Predictive Biomarkers in GI tract tumors

Are we ready for 'off-label' use of IHC?

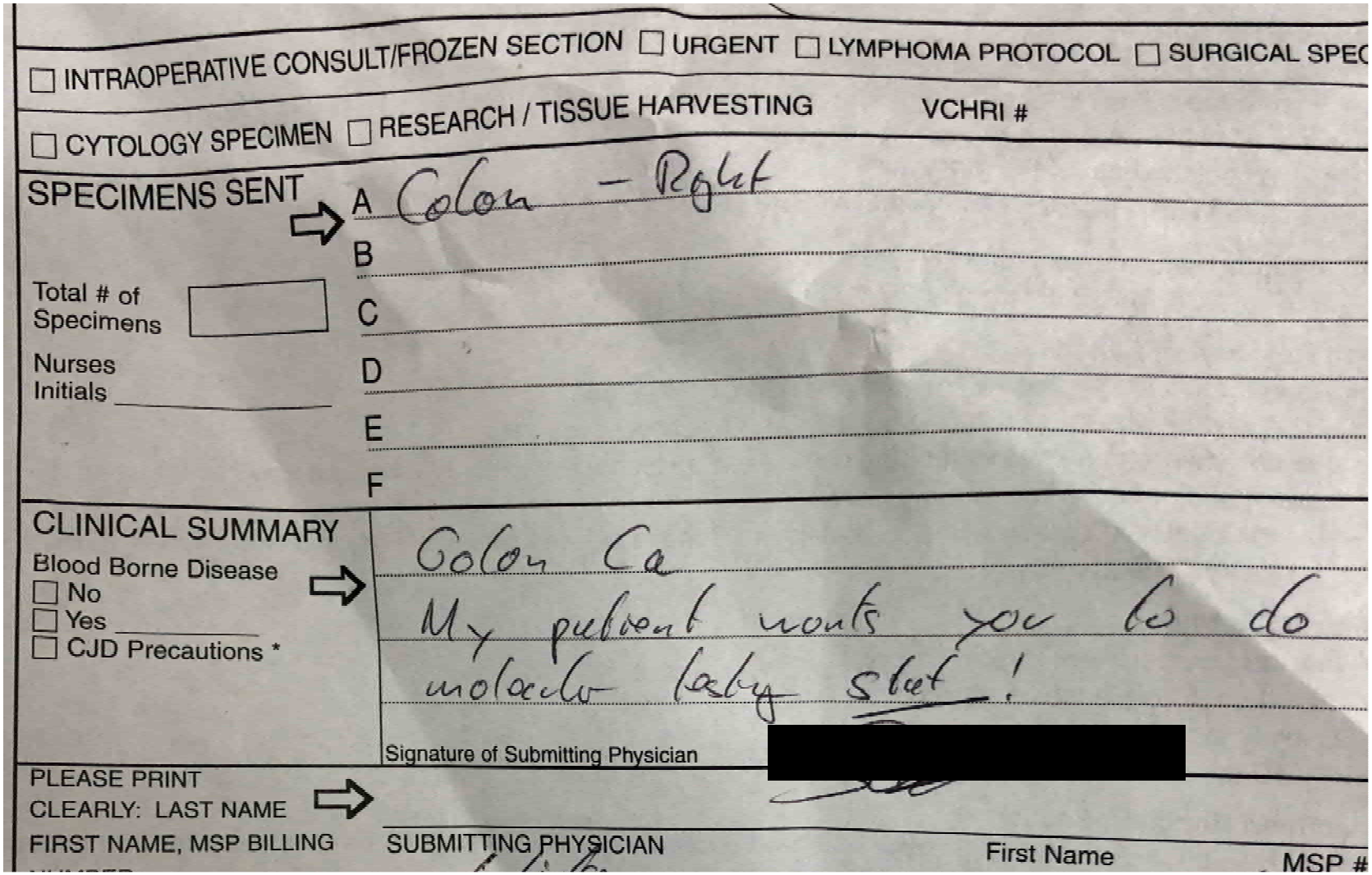
David Schaeffer
Consultant Pathologist and Head, Department of Pathology and Laboratory Medicine, Vancouver General Hospital
Assistant Professor, Department of Pathology and Laboratory Medicine, UBC

Disclosure

No relevant financial relationship with commercial interest to disclose.

Some random facts about Vancouver…. *to start those conversations during the second half of the week*

Personalized Medicine – *we can't escape it*


☐ INTRAOPERATIVE CONSULT/FROZEN SECTION ☐ URGENT ☐ LYMPHOMA PROTOCOL ☐ SURGICAL SPEC

☐ CYTOLOGY SPECIMEN ☐ RESEARCH / TISSUE HARVESTING VCHRI #

SPECIMENS SENT

A Colon - Right

B

C

D

E

F

Total # of Specimens

Nurses Initials

CLINICAL SUMMARY

Blood Borne Disease
☐ No
☐ Yes
☐ CJD Precautions *

Colon Ca

My patient wants you to do molecular baby stuff!

Signature of Submitting Physician [redacted]

PLEASE PRINT CLEARLY: LAST NAME FIRST NAME, MSP BILLING NUMBER

SUBMITTING PHYSICIAN First Name MSP #

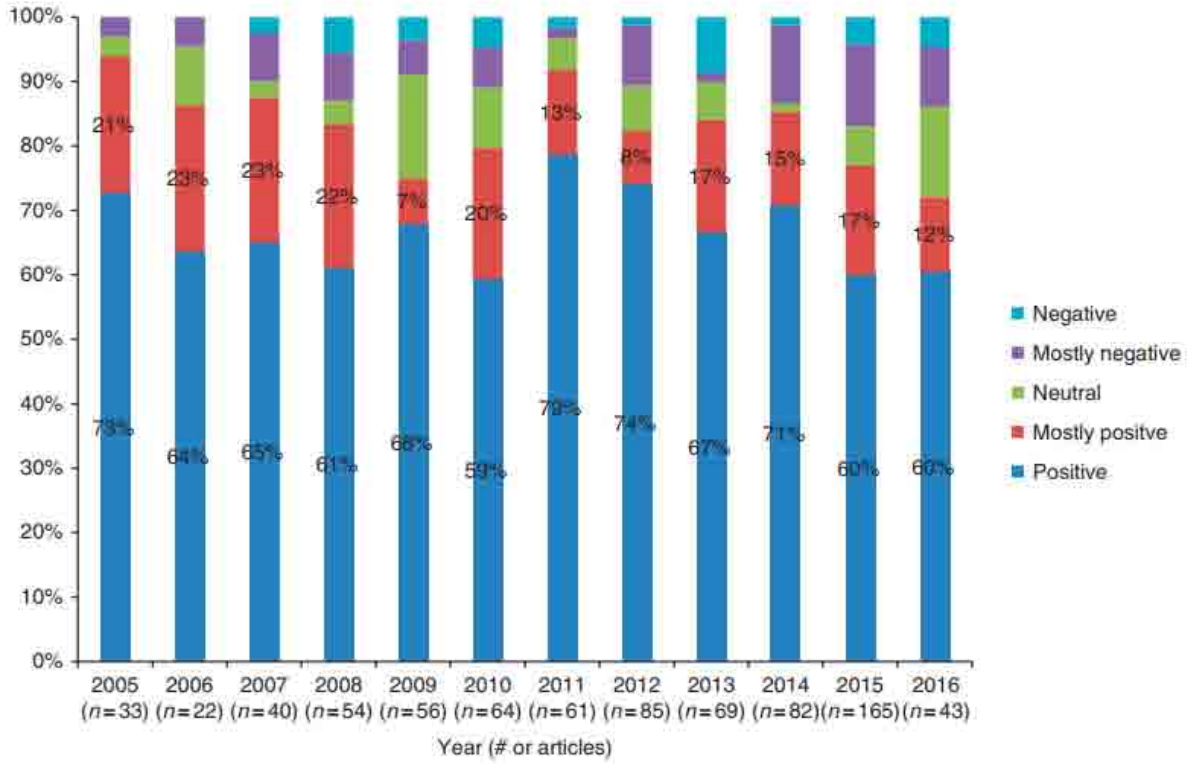
Portrayal of Personalized Medicine in the popular press lacks a balanced and nuanced framing

