MAYO CLINIC

MOLECULAR AND IMMUNOTHERAPY UPDATES IN CRC

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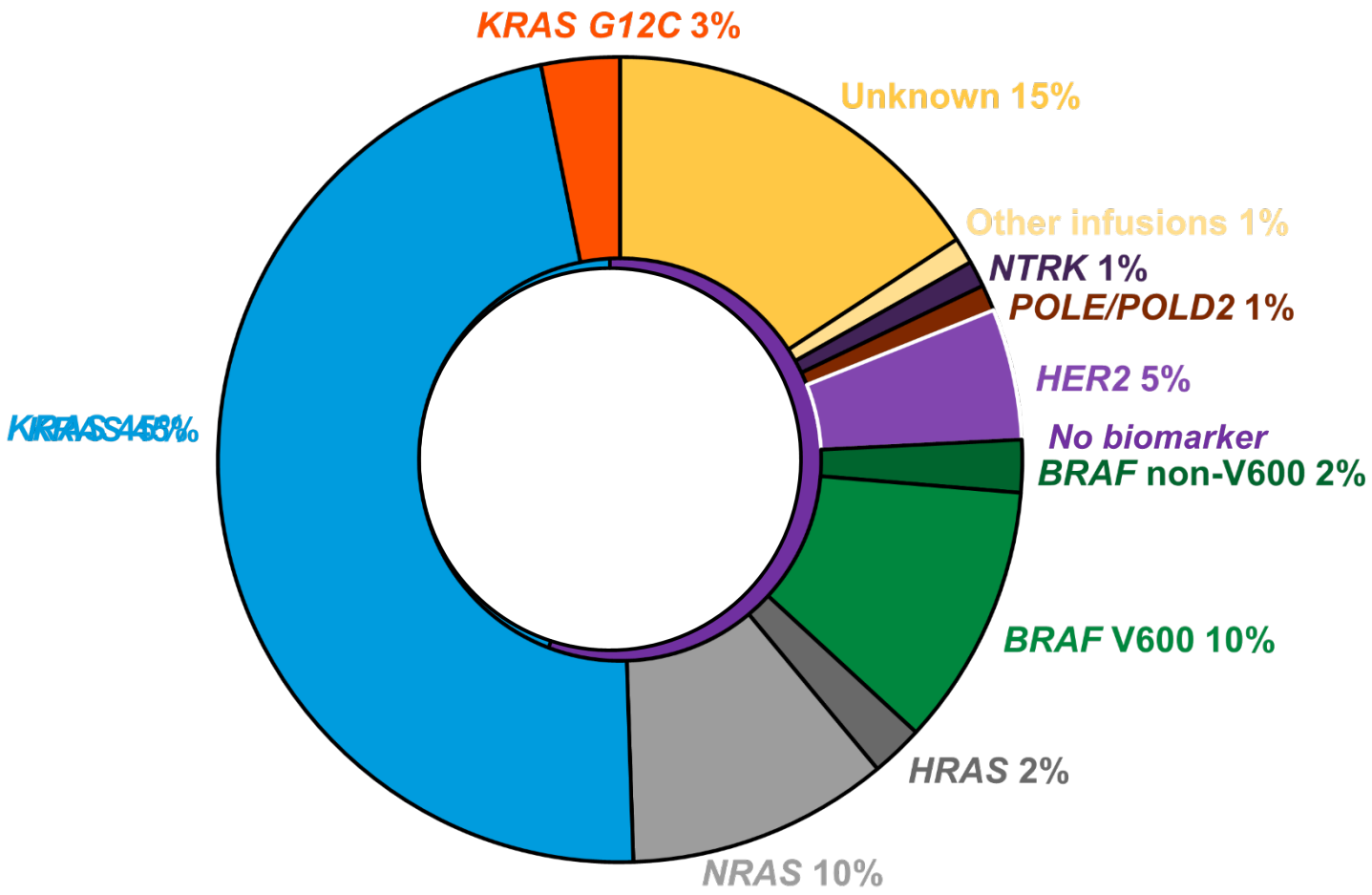
Associate Professor

OUTLINE

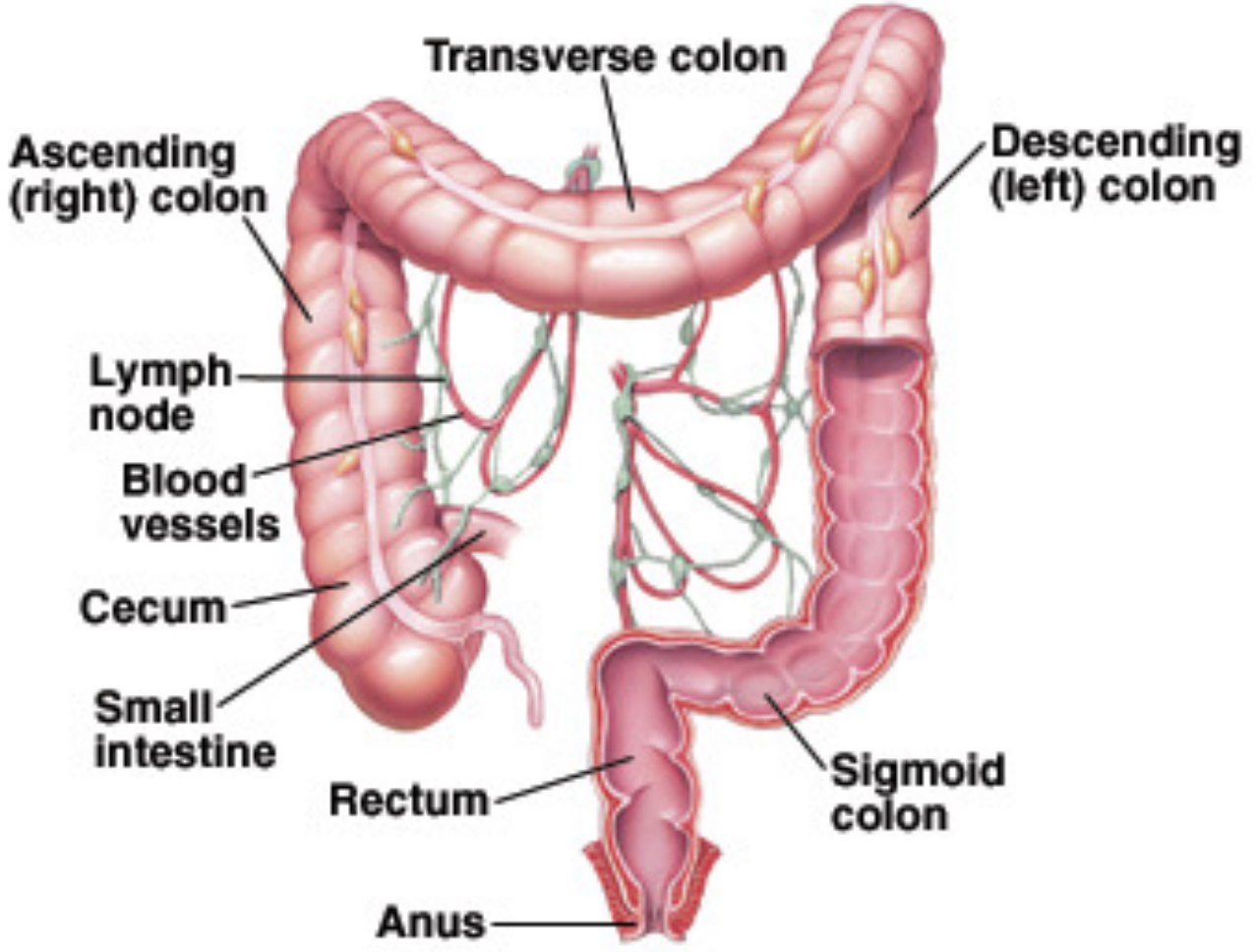
- Updates in target directed therapy in CRC
  - HER-2
  - KRAS G12C
  - KRAS G12D, novel RAS inhibitors
- Updates in IO in MSI-H CRC
- Updates in IO in MSS CRC

ACTIONABLE ALTERATIONS IN MCRC



HER2 IN METASTATIC CRC

- Usually left sided
- Homogeneous HER2 expression
- Primary resistance to EGFR monoclonal antibodies (cetuximab, panitumumab)
- Not mutually exclusive with *RAS* or *BRAF* mutations
- Not associated with worse prognosis



HER2 (*ERBB2*) AMPLIFICATION IS MORE COMMON IN *RAS/RAF* WT CRC PATIENTS

Dataset	Patient population (n)	*ERBB2* amplified
HERACLES	914 KRAS exon 2 WT metastatic CRC patients Source: Sartore-Bianchi, Andrea et al., Lancet Oncol, 17(6) 738 - 746	5.3%
MDACC	114 NRAS/KRAS WT CRC patients 97 KRAS/NRAS/BRAF WT CRC patients Source: J Clin Oncol 34, 2016 (suppl; abstr 3517)	12.2% 14.4%
NCT02008383	76 RAS WT CRC patients at Duke Univ (Guardant360) – unpublished series	11.8%

RECENT DATA OF HER2-TARGETED THERAPIES IN ADVANCED CRC

Regimen	Trial	ORR	PFS	OS	Most common G3 AE's
Trastuzumab + lapatinib	HERACLES-A (N=32)	28%	4.7m	10m	Fatigue- 16%, Decreased LVEF 6%
Trastuzumab + Pertuzumab	MyPathway (N=57)	32%	2.9m	11.5m	Hypokalemia 5%, Abdominal pain 5%
Pertuzumab + T-DM1	HERACLES-B (N=31)	9.7%	4.1m	Not reported	Thrombocytopenia 7%
Trastuzumab + tucatinib	MOUNTAINEER (N=117)	38.1%	8.2m	24.1m	Hypertension 7%, Diarrhea 3.5%
T-DXd	DESTINY-CRC02 (N=82)	37.8%	5.8m	13.4m	Neutropenia 17%, fatigue and anemia (10%)

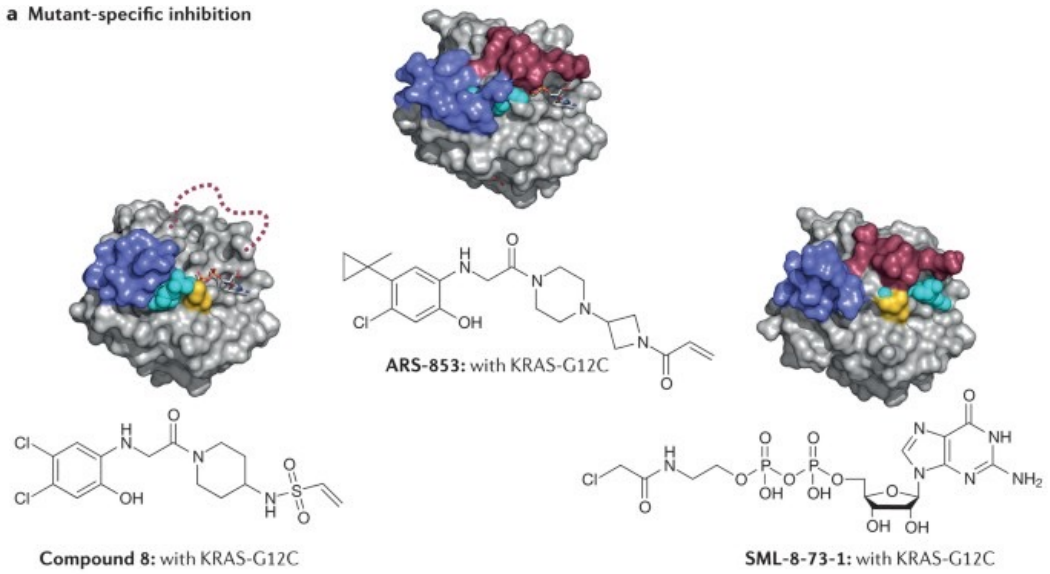
WITH VARIOUS AVAILABLE THERAPIES FOR HER2+ MCRC, HOW DO YOU SEQUENCE TREATMENT?

