Circulating-tumor DNA in GI Malignancies

ASCO 2024 Updates

Christopher Cann, MD

Assistant Professor

Gastrointestinal Cancer Program

Fox Chase Cancer Center

FOX CHASE CANCER CENTER

TEMPLE HEALTH

ctDNA: Background

- Circulating tumor DNA (ctDNA) is extracellular DNA derived from direct release, active secretion or necrosis/apoptosis of tumor cells
- Half life of 0.5-2.5 hours
- Detection rates can vary between tumor types
- Established that ctDNA levels are associated with tumor burden, stage, and response to therapy

Kasi JCO Prec Onc 2022, Lam Frontiers Onc 2022, Pathak ASCO 2024

ctDNA Detection Techniques

Nucleotide Coverage

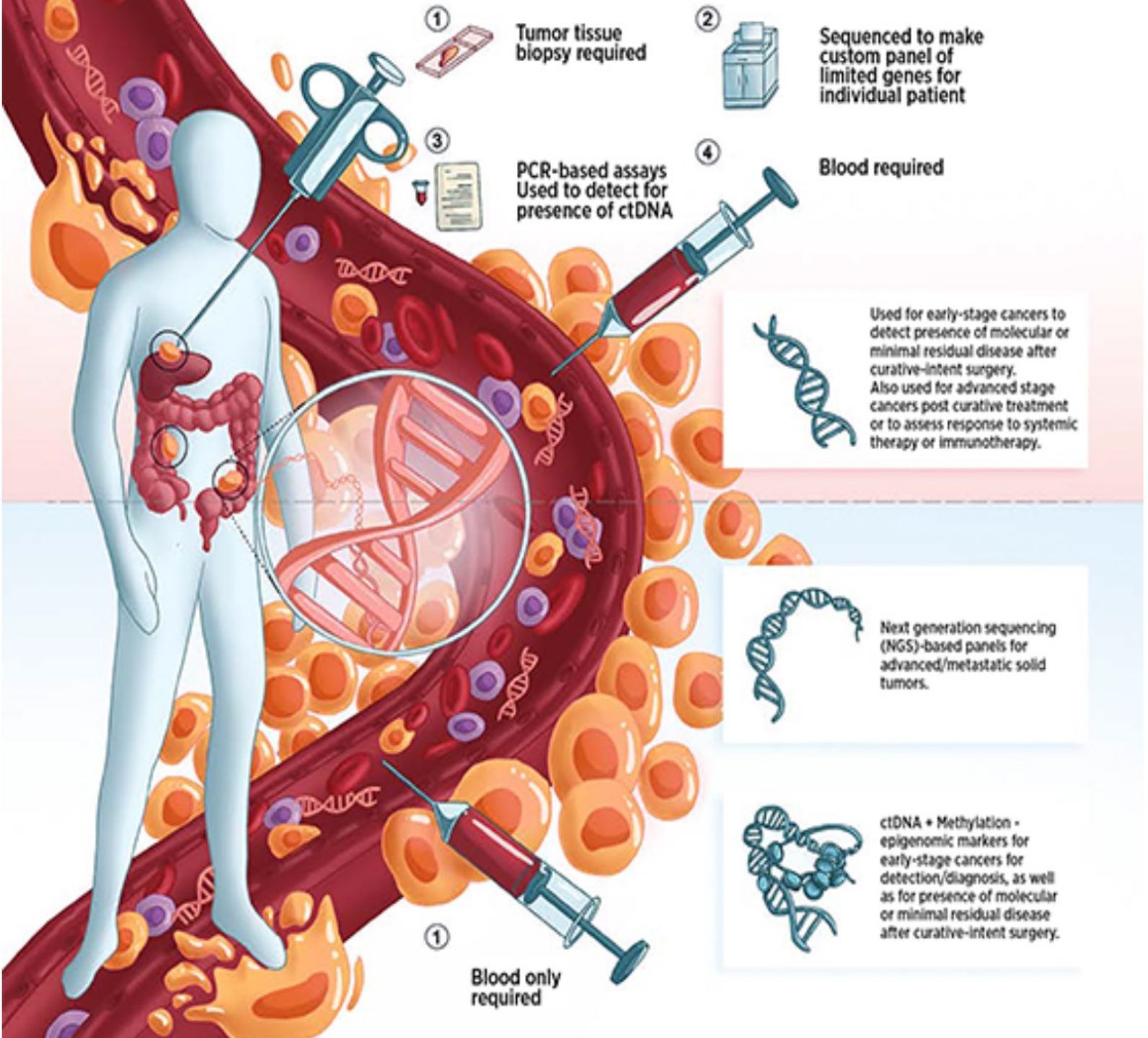
TABLE 1. ctDNA Detection Techniques

Technique	Description	Target	Examples (tumor-informed or tumor-naive)
dPCR	Separating DNA molecules into different reactions enabling high-throughput analysis	Single locus or multiple assays	dPCR[14] (either) ddPCR[19] (either) BEAMing[16] (either)
Multiplex PCR	PCR amplification of multiple targets before NGS analysis	Targeted sequencing	TAm-seq[20] (either) Enhanced Tam-seq (either) Safe-seq[21] (tumor-informed) Signatera[8] (tumor-informed) TARDIS[22]
Hybrid capture	Regions of interest are hybridized to target-specific biotinylated probes and captured for NGS analysis	Targeted sequencing	CAPP-seq[23] (either) TEC-seq[17] (tumor-informed) Guardant360[24,25] (tumor-naive) FoundationOne Liquid[26] (tumor-naive)
WGS	Plasma WGS of genomic alterations	Whole genome	PARE[27] (tumor-naive)

Abbreviations: ctDNA, circulating tumor DNA; ddPCR, droplet digital polymerase chain reaction; dPCR, digital polymerase chain reaction; NGS, next-generation sequencing; WGS, whole-genome sequencing.

