF mutant melanomas commonly involve the use of one or more small molecule tyrosine kinase inhibitors (TKI) (28, 29) or checkpoint immunotherapy (30, 31). While an attractive target, BRAF mutations occur in only 15% of AM as compared to 50% of cutaneous melanomas. The presence of other therapeutically-tractable mutations such as KIT may indicate additional treatment options using other TKIs such as imatinib (32), dasatinib (33) or nilotinib (16). Aberrations in the CDK4 pathway, including amplifications of CDK4 and CCND1, as well as deletions of CDKN2A, may indicate the potential utility of CDK4/6 inhibitors in AM (34-36). Interestingly, our data revealed ATM deep deletions, with or without concurrent shallow deletions of RAD51 and FANCA in 3 of 8 (37.5%) AM patients, supporting the use of PARP inhibitors for their treatment (37). Recent real-world data suggests that the efficacy of immune checkpoint inhibitors is significantly lower in patients with AM as compared to cutaneous melanomas (38). While the lack of high TMB may in part explain this dismal result, genetic gains of CDK4 or CCND1, as well as CDKN2A loss have been identified in melanoma patients with innate resistance to anti-PD1 checkpoint immunotherapy (39). Yu et al. have provided further evidence that this innate resistance may be mediated by the lack of IFNγ and TNFα-NFκB signaling responses in CDK4 pathway-defective tumors, and that the addition of the CDK4/6 inhibitor palbociclib may enhance the efficacy of immunotherapy by upregulating PD-L1 (39).

Our current study is limited by the small patient cohort. Nonetheless, the results are consistent with previously published studies and lend confirmatory evidence to support a therapeutic target landscape for AM. In addition, we described the loss of genes involved in homologous recombination repair in a significant proportion of AM, supporting the use of PARP inhibitors in the treatment of
these patients. Taken together, the recent data suggesting that complex structural alterations represent early events unique to AM pathogenesis opens up further avenues that can be exploited for therapy.

In conclusion, the genomic landscape of AM presents a unique opportunity for applying novel therapies to this group of patients. Future studies are warranted for the direct translation of these findings to the clinic.

Conflicts of Interest
The Authors have no conflicts of interest to declare in relation to this study.

Authors’ Contributions
JYC analyzed the data and drafted the manuscript; CCN processed tissue and performed sequencing experiments; AHI performed the bioinformatic analyses; JYC, CEW, and JWC obtained patient samples and data; BTT, JWC designed the study; JYC, BTT, JWC interpreted the results, and revised the manuscript; JJH was responsible for the CORPG3J0151~2 grant application, sample collection, DNA preparation, clinical data confirmation, and manuscript revisions; and all authors read and approved the final version of the manuscript.

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References
1 Chen YA, Teer JK, Eroglu Z, Wu JY, Koomen JM, Karreth FA, Messina JL and Smalley KSM: Translational pathology, genomics and the development of systemic therapies for acral melanoma. Semin Cancer Biol 61: 149-157, 2020. PMID: 31689494. DOI: 10.1016/j.semcancer.2019.10.017

2 Chi Z, Li S, Sheng X, Si L, Cui C, Han M and Guo J: Clinical presentation, histology, and prognoses of malignant melanoma in ethnic Chinese: a study of 522 consecutive cases. BMC Cancer 11: 83-92 (2021)

3 Shoushtari AN, Munhoz RR, Kuk D, Ott PA, Johnson DB, Tsai KK, Rapisuwon S, Eroglu Z, Sullivan RJ, Luke JJ, Gangadhar TC, Salama AK, Clark V, Burias C, Puzanov I, Atkins MB, Algazi AP, Ribas A, Wolchok JD and Postow MA: The efficacy of anti-PD-1 agents in acral and mucosal melanoma. Cancer 22: 3354-3362, 2016. PMID: 27533633. DOI: 10.1002/cncr.30259

4 Keilholz U, Ascierto PA, Dummer R, Robert C, Lorigan P, van Akkooi A, Arance A, Blank CU, Chiarion Sileni V, Donia M, Faries MB, Gaudy-Marqueste C, Gogas H, Grob JJ, Guckenberger M, Haenen J, Hayes AJ, Hoeller C, Lebbé C, Lugowska I, Mandala M, Márquez-Rodas I, Nathan P, Neyns B, Olofsson Bagge R, Puig S, Rutkowski P, Schilling B, Sondak VK, Tawbi H, Testori A and Michielin O: ESMO consensus conference recommendations on the management of metastatic melanoma: under the auspices of the ESMO Guidelines Committee. Ann Oncol 31: 1435-1448, 2020. PMID: 32763453. DOI: 10.1016/j.annonc.2020.07.004

5 Forschner A, Hilke FJ, Bonzheim I, Gschwind A, Demidov G, Amaral T, Ossowski S, Riess O, Schroeder C, Marutzus P, Klumpp B, Gonzalez-Menendez I, Garbe C, Niessner H and Sinnberg T: MDM2, MDM4 and EGFR amplifications and hyperprogression in metastatic acral and mucosal melanoma. Cancers (Basel) 12: 540, 2020. PMID: 32110946. DOI: 10.3390/cancers12030540.

