MicroRNAs as Bile-Based Biomarkers in Pancreatic Cancer and Biliary Tract Cancers

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1. Introduction

Pancreatic cancer is one of the most lethal diseases in the world with 9,618 new cases and around 8,800 deaths in the UK. It is the fourth most common cause of cancer death in the UK (2014). The lack of efficient diagnostic techniques and limited treatment options leads to a late detection and poor survival rates (less than 1%)[1].Therefore, further investigation is needed to improve the detection process and design specific molecular targeted treatments.

During last decade, a class of non-coding RNAs (microRNAs) are emerging as potential biomarkers and therapeutic targets for different cancers. In this study, the role of miRNAs has been assessed in a large cohort of pancreatic cancer.

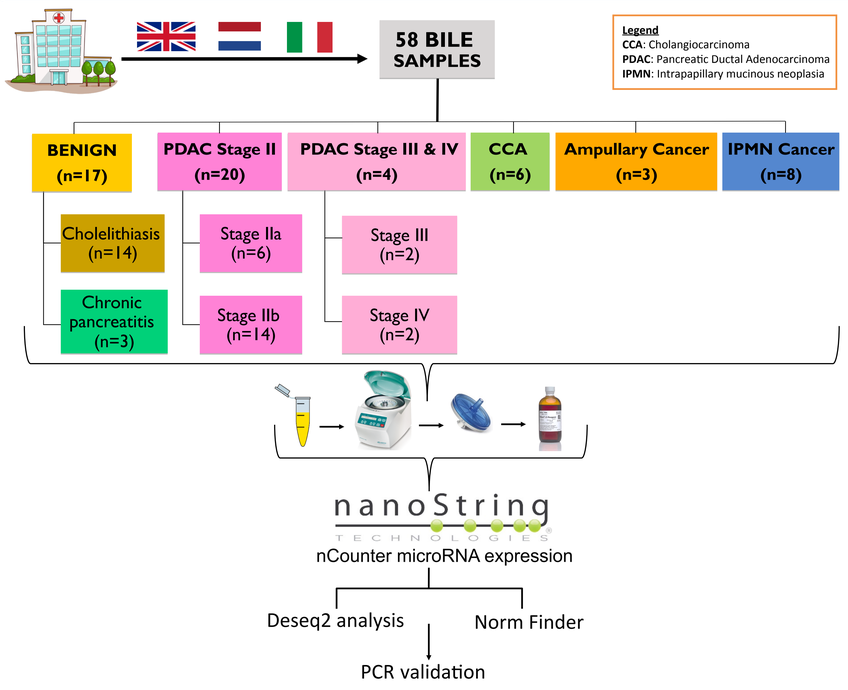
2. Aims of the study

Pancreatic cancer patients present an alteration of the miRNAs when compared to healthy individuals and this could allow us to detect the malignancy. Finding novel miRNAs altered in the bile fluid associated to pancreatic cancer or biliary tract cancers would be a considerable discovery because of the close proximity of this biological fluid to the malignant lesion.

Main objectives:

- Develop a bile-based diagnostic test focused on miRNA expression to detect pancreatic cancer early.
- Establish a bile miRNA signature to stratify pancreatic cancer pre-operatively.

3. Materials and Methods



4. Results

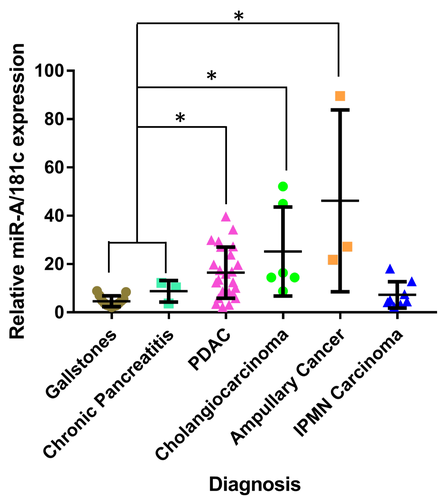


Figure 1. Relative expression of miR-A in benign and malignant human bile samples. Displayed are the expression levels across different diagnosis for miR-A. Scatterplots are shown for this miRNA and the horizontal lines represent the mean expression level and standard deviation. One-way analysis of variance (ANOVA) was used to compare miRNA levels between patient diagnosis, followed by Tukey's multiple comparison test and unpaired t-test, (the asterisk (*) means p-value < 0.05).