Detection Sensitivity

Kasi JCO Prec Onc 2022, Lam Frontiers Onc 2022, Pathak ASCO 2024



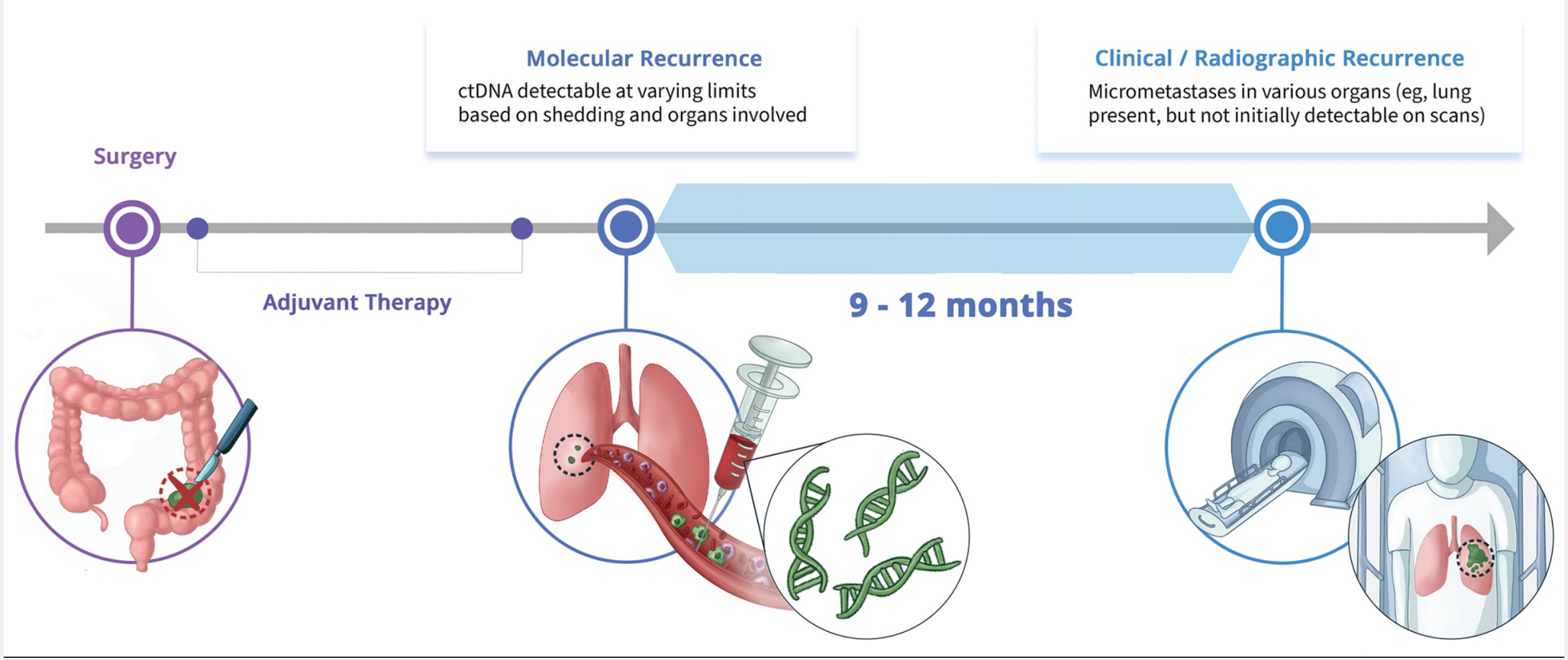
ctDNA Platforms

- **Tumor Informed**
  - Genomic
  - Prior knowledge of specific tumor alterations
  - Initial results: 4-6 weeks

- **Tumor Agnostic**
  - Broad, panel-based assays
  - Genomic and epigenetic
  - Real time tracking of novel mutational changes and cancer specific alterations simultaneously
  - Assays can be disease specific
  - Initial results: 7-10 days

Kasi ASCO Daily News 2022, Lam Frontiers Onc 2022, Pathak ASCO 2024

ctDNA Dynamics



Kasi ASCO Daily News 2023

2024 ASCO® ANNUAL MEETING

Circulating Tumor DNA Analysis Guiding Adjuvant Therapy in Stage II Colon Cancer

Overall Survival and Updated 5-Year Results from the Randomized DYNAMIC Trial

Jeanne Tie

Peter MacCallum Cancer Centre and Walter & Eliza Hall Institute of Medical Research, Melbourne, Australia

On behalf of the DYNAMIC Investigators

Yuxuan Wang, Serigne Lo, Kamel Lahouel, Joshua Cohen, Rachel Wong, Jeremy Shapiro, Samuel Harris, Adnan Khattak, Matthew Burge, Margaret Lee, Marion Harris, Sue-Anne McLachlan, Sumitra Ananda, Craig Underhill, Nickolas Papadopoulos, Cristian Tomasetti, Kenneth Kinzler, Bert Vogelstein, Peter Gibbs

DYNAMIC

Background

- Stage II colon cancer is a heterogenous disease, with 5-year overall survival ranging from 58.4% - 87% depending on risk stratification

- The ideal adjuvant treatment strategy remains unclear
  - Resection alone is curative in most patients
  - Fluoropyrimidine alone vs the addition of oxaliplatin in high-risk disease?

- Colon cancer is considered “high shedding” for ctDNA
  - Can ctDNA dynamics be used to guide adjuvant therapy in stage II disease?

Gunderson JCO 2010, Corcoran NEJM 2018, Baxter JCO 2021, Kasi JCO 2022, NCCN Colon Cancer 2024

DYNAMIC

DYNAMIC Study Design

Stage II Colon Cancer

- R0 resection
- ECOG 0 – 2
- Staging CT within 8 weeks
- Provision of adequate tumor tissue within 4 weeks post-op
- No synchronous colorectal cancer

Tumor-Informed ctDNA Analysis
(Targeted CRC panel)

(N = 455)

ctDNA-Guided Management

- **ctDNA-Positive → Adjuvant Chemo**
  (oxaliplatin-based or single agent FP)
- **ctDNA-Negative → Observation**

ctDNA-Positive = Positive result at week 4 and/or 7

Standard Management

Adjuvant treatment decisions based on conventional clinico-pathologic criteria

Follow-Up

Until recurrence

- CEA: 3-monthly for 24M, then 6-monthly to 5 years
- CT C/A/P: 6-monthly for 24M, then at 36M

Post-recurrence

- Survival: 6-monthly to 5 years

Endpoints	
Primary: RFS at 2 years (non-inferiority margin 8.5%)	**Secondary:** RFS by ctDNA status, EoT ctDNA clearance
Key secondary: Proportion receiving adjuvant chemo, OS	**Exploratory:** Post-op ctDNA levels

PRESENTED BY: Jeanne Tie, MBChB FRACP MD

DYNAMIC

Results: NEJM 2022

Treatment delivery: ctDNA-guided approach significantly reduced chemotherapy use

Treatment Information	ctDNA-Guided N = 294	Standard Management N = 147	P-value
Adjuvant Chemotherapy received, n	45 **(15%)**	41 **(28%)**	0.0017
Chemotherapy regimen, n			
Oxaliplatin-based doublet	28/45 **(62%)**	4/41 **(10%)**	
Single agent FP	17/45 **(38%)**	37/41 **(90%)**	<.0001
Time from surgery to commencing chemotherapy, median (IQR), days	83 (76, 89)	53 (49, 61)	<.0001
Treatment duration, median (IQR), weeks	24 (19, 24)	24 (21, 24)	0.9318

- Relapse Free Survival Rate at 2 years
  - Difference: +1.1%
  - Hazard Ratio: 0.96 (0.51, 1.82)

- ctDNA guided approach **non-inferior** to standard management

DYNAMIC

Results

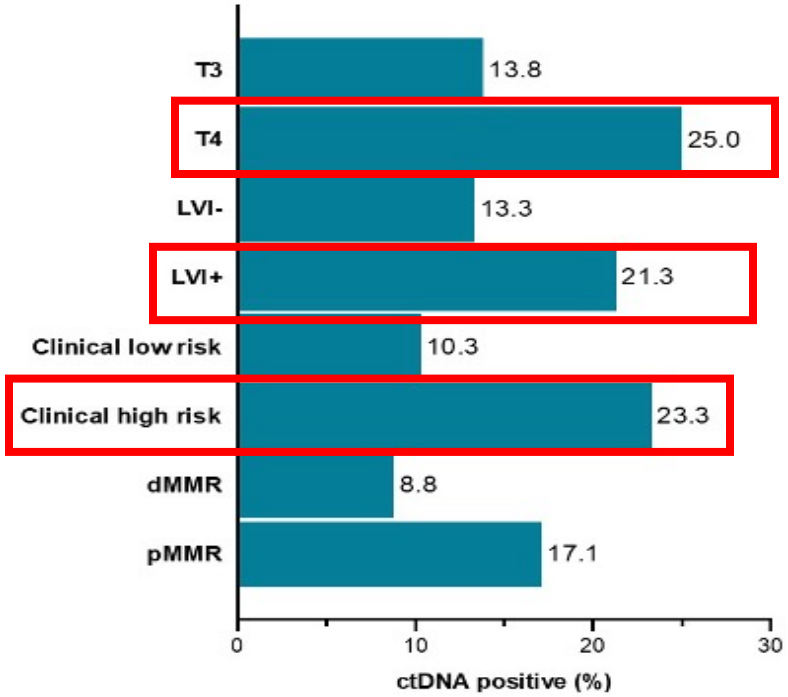
Baseline Characteristics

Characteristics	ctDNA-Guided Management N = 294, N (%)	Standard Management N = 147, N (%)
Age, median (range), yrs	65 (30 , 94)	62 (28 , 84)
Sex, Male	154 (52)	81 (55)
ECOG, 0	226 (77)	124 (84)
Tumor stage, T3	250 (85)	127 (86)
Lymph node yield, < 12	13 (4)	7 (5)
Lymphovascular invasion, present	82 (28)	38 (26)
MMR, deficient	59 (20)	27 (18)
Clinical risk group, high*	116 (40)	60 (41)

