Perthera Report Feedback

Patient: R-3403_28270607.1849

(1) Since receiving the Perthera report, have you initiated treatment (or do you plan to initiate treatment) with a therapy listed in the report?

☐ Yes. If yes, please check the box next to the therapy below:

- ☐ Vemurafenib + irinotecan + cetuximab/panitumumab
- ☐ A BRAF inhibitor and/or MEK/ERK inhibitor on a clinical trial - e.g., NCT02428712 (A Study of PLX8394 as a Single Agent in Patients With Advanced Unresectable Solid Tumors) or NCT02097225 (Onalespib, Dabrafenib, and Trametinib in Treating Patients With BRAF-Mutant Melanoma or Solid Tumors That Are Metastatic or Cannot Be Removed by Surgery) or NCT03377361 (An Investigational Immuno-therapy Study Of Nivolumab In Combination With Trametinib With Or Without Ipilimumab In Patients With Previously Treated Cancer of the Colon or Rectum That Has Spread)
- ☐ Vemurafenib/dabrafenib and/or trametinib/cobimetinib off label
- ☐ Regorafenib - e.g., NCT02466009 (Regorafenib in Metastatic Colorectal Cancer) or NCT02466802 (Study of Regorafenib and Sildenafil for Advanced Solid Tumors) or NCT01896856 (Phase I Study of SGI-110 With Irinotecan Followed by Randomized Phase II Study of SGI-110 With Irinotecan Versus Regorafenib or TAS-102 in Previously Treated Metastatic Colorectal Cancer)
- ☐ FOLFIRI or FOLFOXIRI +/- bevacizumab - e.g., NCT03314935 (A Phase 1/2 Study of INCB001158 in Combination With Chemotherapy in Subjects With Solid Tumors)
- ☐ Trifluridine + tipiracil - e.g., NCT03368963 (TAS102 in Combination With NAL-IRI in Advanced GI Cancers) or NCT02848079 (TAS-OX for Refractory Metastatic Colon Cancer)
- ☐ mTOR inhibitor on a clinical trial - e.g., NCT02124148 (A Study of Prexasertib (LY2606368) With Chemotherapy or Targeted Agents in Participants With Advanced Cancer) or NCT02890069 (A Study of PDR001 in Combination With LCL161, Everolimus or Panobinostat)
- ☐ Everolimus or temsirolimus off label
- ☐ A CHK1 inhibitor on a clinical trial - e.g., NCT03495323 (A Study of Prexasertib (LY2606368), CHK1 Inhibitor, and LY3300054, PD-L1 Inhibitor, in Patients With Advanced Solid Tumors) or NCT03057145 (Combination Study of Prexasertib and Olaparib in Patients With Advanced Solid Tumors) or NCT02873975 (A Study of LY2606368 (Prexasertib) in Patients With Solid Tumors With Replicative Stress or Homologous Repair Deficiency)
- ☐ A SINE/XPO1 inhibitor on a clinical trial - e.g., NCT02667873 (A Phase 1 Trial of a Novel XPO1 Inhibitor in Patients With Advanced Solid Tumors) or NCT02649790 (Study of the Safety, Tolerability and Efficacy of KPT-8602 in Patients With Relapsed/Refractory Cancer Indications)
- ☐ A Wnt inhibitor on a clinical trial - e.g., NCT02675946 (CGX1321 in Subjects With Advanced Solid Tumors and CGX1321 With Pembrolizumab in Subjects With Advanced GI Tumors (Keynote 596)) or NCT02521844 (A Study to Evaluate the Safety and Tolerability of ETC-1922159 in Advanced Solid Tumours) or NCT03355066 (A Study Evaluating the Safety and Pharmacokinetics of Orally Administered SM08502 in Subjects With Advanced Solid Tumors)
- ☐ Mutant p53-directed therapy on a clinical trial - e.g., NCT02617277 (Safety, Tolerability and Pharmacokinetics of AZD1775 Plus MEDI4736 in Patients With Advanced Solid Tumours) or NCT02576444 (OLAParib COmbinations)