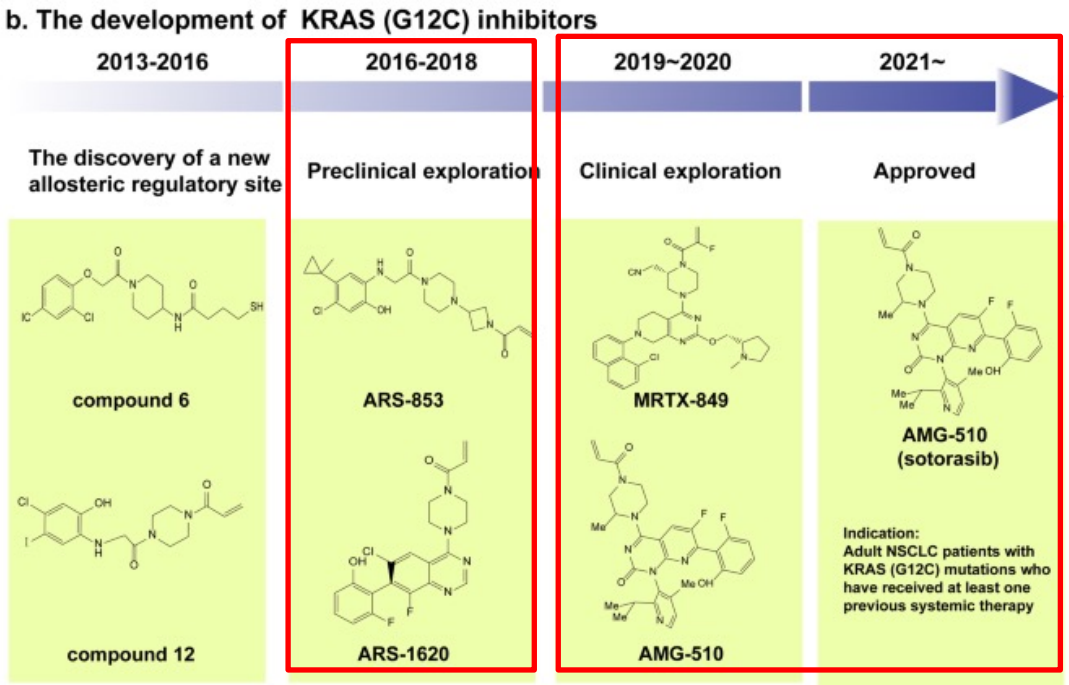
- **Toxicities, prior therapies**
- **RAS mutation status?** 28.6% response observed with T-DxD
- **Prior anti-HER-2 therapy?** 41.2% with T-DxD
- **Optimal sequence of HER-2 directed therapies?**

1. **Tucatinib/trastuzumab** (*only* FDA approved regimen)
2. Pertuzumab/trastuzumab
3. T-DxD (RAS MT/WT and IHC 3+, toxicities may limit utility)

- **Moving forward (MOUNTANEER-3) will establish the role in earlier line setting**

Undruggable to druggable: KRAS G12C





After nearly 40 years of effort, direct targeting of KRAS was difficult and focused on downstream/parallel pathways of RAS signaling…

However, in work published by Ostrem et al (2013):

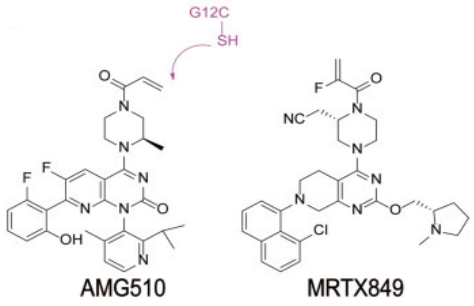
- KRAS G12C harbors a non-native cysteine residue (cysteine = reactive amino acid for forming covalent bonds with drugs), but wild-type RAS does NOT
- Developed irreversible inhibitors by forming a covalent bond to cysteine-12 positioned adjacent to the allosteric switch II region
- Dependent on KRAS G12C actively cycling between GTP and GDP states where switch II pocket restricted only to the GDP-bound state

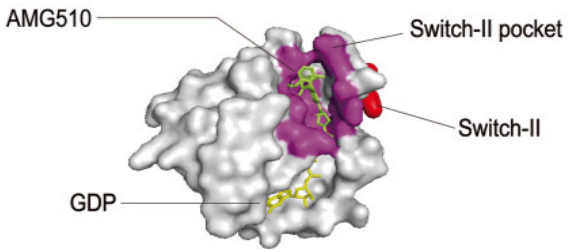
Subsequent switch II pocket covalent KRAS G12C inhibitors →

- Improving potency
- Improved chemical and metabolic stability
- Rate of covalent engagement to G12C must be sufficient to access the GDP-bound state in rapid cycle within a tumor
- Balance optimal potency and selectivity with a favorable PK profile

Huang L et al. *Signal Transduct Target Ther*. 2021;6(1):386;
Ostrem JM et al. *Nature*. 2013;503(7477):548-551;
Ostrem JM et al. *Nat Rev Drug Discov*. 2016;15(11):771-778

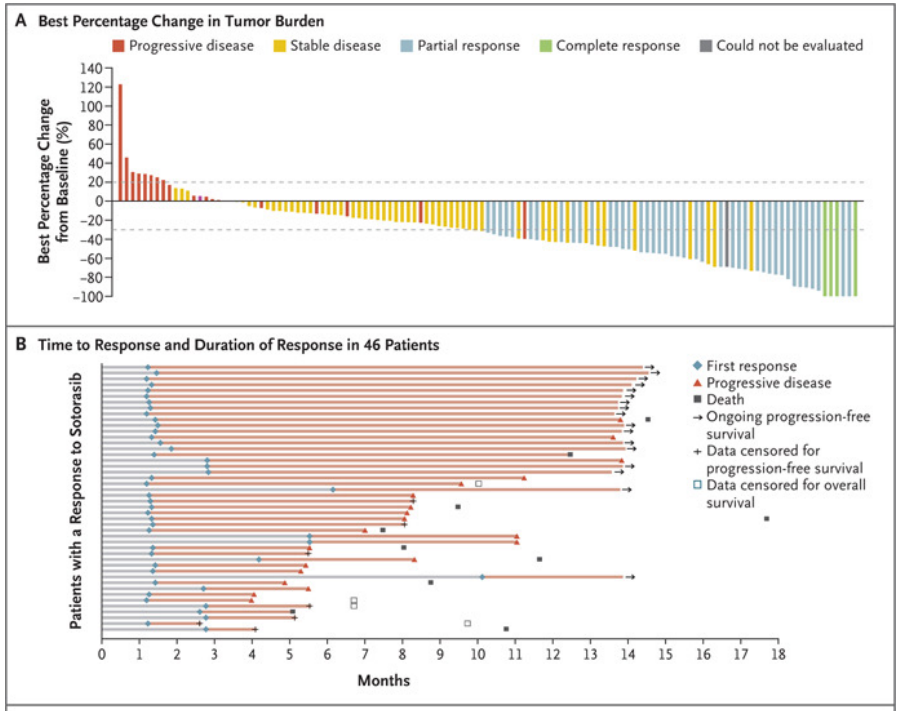
First FDA-approved KRAS G12C inhibitors





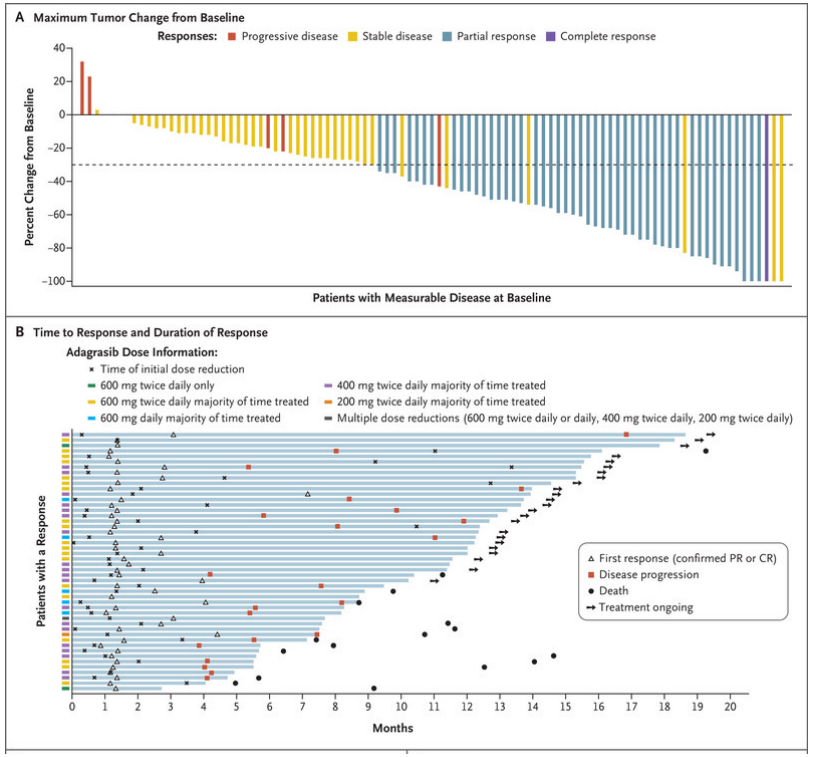
AMG 510 (or sotorasib) and MRTX849 or (adagrasib) → first 2 KRAS G12C inhibitors that form a covalent bond with cysteine12 within the switch-II pocket, locking KRAS in the inactive state to be FDA approved in metastatic KRAS G12C MT NSCLC ≥1 prior line of systemic therapy

Single-arm, open-label, phase 2 trial (CodeBreaK100)



n=126
ORR 37% (3.2% CR, 34% PR)
Median DOR 11.1 mos

Single-arm, open-label, phase 2 trial (KRYSTAL-1)



n=112
ORR 43% (1% CR, 42% PR)
Median DOR 8.5 mos

Jänne PA et al. *N Engl J Med*. 2022;387(2):120-131; Skoulidis F et al. *N Engl J Med*. 2021;384(25):2371-2381; Zhu C et al. *Mol Cancer*. 2022;21(1):159.

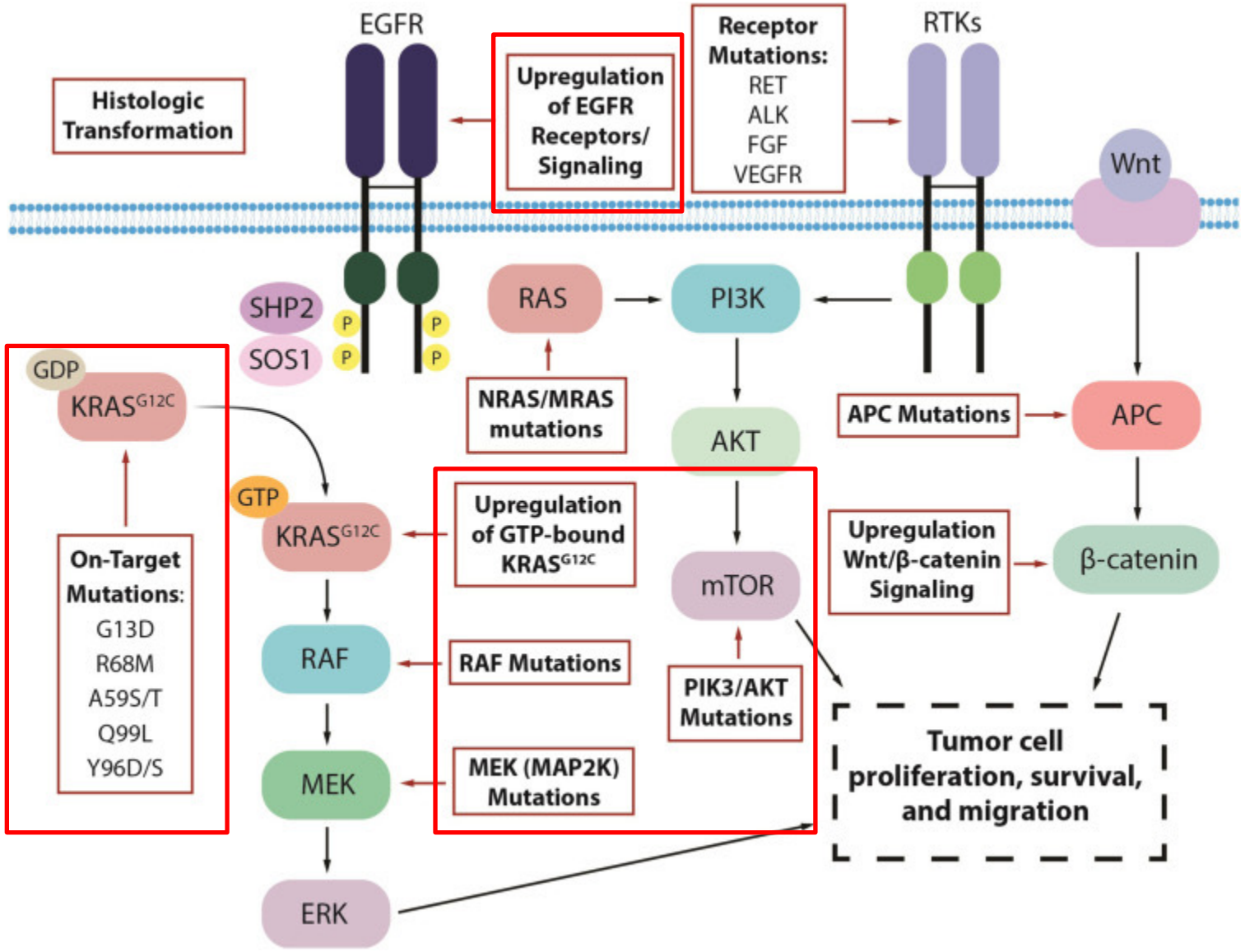
Switch II pocket covalent KRAS G12C inhibitors in CRC

Monotherapy	Sotorasib	Adagrasib	Divarasib
Phase, n subjects	Ph II, n=62	Ph I-II, n=44	Ph I, n=55
Median lines of prior therapy	3	3	2
ORR	9.7% (0 CRs)	23% (0 CRs)	29.1% (1 CR)
Median DOR	4.2 mos	4.3 mos	7.1 mos
Median PFS	4.0 mos	5.6 mos	5.6 mos

Divarasib 5-20X potent and up to 50X as selective in vitro as sotorasib and adagrasib

Fakih MG et al. *Lancet Oncol*. 2022;23(1):115-124; Sacher A et al. *N Engl J Med*. 2023;389(8):710-721; Yaeger R et al. *N Engl J Med*. 2023;388(1):44-54.

Mechanisms of resistance to KRAS G12C inhibitors



Limited activity of monotherapy for KRAS G12C inhibition in CRC

Adaptive feedback loop and resistance similar to BRAF V600E mutant colorectal cancer → targeting EGFR (thought to be the primary driver of feedback reactivation) → most KRAS G12C inhibitors are now paired with anti-EGFR

Ji J et al. *Onco Targets Ther*. 2022;15:747-756.

