Targeted Isolation of Tumor-Derived Extracellular Vesicles Using a Cocktail of Magnetic Nanospheres for Minimally Invasive Pancreatic Cancer Diagnosis

1. INTRODUCTION AND BACKGROUND

Pancreatic Cancer – The King of Cancers

Epidemiology	Prognostics	Diagnosis and treatment
4th leading cause of cancer-related deaths in U.S. today	1st mortality rate	87% diagnosed at advanced stages
2nd leading cause of cancer-related deaths in U.S. by 2030	4 months median overall survival	< 20% eligible for surgery
10th in incidence in U.S. today		

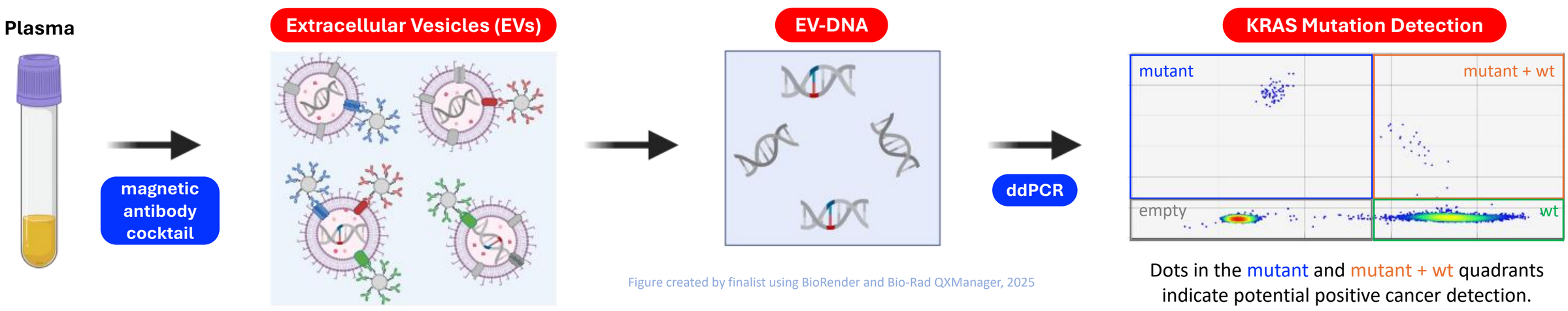
Tables created by finalist in PowerPoint using data from Stoop, T. F., et. al. Pancreatic cancer. *The Lancet* 405, 1182-1202, (2025).

Current Challenges in Pancreatic Cancer Diagnosis

- **Medical imaging** is widely used for cancer detection but is limited by *high cost, low sensitivity, radiation exposure risks, and infrequent applicability*
- **Tissue biopsy** enables molecular diagnosis, yet it is *invasive, expensive, prone to sampling bias, and often inaccessible for pancreatic cancer*
- **Circulating free DNA**-based liquid biopsy offers a minimally invasive alternative but *lacks sensitivity for early cancer diagnosis and has limited multiplexing capability for RNA and protein analysis*

➡ **To win the battle against pancreatic cancer, it is urgent to develop innovative and effective diagnostic methods for early diagnosis, treatment monitoring, and recurrence surveillance.**

2. APPROACH: EV ISOLATION + ddPCR



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Dots in the mutant and mutant + wt quadrants indicate potential positive cancer detection.

3. RESULTS

Comparison with Standard Method (Exploratory Cohort)

KRAS mutations were detected in all 20 pancreatic cancer patients, while 9 of 10 healthy donors were mutation-negative.

