

Cancer colorectal : biopsies tumorales ou biopsies liquides ?

Marc DENIS

Plateforme de Génétique Moléculaire des Cancers

CHU de Nantes

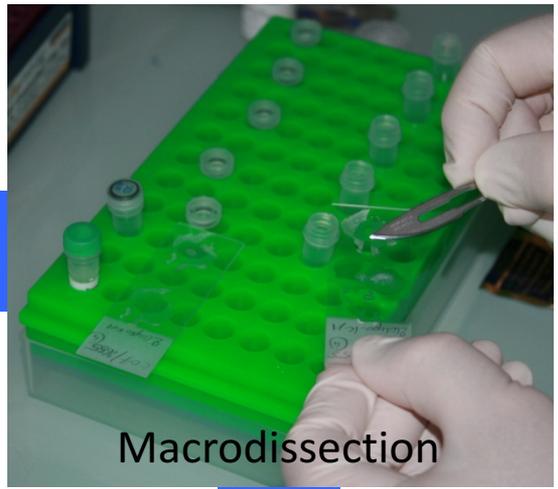


CCR - inhibiteurs d'EGFR

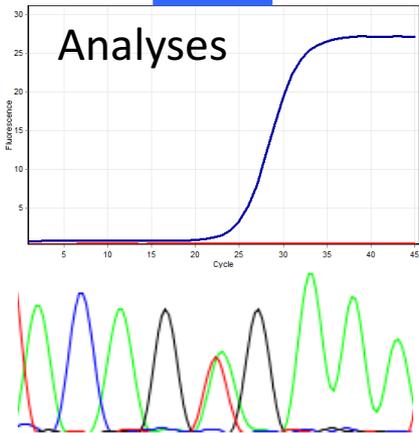
2 molécules disponibles :

- Cetuximab - ERBITUX®
- Panitumumab - VECTIBIX®

→ **testing RAS obligatoire**



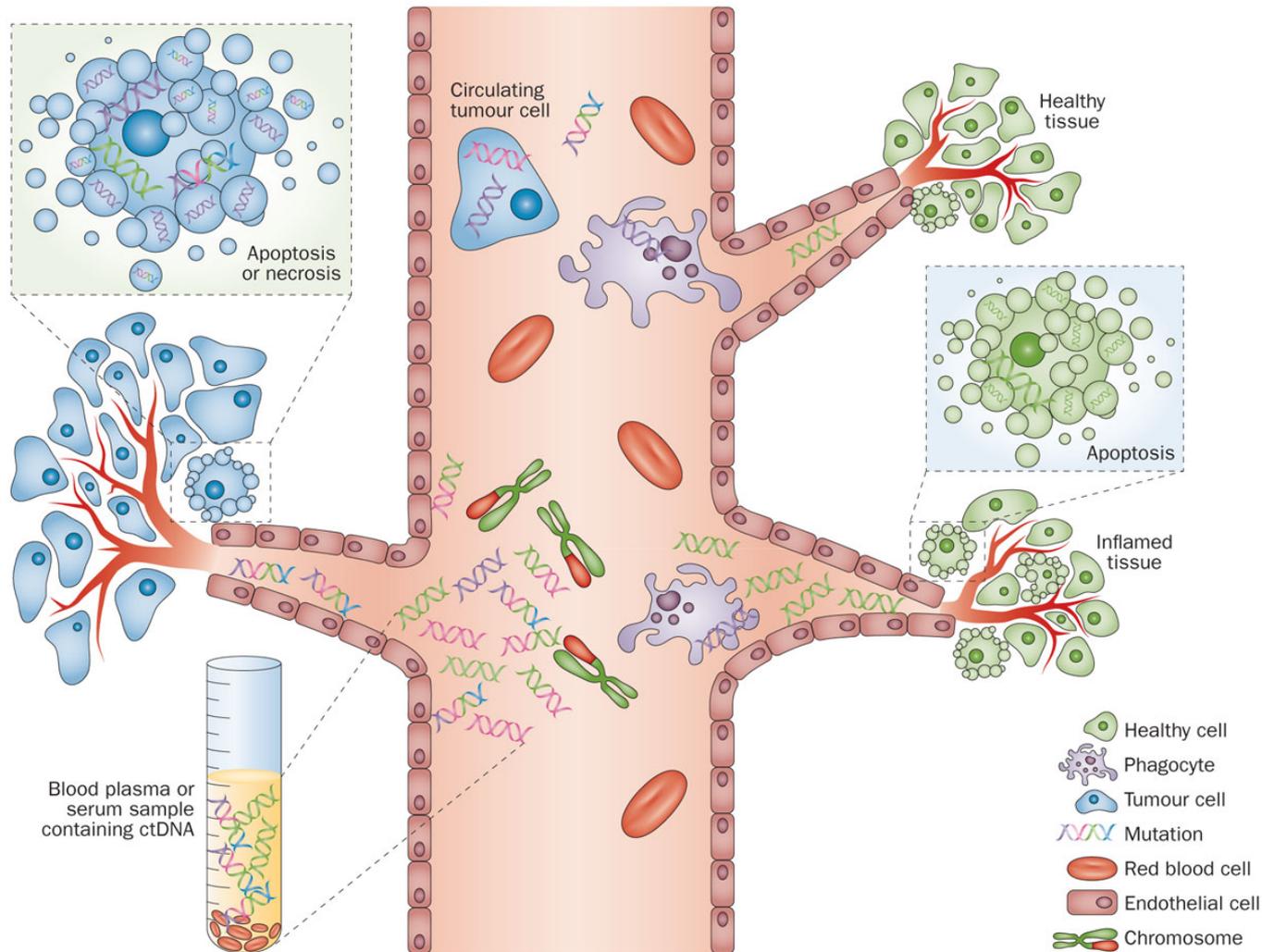
Report



Mais...

