黃明祥先生 您好:

感謝您選用行動基因所提供的檢測服務。行動基因經您的同意，於西元 2021 年 08 月 16 日取得您的檢體，進行ACTOnco®+癌安克™癌症基因檢測與ACTFusion™癌融克™癌症融合基因檢測。行動基因實驗室為通過美國病理學會 (The College of American Pathologists，CAP) (CAP#：9028096) 與臺灣衛生福利部食品藥物管理署「精準醫療分子檢測列冊登錄實驗室」(Laboratory Developed Tests and Services, LDTS) (列冊登錄編號：LDTS0001) 的認證機構。

ACTOnco®+癌安克™癌症基因檢測平台利用次世代定序分析技術，同時檢測 440 個與腫瘤相關的基因變異，並計算腫瘤突變負荷量。

ACTFusion™癌融克™癌症融合基因檢測能檢測 13 個融合基因轉錄片段。

行動基因的專業生物與醫藥資訊團隊根據您的基因檢測結果與參考文獻，評估您對藥物的反應，輔助醫師進行治療與預後分析，以體現癌症精準醫療。

本次檢測於腫瘤檢體偵測到的重要基因變異及其相對應的標靶用藥如下:

基因變異	具敏感性之標靶用藥	具抗藥性之標靶用藥
• PIK3CA M1043I 基因突變 • STK11, TSC2 基因減少	• Everolimus	-
• PIK3CA M1043I 基因突變 • TSC2 基因減少	• Temsirolimus	-
• STK11 基因減少	• Trametinib	-
• PIK3CA M1043I 基因突變	• Lapatinib* • Trastuzumab* • Alpelisib	-

*研究結果顯示，帶有此基因變異者對此藥物的反應率較差。

腫瘤突變負荷量 (TMB): 1.9 mutations/megabase

微小衛星體不穩定性 (MSI): 穩定(stable)

融合基因: 未測得基因融合

詳細變異基因描述與用藥建議，請參閱以下完整基因檢測報告內容。

基因檢測報告所提供的資訊僅作為診療參考依據之一，您必須藉由醫師綜合評估過去的治療紀錄及專業判斷，選擇最適合您的治療策略。

若您對本檢測報告有任何疑問，請隨時與我們聯繫。

行動基因 敬上

ACTOnco®+ Report

REPORT SUMMARY

REPORT DETAILS

APPENDIX

PATIENT AND SAMPLE INFORMATION

PATIENT	SPECIMEN	ORDERING PHYSICIAN
Name: 黃明祥	Type: FFPE tissue	Name: 陳明晃醫師
Gender: Male	Date received: Aug 16, 2021	Facility: 臺北榮總
Date of Birth: Jul 05, 1966	Collection site: Lymph node	Tel: 886-228712121
Patient ID: 47155124	Specimen ID: S11021628	Address: 臺北市北投區石牌路二段 201 號
Diagnosis: Gallbladder adenosquamous cell carcinoma	Lab ID: AA-21-03344	Cooperative ID: NA
	D/ID: NA	

VARIANT(S) WITH CLINICAL RELEVANCE

Only variant(s) with clinical significance are listed. See the "DETAILED TEST RESULTS" section for full details.

SINGLE NUCLEOTIDE AND SMALL INDEL VARIANTS

Gene	Amino Acid Change	Coverage	Allele Frequency	COSMIC ID
MSH6	Splice donor/ Splice region	168	48.8%	COSM9494225
PIK3CA	M1043I	1651	15.3%	COSM94984
RBM10	E750*	218	24.3%	-
TP53	L194F	684	16.4%	COSM10995

COPY NUMBER VARIANTS (CNVS)

Loss of heterozygosity (LOH) information was used to infer tumor cellularity. Copy number alteration in the tumor was determined based on **30%** tumor purity.

Amplification (Copy number ≥ 8)

Chr	Gene	Copy Number
ND	ND	ND

Homozygous deletion (Copy number=0)

Chr	Gene
chr19	*STK11*

Heterozygous deletion (Copy number=1)

Chr	Gene
chr16	*TSC2*

ND, Not Detected

TUMOR MUTATIONAL BURDEN (TMB)

1.9 muts/Mb

Muts/Mb, mutations per megabase

MICROSATELLITE INSTABILITY (MSI)

Microsatellite stable (MSS)

Note:
TMB was calculated by using the sequenced regions of ACTOnco®+ to estimate the number of somatic nonsynonymous mutations per megabase of all protein-coding genes (whole exome). The threshold for high mutation load is set at ≥ 7.5 mutations per megabase. TMB, microsatellite status and gene copy number deletion cannot be determined if calculated tumor purity is < 30%.

Variant Analysis:
醫檢師張筑芃 博士
Chu-Yuan Chang Ph.D.
檢字第 020115 號

Sign Off
醫檢師張筑芃 博士
Chu-Yuan Chang Ph.D.
檢字第 020115 號

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THERAPEUTIC IMPLICATIONS

TARGETED THERAPIES

Genomic Alterations	Therapies	Effect
Level 3B		
PIK3CA M1043I	Alpelisib, Everolimus, Temsirolimus	**sensitive**
Level 4		
STK11 Homozygous deletion	Everolimus, Trametinib	**sensitive**
TSC2 Heterozygous deletion	Everolimus, Temsirolimus	**sensitive**
PIK3CA M1043I	Lapatinib, Trastuzumab	**less sensitive**

‡ Refer to "ONGOING CLINICAL TRIALS" section for detailed trial information.

Note: Therapies associated with benefit or lack of benefit are based on biomarkers detected in this tumor and published evidence.

Level		Description
1		FDA-recognized biomarker predictive of response to an FDA approved drug in this indication
2		Standard care biomarker (recommended as standard care by the NCCN or other expert panels) predictive of response to an FDA approved drug in this indication
3	A	Biomarkers that predict response or resistance to therapies approved by the FDA or professional societies for a different type of tumor
	B	Biomarkers that serve as inclusion criteria for clinical trials
4		Biomarkers that show plausible therapeutic significance based on small studies, few case reports or preclinical studies

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IMMUNE CHECKPOINT INHIBITORS (ICI) THERAPIES

Genomic markers and alterations that are associated with response to ICI therapies

Positive Biomarker	Negative Biomarker
TMB-H: ND	EGFR aberration: ND
MSI-H: ND	MDM2/MDM4 amplification: ND
MMR biallelic inactivation: ND	STK11 biallelic inactivation: **Yes**
PBRM1 biallelic inactivation: ND	PTEN biallelic inactivation: ND
SERPINB3/SERPINB4 mutation: ND	B2M biallelic inactivation: ND
	JAK1/2 biallelic inactivation: ND

MMR, mismatch repair; ND, not detected

Note: Tumor non-genomic factors, such as patient germline genetics, PDL1 expression, tumor microenvironment, epigenetic alterations or other factors not provided by this test may affect ICI response.

As mutational inactivation of STK11 has been shown to be a novel genomic predictor of de novo resistance to immune checkpoint blockade in tumor, homozygous deletion of STK11 in the tumor may render it **resistant** to immune checkpoint inhibitors [1][2][3][4].

CHEMOTHERAPIES

No genomic alterations detected in this tumor predicted to confer sensitivity or lack of benefit to chemotherapies.

HORMONAL THERAPIES

No genomic alterations detected in this tumor predicted to confer sensitivity or lack of benefit to hormonal therapies.

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OTHERS

Pharmacogenomic implication

Gene	Detection Site	Genotype	Drug Impact	Clinical Interpretation	Level of Evidence*
UGT1A1	rs4148323	AG	Irinotecan-based regimens	Patients with the AG genotype and cancer who are treated with irinotecan-based regimens may have an increased risk of diarrhea and neutropenia as compared to patients with the GG genotype, or a decreased risk of diarrhea and neutropenia compared to patients with the AA genotype. Other genetic and clinical factors may also influence a patient's risk of diarrhea and neutropenia.	**Level 1B**

* Level of evidence was defined by PharmGKB (https://www.pharmgkb.org/page/clinAnnLevels)

Level 1A: ***Clinical annotations describe variant-drug combinations that have variant-specific prescribing guidance available in a current clinical guideline annotation or an FDA-approved drug label annotation.***

Level 1B: ***Clinical annotations describe variant-drug combinations with a high level of evidence supporting the association but no variant-specific prescribing guidance in an annotated clinical guideline or FDA drug label.***

Level 2A: ***Variants in Level 2A clinical annotations are found in PharmGKB's Tier 1 Very Important Pharmacogenes (VIPs). These variants are in known pharmacogenes, implying causation of drug phenotype is more likely.***

Note:
Therapeutic implications provided in the test are based solely on the panel of 440 genes sequenced. Therefore, alterations in genes not covered in this panel, epigenetic and post-transcriptional and post-translational factors may also determine a patient's response to therapies. In addition, several other patient-associated clinical factors, including but not limited to, prior lines of therapies received, dosage and combinations with other therapeutic agents, patient's cancer types, sub-types, and/or stages, may also determine the patient's clinical response to therapies.

黃明祥

VARIANT INTERPRETATION

MSH6 Splice donor/Splice region

Biological Impact

The mutS homolog 6 (MSH6) gene encodes the DNA repair mismatch protein MSH6, a tumor suppressor involved in the DNA mismatch repair (MMR) [5]. Germline mutations in MMR pathway, including MSH6, are associated with susceptibility to hereditary nonpolyposis colon cancer (HNPCC), endometrial cancer [6] or sporadic cancers with microsatellite instability (MSI) [7].

MSH6 c.4001+12_4001+15del is a variant located at the splice donor region/splice region, which may result in the exon skipping.

Therapeutic and prognostic relevance

No drugs targeting MSH6 mutations have been currently approved. A screening test for microsatellite instability (MSI) is commonly used to identify a MMR-deficient tumor in clinic [8][9]. Tumors with mismatch-repair defects were associated with a large number of somatic mutations and have been predictive of clinical benefit to certain immune checkpoint blockade therapies [10][11]. The PD-1 antibodies pembrolizumab and nivolumab have been approved by the U.S. FDA for instability-high (MSI-H) or mismatch repair deficient (dMMR) solid tumors and MSI-H or dMMR colorectal cancer respectively. Down-regulation of genes involved in the MMR pathway such as MLH1, MSH2, and MSH6 in high-grade serous epithelial ovarian cancer cell lines rendered cells sensitive to PARP inhibitors [12].

PIK3CA M1043I

Biological Impact

The PIK3CA gene encodes the catalytic subunit (p110α) of phosphatidylinositol 3-kinase (PI3K) that plays a key role in the PI3K/AKT signaling pathway and is involved in the regulation of cellular functions such as proliferation, metabolism and protein synthesis, angiogenesis and apoptosis. PIK3CA has long been described as an oncogene and the PIK3CA gene amplification, deletion, and mutations have been reported in a wide range of cancers, including colorectal, breast, brain, liver, ovarian, stomach and lung cancers [13][14][15][16]. Mutations located in the exon 9 that encodes the PI3K helical (like E542K, E545K) and the exon 20 that encodes the catalytic/kinase domain (like H1047R, H1047L, H1047Y) have been shown to result in the constitutively activated mutant, which could enhance downstream signaling and oncogenic transformation in vitro and in vivo [14][17][18][19].