*High clinical risk = proficient MMR + ≥1 high-risk feature (T4, poor tumor differentiation, <12 lymph node yield, LVI, tumor perforation and/or bowel obstruction)

Post-op ctDNA Detection

(ctDNA-Guided Arm)



PRESENTED BY: Jeanne Tie, MBChB FRACP MD

DYNAMIC

Results

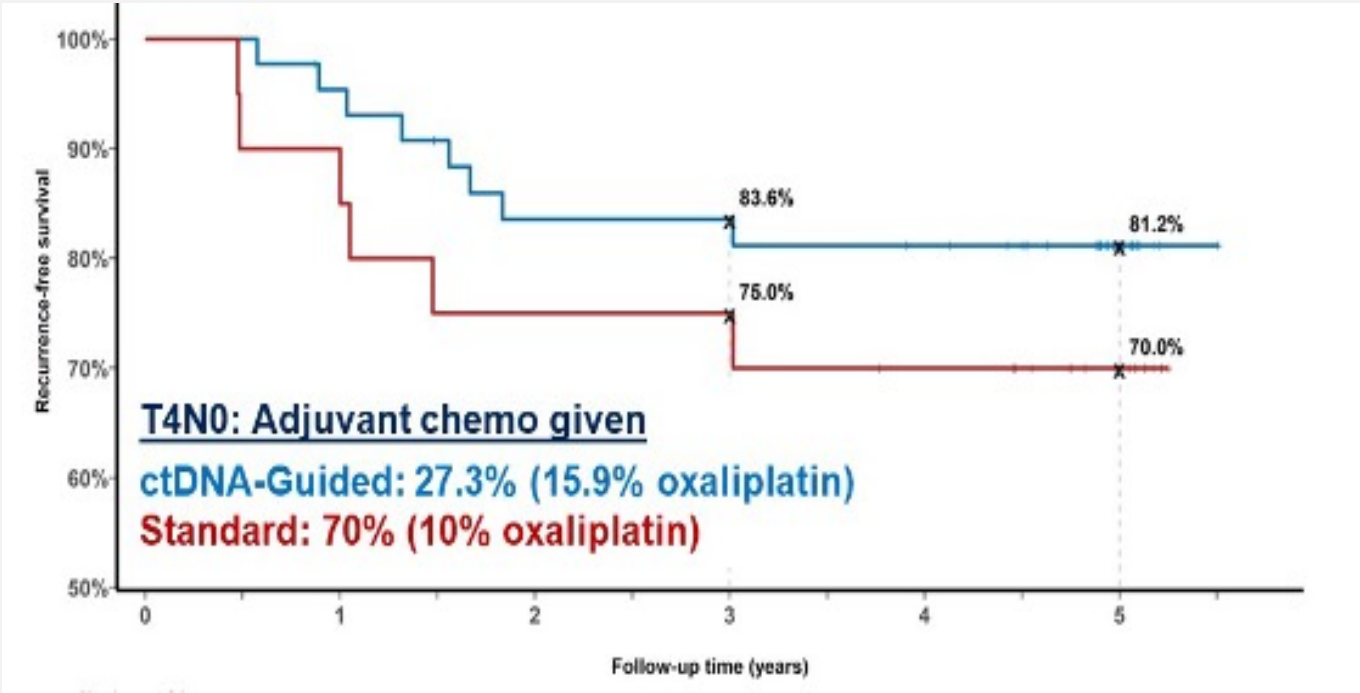
5 Year Relapse Free Survival Rates

Overall (HR 1.01; CI: 0.56, 1.18)

- ctDNA Guided: 88.3%
- Standard of care: 87.2%

T4N0 Disease (HR 1.79; CI: 0.62, 5.15)

- ctDNA Guided: 81.2%
  - 27.3% given chemotherapy
- Standard of care: 70%
  - 70% given chemotherapy



Tie ASCO 2024

DYNAMIC

Results

5-Year Overall Survival Rate

5-Year OS Rate, %		CRC deaths, N	Non-CRC deaths, N
ctDNA (N = 294)	93.8	7	10
SoC (N = 147)	93.3	4	5
HR (95% CI): 1.05 (0.47, 2.37), p = 0.887			

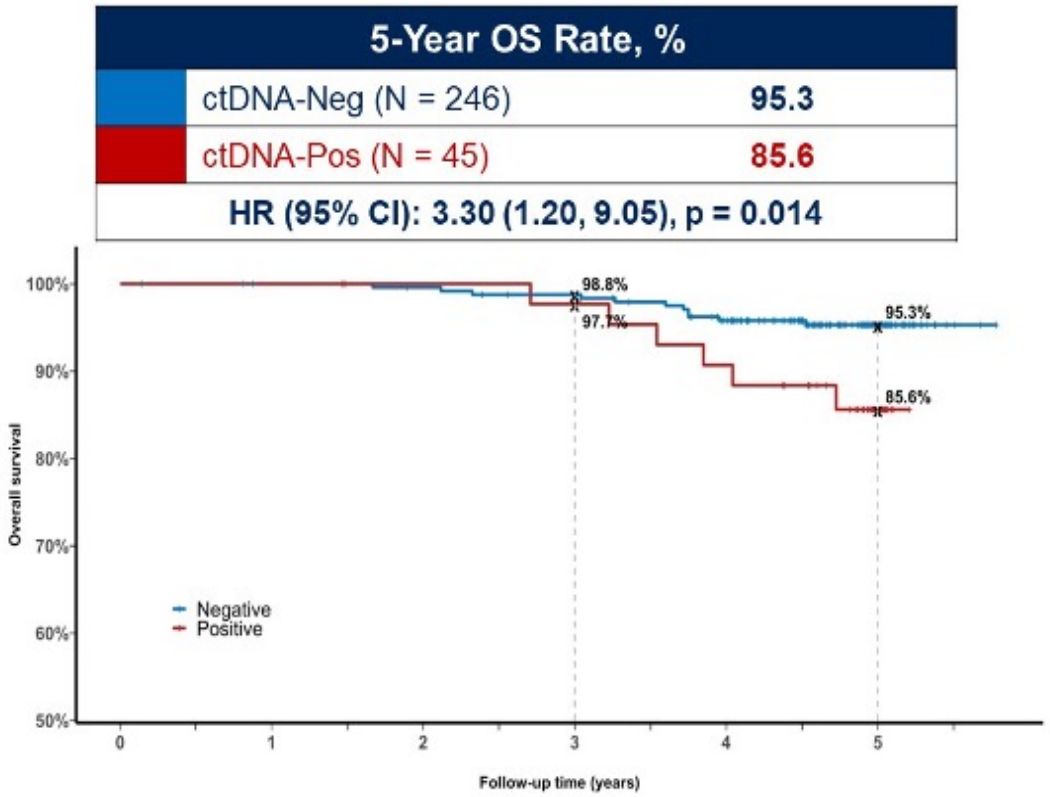
5-Year Disease-Specific Survival Rate, %	
ctDNA (N = 294)	97.9
SoC (N = 147)	97.2
HR (95% CI): 1.19 (0.35, 4.09), p = 0.792	