ORIGINAL RESEARCH ARTICLE | Genetics inMedicine

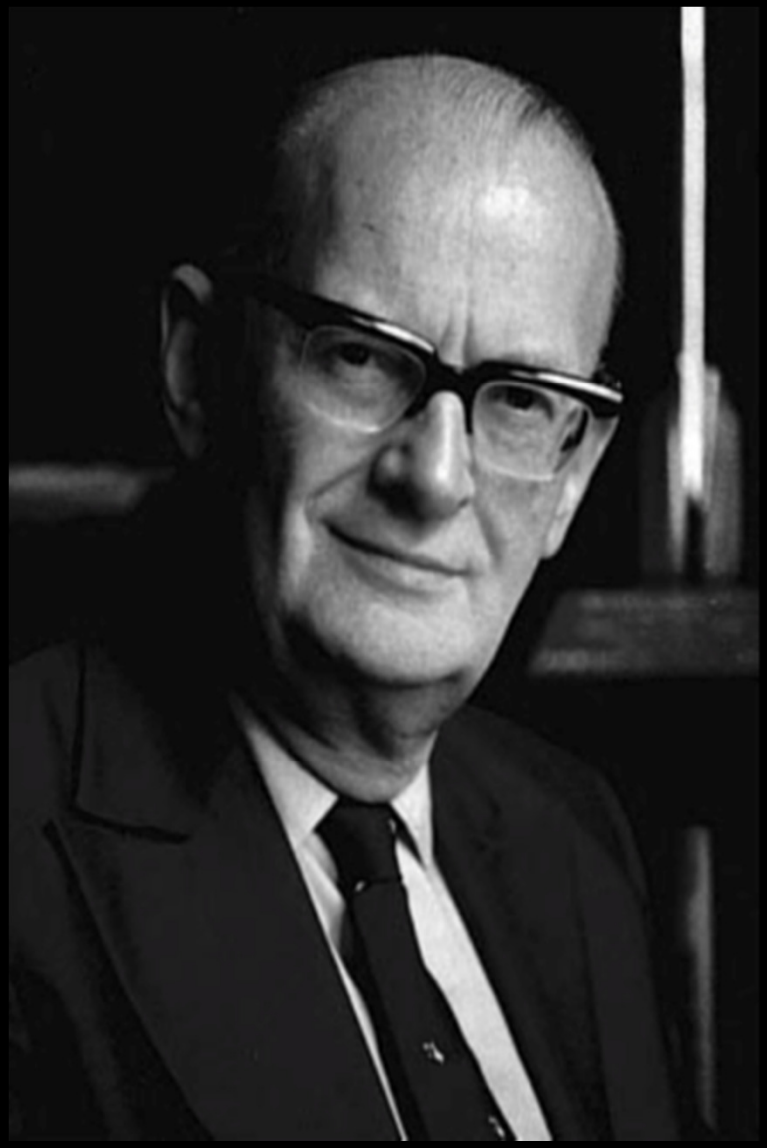
Open

Representing a "revolution": how the popular press has portrayed personalized medicine

Alessandro R Marcon, MA[1], Mark Bieber, BSc[2] and Timothy Caulfield, LLM, FRSC[1,3]



Genetics in Medicine. advance online publication 4 January 2018. doi:10.1038/gim.2017.217



"Any sufficiently advanced technology is indistinguishable from magic."

- Arthur C. Clarke

..and it is just the beginning of the personalized medicine tidal wave

Review Article

Personalized and precision medicine: integrating genomics into treatment decisions in gastrointestinal malignancies

Trang H. Au[1], Kai Wang[2], David Stenehjem[1,3], Ignacio Garrido-Laguna[3,4]

J Gastrointest Oncol 2017;8(3):387-404

While basket studies are gaining momentum, failures remind us that shifting from a biology agnostic (histology-driven) approach to a histology-agnostic approach is unlikely to be a failure-free strategy for a number of tumor types

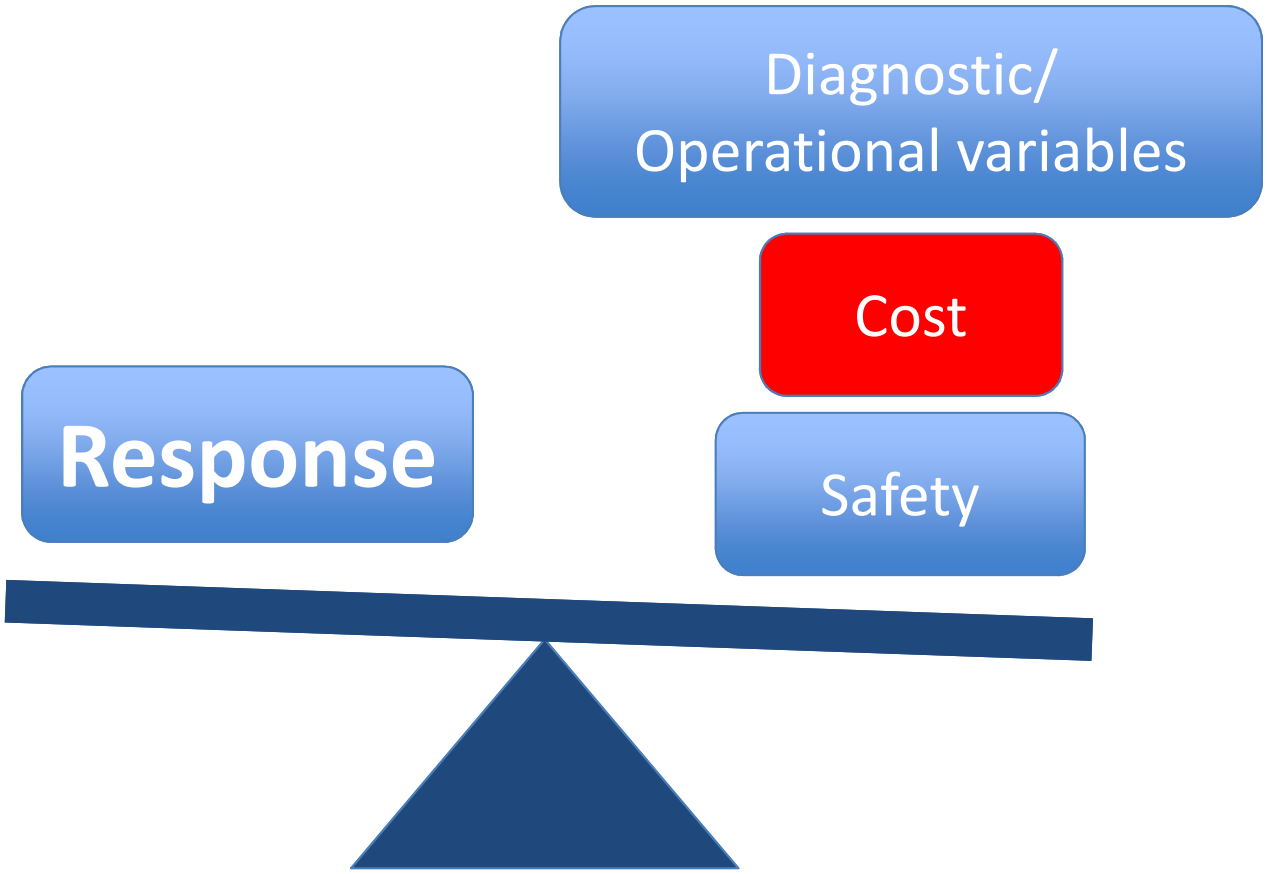
LETTER

doi:10.1038/nature10868

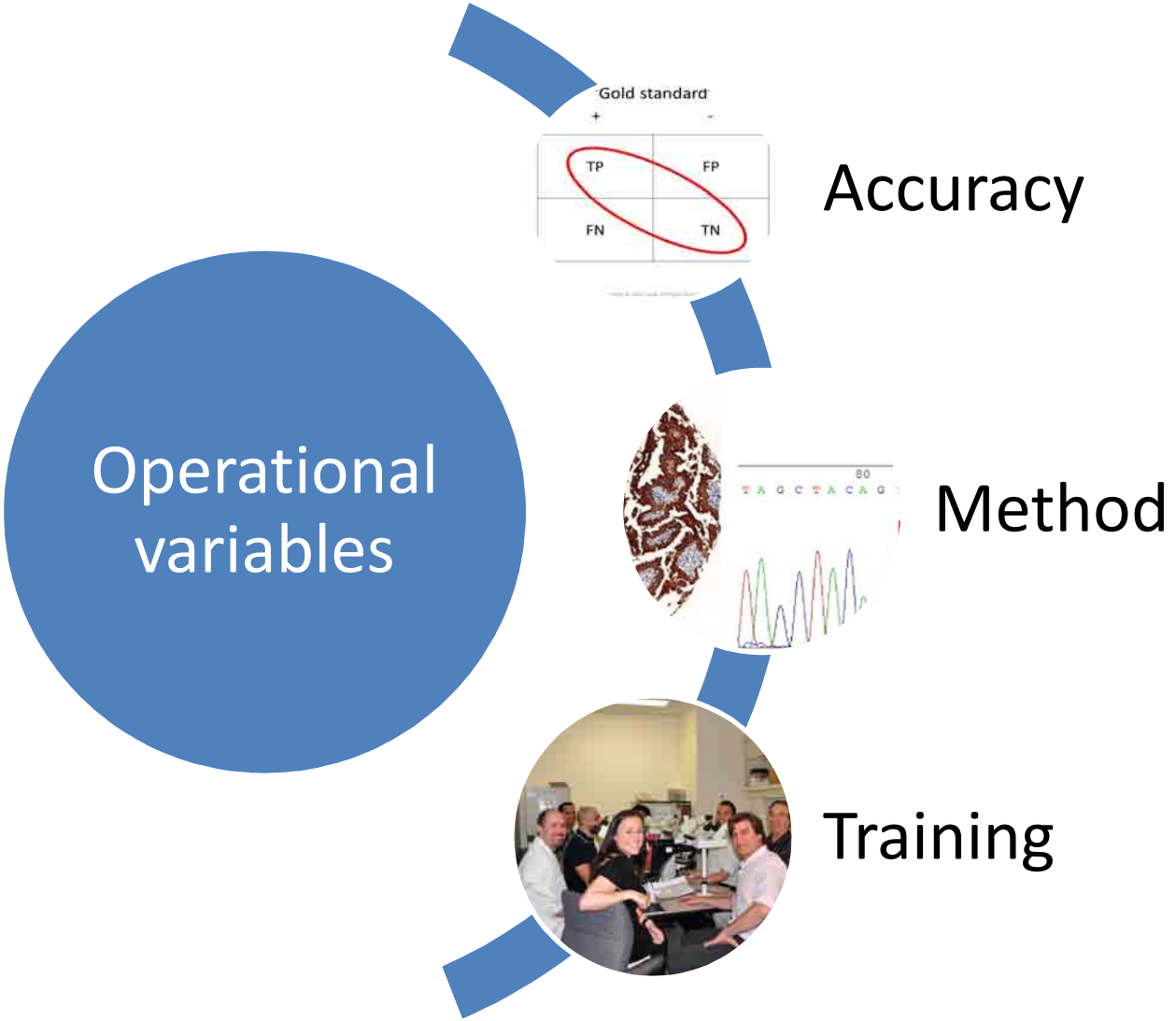
Unresponsiveness of colon cancer to BRAF(V600E) inhibition through feedback activation of EGFR