PIK3CA M1043I is a rare activating mutation that has been shown to result in increased kinase activity, lipid binding, phosphorylation of AKT and cell oncogenic transformation in vitro [20][21].

Therapeutic and prognostic relevance

In 2019 May, alpelisib, the PI3K inhibitor, in combination with fulvestrant received U.S. FDA approval for the treatment of postmenopausal women and men, with HR+, human epidermal growth factor receptor 2 (HER2)-negative, PIK3CA-mutated, advanced breast cancer following progression on or after an endocrine-based regimen. The approval is based on the results of the SOLAR-1 trial (NCT02437318), a randomized phase III trial of 572 patients with breast cancers showed that addition of alpelisib to fulvestrant significantly prolonged progression-free survival (median of 11 months vs. 5.7 months) in patients whose tumors had a PIK3CA mutation including C420R, E542K, E545A, E545D, E545G, E545K, Q546E, Q546R, H1047L, H1047R, and H1047Y [22].

An early phase study demonstrated that alpelisib has potential treatment benefits in a subset of PIK3CA-mutated tumors. Of the 114 patients with PIK3CA-mutated tumors, one patient with endometrial cancer achieved a complete response and seven had partial responses (including cervical, breast, endometrial, colon and rectal cancers). Besides, stable disease was observed in 70 patients and disease control rate was 58.2% [23].

Given that compared to patients with wild-type PIK3CA, those carrying PIK3CA mutations have a favorable response to mTOR inhibitors-containing monotherapy like everolimus and temsirolimus in several clinical studies of advanced malignancies [24][25][26][27][28] or temsirolimus in combination with doxorubicin and bevacizumab [29][30], combining PI3K-targeted agent with endocrine therapy was suggested.

Meanwhile, clinical benefit and response were observed when everolimus, an mTOR kinase inhibitor, was added to trastuzumab for the treatment of patients with HER2-overexpressing metastatic breast cancer, who progressed on trastuzumab-based therapy [31]. BOLERO-1 and BOLERO-3 (Breast Cancer Trials of Oral Everolimus) also suggested that patients with HER2-positive advanced breast cancer carrying PIK3CA mutations, PTEN loss, or hyperactive PI3K pathway could derive progression-free survival (PFS) benefit from everolimus [32]. In addition, BOLERO-3 study also showed that the addition of everolimus to trastuzumab plus vinorelbine significantly prolonged PFS in patients with trastuzumab-resistant and taxane-pretreated, HER2-positive, advanced breast cancer. However, the clinical benefit should be considered in the context of the adverse event profile in this population [33].

PIK3CA mutations have been determined as an inclusion criterion for the trials evaluating everolimus efficacy in patients with malignant glioma and different types of tumors (NCT03834740, NCT01827384) and trials examining temsirolimus in malignant solid tumors, multiple myeloma, B-cell non-Hodgkin lymphoma and malignant uterine neoplasm (NCT03297606, NCT02693535). On the other hand, there are several investigational PI3K inhibitors, including taselisib and buparlisib, which are currently in clinical development [34][35].

A retrospective study indicated that CRC patients with PIK3CA mutation and wild-type KRAS/BRAF showed fair responses to anti-EGFR therapies [36].

Hyperactivation of the PI3K signaling pathway is common and has been implicated in resistance to endocrine therapy [37][34] and HER2-targeting neoadjuvant therapies such as trastuzumab and lapatinib [38][39][40][41][42][43] in patients with advanced breast cancer.

Results from a clinical study showed that PIK3CA mutations occur in approximately 5% (2/37) of EGFR-mutated lung cancers who developed acquired resistance to EGFR TKI therapy [44]. Preclinical data also showed that PI3K/AKT/mTOR signaling was implicated in gefitinib resistance in EGFR-mutated lung cancer cell lines [45]. Although preclinical data suggested that addition of everolimus restored gefitinib sensitivity in NSCLC cancer cell lines [46][47], the efficacy was not evidenced in the clinical settings [48][49].

Two meta-analyses involving five studies demonstrated a significant correlation of PIK3CA mutations with better recurrence-free survival (RFS) in patients with unsorted breast cancer [50][51][41].

In patients with advanced EGFR- or KRAS-mutant lung adenocarcinoma, a concurrent PIK3CA mutation has been reported as a poor prognostic factor [52].

RBM10 E750*

Biological Impact

RBM10 (RNA binding motif protein 10) gene encodes a nuclear protein of the RNA-binding motif gene family which plays essential roles in alternative splicing [53][54]. Loss-of-function of RBM10 has been reported as the causes of TARP syndrome which results in pre- or postnatal death in affected males [55][56][57]. Mutations of RBM10 have been reported in lung adenocarcinoma [58][59][60], bladder and colorectal cancer [61].

E750* mutation results in a premature truncation of the RBM10 protein at amino acid 750 (UniProtKB). This mutation is predicted to lead to a loss of RBM10 function, despite not having characterized in the literature.

Therapeutic and prognostic relevance

Low expression of RBM10 was associated with shorter overall survival in lung adenocarcinoma patients [62].

TP53 L194F

Biological Impact

TP53 encodes the p53 protein, a crucial tumor suppressor that orchestrates essential cellular processes including cell cycle arrest, senescence and apoptosis [63]. TP53 is a proto-typical haploinsufficient gene, such that loss of a single copy of TP53 can result in tumor formation [64].

TP53 L194F lies within the DNA-binding domain (DBD) of the p53 protein (UniProtKB). This mutation has been reported [65], but not been biochemically characterized in the scientific literature; therefore, its effect on the p53 protein function remains unknown.

Therapeutic and prognostic relevance

Despite having a high mutation rate in cancers, there are currently no approved targeted therapies for TP53 mutations. A phase II trial demonstrated that Wee1 inhibitor (AZD1775) in combination with carboplatin was well tolerated and showed promising anti-tumor activity in TP53-mutated ovarian cancer refractory or resistant (< 3 months) to standard first-line therapy (NCT01164995) [66].

In a retrospective study (n=19), advanced sarcoma patients with TP53 loss-of-function mutations displayed improved progression-free survival (208 days versus 136 days) relative to patients with wild-type TP53 when treated with pazopanib [67]. Results from another Phase I trial of advanced solid tumors (n=78) demonstrated that TP53 hotspot mutations are associated with better clinical response to the combination of pazopanib and vorinostat [68].

Advanced solid tumor and colorectal cancer patients harboring a TP53 mutation have been shown to be more sensitive to bevacizumab when compared with patients harboring wild-type TP53 [69][70][71]. In a pilot trial (n=21), TP53-negative breast cancer patients demonstrated increased survival following treatment with bevacizumab in combination with chemotherapy agents, Adriamycin (doxorubicin) and Taxotere (docetaxel) [72]. TP53 mutations were correlated with poor survival of advanced breast cancer patients receiving tamoxifen or primary chemotherapy [73][74]. In a retrospective study of non-small cell lung cancer (NSCLC), TP53 mutations were associated with high expression of VEGF-A, the primary target of bevacizumab, offering a mechanistic explanation for why patients exhibit improved outcomes after bevacizumab treatment when their tumors harbor mutant TP53 versus wild-type TP53 [75].

STK11 Homozygous deletion

Biological Impact

The serine/threonine kinase 11 (STK11, also known as LKB1) gene encodes the multifunctional serine/threonine kinase, a tumor suppressor that functions as an inhibitor for the mTOR signaling pathway [76][77]. STK11 is a haploinsufficient gene with one copy loss may lead to weak protein expression and is insufficient to execute its original physiological functions [78][79]. In the mouse model, loss of STK11 promotes aggressive endometrial and squamous cell carcinomas [80][81]. Mutations in STK11 have been found in lung, breast, cervical, testicular, and liver cancers, as well as malignant melanoma, pancreatic and biliary carcinoma [82]. Germline mutations in STK11 are found in 30-70% of Peutz-Jeghers syndrome [83].

Therapeutic and prognostic relevance

A clinical study in a pancreatic cancer patient with Peutz-Jeghers syndrome whose tumor harboring an STK11 D194E mutation coupled with the loss of heterozygosity of the other STK11 allele displayed partial response to the everolimus treatment [84]. In another clinical case study, an adrenocorticotropic pituitary carcinoma patient whose tumor bearing an STK11 inactivating mutation responded to a combination of everolimus and radiotherapy [85].

Preclinical data suggested that lung cancer cell lines with STK11 inactivating mutations may confer increased sensitivity to the MEK-1 and MEK-2 inhibitor, trametinib [86].

Inactivating mutations of STK11 was shown to be associated with resistance to immune checkpoint blockade in KRAS-mutant lung adenocarcinoma (LUAC) (Journal of Clinical Oncology, 2017. 35(15_suppl): p. 9016-9016) [1][2] and NSCLC [3]. It was proposed that loss of STK11 negatively impacts the number and function of tumor-infiltrating T cells (TILs) and PD-L1 expression on tumor cells and therefore results in an ineffective response to PD-1-targeting antibodies [4].

TSC2 Heterozygous deletion

Biological Impact

The tuberous sclerosis complex 2 (TSC2) gene encodes a protein called tuberin, which interact with a protein called hamartin (encoded by the TSC1 gene). This hamartin-tuberin tumor suppressor complex plays a critical role in growth control as a negative regulator of the mammalian target of rapamycin (mTOR) pathway [87][88]. Mutations in TSC1/TSC2 tumor suppressor genes that result in inactivation of the complex are commonly found in patients with tuberous sclerosis complex [89][90][91], while the loss of heterozygosity (LOH) in TSC1/TSC2 has been identified in head and neck squamous cell carcinoma (HNSCC) [92] and endometrial cancer [93]. TSC2 deletion, splicing-mutant, and inactivating mutations such as A1141T, G305V, S1514X, and R1032X, has been identified in TSC2-null hepatocellular carcinoma (HCC) cell lines, patient-derived xenograft, and primary tumors. Mutations in the TSC1 and TSC2 genes cause the autosomal dominant genetic disorder tuberous sclerosis complex (TSC) [94].

Therapeutic and prognostic relevance

Genomic alterations with activating effects of the mTOR signaling pathway (including deletion/inactivation of TSC1/TSC2) have been shown to confer sensitivity to everolimus across multiple cancer types, such as bladder cancer, gastric cancer, sarcoma, thyroid cancer, hepatocellular carcinoma (HCC) as well as head and neck squamous cell carcinoma (HNSCC) [95][96][97]. Results from one Phase II study of advanced endometrial cancer showed that mutations in AKT1, TSC1, and TSC2 might predict sensitivity to temsirolimus [98]. Recent studies indicated that there are mTORC1-independent signaling pathways downstream of hamartin-tuberin, which may represent new therapeutic targets [99].