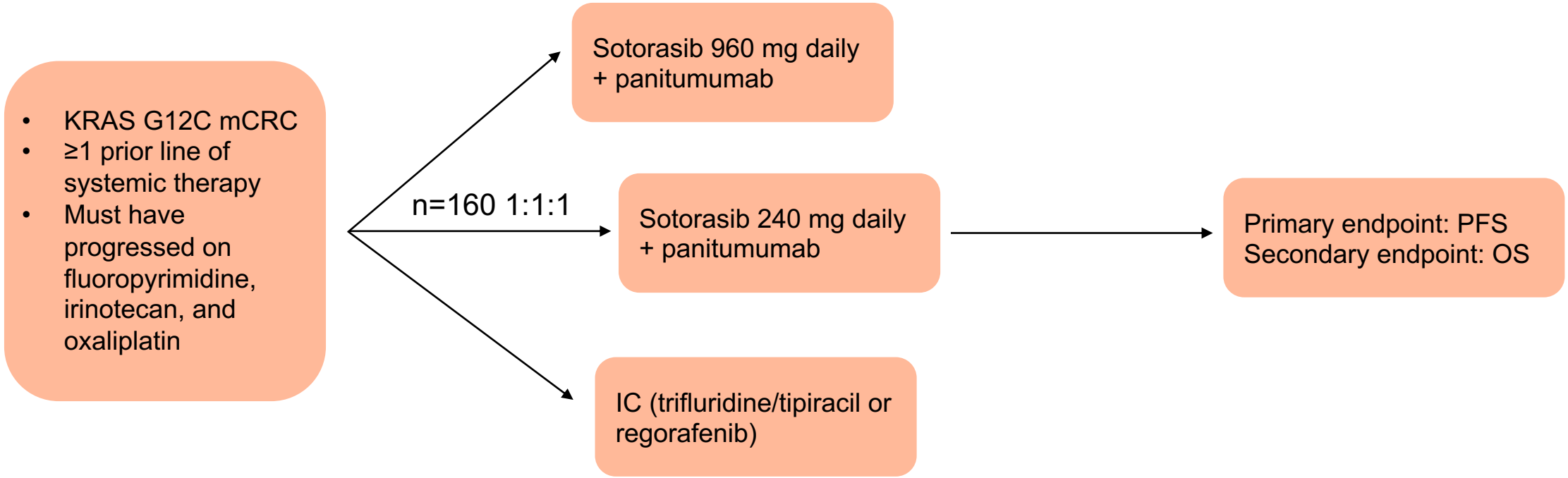
KRAS G12C inhibitors + anti-EGFR in CRC

Combination	Sotorasib + panitumumab	Adagrasib + cetuximab	Divarasib + cetuximab
Phase, n subjects	Ph Ib, n=40	Ph I-II, n=32	Ph Ib, n=29
Dose	960 mg daily + 6 mg/kg IV Q2W	600 mg daily + 500 mg/m^2 Q2W (or 400 mg/m^2 loading → 250 mg/m^2 QW)	200-400 mg daily + 400 mg/m^2 loading → 250 mg/m^2 QW
Median lines of prior therapy	2	3	2
ORR	30%	46% (0 CRs)	62%
Median DOR	NR	7.6 mos	NR
Median PFS	NR	6.9 mos	NR
Median OS	NR	13.4 mos	NR
Grade ≥3 TRAEs	22.5%	16%	38%

Desai J et al. AACR 2023. Abstract CT029; Kuboki Y et al. ESMO 2022. Abstract 3150; Yaeger R et al. *N Engl J Med*. 2023;388(1):44-54.

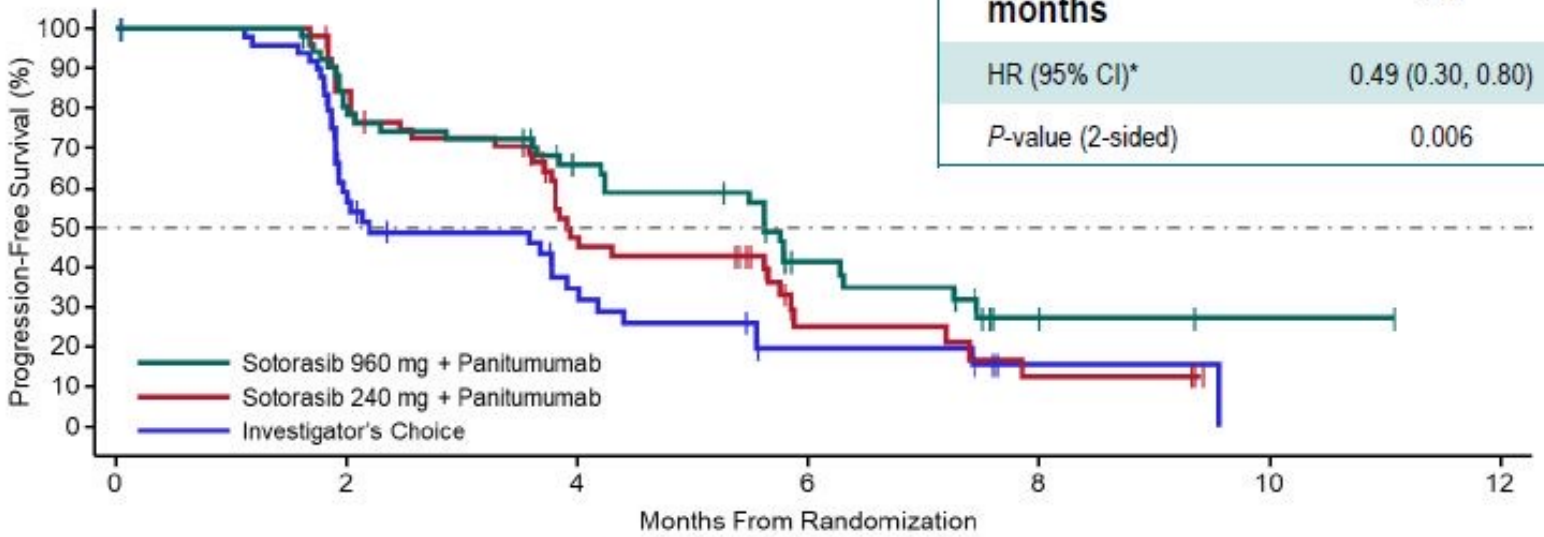
KRAS G12C inhibitors + anti-EGFR in CRC

Global, phase III, randomized, open-label CodeBreaK 300



Primary Endpoint: PFS in Intent-to-Treat Population

	Sotorasib 960 mg + Panitumumab (n = 53)	Sotorasib 240 mg + Panitumumab (n = 53)	Investigator's Choice (n = 54)
Median PFS, months	**5.6**	**3.9**	**2.2**
HR (95% CI)*	0.49 (0.30, 0.80)	0.58 (0.36, 0.93)	–
P-value (2-sided)	0.006	0.030	–



Number of Patients at Risk:							
Sotorasib 960 mg + Panitumumab	53	40	28	13	2	1	0
Sotorasib 240 mg + Panitumumab	53	43	20	6	3	0	
Investigator's Choice	54	24	12	5	1	0	

After a median follow-up of 7.8 months, sotorasib (960 mg and 240 mg) in combination with panitumumab significantly improved PFS by BICR versus investigator's choice

PFS was tested using stratified log-rank test. *HR is sotorasib 960 mg + panitumumab / investigator's choice therapy, or sotorasib 240 mg + panitumumab / investigator's choice therapy.
BICR, blinded independent central review; HR, hazard ratio; PFS, progression-free survival.

Activity Outcomes

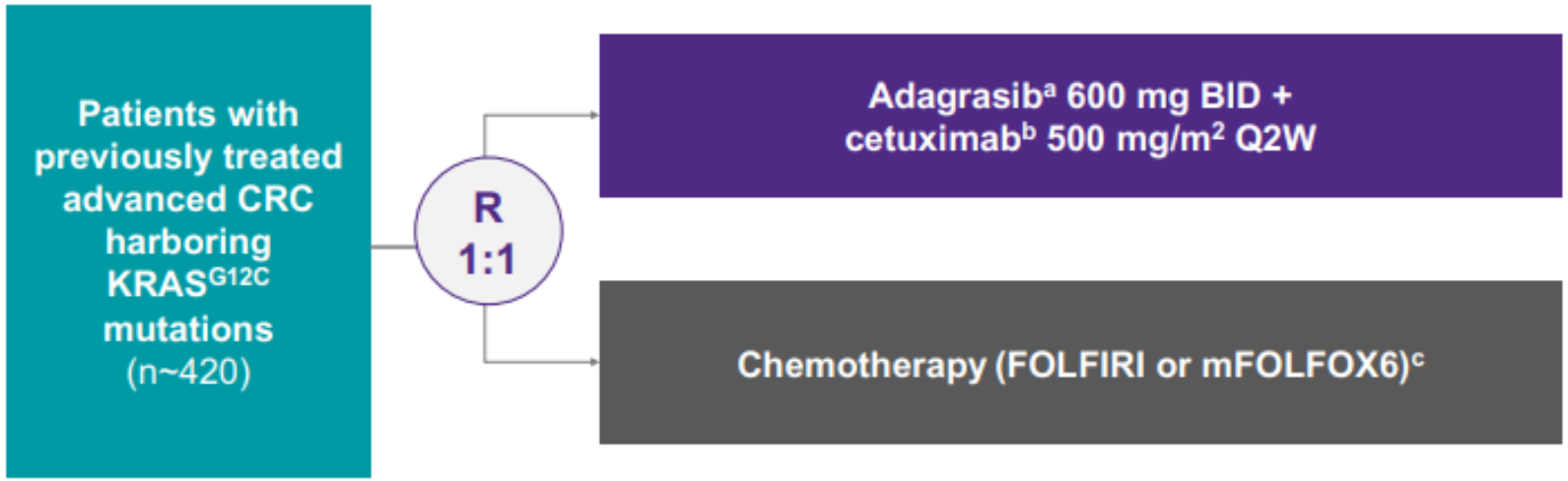
Response by BICR	Sotorasib 960 mg + Panitumumab (n = 53)	Sotorasib 240 mg + Panitumumab (n = 53)	Investigator's Choice (n = 54)
ORR, % (95% CI)*†	26 (15.3–40.3)	6 (1.2–15.7)	0 (0–6.6)
Complete response, n (%)	1 (2)	0	0
Partial response, n (%)	13 (25)	3 (6)	0
Stable disease, n (%)	24 (45)	33 (62)	25 (46)
Progressive disease, n (%)	12 (23)	13 (25)	17 (31)
Not evaluable / not done, n (%)	3 (6)	2 (4)	11 (20)
DCR, % (95% CI)*	72 (57.7–83.2)	68 (53.7–80.1)	46 (32.6–60.4)

ORR and DCR by BICR were higher with sotorasib (960 mg and 240 mg) + panitumumab versus investigator's choice

The intention-to-treat analysis set included all patients who underwent randomization.
*95% CIs were estimated using the Clopper-Pearson method. BICR, blinded independent central review; DCR, disease control rate; ORR, objective response rate
†Two patients (4%) in the 240 mg arm and 1 patient (2%) in the investigator's choice arm had non-complete response/non-progressive disease; these patients had BICR assessed non-target disease only

KRAS G12C inhibitors + anti-EGFR in mCRC

Open label, randomized phase III KRYSTAL-10



Primary endpoint: PFS, OS
Secondary endpoint: Safety, ORR, DOR

IV, intravenously; Q2W, every 2 weeks.
[a]Adagrasib administered orally in 4-week cycles 600 mg BID on a continuous basis until disease progression. [b]Cetuximab administered Q2W by IV 500 mg/m2. [c]A VEGF/VEGFR inhibitor may be given per investigator discretion.

Tabernero J et al. *Ann Oncol*. 2021;32(suppl 3):S121.

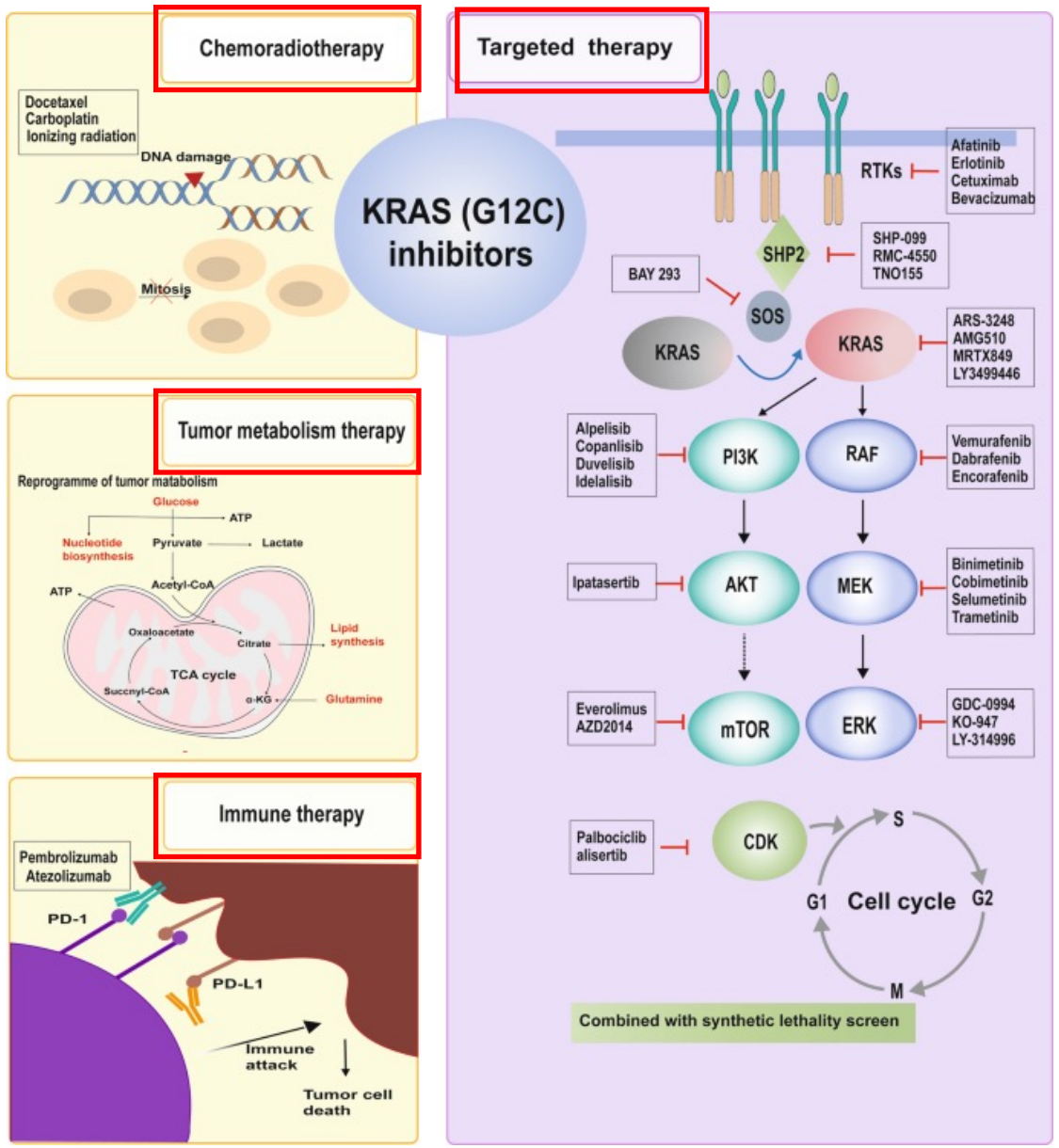
New switch II pocket covalent KRAS G12C inhibitors in CRC

	JAB-21822	JDQ443	LY3537982	D-1553
Trial	NCT05009329	KontRASt-01	LOXO-RAS-20001	NCT04585035
Phase, n subjects	Ph I/II, n=9	Ph Ib/II, n=42	Ph I, n=17	Ph I/II, n=24
Median lines of prior therapy	2	3	2	2
ORR	CRC cohort NR 70% (7/10 pts w/NSCLC)	CRC cohort NR 55% (6/11 pts w/NSCLC)	7% (1/15 evaluable CRC subjects)	20.8% (1 CR)
Median DOR	NR	NR	NR	NR
Median PFS	NR	NR	NR	7.6 mos

- **Ph I trial JNJ-74699157 enrollment stopped due to DLTs and lack of efficacy**
- **LY3499446 discontinued in Ph I trial due to toxicity**

Cassier PA et al. ASCO 2023. Abstract 9007; Li J et al. ASCO 2023. Abstract 3089; Murciano-Goroffet YR et al. AACR 2023. CT028; Peng S-B et al. *Cancer Res*. 2021;81(13_Suppl):1259; Ruan D et al. ASCO 2023. Abstract 3563; Wang J et al. *Oncologist*. 2022;27(7):536-e553.