Anirudh Prahallad[1]*, Chong Sun[1]*, Sidong Huang[1]*, Federica Di Nicolantonio[2,3]*, Ramon Salazar[4], Davide Zecchin[2], Roderick L. Beijersbergen[1], Alberto Bardelli[2,3] & René Bernards[1]

Success of targeted molecular therapy



The tidal wave of molecular specific requests is coming our way – *how do we prepare*



Molecular evaluation of colorectal cancer

Table 4. Guideline Statements and Strength of Recommendations

Guideline Statement	Strength of Recommendation
1. Patients with colorectal carcinoma being considered for anti-EGFR therapy must receive *RAS* mutational testing. Mutational analysis should include *KRAS* and *NRAS* codons 12 and 13 of exon 2, 59 and 61 of exon 3, and 117 and 146 of exon 4 ("expanded" or "extended" *RAS*).	Recommendation
2a. *BRAF* p.V600 (*BRAF* c.1799 [p.V600]) mutational analysis should be performed in colorectal cancer tissue in patients with colorectal carcinoma for prognostic stratification.	Recommendation
2b. *BRAF* p.V600 mutational analysis should be performed in deficient MMR tumors with loss of MLH1 to evaluate for Lynch syndrome risk. Presence of a *BRAF* mutation strongly favors a sporadic pathogenesis. The absence of a *BRAF* mutation does not exclude risk of Lynch syndrome.	Recommendation
3. Clinicians should order mismatch repair status testing in patients with colorectal cancers for the identification of patients at high risk for Lynch syndrome and/or prognostic stratification.	Recommendation
4. There is insufficient evidence to recommend *BRAF* c.1799 p.V600 mutational status as a predictive molecular biomarker for response to anti-EGFR inhibitors.	No recommendation
5. There is insufficient evidence to recommend *PIK3CA* mutational analysis of colorectal carcinoma tissue for therapy selection outside of a clinical trial. *Note:* Retrospective studies have suggested improved survival with postoperative aspirin use in patients whose colorectal carcinoma harbors a *PIK3CA* mutation.	No recommendation
6. There is insufficient evidence to recommend PTEN analysis (expression by immunohistochemistry or deletion by fluorescence in situ hybridization) in colorectal carcinoma tissue for patients who are being considered for therapy selection outside of a clinical trial.	No recommendation
7. Metastatic or recurrent colorectal carcinoma tissues are the preferred specimens for treatment predictive biomarker testing and should be used if such specimens are available and adequate. In their absence, primary tumor tissue is an acceptable alternative and should be used.	Expert consensus opinion
8. Formalin-fixed, paraffin-embedded tissue is an acceptable specimen for molecular biomarker mutational testing in colorectal carcinoma. Use of other specimens (eg, cytology specimens) will require additional adequate validation, as would any changes in tissue-processing protocols.	Expert consensus opinion
9. Laboratories must use validated colorectal carcinoma molecular biomarker testing methods with sufficient performance characteristics for the intended clinical use. Colorectal carcinoma molecular biomarker testing validation should follow accepted standards for clinical molecular diagnostics tests.	Strong recommendation
10. Performance of molecular biomarker testing for colorectal carcinoma must be validated in accordance with best laboratory practices.	Strong recommendation
11. Laboratories must validate the performance of IHC testing for colorectal carcinoma molecular biomarkers (currently IHC testing for *MLH1, MSH2, MSH6,* and *PMS2*) in accordance with best laboratory practices.	Strong recommendation
12. Laboratories must provide clinically appropriate turnaround times and optimal utilization of tissue specimens by using appropriate techniques (eg, multiplexed assays) for clinically relevant molecular and immunohistochemical biomarkers of colorectal cancer.	Expert consensus opinion
13. Molecular and IHC biomarker testing in colorectal carcinoma should be initiated in a timely fashion based on the clinical scenario and in accordance with institutionally accepted practices. *Note:* Test ordering can occur on a case-by-case basis or by policies established by the medical staff.	Expert consensus opinion
14. Laboratories should establish policies to ensure efficient allocation and utilization of tissue for molecular testing, particularly in small specimens.	Expert consensus opinion
15. Members of the patient's medical team, including pathologists, may initiate colorectal carcinoma molecular biomarker test orders in accordance with institutionally accepted practices.	Expert consensus opinion
16. Laboratories that require send-out of tests for treatment predictive biomarkers should process and send colorectal carcinoma specimens to reference molecular laboratories in a timely manner. *Note:* It is suggested that a benchmark of 90% of specimens should be sent out within 3 working days.	Expert consensus opinion
17. Pathologists must evaluate candidate specimens for biomarker testing to ensure specimen adequacy, taking into account tissue quality, quantity, and malignant tumor cell fraction. Specimen adequacy findings should be documented in the patient report.	Expert consensus opinion
18. Laboratories should use colorectal carcinoma molecular biomarker testing methods that are able to detect mutations in specimens with at least 5% mutant allele frequency, taking into account the analytical sensitivity of the assay (limit of detection [LOD]) and tumor enrichment (eg, microdissection). *Note:* It is recommended that the operational minimal neoplastic carcinoma cell content tested should be set at least two times the assay's LOD.	Expert consensus opinion
19. Colorectal carcinoma molecular biomarker results should be made available as promptly as feasible to inform therapeutic decision making, both prognostic and predictive. *Note:* It is suggested that a benchmark of 90% of reports be available within 10 working days from date of receipt in the molecular diagnostics laboratory.	Expert consensus opinion
20. Colorectal carcinoma molecular biomarker testing reports should include a results and interpretation section readily understandable by oncologists and pathologists. Appropriate Human Genome Variation Society and Human Genome Organisation nomenclature must be used in conjunction with any historical genetic designations.	Expert consensus opinion
21. Laboratories must incorporate colorectal carcinoma molecular biomarker testing methods into their overall laboratory quality improvement program, establishing appropriate quality improvement monitors as needed to ensure consistent performance in all steps of the testing and reporting process. In particular, laboratories performing colorectal carcinoma molecular biomarker testing must participate in formal proficiency testing programs, if available, or an alternative proficiency assurance activity.	Strong recommendation

EGFR, epidermal growth factor receptor; IHC, immunohistochemistry; MMR, mismatch repair; PTEN, phosphatase and tensin homolog.

Arch Pathol Lab Med 2017 2017 May;141(5):625-657.

Guideline Statements:

- Members of the patient's clinical team, including pathologists, may initiate molecular testing
- Reports should include a results and interpretation section readily understandable by oncologists and pathologists

Predictive biomarkers in gastrointestinal tract tumours – *focusing on IHC*

Biomarker	Established use[1]	'Off-label' use
Her2 (*ERBB2*)	Stomach Esophagus	Colon Small bowel adenocarcinoma Cholangiocarcinoma PDAC
MMR/PDL-1	Colon Gastric	Small bowel adenocarcinoma Esophageal adenocarcinoma PDAC/Ampullary
BRAFV600E	Colon	Small bowel adenocarcinoma Cholangiocarcinoma Gastric adenocarcinoma
ROS1	-	Colon Small bowel adenocarcinoma Cholangiocarcinoma Gastric adenocarcinoma PDAC

[1] For the purpose of this presentation 'established' was defined as the biomarkers that have a CAP template.

Disclosure – *more important than the first*

I reviewed selected publications only and I apologize in advance for missing key papers…….

If there are any glaring mistakes I am happy to blame my residents Drs. Basile Tessier Cloutier and Daniel Owen who kindly helped with some of the literature search.