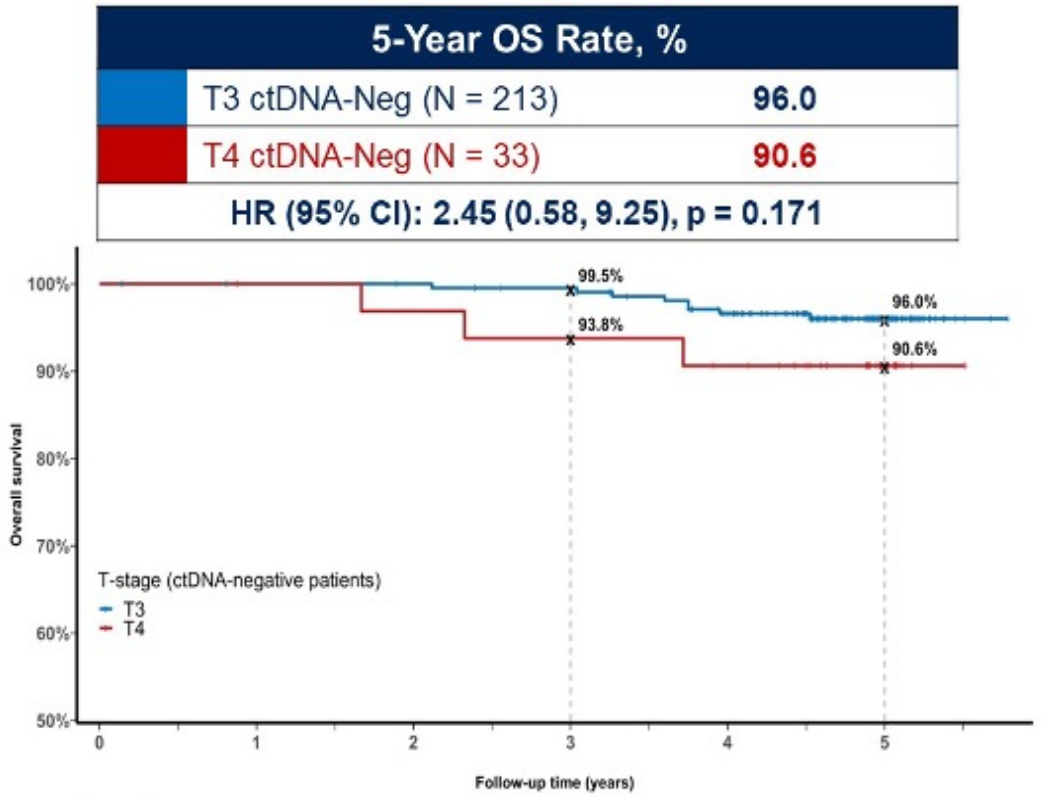
Tie ASCO 2024

DYNAMIC

Results

Overall Survival ctDNA Guided Arm





Tie ASCO 2024

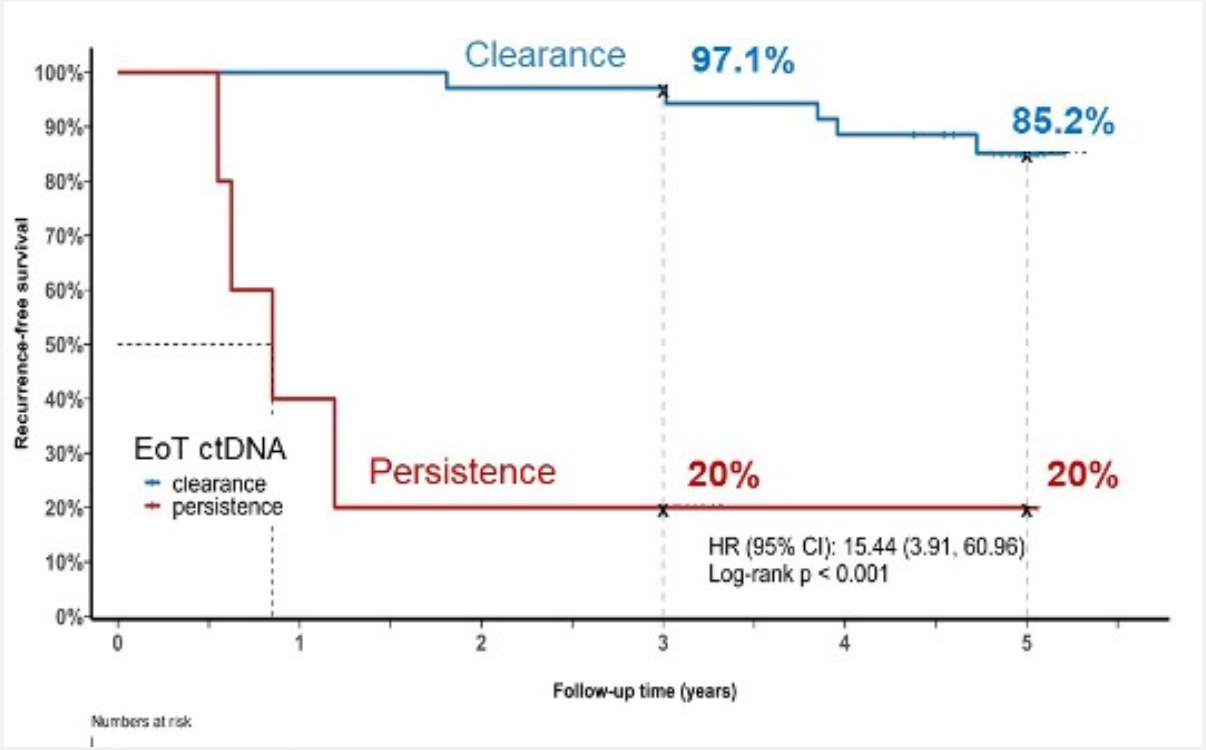
DYNAMIC

Results

End of Treatment ctDNA Clearance

- **Of those ctDNA+ after surgical resection**
  - 92.3% had clearance of ctDNA with the use of oxaliplatin
  - 78.6% had clearance of ctDNA with use of fluoropyrimidine monotherapy

- **At the end of adjuvant treatment**
  - 8.6% of those who were *ctDNA -* developed recurrence
  - 80% of those who were *ctDNA +* developed recurrence



DYNAMIC

Conclusions

- In stage II colon cancer, ctDNA guidance can allow for reduction in chemotherapy exposure without impacting survival outcomes
  - Including those with high-risk disease (ex T4)
- The use of adjuvant therapy results in ctDNA clearance in the majority of patients, which leads to improvement in relapse free survival
- Will these results change the treatment paradigm of stage II colon cancer?