Future of KRAS G12C inhibitors in CRC: Combinations



CodeBreaK-101

- n=46 mCRC (2 median prior lines of therapy) FOLFIRI-pmab + sotorasib 960 mg oral QD
- ORR 58.1%, 45.5% grade ≥3 AEs

Proffered Paper session - Developmental therapeutics

Date Sun, 22.10.2023
Time 08:30 - 10:00
Chairs Lillian L. Siu (Toronto, Canada), Elena Garralda (Barcelona, Spain)
Room Málaga Auditorium - Hall 10
Session Type Proffered Paper session

Proffered Paper session

652O - Preliminary clinical activity of RMC-6236, a first-in-class, RAS-selective, tri-complex RAS-MULTI(ON) inhibitor in patients with KRAS mutant pancreatic ductal adenocarcinoma (PDAC) and non-small cell lung cancer (NSCLC)

Presentation Number 652O
Speakers Kathryn C. Arbour (New York, United States of America)
Lecture Time 08:30 - 08:40

Proffered Paper session

653O - Glecirasib (KRAS G12C inhibitor) in combination with JAB-3312 (SHP2 inhibitor) in Patients with KRAS p.G12C mutated solid tumors.

Presentation Number 653O
Speakers Jie Wang (Beijing, China)
Lecture Time 08:40 - 08:50

Proffered Paper session

LBA33 - A first-in-human phase 1 study of a novel KRAS G12D inhibitor HRS-4642 in patients with advanced solid tumors harboring KRAS G12D mutation

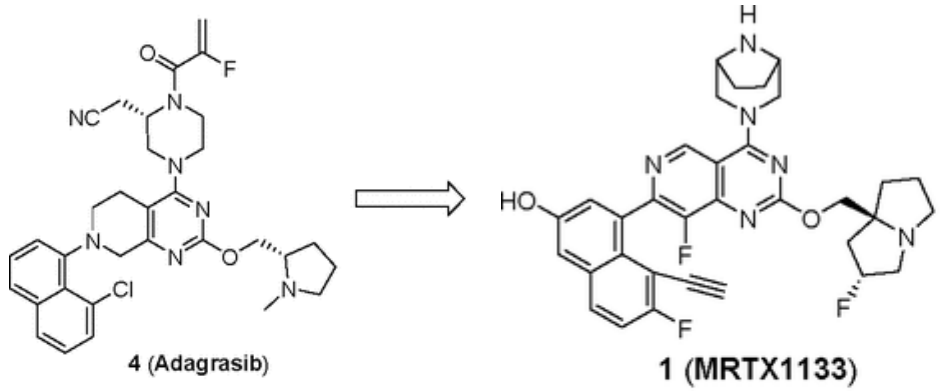
Presentation Number LBA33
Speakers Caicun Zhou (Shanghai, China)

Huang L et al. Signal Transduct Target Ther. 2021 Nov;6(1):386
Hong DS, et al. J Clin Oncol. 2023;41(16 suppl):3513

KRAS G12D inhibitor in CRC

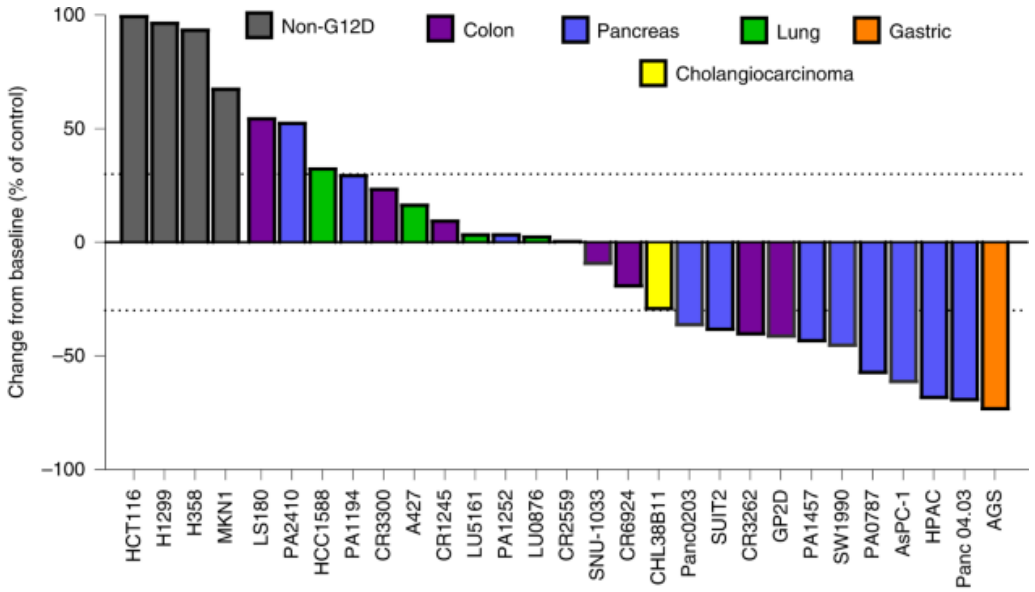
MRTX1133, a KRAS G12D inhibitor, is a noncovalent, potent, selective inhibitor that binds to the switch II pocket:

- Builds on adagrasib's structure → pyrido[4,3-d]pyrimidine scaffold instead of tetrahydropyridopyrimidine core
- Optimized three substituents on the 2-, 4-, and 7-positions of pyrido[4,3-d]pyrimidine core
  - Protonated piperazinyl group at the C4-position → an ionic pair with the mutant Asp12
  - + charged pyrrolidinyl moiety at the C2-position → ionic interaction with Glu62
  - C7-naphthyl occupies a deep hydrophobic pocket as reported previously for adagrasib



High-affinity interaction with GDP-loaded KRAS G12D

- **IC_{50} <2 nM**
- **Approx 700-fold selectivity for KRAS G12D over KRAS WT**



A Phase 1/2 Multiple Expansion Cohort Trial of MRTX1133 in Patients With Advanced Solid Tumors Harboring a KRAS G12D Mutation (NCT05737706)

Hallin J et al. *Nat Med*. 2022;28(10):2171-2182.

Wang X et al. *J Med Chem*. 2022;65(4):3123-3133.

Tri-Complex RAS(ON) inhibitors

1. Small molecule inhibitor binds non-covalently to an abundant intracellular chaperone protein, cyclophilin A (CypA)

2. RMC-XXXX-CypA binary complex engages RAS(ON) or GTP-bound state → stable tri-complex that inhibits RAS binding to effectors
   - Tri-complex enables stable binding: a) non-covalent contacts between RMC-XXXX and switch I and II motifs, b) neomorphic contacts between CypA and KRAS, c) covalent modification of cysteine and asp residues via RMC-XXXX warhead

RMC-6236
First-in-class, potent, oral RAS-selective tri-complex RAS^{MULTI}(ON) inhibitor induced tumor regression in preclinical models across KRAS G12D, G12V, G12R tumors

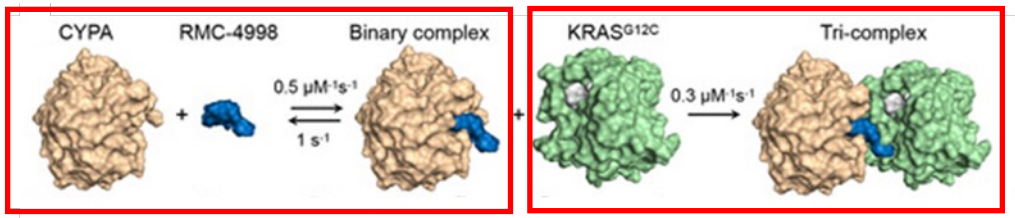


Table S2. Antiproliferative potency of RMC-4998, RMC-6291 and adagrasib

	Cell Line	Cancer type	Mutation	RMC-4998 EC_{50} (nM)	RMC-6291 EC_{50} (nM)	Adagrasib EC_{50} (nM)
KRAS^{G12C}	MIA Paca-2	Pancreas	*KRAS^{G12C}*	0.180	0.0661	0.324
	NCI-H1373	Lung	*KRAS^{G12C}*	0.179	0.0708	8.91
	NCI-H2030	Lung	*KRAS^{G12C}*	0.266	0.123	3.16
	NCI-H2122	Lung	*KRAS^{G12C}*	0.319	0.145	8.71
	SW1463	Intestine/ Colorectum	*KRAS^{G12C}*	0.470	0.219	17.4
	SW1573	Lung	*KRAS^{G12C}*	> 302	> 302	> 302
	UMUC3	Urinary/ Bladder	*KRAS^{G12C}*	0.0618	0.0355	1.23
	Calu1	Lung	*KRAS^{G12C}*	0.263	0.0891	8.13
	NCI-H1792	Lung	*KRAS^{G12C}*	0.141	0.0631	2.63
	NCI-H23	Lung	*KRAS^{G12C}*	0.304	0.178	3.47
	NCI-H358	Lung	*KRAS^{G12C}*	0.278	0.107	1.51
	SW837	Intestine/ Colorectum	*KRAS^{G12C}*	0.293	0.102	3.16
	HCC-44	Lung	*KRAS^{G12C}*	0.230	0.0813	19.1
	IA-LM	Lung	*KRAS^{G12C}*	0.556	0.251	3.98
	KYSE-410	Esophageal	*KRAS^{G12C}*	> 302	> 302	> 302
	LU65	Lung	*KRAS^{G12C}*	0.184	0.0813	5.75
	OV56	Ovarian	*KRAS^{G12C}*	> 302	> 302	> 302
			Median EC_{50}	**0.278**	**0.107**	**5.75**

Koltun ES et al. *Cancer Res*.2022;82(12_Supplement):3597.
Schulze CJ et al. *Science*. 2023 Aug; 381(6659):794-799.

Tri-Complex RAS(ON) inhibitors

Phase I/Ib open-label, multicenter trial of oral RMC-6236 in advanced solid tumors with KRAS p.G12 mutations

- Histologically confirmed advanced solid tumor with KRAS p.G12A, KRAS p.G12D, KRAS p.G12R, KRAS p.G12S, or KRAS p.G12V mutations
- KRASG12C excluded
- Received prior standard therapy appropriate for tumor type and stage

RMC-6236-001
n=141 →

Primary endpoint: Safety and DLTs

	Cell Line	Cancer type	Mutation	RMC-4998 EC_{50} (nM)	RMC-6291 EC_{50} (nM)	Adagrasib EC_{50} (nM)
Non-$KRAS^{G12C}$	HCC827	Lung	$EGFR^{E746\_A750del}$	730	1450	2290
	NCI-H1975	Lung	$EGFR^{L858R/T790M}$	1600	4370	4900
	AsPC1	Pancreas	$KRAS^{G12D}$	998	2340	1450
	A427	Lung	$KRAS^{G12D}$	217	562	1320
	SW480	Intestine/ Colorectum	$KRAS^{G12V}$	387	813	1230
	A549	Lung	$KRAS^{G12S}$	604	1660	1820
	MeWo	Skin	$NF1^{LOF}$	401	891	2090
	HCT116	Intestine/ Colorectum	$KRAS^{G13D}$	356	603	1860
	NCI-H1299	Lung	$NRAS^{Q61K}$	1600	4370	4900
	COLO205	Intestine/ Colorectum	$BRAF^{V600E}$	393	2880	871
	A375	Skin	$BRAF^{V600E}$	339	912	1290
			Median EC_{50}	**401**	**1450**	**1820**
			Fold selectivity*	**1450**	**13500**	**316**

*Fold selectivity was calculated by (median EC_{50} non-$KRAS^{G12C}$)/(median EC_{50} $KRAS^{G12C}$) and rounded to 3 significant figures

Questions:

- **Possible that a dependency on CypA expression could limit the therapeutic utility of tri-complex inhibitors?**
- **Impact on RAS WT protein → safety/tolerability?**