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US FDA-APPROVED DRUG(S)

Alpelisib (PIQRAY)

Alpelisib is an inhibitor of phosphatidylinositol-3-kinase (PI3K) with inhibitory activity predominantly against PI3Kα. Gain-of-function mutations in the gene encoding the catalytic α-subunit of PI3K (PIK3CA) lead to activation of PI3Kα and Akt-signaling, cellular transformation and the generation of tumors in in vitro and in vivo models. Alpelisib is developed and marketed by Novartis under the trade name PIQRAY.

FDA Approval Summary of Alpelisib (PIQRAY)

SOLAR-1[22] NCT02437318	**Breast cancer** (Approved on 2019/05/24)
	PIK3CA
	Alpelisib plus fulvestrant vs. Placebo plus fulvestrant [PFS(M): 11 vs. 5.7]

Everolimus (AFINITOR)

Everolimus, a derivative of sirolimus, works as an inhibitor of mammalian target of rapamycin complex 1 (mTORC1) and blocks mTORC1-mediated downstream signals for cell growth, proliferation, and survival. Everolimus is developed and marketed by Novartis under the trade name AFINITOR.

FDA Approval Summary of Everolimus (AFINITOR)

RADIANT-4[100] NCT01524783	**Lung or gastrointestinal neuroendocrine tumor** (Approved on 2016/02/26)
	-
	Everolimus vs. Placebo [PFS(M): 11 vs. 3.9]
BOLERO-2[101] NCT00863655	**Breast cancer** (Approved on 2012/07/20)
	ER+/HER2-
	Everolimus + exemestane vs. Placebo + exemestane [PFS(M): 7.8 vs. 3.2]
RADIANT-3[102] NCT00510068	**Pancreatic neuroendocrine tumor** (Approved on 2011/05/05)
	-
	Everolimus vs. Placebo [PFS(M): 11 vs. 4.6]
EXIST-1[103] NCT00789828	**Subependymal giant cell astrocytoma** (Approved on 2010/10/29)
	-
	Everolimus vs. Placebo [ORR(%): 35.0]

RECORD-1[104] NCT00410124	**Renal cell carcinoma** (Approved on 2009/05/30)
	-
	Everolimus vs. Placebo [PFS(M): 4.9 vs. 1.9]

Temsirolimus (TORISEL)

Temsirolimus is a soluble ester of sirolimus (rapamycin, brand-name drug Rapamune) and functions as an inhibitor of mammalian target of rapamycin complex (mTORC). The inhibitory molecular mechanism is similar to Everolimus. Temsirolimus is developed by Wyeth Pharmaceuticals and marketed by Pfizer under the trade name TORISEL.

FDA Approval Summary of Temsirolimus (TORISEL)

[105] NCT00065468	**Renal cell carcinoma** (Approved on 2007/05/30)
	-
	Temsirolimus vs. Ifn-α [OS(M): 10.9 vs. 7.3]

Trametinib (MEKINIST)

Trametinib is an anti-cancer inhibitor which targets MEK1 and MEK2. Trametinib is developed and marketed by GlaxoSmithKline (GSK) under the trade name MEKINIST.

FDA Approval Summary of Trametinib (MEKINIST)

BRF117019[106] NCT02034110	**Anaplastic thyroid cancer** (Approved on 2018/05/04)
	BRAF V600E
	Dabrafenib + trametinib [ORR(%): 61.0]
BRF113928[107] NCT01336634	**Non-small cell lung cancer** (Approved on 2017/06/22)
	BRAF V600E
	Trametinib + dabrafenib vs. Dabrafenib [ORR(%): 63.0 vs. 27.0, DOR(M): 12.6 vs. 9.9]
COMBI-d[108] NCT01584648	**Melanoma** (Approved on 2014/01/10)
	BRAF V600E/K
	Trametinib + dabrafenib vs. Dabrafenib + placebo [PFS(M): 9.3 vs. 8.8]

METRIC[109] NCT01245062	**Melanoma** (Approved on 2013/05/29)
	BRAF V600E/K
	Trametinib vs. Dacarbazine or paclitaxel [PFS(M): 4.8 vs. 1.5]

d=day; w=week; m=month

黃明祥

ONGOING CLINICAL TRIALS

Clinical trials shown below were selected by applying filters: study status, patient's diagnosis, intervention, location and/or biomarker(s). Please visit https://clinicaltrials.gov to search and view for a complete list of open available and updated matched trials.

No trial has been found.

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DETAILED TEST RESULTS

SINGLE NUCLEOTIDE AND SMALL INDEL VARIANTS

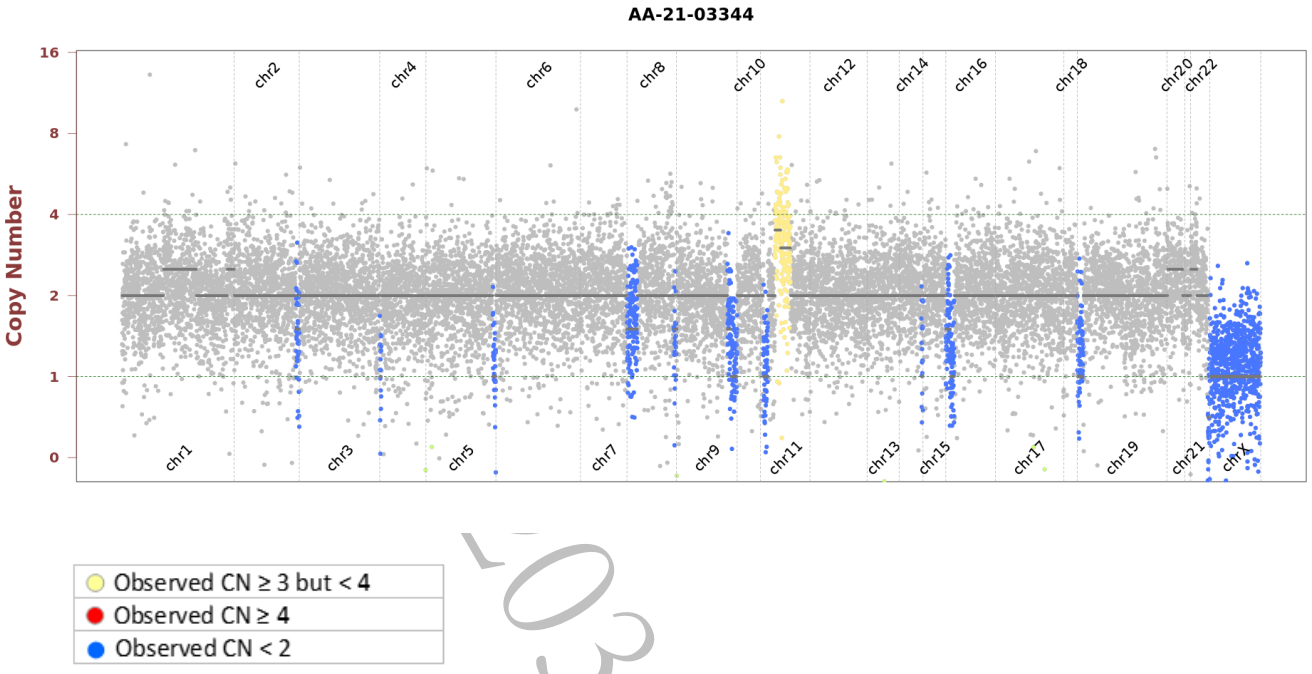
Gene	Chr	Exon	Accession Number	cDNA Change	Amino Acid Change	Coverage	Allele Frequency	COSMIC ID
ADAMTS13	9	-	NM_139025	c.173-7C>G	Splice region	585	45.8%	-
CCNA1	13	1	NM_001111045	c.108+1del	-	418	58.1%	-
FGF10	5	1	NM_004465	c.302C>T	T101I	238	12.6%	-
IRS2	13	1	NM_003749	c.3806A>C	Q1269P	169	39.1%	COSM5513012
MSH6	**2**	**-**	**NM_000179**	**c.4001+12_4001+15del**	**Splice donor/ Splice region**	**168**	**48.8%**	**COSM9494225**
PDGFRA	4	14	NM_006206	c.1907G>A	S636N	930	42.9%	-
PIK3CA	**3**	**21**	**NM_006218**	**c.3129G>C**	**M1043I**	**1651**	**15.3%**	**COSM94984**
POLD1	19	16	NM_001256849	c.1932C>G	D644E	564	43.4%	-
POLE	12	15	NM_006231	c.1524G>T	Q508H	599	50.8%	-
RBM10	**X**	**20**	**NM_005676**	**c.2247_2248delinsTT**	**E750***	**218**	**24.3%**	**-**
TP53	**17**	**6**	**NM_000546**	**c.580C>T**	**L194F**	**684**	**16.4%**	**COSM10995**

Mutations with clinical relevance are highlighted in red.

黃明祥
Project ID: C21-M001-00456
Report No.: AA-21-03344_ONC
Date Reported: Aug 27, 2021

COPY NUMBER VARIANTS (CNVS)

Observed copy number (CN) for each evaluated position is shown on the y-axis. Regions referred to as amplification or deletion are shown in color. Regions without significant changes are represented in gray.



黃明祥
Project ID: C21-M001-00456
Report No.: AA-21-03344_ONC
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HOTSPOT GENOTYPES

Listed variants are biomarkers or hotspots that are recommended as standard care by the NCCN or other expert panels and not necessarily FDA-recognized for a particular indication. The genotypes have been manually checked to ensure sufficient coverage for each hotspot of the target gene.

Gene	Variant	Genotype Detected
BRAF	V600X	Not detected
EGFR	A763_Y764insFQEA, E709K, E709_T710delinsD, Exon 19 deletion, Exon 19 insertion, Exon 20 insertion, G719A/C/D/S, L747P, L833V, L858R, L861Q/R, S768I, T790M	Not detected
IDH2	R140Q, R172G/K/M/S	Not detected
KIT	A502_Y503dup, D419del, D579del, D816F/V/Y, D820A/E/G/Y, E554_I571del, E554_K558del, E554_V559del, Exon 11 mutation, F522C, H697Y, I563_L576del, I653T, K550_W557del, K558N, K558_E562del, K558_V559del, K558delinsNP, K642E, M552_W557del, N505I, N564_Y578del, N822H/I/K/Y, P551_M552del, P573_D579del, P577_D579del, P577_W582delinsPYD, P838L, Q556_K558del, T417_D419delinsI, T417_D419delinsRG, T574_Q575insTQLPYD, V530I, V555_L576del, V555_V559del, V559A/C/D/G, V559_V560del, V559del, V560D/G, V560del, V569_L576del, V654A, W557G/R, W557_K558del, Y553N, Y553_K558del, Y570H, Y578C	Not detected
KRAS	A146T/V/P, G12X, G13X, Q61X	Not detected
MET	D1028H/N/Y	Not detected
NRAS	G12X, G13X, Q61X	Not detected
PDGFRA	A633T, C450_K451insMIEWMI, C456_N468del, C456_R481del, D568N, D842I/V, D842_H845del, D842_M844del, D846Y, E311_K312del, G853D, H650Q, H845Y, H845_N848delinsP, I843del, N659K/R/S, N848K, P577S, Q579R, R560_V561insER, R748G, R841K, S566_E571delinsR, S584L, V469A, V536E, V544_L545insAVLVLLVIVIISLI, V561A/D, V561_I562insER, V658A, W559_R560del, Y375_K455del, Y555C, Y849C/S	Not detected
PIK3CA	C420R, E542K/V, E545A/D/G/K, H1047X, Q546E/R	Not detected

V600X= any mutation in the valine (V) at amino acid 600 being replaced by a different amino acid.
G12X = any mutation in the glycine (G) at amino acid 12 being replaced by a different amino acid.
G13X= any mutation in the glycine (G) at amino acid 13 being replaced by a different amino acid.
Q61X = any mutation in the glutamine (Q) at amino acid 61 being replaced by a different amino acid.
H1047X = any mutation in the histidine (H) at amino acid 1047 being replaced by a different amino acid.

