Detection of circulating tumor DNA following neoadjuvant chemotherapy and surgery to anticipate early relapse in ER positive and HER2 negative breast cancer: Analysis from the PENELOPE-B trial

Nicholas Turner, Frederik Marmé, Sung-Bae Kim, Hervé Bonnefoi, Jose Angel García-Sáenz, Antonio Antón Torres, Harry Bear, Hans Tesch, Mireia Melé Olivé, Nicole McCarthy, Josefina Cruz Jurado, Seock-Ah Im, Yuan Liu, Zhe Zhang, Karsten Weber, Bärbel Felder, Valentina Nekljudova, Toralf Reimer, Carsten Denkert, Sibylle Loibl

Disclosures

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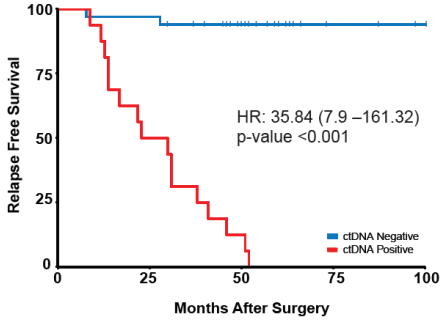
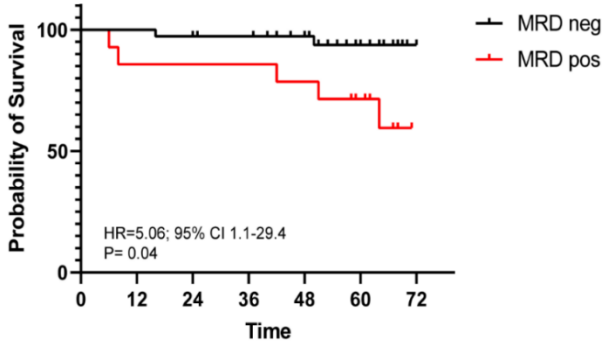
Background – ctDNA in early breast cancer

- Tumors release circulating tumor DNA (ctDNA) into the circulation
- Detection of ctDNA in follow-up anticipates future relapse with high accuracy[1,2]
- Limited data suggest detection of molecular residual disease immediately after surgery
- Potential use of ctDNA in selection of adjuvant CDK4/6i unclear

Molecular relapse monitoring[2]



MRD detection after primary suregry[3]



1. Garcia-Murillas *et al* STM 2015,
2. Coombes *et al* CCR 2019,
3. Garcia-Murillas SABCS 2021

Background

PENELOPE^{B}

N=1250
- HR+/HER2- breast cancer
- no pCR after NACT
- CPS-EG score ≥3 or ≥2 with ypN+

Primary Endpoint: iDFS

Stratification factors
- Nodal status: ypN 0-1 vs ypN2-3
- Age: ≤50 vs >50 yrs
- Ki-67: >15% vs ≤ 15%
- Region: Asian vs non Asian
- CPS-EG Score: ≥3 vs 2 and ypN+

Neoadjuvant Chemotherapy → **Surgery +/- Radiotherapy** → **R 1:1**

Palbociclib
125 mg once daily p.o.
d1-21, q28d for 13 cycles

Placebo
d1-21, q28d for 13 cycles

All patients received concomitantly endocrine therapy according to local standards.

Samples for ctDNA analysis: Baseline, Cycle 7, EoT

PenelopeB ET + Palbociclib/Placebo

1. Loibl S et al. J Clin Oncol 2021

Objectives

- To assess the potential of ctDNA analysis to predict future clinical relapse of patients enrolled in the PENELOPE-B trial
- To assess the potential role of sequential ctDNA analysis, ctDNA dynamics, in assessing future clinical relapse
- To assess whether a full analysis of baseline samples is indicated to assess whether palbociclib has benefit in ctDNA positive patients

Methods

- Patients endocrine naïve at the time of study entry were selected (129 of 1250)

- Biomaterial was available and ctDNA analysis was performed for 83 patients
  - 78 patient successful determinations at baseline
  - ctDNA analysis set was representative of the overall endocrine naïve group, with median follow-up of 42.9 months

- 210 plasma samples were collected in Streck tubes and processed

- Association of ctDNA with invasive disease-free survival (iDFS) and distant metastasis-free survival was analysed using Cox proportional hazard models.

ctDNA analysis methods

A tumor sample was exome sequenced, and up to 48 tumor variants were tracked in plasma using error-corrected sequencing for ctDNA detection (RaDaR assay).