- ▣ Test tissulaire non réalisable
 - ❖ Pas de prélèvement
 - ❖ Bloc épuisé
- ▣ Test non contributif
 - ❖ Faible cellularité
 - ❖ Mauvaise qualité ADN
- ▣ Délai rendu résultat (TAT)
- ▣ Suivi en cours de traitement

ADN circulat – ADN tumoral circulat



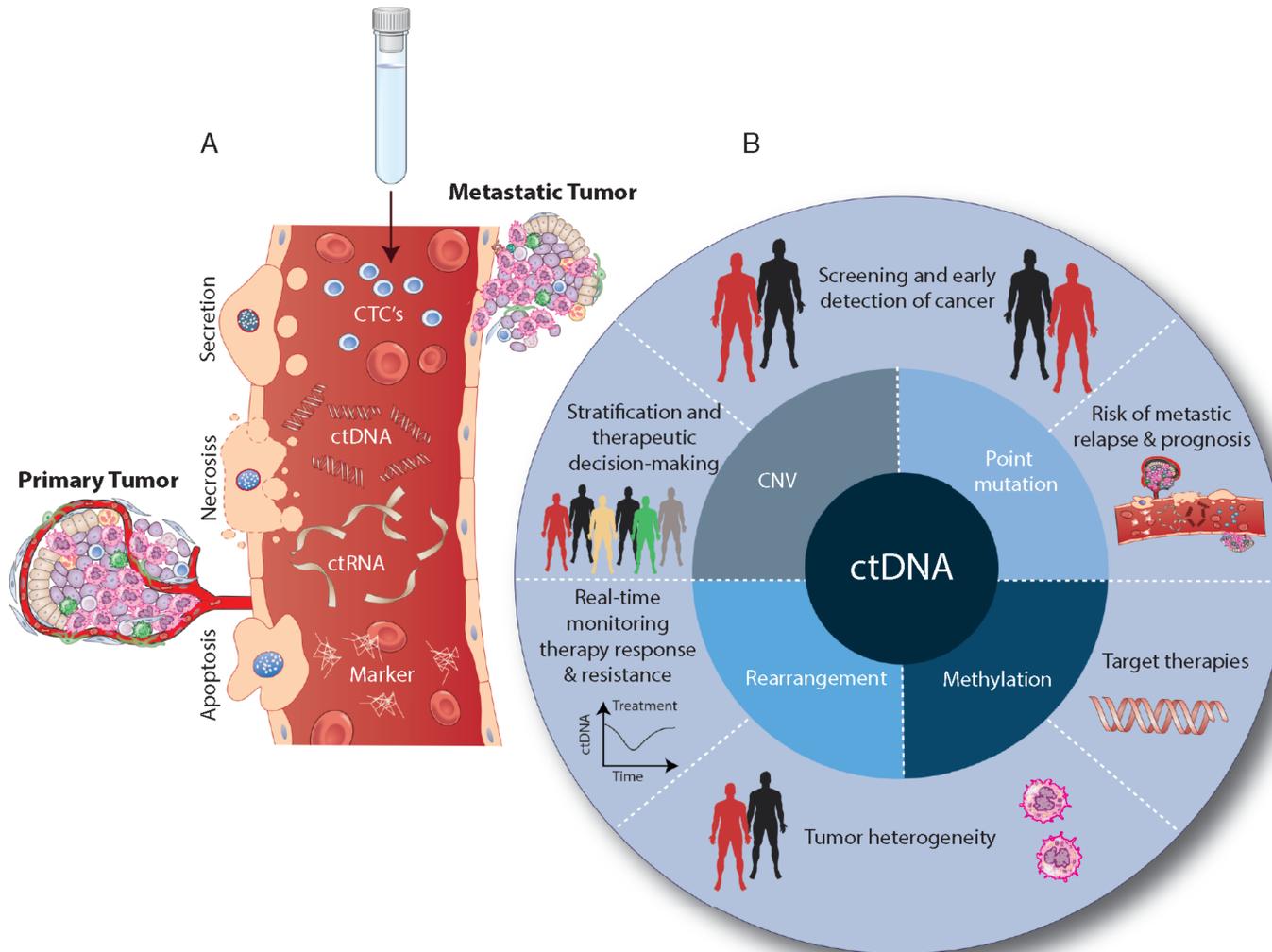
Une notion déjà ancienne...

Les acides nucléiques du plasma sanguin chez l'Homme,

par P. MANDEL et P. MÉTAIS.

Sujet	Sexe	Age	Affection	P phospho- protéine mg.	P ribonu- cléique mg.	P desoxyri- bonu- cléique mg.	P total acides nucléiques mg.
1	F	42	Normal	0	5,0	1,2	6,2
2	F	22	»	0	4,0	0,4	4,4
3	H	24	»	0	5,2	1,3	6,5
4	F	27	»	0	4,7	0,3	5,0
5	F	20	»	0	3,7	0,8	4,5
6	H	48	»	0	4,6	1,3	5,9
7	H	45	»	0	4,5	0,6	5,1
8	F	26	»	0	5,0	0,2	5,2
9	F	37	»	0	4,8	0,6	5,4
10	H	39	»	0	5,0	0,9	5,9
11	H	62	Insuffis. card.	0	3,8	0,7	4,5
12	H	62	»	0	3,8	0,45	4,25
13	H	42	»	0	5,1	0,9	6,0
14	F	33	Endocard. maligne	0	3,35	0,65	4,0
15	H	19	»	0	3,5	0,8	4,3
16	F	5	Goutte	0	5,6	0,4	6,0
17	F	5	Basedow	0	3,6	0,3	3,9
18	H	48	Diabète	0	3,6	0,4	4,0
19	H	61	»	0	3,5	0,4	3,9
20	H	48	Cirrhose	0	5,3	1,2	6,5
21	F	52	Ictère	0	3,6	0,4	4,0
22	H	48	Goutte	0	3,5	1,0	4,5
23	H	33	»	0	2,66	0,8	3,46
24	H	33	Goutte	0	5,5	0,5	6,0
25	H	»	»	0	4,75	0,75	5,5
26	H	26	Néphrite	0	3,75	0,7	4,45
27	H	37	Tuberculose	0	3,5	0,45	3,95
28	F	23	Grossesse 7 ^e m.	0	7,65	1,35	9,0
29	H	»	»	0	7,25	1,00	8,25

...et de nombreuses applications



Quelles applications ?