6 Lee M, Yoon J, Chung YJ, Lee SY, Choi JY, Shin OR, Park HY, Bahk WJ, Yu DS and Lee YB: Whole-exome sequencing reveals differences between nail apparatus melanoma and acral melanoma. Am Acad Dermatol 79: 559-561.e1, 2018. PMID: 29438763. DOI: 10.1016/j.jaad.2018.02.019

7 Krauthammer M, Kong Y, Ha BH, Evans P, Bacchiocchi A, McCusker JP, Cheng E, Davis MJ, Goh G, Choi M, Aryan S, Narayan D, Dutton-Regester K, Capatan A, Holman EC, Bosenberg M, Sznol M, Kluger HM, Brash DE, Stern DF, Materin MA, Lo RS, Mane S, Ma S, Kidd KK, Hayward NK, Lifton RP, Schlessinger J, Boggon TJ and Halaban R: Exome sequencing identifies recurrent somatic RAC1 mutations in melanoma. Nat Genet 44: 1006-1014, 2012. PMID: 22842228. DOI: 10.1038/ng.2359

8 Newell F, Wilmott JS, Johansson PA, Nones K, Addala V, Mukhopadhay P, Broit N, Amato CM, Van Gulick R, Kazakoff SH, Patch AM, Kosfariotis LT, Lakis V, Leonard C, Wood S, Holmes O, Xu Q, Lewis K, Medina T, Gonzalez R, Saw RPM, Spillane AJ, Stretch JR, Rawson RV, Ferguson PM, Dodds TJ, Thompson JF, Long GV, Levesque MP, Robinson WA, Pearson JV, Mann GI, Scolyer RA, Waddell N and Hayward NK: Whole-genome sequencing of acral melanoma reveals genomic complexity and diversity. Nat Commun 11: 5259, 2020. PMID: 33067454. DOI: 10.1038/s41467-020-18988-3

9 Hadi K, Yao X, Behr JM, Deshpande A, Xanthopoulakis C, Tian Z, L filmed under the auspices of the ESMO Guidelines Committee. Ann Oncol 31: 1435-1448, 2020. PMID: 32763453. DOI: 10.1016/j.annonc.2020.07.004

10 ICPC/CGCA Pan-cancer analysis of whole genomes consortium pan-cancer analysis of whole genomes. Nature 578: 82-93, 2020. PMID: 23025007. DOI: 10.1038/s41586-020-1969-6

11 Hayward NK, Wilmott JS, Waddell N, Johansson PA, Field MA, Nones K, Patch AM, Kakavand H, Alexander LB, Burke H, Jakrot V, Kazakoff S, Holmes O, Leonard C, Saharianathan R, Mularoni L, Wood S, Xu Q, Waddell N, Temhe V, Pupo GM, De Paoli-Iseppi R, Vilain RE, Shang P, Lau LMS, Dagg RA,
Schramm SJ, Pritchard A, Dutton-Regester K, Newell F, Fitzgerald A, Shang CA, Grimmond SM, Pickett HA, Yang JY, Stretch JR, Behren A, Kefferd RF, Hersey P, Long GV, Cebon J, Shackleton M, Spillane AJ, Saw RM, López-Bigas N, Pearson JV, Thompson JF, Scoller RA and Mann GJ: Whole-genome landscapes of major melanoma subtypes. Nature 545: 175-180, 2017. PMID: 28467829. DOI: 10.1038/nature22071

12 Liang WS, Hendricks W, Kiefer J, Schmidt J, Sekar S, Carpten J, Craig DW, Adkins J, Cuyugan L, Manojlovic Z, Halperin RF, Helland A, Nasser S, Legendre C, Hurley LH, Sivap拉卡斯K, Johnson DB, Crandall H, Busam KJ, Zismann V, Deluca V, Lee J, Sekulic A, Ariyan CE, Kosman J and Trent J: Integrated genomic analyses reveal frequent TERT aberrations in acral melanoma. Genome Res 27: 524-532, 2017. PMID: 28373299. DOI: 10.1101/gr.213348.116