Step 1

Patient's tumor sample (FFPE) is sent to the NeoGenomics laboratory

Step 2

Patient's tumor DNA is sequenced to determine the tumor's unique mutation profile

Step 3

A personalized RaDaR panel is designed for the patient

Step 4

After initial panel design, ctDNA is tested using blood samples and the patient's custom RaDaR panel

Step 5

Report is generated

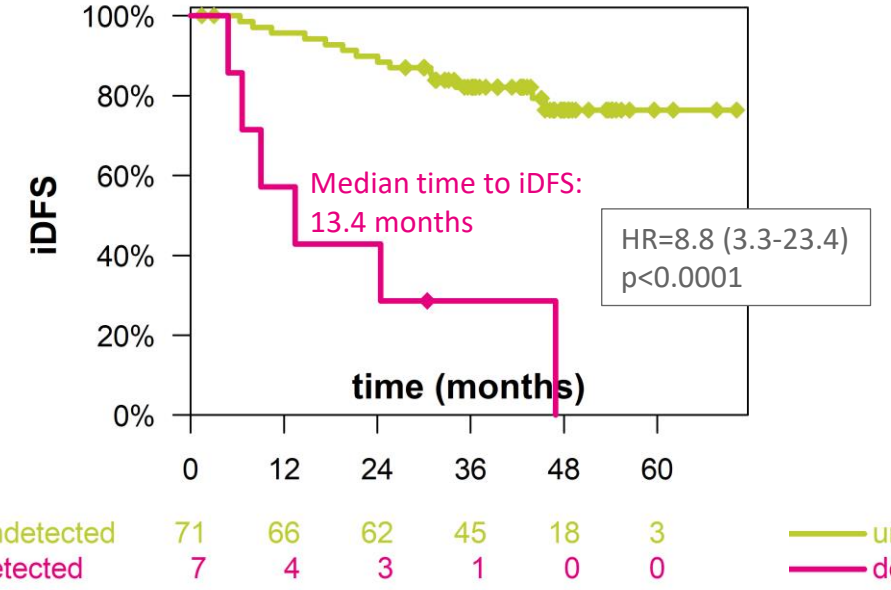
ctDNA detection

- Baseline ctDNA detection 9% (7/78) patients
- Of patients undetected at baseline 5% (3/66) had ctDNA detected in later samples
- Of patients detected at baseline, 29% (2/7) became undetected in later samples

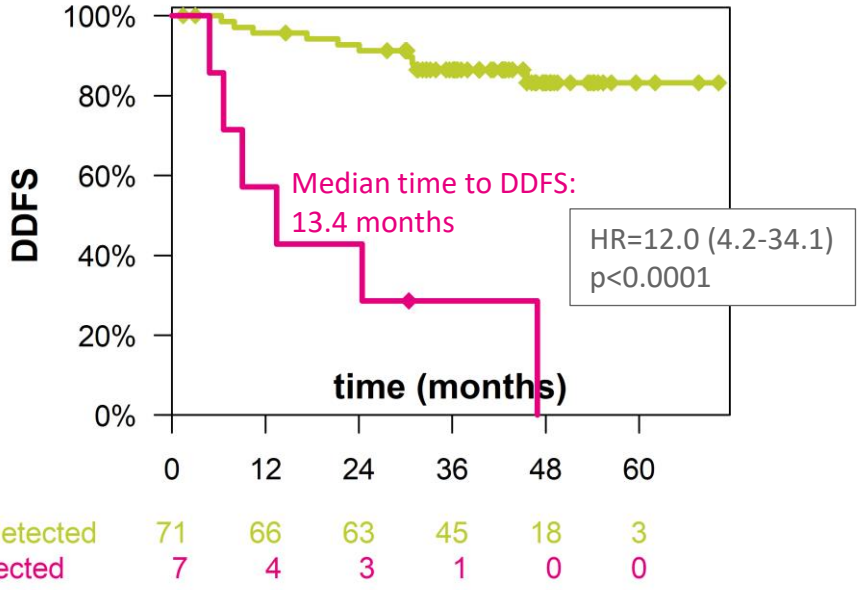
n	baseline	before cycle 7	EoT	Dynamics classification
48	undetected	undetected	undetected	all undetected
8	undetected	undetected		all undetected
7	undetected		undetected	all undetected
5	undetected			Not classified (one sample)
1	detected	undetected	undetected	becoming undetected
1	detected	undetected		becoming undetected
2	undetected	undetected	detected	becoming detected
1	undetected	detected	detected	becoming detected
2	detected		detected	all detected
3	detected	detected	detected	all detected