2024 ASCO ANNUAL MEETING

The potential role of serial circulating tumor DNA (ctDNA) testing after upfront surgery to guide adjuvant chemotherapy for early-stage pancreatic cancer: The AGITG DYNAMIC-Pancreas Trial

Belinda Lee[1-4], Jeanne Tie[1-2,4], Yuxuan Wang[5], Joshua D Cohen[5], Jeremy D Shapiro[6,7], Rachel Wong[7-9], Morteza Aghmesheh[10-11], Andrew D Kiberu[12], Alessandra Francesconi[13], Matthew E Burge[14], Amitesh Roy[15], Lisa Dobbyn[5], Janine Ptak[5], Natalie Silliman[5], Nickolas Papadopoulos[5], Kenneth W Kinzler[5], Bert Vogelstein[5], Peter Gibbs[1,4,16]

[1]Walter & Eliza Hall Institute of Medical Research, Melbourne, Victoria, [2]Peter MacCallum Cancer Centre, Melbourne, Victoria, [3]Northern Health, Melbourne, Victoria, [4]University of Melbourne, Victoria, [5]Ludwig Centre for Cancer Genetics and Therapeutics, Johns Hopkins University School of Medicine, Baltimore, USA, [6]Cabrini Health, Melbourne, Victoria, [7]Monash University, Melbourne, Victoria, [8]Eastern Health, Melbourne, Victoria [9]Epworth Healthcare, Melbourne, Victoria [10] Illawarra Cancer Care Centre, Wollongong Hospital, New South Wales, [11]Prince of Wales Hospital, New South Wales, [12]Fiona Stanley Hospital, Western Australia, [13]Sunshine Coast University Hospital, Queensland, [14]Royal Brisbane and Women's Hospital, Queensland, [15]Flinders Centre for Innovation in Cancer, Adelaide, South Australia, [16]Western Health, Melbourne, Australia

AGITG DYNAMIC-Pancreas

Background

- Pancreas cancer is a formidable disease, with a 5-year overall survival of 13%
- Minority of patients (14%) are considered localized at diagnosis and eligible for potentially curative resection
- Despite the use of adjuvant chemotherapy
  - 80% relapse within 2 years
  - 5-year OS remains <50%
- Can ctDNA be used to indicate the benefit of adjuvant therapy and risk of recurrence?

Conroy NEJM 2018, Seigel CA Cancer J Clin 2023, SEER 2024

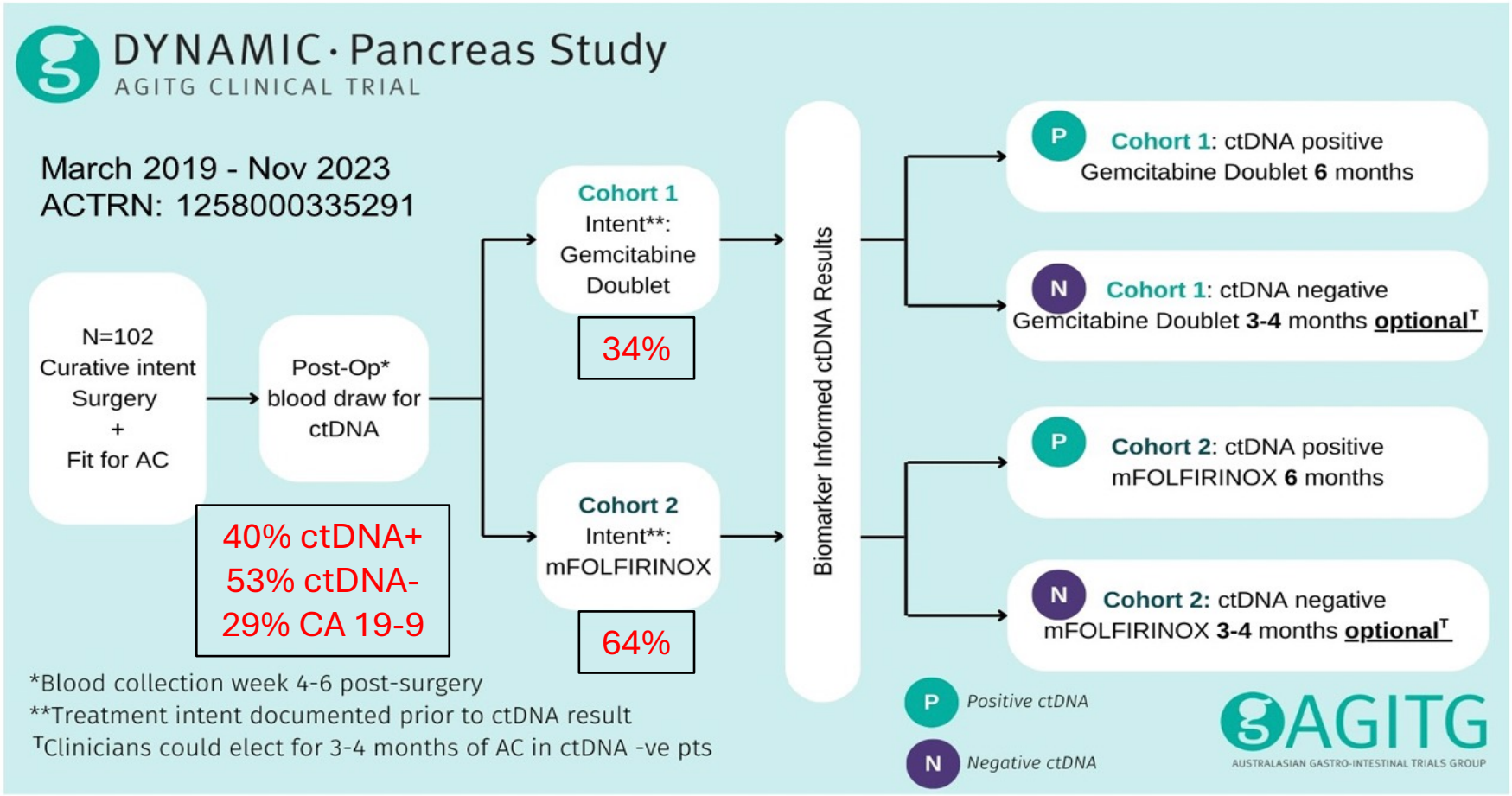
AGITG DYNAMIC-Pancreas

Methods

- Prospective multi-center biomarker driven treatment study
- Resectable pancreas adenocarcinoma
- Objective: determine feasibility of ctDNA-informed risk stratified approach in resectable pancreas cancer
- Endpoint: 2-year recurrence free survival

- **<u>Questions:</u>**
- Risk of recurrence in *ctDNA* – vs *ctDNA* + receiving adjuvant therapy?
- Can duration or regimen change based on ctDNA results?
- Implications of ctDNA dynamics?

AGITG DYNAMIC-Pancreas



Lee et al ASCO 2024

AGITG DYNAMIC-Pancreas

ctDNA status was not associated with known risk factors

Tumor assessed, N=98 (*KRAS* wildtype, N=4, subsequently excluded)	ctDNA Negative	ctDNA Positive	P value	Overall Cohort
ctDNA analysed, N=94	N=54	N=40		
Age: Median [range (years)]	68 [47-86]	66 [41-81]	0.55	68
Gender: Male / Female	30 / 24	16 /24	0.14	M: 50%
Location in Pancreas Head / Body & Tail	35 / 19	33 / 7	0.06	HOP: 72%
Pathological Tumor stage: T1 / T2-3	12 / 42	5 / 35	0.23	
Pathological Nodal stage: N0 / N+	17 / 37	10 / 30	0.49	N+ 71%
Resection margin status: R0 / R1	42 / 12	31 / 9	0.97	R0 77%
Lymphovascular invasion: Present / Absent	35 / 19	31 / 9	0.18	LVI+ 67%
Perineural invasion: Present / Absent	48 / 6	33 / 7	0.38	PNI+ 83%
Tumor grade: G1 / G2 / G3	4 / 33 / 17	2 / 23 /15	0.71	