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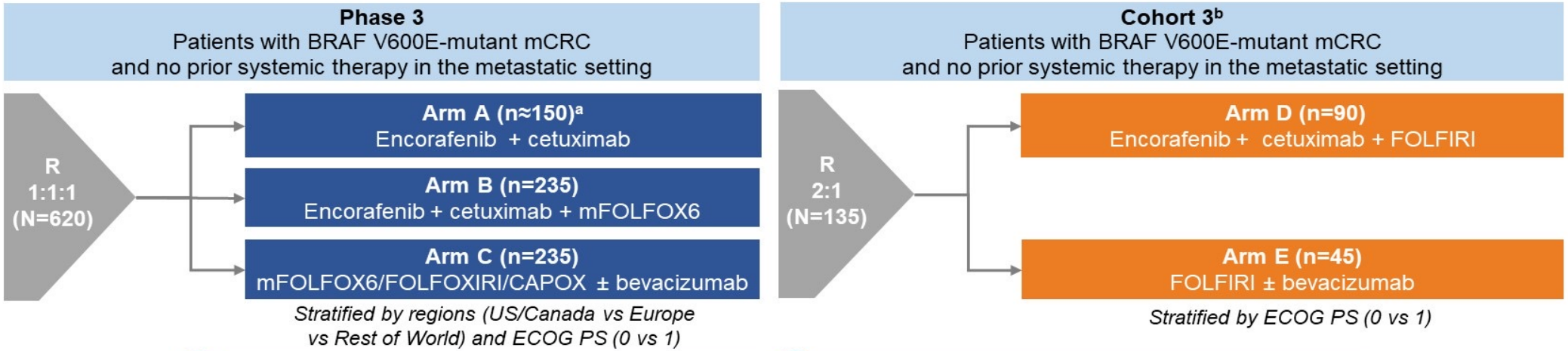
Koltun ES et al. Cancer Res.2022;82(12_Supplement):3597
NCT05379985
Schulze CJ et al. Science. 2023 Aug; 381(6659):794-799

BRAF V600E mCRC

Phase 3 and Cohort 3 Study Design

- BREAKWATER (NCT04607421) is an ongoing, open-label, multicenter, randomized, phase 3 study evaluating 1L EC ± CT vs SOC CT alone in patients with BRAF V600E-mutant mCRC
  - In the BREAKWATER SLI, which evaluated 57 patients with mCRC who had received ≤1 prior treatment, EC + CT showed encouraging antitumor activity
  - Based on these SLI results, EC + mFOLFOX6 was selected as the recommended phase 3 regimen



Inclusion criteria

- Age ≥16 years (or ≥18 years based on country)
- No prior systemic treatment for metastatic disease
- Measurable disease (RECIST 1.1)
- BRAF V600E-mutant mCRC (based on blood or tumor tissue)
- ECOG PS 0 or 1
- Adequate bone marrow, hepatic, and renal function

Exclusion criteria

- Prior BRAF or EGFR inhibitors
- Symptomatic brain metastases (unless stable for ≥4 weeks prior to randomization)
- MSI-H/dMMR tumors (unless ineligible to receive immune checkpoint inhibitors due to a pre-existing medical condition)

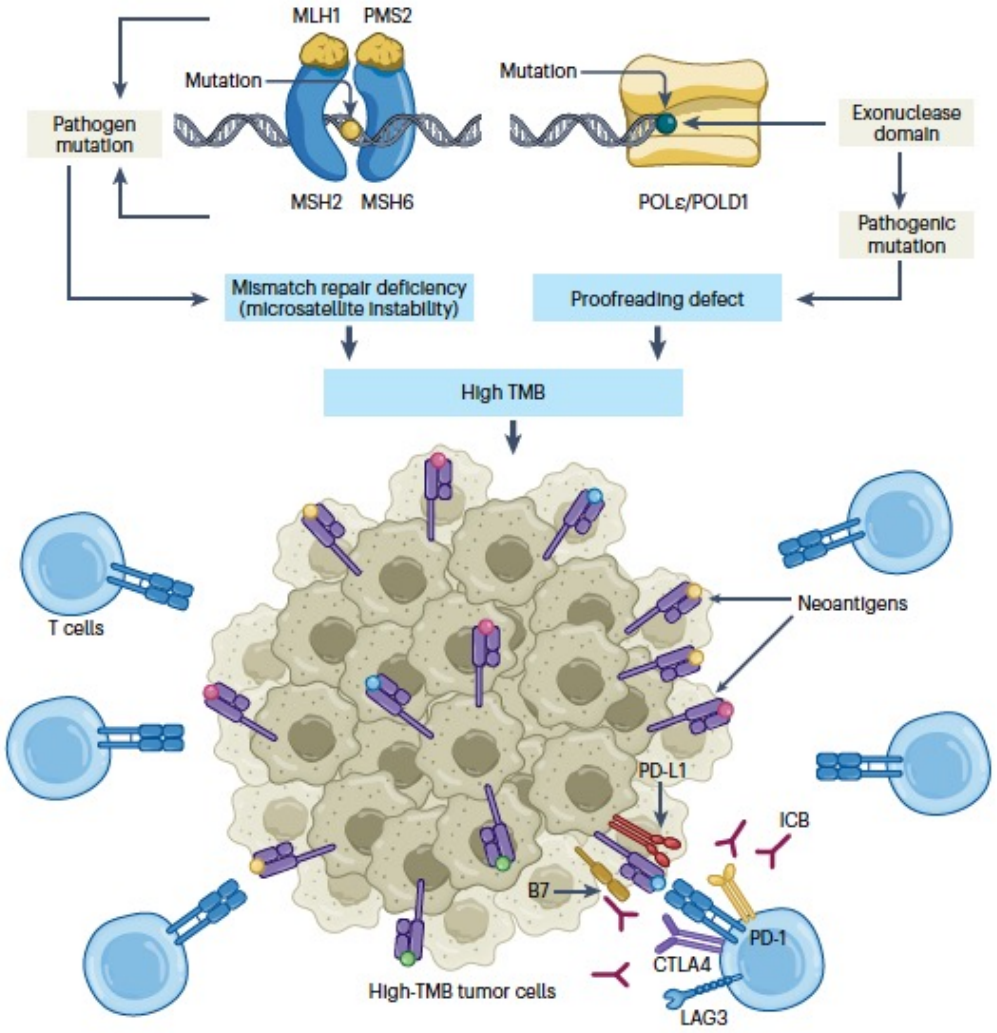
[a] In the phase 3 portion of the study, randomization to arm A will cease upon site institutional review board/ethics committee and competent authority approval of protocol amendment 5. Following this protocol amendment, randomization is 1:1 into arms B and C. [b] Cohort 3 will begin after phase 3 enrollment is complete.
CAPOX, capecitabine/oxaliplatin; dMMR, deficient mismatch repair; ECOG PS, Eastern Cooperative Oncology Group performance status; EGFR, epidermal growth factor receptor; FOLFIRI, fluorouracil/leucovorin/irinotecan; mFOLFOX6, modified fluorouracil/leucovorin/oxaliplatin; FOLFOXIRI, fluorouracil/leucovorin/oxaliplatin/irinotecan; mCRC, metastatic colorectal cancer; MSI-H, microsatellite instability-high cancer; RECIST, Response Evaluation Criteria in Solid Tumors.

Targeted Therapies in mCRC

- Recent advances have demonstrated the relevance of actionable genomic alterations in mCRC with the recent approval of several agents against mutations including HER-2, BRAFv600E, KRAS G12C
- Future directions:
  - Evaluation of target directed therapy in the early lines of therapy (BREAKWATER, MOUNTANEER3)
  - In combination with IO (SWOG 2107) – enco/cetux +/- nivo, chemotherapy (BREAKWATER)

Why Is It Important to Test for MSI-H/dMMR in CRC?

- Lynch syndrome diagnosis/screening
- Prognostic marker
- Predictive biomarker for response to (neo)adjuvant chemotherapy
- Predictive biomarker for response to immunotherapy



- Alouani E, et al., *Nat Cancer* 2022

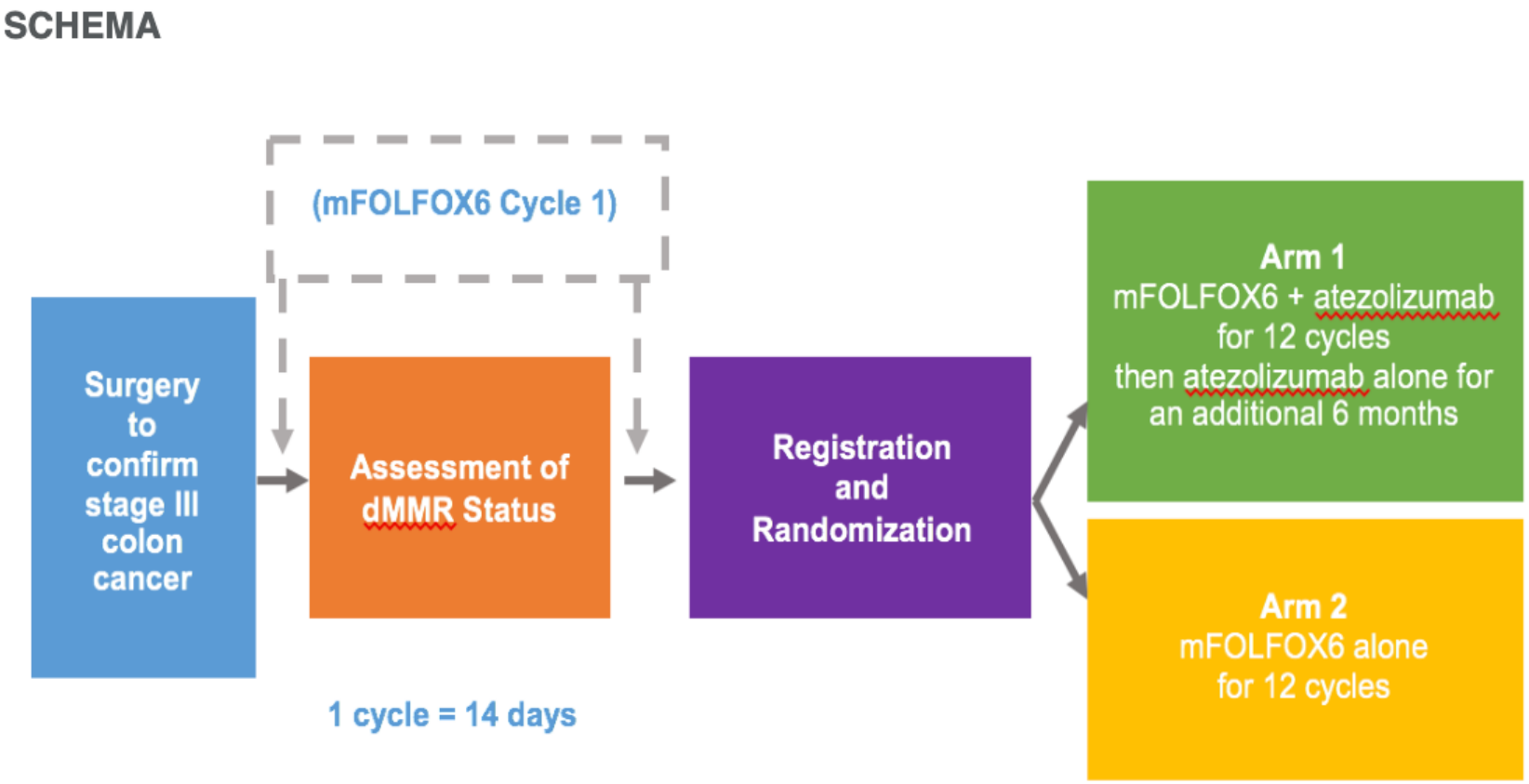
ICI Trials in Localized MSI-H/dMMR CRC

Clinical Trial	Eligible Patients	ICI Therapy	Demographics	Efficacy Endpoints	Adverse Events
NICHE (Chalabi, *Nature Med* 2020)	Localized colon cancer	Neoadjuvant nivo d1,15 + ipilimumab d1 (n = 21)	• 81% stage III • 33% Lynch syndrome	• 100% path response • 95% MPR • 60% pCR	• 10% grade 3 • 2% grade 4
NICHE-2 (Chalabi, ESMO 2022)	cT3 and/or N+ colon cancer	Neoadjuvant nivo d1,15 + ipilimumab d1 (n = 112)	• 74% high-risk stage III • 31% Lynch syndrome	• 95% MPR • 67% pCR • 100% R0 sx	• 61% any AEs • 4% grade ≥ 3 AEs
Dostarlimab (Cercek, ASCO 2022, *NEJM* 2022)	Stage II/III rectal cancer	Neoadjuvant dostarlimab q3 weeks x 6 months (n = 14)	• 78% T3/T4 • 94% N+ • 59% germline MMR mutation (100% BRAF wt)	• 100% cCR	• 75% any AEs • 0% grade ≥ 3 AEs
Sintilimab (Chen, *Lancet GH* 2023)	Stage II/III rectal cancer	Neoadjuvant sintilimab q3 weeks x 2-8 cycles (n = 17)	• 59% T3, 29% T4 • 82% N+ • 35% Lynch syndrome	• pCR + cCR = 75% (3/6 pts s/p sx w/ pCR) • 6% PD	• 6% grade 3 AE (stopped tx; no cCR)
Toripalimab (Hu, *Lancet Gastro Hep* 2022)	cT3/T4 and/or N+ colon cancer	Neoadjuvant toripalimab +/- celecoxib x2, +/- adjuvant T +/- C (n = 34)	• 100% T3/T4 • 84% N+ (T+C), 95% (T) • 24% Lynch (T+C), 6% Lynch (T)	• 84% pCR (T+C) • 65% pCR (T) • 94% MPR (T+C) • 100% MPR (T)	• 59% grade 1-2 AEs • 3% grade 3 neoadj, 3% grade 3 adj)
Pembrolizumab (Ludford, *JCO* 2023)	Localized unresectable or high-risk resectable solid tumors	Neoadjuvant pembrolizumab q3 weeks x 6 mos +/- add'l therapy (n = 27 CRC, 35 total)	• 74% stage III • 46% Lynch syndrome	• 67% pCR (79% CRC) • ORR 82%	• 37% grade 1-2 AEs • 6% grade 3 AEs