- ▣ Choix d'une thérapie ciblée
- ▣ Détection maladie résiduelle
- ▣ Indicateur efficacité traitement

Concordance status RAS tissuS/ADNtc

Reference	Detection method	Number of patients	Mutations in tumor tissues	Mutations in plasma	Sensibility Specificity	Concordance level between tissue and plasma
Spindler et al. 2012 [29]	qPCR	N = 95	<i>KRAS</i> 41/95 (43%)	<i>KRAS</i> 32/95 (34%)	78% 100%	
Taly et al. 2013 [10]	ddPCR	N = 50	<i>KRAS</i> 19/50 (38%)	<i>KRAS</i> 14/50 (28%)	74% 94%	89% Duplex dPCR 78% Multiplex dPCR
Bettegowda et al. 2014 [32]	ddPCR	N = 206	<i>KRAS</i> 78/206 (38%)	<i>KRAS</i> 69/206 (33%)	87.2% 99.2%	95%
Thierry et al. 2014 [11]	Intplex qPCR	N = 95	<i>KRAS</i> 39/95 (41%) <i>BRAF</i> 5/95 (100%)	<i>KRAS</i> 37/95 (39%) <i>BRAF</i> 5/95 (100%)	92% 98% 100%	96% 100%
Spindler et al. 2015 [31]	qPCR	N = 140	<i>KRAS</i> 48/140 (34%)	<i>KRAS</i> 30/140 (23%)		
Bachet et al. 2017 [30]	NGS-BEPER and DNA methylation	N = 406 N = 121	<i>RAS</i> 222/406 (55%) <i>KRAS</i> exon 2 53/121 (44%)	<i>RAS</i> 172/406 (42%) <i>KRAS</i> exon 2 71/121 (59%)	92% 94% 85% 62%	93% 72%
Thierry et al. 2017 [33]	Intplex qPCR	N = 34 N = 97	<i>NRAS</i> exons 2/3 3/34 (9%) <i>BRAF</i> 7/97 (7%)	<i>NRAS</i> exons 2/3 4/34 (12%) <i>BRAF</i> 14/97 (14%)	67% 94% 57% 89%	92% 87%

Etude RASANC

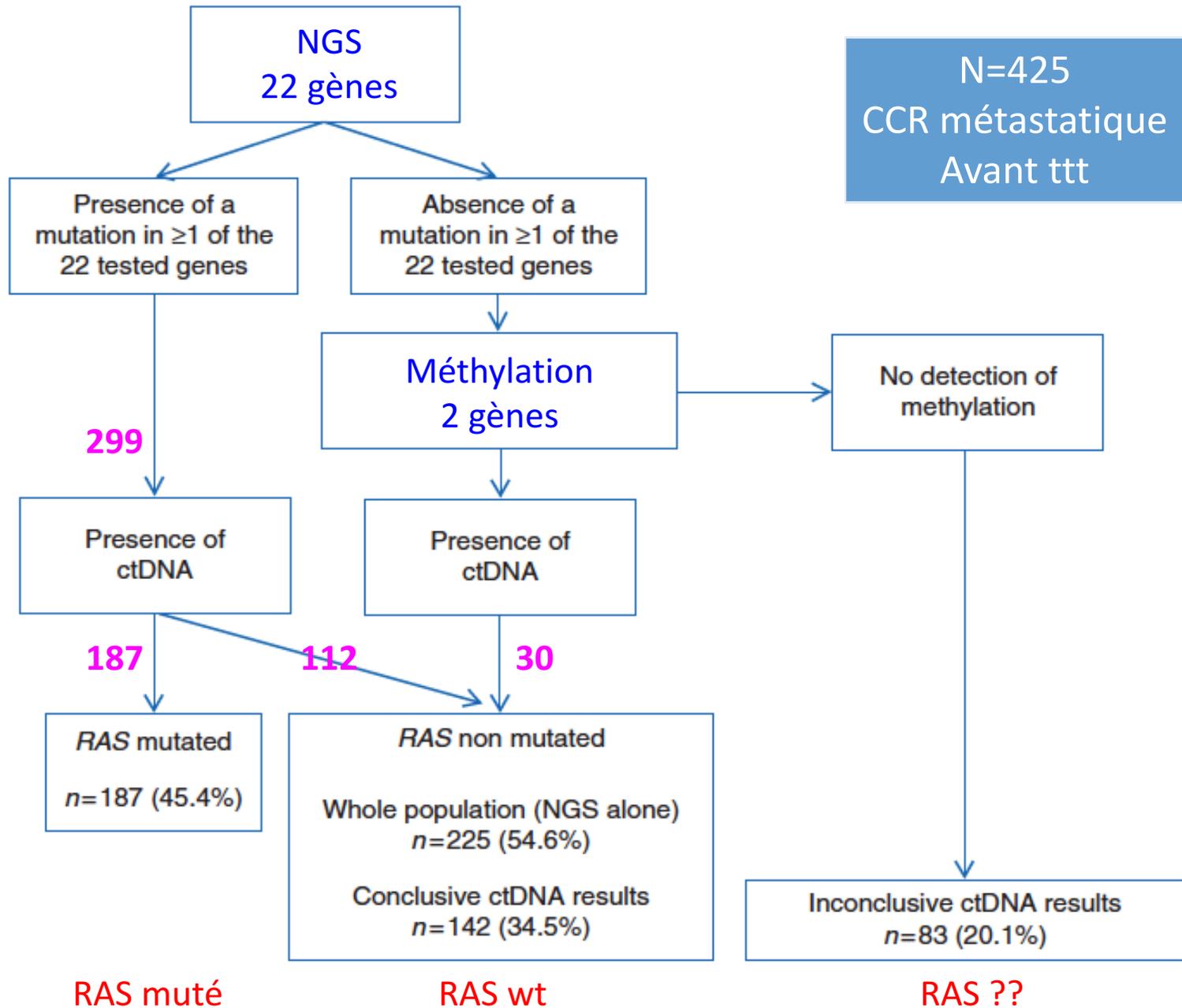


Annals of Oncology 29: 1211–1219, 2018
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ORIGINAL ARTICLE