AGITG DYNAMIC-Pancreas

Results

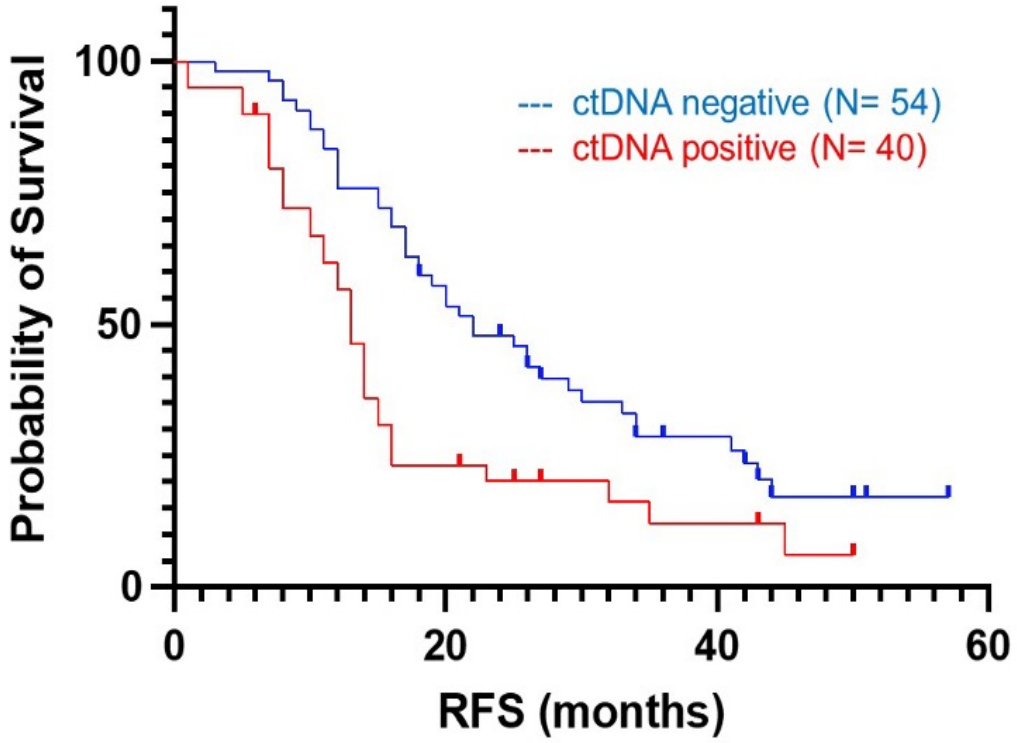
- **ctDNA negative cohort**
  - Planned: 55% of physicians planned to de-escalate
  - Actual: 67% received <4 months of therapy

- **ctDNA positive cohort**
  - Planned: 100% planned 6 months of adjuvant therapy
  - Actual: 50% received >6 months of therapy
    - Toxicity, relapse

Lee et al ASCO 2024

AGITG DYNAMIC-Pancreas

Recurrence Free Survival stratified by post operative ctDNA status (median FU = 36 months)

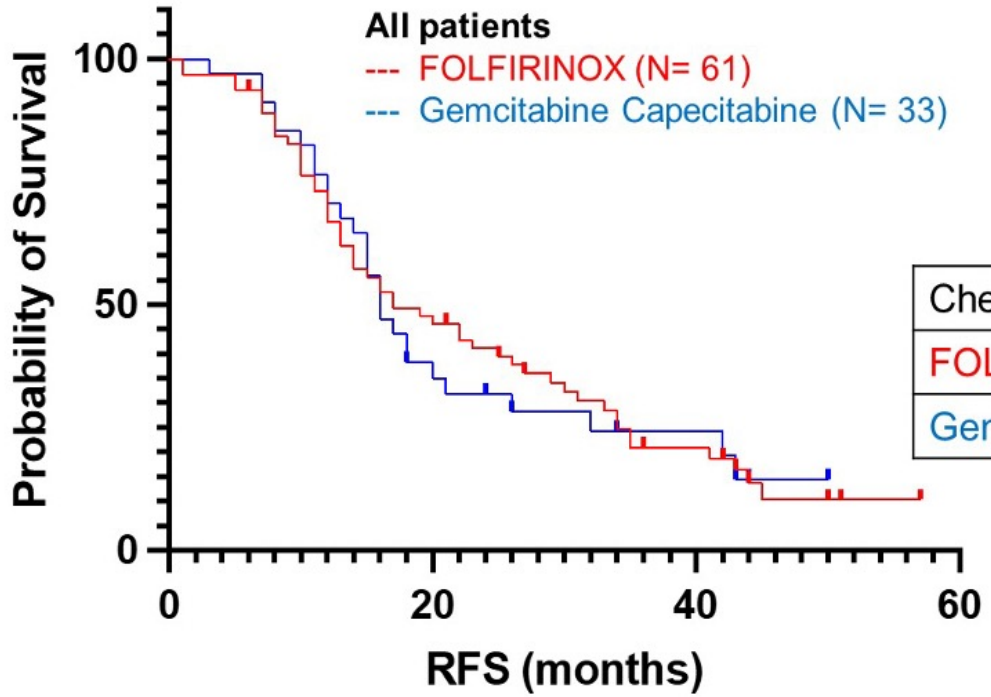


ctDNA status	RFS (months)	P value
ctDNA negative	22	P=0.003 HR 0.28
ctDNA positive	13	

Lee et al ASCO 2024

AGITG DYNAMIC-Pancreas

RFS in all patients stratified by adjuvant chemotherapy regimen administered

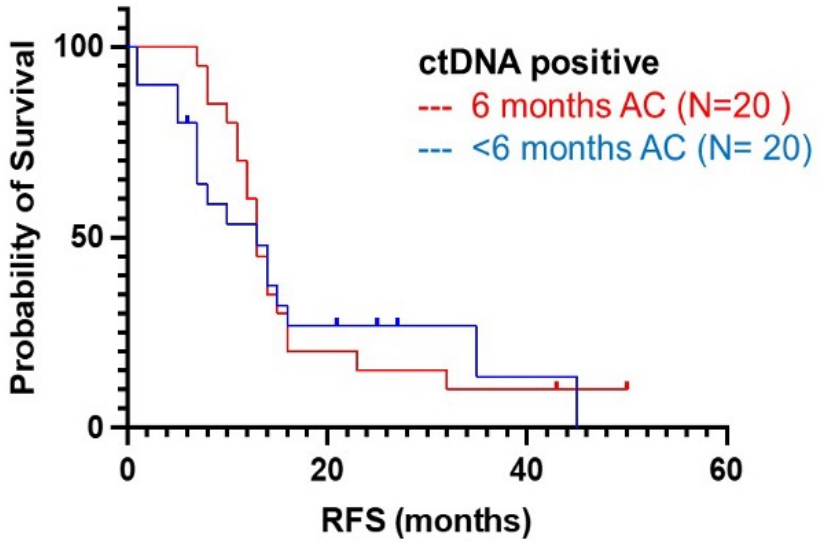


Chemotherapy	RFS (months)	P value
FOLFIRINOX	17	P=0.86 HR 0.96
Gemcitabine Capecitabine	16	