Her2 (*ERBB2*)

Her2 (*ERBB2*) amplification in CRC

- Detected in approximately 2-4% of unselected CRC[1,2,3]
- Probably exclusively (or almost exclusively) occurs in MSS CRC[3]
- Appears to occur almost exclusively in RAS-wild type CRC[4,5,6,7,9]
- No correlation with type, localization, grade, p stage or survival[1]
- Correlates with resistance to EGFR-directed therapy in CRC[5,7,8]

1. Marx AH et al. Hum Pathol 41(11): 1577-85.
2. Ooi A et al. Mod Pathol 17(8): 895–904.
3. Cancer Genome Atlas Network. 2012. Nature 487(7407): 330-7.
4. Bertotti A, et al. Cancer Discov 1(6): 508-23.
5. Yonesaka K, et al. Sci Transl Med 3(99): 99ra86.
6. Sartore-Bianchi A, et al. Lancet Oncol 17(6): 738-746.
7. Jeong JH, et al. Clin Colorectal Cancer 16(3): e147-e152.
8. Martin V, et al. Br J Cancer 108(3): 668-75.
9. Valtorta E, et al. Mod Pathol 28(11): 1481-91.

Her2 (*ERBB2*) amplification in CRC – *HERACLES trial*

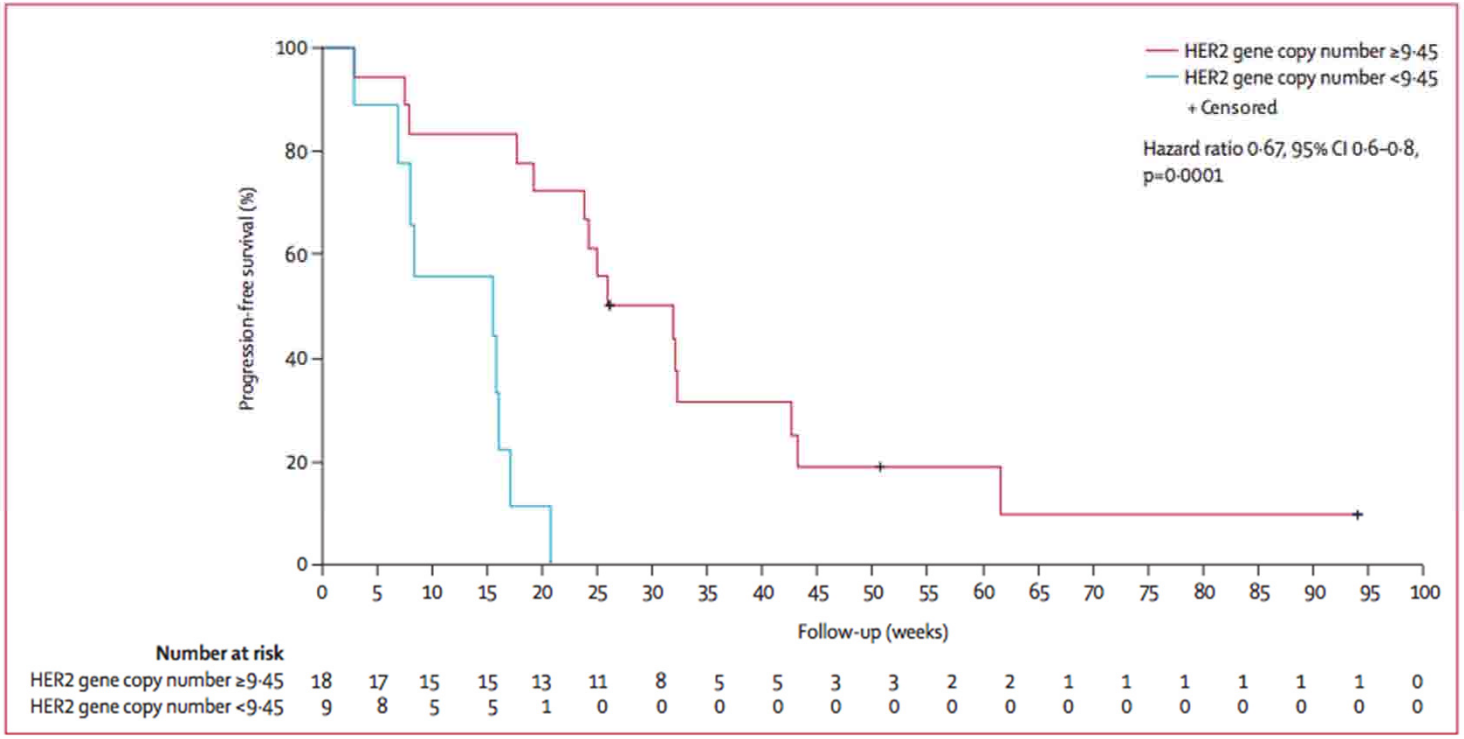


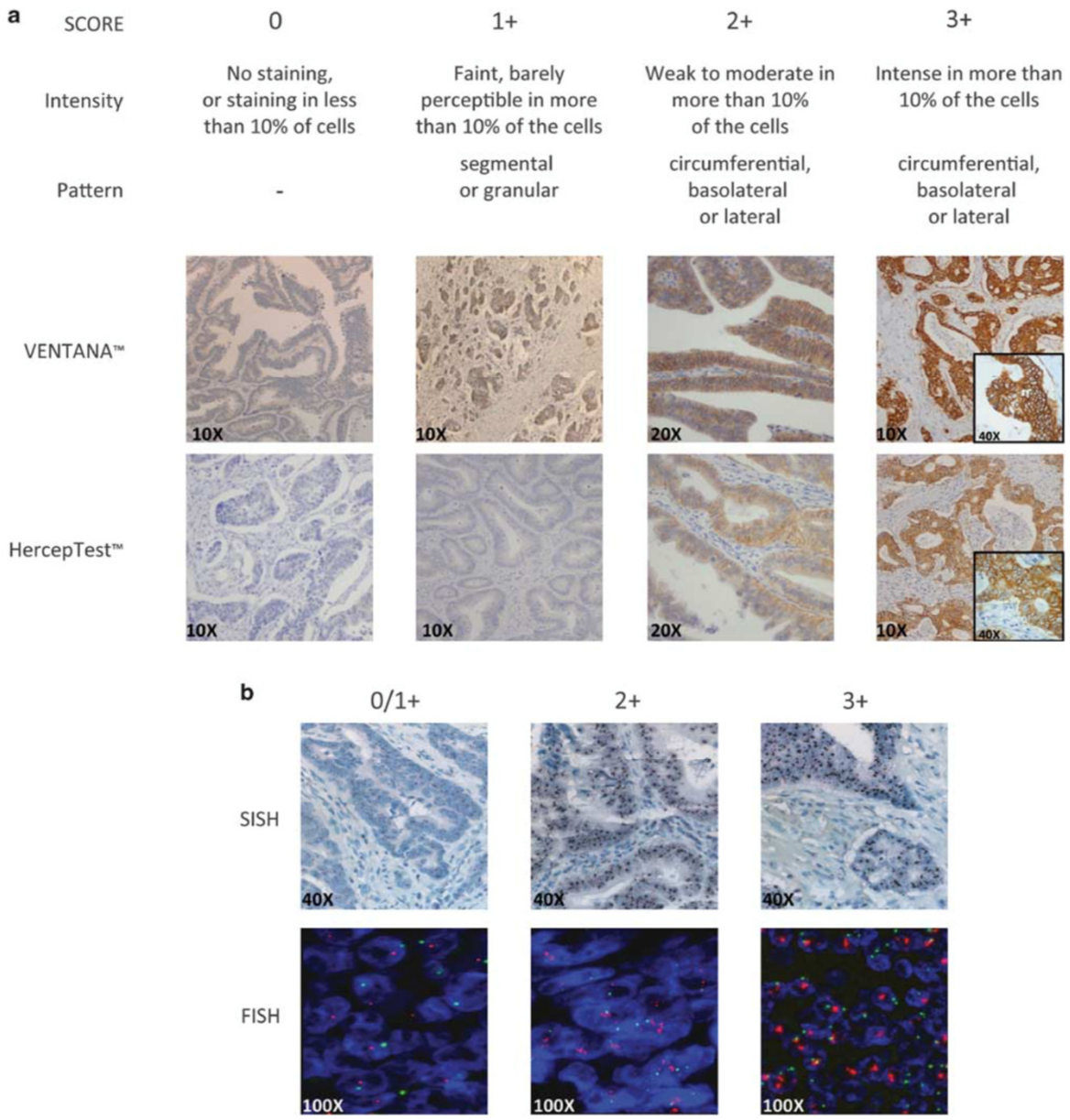
***Figure 2*: Progression-free survival by *HER2* gene copy number variation**
Data from three patients, who remained in follow-up for progression-free survival at the time of data cutoff, were censored.

- Heterogeneity of *ERBB2*-amplification; activating *PIK3CA* mutation; decreased expression of *PTEN*; increased expression of *MUC1* or *MET*

Her2 (*ERBB2*) amplification in CRC - *HERACLES Diagnostic criteria*

Assessn
colorect
validatio

Emanuele Valto
Frédérique Pena
Walter Grigioni[6]
Johannes Noe[8],
Stefania Moscor
Fotios Loupakis
Silvio Veronese
Marcello Gamba



Should one test all CRC for Her2 (*ERBB2*) amplification?

- No definitive answer to this question is published
- *ERBB2* amplification testing will be necessary to select patients most likely to benefit from HER2-directed therapy
- Current CAP guidelines do not recommend *ERBB2* amplification testing for the purpose of selecting patients who may be eligible for EGFR-directed therapy

Personal Opinion: Select cases could presently be tested under specific circumstances.