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- ☐ An immunotherapy or tumor vaccine on a clinical trial - e.g., NCT02499328 (Study to Assess MEDI4736 With Either AZD9150 or AZD5069 in Advanced Solid Tumors & Relapsed Metastatic Squamous Cell Carcinoma of Head & Neck) or NCT02660034 (The Safety, Pharmacokinetics and Antitumor Activity of BGB-A317 in Combination With BGB-290 in Subjects With Advanced Solid Tumors) or NCT02646748 (Pembrolizumab Combined With Itacitinib (INCB039110) and/or Pembrolizumab Combined With INCB050465 in Advanced Solid Tumors)
- ☐ A non-biomarker-based clinical trial - e.g., NCT01483027 (Efficacy Evaluation of TheraSphere Following Failed First Line Chemotherapy in Metastatic Colorectal Cancer) or NCT02974738 (A Trial of PT2977 Tablets In Patients With Advanced Solid Tumors) or NCT03037385 (Phase 1 Study of the Highly-selective RET Inhibitor BLU-667 in Patients With Thyroid Cancer, Non-Small Cell Lung Cancer, and Other Advanced Solid Tumors)

ON LABEL OFF LABEL CLINICAL TRIAL

☐ No

(2) Please rate your overall experience with Perthera (circle one):

Poor Below Average Average Above Average Excellent

Comment: _____

(3) Would you refer another physician to use Perthera?

☐ Yes ☐ No

Comment: _____

We value any additional feedback you would like to give. If you'd like to be contacted by Perthera's medical director for further discussion, please leave your preferred contact information below:

☐ Email: _____

☐ Phone: _____

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SUMMARY

BRIEF HISTORY: A 72 year old female with recently diagnosed colon adenocarcinoma metastatic to the lung after resection in 1/2017 of a T3N2b right sided colon cancer (9/17 LN+, PNI, LVI, MS-stable, BRAFV600E) followed by adjuvant FOLFOX who is now on 1st line capecitabine.

SUMMARY OF GENOMIC FINDINGS: (Note this testing was done on the colonic primary)

- **BRAF** V600E activating mutation associated with excessive MAPK pathway activity and possible sensitivity to BRAF V600 inhibitors and MEK inhibitors (separately or in combination).
- **p53** inactivating mutation associated with impaired apoptotic responses to genomic and cellular damage and possible sensitivity to Wee1 and CHK1 inhibitors, although targeting p53 directly in colon cancer has not been successful to date.
- **FBXW7** inactivating mutation associated with impaired ubiquitin/proteasome degradation of target proteins and increased Wnt pathway activity, excessive mTORC1/2 pathway activity, and increased replicative stress leading to possible sensitivity to mTOR inhibitors, Wnt inhibitors, and CHK1 inhibitors.
- **TMB** intermediate associated with possible increase in neoantigens that can be recognized by the immune system and modestly increased response rates to immunotherapies, including checkpoint inhibitors.
- **Microsatellite stable**

SUMMARY OF PROTEOMIC FINDINGS: (Note that this testing was done on the lung biopsy)

- Possible decreased sensitivity to 5FU (TS positive) and gemcitabine (RRM1 positive)
- Possible increased sensitivity to platinum agents (ERCC1 low)
- pAKT positive
- HER2 negative by IHC and indeterminate by FISH
- PD-L1 negative by IHC (22C3, pembrolizumab antibody)
- No evidence of mismatch repair (MMR) enzyme deficiency

SUMMARY RECOMMENDATIONS: If this patient pursues therapy beyond capecitabine, therapies including dabrafenib and/or trametinib or other BRAF/MEK inhibitors in clinical trials are the primary consideration. Trials including mTOR inhibitors, Wnt inhibitors, CHK1 inhibitors, or SINE inhibitors could be considered.