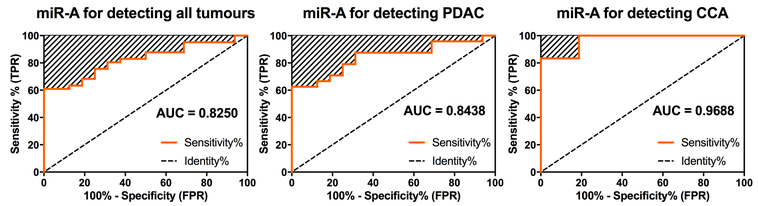


Figure 2. Performance of ROC curves of miR-A for detecting pancreatic malignancy in human bile samples. Displayed are ROC and AUC curves for miR-A (from left to right)when comparing malignant vs benign, PDAC vs benign and CCA vs benign.

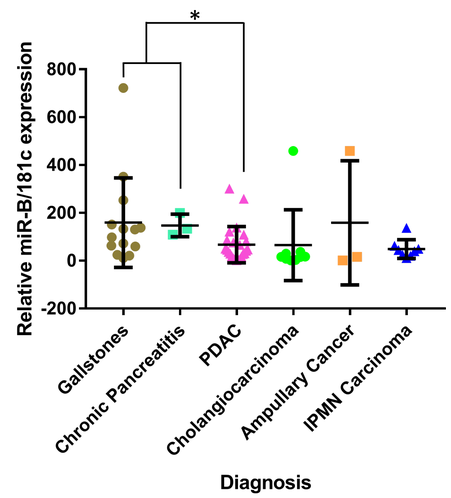


Figure 3. Relative expression of miR-B in benign and malignant human bile samples. . Displayed are the expression levels across different diagnosis for miR-A. Scatterplots are shown for this miRNA and the horizontal lines represent the mean expression level and standard deviation. One-way analysis of variance (ANOVA) was used to compare miRNA levels between patient diagnosis, followed by Tukey's multiple comparison test and unpaired t-test, (the asterisk (*) means p-value < 0.05).

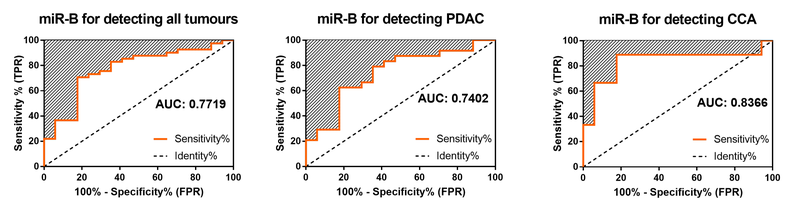


Figure 4. Performance of ROC curves miR-B for detecting pancreatic malignancy in human bile samples. Displayed are ROC and AUC curves for miR-B (from left to right) when comparing malignant vs benign, PDAC vs benign and CCA vs benign.

5. Conclusions

- These two candidate miRNAs (miR-A and miR-B) can distinguish between malignant and benign disease (AUC=0.8250, p-value=0.0002 ;AUC=0.7719, p-value=0.0012, respectively).
- In PDAC cases, miR-A exhibited an AUC higher than 0.75 (p-value=0.0003). It could be a more specific PDAC biomarker, although both candidates are significantly differentially expressed in comparison with benign cases.
- miR-A and miR-B are good candidates for detecting cholangiocarcinoma because they exhibited an AUC > 0.75 (p-value=0.0009 and p-value=0.0055, respectively).

Furthermore, these miRNAs could be used as bile-based biomarkers in order to help clinicians in cases of diagnostic uncertainty. Moreover, these bile-biomarkers may have a role in stratifying cancer patients pre-operatively in order to personalise their treatments and improve their survival outcomes.

6. Future Work

The current interest of the project has been focussed on:

1) Validate our candidate bile-miRNAs by RT-qPCR in the same cohort.
2) Use a large and independent series of bile samples to validate the initial results (cases and controls, n=20/condition).
3) Evaluate the functional role of these candidate miRNAs by *in vitro* assays (in commercial and primary cancer cell lines), performing proliferation, migration and invasion assays.

References

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