Results – baseline ctDNA detection

Invasive disease free survival



Distant disease free survival



Results – baseline ctDNA detection

Multivariable analysis:

Invasive disease free survival

variable	comparison	HR	P
ctDNA at baseline	detected vs undetected	6.47 (2.19-19.12)	0.0007
Ki-67	>15% vs ≤15%	1.90 (0.70- 5.14)	0.2054
ypT	ypT3-4 vs ypT0-2	2.03 (0.75- 5.52)	0.1644

Distant disease free survival

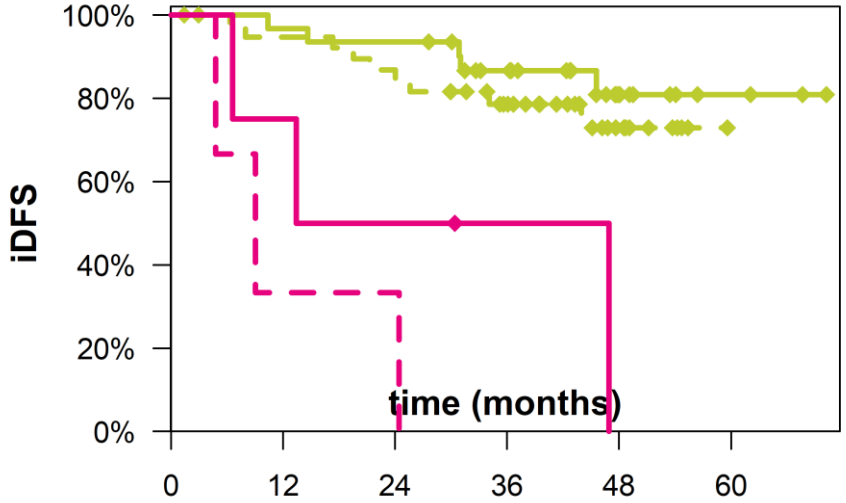
variable	comparison	HR	P
ctDNA at baseline	detected vs undetected	10.93 (3.47-34.48)	<0.0001
Ki-67	>15% vs ≤15%	1.17 (0.37- 3.74)	0.7875
ypT	ypT3-4 vs ypT0-2	1.96 (0.63- 6.11)	0.2434

ctDNA analysis dominated multivariable analysis

Results – baseline ctDNA detection

Invasive disease free survival

Test for interaction: p=0.5456



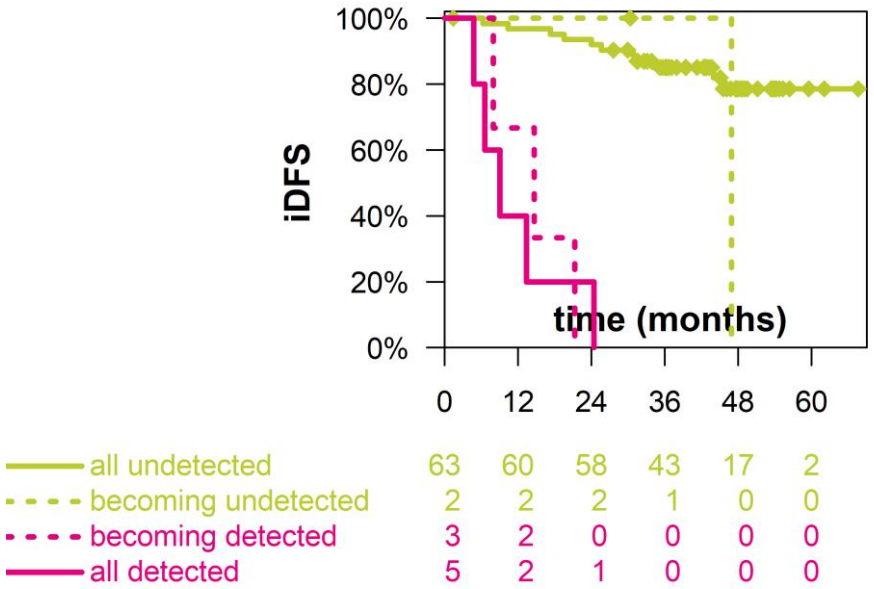
	0	12	24	36	48	60
undetected, palbociclib	33	30	29	21	9	3
undetected, placebo	38	36	33	24	9	0
detected, palbociclib	4	3	2	1	0	0
detected, placebo	3	1	1	0	0	0