Lee et al ASCO 2024

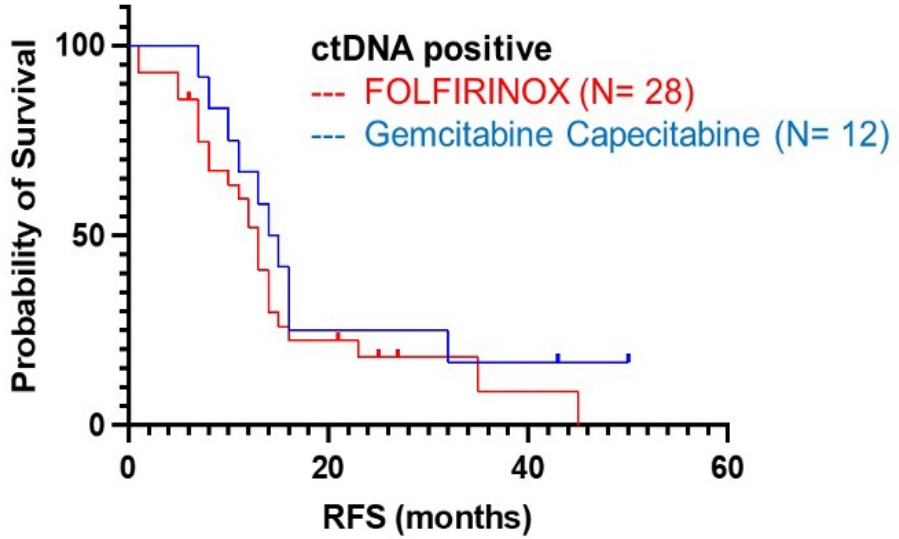
AGITG DYNAMIC-Pancreas

RFS in ctDNA+ve cohort stratified by AC duration



Duration of AC	RFS (months)	P value
6 months	13	P=0.84 HR 0.93
<6 months	13	

RFS stratified by AC Doublet vs Triplet therapy

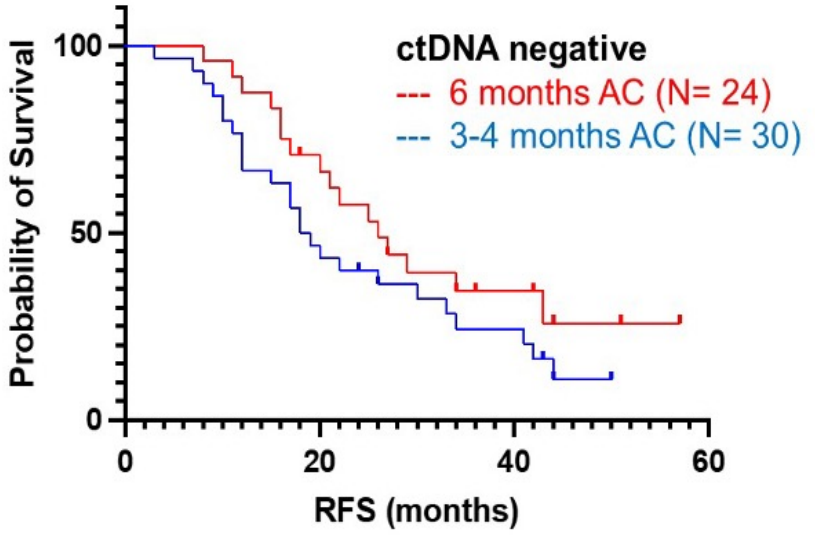


Chemotherapy	RFS (months)	P value
FOLFIRINOX	13	P=0.29 HR 0.69
Gemcitabine Capecitabine	14.5	

Lee et al ASCO 2024

AGITG DYNAMIC-Pancreas

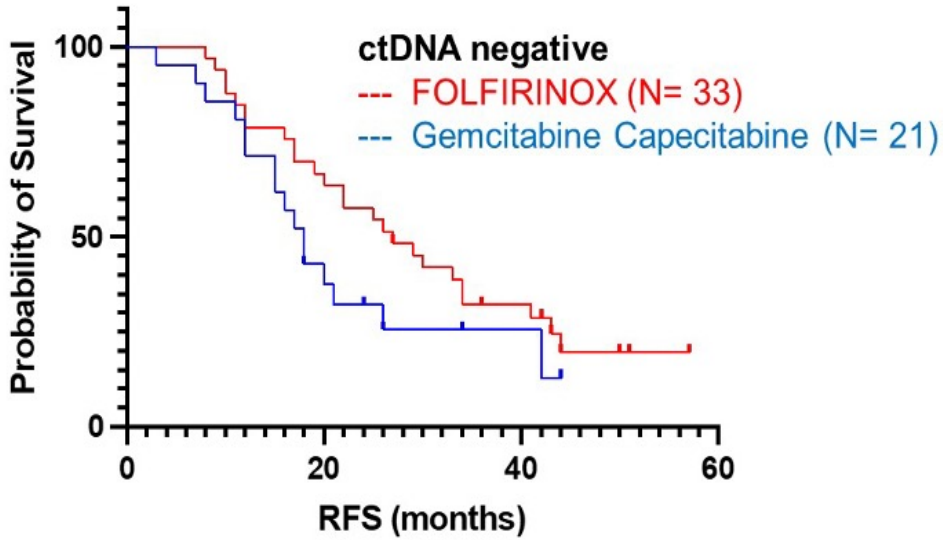
RFS in ctDNA-ve cohort stratified by AC duration



Duration of AC	RFS (months)	P value
6 months	26	P=0.159 HR 0.64
3-4months	18.5	

(shorter treatment duration at clinician discretion)

RFS stratified by AC Triplet vs Doublet therapy



Chemotherapy	RFS (months)	P value
FOLFIRINOX	27	P=0.09 HR 0.63
Gemcitabine Capecitabine	18	

Lee et al ASCO 2024

AGITG DYNAMIC-Pancreas

Conclusions

- In early pancreas cancer, ctDNA can serve as a prognostic marker, with *ctDNA+* indicating a shorter RFS
- However, regardless of ctDNA results, recurrence rate **remains very high**
- In *ctDNA –* patients, the use of 6 months of adjuvant therapy and the use of FOLFIRINOX had a **numerically higher RFS**, but were **NOT statistically significant**
- Awaiting additional results for
  - RFS correlation with ctDNA on treatment and end of treatment
  - Radiographic and ctDNA correlation
  - Survival data
- **ctDNA for prognostic purposes only, but it should not direct treatment decisions at this time**

2024 ASCO® ANNUAL MEETING

Time dependency for HPV ctDNA detection as a prognostic biomarker for anal cancer

Van K. Morris[1], Weihong Xiao[2], Emma B. Holliday[3], Kangyu Lin[1], Ryan W. Huey[1], Sonal S. Noticewala[3], Ethan B. Ludmir[3], Alisha H. Bent[1], Victoria Higbie[1], Eugene J. Koay[3], Albert C. Koong[3], Prajnan Das[3], Maura L. Gillison[2]

Departments of [1]Gastrointestinal Medical Oncology, [2]Thoracic/Head & Neck Medical Oncology, and [3]Gastrointestinal Radiation Oncology, The University of Texas- MD Anderson Cancer Center, Houston, TX

HPV ctDNA in Anal Cancer

Background

- Anal cancer is a relatively rare disease, however incidence has steadily risen over the past two decades
- Over 90% secondary to HPV infection
- Majority (77%) present with localized or regional disease that is eligible for curative intent chemoradiation therapy
- Clinical validity of HPV ctDNA has been shown in both oropharyngeal and anal cancers
- What is the optimal time point for HPV ctDNA evaluation to assess risk of recurrence?

