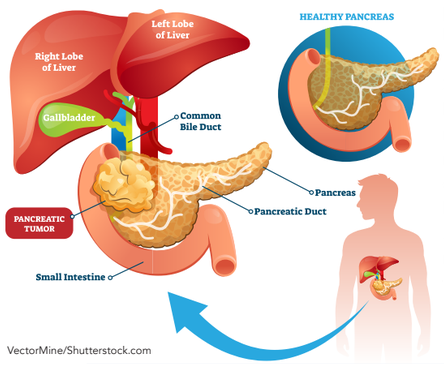
Blood biomarker breakthrough for early detection of pancreatic cancer

Nearly half a million people each year globally are diagnosed with pancreatic cancer (PC). Timely intervention is essential for the best possible prognosis. However, early-stage PC is notoriously difficult to diagnose. The dearth of diagnostics available for detecting early-stage PC means many lives are lost prematurely. Now there is hope for a new, non-invasive diagnostic test. Professor Kazufumi Honda at Nippon Medical School in Tokyo, Japan, leads research on the use of apolipoprotein A2-isoforms (apoA2-i) as blood biomarkers for early detection of PC and identification of cancer risk.

Pancreatic cancer (PC) was reported as the 12th most common form of cancer in 2020 – with over 495,000 new cases of the incurable disease reported worldwide. As populations across the globe increase, along with rising levels of obesity and longer lifespans, PC is predicted to become more common and claim more lives. Other risk factors for PC include smoking, physical inactivity, diet, inheritance, and genetic predisposition. Global Cancer Observatory (GLOBOCAN) estimates that in 20 years, figures for PC will increase by 70%, with 844,000 new cases each year.



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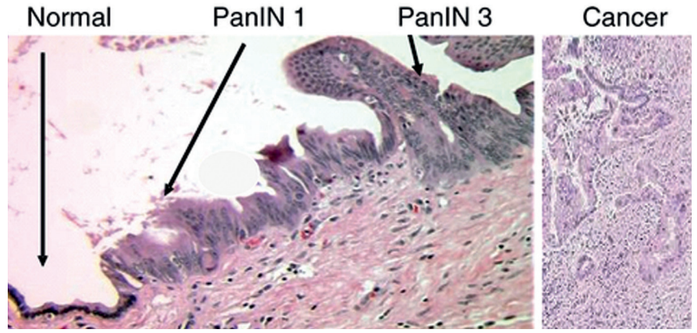
To tackle PC and reduce the rate of mortality worldwide, it's important to catch the disease early through an appropriate detection method. However, PC is an uncommon cancer with an age-adjusted annual incidence of 12.9 cases per 100,000 person-years (which accounts for 100,000 people across one year of study). Age-adjustment for yearly incidence of PC ensures that differences in incidence from one year to another (or between one geographic location and another) are not due to differences in the age distribution of the populations that are being compared. In the general population, the lifetime risk of developing PC is estimated at approximately 1.6% based on US data from 2015 to 2017. The International Cancer of the Pancreas Screening (CAPS) Consortium recommends targeted screening using magnetic resonance imaging (MRI) and endoscopic ultrasonography (EUS) for high-risk individuals having either over 5% lifetime risk or a five-fold increase in relative risk. An MRI is a medical imaging technique that uses a magnetic field and computer-generated radio waves to produce detailed images of body tissues or organs. The EUS technique uses ultrasound and camera data to generate detailed images for the purpose of assessing digestive and lung diseases.

The standardised incidence ratio for individuals in which there is familial PC in two first-degree relatives is reportedly 6.4-fold greater than that of the general population (95% confidential interval 1.8–16.4). However, it has been suggested that only 5–10% of PC patients have a familial basis, and most cases of PC are sporadic. Therefore, efficient screening strategies using non-invasive methods to distinguish high-risk individuals from the general population are urgently needed.