THERAPEUTIC ASSOCIATIONS:

- **CLEAR** Associations:
  - **BRAF** V600E mutation = BRAF inhibitors, MEK inhibitors, ERK inhibitors
- **POSSIBLE** Associations:
  - **p53** inactivating mutation = Wee1 inhibitors, CHK1 inhibitors
  - **FBXW7** inactivating mutation = mTOR inhibitors, CHK1 inhibitors, exportin 1 inhibitors, Wnt inhibitors
  - **TMB** intermediate = immune checkpoint inhibitors
- **NO** Associations:
  - **MS**-Stable

STANDARD THERAPIES to consider:

- Vemurafenib and irinotecan plus panitumumab or cetuximab
- FOLFIRI (or FOLFOXIRI) +/- bevacizumab (or ziv-aflibercept or ramucirumab)
- Regorafenib
- Trifluridine plus tipiracil

OFF LABEL use of FDA APPROVED agents to consider:

- Vemurafenib/dabrafenib and/or trametinib/cobimetinib (BRAF V600E)
- Everolimus or temsirolimus (FBXW7, pAKT positive)

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SELECT CLINICAL TRIALS:

- **NCT03377361**; An Investigational Immuno-therapy Study Of Nivolumab In Combination With Trametinib With Or Without Ipilimumab In Patients With Previously Treated Cancer of the Colon or Rectum That Has Spread.
  - Rationale: BRAF V600E mutation, TMB-intermediate
- **NCT02097225**; Onalespib, Dabrafenib, and Trametinib in Treating Patients With BRAF-Mutant Melanoma or Solid Tumors That Are Metastatic or Cannot Be Removed by Surgery.
  - Rationale: BRAF V600E mutation
- **NCT02890069**; A Study of PDR001 in Combination With LCL161, Everolimus or Panobinostat.
  - Rationale: FBXW7 mutation, TMB-intermediate
- **NCT02873975**; A Study of LY2606368 (Prexasertib) in Patients With Solid Tumors With Replicative Stress or Homologous Repair Deficiency.
  - Rationale: FBXW7 mutation, p53 mutation

NCCN believes that the best management for any patient with cancer is in a clinical trial. Participation in clinical trials is especially encouraged.

The scores given to each treatment option are intended to highlight how strongly that treatment option aligns with the molecular findings, the available evidence, and patient history, but is NOT meant to indicate or imply that any one option has a greater chance of clinical success.

The addition of palliative care support services, where available, are recommended to help aggressively manage symptoms and side effects. Palliative care can be added as a complement to active treatment prior to terminal or end of life care.

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Perthera

CASE NUMBER	DIAGNOSIS	DATE
R-3403_28270607.1849	Colon Adenocarcinoma	24 August 2018

FINDINGS

Date of Birth	__/__/____	Client	_____	MRN	_____
Gender	**Female**			Specimen Date	**06/07/2018**
Case #	**R-3403_28270607.1849**	Physician	_____	Specimen Site	**Colon and Lung**

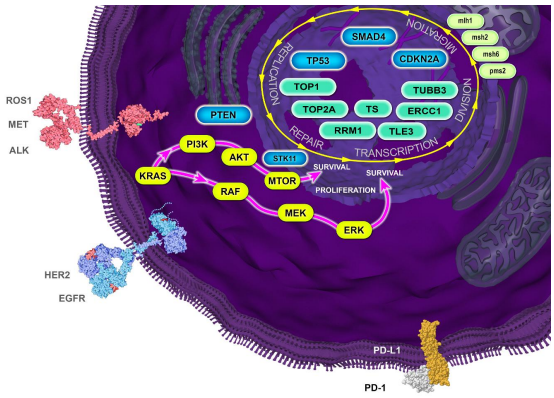
GENOMIC FINDINGS

Gene	Result	Biopsy
BRAF	**V600E**	Solid
FBXW7	**W649***	Solid
TP53	**E204***	Solid
ALK	Negative	Solid
HER2	Negative	Solid
MET	Negative	Solid
ROS1	Negative	Solid
TMB	intermediate	Solid
MSI Status	MSS	Solid

positive predictor
negative predictor
pathogenic alteration
pertinent negative

PROTEIN FINDINGS

Protein	Result	Int/%
pAKT	Positive	1 / 40%
ERCC1	Low	1 / 30%
RRM1	Positive	3 / 70%
TS	Positive	2 / 33%
HER2	Negative	1 / --
MLH1	No loss of expression	3 / 75%
MSH2	No loss of expression	3 / 80%
MSH6	Equivocal	3 / 48%
PD-L1	No expression	0 / 0%
PMS2	No loss of expression	3 / 70%