Daling Cancer 2004, Chera Clin Cancer Res 2019, Bernard-Tessier Clin Cancer Res 2019, Deshmukh JNCI 2020, SEER 2024

HPV ctDNA in Anal Cancer

Methods

- Stage I-III anal cancer at MDACC → curative intent chemoradiation
  - 50-58 Gy to primary tumor and 43-47 Gy to elective nodes over 25-29 fractions
  - 5 FU 300mg/m2/day on days of radiation plus cisplatin
- All patients had HPV ddPCR assay evaluating 13 oncogenic HPV types
  - Baseline
  - End of treatment
  - Month 3, 6, 12, 18, 24
- ≥ 16 copies considered detection threshold

HPV ctDNA in Anal Cancer

Results

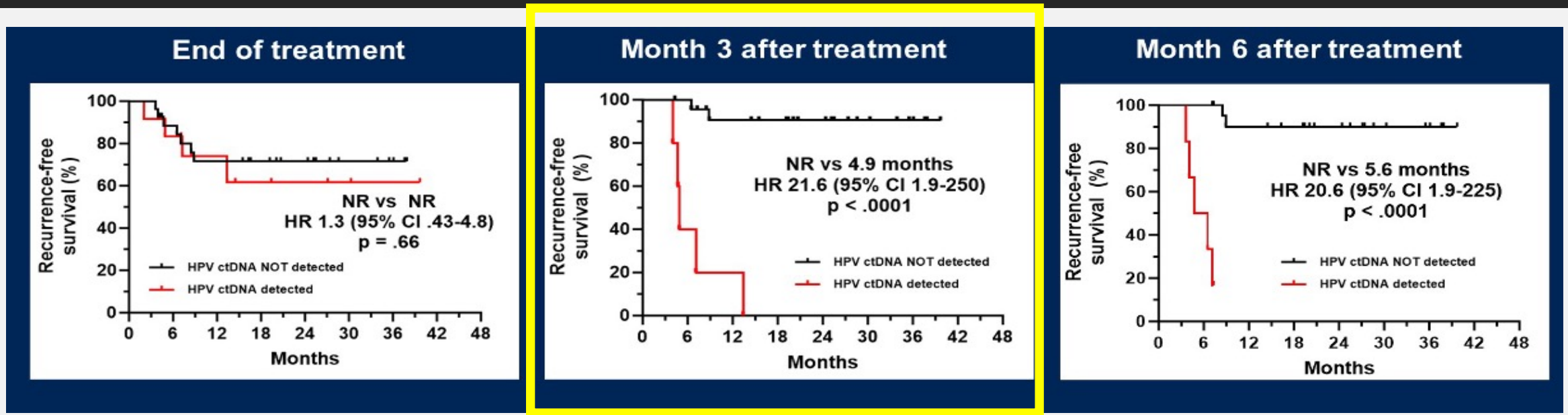
- 66 total patients
  - 33% *ctDNA –*
  - 67% *ctDNA+*
- **97.7%** of *ctDNA+* were HPV 16

- ***ctDNA –***
  - 82% T stage 1-2
  - 68% N0
  - 81% Stage I-II
- ***ctDNA+***
  - 55% T stage 3-4
  - 68% N+
  - 57% Stage III

Baseline HPV ctDNA status (N=66)			
	NOT DETECTED (< 16 copies/mL) N=22	DETECTED (≥ 16 copies/mL) N=44	P-value
Age (years, SD)	62.4 (11.2)	61.0 (9.2)	.682
Gender (%)			
Female	17 (77)	33 (75)	1
Male	5 (23)	11 (25)	
T stage (%)			.01
1-2	18 (82)	20 (45)	
3-4	4 (18)	24 (55)	
N stage (%)			.01
0	15 (68)	14 (32)	
1	7 (32)	30 (68)	
Clinical stage (%)			.009
1-2	17 (81)	19 (43)	
3	4 (19)	25 (57)	

Morris ASCO 2024

HPV ctDNA in Anal Cancer

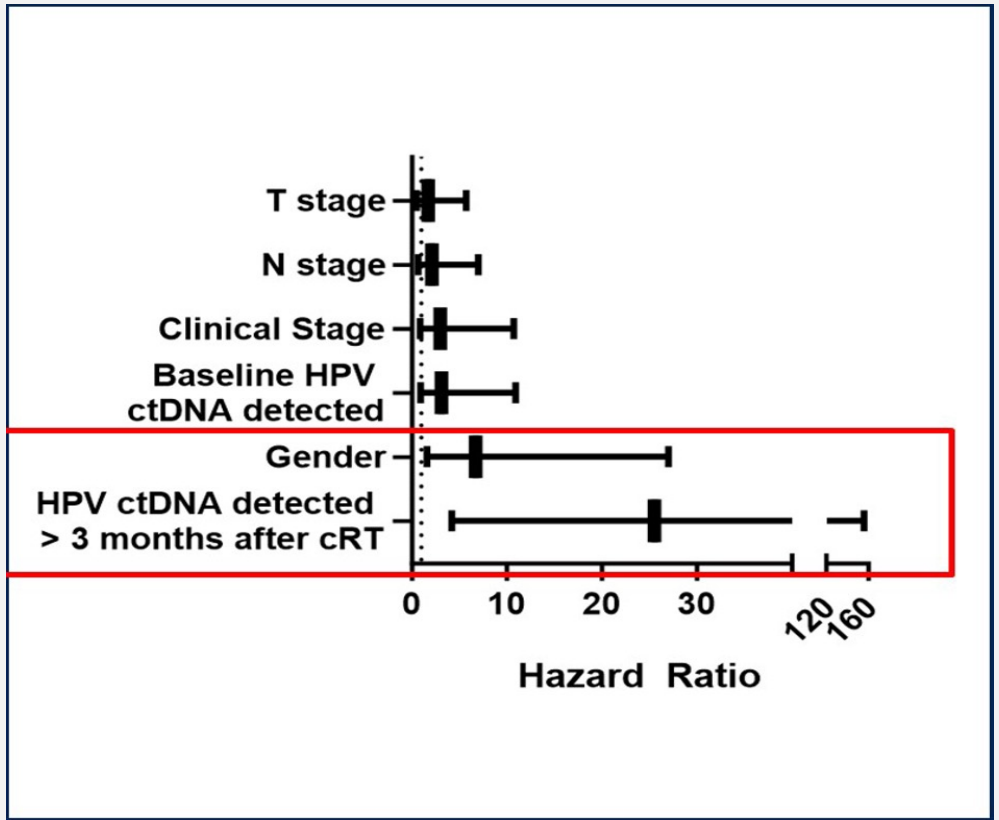


- ctDNA kinetics shows more positive ctDNA at end of treatment, but clearance by 3 months
- **3 months after treatment** appears to be the first prognostic time point
  - Sensitivity 80%, Specificity 96%, PPV 89%, NPV 93%
- This is in line with known anti-tumor effects of chemoradiation in this tumor type (up to 6 months)

Morris ASCO 2024

HPV ctDNA in Anal Cancer: Results

- **HPV ctDNA detection ≥ 3 months**: Best clinicopathologic factor for risk of recurrence in this study



HPV ctDNA in Anal Cancer

Conclusions

- HPV ctDNA detection via a ddPCR assay can be prognostic for recurrence in patients with anal cancer treated with chemoradiation
- 3 months after the end of treatment appears to be the first time for assessing recurrence risk
- As ctDNA positivity is associated with higher clinical stage, can this be successfully used in early stage/node negative disease?
- **ctDNA for prognostic purposes only, but it should not direct treatment decisions at this time**

Additional ctDNA Studies and Discussions

- Wullaert et al: “Circulating tumor cells and tumor DNA in patients with resectable colorectal liver metastases: The MIRACLE”
- Pathak: “Circulating tumor DNA in CRC: Current Practices and future Directions”
- Cohen: “Clinical Utility of ctDNA in Gastrointestinal Cancers: Is it Ready for Prime Time?”

Thoughts

- Standardization of ctDNA assays/commercial availability
- Tumor informed vs tumor agnostic assays
- Chemotherapy heterogeneity