

**VERSION 2** 

DEC 20, 2023

# OPEN ACCESS



#### DOI:

dx.doi.org/10.17504/protocol s.io.j8nlkoy56v5r/v2

Collection Citation: ronan.o cualain 2023. Modular automated sample processing of biological samples using ultrasonication and a workstation for high-throughput proteomics.

#### protocols.io

https://dx.doi.org/10.17504/p rotocols.io.j8nlkoy56v5r/v2Ve rsion created by ronan.ocualain

License: This is an open access collection distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Modular automated sample processing of biological samples using ultrasonication and a workstation for high-throughput proteomics V.2

ronan.ocualain<sup>1</sup>

<sup>1</sup>University of Manchester

protocols.io Ambassadors

BioMS CRF, UoM



## ronan.ocualain

#### **ABSTRACT**

Sample preparation for mass spectrometry analysis involves **numerous** liquid transfer steps.

These include

- sample lysis,
- protein extraction,
- solubilisation.
- estimation,
- reduction and alkylation,
- normalisation,
- clean-up,
- enzymatic digestion,
- and desalting.

Adapting these steps onto an automated workstation can increase efficiency, throughput, and reduce coefficients of variance (%CV) thereby providing reliable reproducible data for statistical comparisons.

This protocol is part of a modular collection for the processing of biological samples for proteomics.

The entry point is ultrasonication of biological samples (cells, tissues, laser captured FFPE sections) using a plate based LE220+ system from Covaris, followed by sample processing with a Biomek NxP workstation.

Technical measurement of workflow percentage coefficients of variation (%CVs) using HeLa extract at the entry stage, to measurement of the ion intensities at the data processing stage with data collected by DDA using a QE-HF and processed using Progenesis-QI for proteomics has shown that one third of peptides have **%CVs** 

Oct 20 2023

**Protocol status:** Working We use this collection and it's working

below **20%**, and with 80% of peptides having %CVs below **30%**. Using Progenesis QI for proteomics indicates that 98% of peptides are found in all 7 replicates processed.

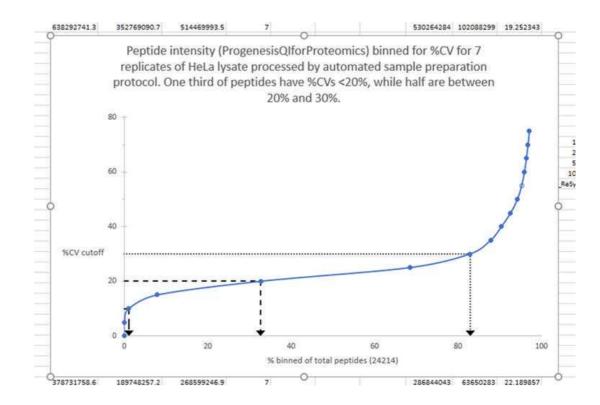
Created: Dec 20, 2023

Last Modified: Dec 20,

2023

**COLLECTION** integer ID:

92551



In addition, the whole procedure (80 samples) may be completed in a day, if shorter digestion times are utilised.



### **Protocol**



NAME

Automated 96 well plate based protein reduction and alkylation using a Beckman Biomek™ NxP workstation

VERSION 1

**CREATED BY** 



OPEN →

# **Protocol**



Automated 96 well plate based protein quantitation using a Beckman Biomek™ NxP workstation and a Pierce™ 660nm Protein Assay Kit

VERSION 1

**CREATED BY** 



OPEN →

## **Protocol**



Protein aggregation capture (PAC) and minimal automated processing for proteomics using magnetic beads and a Beckman Biomek™ NxP workstation for 96 well plates.

VERSION 1

**CREATED BY** 



OPEN  $\rightarrow$