Cancer Signaling Map

RANKED THERAPY OPTIONS

ON LABEL | OFF LABEL | CLINICAL TRIAL

Therapy

A	**Vemurafenib + irinotecan + cetuximab/panitumumab** BRAF V600 mutation; This regimen was recently added to the NCCN guidelines for BRAF-mutant colon cancer; The patient has not had these agents
B	**A BRAF inhibitor and/or MEK/ERK inhibitor on a clinical trial - e.g., NCT02428712 or NCT02097225 or NCT03377361** BRAF V600 mutation; Activating BRAF mutations experimentally sensitize tumor cells to BRAF/MEK/ERK inhibitors; The patient has not had this class of agents ((51 miles) University of Miami - Miami, FL - Michael Pelayo, mpelayo@plexxikon.com)
C	**Vemurafenib/dabrafenib and/or trametinib/cobimetinib off label** BRAF V600 mutation; BRAF inhibitors combined with MEK inhibitors have demonstrated benefit in BRAF-mutant melanoma; The patient has not had this class of agents
D	**Regorafenib - e.g., NCT02466009 or NCT02466802 or NCT01896856** BRAF mutation; Regorafenib is a standard agent in this disease; The patient has not had this class of agents ((660 miles) University of North Carolina at Chapel Hill - Chapel Hill, NC - Grant R Williams, MD, Grant.Williams@unchealth.unc.edu, 919-966-0000)
E	**FOLFIRI or FOLFOXIRI +/- bevacizumab - e.g., NCT03314935** ERCC1 low, TS positive; These are standard combinations for colorectal cancer; The patient has had adjuvant FOLFOX ((954 miles) The University of Texas MD Anderson Cancer Center - Houston, TX - Incyte Corporation Call Center (US), medinfo@incyte.com, 1.855.463.3463)
F	**Trifluridine + tipiracil - e.g., NCT03368963 or NCT02848079** No implicated markers; Standard agents; The patient has not had this class of agents ((567 miles) Emory University Hospital Midtown - Atlanta, GA - Shabnam Montazeri, shabnam.montazeri@emory.edu, 404-686-0242)
G	**mTOR inhibitor on a clinical trial - e.g., NCT02124148 or NCT02890069** FBXW7 mutation, pAKT positive; PI3K/AKT/mTOR inhibitors have not shown clinical activity as single agents; Patient has not had this type of therapy ((153 miles) Florida Cancer Specialists - Sarasota, FL - 941-377-9993)
H	**Everolimus or temsirolimus off label** FBXW7 mutation, pAKT positive; mTOR inhibitors have shown clinical activity in a variety of tumors with PI3K/AKT/mTOR pathway alterations; Patient has not had this type of therapy
I	**A CHK1 inhibitor on a clinical trial - e.g., NCT03495323 or NCT03057145 or NCT02873975** FBXW7 mutation, p53 mutation; This class of agents has limited clinical evidence; The patient has not had this class of agents ((1206 miles) Dana Farber Cancer Institute - Boston, MA - Adrienne Anderson, RN, BSN, aanderson10@partners.org, 617-632-6594)