- MSS, *KRAS* and *BRAF* wild-type advanced CRC
- Well established lines of therapy ineffective (including but not limited to chemotherapy and EGFR-directed therapy)
- Patient has the potential to receive trastuzumab and lapatinib (*e.g.* via special funding or clinical trial)

Her2 (*ERBB2*) amplification in extracolonic sites

Site	Rate
PDAC	2-7%
Small bowel adenocarcinoma	2-3%
Biliary tract adenocarcinoma	1-9%

- No RTC looked at HER2 inhibitors in HER2 amplified tumours outside of colon, stomach, and esophagus
- Only anecdotal report of response in small bowel cancer
- RTC in PDAC, without HER2 amplification status, failed to show significant response.

Oncotarget, Vol. 6, No. 14

Human Epidermal Growth Factor Receptor 2-Positive Duodenal Adenocarcinoma: A Case Report and Review of the Literature

Virginia Moreira Braga[a] Marcos Belotto de Oliveira[b]
Caio Coelho Netto[b] Roberto El Ibrahim[c] Renata D'Alpino Peixoto[a]

with cetuximab and
ıncreatic cancer after
PY"phase 1-2 trial

Rosine Guimbaud[7], Nicole
Tubiana-Mathieu[8], Denis Smith[9], Jean-Pierre Delord[10], Emmanuelle Samalin[2],
Fabienne Portales[2], Christel Larbouret[3,4,5,6], Bruno Robert[3,4,5,6], Frédéric Bibeau[2],
Jean-Pierre Bleuse[2], Evelyne Crapez[2], Marc Ychou[1,2,3,4,5,6,*] and André Pèlegrin[3,4,5,6,*]

Mismatch repair (MMR) deficiency

Mismatch repair (MMR) deficiency in extracolonic sites

Site	Rate
PDAC	6-15%

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

FDA grants accelerated approval to pembrolizumab for first tissue/site agnostic indication

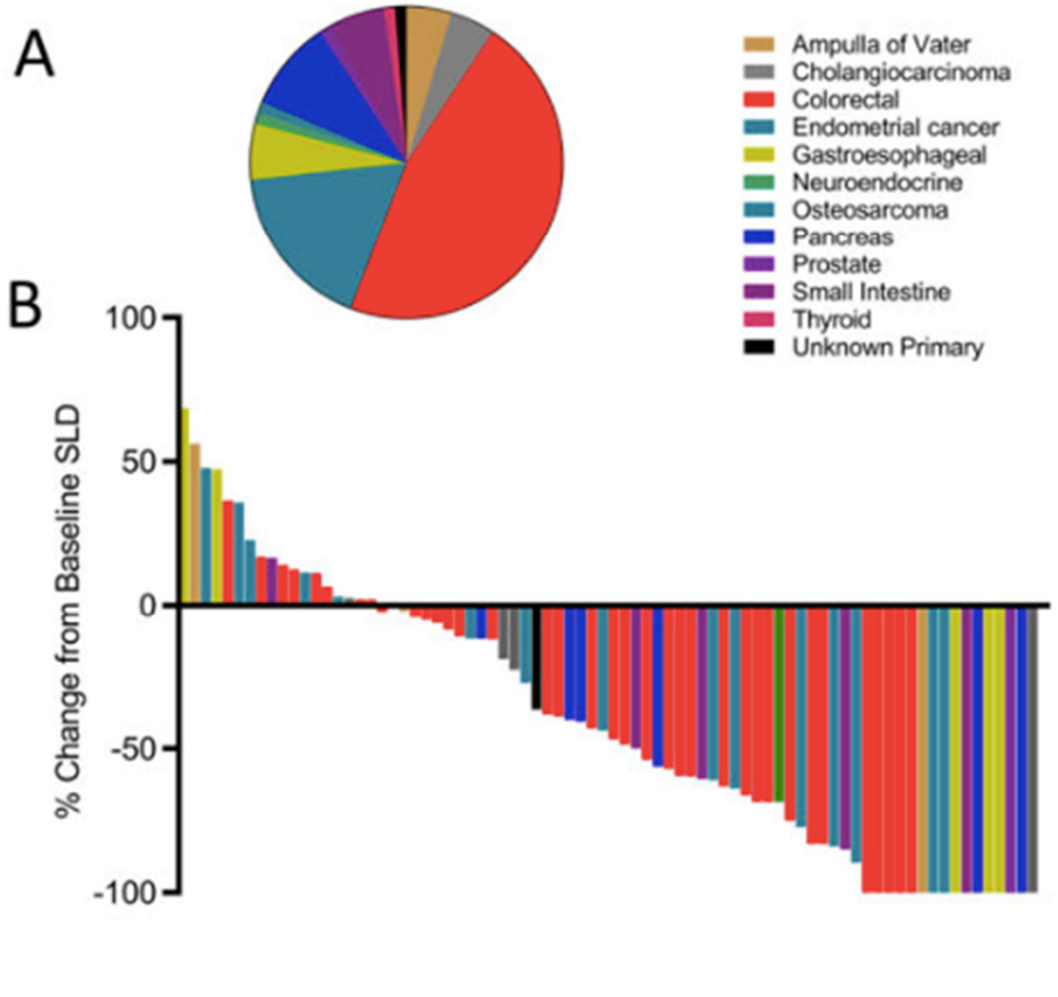
- FDA approved pembrolizumab (PD-1 inhibitor) in unresectable/metastatic MSI-H or dMMR tumors agnostic of site.
- The predictive role of dMMR was never tested prospectively in extracolonic GI cancers

This is the FDA's first tissue/site-agnostic approval.

Dung T. Le[1,2,3], Jennifer N. Durham[1,2,3,*], Kellie N. Smith[1,3,*], Hao Wang[3,*], Bjarne R. Bartlett[2,4,*], Laveet K. Aulakh[2,4], Steve Lu[2,4], Holly Kemberling[3], Cara Wilt[3], Brandon S. Luber[3], Fay Wong[2,4], Nilofer S. Azad[1,3], Agnieszka A. Rucki[1,3], Dan Laheru[3], Ross Donehower[3], Atif Zaheer[5], George A. Fisher[6], Todd S. Crocenzi[7], James J. Lee[8], Tim F.

Mismatch repair (MMR) deficiency in extracolonic sites

Le et al.



Mismatch repair (MMR) deficiency – *Gastric cancer*

uicc
global cancer control

IJC
International Journal of Cancer

Int. J. Cancer: **128**, 1606–1613 (2011) © 2010 UICC

MSI phenotype and MMR alterations in familial and sporadic gastric cancer

Marina Leite[1], Giovanni Corso[1,2,3], Sónia Sousa[1], Fernanda Milanezi[1], Luís P. Afonso[4], Rui Henrique[4], José Manuel Soares[5], Sérgio Castedo[6,7], Fátima Carneiro[1,7], Franco Roviello[2,3], Carla Oliveira[1,7] and Raquel Seruca[1,7]

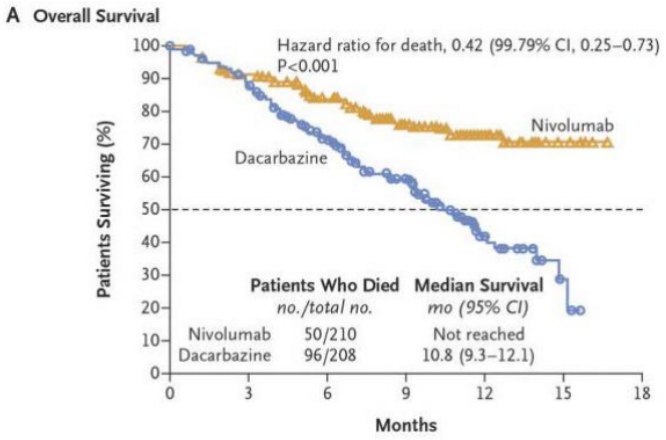
[1] IPATIMUP, Institute of Molecular Pathology and Immunology of the University of Porto, Portugal
[2] Unit of Surgical Oncology, University of Siena, Siena, Italy
[3] Institute of Tumours of Tuscany (ITT), Tuscany, Italy
[4] Pathologic Anatomy Service, Portuguese Oncology Institute-Porto (IPOP), Porto, Portugal
[5] Gastroenterology Service, Santo Antonio General Hospital (HGSA), Porto, Portugal
[6] GDPN, Genética Médica e Diagnóstico Pré-Natal, Porto, Portugal
[7] FMUP, Faculty of Medicine of the University of Porto, Porto, Portugal

We verified that the frequency of MSI was similar in familial and sporadic GC settings, demonstrating that this molecular phenotype is not a hallmark of familial GC in contrast to what is verified in HNPCC.
Moreover, we observed that the frequency of MLH1 hypermethylation is similar in sporadic and familial cases suggesting that in both settings MSI is not associated to MMR genetic alterations but in contrast to epigenetic deregulation.

What about PDL-1 then…….