For screening of PC, especially two important precursor lesions (areas of abnormal tissue or cells) can signal the early stages of the disease: pancreatic intraepithelial neoplasia (PanIN) and intraductal papillary mucinous neoplasm (IPMN). IPMN is an especially important precursor because it is the only lesion that can be detected by non-invasive imaging. Additionally, individuals with pancreatic cyst or dilatation of the pancreatic duct have been considered to be at high-risk individuals (HRIs) of PC. Therefore, the development of biomarkers along with non-invasive methods is the unmet medical need to increase the diagnostic probabilities of subjects with high risk factors before the imaging for efficient PC screening.

Apolipoprotein A2-isoforms (apoA2-i) are also associated with PC and its precancerous lesions – and could be used as future biomarkers. Apolipoproteins like apoA2 are known for their involvement in lipoprotein metabolism. More specifically, apoA2 has been identified as an important component of high-density lipoprotein (HDL) and a mediator of lipid transportation. HDL is often called 'good' cholesterol, helping to absorb cholesterol and take it to the liver where it is processed for removal from the body. Research indicates that high levels of HDL cholesterol can lower an individual's risk of heart disease or stroke. On the other hand, apoA2-i levels may be associated with higher levels of 'bad' cholesterol (or low-density lipoproteins, LDLs) in the blood. Chief Professor Kazufumi Honda at Nippon Medical School in Tokyo, Japan, is working with a team on how to use these isoforms to screen patients with a high risk of PC.



Pancreas neoplasia carcinoma sequence.

Photo Credit: Palladin_expression_neoplasia.png: see above derivative work: Nephron TIC, CC BY 1.0 , via Wikimedia Commons

BLOOD BIOMARKER BREAKTHROUGH

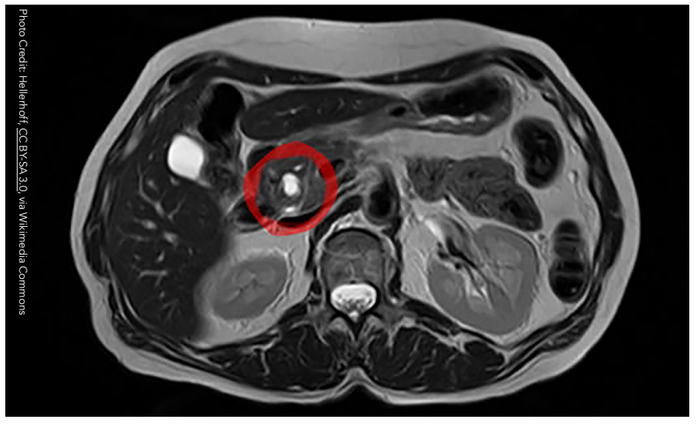
In a 2016 review, Honda details the use of top-down proteomics to screen expression profiles of proteins extracted from plasma samples, which are taken from the blood of healthy volunteers and patients with PC. Top-down proteomics involves the use of mass spectrometry (MS), an analytical technique that allows molecule mass to be measured for identification; expression profiling reveals what is happening in the protein. The method can be used to identify suitable biomarkers for the early detection of PC. In a 2005 study, conducted by Honda, a protein with a molecular weight of 17.3 kilodaltons (kDa) was identified in patients with the most common form of pancreatic tumours: pancreatic ductal adenocarcinoma (PDAC). The protein biomarker was found to be significantly decreased in concentration in patients with PDAC, compared to healthy controls. This finding was supported by results from other studies in Germany and Australia. The protein was an apoA2 isoform (or another form of the parent apoA2 homodimer) that lacked an amino acid at the end of the protein, known as the C-terminal end. A homodimer is a protein complex where two identical protein subunits are bound together.

Through further study, Honda and colleagues were able to identify five isoforms of apoA2 homodimers, two of which were newly identified. Three isoforms were identified in healthy blood: apoA2-AT/AT, apoA2-ATQ/ATQ, and apoA2-ATQ/AT. The remaining two novel isoforms were identified in the plasma of PC and risk diseases of PC: apoA2-AT/A and apoA2-A/A. Further analysis using MS revealed that in the plasma of patients with PDAC (at any clinical stage) the expression level of apoA2-ATQ/AT was significantly decreased compared to healthy controls. An immunological test, known as ELISA (or enzyme-linked immunosorbent assay), was conducted to measure apoA2 isoforms in clinical samples. Results of the assay point toward a significant reduction in plasma levels of apoA2-ATQ/AT at any stage of PDAC. Honda went on to explore the feasibility of using apoA2-ATQ/AT as a biomarker for the identification of the early stages of PC and related symptoms in clinical settings.