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Therapy

J	**A SINE/XPO1 inhibitor on a clinical trial - e.g., NCT02667873 or NCT02649790** FBXW7 mutation; This class of agents has limited clinical evidence in solid tumors; The patient has not had this class of agents ((136 miles) Florida Cancer Specialist - Sarasota, FL - Shay Shemesh, MS, Trials@stemline.com, 646-502-2310)
K	**A Wnt inhibitor on a clinical trial - e.g., NCT02675946 or NCT02521844 or NCT03355066** FBXW7 mutation; This class of agents has limited clinical evidence; The patient has not had this class of agents ((873 miles) Lombardi Comprehensive Cancer Center - Washington, DC - Jennifer Montcalm, jem257@georgetown.edu, 202-687-8974)
L	**Mutant p53-directed therapy on a clinical trial - e.g., NCT02617277 or NCT02576444** FBXW7 mutation, p53 mutation; Targeting p53 directly has not been successful to date; Patient has not had this type of therapy ((153 miles) Research Site - Sarasota, FL - AstraZeneca Clinical Study Information Center, information.center@astrazeneca.com, 1-877-240-9479)
M	**An immunotherapy or tumor vaccine on a clinical trial - e.g., NCT02499328 or NCT02660034 or NCT02646748** TMB intermediate; Immunotherapy is a promising approach in many cancers; The patient has not had this class of agents ((29 miles) Research Site - Plantation, FL - AstraZeneca Clinical Study Information Center, information.center@astrazeneca.com, 1-877-240-9479)
N	**A non-biomarker-based clinical trial - e.g., NCT01483027 or NCT02974738 or NCT03037385** No predictive biomarkers are being used for eligibility; Activity is unknown; Patient has not had these experimental agents ((12 miles) Lynn Clinical Research Center, Boca Raton Regional Hospital - Boca Raton, FL - Sylvie Godbout, sgodbout@brrh.com, 561-955-4539)

Perthera treatment scoring system uses detailed molecular, disease-specific and patient medical history to rank therapies that have the highest probability of best outcome for the patient. Higher scores do not guarantee a greater chance of treatment success. Other considerations must be taken into account by the treating Physician.

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PATIENT CONSIDERATIONS

Only those patient factors provided to Perthera have been considered.

Treating oncologist will consider complete patient history and current conditions.

RELEVANT MEDICAL HISTORY

- 72 year old female with recently diagnosed colon cancer metastatic to the lung.
- Originally had resection of T3N2b colon cancer in 1/2017 (MS-stable, BRAFV600E).
- She received adjuvant FOLFOX 2/2017 to 7/2017.
- Small pulmonary nodules were first discovered in 2/2018 but were small and were initially followed expectantly but enlarged prompting recent needle biopsy.
- She has been placed on capecitabine as a single agent given that progression is slow and she has extensive travel plans.
- **This analysis is based on a specimen (Colon) from 05 Jan 2017**

PERTHERA REPORT THERAPY SCORING MODEL

Molecular Rationale and Scoring: 0-3

0- No predictive biomarkers

1- Preclinical evidence or conflicting clinical evidence of predictive value of this biomarker

2- Mixed evidence (pre-clinical and clinical) of predictive value of this biomarker

3- Clear clinical evidence of the predictive value of this biomarker

Disease Rationale and Scoring: 0-4

0- Agents tested in this cancer type and failed to demonstrate any activity/benefit

1- Agents with limited clinical evidence (or only pre-clinical evidence)

2- Agents with promising clinical evidence

3- Agents with strong clinical evidence in another indication (off-label)

4- Agents with strong clinical evidence in this cancer type (on-label)

Patient Rationale and Scoring: 0-2

0- Patient's disease progressed on, or patient is currently receiving, this drug/drug combination

1- Patient previously exposed to this drug/drug combination >6 months ago without disease progression

2- Patient was never exposed to this drug/drug combination

The Perthera scoring model was developed using best practices for scoring predictive somatic variants published by ClinGen-{PMID: 27814769}, Association of Molecular Pathologists-{PMID: 27993330} and OncoKB- {DOI: 10.1200}

