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VAMAS TWA 2:
Project A19: Inter-laboratory study of the measurement of chemistry and thickness of nanoparticle coatings: Protocol for sample preparation.

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VAMAS TWA 2, 2015: Project A19: Inter-laboratory Study: Measurement of chemistry and thickness of nanoparticle coatings - Protocol for sample preparation.

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ABSTRACT

This document describes the protocol for sample preparation in the VAMAS TWA 2, 2015 study on the measurement of chemistry and thickness of nanoparticle coatings. Samples are supplied for this study, comprising a ready-deposited sample of peptide-coated nanoparticles on silicon wafer, in addition to a concentrated aqueous suspension of the same nanoparticle sample for preparation 'in-house'. An additional flat gold surface with ready-deposited self-assembled monolayer of the same peptides utilised to functionalise the nanoparticles will be provided as a reference sample for participants using LEIS and MEIS. Specific guidance is given on sample storage and handling.

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Approved on behalf of NPLML by Ian Gilmore, NPL Fellow..

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1 INTRODUCTION

High vacuum techniques such as X-ray Photoelectron Spectroscopy (XPS) and Low and Medium Energy Ion Scattering (LEIS and MEIS respectively) have shown promise for the accurate measurement of chemistry and average thickness of nanoparticle coatings. However, the accuracy of these measurements heavily relies upon (1) correct modelling to support data analysis and (2) quality and reproducibility of sample preparation for surface analysis. To date, there is no uniform approach to these two measurement issues and the nanoparticle community will highly benefit from this comparative study on these subjects.

Results from this inter-laboratory comparison will aid the development of a new ISO standard for the measurement of chemistry and average thickness of surface coatings of nanoparticles. Furthermore, the study will provide information of direct relevance for the ISO standard for reporting information related to the history, preparation, handling and mounting of nanomaterials prior to analysis which is currently in development under ISO TC201. The production of guidelines for the measurement of nanoparticle surface coatings and the handling of nanoparticle samples will be highly beneficial to those industries which rely on the high performance of nanoparticle coatings. Accurate knowledge of nanoparticle surface chemistry will also impact on the ability to perform accurate risk assessment of the materials, with important implications for the commercialisation and safe use of the products.

The specific objectives of this inter-laboratory study are: (i); assessment of inter-laboratory variability in measuring the thickness of nanoparticle coatings (ii) comparison of sample preparation techniques (iii) testing literature or ‘in-house’ procedures for quantitative analysis (optional).

If you are unsure of any part of this protocol please contact Natalie Belsey (natalie.belsey@npl.co.uk) or Alex Shard (alex.shard@npl.co.uk).

2 TIMETABLE

You should complete the analysis for this work by 6th March 2015. If you cannot do so and you need extra time please inform Natalie Belsey.

3 THIS PACKAGE

This package contains this protocol, a pre-deposited nanoparticle sample on silicon wafer in addition to a concentrated solution for ‘in-house’ sample preparation (a Viton rubber O-ring and a silicon wafer are provided for sample preparation). In addition, participants using LEIS or MEIS are provided with a peptide-functionalised flat gold substrate and a cleaned flat gold substrate as a reference. Please inspect the packaging to check if it has been opened by customs and if the integrity of the samples has been compromised; for example the Freeze Watch™ Indicator paper should remain white. If the paper has been stained black, or you are in doubt, please contact NPL. Upon receipt of the samples, please notify us that everything is received in good order.

☐ I have emailed NPL that all is OK with the sample(s) on / / 2015

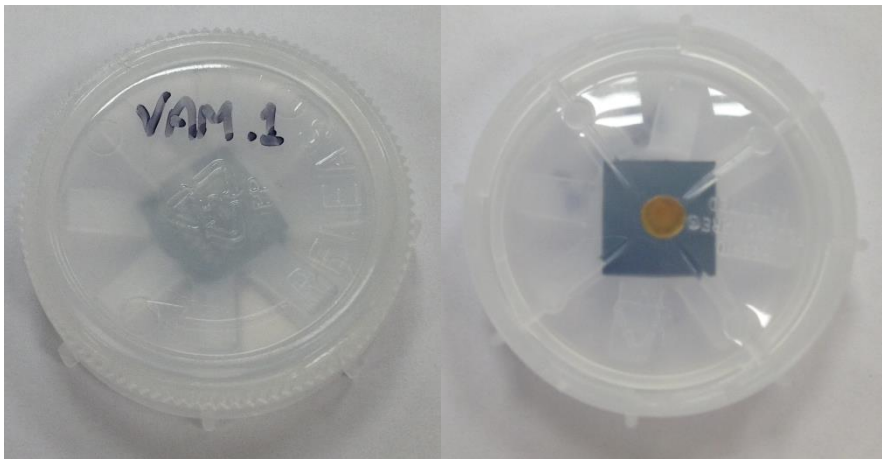
4 SAMPLES, HANDLING AND STORAGE

Three types of sample are provided:

1. Peptide-functionalised gold nanoparticles (59 nm diameter gold core) ready-deposited (dried) onto silicon wafer. The nanoparticles should appear as a circular gold area approximately 3 mm in diameter on the polished side of the silicon wafer.
2. A concentrated aqueous solution of the same peptide-functionalised gold nanoparticles for 'in-house' sample preparation. A pre-cleaned silicon wafer is included for use as a substrate, along with a rubber O-ring to aid sample deposition.
3. A peptide-functionalised flat gold surface and a cleaned flat gold substrate (for participants performing LEIS and MEIS measurements only).

The samples will be transported within sealed bags containing an argon atmosphere, and upon receipt, the whole unopened bag should immediately be placed directly into a refrigerator (2-5 °C) for storage. If the bag is opened, it can be simply re-flushed with argon gas and sealed before refrigerated storage. The samples must NOT be frozen under any circumstances. The samples are stable for at least 1 month under such conditions, but should be analysed as soon as is convenient within this timeframe.

When the samples are removed from the refrigerator for characterisation, please allow at least one hour for them to return to room temperature before opening the Fluoroware container. It is vital that the samples are kept clean and are analysed as soon as possible after the Fluoroware containers are opened. The samples are stored 'upside-down' within the Fluoroware containers, with the nanoparticles located on the side of the silicon wafer facing the concave bottom half of the container. A flexible plastic 'spider' is pressed against the back of the sample, holding it in place for transit, and will need to be carefully removed before the sample can be extracted. Samples should only be handled at their edge using cleaned metal tweezers held using powder-less polyethylene gloves. Vinyl gloves, often used in clean rooms, are coated with a release agent from the moulding process, and should not be used.



Above: Photograph of a sample oriented face-down within a Fluoroware container, as viewed from the top (left) and the underside (right).

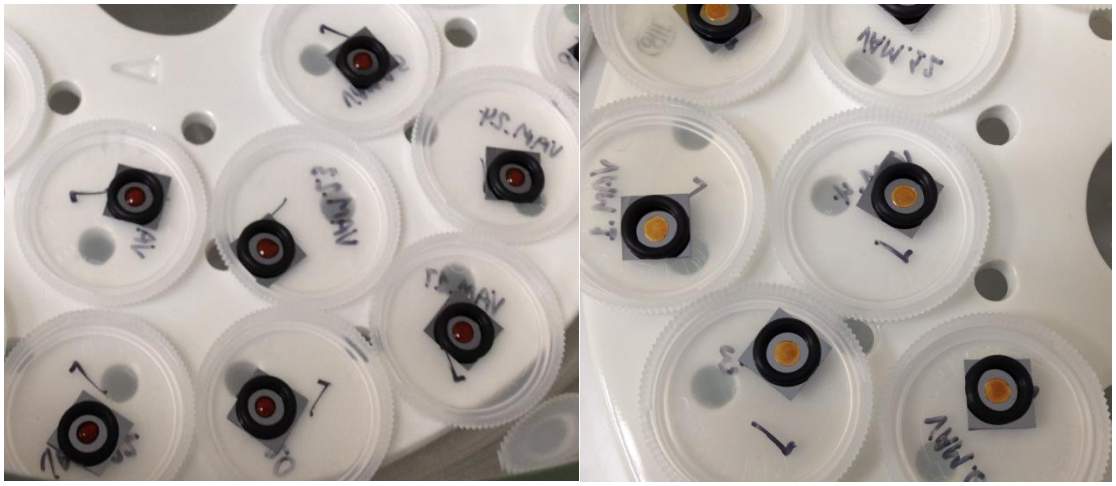
The silicon wafer sample with nanoparticles is 10 mm × 10 mm within which the sample spot is centrally located with a diameter of ~3 mm. Please avoid handling the coated area.
The peptide-functionalised gold coated silicon wafer sample (for MEIS and LEIS only) is 10 mm × 10 mm and coated uniformly. Please avoid handling the areas where measurement will be performed.