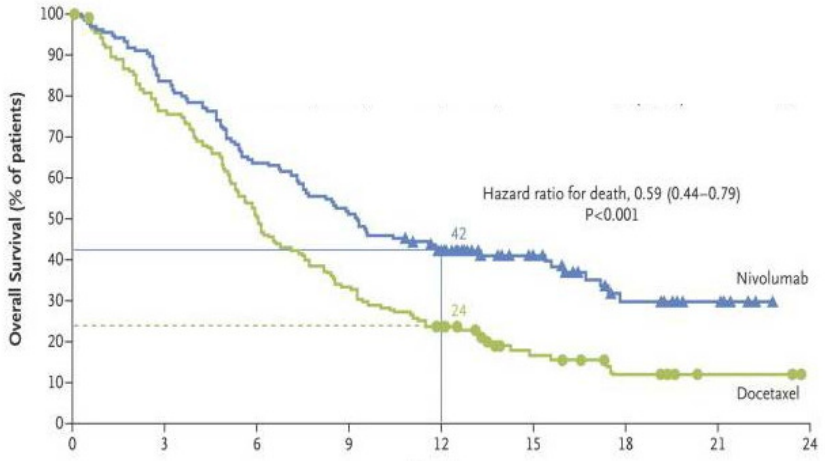
Drug	Target	Vendor
Nivolumab	PD-1	Bristol Myers Squibb
Pembrolizumab	PD-1	Merck
Durvalumab	PD-L1	Astra Zeneca
Atezolizumab	PD-L1	Roche

Melanoma



Garon EB et al. N Engl J Med 2015;372:2018-2028.

Non-Small Cell Lung Cancer



Brahmer J et al. N Engl J Med 2015;373:123-135.

PDL-1 outside the colon?

PD-L1 immunohistochemistry has most recently been advocated to select patients with upper GI tract carcinoma (gastric/esophagus) for anti-PD1 therapy.

Combined Positive Score (CPS)

$$\text{CPS} = \frac{\text{Number of } \mathbf{all} \text{ PD} - \text{L1 staining cells}}{\text{Total number of tumor cells}}$$

Heterogeneity in PD-L1 biomarker tests

Programmed Death L
A New Cha

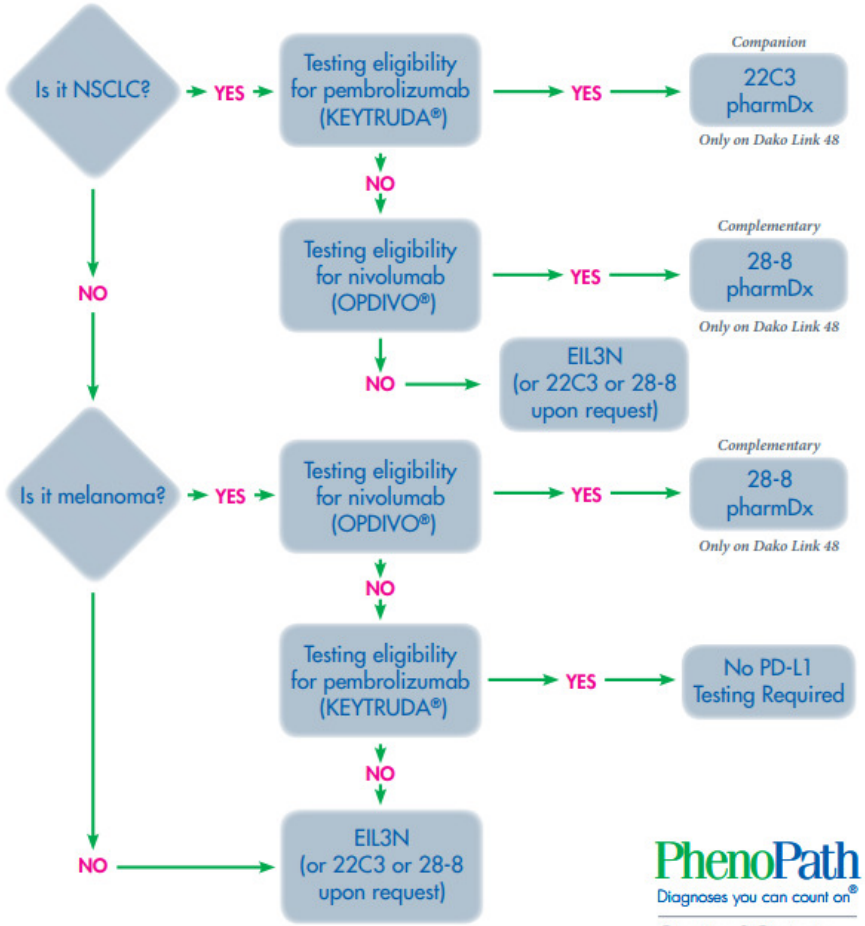
A Perspective From Mem

Lynette M. Sholl, MD; Dara L. Aisner, MD; Tim
Philip T. Cagle, MD; Vera Capelozzi, MD, PhD; S
FRCPath, FRCPE; Sylvie Lantuejoul, MD, PhD; Ma
Sinchita Roy-Chowdhuri, MD, PhD; Eric Thunnissen
of th

Arch Pathol Lab Med—Vol 140, April 2016

Anti-PD-L1 antibodies 22C3, 28-8, and E1L3N

Which PD-L1 test should I order?



PhenoPath
Diagnoses you can count on®

Questions? Contacts:
Allen M. Gown, MD or
Regan Fulton, MD, PhD
206-374-9000

www.phenopath.com

Drug	A	-examined
Nivolumab	2	or cells
Pembrolizumab	2	or cells
Durvalumab	S	or cells or immune cells
Atezolizumab	S	or cells

Does PD-L1 expression even matter in the colon?

a scale from 0 to 100, with higher scores indicating better health status.

Tumour MMR/MSI was assessed per local guidelines (immunohistochemistry or PCR) before screening. MSI was subsequently evaluated on mandatory fresh tumour biopsies collected at enrolment by a central laboratory using PCR (modified Bethesda panel including TGFβR

Discussion

In this open-label, multicentre, phase 2 study, nivolumab showed encouraging activity in patients with dMMR/MSI-H tumours. Responses were recorded across all patient subgroups, including those with (≥1%) and without (<1%) tumour PD-L1 expression, suggesting that PD-L1 is not a predictive biomarker in these patients. Additionally, responses were reported in patients with and without a clinical history of Lynch syndrome, or *KRAS* or *BRAF* mutations. *BRAF*^{V600E} mutations are associated with sporadic dMMR/MSI-H metastatic colorectal cancer and are rarely reported in patients with Lynch syndrome.[20,21] In this study, an investigator-assessed objective response of 25% was recorded in patients with *BRAF*-mutant tumours, which is higher than those historically reported with chemotherapy (<10%)[22,23] or combination treatment including BRAF, EGFR, or MEK inhibitors (10–16%)[24,25] in patients with

	Objective response	Disease control for ≥12 weeks
Tumour PD-L1 expression		
≥1% (n=21)	6 (29%)	11 (52%)
<1% (n=47)	13 (28%)	35 (75%)
Immune cell PD-L1 expression		
Rare (n=24)	5 (21%)	14 (58%)
Intermediate (n=21)	5 (24%)	17 (81%)
Numerous (n=23)	9 (39%)	15 (65%)
Mutation status		
BRAF mutant (n=12)	3 (25%)	9 (75%)
KRAS mutant (n=26)	7 (27%)	16 (62%)
Both *BRAF* and *KRAS* wild type (n=29)	12 (41%)	23 (79%)
Clinical history of Lynch syndrome*		
Yes (n=27)	9 (33%)	19 (70%)
No (n=28)	8 (29%)	21 (75%)

Data are n (%). dMMR/MSI-H=DNA mismatch repair deficient/microsatellite instability–high. *Lynch syndrome designation was based on the clinical records of the patients at sites in countries where this reporting was permitted (excluded Italy).

Table 3: **Investigator-assessed objective response and disease control in patients locally assessed as having dMMR/MSI-H (n=74)**

BRAFV600E

BRAFV600E mutation outside the colon

MODERN PATHOLOGY (2014) 27, 1028–1034

BRAF V600E-specific immunohistochemistry reveals low mutation rates in biliary tract cancer and restriction to intrahepatic cholangiocarcinoma

Benjamin Goeppert[1], Lena Frauenschuh[1], Marcus Renner[1], Stephanie Roessler[1], Albrecht Stenzinger[1], Frederick Klauschen[2], Arne Warth[1], Monika Nadja Vogel[3], Arianeb Mehrabi[4], Mohammadreza Hafezi[4], Katja Boehmer[5], Andreas von Deimling[5,6], Peter Schirmacher[1], Wilko Weichert[1] and David Capper[5,6]

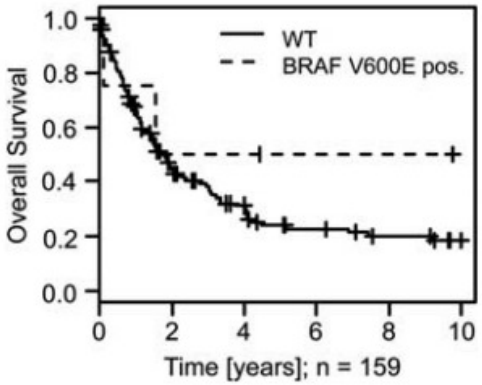


Figure 2 Overall survival probability in intrahepatic cholangiocarcinoma patients in correlation with *BRAF* V600E status. Kaplan–Meier curves show no difference in overall survival of patients in correlation with *BRAF* V600E status in intrahepatic cholangiocarcinoma ($P = 0.38$). The *P*-values were calculated with a log-rank test.