Sensitivity and specificity

	Patients	Healthy donors
Test positive	TP = 20	FP = 1
Test negative	FN = 0	TN = 9

$Sensitivy = 100\%$

$Specificity = 90\%$

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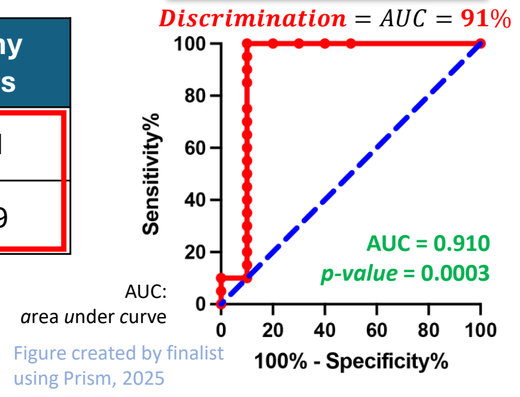
Discrimination



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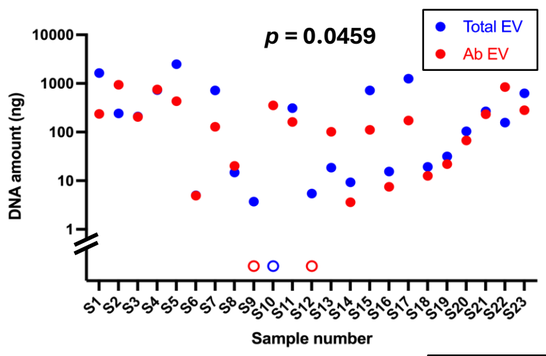
EV-DNA amount

Antibody cocktail–based EV isolation yielded **significantly less EV DNA** than total EV isolation (**Wilcoxon matched-pairs signed rank test** p = 0.0459).



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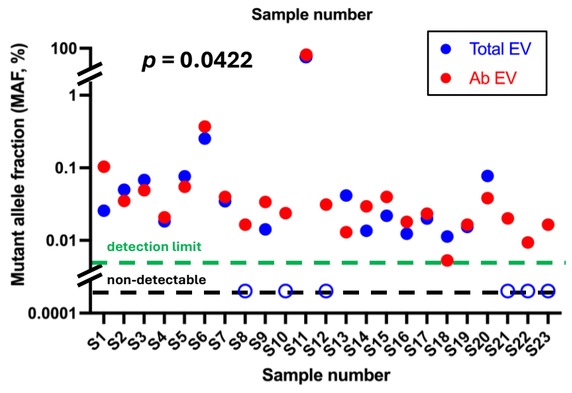
Clinical sensitivity

The antibody cocktail-based EV isolation achieved **100%** KRAS detection versus **73.9%** with total EV isolation, with **significantly higher detectability** (**Wilcoxon matched-pairs signed rank test** p = 0.0422).



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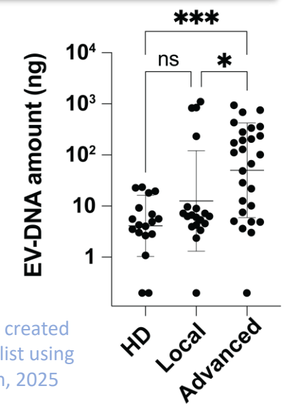
Impact of Cancer Stage on EV-based Clinical Diagnosis

Pooling training and validation cohorts, PDAC patients were grouped as local/early (stages I–II) or advanced (stages III–IV) stage disease.

EV-DNA amount



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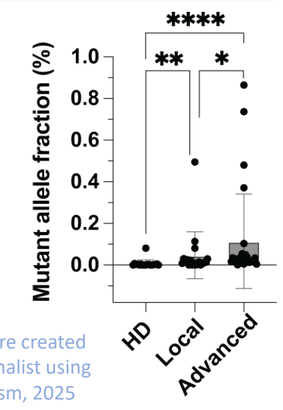
EV-DNA yields were higher in advanced versus local disease (p = 0.0444, Mann–Whitney).

KRAS mutation MAF



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KRAS MAF was higher in advanced versus local disease (p = 0.0469, Mann-Whitney).