5 PROPOSED METHOD

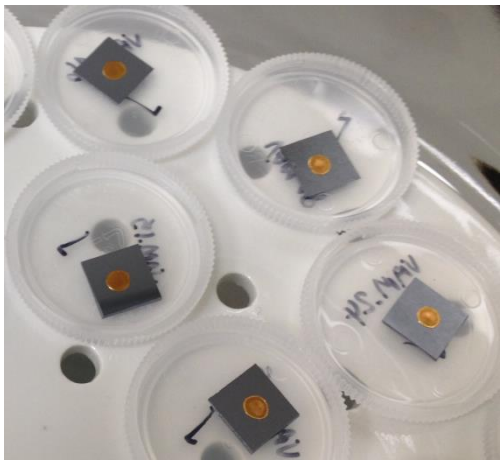
The following method was used to prepare the pre-deposited samples, and is provided as a suggested method for depositing the solution provided. If you have another method you prefer to use to deposit the nanoparticles, please feel free to do so, and describe your method in full within the data reporting spreadsheet.

5.1 METHOD FOR SAMPLE PREPARATION FROM CONCENTRATED SOLUTION.

The aqueous solution of nanoparticles should be applied to the silicon wafer supplied using the Viton rubber O-ring to reduce ‘coffee ring’ drying effects. We recommend placing the O-ring at the centre of the silicon wafer and applying a 3 µL aliquot of sample solution centrally, within the confines of the O-ring (without touching the sides). The O-ring serves to reduce differential evaporation rates, so do not simply fill the O-ring with liquid; the sample solution must not contact the O-ring. Allow the solution to dry before applying subsequent aliquots over the top of the dried residue. Continue this process until all of the solution has been utilised. This process may be performed in a desiccator to speed up the drying process, however please ensure the environment is clean and application/removal of vacuum is applied with extreme caution to avoid disturbing the deposited dried material.



Above: Photographs of nanoparticle solution during the deposition process - droplets should be placed within the O-ring as shown in the left picture. After multiple cycles of deposition and drying, a circular area of deposited nanoparticles as shown in the right picture should begin to be seen. Upon completing the deposition, carefully remove the O-ring, and the sample should appear as in the picture below.



For participants using MEIS: please use your best judgement with regard to the deposition process: The volume of material supplied is intended to produce a thick layer for XPS measurements, so in order to achieve the most suitable sample for MEIS (i.e. a monolayer), please feel free to use as much or little of the solution as you see fit. Please be sure to supply full details of your method including the volume of solution applied in the supplied spreadsheet for data reporting.

If analysis will not be performed immediately, the sample may be stored in an argon-filled plastic bag sealed and placed in a fridge.

6 INSTRUMENT OPERATING CONDITIONS

The instrument should be operated under conditions that give the most stable performance. For the optimum inter-comparison, a common set of operating conditions is needed, however due to the variety of instruments available, no single set of conditions can do this. Instead we give general conditions for guidance to improve comparability.

6.1 SAMPLE TEMPERATURE.

Analysis at room temperature is preferred. However, if there is a likelihood that samples will be heated during analysis, e.g. through the proximity of filaments and other heat sources, sample cooling is recommended. Under no circumstances should the samples be exposed to temperatures of 40°C or higher. If the temperature at which the sample is analysed differs significantly from 20°C, please give details in the 'conditions' section of the data reporting spreadsheet.

6.2 CHARGE COMPENSATION

It may be necessary to employ low energy electrons to compensate for sample charging that occurs during analysis. However, it is preferable to minimise exposure to electron irradiation especially if more than one analysis is performed on the same sample. Where possible, please report the duration of any prior exposure to electron irradiation (as well as present charge compensation details) for each analysis undertaken.

6.3 XPS ANALYSIS

Ideally, XPS analysis should be performed in triplicate by analysing three non-overlapping regions within the sample spot. Where this is not possible due to instrumental constraints (i.e. x-ray beam area size, analysis area size), selecting regions with minimal overlap is preferred. Please indicate the order in which regions were analysed, and where possible, if a region has already been exposed to x-rays before analysis, please note the extent of prior exposure in the 'conditions' section of the data reporting spreadsheet. For the most accurate measurement of thickness, areas should be chosen that show no or minimal signal from the silicon substrate. Please allow a suitable 'warm-up' time for your x-ray source prior to any analysis, to ensure stable beam intensity throughout.