ICI Trials in Localized MSI-H/dMMR CRC

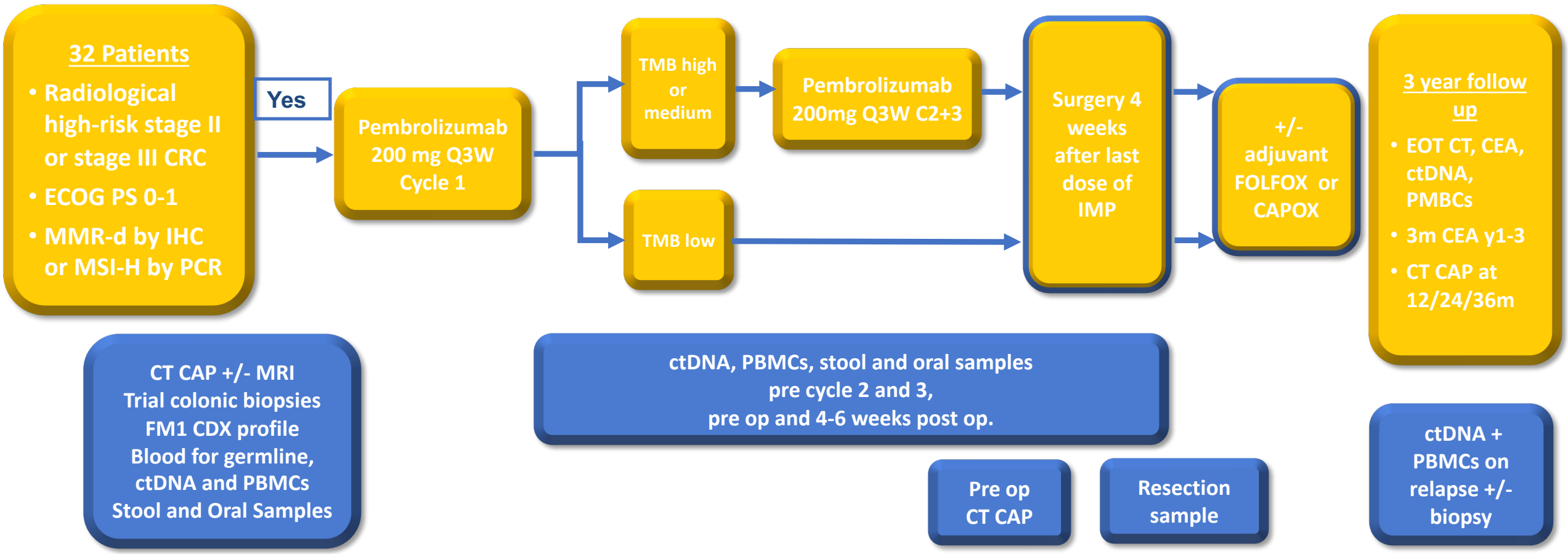
- Potential for nonoperative management in localized disease
  - Feasibility of colonoscopic surveillance, NOM
  - Differences between colon and rectal cancer treatment, risk/benefit profile
- Radiographic and pathologic discordance
  - Potential for overtreatment with neoadjuvant therapy?
  - “Neoadjuvant surgery” for patients with less-than-optimal response to ICIs?
- Responses remarkable in localized disease, but not ubiquitous
  - Lynch vs. non-Lynch and impact on efficacy
  - How to predict primary progressors?
- Optimal duration of ICI therapy unclear
  - Variable treatment courses in completed trials – may be role for personalization
- Need for circulating tumor DNA (ctDNA) or other predictive biomarkers to assess response to ICI therapy

ATOMIC: Adjuvant Trial of Microsatellite Instability Colon Cancer [Alliance A021502]



- PI: Frank Sinicrope

NEOPRISM-CRC: Neoadjuvant Pembrolizumab for early stage dMMR/MSI-H CRC stratified to TMB. A Multisite Phase 2 investigator-initiated trial

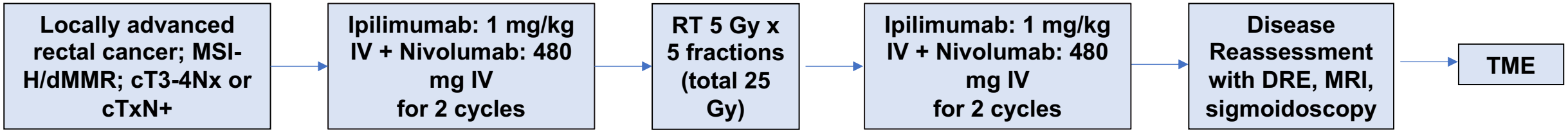


Primary endpoint: Pathological complete response rate

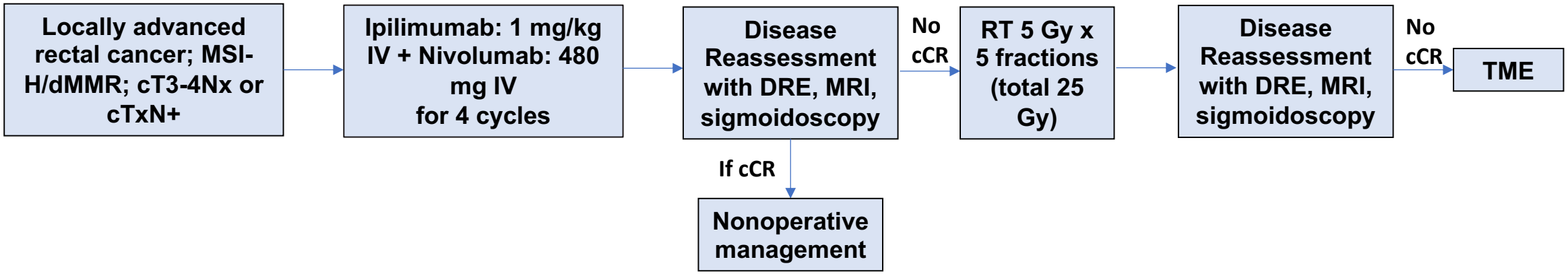
Secondary endpoints: 3-year RFS, OS, ctDNA response to neoadjuvant therapy, minimal residual disease monitoring, genomic and microbiome signatures to determine immunotherapy resistance/sensitivity

KK Shiu UCL 2022
MISP 58807
https://clinicaltrials.gov/ct2/show/NCT05197322

EA2201: A Phase II Study of Neoadjuvant Nivolumab Plus Ipilimumab and Short Course Radiation in MSI-H Locally Advanced Rectal Cancer



Current primary endpoint: Pathologic complete response rate (pCR)



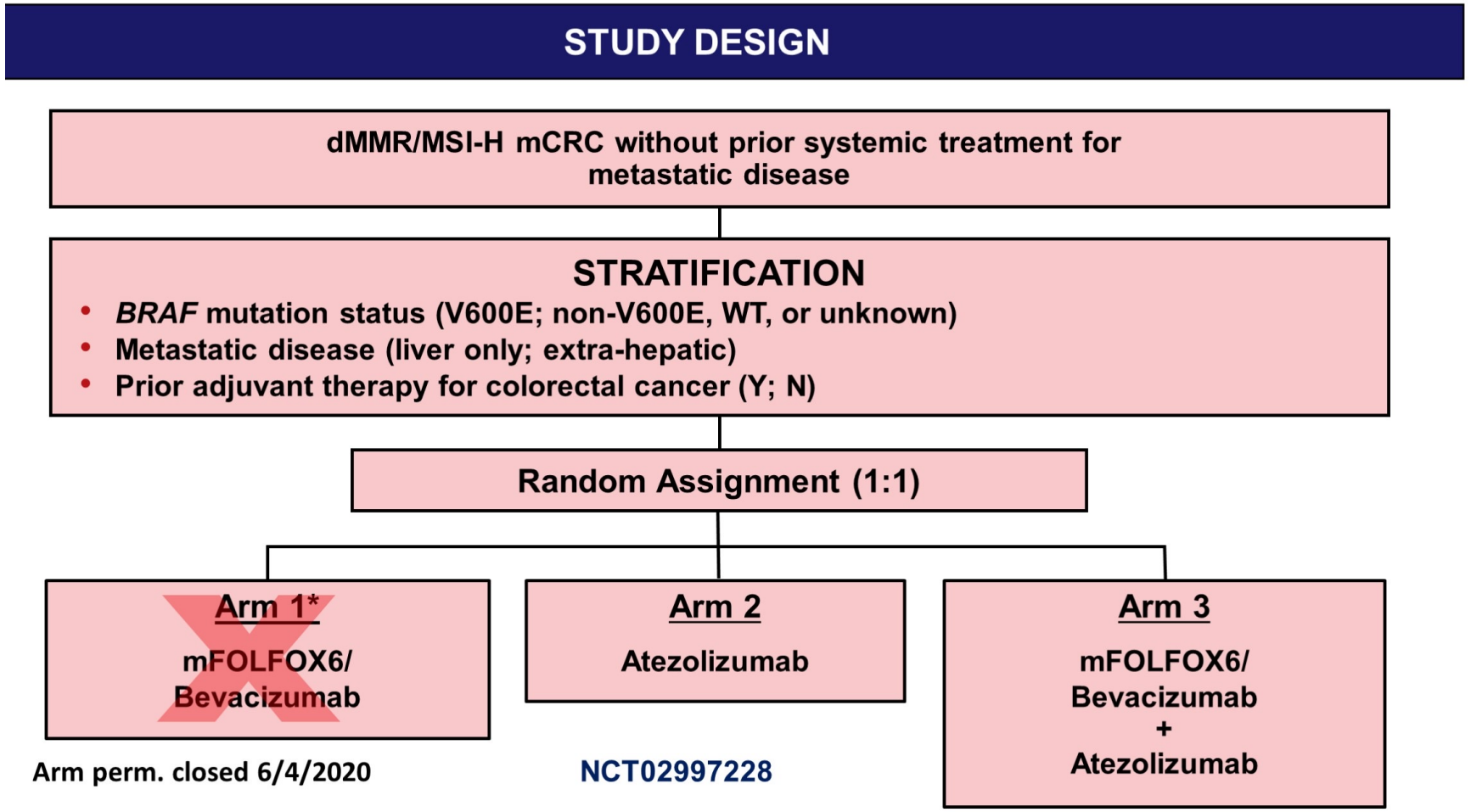
Proposed primary endpoint: Clinical complete response rate (cCR)

PI: Kristen Ciombor

Statistical design:

- Two-stage single-arm phase II study (n=31)

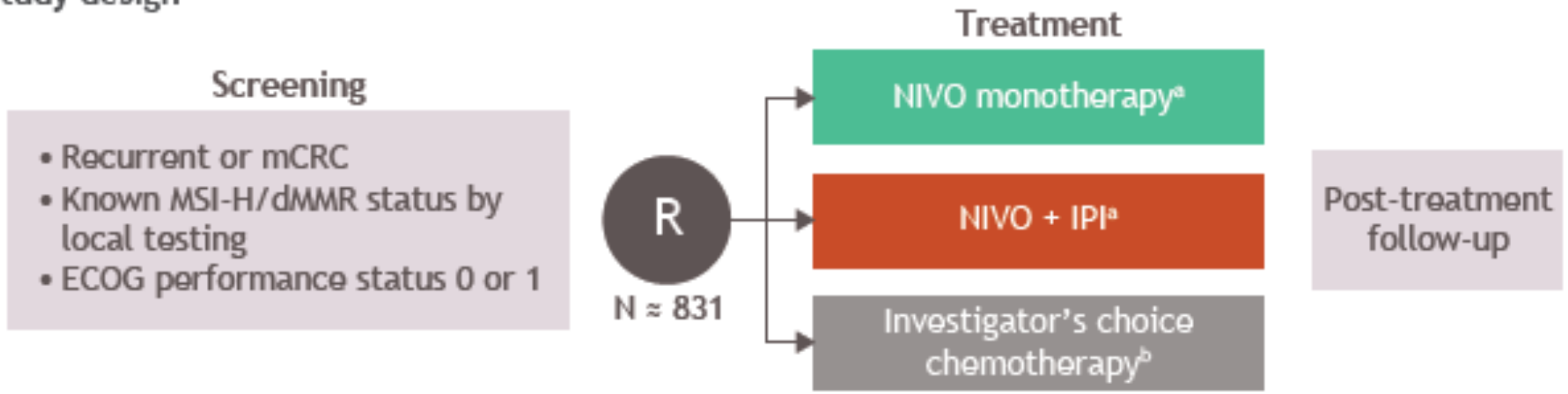
NRG-GI004/SWOG-S1610: Colorectal Cancer Metastatic dMMR Immuno-Therapy (COMMIT) Study: A Randomized Phase III Study of Atezolizumab with or without mFOLFOX6/Bevacizumab Combination Chemotherapy in the First-line Treatment of Patients with Deficient DNA Mismatch Repair (dMMR) or Microsatellite Instability High (MSI-H) Metastatic Colorectal Cancer



- Presented by MJ Overman, ASCO GI 2023

A phase 3 study of nivolumab, nivolumab + ipilimumab, or chemotherapy for microsatellite instability-high/mismatch repair-deficient metastatic colorectal cancer: CheckMate 8HW

Figure 1. Study design



PRESS RELEASE on 12/7/2023 – STUDY POSTIVE FOR BOTH PRIMARY ENDPOINTS

- Dual primary endpoints: PFS by BICR (nivo/ipi vs nivo across all lines)
- Other key endpoints: PFS by BICR (nivo/ipi vs. nivo in 1L), PFS by investigator assessment, ORR, OS, DCR, TTR, DOR, safety

- Presented by T Andre, World GI 2022

IN SUMMARY

- Test for mismatch repair deficiency/microsatellite instability at diagnosis for every stage of CRC
  - Ensure adequate diagnostic testing methods
- ICIs a very helpful therapeutic strategy in most MSI-H/dMMR CRC
  - Efficacy increases with earlier stages of disease
  - Toxicity profile favorable for majority of patients
- Need biomarkers to help predict response, non-response to ICIs
  - Molecular markers, Lynch vs. non-Lynch, tumor microenvironment, others all potential impacting factors
- Optimal duration of therapy and role of singlet vs. doublet ICIs unclear
  - Circulating tumor DNA may be helpful for assessing/quantifying response
- ICIs may spare patients from morbidity from other treatment modalities (surgery, radiation, chemotherapy) and lead to durable responses/cure
- Enroll patients in ongoing and upcoming prospective trials to answer these questions!