RAS mutation analysis in circulating tumor DNA from patients with metastatic colorectal cancer: the AGEO RASANC prospective multicenter study

J. B. Bachet^{1,2,3,4}, O. Bouché^{4,5}, J. Taieb^{4,6}, O. Dubreuil^{3,4}, M. L. Garcia^{4,7}, A. Meurisse⁸, C. Normand², J. M. Gornet^{4,9}, P. Artru^{4,10}, S. Louafi^{4,11,12}, F. Bonnetain⁸, A. Thiot-Bidault^{4,13}, I. Baumgaertner^{4,14}, R. Coriat^{4,15}, D. Tougeron^{4,16}, T. Lecomte^{4,17}, F. Mary^{4,18}, T. Aparicio^{4,9,18}, L. Marthey^{4,19}, V. Taly², H. Blons^{2,20}, D. Vernerey⁸ & P. Laurent-Puig^{2*}



Facteurs associés à la détection d'ADNtc

Parameters		Patients <i>n</i>	Patients with inconclusive ctDNA results <i>n</i>	Odds ratio	Hazard ratio 95% CI	<i>p</i> -value
Primary tumor resection	No	197	20	1		
	Yes	216	63	3.67	2.12 to 6.34	<0.0001
T Stage	T1	4	1	1		
	T2	16	5	1.36	0.11 to 16.58	
	T3	116	38	1.46	0.15 to 14.52	
	T4	88	19	0.83	0.08 to 8.40	
	Tx	186	19	0.34	0.03 to 3.45	0.0002
N stage	N0	74	23	1		
	N1	70	22	1.02	0.50 to 2.06	
	N2	74	17	0.66	0.32 to 1.37	
	Nx	192	20	0.26	0.13 to 0.51	<0.0001
Metastases	Synchronous	310	41	1		
	Metachronous	102	42	4.59	2.75 to 7.67	<0.0001
Liver metastases	No	119	59	1		
	Yes	293	24	0.09	0.05 to 0.16	<0.0001
Albumin (g/L)		315	58	1.12	1.06 to 1.18	<0.0001
CEA (ng/mL)		364	69	0.99	0.98 to 0.99	0.0001
CA19.9 (log10)		315	58	0.56	0.42 to 0.76	0.0001
LDH (x ULN)		220	35	0.15	0.04 to 0.48	0.001
ALP (x ULN)		359	68	0.18	0.08 to 0.38	<0.0001

Quelles applications ?

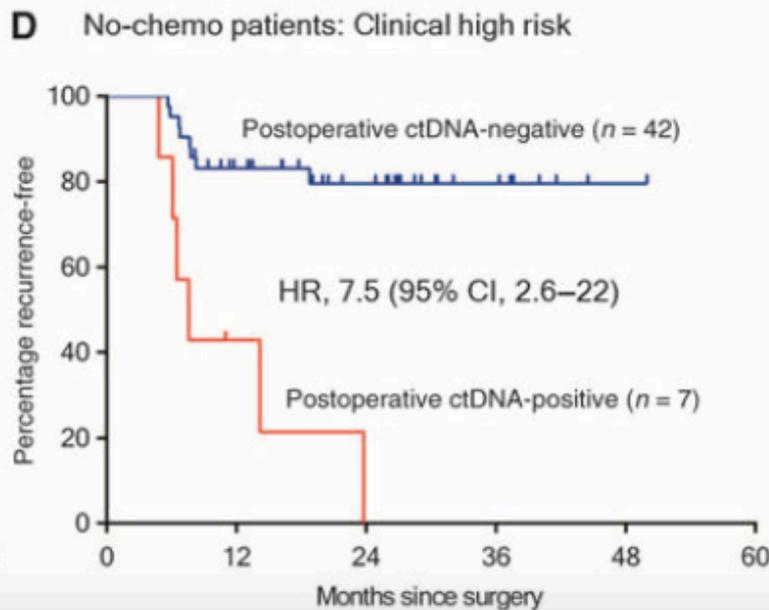
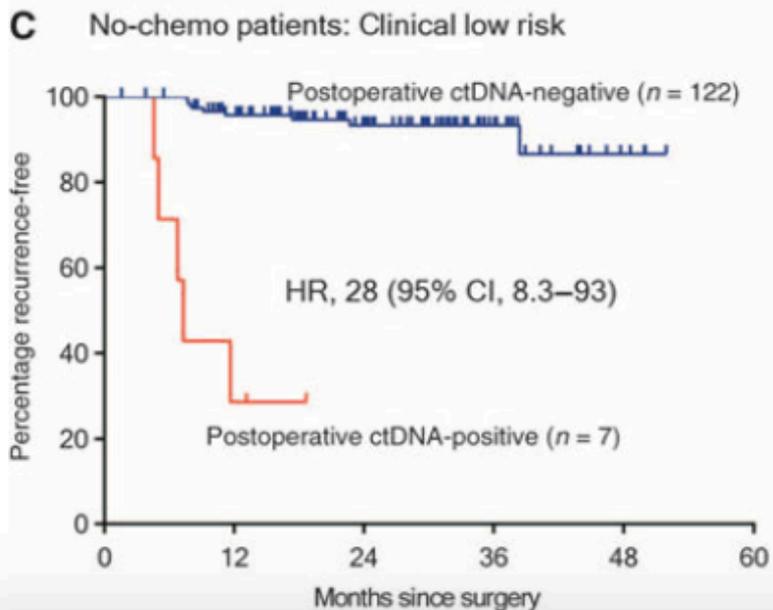
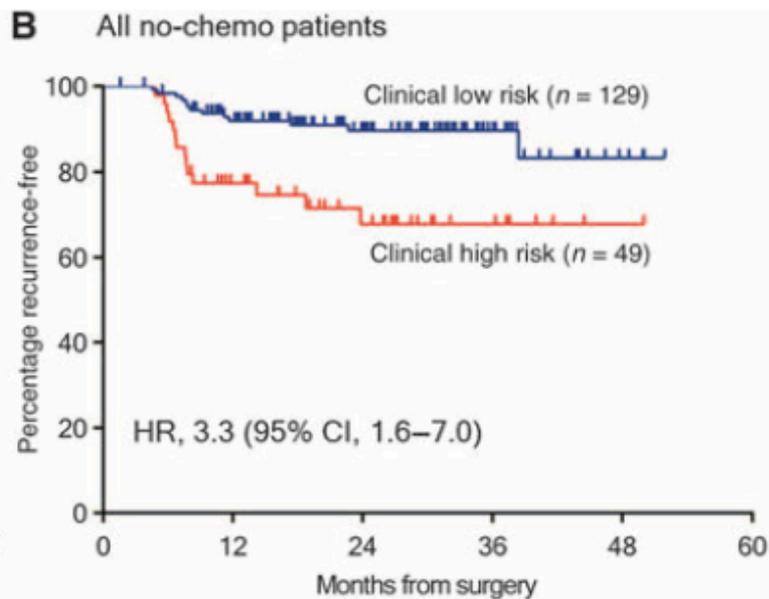
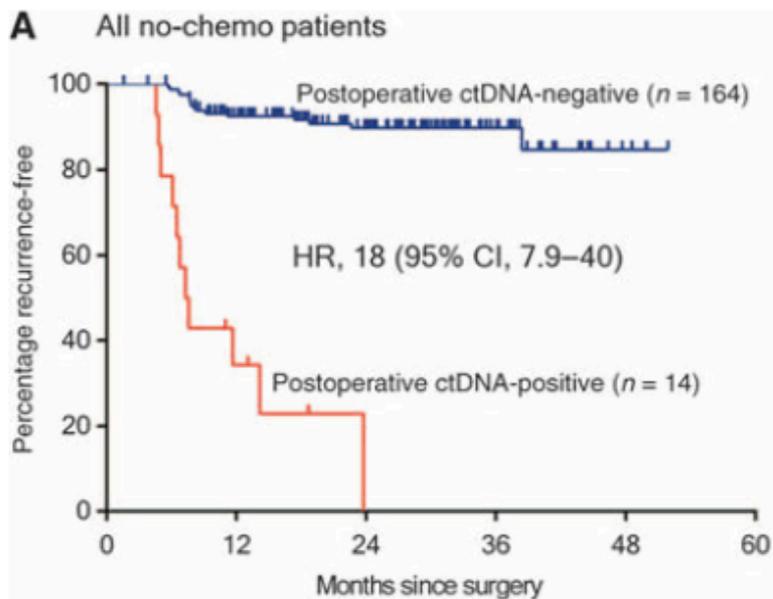
- ▣ Choix d'une thérapie ciblée
- ▣ Détection maladie résiduelle
- ▣ Indicateur efficacité traitement