For each sample a 'survey' or 'wide' scan should be acquired followed by narrow scans for the following elements: Au 4f, C 1s, O 1s, N 1s, S 2p, Si 2p.
Please use your judgment with regard to settings; but as a guide the following settings were used for analysis on a Kratos Axis Ultra spectrometer: a pass energy of 160 eV with a 1000 meV step size and 300 ms dwell time for survey scans. For narrow scans: a 40 eV pass energy with a step size of 100 meV and a dwell time of 500 ms. One sweep was sufficient for survey scans; additional sweeps were performed for narrow scans.

The data should be reported as raw intensities without the application of an instrumental transmission function and/or sensitivity factors, and also as atomic% using your standard procedures. If any other elements are detected, please contact Natalie Belsey or Alex Shard immediately.

6.4 LEIS/MEIS ANALYSIS

Ideally, analysis should be performed in triplicate by analysing three non-overlapping regions within the sample spot. Where instrumental constraints prevent this, selecting regions with minimal overlap is preferred (please note the order in which the regions were measured). Please use your best judgement to select parameters such as beam energy, current area of analysis, and give as full a description of these as possible in the reporting spreadsheet.

7 DATA AQCUISITION

7.1 USEFUL DATA

Examples of both a survey/wide scan and a carbon 1s narrow scan for a sample of nanoparticles deposited on silicon are shown below as a guide.

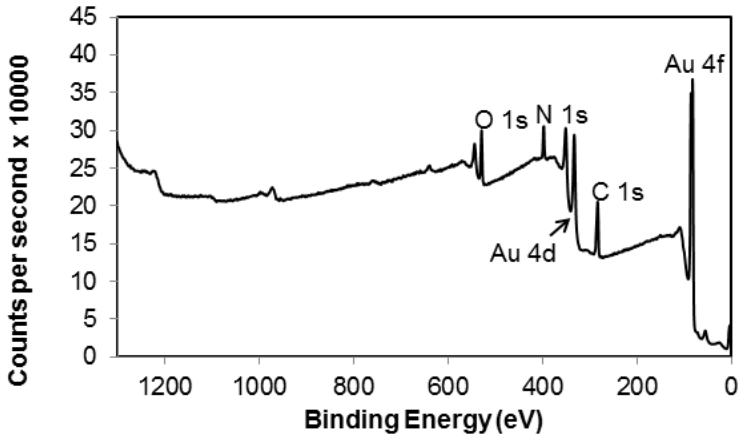


Figure 1 : Example survey/wide spectrum of nanoparticles deposited on silicon wafer.

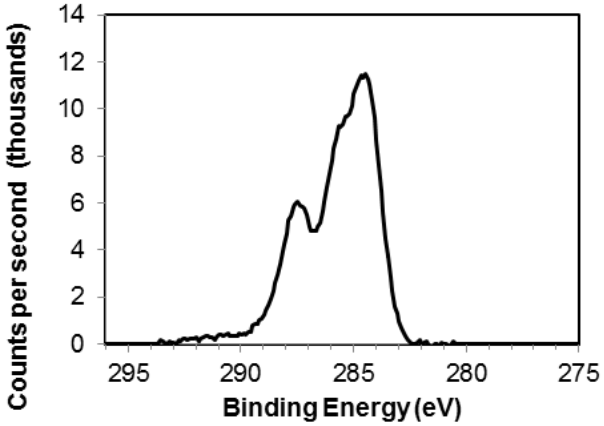


Figure 2 : Example Carbon 1s narrow scan from nanoparticles deposited on silicon wafer.

In addition to the suggestions for the narrow scan elemental analysis, please report any additional elements detected in the sample.

7.2 DATA REPORTING

An Excel electronic reporting form is supplied as part of this study. Please report the details of your analysis conditions and methods, alongside your results, in the appropriate section of the spreadsheet. Please also return raw data in .vms format, with the filename for each dataset clearly noted in the 'data summary' part of the data reporting spreadsheet.

7.3 DATA INTERPRETATION

Use your own in-house procedures to calculate the thickness of the peptide coating. The diameter of the gold core of the nanoparticles (as measured by the manufacturer) is 59 nm. Please report your results and the method you used.

8 AFTER ANALYSIS

When all analyses are complete, please return the samples to NPL at your earliest convenience.

9 CONFIDENTIALITY

The samples supplied in this inter-laboratory study are not certified reference materials, but have been produced to the best of our abilities. They are sent to you in confidence, and if there are any problems with them we ask that you contact us immediately so that we can determine whether the problem is generic or restricted to a single batch or sample. Please do not to publish your individual results, or any further or different analyses of these materials without consulting or informing NPL. If, for commercial reasons, you do not wish to be identified in our final report, please note this in your report.

10 ACKNOWLEDGEMENTS

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