UPDATES IN IO IN MSS MCRC

Resistance Mechanisms to Immune Checkpoint Inhibitor Therapy in MSS mCRC

- Low immunogenicity for $CD8^+$ T cell recognition (low tumor mutational burden)
- Defects in antigen presentation machinery
- Overexpression of intrinsic immunosuppressive oncogenic pathways
- Immunosuppressive effects of the tumor microenvironment
- Key ? = efforts aimed at molecular pathways to enhance T cell recruitment to the TME

ASCO Gastrointestinal Cancers Symposium

Results from a phase 1a/1b study of botensilimab (BOT), a novel innate/adaptive immune activator, plus balstilimab (BAL; anti-PD-1 antibody) in metastatic heavily pretreated microsatellite stable colorectal cancer (MSS CRC)

Authors: Anthony B. El-Khoueiry, MD[1], Marwan G. Fakih, MD[2], Michael S. Gordon, MD[3], Apostolia M. Tsimberidou, MD, PhD[4], Andrea J. Bullock, MD, MPH[5], Breelyn A. Wilky, MD[6], Jonathan C. Trent, MD, PhD[7], Kim A. Margolin, MD, FACP, FASCO[8], Daruka Mahadevan, MD, PhD[9], Ani S. Balmanoukian, MD[10], Rachel E. Sanborn, MD[11], Gary K. Schwartz, MD[12], Bruno Bockorny, MD[5], Justin C. Moser, MD[3], Joseph E. Grossman, MD[13], Waldo Ortuzar Feliu, MD[13], Katherine Rosenthal, RN, MSN, OCN, CCRP[13], Steven J. O'Day, MD[13], Heinz-Josef Lenz, MD, FACP[1], Benjamin L. Schlechter, MD[14]

[1]University of Southern California Norris Comprehensive Cancer Center, Los Angeles, CA, USA, [2]City of Hope Comprehensive Cancer Center, Duarte, CA, USA, [3]Honor Health Research and Innovation Institute, Scottsdale, AZ, USA, [4]The University of Texas MD Anderson Cancer Center, Houston, TX, USA, [5]Beth Israel Deaconess Medical Center, Boston, MA, USA, [6]University of Colorado Cancer Center, Aurora, CO, USA, [7]Sylvester Comprehensive Cancer Center, University of Miami, Miami, FL, USA, [8]Providence Saint John's Cancer Institute, Santa Monica, CA, USA, [9]The University of Texas Health Sciences Center at San Antonio, San Antonio, TX, USA, [10]The Angeles Clinic and Research Institute, Los Angeles, CA, USA, [11]Earle A. Chiles Research Institute, Providence Cancer Institute, Portland, OR, USA, [12]Herbert Irving Comprehensive Cancer Center, Columbia University Medical Center, New York, NY, USA, [13]Agenus Inc., Lexington, MA, USA, [14]Dana-Farber Cancer Institute, Boston, MA, USA

Presented by: Anthony B. El-Khoueiry, MD

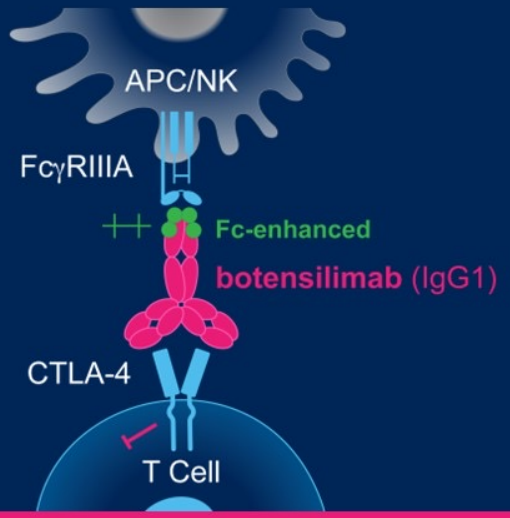
University of Southern California
Norris Comprehensive Cancer Center
Los Angeles, California, United States

January 21, 2023
Abstract Number: LBA8

Active in 'Cold' and IO Refractory Tumors

botensilimab

Fc-enhanced CTLA-4 Inhibitor

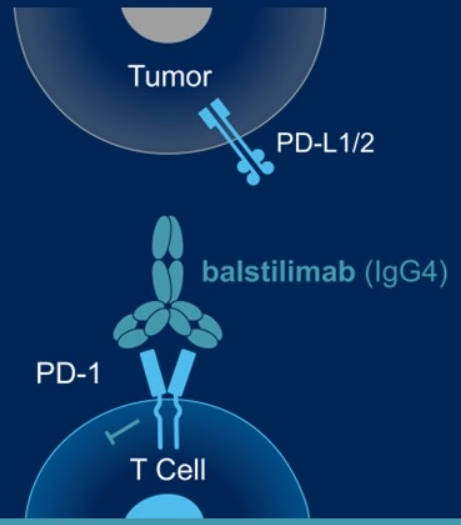


Active in 'cold' and IO refractory tumors[1,2]

- >300 patients treated across 4 trials
- ↑ T cell priming, expansion, memory[3,4]
- ↑ Frequency of activated APCs
- ↑ Treg depletion
- ↓ Complement mediated toxicity

balstilimab

PD-1 Inhibitor

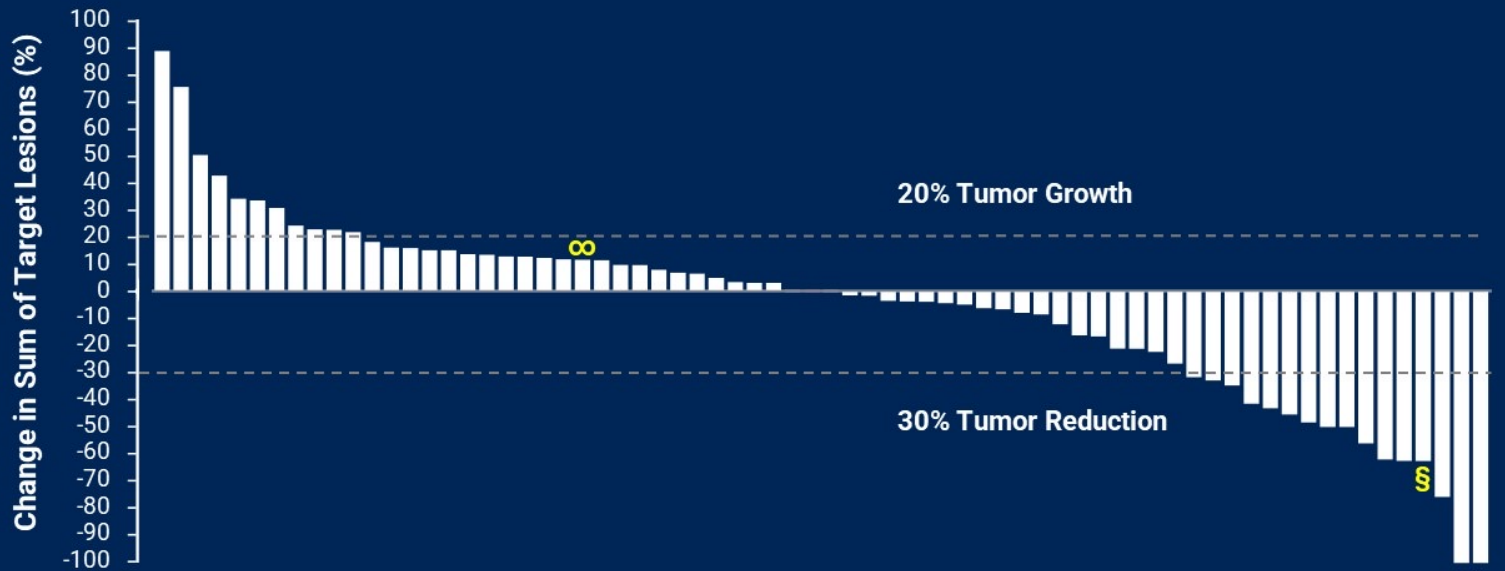


Safety and efficacy analogous to approved anti-PD-1 mAbs[5,6]

- >750 patients treated; 10 ongoing trials / 2 completed
- Complete blocker of PD-1-PD-L1/2 interactions
- Enhanced T cell activation and effector function

1. El-Khoueiry AB. SITC 2021 Annual Meeting. Poster #479. 2. Wilky B. SITC 2022 Annual Meeting. Oral #778. 3. Waight et al. Cancer Cell. 2018;33(6): 1033-1047. 4. Levey D. SITC 2022. Annual Meeting. Oral #470. 5. O'Malley, et al. Gynecol Oncol. 2021; 163: 274-280. 6. O'Malley et al, J Clin Oncol. 2022; 40(7): 762-771.

Deep Objective Responses



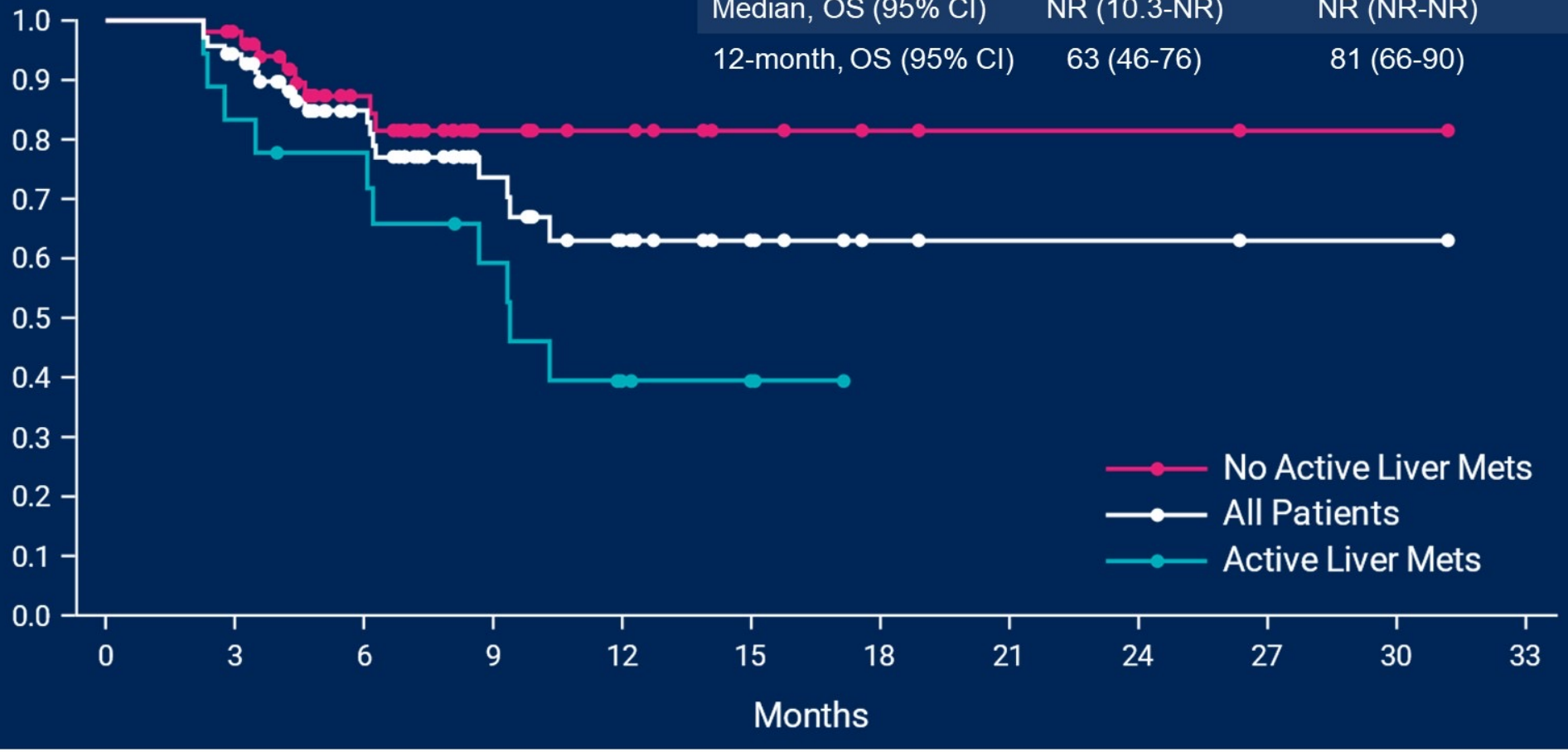
Efficacy	N=70
ORR*, % (95% CI)	23 (14-34)
BOR, n (%)	
CR	1 (1)
PR	15 (21)
SD	37 (53)
DCR (CR + PR + SD), % (95% CI)	76 (64-85)
Median, OS (95% CI)	NR (10.3-NR)
Median PFS, months (95% CI)	4.1 (2.8-5.5)
Median F/U, months (Min, Max)	7 (2, 31)

*Includes unconfirmed responses. ∞ Resected target lesions showed complete pathologic response. § Response by iRECIST.