Maladie résiduelle

Sci Transl Med. 2016 July 06; 8(346): 346ra92. doi:10.1126/scitranslmed.aaf6219.

Circulating tumor DNA analysis detects minimal residual disease and predicts recurrence in patients with stage II colon cancer

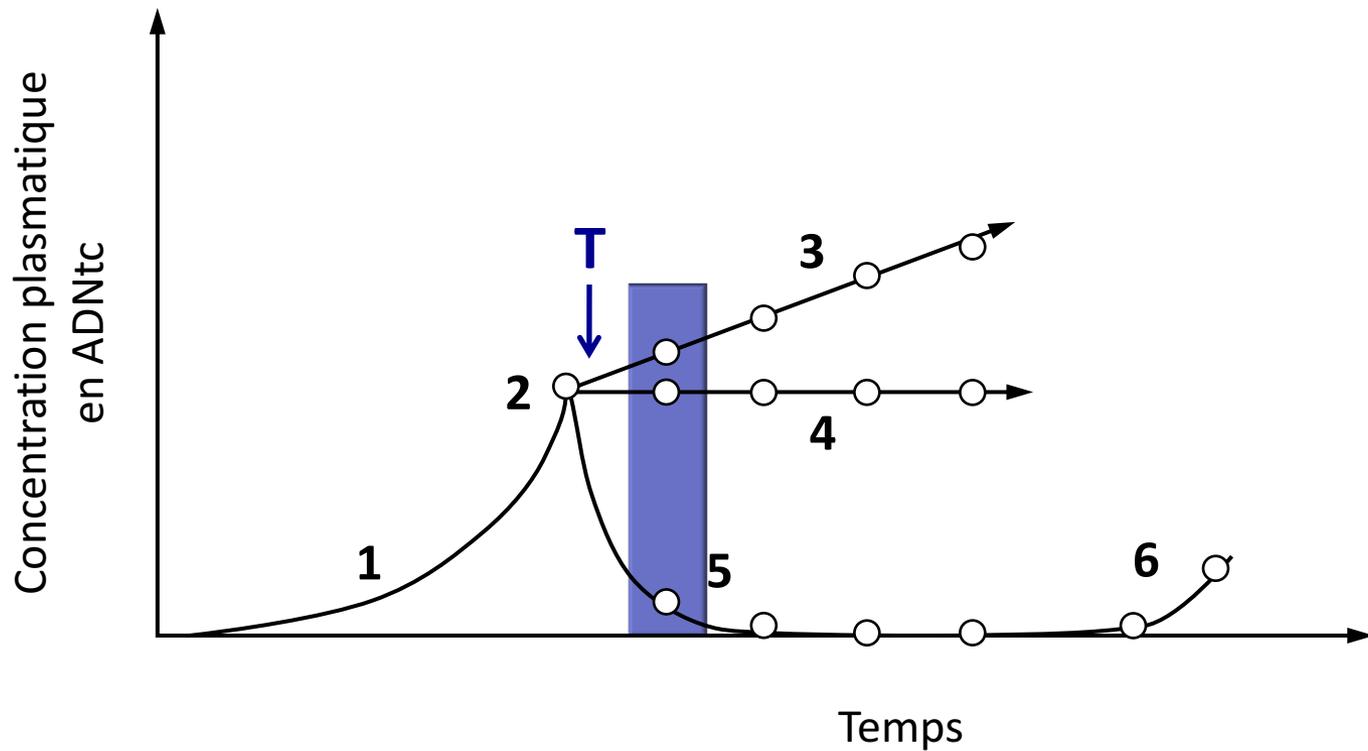
Jeanne Tie^{1,2,3,4,*†}, **Yuxuan Wang**^{5,†}, **Cristian Tomasetti**^{6,7}, **Lu Li**⁶, **Simeon Springer**⁵, **Isaac Kinde**⁸, **Natalie Silliman**⁵, **Mark Tacey**⁹, **Hui-Li Wong**^{1,3,4}, **Michael Christie**^{1,3,10}, **Suzanne Kosmider**², **Iain Skinner**², **Rachel Wong**^{1,11,12}, **Malcolm Steel**¹¹, **Ben Tran**^{1,2,3,4}, **Jayesh Desai**^{1,3,4}, **Ian Jones**^{4,13}, **Andrew Haydon**¹⁴, **Theresa Hayes**¹⁵, **Tim J. Price**¹⁶, **Robert L. Strausberg**¹⁷, **Luis A. Diaz Jr.**⁵, **Nickolas Papadopoulos**⁵, **Kenneth W. Kinzler**⁵, **Bert Vogelstein**^{5,*†}, and **Peter Gibbs**^{1,2,3,4,17,*†}