Gene	Copy Number Detected
CDK4	2
EGFR	2
ERBB2	2
MET	2

Copy number ≥ 8 is considered amplification

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Other known alterations that are associated with sensitivity, resistance, and toxicity to therapies.

Gene	Variant	Genotype Detected
AKT1	E17K	Not detected
ALK	C1156Y, D1203N, G1202R, L1152R, S1206Y, T1151_L1152insT	Not detected
BRAF	K601E, L597V/Q/R/S	Not detected
DPYD	D949V, I560S, splice-site mutation	Not detected
EGFR	A750P, C797S/Y, S492R	Not detected
ERBB2	V659E	Not detected
ESR1	D538G, E380Q, L469V, L536H/P/Q/R, S432L, S463P, V422del, V534E, Y537C/N/S	Not detected
FGFR3	G370C, G380R, K650E/N/R/M/T/Q, R248C, S249C, S371C, Y373C	Not detected
IDH1	R132C/G/H/Q/S	Not detected
MAP2K1	D67N, E203K, F53L, K57E/N, P124S, Q56P, Q56_V60del, R47Q, R49L, S222D	Not detected
PTEN	R130*/fs/G/L/P/Q	Not detected
TPMT	A154T, Y240C	Not detected

Gene	Copy Number Detected
FGFR1	2
MDM2	2
MDM4	2

Copy number ≥ 8 is considered amplification

TEST DETAILS

ABOUT ACTOnco®+

The test is a next-generation sequencing (NGS)-based assay developed for efficient and comprehensive genomic profiling of cancers. This test interrogates coding regions of 440 genes associated with cancer treatment, prognosis and diagnosis. Genetic mutations detected by this test include small-scale mutations like single nucleotide variants (SNVs), small insertions and deletions (INDELs) (≤ 15 nucleotides) and large-scale genomic alterations like copy number variations (CNVs).

See ACTOnco®+ Gene List' Section for details of gene sequenced.

DATABASE USED

- Reference genome: human genome sequence hg19
- COSMIC v.92
- Genome Aggregation database r2.1.1
- ClinVar (version 20210208)
- ACT Genomics in-house database

NEXT-GENERATION SEQUENCING (NGS) METHODS

Extracted genomic DNA was amplified using four pools of primer pairs targeting coding exons of analyzed genes. Amplicons were ligated with barcoded adaptors. Quality and quantity of amplified library were determined using the fragment analyzer (AATI) and Qubit (Invitrogen). Barcoded libraries were subsequently conjugated with sequencing beads by emulsion PCR and enriched using Ion Chef system (Thermo Fisher Scientific) according to the Ion PI Hi-Q Chef Kit protocol (Thermo Fisher Scientific) or Ion 540 Kit-Chef protocol (Thermo Fisher Scientific). Sequencing was performed on the Ion Proton or Ion S5 sequencer (Thermo Fisher Scientific).

Raw reads generated by the sequencer were mapped to the hg19 reference genome using the Ion Torrent Suite (version 5.10). Coverage depth was calculated using Torrent Coverage Analysis plug-in. Single nucleotide variants (SNVs) and short insertions/deletions (INDELs) were identified using the Torrent Variant Caller plug-in (version 5.10). The coverage was down-sampled to 4000. VEP (Variant Effect Predictor) (version 100) was used to annotate every variant using databases from Clinvar (version 20210208), COSMIC v.92 and Genome Aggregation database r2.1.1. Variants with coverage ≥ 25, allele frequency ≥ 5% and actionable variants with allele frequency ≥ 2% were retained.

This test provides uniform coverage of the targeted regions, enabling target base coverage at 100x ≥ 85% with a mean coverage ≥ 500x.

Variants reported in Genome Aggregation database r2.1.1 with > 1% minor allele frequency (MAF) were

considered as polymorphisms. ACT Genomics in-house database was used to determine technical errors. Clinically actionable and biologically significant variants were determined based on the published medical literature.

The copy number variations (CNVs) were predicted as described below:
Amplicons with read counts in the lowest 5th percentile of all detectable amplicons and amplicons with a coefficient of variation ≥ 0.3 were removed. The remaining amplicons were normalized to correct the pool design bias. ONCOCNV (an established method for calculating copy number aberrations in amplicon sequencing data by Boeva et al., 2014) was applied for the normalization of total amplicon number, amplicon GC content, amplicon length, and technology-related biases, followed by segmenting the sample with a gene-aware model. The method was used as well for establishing the baseline of copy number variations from samples in ACT Genomics in-house database.

Tumor mutational burden (TMB) was calculated by using the sequenced regions of ACTOnco®+ to estimate the number of somatic nonsynonymous mutations per megabase of all protein-coding genes (whole exome). The TMB calculation predicted somatic variants and applied a machine learning model with a cancer hotspot correction. TMB may be reported as “TMB-High”, “TMB-Low” or “Cannot Be Determined”. TMB-High corresponds to ≥ 7.5 mutations per megabase (Muts/Mb); TMB-Low corresponds to < 7.5 Muts/Mb. TMB is reported as “Cannot Be Determined” if the tumor purity of the sample is < 30%.

Classification of microsatellite instability (MSI) status is determined by a machine learning prediction algorithm. The change of a number of repeats of different lengths from a pooled microsatellite stable (MSS) baseline in > 400 genomic loci are used as the features for the algorithm. The final output of the results is either microsatellite Stable (MSS) or microsatellite instability high (MSI-H).

STANDARD OPERATING PROCEDURES (SOPS)

Standard operating procedures (SOPs) are shown below:

- AG2-QP-15 Specimen Management Procedure
- AG3-QP16-03 SOP of Cancer Cell DNA and RNA Extraction
- AG3-QP16-07 SOP of Nucleic Acid Extraction with QIAsymphony SP
- AG3-QP16-08 SOP of FFPE Nucleic Acid Extraction
- AG3-QP16-10 SOP of HE Staining
- AG3-QP16-13 SOP of Library Construction and Preparation
- AG3-QP16-17 SOP of DNA Quantification with Qubit Fluorometer
- AG3-QP16-20 SOP of CE-Fragment Analysis
- AG3-QP16-22 SOP of Variant Calling
- AG3-QP16-24 SOP of Ion Torrent System Sequencing Reaction
- AG3-QP16-26 SOP of Ion Chef Preparation

- AG3-QP16-35 SOP of Variant Annotation
- AG3-QP16-96 SOP of Manual Inspection for SNVIndel Variant
- AG3-QP16-95 SOP of Manual Inspection for Copy Number Variant
- AG3-QP40-08 (02) Standard protocol for variant interpretation, curation and classification
- AG3-QP16-41 SOP of The user manual for clinical report system (CRS)

LIMITATIONS

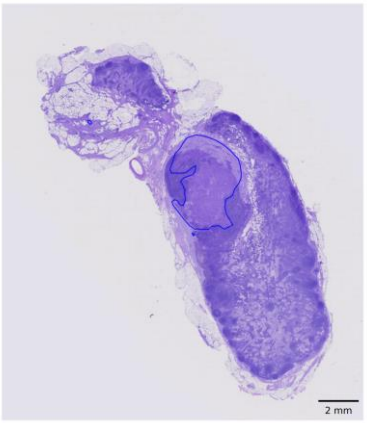
This test does not provide information of variant causality and does not detect variants in non-coding regions that could affect gene expression. This report does not report polymorphisms and we do not classify whether a mutation is germline or somatic. Variants identified by this assay were not subject to validation by Sanger or other technologies.

NOTES

We do not exclude the possibility that pathogenic variants may not be reported by one or more of the tools and the parameters used.

PATHOLOGY EVALUATION

- H&E-stained section No.: S11021628
- Collection site: Lymph node
- Examined by: Dr. Pei-Yi Chu
- Estimated neoplastic nuclei (whole sample): The percentage of viable tumor cells in total cells in the whole slide (%): 10%
  The percentage of viable tumor cells in total cells in the encircled areas in the whole slide (%): 80%
  The percentage of necrotic cells (including necrotic tumor cells) in total cells in the whole slide (%): 0%
  The percentage of necrotic cells (including necrotic tumor cells) in total cells in the encircled areas in the whole slide (%): 0%
  Additional comment: NA
- Manual macrodissection: Performed on the highlighted region



The outline highlights the area of malignant neoplasm annotated by a pathologist.