Pancreatic cancer remains one of the deadliest cancers worldwide, typically diagnosed late with a poor prognosis.

PROMISING TEST RESULTS: apoA2-i

In a prospective study published in 2020, Honda and his research team screened 5,120 people from the general population. The sample had a median age of 52 years old and was 51% male. The researchers evaluated the positive predictive value (PPV) of the plasma apoA2-ATQ/AT level of ≤35 μg/mL for detecting PC and individuals at high risk for PC. Blood plasma constitutes the liquid component of blood that does not contain red blood cells. Of the study participants enrolled, 84 individuals (1.3%) exhibited positive results for apoA2-ATQ/AT. Further imaging was undertaken for 54 of these individuals and found pancreatic abnormalities in 26 people (48%). Furthermore, 18 of these abnormalities were PC and high-risk diseases. This means that the PPV of apoA2-ATQ/AT for detecting PC and high risk was 33% (18/54), and if the test is positive, one in three people has PC or a high risk of developing the disease. In particular, the PPV of the plasma apoA2-ATQ/AT level for detecting pancreatic cystic lesions (PCL) was 25.9% (14/54, including 9 IPMN). These results indicated that the measurement of plasma apoA2-ATQ/AT levels could be used as an effective initial screening method to enhance the detection of PCL. Receiver Operating Characteristic (ROC) curve analysis serves as a measure for diagnostic test accuracy. ROC curve analysis revealed that the test, using plasma apoA2-ATQ/AT level for detecting PC and high-risk status, has a sensitivity of 40.6% adjusted for bias. Overall, Honda's study reveals the potential of measuring plasma apoA2-



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Intraductal papillary mucinous neoplasm in magnetic resonance imaging.

ATQ/AT levels to help diagnose PC and determine high-risk status before imaging examinations.

In a later study published in 2021, Honda and colleagues assessed the diagnostic performance of apoA2-i and compared it to that of the routine clinical biomarker carbohydrate antigen 19-9 (CA19-9). The latter biomarker is United States Food and Drug Administration (US FDA) approved for in vitro diagnostics (test tube tests) and is most commonly used for monitoring PC patient responses to therapy. However, among the scientific community, the effectiveness of CA19-9 is deemed limited, as it only reacts during the advanced stage of PC. It is therefore not considered sensitive enough for detecting early PC. Rather than clinical professionals discovering IPMN incidentally during abdominal screenings of patients, Honda and colleagues highlight the potential use of serum apoA2-ATQ/AT for the early detection of IPMNs with a high risk of progressing to PC. The term serum is usually used in biological and biomedical research to describe the clear, liquid part of blood that is left over after blood cells and clotting proteins have been removed.

The 2021 retrospective study reported that ROC curve analysis of IPMNs with high-grade dysplasia, and IPMNs with associated carcinoma, revealed the area under the curve (AUC) at 0.91 and >0.94, respectively. The term dysplasia is used to refer to the presence of abnormal cells within a tissue or organ of the body. Carcinoma describes a type of cancer that can begin to manifest in cells of the skin or the tissue lining organs, such as the liver or kidneys. The AUC values mentioned here indicate a highly accurate quantitative diagnostic test result. The respective sensitivities were 70% and 83% with a specificity of 95%. This result was significantly higher than for the gold standard biomarker, CA19-9. AUC values of apoA2-i were also high for IPMN-associated carcinoma of colloid and ductal subtypes. Overall, this study suggests that, excitingly, the serological apoA2-i test – with clinical imaging techniques – could help identify high risk in individuals with IPMN diagnosis.

The serological apoA2-i test – with clinical imaging techniques – could help identify high risk in individuals with IPMN diagnoses.