Quelles applications ?

- ▣ Choix d'une thérapie ciblée
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Evolution ADNtc en cours de traitement



Etude PLACOL

Personalized Medicine and Imaging

Clinical
Cancer
Research

Early Evaluation of Circulating Tumor DNA as Marker of Therapeutic Efficacy in Metastatic Colorectal Cancer Patients (PLACOL Study)

Fanny Garlan¹, Pierre Laurent-Puig^{1,2}, David Sefrioui³, Nathalie Siauve⁴, Audrey Didelot¹, Nasrin Sarafan-Vasseur⁵, Pierre Michel³, Geraldine Perkins^{1,6}, Claire Mulot¹, Hélène Blons^{1,2}, Julien Taieb⁶, Frederic Di Fiore⁷, Valerie Taly¹, and Aziz Zaanani^{1,6}



Patients with mCRC ($n = 82$)
First-line ($n = 68$) or second-line ($n = 14$) chemotherapy regimen

Mutation analysis in tumor

Identification of
targetable mutation* in the tumor
 $n = 43$ (52.4%)

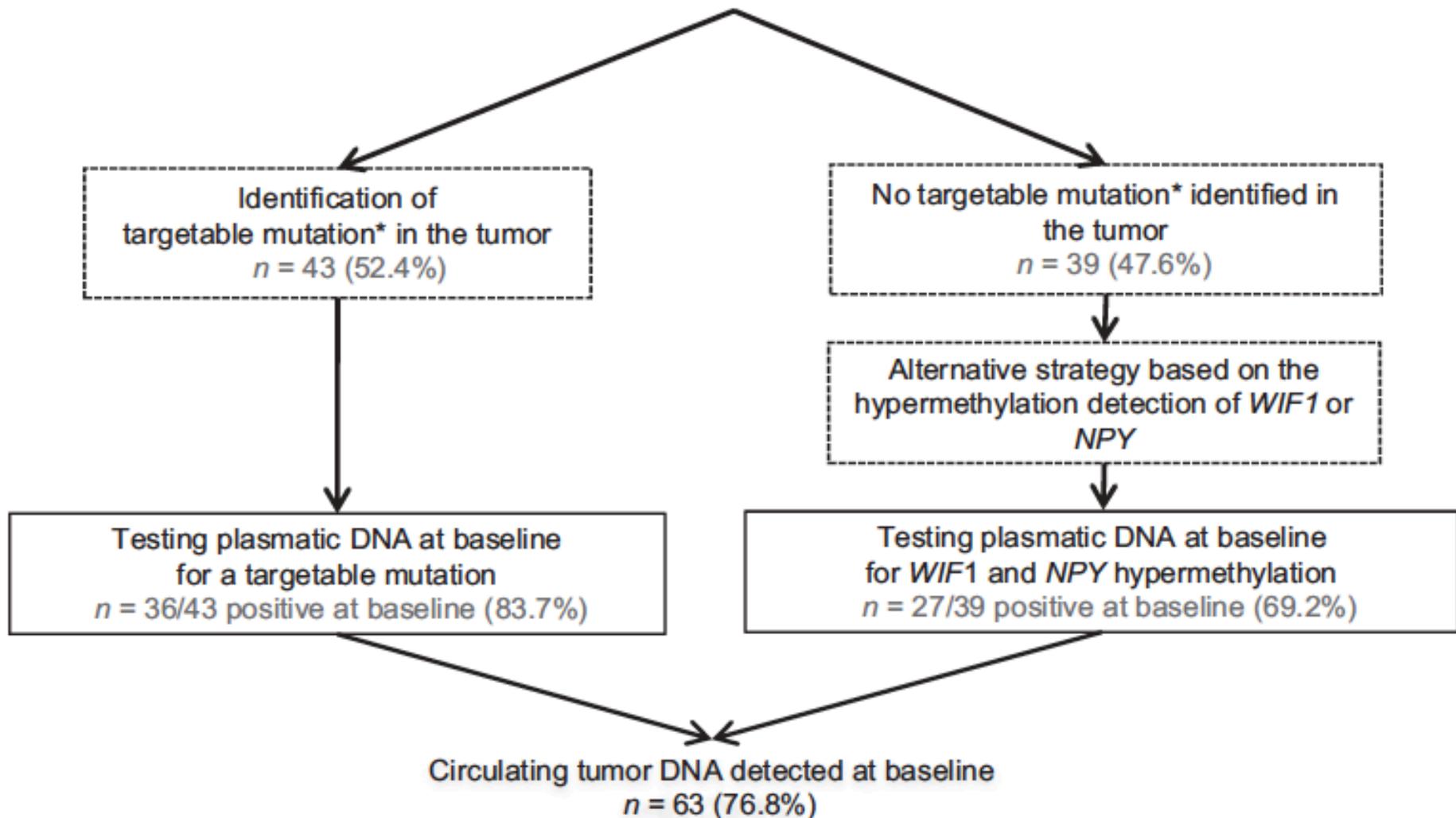
No targetable mutation* identified in
the tumor
 $n = 39$ (47.6%)

