ASX Release
7 February 2022

ASX code: PIQ

Novel biomarkers identified for Asthma and COPD

- **Proof-of-concept study identifies multiple protein biomarkers with potential to test for obstructive airway disease, including asthma and COPD**
- **Research is part of a collaboration with the Busselton Population Medical Research Institute to improve diagnosis of obstructive airway diseases**
- **Biomarkers identified in a pilot study of 75 individuals**
- **Proteomics International to file a new patent application for the invention covering screening, diagnostic and prognostic methods of using the airway disease biomarkers**
- **New biomarkers highlight the power of Proteomics International's Promarker™ technology platform to develop novel diagnostics tests in areas of significant unmet medical need**

Proteomics International Laboratories Ltd (Proteomics International; ASX: PIQ) is pleased to announce its proof-of-concept study has identified multiple novel protein biomarkers for obstructive airway disease. These biomarkers, once validated, have the potential to deliver a new diagnostic test for asthma and chronic obstructive pulmonary disease (COPD). Proteomics International will now file a patent application covering screening, diagnostic and prognostic methods of using these airway disease biomarkers.

The results of the study were presented at the 27th Lorne Proteomics Symposium, Victoria, the annual conference of the Australasian Proteomics Society (see copy of presentation attached, titled: *Protein Biomarkers of Obstructive Airway Disease*). The results showed multiple protein biomarkers were statistically significant in identifying specific types of airway disease.

Obstructive airway disease is a heterogeneous condition that includes asthma, COPD and bronchitis with many common symptoms including breathing difficulty, chronic cough, and respiratory infection. Diagnosis is often subjective and can rely upon clinical judgement rather than objective findings, with early and more accurate diagnosis needed for improved patient outcomes.

The proof-of-concept study was performed in collaboration with the Busselton Population Medical Research Institute, and analysed plasma samples from 75 individuals with a range of symptoms including airway obstruction, atopy, bronchial hyper-responsiveness and healthy controls.

Clinical Professor Alan James, a consultant respiratory physician and chair of the Busselton Population Medical Research Institute, said doctors currently use physiological tests to diagnose and assess severity of airway disease. "*These tests of lung function and structure are useful in investigating common symptoms such as cough and breathlessness. However they do not necessarily reflect the various underlying pathologies which cause abnormal structure and function and which may respond differently to different treatments.*"

Once the disease is advanced, interventions are less effective because damage to the lungs from COPD cannot be reversed with current treatments. "*Accurate and early identification of these common conditions and differentiation of phenotypes (sub-types) of airway disease can allow early intervention with directed therapy, resulting in improved patient outcomes*," Professor James said.

In Australia, asthma affects 11% of the total population, whilst COPD affects 4.8% of adults over 45 years old - approximately 3.2 million people in total - and collectively asthma and COPD cost the Australian health system an estimated $1.7 billion in 2016[1]. Worldwide, COPD is the third leading cause of death, causing 3.2 million deaths in 2019[2].

The Company will now work with its collaborators to validate the biomarkers in larger clinical cohorts and refine the panel of biomarkers into a working diagnostic test for obstructive airway disease. This additional analysis is expected to be performed over the next 12-18 months.

Proteomics International managing Director Dr Richard Lipscombe said these latest biomarker results further highlight the power of the Company's Promarker™ technology platform, and build upon our experience acquired in developing the PromarkerD assay, the world's first test for predicting diabetic kidney disease. *"The recent success validating biomarkers for oesophageal cancer (ASX: 4 February) and the clinical validation study progressing for an endometriosis diagnostic (ASX: 4 August) are part of a rich pipeline of novel diagnostic tests in development. These all target areas of significant unmet medical need and each has the potential to generate significant value for our Company."*

Authorised by the Board of Proteomics International Laboratories Ltd (ASX.PIQ).

ENDS

About Busselton Health Study (www.bpmri.org.au)
The Busselton Health Study is one of the longest running epidemiological studies in the world. Since 1966, over 30,000 residents of Busselton, located in Western Australia's south-west, have participated in surveys concerning such health topics as cardiovascular disease, pulmonary function, diabetes, and allergies, resulting in over 400 academic publications. The program is administered by Busselton Population Medical Research Institute. The Busselton Health Studies have been described as a national treasure and continue to provide important information about the prevalence and lifestyle risk factors of many common diseases and health conditions in a general Australian population. Current research activities continue to explore multimorbidity, healthy ageing, genetic causes of disease, mental health and Covid-19, and the prevalence of specific types of airway diseases in the community.