SPECIMEN PHOTO(S)

- Collection date: Jul 2021
- Facility retrieved: 臺北榮總

RUN QC

- Panel: ACTOnco®+
- Mean Depth: 819x
- Target Base Coverage at 100x: 92%

黃明祥
Project ID: C21-M001-00456
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ACTOnco®+ GENE LIST

ABCB1*	AURKB	CBL	CDKN2B	E2F3	FAT1	GRIN2A	JAK2	MED12	NOTCH4	PMS1	RAD51D	SLCO1B3*	TNFRSF14
ABCC2*	AXIN1	CCNA1	CDKN2C	EGFR	FBXW7	GSK3B	JAK3	MEF2B	NPM1	PMS2	RAD52	SMAD2	TNFSF11
ABCG2*	AXIN2	CCNA2	CEBPA*	EP300	FCGR2B	GSTP1*	JUN*	MEN1	NQO1*	POLB	RAD54L	SMAD3	TOP1
ABL1	AXL	CCNB1	CHEK1	EPCAM	FGF1*	GSTT1*	KAT6A	MET	NRAS	POLD1	RAF1	SMAD4	TP53
ABL2	B2M	CCNB2	CHEK2	EPHA2	FGF10	HGF	KDM5A	MITF	NSD1	POLE	RARA	SMARCA4	TPMT*
ADAMTS1	BAP1	CCNB3	CIC	EPHA3	FGF14	HIF1A	KDM5C	MLH1	NTRK1	PPARG	RB1	SMARCB1	TSC1
ADAMTS13	BARD1	CCND1	CREBBP	EPHA5	FGF19*	HIST1H1C*	KDM6A	MPL	NTRK2	PPP2R1A	RBM10	SMO	TSC2
ADAMTS15	BCL10	CCND2	CRKL	EPHA7	FGF23	HIST1H1E*	KDR	MRE11	NTRK3	PRDM1	RECQL4	SOCS1*	TSHR
ADAMTS16	BCL2*	CCND3	CRLF2	EPHB1	FGF3	HNF1A	KEAP1	MSH2	PAK3	PRKAR1A	REL	SOX2*	TYMS
ADAMTS18	BCL2L1	CCNE1	CSF1R	ERBB2	FGF4*	HR	KIT	MSH6	PALB2	PRKCA	RET	SOX9	U2AF1
ADAMTS6	BCL2L2*	CCNE2	CTCF	ERBB3	FGF6	HRAS*	KMT2A	MTHFR*	PARP1	PRKCB	RHOA	SPEN	UBE2A*
ADAMTS9	BCL6	CCNH	CTLA4	ERBB4	FGFR1	HSP90AA1	KMT2C	MTOR	PAX5	PRKCG	RICTOR	SPOP	UBE2K
ADAMTSL1	BCL9	CD19	CTNNA1	ERCC1	FGFR2	HSP90AB1	KMT2D	MUC16	PAX8	PRKCI	RNF43	SRC	UBR5
ADGRA2	BCOR	CD274	CTNNB1	ERCC2	FGFR3	HSPA4	KRAS	MUC4	PBRM1	PRKCQ	ROS1	STAG2	UGT1A1*
ADH1C*	BIRC2	CD58	CUL3	ERCC3	FGFR4	HSPA5	LCK	MUC6	PDCD1	PRKDC	RPPH1	STAT3	USH2A
AKT1	BIRC3	CD70*	CYLD	ERCC4	FH	IDH1	LIG1	MUTYH	PDCD1LG2	PRKN	RPTOR	STK11	VDR*
AKT2	BLM	CD79A	CYP1A1*	ERCC5	FLCN	IDH2	LIG3	MYC	PDGFRA	PSMB8	RUNX1	SUFU	VEGFA
AKT3	BMPR1A	CD79B	CYP2B6*	ERG	FLT1	IFNL3*	LMO1	MYCL	PDGFRB	PSMB9	RUNX1T1	SYK	VEGFB
ALDH1A1*	BRAF	CDC73	CYP2C19*	ESR1	FLT3	IGF1	LRP1B	MYCN	PDIA3	PSME1	RXRA	SYNE1	VHL
ALK	BRCA1	CDH1	CYP2C8*	ESR2	FLT4	IGF1R	LYN	MYD88	PGF	PSME2	SDHA	TAF1	WT1
AMER1	BRCA2	CDK1	CYP2D6	ETV1	FOXL2*	IGF2	MALT1	NAT2*	PHOX2B*	PSME3	SDHB	TAP1	XIAP
APC	BRD4	CDK12	CYP2E1*	ETV4	FOXP1	IKBKB	MAP2K1	NBN	PIK3C2B	PTCH1	SDHC	TAP2	XPO1
AR	BRIP1	CDK2	CYP3A4*	EZH2	FRG1	IKBKE	MAP2K2	NEFH	PIK3C2G	PTEN	SDHD	TAPBP	XRCC2
ARAF	BTG1*	CDK4	CYP3A5*	FAM46C	FUBP1	IKZF1	MAP2K4	NF1	PIK3C3	PTGS2	SERPINB3	TBX3	ZNF217
ARID1A	BTG2*	CDK5	DAXX	FANCA	GATA1	IL6	MAP3K1	NF2	PIK3CA	PTPN11	SERPINB4	TEK	
ARID1B	BTK	CDK6	DCUN1D1	FANCC	GATA2	IL7R	MAP3K7	NFE2L2	PIK3CB	PTPRD	SETD2	TERT	
ARID2	BUB1B	CDK7	DDR2	FANCD2	GATA3	INPP4B	MAPK1	NFKB1	PIK3CD	PTPRT	SF3B1	TET1	
ASXL1	CALR	CDK8	DICER1	FANCE	GNA11	INSR	MAPK3	NFKBIA	PIK3CG	RAC1	SGK1	TET2	
ATM	CANX	CDK9	DNMT3A	FANCF	GNA13	IRF4	MAX	NKX2-1*	PIK3R1	RAD50	SH2D1A*	TGFBR2	
ATR	CARD11	CDKN1A	DOT1L	FANCG	GNAQ	IRS1	MCL1	NOTCH1	PIK3R2	RAD51	SLC19A1*	TMSB4X*	
ATRX	CASP8	CDKN1B	DPYD	FANCL	GNAS	IRS2*	MDM2	NOTCH2	PIK3R3	RAD51B	SLC22A2*	TNF	
AURKA	CBFB	CDKN2A	DTX1	FAS	GREM1	JAK1	MDM4	NOTCH3	PIM1	RAD51C	SLCO1B1*	TNFAIP3	

*Analysis of copy number alteration not available.

DISCLAIMER

Legal Statement

This test was developed by ACT Genomics and its performing characteristics were determined by ACT Genomics. This test result is to be used for clinical consultative purposes only and is not intended as a substitute for a clinical guidance of your doctor or another qualified medical practitioner. It should not be regarded as investigational or used for research.

The detection of genomic alterations does not necessarily indicate pharmacologic effectiveness (or lack thereof) of any drug or treatment regimen; the detection of no genomic alteration does not necessarily indicate lack of pharmacologic effectiveness (or effectiveness) of any drug or treatment regimen.

Treatment Decisions are the Responsibility of the Physician

Decisions on clinical care and treatment should be based on the independent medical judgment of the treating physician, taking into consideration all applicable information concerning the patient's condition, including physical examinations, information from other diagnostics tests and patient preferences, in accordance with the standard of care in a given community. A treating physician's decisions should not be based on a single test, such as this test, or the information contained in this report.

In terms of consulting a different treating physician, the patient must file an application and fulfill the listed criteria for ACT Genomics to provide the patient's report to the assigned physician. The report may not be copied or reproduced except in its totality.

Genetic Alterations and Drugs Not Presented in Ranked Order

In this report, neither any biomarker alteration nor any drug associated with a potential clinical benefit (or potential lack of clinical benefit), are ranked in order of potential or predicted efficacy.

Level of Evidence Provided

Drugs with a potential clinical benefit (or potential lack of clinical benefit) are evaluated for level of published evidence with at least one clinical efficacy case report or preclinical study. We endeavor to keep the information in the report up to date. However, customers must be aware that scientific understanding and technologies change over time, and we make no warranty as to the accuracy, suitability or currency of information provided in this report at any time.

No Guarantee of Clinical Benefit

This report makes no promises or guarantees about the effectiveness of a particular drug or any treatment procedure in any disease or in any patient. This report also makes no promises or guarantees that a drug without an association of reportable genomic alteration will, in fact, provide no clinical benefit.

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本檢驗報告僅提供專業醫療參考，結果需經專業醫師解釋及判讀。基因突變資訊非必具備藥物或治療有效性指標，反之亦然。本檢驗報告提供之用藥指引不聲明或保證其臨床有效性，反之亦然。本基因檢測方法係由本公司研究開發，已經過有效性測試。

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醫療決策需由醫師決定

任何治療與用藥需經由醫師在考慮病患所有健康狀況相關資訊包含健檢、其他檢測報告和病患意願後，依照該地區醫療照護標準由醫師獨立判斷。醫師不應僅依據單一報告結果(例如本檢測或本報告書內容)做決策。

基因突變與用藥資訊並非依照有效性排序

本報告中列出之生物標記變異與藥物資訊並非依照潛在治療有效性排序。

證據等級

藥物潛在臨床效益(或缺乏潛在臨床效益)的實證證據是依據至少一篇臨床療效個案報告或臨床前試驗做為評估。本公司盡力提供適時及準確之資料，但由於醫學科技之發展日新月異，本公司不就本報告提供的資料是否為準確、適宜或最新作保證。

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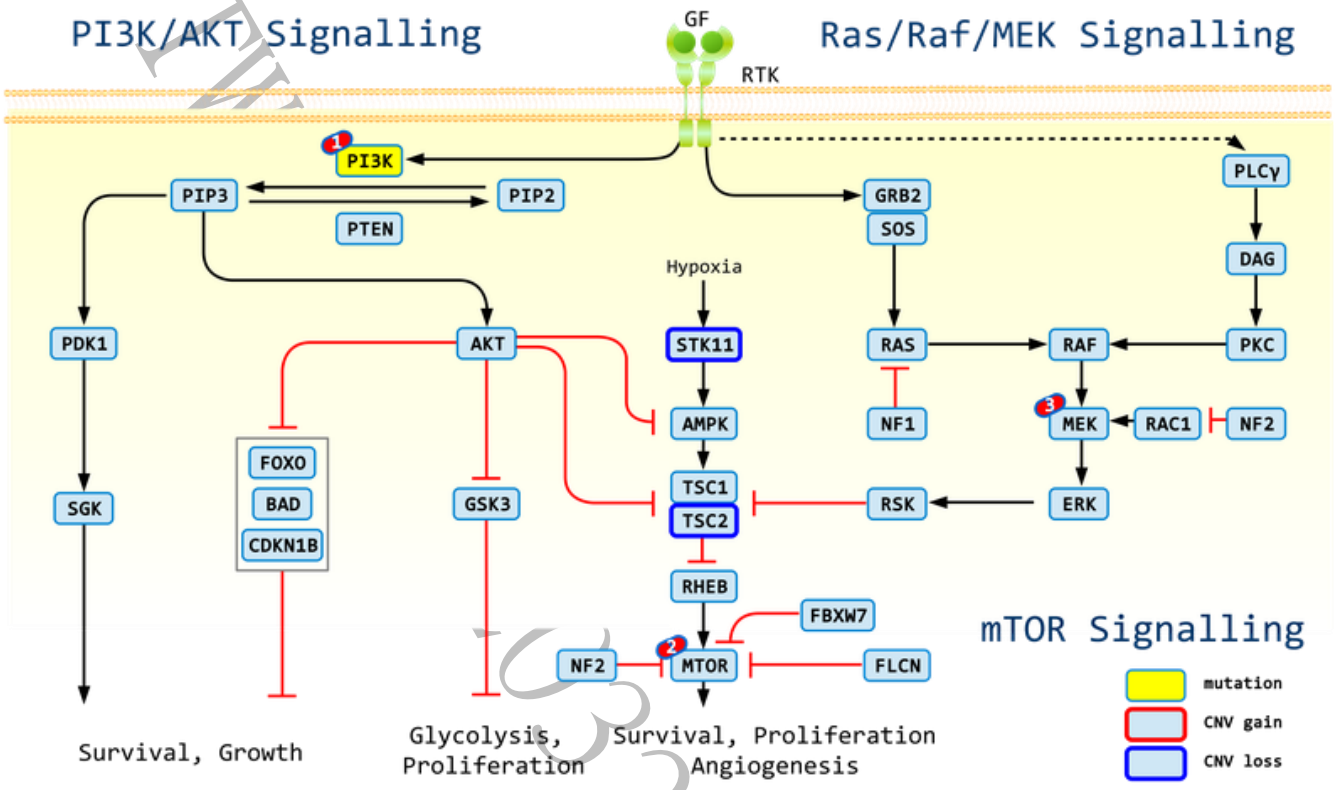
SIGNALING PATHWAYS AND MOLECULAR-TARGETED AGENTS



1: Alpelisib; 2: Everolimus, Temsirolimus; 3: Trametinib

REFERENCES

1. PMID: 29764856; 2018, Clin Cancer Res;24(22):5710-5723
TP53, STK11, and EGFR Mutations Predict Tumor Immune Profile and the Response to Anti-PD-1 in Lung Adenocarcinoma.