Alternative strategy based on the
hypermethylation detection of *WIF1* or
NPY

Testing plasmatic DNA at baseline
for a targetable mutation
 $n = 36/43$ positive at baseline (83.7%)

Testing plasmatic DNA at baseline
for *WIF1* and *NPY* hypermethylation
 $n = 27/39$ positive at baseline (69.2%)

Circulating tumor DNA detected at baseline
 $n = 63$ (76.8%)



[ADNtc], facteur pronostique

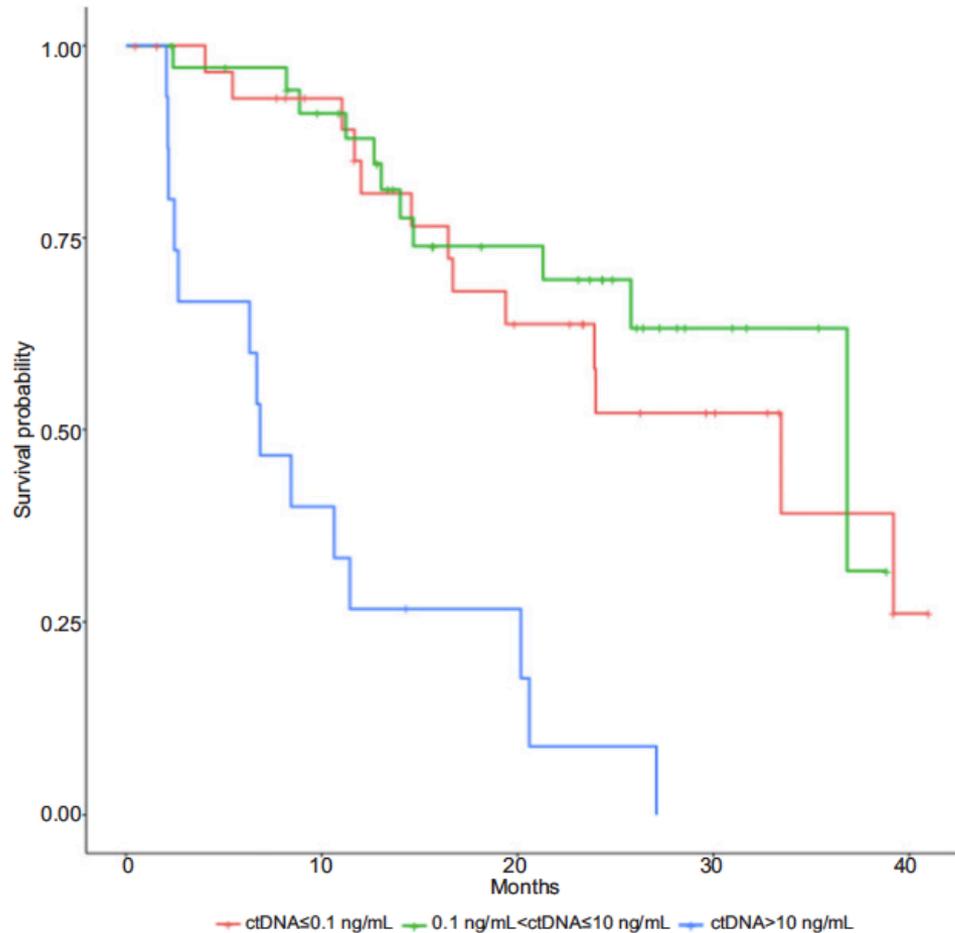
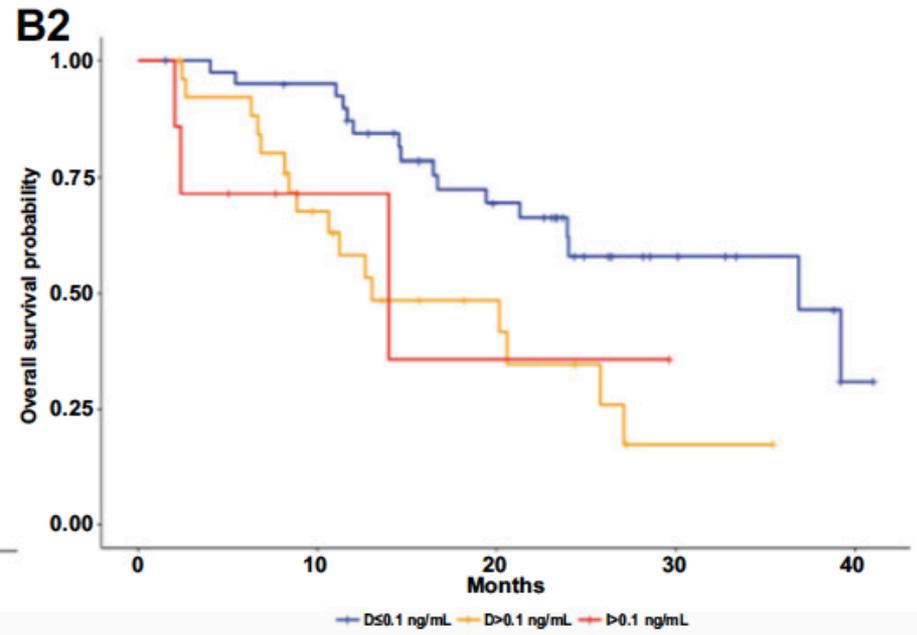
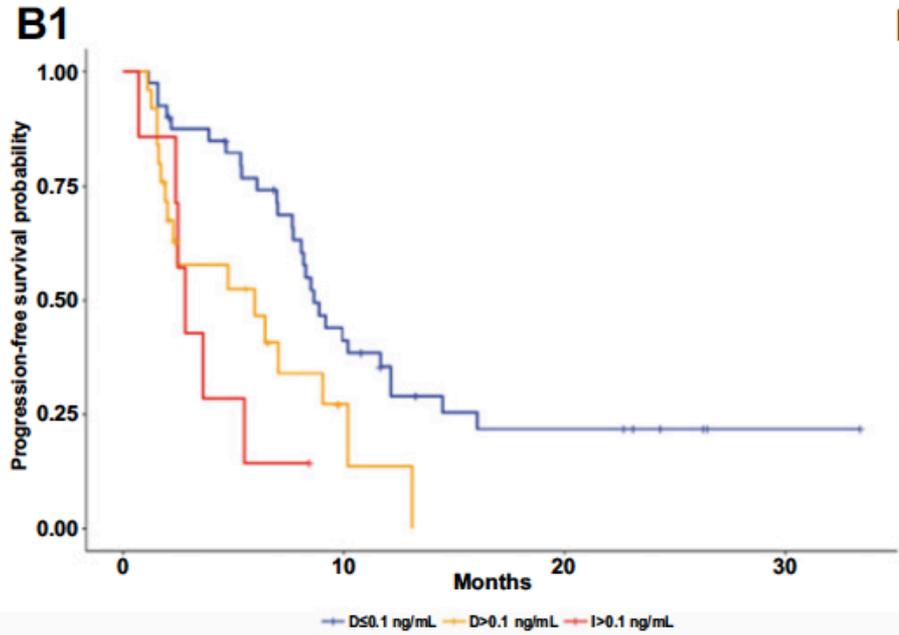
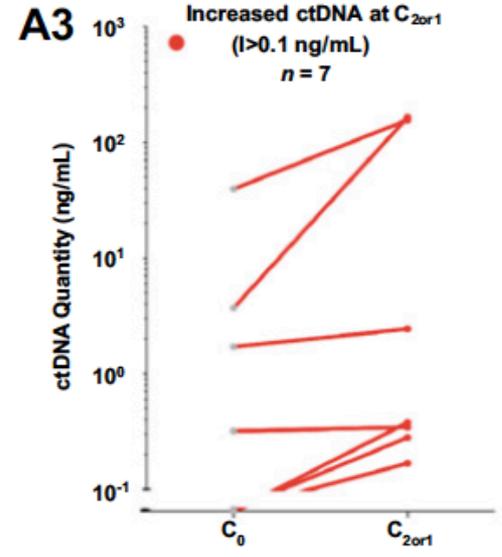
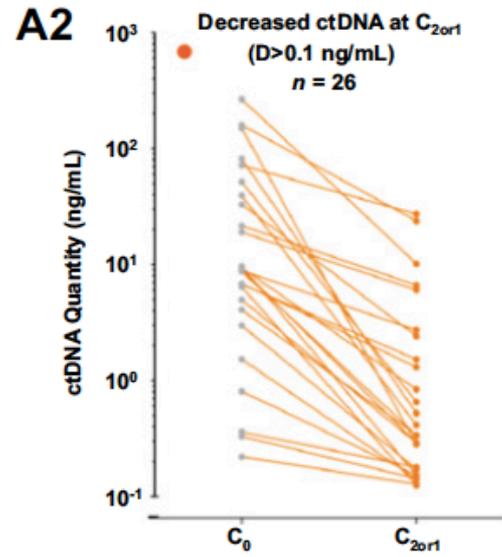
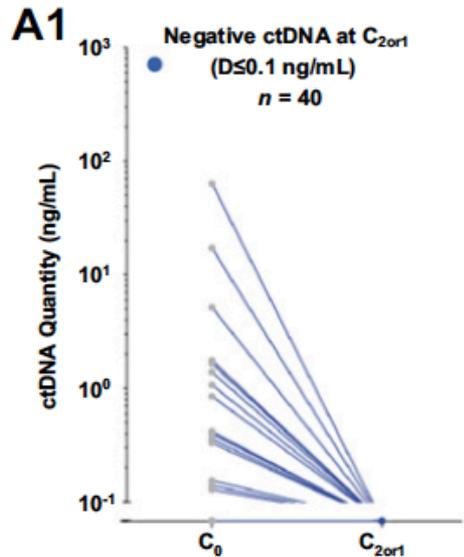