Late-stage sensitivity, specificity and discrimination

Sensitivity and specificity

	Patients	Healthy donors
Test positive	TP = 25	FP = 2
Test negative	FN = 2	TN = 16

$Sensitivy = 92.6\%$

$Specificity = 88.9\%$

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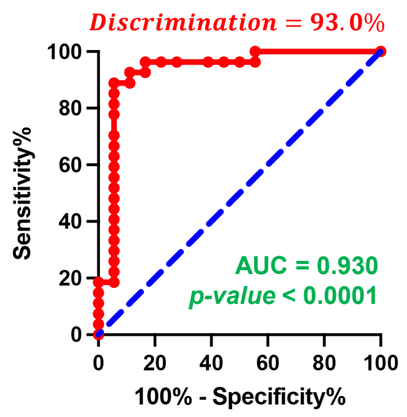
Discrimination



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Sensitivity and specificity

	Patients	Healthy donors
Test positive	TP = 15	FP = 2
Test negative	FN = 4	TN = 16

$Sensitivy = 78.9\%$

$Specificity = 88.9\%$

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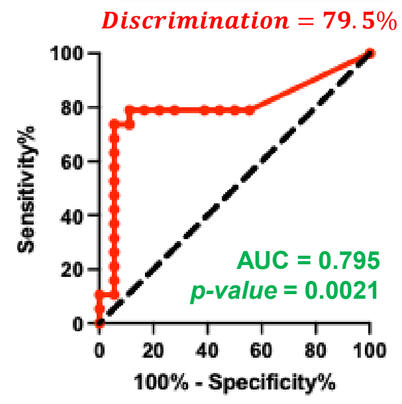

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Evaluation in Validation Cohort

KRAS mutations were detected in 20 pancreatic cancer patients (6 false negatives), while 7 of 8 healthy donors were mutation-negative.

Sensitivity and specificity

	Patients	Healthy donors
Test positive	TP = 20	FP = 1
Test negative	FN = 6	TN = 7

$Sensitivy = 76.9\%$

$Specificity = 87.5\%$

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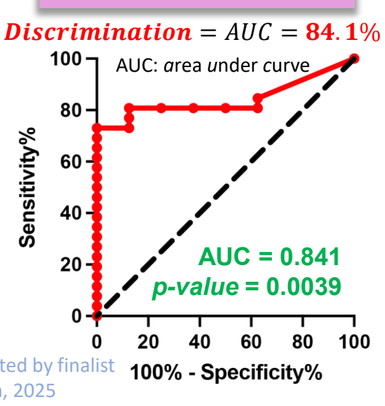

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Overall sensitivity, specificity and discrimination

Sensitivity and specificity

	Patients	Healthy donors
Test positive	TP = 40	FP = 2
Test negative	FN = 6	TN = 16

$Sensitivy = 87.0\%$

$Specificity = 88.9\%$

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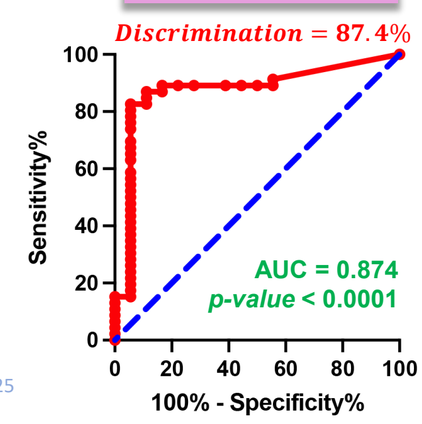
Discrimination



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4. CONCLUSIONS

- ✓ **87.0% sensitivity and 88.9% specificity** for 46 pancreatic cancer patients and 18 healthy donors.
- ✓ **Compared to total EV isolation**, cancer-specific EV increased clinical sensitivity (100% vs 73.9%).
- ✓ **Overall method** offers a highly sensitive, minimally invasive approach for molecular diagnosis.

KEY REFERENCES

1. Kalluri, R., and LeBleu, V. S. (2020), *Science*, 367(6478), eaau6977.
2. Shao, H., et. al. *Chemical Reviews*, 118(4), 1917-1950.
3. Weinberg, R. A. (2013). *Biology of Cancer*, Second Edition. Garland Science.

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