13 Rawson RV, Johansson PA, Hayward NK, Waddell N, Patch AM, Lo S, Pearson JV, Thompson JF, Mann GJ, Scoller RA and Wilmott JS: Unexpected UVR and non-UVR mutation burden in some acral and cutaneous melanomas. Lab Invest 97: 130-145, 2017. PMID: 28067894. DOI: 10.1038/labinvest.2016.143

14 Furney SJ, Turajlic S, Stamp G, Thomas JM, Hayes A, Strauss D, Gavrielides M, Xing W, Gore M, Larkin J and Marais R: The mutational burden of acral melanoma revealed by whole-genome sequencing and comparative analysis. Pigment Cell Melanoma Res 27: 835-838, 2014. PMID: 24913711. DOI: 10.1111/pcm.12279

15 Hodi FS, Corless CL, Gibbou-Hurder A, Fletcher JA, Zhu M, Marino-Enriquez A, Friedlander P, Gonzalez R, Weber JS, Gajewski TF, O'Day SJ, Kim KB, Lawrence D, Flaherty KT, Luke J, Collilcio EA, Earnst SS, Heinrich MC, Beadling C, Zukotynski KA, Yap JT, Van den Abbeele AD, Demetri GD and Fisher DE: Imatinib for melanomas harboring mutationally activated or amplified KIT arising on mucosal, acral, and chronically sun-damaged skin. J Clin Oncol 37: : 3182-3190, 2013. PMID: 23775962. DOI: 10.1200/JCO.2012.47.7836

16 Guo J, Carvalaj RD, Dummer R, Haushofer A, Daud A, Bastian BC, Markovic SN, Queirolo P, Arance A, Berking C, Camargo V, Herchenhorn D, Petrella TM, Schadendorf D, Shafirn W, Testori A, Novick S, Hertle S, Novyr C, Chen Q and Hodi FS: Efficacy and safety of nilotinib in patients with KIT-mutated metastatic or inoperable melanoma: final results from the global, single-arm, phase II TEAM trial. Ann Oncol 28: 1380-1387, 2017. PMID: 28327988. DOI: 10.1093/annonc/mdx079

17 Carvalaj RD, Lawrence DP, Weber JS, Gajewski TF, Gonzalez R, Lutzky J, O’Day SJ, Hamid O, Wolchok JD, Chapman PB, Sullivan RJ, Teitich JB, Ramayila N, Gibbou-Hurder A, Antonescu CR, Heinrich MC, Bastian BC, Corless CL, Fletcher JA and Hodi FS: Phase II study of nilotinib in melanoma harboring KIT alterations following progression to prior KIT inhibition. Clin Cancer Res 27: 2289-2296, 2015. PMID: 25695690. DOI: 10.1158/1078-0432.CCR-14-1630

18 Kalinsky K, Lee S, Rubim K, Lawrence DM, Dfrape J, Arge DR, Margolin KA, Leita MM Jr, Farhini AA, Koon HB, Pecora AL, Jaslowski AJ, Cohen GI, Kuzel TM, Lao CD and Kirkwood J: A phase 2 trial of dasatinib in patients with locally advanced or stage IV mucosal, acral, or vulvovaginal melanoma: A trial of the ECOG-ACRIN Cancer Research Group (E2607). Cancer 123: 2688-2697, 2017. PMID: 28334439. DOI: 10.1002/cncr.30663

19 Mao L, Cao Y, Sheng X, Bui X, Chi Z, Cui C, Wang X, Tang B, Tian B, Yan L, Li S, Zhou L, Wei X, Li X, Qi ZH, Si L, and Guo J: Palbociclib (P) in advanced acral lentiginous melanoma (ALM) with CDK4 pathway gene aberrations. J Clin Oncol 37: 9528-9528, 2019. DOI: 10.1200/JCO.2019.37.15_suppl.9528

20 Balch CM, Gershenson JD, Soong SJ, Thompson JF, Atkins MB, Byrd DR, Buzaid AC, Cochran AJ, Coit DG, Ding S, Eggermont AM, Flaherty KT, Gimotty PA, Kirkwood JM, McMasters KM, Mihm MC Jr, Morton DL, Ross ME, Sober AJ and Sondak VK: Final version of 2009 AJCC melanoma staging and classification. J Clin Oncol 27: 6199-6206, 2009. PMID: 19917835. DOI: 10.1200/JCO.2009.23.4799

21 Li H and Durbin R: Fast and accurate short read alignment with Burrows-Wheeler transform. Bioinformatics 25: 1754-1760, 2009. PMID: 19451168. DOI: 10.1093/bioinformatics/btp324