2. PMID: 29773717; 2018, Cancer Discov;8(7):822-835
STK11/LKB1 Mutations and PD-1 Inhibitor Resistance in KRAS-Mutant Lung Adenocarcinoma.

3. PMID: 29337640; 2018, J Clin Oncol;36(7):633-641
Molecular Determinants of Response to Anti-Programmed Cell Death (PD)-1 and Anti-Programmed Death-Ligand 1 (PD-L1) Blockade in Patients With Non-Small-Cell Lung Cancer Profiled With Targeted Next-Generation Sequencing.

4. PMID: 26833127; 2016, Cancer Res;76(5):999-1008
STK11/LKB1 Deficiency Promotes Neutrophil Recruitment and Proinflammatory Cytokine Production to Suppress T-cell Activity in the Lung Tumor Microenvironment.

5. PMID: 23391514; 2013, Mutat Res;743-744():53-66
Structural, molecular and cellular functions of MSH2 and MSH6 during DNA mismatch repair, damage signaling and other noncanonical activities.

6. PMID: 20028993; 2010, J Natl Cancer Inst;102(3):193-201
Risks of Lynch syndrome cancers for MSH6 mutation carriers.

7. PMID: 12658575; 2003, Am J Hum Genet;72(5):1088-100
Molecular analysis of hereditary nonpolyposis colorectal cancer in the United States: high mutation detection rate among clinically selected families and characterization of an American founder genomic deletion of the MSH2 gene.

8. PMID: 9823339; 1998, Cancer Res;58(22):5248-57
A National Cancer Institute Workshop on Microsatellite Instability for cancer detection and familial predisposition: development of international criteria for the determination of microsatellite instability in colorectal cancer.

9. PMID: 12454837; 2002, Gastroenterology;123(6):1804-11
Evaluation of tumor microsatellite instability using five quasimonomorphic mononucleotide repeats and pentaplex PCR.

10. PMID: 26028255; 2015, N Engl J Med;372(26):2509-20
PD-1 Blockade in Tumors with Mismatch-Repair Deficiency.

11. PMID: 25409260; 2014, N Engl J Med;371(23):2189-2199
Genetic basis for clinical response to CTLA-4 blockade in melanoma.

12. PMID: 27374179; 2017, Oncotarget;8(25):40152-40168
Cumulative defects in DNA repair pathways drive the PARP inhibitor response in high-grade serous epithelial ovarian cancer cell lines.

13. PMID: 16449998; 2006, Br J Cancer;94(4):455-9
Mutation of the PIK3CA oncogene in human cancers.

14. PMID: 15016963; 2004, Science;304(5670):554
High frequency of mutations of the PIK3CA gene in human cancers.

15. PMID: 12094235; 2002, Nat Rev Cancer;2(7):489-501
The phosphatidylinositol 3-Kinase AKT pathway in human cancer.

16. PMID: 9916799; 1999, Nat Genet;21(1):99-102
PIK3CA is implicated as an oncogene in ovarian cancer.

17. PMID: 16168105; 2005, Breast Cancer Res;7(5):R609-16
Somatic mutation and gain of copy number of PIK3CA in human breast cancer.

18. PMID: 16432179; 2006, Proc Natl Acad Sci U S A;103(5):1475-9
Cancer-specific mutations in PIK3CA are oncogenic in vivo.

19. PMID: 23411347; 2013, Trends Pharmacol Sci;34(3):149-53
Novel approaches to inhibitor design for the p110β phosphoinositide 3-kinase.

20. PMID: 22120714; 2012, Oncogene;31(32):3655-66
Regulation of lipid binding underlies the activation mechanism of class IA PI3-kinases.

21. PMID: 17376864; 2007, Proc Natl Acad Sci U S A;104(13):5569-74
Rare cancer-specific mutations in PIK3CA show gain of function.

22. PMID: 31091374; 2019, N Engl J Med;380(20):1929-1940
Alpelisib for PIK3CA-Mutated, Hormone Receptor-Positive Advanced Breast Cancer.

23. PMID: 29401002; 2018, J Clin Oncol;36(13):1291-1299
Phosphatidylinositol 3-Kinase α-Selective Inhibition With Alpelisib (BYL719) in PIK3CA-Altered Solid Tumors: Results From the First-in-Human Study.

24. PMID: 22271473; 2012, J Clin Oncol;30(8):777-82
PI3K/AKT/mTOR inhibitors in patients with breast and gynecologic malignancies harboring PIK3CA mutations.

25. PMID: 23066039; 2013, Cancer Res;73(1):276-84
PIK3CA mutation H1047R is associated with response to PI3K/AKT/mTOR signaling pathway inhibitors in early-phase clinical trials.

26. PMID: 24440717; 2014, Cell Rep;6(2):377-87
Assessing PIK3CA and PTEN in early-phase trials with PI3K/AKT/mTOR inhibitors.

27. PMID: 24748656; 2014, Mol Cancer Ther;13(5):1021-31
Picking the point of inhibition: a comparative review of PI3K/AKT/mTOR pathway inhibitors.

28. PMID: 25878190; 2015, Ann Oncol;26(7):1346-52
Inhibition of the phosphoinositide 3-kinase pathway for the treatment of patients with metastatic metaplastic breast cancer.

29. PMID: 22927482; 2012, Clin Cancer Res;18(20):5796-805
Phase I study of the antiangiogenic antibody bevacizumab and the mTOR/hypoxia-inducible factor inhibitor temsirolimus combined with liposomal doxorubicin: tolerance and biological activity.

30. PMID: 21890452; 2011, Clin Cancer Res;17(21):6840-6
A phase I trial of liposomal doxorubicin, bevacizumab, and temsirolimus in patients with advanced gynecologic and breast malignancies.

31. PMID: 21730275; 2011, J Clin Oncol;29(23):3126-32
Phase I/II study of trastuzumab in combination with everolimus (RAD001) in patients with HER2-overexpressing metastatic breast cancer who progressed on trastuzumab-based therapy.

32. PMID: 27091708; 2016, J Clin Oncol;34(18):2115-24
Molecular Alterations and Everolimus Efficacy in Human Epidermal Growth Factor Receptor 2-Overexpressing

Metastatic Breast Cancers: Combined Exploratory Biomarker Analysis From BOLERO-1 and BOLERO-3.

33. PMID: 24742739; 2014, Lancet Oncol;15(6):580-91
Everolimus for women with trastuzumab-resistant, HER2-positive, advanced breast cancer (BOLERO-3): a randomised, double-blind, placebo-controlled phase 3 trial.

34. PMID: 30651399; 2019, Oncologist;24(3):305-312
Exploring Biomarkers of Phosphoinositide 3-Kinase Pathway Activation in the Treatment of Hormone Receptor Positive, Human Epidermal Growth Receptor 2 Negative Advanced Breast Cancer.

35. PMID: 27986714; 2017, Cancer Discov;7(2):121
Benefit Mixed with Caution for Buparlisib.

36. PMID: 25714871; 2015, Mol Cancer Res;13(6):1003-8
AKT1 E17K in Colorectal Carcinoma Is Associated with BRAF V600E but Not MSI-H Status: A Clinicopathologic Comparison to PIK3CA Helical and Kinase Domain Mutants.

37. PMID: 29086897; 2018, Breast Cancer;25(4):392-401
Mechanism of resistance to endocrine therapy in breast cancer: the important role of PI3K/Akt/mTOR in estrogen receptor-positive, HER2-negative breast cancer.

38. PMID: 17936563; 2007, Cancer Cell;12(4):395-402
A functional genetic approach identifies the PI3K pathway as a major determinant of trastuzumab resistance in breast cancer.

39. PMID: 21594665; 2011, Breast Cancer Res Treat;128(2):447-56
Evaluation of the association of PIK3CA mutations and PTEN loss with efficacy of trastuzumab therapy in metastatic breast cancer.

40. PMID: 25559818; 2015, J Clin Oncol;33(12):1334-9
PIK3CA mutations are associated with decreased benefit to neoadjuvant human epidermal growth factor receptor 2-targeted therapies in breast cancer.

41. PMID: 26028978; 2015, Breast Cancer (Dove Med Press);7():111-23
PI3K mutations in breast cancer: prognostic and therapeutic implications.

42. PMID: 26920887; 2016, Clin Cancer Res;22(15):3755-63
Relationship between Tumor Biomarkers and Efficacy in EMILIA, a Phase III Study of Trastuzumab Emtansine in HER2-Positive Metastatic Breast Cancer.

43. PMID: 27177864; 2016, Ann Oncol;27(8):1519-25
PIK3CA mutations are associated with reduced pathological complete response rates in primary HER2-positive breast cancer: pooled analysis of 967 patients from five prospective trials investigating lapatinib and trastuzumab.

44. PMID: 21430269; 2011, Sci Transl Med;3(75):75ra26
Genotypic and histological evolution of lung cancers acquiring resistance to EGFR inhibitors.

45. PMID: 16906227; 2006, J Clin Invest;116(10):2695-706
Allelic dilution obscures detection of a biologically significant resistance mutation in EGFR-amplified lung cancer.

46. PMID: 19427302; 2009, Biochem Pharmacol;78(5):460-8
Everolimus restores gefitinib sensitivity in resistant non-small cell lung cancer cell lines.

47. PMID: 22941374; 2012, Cancer Chemother Pharmacol;70(5):707-16
Everolimus synergizes with gefitinib in non-small-cell lung cancer cell lines resistant to epidermal growth factor receptor tyrosine kinase inhibitors.

48. PMID: 24368400; 2014, Ann Oncol;25(2):409-15
Phase II study of everolimus-erlotinib in previously treated patients with advanced non-small-cell lung cancer.

49. PMID: 20871262; 2010, J Thorac Oncol;5(10):1623-9
Phase II trial of gefitinib and everolimus in advanced non-small cell lung cancer.

50. PMID: 24748804; 2014, Onco Targets Ther;7():543-52
PIK3CA mutations define favorable prognostic biomarkers in operable breast cancer: a systematic review and meta-analysis.

51. PMID: 25176561; 2014, Sci Rep;4():6255
Prognostic role of PIK3CA mutations and their association with hormone receptor expression in breast cancer: a meta-analysis.