About the Promarker™ Platform
Proteomics International's diagnostics development is made possible by the Company's proprietary biomarker discovery platform called Promarker, which searches for protein 'fingerprints' in a sample. This disruptive technology can identify proteins that distinguish between people who have a disease and people who do not, using only a simple blood test. It is a powerful alternative to genetic testing. The technology is so versatile it can be used to identify fingerprints from any biological source, from wheat seeds to human serum. The Promarker platform was previously used to develop PromarkerD, a world-first predictive test for diabetic kidney disease, that is currently being commercialised.

About Proteomics International Laboratories (PILL) (www.proteomicsinternational.com)
Proteomics International (Perth, Western Australia) is a wholly owned subsidiary and trading name of PILL (ASX: PIQ), a medical technology company at the forefront of predictive diagnostics and bio-analytical services. The Company specialises in the area of proteomics – the industrial scale study of the structure and function of proteins. Proteomics International's mission is to improve the quality of lives by the creation and application of innovative tools that enable the improved treatment of disease.

[1] Australian Institute of Health and Welfare (AIHW) 2020, www.aihw.gov.au/reports/chronic-respiratory-conditions/asthma

[2] WHO, www.who.int/news-room/fact-sheets/detail/chronic-obstructive-pulmonary-disease-(copd)

For further information please contact:

Dr Richard Lipscombe
Managing Director
Proteomics International Laboratories Ltd
T: +61 8 9389 1992
E: enquiries@proteomicsinternational.com

Dirk van Dissel
Investor Relations & Corporate Advisor
Candour Advisory
T: +61 408 326 367
E: dirk@candouradvisory.com.au

Kyle Moss
Corporate Advisor
Euroz Hartleys
T: +61 8 9488 1400
E: kmoss@eurozhartleys.com

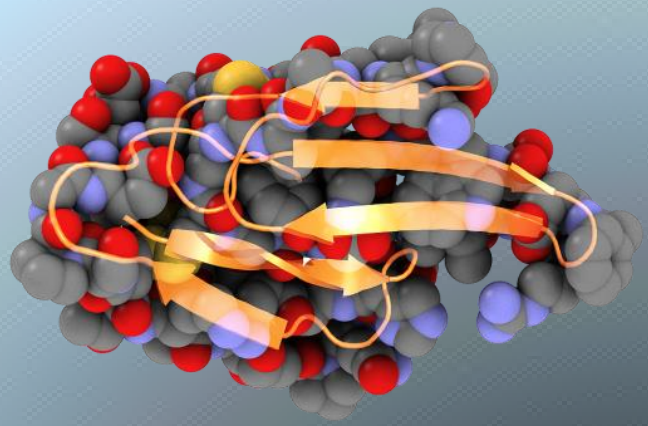
Lorne Proteomics 2022

e-Poster | February 3rd – 6th 2022

Protein Biomarkers of Obstructive Airway Diseases

Jason Ito [1] , Scott Bringans [1] , Lianzhi Chen [1] , Tasha S. Lumbantobing [1], Alan L. James [2] , Bill A.W. Musk [2] , Jennie Hui [2] , Richard J. Lipscombe [1]

1. *Proteomics International, Nedlands, WA, Australia*
2. *Busselton Population Medical Research Institute, Bussleton, WA, Australia*



- **Background:** Obstructive airway disease is heterogenous (including asthma, chronic obstructive pulmonary disease and bronchitis) with many overlapping symptoms. Diagnosis is often subjective, early and more accurate diagnosis needed for improved patient outcomes.

- **Hypothesis:** Plasma from individuals with obstructive airway diseases (Busselton Health Study Cohort) are stratified by quantitative variables, rather than clinical groupings; these samples may contain proteins at significantly different concentrations when compared with control plasma from healthy individuals.