Table 2 Clinicopathological data of biliary tract cancer cohort with complete clinicopathological data and correlation with the *BRAF* V600E status

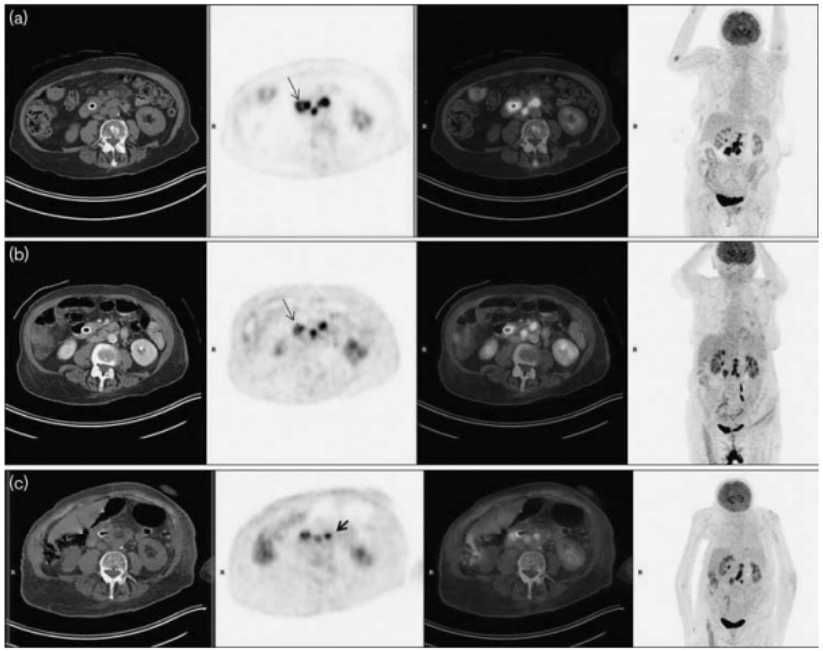
		Number (%)	
	Number (%)	*BRAF* V600E	Fisher's exact test
BTC patients	377 (100 %)	5 (1%)	NS
Age			
64–92 Years	187 (50%)	2 (1%)	NS
31–64 Years	190 (50%)	3 (2%)	NS
Sex			
M	190 (50%)	1 (1%)	0.37
W	187 (50%)	4 (2%)	NS
UICC (N = 296)			
UICC 1	40 (14%)	2 (5%)	NS
UICC 2	75 (25%)	1 (1%)	NS
UICC 3	82 (28%)	0 (0%)	NS
UICC 4	99 (33%)	2 (2%)	NS
pT			
T1	80 (21%)	1 (1%)	NS
T2	148 (39%)	3 (2%)	NS
T3	117 (31%)	1 (1%)	NS
T4	32 (9%)	0 (0%)	NS
pN (N = 286)			
N0	129 (45%)	2 (2%)	NS
N1	157 (55%)	2 (1%)	NS
M			
M0	354 (94%)	5 (1%)	NS
M1	23 (6%)	0 (0%)	NS
G			
G1	20 (5%)	0 (0%)	NS
G2	255 (68%)	4 (2%)	NS
G3	102 (27%)	1 (1%)	NS
L			
L0	174 (46%)	2 (1%)	NS
L1	203 (54%)	3 (2%)	NS
V			
V0	275 (73%)	3 (1%)	NS
V1	102 (27%)	2 (2%)	NS
Pn			
Pn0	294 (78%)	5 (2%)	NS
Pn1	83 (22%)	0 (0%)	NS
Biliary tract cancer subgroups			
Intrahepatic cholangiocarcinoma	159 (42%)	5 (3%)	0.01
Extrahepatic cholangiocarcinoma	149 (40%)	0 (0%)	NS
Adenocarcinomas of the gallbladder	69 (18%)	0 (0%)	NS
Histology			
Ductal	308 (82%)	4 (1%)	NS
Papillary	25 (7%)	1 (4%)	NS
Mucinous	10 (3%)	0 (0%)	NS
Intestinal	10 (3%)	0 (0%)	NS
Other	24 (6%)	0 (0%)	NS

BRAFV600E mutation outside the colon – ampullary carcinoma

MEK inhibitor treatment is effective in a patient with metastatic carcinoma of the ampulla of Vater with BRAF and NRAS mutations shown by next-generation sequencing

Esther Tahover[a,b], Rachel Bar Shalom[a], Naama Bogot[a], David Kelsen[a,c] and Alberto Gabizon[a,b]

Anti-Cancer Drugs 2016, Vol 27 No 6



(a) Selected images of ^{18}F-FDG PET/CT at staging show pathological uptake in the primary periampullar mass (arrow, SUV_{max} 16) and in enlarged right retroperitoneal lymph nodes (SUV_{max} 15). (b) Selected images of the second ^{18}F-FDG PET/CT during therapy show interval reduction in size and ^{18}F-FDG uptake intensity in the primary periampullar mass (arrow, SUV_{max} 10) and in the right retroperitoneal lymph nodes (SUV_{max} 14). (c) Selected images of the third ^{18}F-FDG PET/CT during therapy show further interval reduction in ^{18}F-FDG uptake intensity in the periampullar mass (SUV_{max} 8) and in one of the right retroperitoneal lymph nodes (short arrow, SUV_{max} 8). The ^{18}F-FDG uptake in the other adjacent lymph node (only partially presented in this slice) was unchanged (SUV_{max} 14). CT, computed tomography; ^{18}F-FDG, fluorine-18-fluorodeoxyglucose; SUV_{max}, maximum standardized uptake value.

ROS1 rearrangement

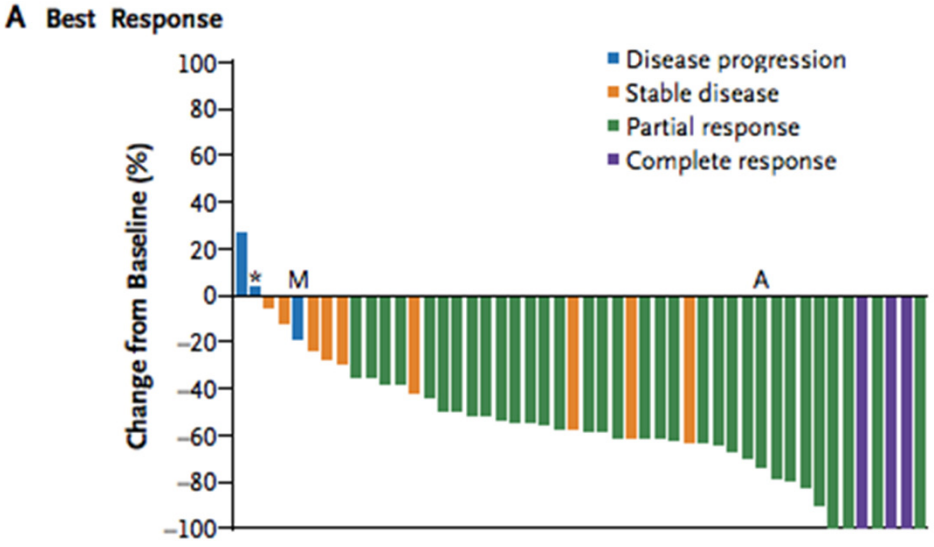
ROS1 rearrangement in gastrointestinal cancers

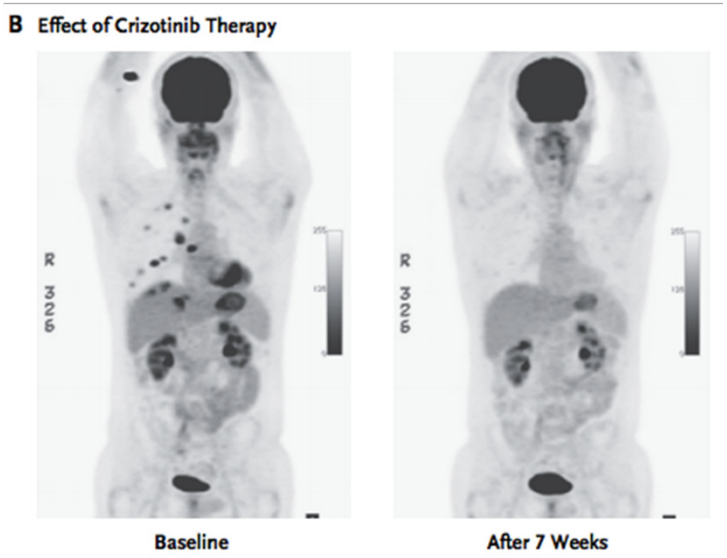
The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 NOVEMBER 20, 2014 VOL. 371 NO. 21

Crizotinib in *ROS1*-Rearranged Non–Small-Cell Lung Cancer

Alice T. Shaw, M.D., Ph.D., Sai-Hong I. Ou, M.D., Ph.D., Yung-Jue Bang, M.D., Ph.D., D. Ross Camidge, M.D., Ph.D., Benjamin J. Solomon, M.B., B.S., Ph.D., Ravi Salgia, M.D., Ph.D., Gregory J. Riely, M.D., Ph.D., Marileila Varella-Garcia, Ph.D., Geoffrey I. Shapiro, M.D., Ph.D., Daniel B. Costa, M.D., Ph.D., Robert C. Doebele, M.D., Ph.D., Long Phi Le, M.D., Ph.D., Zongli Zheng, Ph.D., Weiwei Tan, Ph.D., Patricia Stephenson, Sc.D., S. Martin Shreeve, M.D., Ph.D., Lesley M. Tye, Ph.D., James G. Christensen, Ph.D., Keith D. Wilner, Ph.D., Jeffrey W. Clark, M.D., and A. John Iafrate, M.D., Ph.D.