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THERAPEUTIC ASSOCIATIONS

Genomics

- **BRAFV600E** mutation was found. BRAF encodes a serine/threonine-protein kinase called B-Raf. The B-Raf protein is involved in sending signals inside cells which are involved in directing cell growth. B-Raf phosphorylates MAP2K leading to activation of the MAP kinase signal transduction pathway. BRAF mutations in pancreatic cancer have been shown to be mutually exclusive with KRAS mutations [1]. The V600E mutation results in an amino acid substitution at position 600 resulting in increased kinase activity and is transforming in vitro. BRAF V600E mutations are associated with increased sensitivity to BRAF inhibitors, vemurafenib and dabrafenib [2] and the MEK inhibitors, trametinib or cobimetinib. Combination therapy with dabrafenib and trametinib is FDA approved for melanoma patients with V600E or V600K mutations based on phase I/II and phase III trials showing improved response rates and response duration compared to vemurafenib [3] or dabrafenib alone [4],[5]. Similarly, vemurafenib and cobimetinib [6] show improved response rates and progression-free survivals compared to vemurafenib alone. A clinical trial including a BRAF inhibitor and a MEK inhibitor in combination could be considered.
- **FBXW7** inactivating mutation was found. FBXW7 has been shown to function as a tumor suppressor through loss of its ubiquitin/proteosome degradation of multiple important oncoproteins, including c-Myc, c-Jun, cyclinE, HIF-1, mTOR, Mcl-1, NF-kB2, Notch-1, Aur-K, RAS, BCL-2, Eno1, c-Myb, and p63 [7]. FBXW7 has also been shown to be a regulator of WNT/beta-catenin signaling, and inactivation of FBXW7 in pancreatic cancer tissues has been shown to lead to aberrant activation of WNT signaling through targeting beta-catenin for degradation [8]. FBXW7 specifically regulates c-Myc by recognizing a GSK3-beta-mediated phosphorylated threonine residue, leading to increased c-Myc turnover [9]. Experimental studies have linked loss of FBXW7 function with elevated mTOR function [10], suggesting that patients with this mutation may have increased sensitivity to mTOR inhibitors; however, few responses have been observed in FBXW7-mutated patients [11]. Alterations in FBXW7 have also been associated with increased sensitivity to CHK1 inhibitors due to increased replicative stress. Thus, a clinical trial including a WNT inhibitor, a CHK1 inhibitor, an mTOR inhibitor, or a SINE inhibitor, such as selinexor, could be considered.
- **TP53** (p53) inactivating mutation was present. The p53 system acts as a tumor suppressor by normally controlling cellular fate after DNA damaging exposures. Dysregulation of p53 confers tumor resistance to programmed cell death despite DNA changes. p53 is dysregulated in the majority of cancers, and there are currently no clinically successful agents directly reversing p53 dysregulation. Experimentally, p53-deficient tumors may be selectively sensitive to Wee1 inhibitors [12],[13],[14],[15], CHK1 inhibitors [12]), or p53-directed vaccine therapies.
- **TMB-Intermediate:** An increased tumor mutational burden (TMB) has been shown to be a response biomarker for PD-1/PD-L1 blockade in tumors such as melanoma and non-small cell lung cancer (NSCLC) and higher TMB has been shown to be independently associated with better outcome parameters across a broad range of tumors [16],[17]. The association if intermediate TMB (5-15 mut/mb) has been less clear but is suggested in some tumor types. TMB has not been specifically validated as a therapeutically predictive biomarker in colon adenocarcinoma.

Proteomics

Positive Predictors

- **Platinum agents**: ERCC1 expression was low by IHC, suggesting potentially increased responsiveness to platinum agents [18],[19],[20],[21].
- **PI3K/AKT/mTOR inhibitors**: pAKT positivity suggests activity of the PI3K/AKT/mTOR pathway [22];[23] as predicted with FXBW7 mutation.

Negative Predictors

- **Gemcitabine**: RRM1 expression was positive by IHC. Despite evidence supporting the role of RRM1 expression as a biomarker of responsiveness to gemcitabine [24],[25],[26],[27], recent clinical trials have demonstrated that RRM1 expression did not correlate with PFS or OS in treating NSCLC with gemcitabine [28],[29].
- **5FU**: TS expression was positive by IHC, suggesting a possible LACK of responsiveness to 5FU-based therapies [30],[31],[18][32].
- **Checkpoint Inhibitors**: No evidence of mismatch repair (MMR) deficiency, PD-L1 negative, and MS-stable.

Testing Performed at Foundation Medicine and NeoGenomics

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