Overall Survival

Efficacy evaluable population, N=70

	Overall	No Active Liver Mets	Active Liver Mets
Median, OS (95% CI)	NR (10.3-NR)	NR (NR-NR)	9.4 (6.1-NR)
12-month, OS (95% CI)	63 (46-76)	81 (66-90)	40 (17-62)

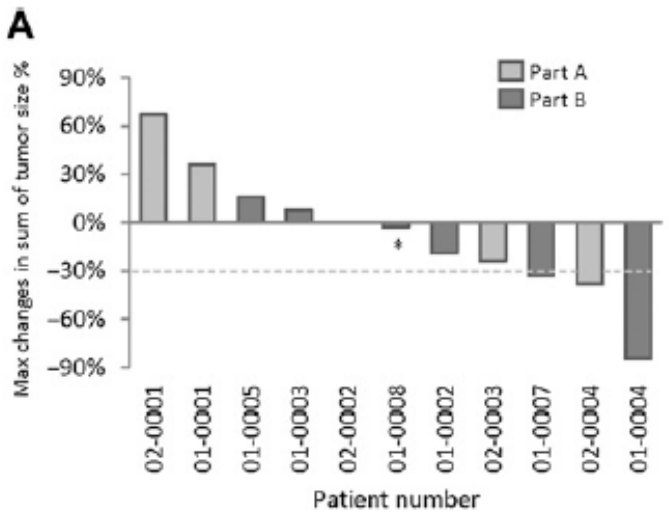


CANCER VACCINES FOR MSS MCRC

- Direct administration of cells or antigens to the patient, indirect antigen delivery or delivery of APCs trained ex vivo to recognize cancer cells
  - Peptide vaccines
  - Viral vectors
  - Dendritic cell vaccines
    - Can stimulate tumor antigen-specific cytotoxic T cells
    - DCs can elicit adaptive antitumor immunity

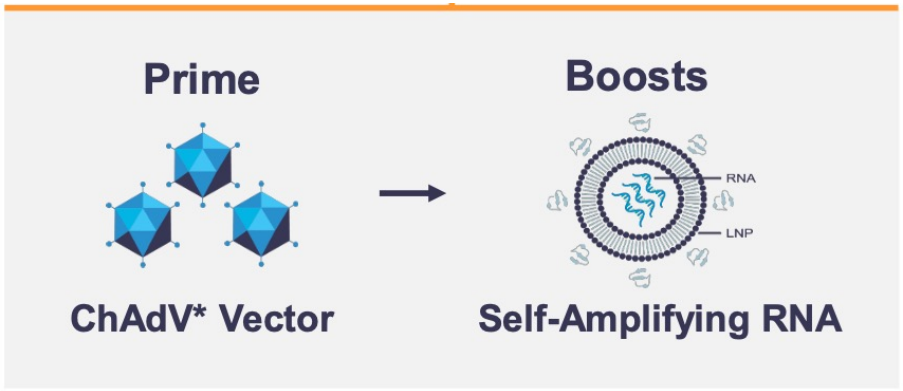
Example: Phase 1b OBERTO-101 study

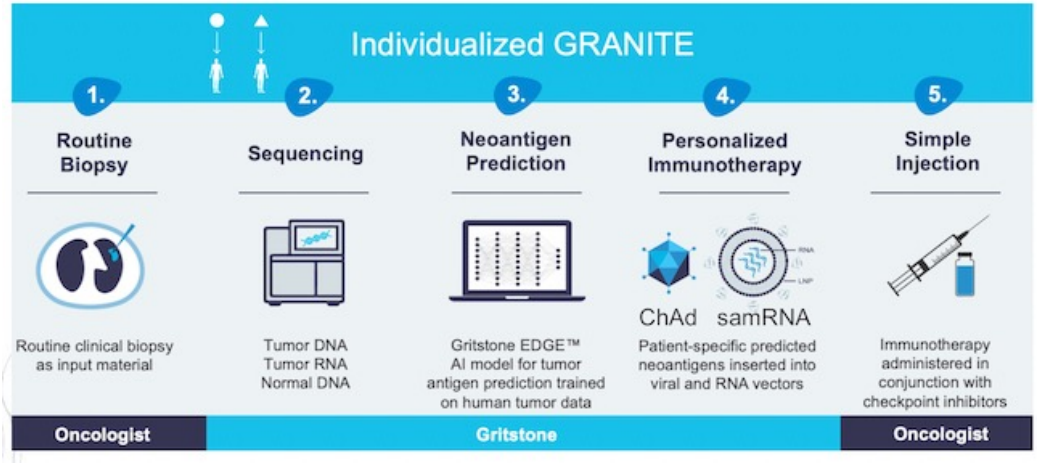
- PolyPEPI1018 vaccine + Montanide ISA51VG adjuvant + 5FU-based maintenance chemo



Hubbard JM et al. *Clin Cancer Res* 2022

Cancer Vaccines for MSS mCRC



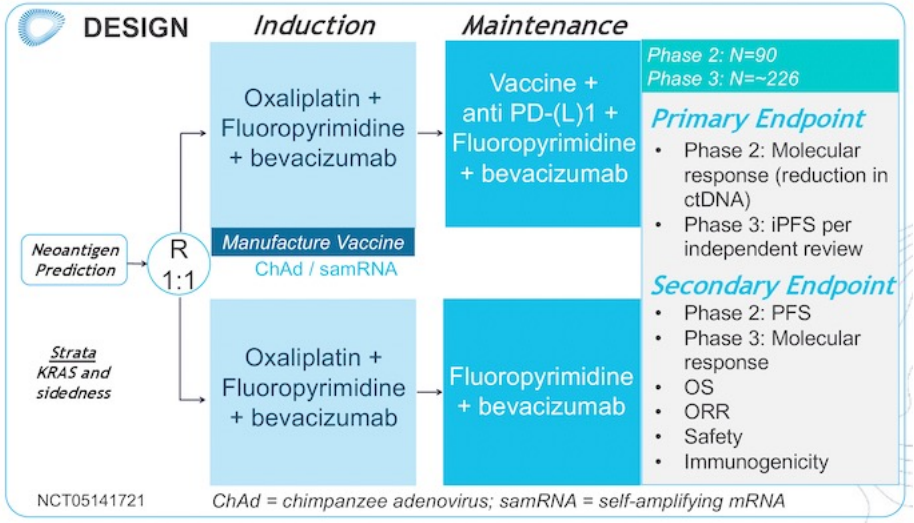


GRANITE/GO-004: Phase 1/2 study

	All N=22[1]	MSS-CRC N=11
Overall Response Rate per RECIST		
CR/PR	1 (CR, 4%)	0
Stable Disease (SD)	5 (23%)	4 (36%)
Progressive Disease (PD)	16 (73%)	7 (64%)[2]

1. 2 patients had no evaluable disease at time of vaccine availability
2. In patients treated beyond PD, 4 patients did not have confirmed progression at the next scan

Catenacci DVT, ESMO 2021



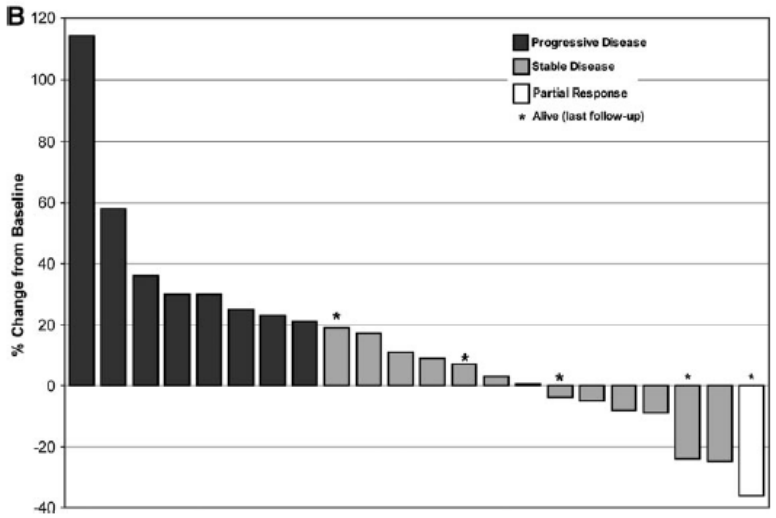
Hecht JR, 2022 ASCO, abstract TPS3635

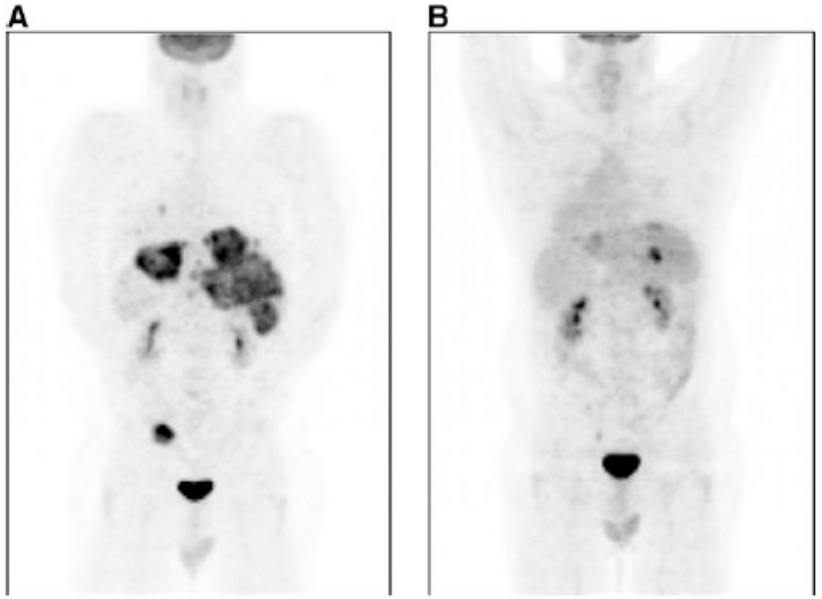
INTRATUMORAL THERAPIES FOR MSS MCRC

- Oncolytic viruses
  - Viral replication in transformed cells → cytolytic cell death → progeny virions infect adjacent tumor cells

Example: NV1020 (HSV virus)

- Ph I/II study of NV1020 in patients with liver-dominant mCRC
- 4 doses of NV1020 via HAIP infusion, then 2 or more chemo cycles



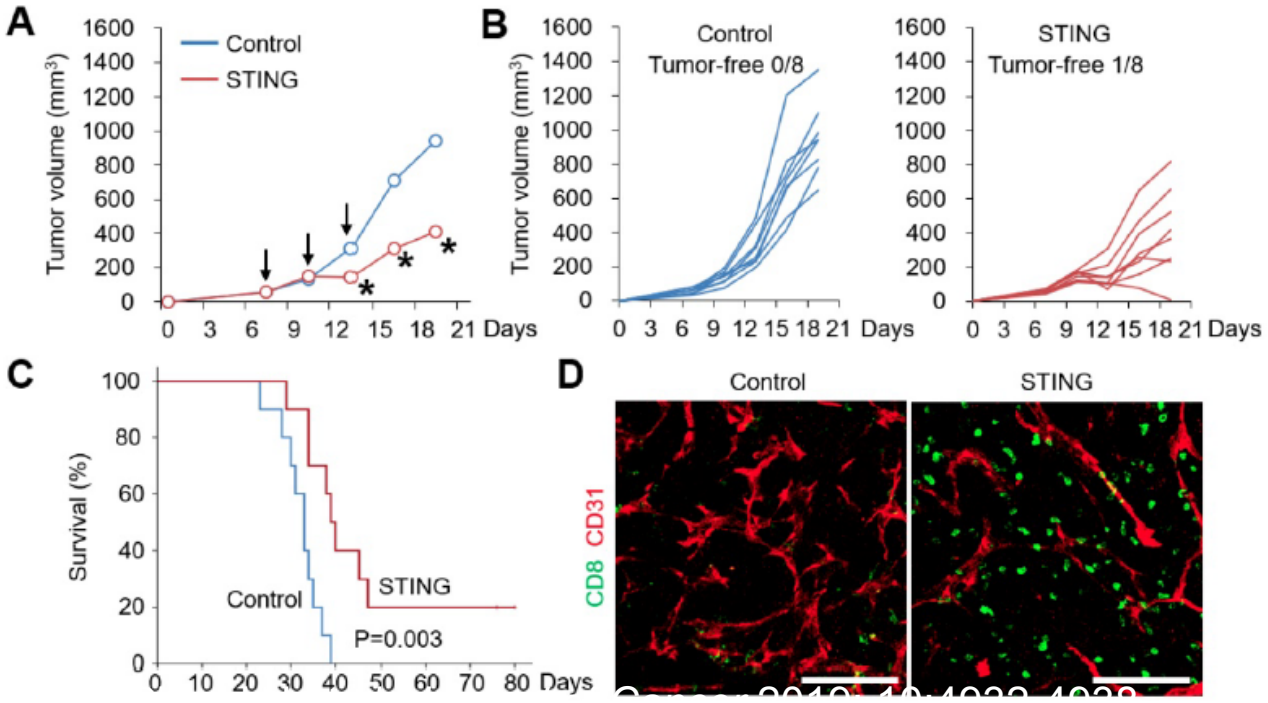


Geevarghese SK et al, *Hum Gene Ther* 2010; 21:1119-1128

Intratumoral Therapies for MSS mCRC

- STING (Stimulator of Interferon Genes)
  - Adaptor transmembrane protein in the ER
  - Innate immune sensor that detects tumor-derived DNA
  - STING pathway activation → type I interferon production → DC activation for cross-priming of T cells → adaptive antitumor immune response
- Phase I FIH trial: GSK3745417 +/- dostarlimab ongoing (NCT03843359)

Example: Intratumoral injection of synthetic STING agonists impair tumor growth in mice by increase in CD8+ T cell infiltration



Conclusions

- Failure of immune checkpoint blockade monotherapy in MSS mCRC well established
  - Reasons: lack of T-cell inflamed phenotype (inadequate T-cell infiltration and activation, T-cell suppression)
- Immunotherapy combinations are under investigation to try to overcome this failure
  - Need better understanding of primary and adaptive immune resistance, biomarkers for optimal patient selection, unique mechanisms to induce an antitumor immune response
  - Harnessing the immune system appropriately is difficult but holds promise for the future