52. PMID: 26334752; 2015, J Thorac Oncol;10(12):1713-9
Impact of Concurrent PIK3CA Mutations on Response to EGFR Tyrosine Kinase Inhibition in EGFR-Mutant Lung Cancers and on Prognosis in Oncogene-Driven Lung Adenocarcinomas.

53. PMID: 24530524; 2014, FEBS Lett;588(6):942-7
RBM10 regulates alternative splicing.

54. PMID: 24332178; 2013, Mol Cell;52(5):720-33
RBM5, 6, and 10 differentially regulate NUMB alternative splicing to control cancer cell proliferation.

55. PMID: 20451169; 2010, Am J Hum Genet;86(5):743-8
Massively parallel sequencing of exons on the X chromosome identifies RBM10 as the gene that causes a syndromic form of cleft palate.

56. PMID: 21910224; 2011, Am J Med Genet A;155A(10):2516-20
Long-term survival in TARP syndrome and confirmation of RBM10 as the disease-causing gene.

57. PMID: 24000153; 2013, EMBO Mol Med;5(9):1431-42
Integrative analysis revealed the molecular mechanism underlying RBM10-mediated splicing regulation.

58. PMID: 26853560; 2016, RNA Biol;13(4):466-72
Tumor suppressor properties of the splicing regulatory factor RBM10.

59. PMID: 22980975; 2012, Cell;150(6):1107-20
Mapping the hallmarks of lung adenocarcinoma with massively parallel sequencing.

60. PMID: 25079552; 2014, Nature;511(7511):543-50
Comprehensive molecular profiling of lung adenocarcinoma.

61. PMID: 28481359; 2017, Nat Med;23(6):703-713
Mutational landscape of metastatic cancer revealed from prospective clinical sequencing of 10,000 patients.

62. PMID: 28091594; 2017, Sci Rep;7():40488
Functional analysis reveals that RBM10 mutations contribute to lung adenocarcinoma pathogenesis by deregulating splicing.

63. PMID: 24739573; 2014, Nat Rev Cancer;14(5):359-70
Unravelling mechanisms of p53-mediated tumour suppression.

64. PMID: 21125671; 2011, J Pathol;223(2):137-46
Haplo-insufficiency: a driving force in cancer.

65. PMID: 30577483; 2018, Cancers (Basel);11(1):
Regulators of Oncogenic Mutant TP53 Gain of Function.

66. PMID: 27998224; 2016, J Clin Oncol;34(36):4354-4361
Phase II Study of WEE1 Inhibitor AZD1775 Plus Carboplatin in Patients With TP53-Mutated Ovarian Cancer Refractory or Resistant to First-Line Therapy Within 3 Months.

67. PMID: 26646755; 2016, Ann Oncol;27(3):539-43
TP53 mutational status is predictive of pazopanib response in advanced sarcomas.

68. PMID: 25669829; 2015, Ann Oncol;26(5):1012-8
Phase I study of pazopanib and vorinostat: a therapeutic approach for inhibiting mutant p53-mediated angiogenesis and facilitating mutant p53 degradation.

69. PMID: 27466356; 2016, Mol Cancer Ther;15(10):2475-2485
TP53 Alterations Correlate with Response to VEGF/VEGFR Inhibitors: Implications for Targeted Therapeutics.

70. PMID: 23670029; 2013, Oncotarget;4(5):705-14
P53 mutations in advanced cancers: clinical characteristics, outcomes, and correlation between progression-free survival and bevacizumab-containing therapy.

71. PMID: 17145525; 2006, Semin Oncol;33(5 Suppl 10):S8-14
Bevacizumab in combination with chemotherapy: first-line treatment of patients with metastatic colorectal cancer.

72. PMID: 21399868; 2011, Int J Oncol;38(5):1445-52
p53, HER2 and tumor cell apoptosis correlate with clinical outcome after neoadjuvant bevacizumab plus chemotherapy in breast cancer.

73. PMID: 20549698; 2011, Int J Cancer;128(8):1813-21
p53 status influences response to tamoxifen but not to fulvestrant in breast cancer cell lines.

74. PMID: 10786679; 2000, Cancer Res;60(8):2155-62
Complete sequencing of TP53 predicts poor response to systemic therapy of advanced breast cancer.

75. PMID: 25672981; 2015, Cancer Res;75(7):1187-90
VEGF-A Expression Correlates with TP53 Mutations in Non-Small Cell Lung Cancer: Implications for Antiangiogenesis Therapy.

76. PMID: 19029933; 2008, Oncogene;27(55):6908-19
LKB1; linking cell structure and tumor suppression.

77. PMID: 19584313; 2009, Physiol Rev;89(3):777-98
LKB1 and AMPK family signaling: the intimate link between cell polarity and energy metabolism.

78. PMID: 20142330; 2010, Dis Model Mech;3(3-4):181-93
Lkb1 inactivation is sufficient to drive endometrial cancers that are aggressive yet highly responsive to mTOR inhibitor monotherapy.

79. PMID: 17676035; 2007, Nature;448(7155):807-10
LKB1 modulates lung cancer differentiation and metastasis.

80. PMID: 18245476; 2008, Cancer Res;68(3):759-66
Loss of Lkb1 provokes highly invasive endometrial adenocarcinomas.

81. PMID: 18172296; 2008, Cancer Res;68(1):55-63
LKB1 deficiency sensitizes mice to carcinogen-induced tumorigenesis.

82. PMID: 25244018; 2014, Int J Mol Sci;15(9):16698-718
Recent progress on liver kinase B1 (LKB1): expression, regulation, downstream signaling and cancer suppressive function.

83. PMID: 9425897; 1998, Nat Genet;18(1):38-43
Peutz-Jeghers syndrome is caused by mutations in a novel serine threonine kinase.

84. PMID: 21189378; 2011, J Clin Oncol;29(6):e150-3
mTOR inhibitor treatment of pancreatic cancer in a patient With Peutz-Jeghers syndrome.

85. PMID: 27615706; 2016, CNS Oncol;5(4):203-9
Widely metastatic atypical pituitary adenoma with mTOR pathway STK11(F298L) mutation treated with everolimus therapy.

86. PMID: 27821489; 2017, Cancer Res;77(1):153-163
A Transcriptional Signature Identifies LKB1 Functional Status as a Novel Determinant of MEK Sensitivity in Lung Adenocarcinoma.

87. PMID: 21157483; 2011, Nat Rev Mol Cell Biol;12(1):21-35
mTOR: from growth signal integration to cancer, diabetes and ageing.

88. PMID: 12271141; 2002, Proc Natl Acad Sci U S A;99(21):13571-6
Tuberous sclerosis complex-1 and -2 gene products function together to inhibit mammalian target of rapamycin (mTOR)-mediated downstream signaling.

89. PMID: 9242607; 1997, Science;277(5327):805-8
Identification of the tuberous sclerosis gene TSC1 on chromosome 9q34.

90. PMID: 8269512; 1993, Cell;75(7):1305-15
Identification and characterization of the tuberous sclerosis gene on chromosome 16.

91. PMID: 1303246; 1992, Nat Genet;2(1):37-41
Linkage of an important gene locus for tuberous sclerosis to a chromosome 16 marker for polycystic kidney disease.

92. PMID: 18538015; 2008, BMC Cancer;8():163
Involvement of TSC genes and differential expression of other members of the mTOR signaling pathway in oral squamous cell carcinoma.

93. PMID: 28339086; 2017, Int J Oncol;50(5):1778-1784
Identification of novel mutations in endometrial cancer patients by whole-exome sequencing.

94. PMID: 17005952; 2006, N Engl J Med;355(13):1345-56
The tuberous sclerosis complex.

95. PMID: 22923433; 2012, Science;338(6104):221
Genome sequencing identifies a basis for everolimus sensitivity.

96. PMID: 26859683; 2016, Oncotarget;7(9):10547-56
Next-generation sequencing reveals somatic mutations that confer exceptional response to everolimus.

97. PMID: 25724664; 2015, Mol Cancer Ther;14(5):1224-35
Loss of Tuberous Sclerosis Complex 2 (TSC2) Is Frequent in Hepatocellular Carcinoma and Predicts Response to mTORC1 Inhibitor Everolimus.

98. PMID: 27016228; 2016, Gynecol Oncol;141(1):43-8
Tumor mutational analysis of GOG248, a phase II study of temsirolimus or temsirolimus and alternating megestrol

acetate and tamoxifen for advanced endometrial cancer (EC): An NRG Oncology/Gynecologic Oncology Group study.

99. PMID: 26412398; 2015, Sci Rep;5():14534
PAK2 is an effector of TSC1/2 signaling independent of mTOR and a potential therapeutic target for Tuberous Sclerosis Complex.

100. PMID: 26703889; 2016, Lancet;387(10022):968-977
Everolimus for the treatment of advanced, non-functional neuroendocrine tumours of the lung or gastrointestinal tract (RADIANT-4): a randomised, placebo-controlled, phase 3 study.

101. PMID: 22149876; 2012, N Engl J Med;366(6):520-9
Everolimus in postmenopausal hormone-receptor-positive advanced breast cancer.

102. PMID: 21306238; 2011, N Engl J Med;364(6):514-23
Everolimus for advanced pancreatic neuroendocrine tumors.

103. PMID: 23158522; 2013, Lancet;381(9861):125-32
Efficacy and safety of everolimus for subependymal giant cell astrocytomas associated with tuberous sclerosis complex (EXIST-1): a multicentre, randomised, placebo-controlled phase 3 trial.

104. PMID: 18653228; 2008, Lancet;372(9637):449-56
Efficacy of everolimus in advanced renal cell carcinoma: a double-blind, randomised, placebo-controlled phase III trial.

105. PMID: 17538086; 2007, N Engl J Med;356(22):2271-81
Temsirolimus, interferon alfa, or both for advanced renal-cell carcinoma.

106. PMID: 29072975; 2018, J Clin Oncol;36(1):7-13
Dabrafenib and Trametinib Treatment in Patients With Locally Advanced or Metastatic BRAF V600-Mutant Anaplastic Thyroid Cancer.

107. PMID: 27080216; 2016, Lancet Oncol;17(5):642-50
Dabrafenib in patients with BRAF(V600E)-positive advanced non-small-cell lung cancer: a single-arm, multicentre, open-label, phase 2 trial.

108. PMID: 25265492; 2014, N Engl J Med;371(20):1877-88
Combined BRAF and MEK inhibition versus BRAF inhibition alone in melanoma.