Split by treatment allocation

Groups too small to draw conclusions

Results – ctDNA dynamics

Invasive disease free survival



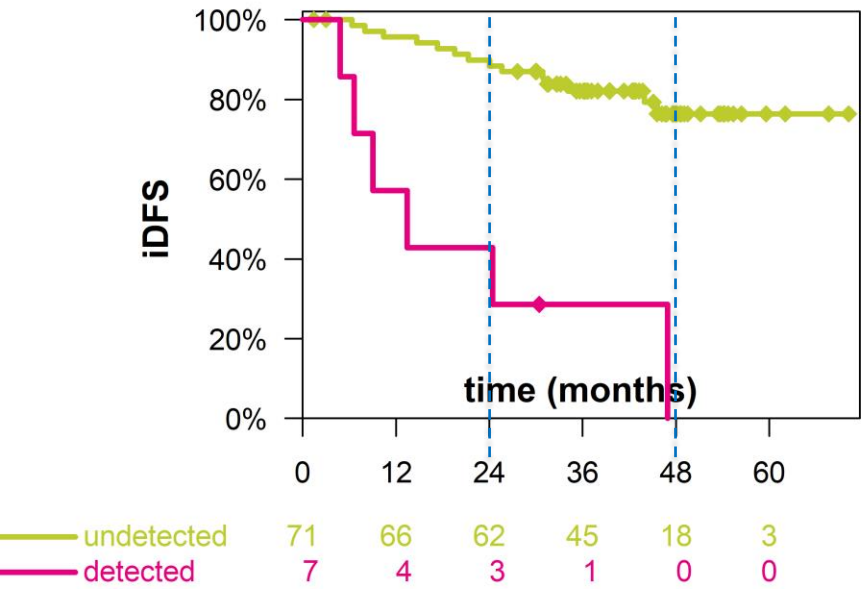
iDFS by ctDNA dynamic groups

Patients with undetected baseline ctDNA, who become positive during treatment have poor outcome

Analysis limited by small groups

Excludes 5 patients with a baseline sample and no subsequent samples

Limitations of baseline ctDNA analysis



'Sensitivity' for relapse analysed in time windows

Higher sensitivity (49%) for relapses within 12 months

Low/Moderate sensitivity for early relapses within 24 months (36% of currently observed)

Very low sensitivity for relapses >48 months (relapses not yet observed, but will occur with longer follow-up)

Analysis limited by follow-up

Conclusions

- Detection of ctDNA following neoadjuvant chemotherapy, and surgery, is associated with a very high risk of early relapse suggesting limited efficacy of adjuvant endocrine therapy
  - Studies of clinical imaging and experimental therapy are warranted for these patients

- 'Sensitivity' for future relapse is imperfect, in particular for later relapses, in patients who had prior neoadjuvant chemotherapy and surgery
  - Response to prior neoadjuvant chemotherapy may reduce ctDNA detection rates
  - Sequential testing improves 'sensitivity' for relapse

- Although Signatera ctDNA analysis has been approved by Medicare, use is likely not appropriate in deciding whether to give adjuvant CDK4/6 inhibitor in patients otherwise eligible, after neoadjuvant chemotherapy

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Cooperating partners

Collaborating study groups

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BioBank team at Philipps Universität Marburg

Central Lab team at Inivata A Leader in Liquid Biopsy Subsidiary of NeoGenomics

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GBG

Members of the Subboard GBG and AGO-B

Data management:
Christiane Praetor
Keyur Mehta

Medical Team:
Sabine Seiler
Johannes Holtschmidt

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