ROS1 rearrangement in gastrointestinal cancers

Site	Rate
PDAC	0.2%
Colorectal adenocarcinoma	0-0.8%
Biliary tract adenocarcinoma	0-3.7%
Gastric cancer	0-2.6%
Esophageal adenocarcinoma	0-2%

- Contrary to lung adenocarcinoma the ROS1 overexpression is not specific for the rearrangement in GC (3%) and cholangiocarcinoma (0%).
- No evidence of a predictive role to ROS1 in GIT cancers.

Lee *et al. BMC Cancer* (2015) 15:721
DOI 10.1186/s12885-015-1737-4

RESEARCH ARTICLE Open Access

Clinical and pathological significance of ROS1 expression in intrahepatic cholangiocarcinoma

Kyung-Hun Lee[1,2], Kyoung-Bun Lee[3*], Tae-Yong Kim[1,2], Sae-Won Han[1,2], Do-Youn Oh[1,2], Seock-Ah Im[1,2], Tae-You Kim[1,2], Nam-Joon Yi[4], Kwang-Woong Lee[4], Kyung-Suk Suh[4], Ja-June Jang[3] and Yung-Jue Bang[1,2]

ROS1 rearrangement in gastrointestinal cancers

- ROS1 is extremely rare in GI malignancies, except in some biliary malignancies (0-3%).
- Lack of evidence supporting ROS1 as a predictive biomarker of response to crizotinib in GI malignancies.
- Currently there is no role for ROS1 IHC to predict response to crizotinib in GIT.

How to deal with molecular pathology results in daily surgical pathology practice…

FOUNDATIONONE

Patient Name	Report Date	Tumor Type
Martina Adams	12 May 2014	Lung adenocarcinoma

Date of Birth	13 January 1943	Medical Facility	Not Given	Specimen Received	17 May 2014
Sex	Female	Ordering Physician	Dr. Patel	Specimen Site	Lung
FMI Case #	13000000	Additional Recipient	Not Given	Date of Collection	22 November 2013
Medical Record #	5	Medical Facility ID #	-1	Specimen Type	Block
Specimen ID	Not Given	Pathologist	Not Provided		

ABOUT THE TEST:
FoundationOne™ is a next-generation sequencing (NGS) based assay that identifies genomic alterations within hundreds of cancer-related genes.

PATIENT RESULTS

8 genomic alterations
5 therapies associated with potential clinical benefit
0 therapies associated with lack of response
15 clinical trials

TUMOR TYPE: LUNG ADENOCARCINOMA

Genomic Alterations Identified†
MET amplification
AKT2 amplification – equivocal
KRAS amplification
TP53 I162fs*8
CDKN2A/B loss
MYCN amplification – equivocal
MYST3 amplification
ARID2 loss

Additional Disease-relevant Genes with No Reportable Alterations Detected
ALK
EGFR

†For a complete list of the genes assayed, please refer to the Appendix
See Appendix for details

THERAPEUTIC IMPLICATIONS

Genomic Alterations Detected	FDA Approved Therapies (in patient's tumor type)	FDA Approved Therapies (in another tumor type)	Potential Clinical Trials
MET amplification	Crizotinib	Cabozantinib	Yes, see clinical trials section
AKT2 amplification - equivocal	None	Everolimus Temsirolimus	Yes, see clinical trials section
KRAS amplification	None	Trametinib	Yes, see clinical trials section
TP53 I162fs*8	None	None	Yes, see clinical trials section
CDKN2A/B loss	None	None	Yes, see clinical trials section
MYCN amplification - equivocal	None	None	Yes, see clinical trials section
MYST3 amplification	None	None	None

Electronically Signed by Jeffrey S. Ross M.D., Medical Director | CLIA Number: 22D2027531 | 12 May 2014
Foundation Medicine, Inc., 150 3rd Street, Cambridge, MA 02141 | 1.888.988.3639

page 1 of 21

genomic health | oncotype DX® Breast Cancer Assay

Page 1 of 3

Genomic Health, Inc.
301 Penobscot Drive
Redwood City, CA 94063
Tel (866) ONCOTYPE (866-662-6897)
www.oncotypeDX.com

PATIENT REPORT

Patient: Doe, Jane
Sex: Female
DOB: 01/01/1950
Medical Record/Patient #: 556677771
Date of Surgery: 1/25/2008
Specimen ID/Block ID: SURG-0001

Requisition: R00003G
Order Received: 2/01/2008
Date Reported: 2/13/2008
Client: Community Medical Center
Treating Physician: Dr. Harry D Smith
Submitting Pathologist: Dr. John P Williams
Additional Recipient: Dr. Sally M Jones

ASSAY DESCRIPTION

Oncotype DX® Breast Cancer Assay uses RT-PCR to determine the expression of a panel of 21 genes in tumor tissue. The Recurrence Score™ is calculated from the gene expression results. The Recurrence Score range is from 0-100.

RESULTS

Recurrence Score = 5

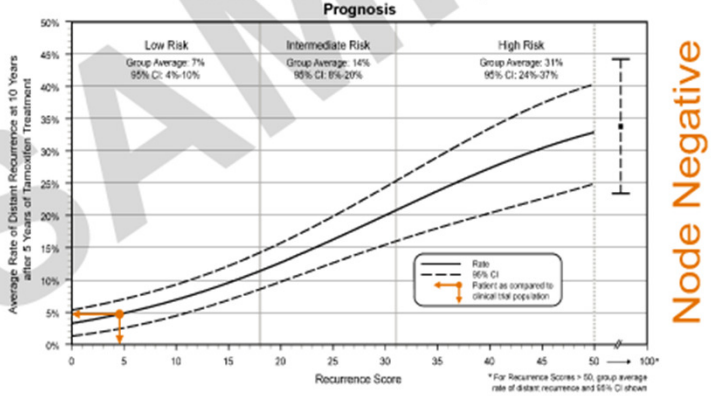
Test Results should be interpreted using the Clinical Experience information contained in this report which is derived from clinical studies involving patient populations with specific clinical features as noted in each section of the Clinical Experience. It is unknown whether the findings summarized in the Clinical Experience are applicable to patients with features different from those described.

CLINICAL EXPERIENCE: PROGNOSIS FOR NODE NEGATIVE, ER-POSITIVE PATIENTS

The Clinical Validation study included female patients with Stage I or II, Node Negative, ER-Positive breast cancer treated with 5 years of tamoxifen. Those patients who had a Recurrence Score of 5 had an Average Rate of Distant Recurrence of 5% (95% CI: 2%-7%)

The following results are from a clinical validation study of 668 patients from the NSABP B-14 study. *N Engl J Med* 2004; 351: 2817-26.

Recurrence Score vs Distant Recurrence in NODE NEGATIVE, ER-Positive Breast Cancer Prognosis



Laboratory Director: Patrick Joseph, MD

CLIA Number 05D1018272

This test was developed and its performance characteristics determined by Genomic Health, Inc. The laboratory is regulated under the Clinical Laboratory Improvement Amendments of 1988 (CLIA) as qualified to perform high-complexity clinical testing. This test is used for clinical purposes. It should not be regarded as investigational or for research. These results are adjunctive to the ordering physician's workup.

301 Penobscot Drive Redwood City, CA 94063 (866) ONCOTYPE (866-662-6897) www.oncotypeDX.com

GH-004 Rev014 6/1/2008

How to deal with molecular pathology results in daily surgical pathology practice…

Offer evidence based recommendation on the predictive role of biomarkers.

Be mindful of the limitations of IHC.

Not every NGS/panel finding is easily translatable in protein expression.

While not wanting to be a barrier, sometimes a cautionary approach to requests by oncologist is prudent to ensure our patients are treated accurately.

Summary

MMR: Should we do reflex MMR IHC on all GIT cancer ?

- Early evidence for that, but in patients who failed previous treatment options, MMR testing should not be discouraged.

PDL-1: I'll let you decide…..

ROS1: The lack of ROS1 prospective data and its rare occurrence supports no predictive role for ROS1 IHC in GIT cancers.

BRAFV600E: When/if previous lines of treatment fail, there is a potential predictive role of BRAFV600E IHC in ampullary carcinoma /cholangiocarcinoma based on anecdotal BRAF inhibitor response.

Her2 *(ERBB2)*: Reasonable to look for Her2 (*ERBB2)* amplification using IHC or FISH in advanced MSS, *KRAS*/*BRAF* wild type tumors that progress on well established therapies, including EGFR-directed therapy.

Thank you!