22 McKenna A, Hanna M, Banks E, Sivachenko A, Cibulskis K, Kentsyrs A, Garinelli K, Altshuler D, Gabriel S, Daly M and DePristo MA: The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. Genome Res 20: 1297-1303, 2010. PMID: 20644199. DOI: 10.1101/gr.107524.110

23 McLaren W, Gil L, Hunt SE, Riit HS, Ritchie GR, Thormann A, Flicek P and Cunningham F: The Ensembl variant effect predictor. Genome Biol 17: 122, 2016. PMID: 27268795. DOI: 10.1186/s13059-016-0974-4

24 Alexandrov LB, Kim J, Haradhvala NJ, Huang MN, Tian Ng AW, Wu Y, Boot A, Covington KR, Gordenin DA, Bergstrom EN, Islam SMA, Lopez-Bigas N, Klimczak LJ, McPherson JR, Morganella S, Sabarinathan R, Wheeler DA, Mustonen V, PCAWG Mutational Signatures Working Group, Getz G, Rozen SG, Stratton MR and PCAWG Consortium: The repertoire of mutational signatures in human cancer. Nature 578: 94-101, 2020. PMID: 32025018. DOI: 10.1038/s41586-020-1943-3

25 Merrel CH, Schumacher SE, Hill B, Meyerson ML, Beroukhim R and Getz G: GISTIC2.0 facilitates sensitive and confident localization of the targets of focal somatic copy-number alteration in human cancers. Genome Biol 12: R41, 2011. PMID: 21527027. DOI: 10.1186/gb-2011-12-4-41

26 Ha G, Roth A, Khattra J, Ho I, Yap D, Prentice LM, Melnyk N, McPherson A, Bashashati A, Laks E, Biele J, Ding J, Le A, Rosner J, Shumansky K, Marra MA, Gilks CB, Huntsman DG, McAlpine JN, Aparicio S and Shah SP: TITAN: inference of copy number architectures in clonal cell populations from tumor whole-genome sequence data. Genome Res 24: 1881-1893, 2014. PMID: 25060187. DOI: 10.1101/gr.180281

27 Noujaim J, Gonzalez D, Thway K, Jones RL and Judson I: p.(L576P) -KIT mutation in GIST: Favorable prognosis and sensitivity to imatinib? Cancer Biol Ther 17: 543-545, 2016. PMID: 26942271. DOI: 10.1080/15384047.2016.1156263

28 Long GV, Stroyakovskiy D, Gogas H, Levchenko E, de Braud F, Larkin J, Garbe C, Jouary T, Haushofer A, Grob J, Chiorion Silen V, Lebbe C, Mandala M, Millward M, Arance A, Bondarenko I, Haanen JB, Hansson J, Utikal J, Ferraresi V, Kovalenko N, Mohr P, Probachai V, Schadendorf D, Nathan P, Robert C, Ribas A, DeMarini DJ, Irani JG, Casey M, Ouellet D, Martin AM, Le N, Patel K and Flaherty K: Combined BRAF and MEK inhibition versus BRAF inhibition alone in melanoma. N Engl J Med 371: : 1877-1888, 2014. PMID: 25265492. DOI: 10.1056/NEJMoa1406037

29 Larkin J, Ascierto PA, Dréno B, Atkinson V, DeMarini DJ, Irani JG, Casey M, Ouellet D, Martin AM, Le N, Patel K and Flaherty K: Combined BRAF and MEK inhibition versus BRAF inhibition alone in melanoma. N Engl J Med 371: : 1877-1888, 2014. PMID: 25265492. DOI: 10.1056/NEJMoa1406037
vemurafenib and cobimetinib in BRAF-mutated melanoma. N Engl J Med 374: 1867-1876, 2014. PMID: 25265494. DOI: 10.1056/NEJMoa1408868

30 Robert C, Schachter J, Long GV, Arance A, Grob JJ, Mortier L, Daud A, Carlino MS, McNeil C, Lotem M, Larkin J, Lorigan P, Neyns B, Blank CU, Hamid O, Mateus C, Shapira-Frommer R, Koshy M, Zhou H, Ibrahim N, Ebbinghaus S, Ribas A and KEYNOTE-006 investigators: Pembrolizumab versus ipilimumab in advanced melanoma. N Engl J Med 372: 2521-2532, 2015. PMID: 25891173. DOI: 10.1056/NEJMoa1503093