- **Objective:** Use iTRAQ quantitative proteomics to discover plasma protein biomarker candidates of airway obstruction diseases.

Study design (iTRAQ 4-plex)

Proteomics International

Pooled Group 1 (n=15) Non-Atopic Airflow Obstruction

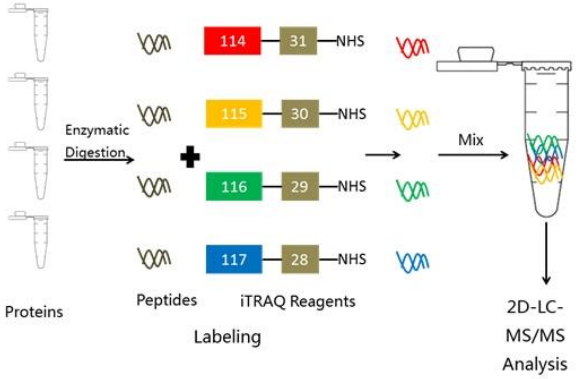
Pooled Group 2 (n=20) Atopic with High eNO

Pooled Group 3 (n=20) Atopic with BHR

Pooled Group 4 (n=20) Healthy Controls

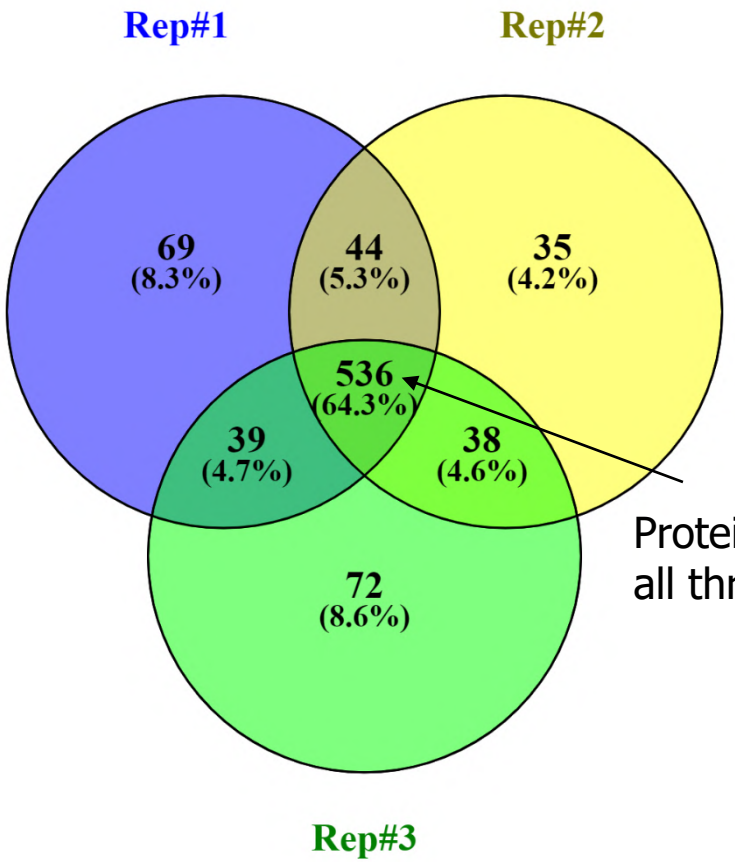
Plasma sample preparation and MS analysis

- Immunodepleted plasma proteins are reduced + alkylated + digested with trypsin
- iTRAQ 4-plex labeling of peptides (3x technical replicates)
- Thermo Q Exactive HF mass spectrometer with Thermo U3000 nano UHPLC
- Protein Pilot Ver. 5.0 software used for iTRAQ MS data analysis