109. PMID: 22663011; 2012, N Engl J Med;367(2):107-14
Improved survival with MEK inhibition in BRAF-mutated melanoma.

ACTFusion™ Report

黃明祥
Project ID: C21-M001-00456
Report No.: AA-21-03344_FUS
Date Reported: Aug 27, 2021

PATIENT	SPECIMEN	ORDERING PHYSICIAN
Name: 黃明祥	Type: FFPE tissue	Name: 陳明晃醫師
Gender: Male	Date received: Aug 16, 2021	Facility: 臺北榮總
Date of Birth: Jul 05, 1966	Collection site: Lymph node	Tel: 886-228712121
Patient ID: 47155124	Specimen ID: S11021628	Address: 臺北市北投區石牌路二段 201 號
Diagnosis: Gallbladder adenosquamous cell carcinoma	Lab ID: AA-21-03344	Cooperative ID: NA
	D/ID: NA	

ABOUT ACTFusion™

The test is a next-generation sequencing (NGS) based in vitro diagnostic assay to detect fusion transcripts of 13 genes, including *ALK, BRAF, EGFR, FGFR1, FGFR2, FGFR3, MET, NRG1, NTRK1, NTRK2, NTRK3, RET,* and *ROS1*.

VARIANT(S) WITH CLINICAL RELEVANCE

FUSION RESULTS

No fusion gene detected in this sample.

Variant Analysis:
醫檢師張筑芃 博士
Chu-Yuan Chang Ph.D.
檢字第 020115 號

Sign Off
醫檢師張筑芃 博士
Chu-Yuan Chang Ph.D.
檢字第 020115 號

黃明祥
Project ID: C21-M001-00456
Report No.: AA-21-03344_FUS
Date Reported: Aug 27, 2021

THERAPEUTIC IMPLICATIONS

TARGETED THERAPIES

Not Applicable.

VARIANT INTERPRETATION

Not Applicable.

US FDA-APPROVED DRUG(S)

Not Applicable.

黃明祥
Project ID: C21-M001-00456
Report No.: AA-21-03344_FUS
Date Reported: Aug 27, 2021

ONGOING CLINICAL TRIAL(S)

Clinical trials shown below were selected by applying filters: study status, patient's diagnosis, intervention, location and/or biomarker(s). Please visit https://clinicaltrials.gov to search and view for a complete list of open available and updated matched trials.

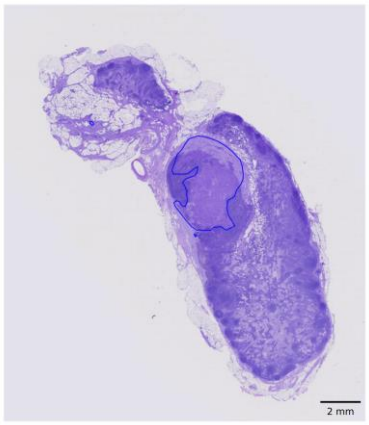
No trial has been found.

ACTFusion™ GENE LIST

ALK	*BRAF*	*EGFR*	*FGFR1*	*FGFR2*	*FGFR3*	*MET*	*NRG1*
NTRK1	*NTRK2*	*NTRK3*	*RET*	*ROS1*	-	-	-

TEST DETAILS

SPECIMEN RECEIVED



- H&E-stained section No.: S11021628
- Collection date: Jul 2021
- Collection site: Lymph node
- Facility retrieved: 臺北榮總
- Examined by: Dr. Pei-Yi Chu
- Estimated neoplastic nuclei (whole sample): The percentage of viable tumor cells in total cells in the whole slide (%): 10%
  The percentage of viable tumor cells in total cells in the encircled areas in the whole slide (%): 80%
  The percentage of necrotic cells (including necrotic tumor cells) in total cells in the whole slide (%): 0%
  The percentage of necrotic cells (including necrotic tumor cells) in total cells in the encircled areas in the whole slide (%): 0%
  Additional comment: NA
- Manual macrodissection: Performed on the highlighted region

The outline highlights the area of malignant neoplasm annotated by a pathologist.

黃明祥
Project ID: C21-M001-00456
Report No.: AA-21-03344_FUS
Date Reported: Aug 27, 2021

NEXT-GENERATION SEQUENCING (NGS) METHODS

The extracted RNA was reverse-transcribed and subjected to library construction. The quality and quantity of the amplified library was determined using the fragment analyzer (AATI) and Qubit (Invitrogen). Sequencing was performed on the Ion 540™ Chip/ Ion 550™ Chip / Ion P1™ Chip and Ion GeneStudio™ S5 Prime System / Ion Proton™ System (Life Technologies). All assays were performed in accordance with ACT Genomics testing SOPs.

Data processing and statistical analysis for the identification of relevant fusions was performed using in-house fusion calling pipeline with default parameter setting. The four internal controls for the purpose of monitoring the overall sequencing quality of the sample were built into the assay, including CHMP2A, RABA7A, GPI, and VCP. Amplification of these genes using gene specific primers was performed, and the sequencing results were applied to the analysis pipeline to assess RNA quality. The inability of the software to detect these genes was considered a run failure. To ensure optimal sequencing quality for variant analysis, all samples had to meet the following sample quality control (QC) criteria: 1) Average unique RNA Start Sites (SS) per control Gene Specific Primer 2 (GSP 2) ≥ 10 (default), and 2) Total reads after sequencing ≥ 500,000 (recommended).

Samples passed the sample QC would be subjected to the fusion analysis pipeline for fusion transcript calling. Briefly, the analysis pipeline aligned sequenced reads to a reference genome, identified regions that map to noncontiguous regions of the genome, and applied filters to exclude probable false-positive events and annotate previously characterized fusion events. A minimum of 5 reads with 3 unique sequencing start sites that cross the breakpoints was set as the cutoff value to indicate strong evidence of fusions. RNA fusions would need to be in frame in order to generate productive transcripts. In addition, databases with details for documented fusions were used to authenticate the fusion sequence identified. Known fusions were queried using Quiver Gene Fusion Database, a curated database owned and maintained by ArcherDX. In summary, samples with detectable fusions had to meet the following criteria: 1) Number of unique start sites (SS) for the GSP2 ≥ 3. 2) Number of supporting reads spanning the fusion junction ≥ 5. 3) Percentage of supporting reads spanning the fusion junction ≥ 10%. 4) Fusions annotated in Quiver Gene Fusion Database.

DATABASE USED

Quiver Gene Fusion Database version 5.1.18

LIMITATIONS

This test has been designed to detect fusions in 13 genes sequenced. Therefore, fusion in genes not covered by this test would not be reported. For novel fusions detected in this test, Sanger sequencing confirmation is recommended if residue specimen is available.

STANDARD OPERATING PROCEDURES (SOPs)

Standard operating procedures (SOPs) are shown below:

- AG2-QP-15 Specimen Management Procedure
- AG3-QP16-08 SOP of FFPE Nucleic Acid Extraction
- AG3-QP16-10 SOP of HE Staining
- AG3-QP16-17 SOP of DNA Quantification with Qubit Fluorometer
- AG3-QP16-20 SOP of CE-Fragment Analysis
- AG3-QP16-24 SOP of Ion Torrent System Sequencing Reaction
- AG3-QP16-26 SOP of Ion Chef Preparation
- AG3-QP40-08 (02) Standard protocol for variant interpretation, curation and classification
- AG3-QP16-94 (01) SOP of ACTFusion v3 Library Construction and Preparation
- AG3-QP16-36(02) SOP of Fusion Gene Detection
- AG3-QP16-41 SOP of The user manual for clinical report system (CRS)

RUN QC

- Panel: ACTFusion™
- Total reads: 1666642
- Average unique RNA Start Sites per control GSP2: 31

DISCLAIMER

Legal Statement

This test was developed by ACT Genomics and its performing characteristics were determined by ACT Genomics. This test result is to be used for clinical consultative purposes only and is not intended as a substitute for a clinical guidance of your doctor or another qualified medical practitioner. It should not be regarded as investigational or used for research.

The detection of genomic alterations does not necessarily indicate pharmacologic effectiveness (or lack thereof) of any drug or treatment regimen; the detection of no genomic alteration does not necessarily indicate lack of pharmacologic effectiveness (or effectiveness) of any drug or treatment regimen.

Treatment Decisions are the Responsibility of the Physician

Decisions on clinical care and treatment should be based on the independent medical judgment of the treating physician, taking into consideration all applicable information concerning the patient's condition, including physical examinations, information from other diagnostics tests and patient preferences, in accordance with the standard of care in a given community. A treating physician's decisions should not be based on a single test, such as this test, or the information contained in this report.

In terms of consulting a different treating physician, the patient must file an application and fulfill the listed criteria for ACT Genomics to provide the patient's report to the assigned physician. The report may not be copied or reproduced except in its totality.

Genetic Alterations and Drugs Not Presented in Ranked Order

In this report, neither any biomarker alteration nor any drug associated with a potential clinical benefit (or potential lack of clinical benefit), are ranked in order of potential or predicted efficacy.

Level of Evidence Provided

Drugs with a potential clinical benefit (or potential lack of clinical benefit) are evaluated for level of published evidence with at least one clinical efficacy case report or preclinical study. We endeavor to keep the information in the report up to date. However, customers must be aware that scientific understanding and technologies change over time, and we make no warranty as to the accuracy, suitability or currency of information provided in this report at any time.

No Guarantee of Clinical Benefit

This report makes no promises or guarantees about the effectiveness of a particular drug or any treatment procedure in any disease or in any patient. This report also makes no promises or guarantees that a drug without an association of reportable genomic alteration will, in fact, provide no clinical benefit.

Liability

ACT Genomics is not affiliated with any medical facility or medical practitioner. We provide information for informational purposes only, therefore, ACT Genomics and their employees cannot be held responsible for any direct, indirect, special, incidental or consequential damages that may arise from the use of information provided in the report.

免責聲明

法律聲明

本檢驗報告僅提供專業醫療參考，結果需經專業醫師解釋及判讀。基因突變資訊非必具備藥物或治療有效性指標，反之亦然。本檢驗報告提供之用藥指引不聲明或保證其臨床有效性，反之亦然。本基因檢測方法係由本公司研究開發，已經過有效性測試。

本檢驗報告非經本公司許可，不得私自變造、塗改，或以任何方式作為廣告及其他宣傳之用途。
本公司於提供檢驗報告後，即已完成本次契約義務，後續之報告解釋、判讀及用藥、治療，應自行尋求相關專業醫師協助，若需將報告移件其他醫師，本人應取得該醫師同意並填寫移件申請書，主動告知行動基因，行動基因僅能配合該醫師意願與時間提供醫師解說。

醫療決策需由醫師決定

任何治療與用藥需經由醫師在考慮病患所有健康狀況相關資訊包含健檢、其他檢測報告和病患意願後，依照該地區醫療照護標準由醫師獨立判斷。醫師不應僅依據單一報告結果(例如本檢測或本報告書內容)做決策。

基因突變與用藥資訊並非依照有效性排序

本報告中列出之生物標記變異與藥物資訊並非依照潛在治療有效性排序。

證據等級

藥物潛在臨床效益(或缺乏潛在臨床效益)的實證證據是依據至少一篇臨床療效個案報告或臨床前試驗做為評估。本公司盡力提供適時及準確之資料，但由於醫學科技之發展日新月異，本公司不就本報告提供的資料是否為準確、適宜或最新作保證。

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REFERENCES

Not Applicable.