31 Larkin J, Chiarion-Sileni V, Gonzalez R, Grob JJ, Rutkowski P, Lao CD, Cowey CL, Schadendorf D, Wagstaff J, Dummer R, Ferrucci PF, Smylie M, Hogg D, Hill A, Marquez-Rodas I, Haenen J, Guidoboni M, Maio M, Schöffski P, Carlino MS, Lebbé C, McArthur G, Atasekic PA, Daniels GA, Long GV, Bastholt L, Rizzo JI, Balogh A, Moshyk A, Hodi FS and Wolchok JD: Five-year survival with combined nivolumab and ipilimumab in advanced melanoma. N Engl J Med 381: 1535-1546, 2019. PMID: 31562797. DOI: 10.1056/NEJMoa1910836

32 Carvajal RD, Antonescu CR, Wolchok JD, Chapman PB, Roman RA, Teitcher J, Panagia KS, Busam KJ, Chmielowski B, Lutzky J, Pavlick AC, Fusco A, Cane L, Takebe N, Vemula S, Brough R, Bastholt L, Rizzo JI, Balogh A, Moshyk A, Hodi FS and Wolchok JD: Five-year survival with combined nivolumab and ipilimumab in advanced melanoma. N Engl J Med 372: 2521-2532, 2015. PMID: 25891173. DOI: 10.1056/NEJMoa1503093

33 Woodman SE, Trent JC, Stemke-Hale K, Lazar AJ, Pricl S, Pavan GM, Fermelegia M, Gopal YN, Yang D, Podoloff DA, Ivan D, Kim KB, Papadopoulos N, Hwu P, Mills GB and Davies MA: Activity of dasatinib against L576P KIT mutant melanoma: molecular, cellular, and clinical correlates. Mol Cancer Ther 8: 2079-2085, 2009. PMID: 19671763. DOI: 10.1158/1535-7163.MCT-09-0459

34 Kong Y, Sheng X, Wu X, Yan J, Ma M, Yu J, Si L, Chi Z, Cui C, Dai J, Li Y, Yu H, Xu T, Tang H, Tang B, Mao L, Lian B, Wang X, Yan X, Li S and Guo J: Frequent genetic aberrations in the CDK4 pathway in acral melanoma indicate the potential for CDK4/6 inhibitors in targeted therapy. Clin Cancer Res 23: 6946-6957, 2017. PMID: 28830923. DOI: 10.1158/1078-0432.CCR-17-0070

35 young RJ, Waldeck K, Duffin C, Foo JH, Cameron DP, Kirby L, Do H, Mitchell C, Cullinan C, Liu W, Fox SB, Dutton Regester K, Hayward NK, Jene N, Dobrovic A, Pearson RB, Christensen JG, Randolph S, McArthur GA and Sheppard KE: Loss of CDKN2A expression is a frequent event in primary invasive melanoma and correlates with sensitivity to the CDK4/6 inhibitor PD0332991 in melanoma cell lines. Pigment Cell Melanoma Res 27: 590-600, 2014. PMID: 24495407. DOI: 10.1111/pcmr.12228

36 Mateo J, Carreira S, Sandhu S, Miranda S, Moppiah H, Perez-Lopez R, Nava Rodrigues D, Robinson D, Omlin A, Tunariu N, Boysen G, Porta N, Flehr P, Gillman A, Figueiredo I, Paulding C, Seed G, Jain S, Ralph C, Protheroe A, Hussain S, Jones R, Elliott T, McGovern J, Bianchini D, Goodall J, Zafeiriou Z, Williamson CT, Ferraleschi R, Riisnaes R, Ebbbs F, Fowler G, Roda D, Yuan W, Wu YM, Cao X, Brough R, Permehiton H, A'Hern R, Swain A, Kunju LP, Eccles R, Attard G, Lord CJ, Ashworth A, Rubin MA, Knudsen KE, Feng FY, Chimnaiyan AM, Hall E and de Bono JS: DNA-repair defects and olaparib in metastatic prostate cancer. N Engl J Med 373: 1697-1708, 2015. PMID: 26510020. DOI: 10.1056/NEJMoa1506859

37 Nakamura Y, Namikawa K, Yoshino K, Yoshikawa S, Uchi H, Goto K, Nakamura Y, Fukushima S, Kiniwa Y, Takenouchi T, Ubara H, Kawai T, Hatta N, Funakoshi T, Teramoto Y, Otsuka A, Doi H, Ogata D, Matsushita S, Isei T, Hayashi T, Shibayama Y and Yamazaki N: Anti-PD1 checkpoint inhibitor therapy in acral melanoma: a multicenter study of 193 Japanese patients. Ann Oncol 31: 1198-1206, 2020. PMID: 32522691. DOI: 10.1016/j.annonc.2020.05.031