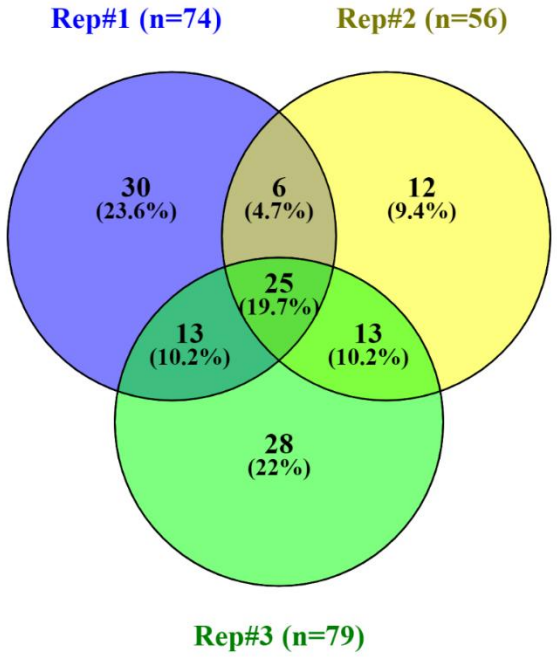
iTRAQ results – Total Protein Identification

	Replicate 1	Replicate 2	Replicate 3
# of total spectra	354,028	352,793	353,382
# distinct peptides	34,858	35,036	35,941
# identified Proteins	688	653	685

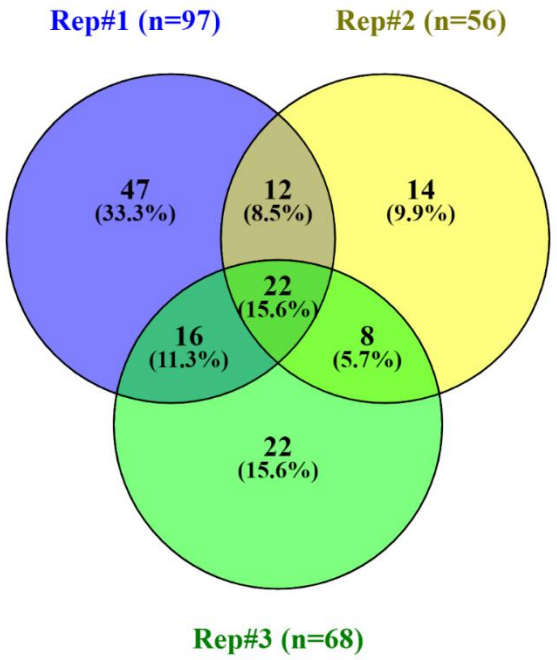


Proteins common to all three replicates

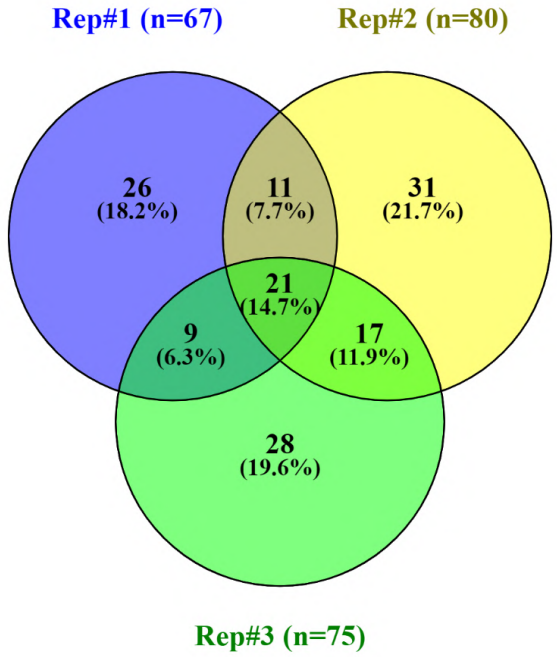
iTRAQ results – Differential Expression



Non-Atopic Airflow Obstruction vs. Healthy Control



Atopic with High eNO vs. Healthy Control



Atopic with BHR vs. Healthy Control

- *p*-value of <0.05 was considered significant for differential expression.
- Fold change of 10% or more was a requirement.
- All proteins considered had 2 or more supporting peptides.

Conclusions

- Triplicate iTRAQ experiments had robust and reproducible sets of differentially expressed biomarker candidates for non-atopy, atopy + high eNO, or atopy + BHR.
- Candidate biomarkers to be validated with cohort analysis by targeted mass spectrometry assays (i.e., multiple reaction monitoring-MRM).
- Validation by MRM can lead to developing a clinical test for early diagnosis and precise treatment for obstructive airway diseases.