Figure 2.

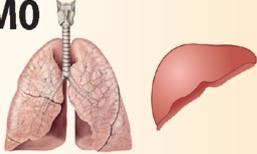
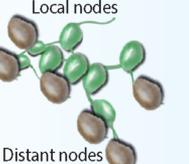
Impact of baseline ctDNA concentration on overall survival (OS). Three groups of patients were identified according to ctDNA concentrations at baseline: below 0.1 ng/mL (red), between 0.1-10 ng/mL (green), and above 10 ng/mL (blue).





Biopsies liquides (ADN circulant) :

- Matériel facilement accessible
- Alternative si tissus non disponible pour choix traitement ciblé
- ADNtc présent dans tous les liquides biologiques
- Délai de rendu de résultat inférieur / tissus
- Moindre sensibilité à l'hétérogénéité tumorale
- Suivi possible : mesure précoce de réponse au traitement et rechute ?

T Tumor Size	N Lymph Node	M Metastasis	B Blood
T1  Tumor size/local invasion	N0 Local nodes  Distant nodes No regional lymph node invasion	M0  No distant metastasis	B0  ctDNA No ctDNA mutations in blood
T2  Tumor size/local invasion	N1 Local nodes  Distant nodes Tumor spread to closest or small number of regional lymph nodes	M1  Distant metastasis	B1  ctDNA mutations in blood (can be further defined with more detailed quantification in the future)
T3  Tumor size/local invasion	N2 Local nodes  Distant nodes Tumor spread to an extent between N1 and N3		
T4  Tumor of any size that invades to other organs	N3 Local nodes  Distant nodes Tumor spread to more distant or regional numerous lymph nodes		

*Incorporating blood-based liquid biopsy information into cancer staging:
time for a TNMB system?*