THE FUTURE FOR PC SCREENING

The use of blood biomarkers and medical imaging modalities to screen for PC in its early stages can improve the accuracy and cost-effectiveness of early diagnosis, as well as increase patient survival rates. At present, the medical and scientific communities are turning to the use of multi-analyte blood tests and other methods that combine several existing analyses to provide a solution. Some combination analyses involve the use of radiomic and/or radiogenic approaches to help identify signs of early-stage PC. Radiomics is a method that involves the use of data-characterisation algorithms to extract radiomic features from medical images. Through these features, tumoral patterns and characteristics can be uncovered. Radiogenomics involves the study of genetic variation associated with response to radiation. This method can also refer to the potential correlation between cancer imaging features and gene expression.

Despite the performance of these methods, they are expensive and may not be considered cost-effective for early-stage PC. Combining apoA2-i testing with suitable medical imaging techniques could be the answer. The use of these isoforms has been validated in several studies – and apoA2-ATQ/AT, in particular, has been revealed to be a credible candidate to use as a biomarker for the early detection of PC and identification of high-risk individuals.

Honda and his team have made significant progress in an area urgently requiring life-saving early diagnostics. To combat the deadly disease worldwide, further research to develop an early detection programme must be a top priority of scientists, healthcare providers, and governments internationally. Currently, the US Preventive Services Task Force (USPSTF) does not recommend screening for PC using imaging, because the positive response rate is too small to outweigh the benefit of screening asymptomatic subjects using imaging tests. However, thanks to Honda and his team, this programme can now consider the use of apoA2-i testing, with a biomarker such as apoA2-ATQ/AT, alongside non-invasive or minimally invasive imaging modalities to enable the early detection of PC. Hopefully, this means that early-stage PC and its risk factors could be caught earlier by blood tests, with secondary examinations using precise, but expensive, imaging tests only where needed. The researchers' approach offers the foundation for an efficient and cost-effective PC screening programme, which could beat back PC mortality rates with a breakthrough blood test.

Behind the Research

Professor Kazufumi Honda

E: k-honda@nms.ac.jp **T:** +81 3 5814 6835 **W:** www.nmsbiomarker.com/english.html

Research Objectives

Chief Professor Kazufumi Honda aims to isolate blood biomarkers that can be used to detect early-stage pancreatic cancer (PC), enabling the identification of individuals at high risk of developing PC or with associated disease symptoms.

Detail

Address
Chief Professor Kazufumi Honda
1-1-5 Sendagi, Bunkyo-ku, Tokyo 113-8602, Japan

Bio
Kazufumi Honda DDS, PhD, is Chief Professor at the Department of Bioregulation, Graduate School of Medicine, Nippon Medical School, Japan. From 2019 to 2021, Honda was Chief of the Division of Biomarkers for Cancer Early Detection at the National Cancer Center Research Institute (NCCRI), Laboratory Head of NCCRI between 2005 and 2019, and technical officer of Ministry of Health, Labour and Welfare, Japan.

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Collaborators
• Toray Industries, Inc.

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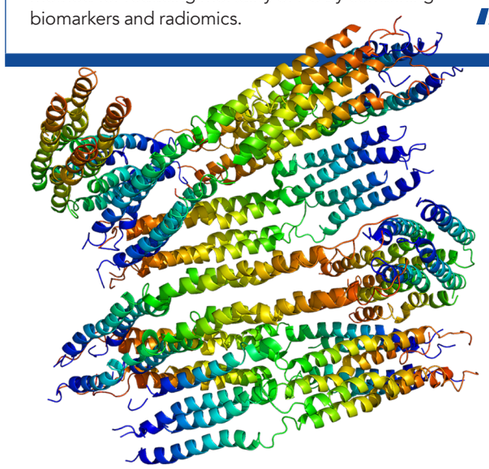
Personal Response

What are your plans for future research? Will you investigate how to improve the accuracy rates of apoA2-i testing?

"When we consider the incidence of PC in the general population, we should identify the most appropriate population for efficient PC screening. We have started to examine a large-scale prospective study for PC screening using apoA2-i blood tests to detect PC and its HRIs in Japan. From this result, we will be able to identify the clinical characteristics of apoA2-i for PC and its HRIs. Ultimately, we hope to develop an effective screening method for reducing mortality of PC by combining biomarkers and radiomics."



Protein apoA2-i. Photo Credit: Emw, CC BY-SA 3.0 , via Wikimedia Commons