TITLE OF THE PROTOCOL: RAMAN SPECTROSCOPY AND COLORECTAL CANCER: TOWARDS EARLY DIAGNOSIS AND PERSONALISED MEDICINE

Short title/Acronym: Raman spectroscopy and colorectal cancer

Sponsor:

Representative of the Sponsor:
Jemma Hughes
R&D Manager
R&D Department
Morriston Hospital
Swansea
SA6 6NL

REC reference: 14/WA/0028

STUDY SUMMARY/SYNOPSIS

TITLE	Raman spectroscopy and colorectal cancer: towards early diagnosis and personalised medicine
SHORT TITLE	Raman spectroscopy and colorectal cancer
Protocol Version Number and Date	ABM/RS/v4 Version 4;08/2/2017
Methodology	Prospective cohort observational study
Study Duration	5 years 6 months
Study Centre	ABM University LHB and Swansea University
Objectives	To explore the feasibility of detecting presence of differing stages of colorectal cancer at different phases of treatment using Raman spectroscopy
Number of Subjects/Patients	1,800 across 8 study groups
Main Inclusion Criteria	Stage T3/4 colorectal cancer (group 1) n=300 T1/2 early colorectal cancer (T1/2) from Bowel Screening Wales programme (group 2) n=300 Benign colorectal polyps (>1cm size)(group 3) n=200 Matched controls (without history of inflammatory bowel disease or familial bowel cancer)(group 4) n=300 Locally advanced rectal cancer requiring chemoradiotherapy (group 5) n=80 Bowel cancer patients at high risk of developing metastatic disease (group 6) n=20 Patient s diagnosed with other cancer types (e.g. lung, pancreas)(group 7) n=300 Patients in primary care with colorectal symptoms (group 8) n=300
Statistical Methodology and Analysis	Principal Components Analysis (PCA) and hierarchical cluster analysis to measure accuracy, sensitivity and specificity measures for each patient group.

Protocol Agreement Page

The clinical study as detailed within this research protocol **(Version 2, dated 9th May 2014)**, or any subsequent amendments will be conducted in accordance with the Research Governance Framework for Health & Social Care (2005), the World Medical Association Declaration of Helsinki (1996) and the current applicable regulatory requirements and any subsequent amendments of the appropriate regulations.

Chief Investigator Name: Prof. Dean A Harris (Consultant Surgeon)

Chief Investigator Site: Singleton Hospital, Swansea

Contact details: Tel No: 01792 285459

Email: Dean.A.Harris2@wales.nhs.uk

Fax:

Signature and Date: …….

Statistician Name: Prof Greg Fegan, Swansea Trials Unit

Contact details: Tel No:

Email:g.fegan@swansea.ac.uk

Fax:

Signature and Date: …….

Principal Investigator Name: Dr Peter Dunstan, Associate Professor

Principal Investigator Site: Swansea University

Contact details: Tel No: 01792 513052

Email: p.r.dunstan@swan.ac.uk

Fax:

Signature and Date: …….

Co investigator: Prof Gareth Jenkins, Prof. of Molecular Carcinogenesis

Investigator Site: Institute of Life Sciences, Swansea University

Contact details: Tel No:01792602512

Email: g.j.jenkins@swansea.ac.uk

Signature and date:.....

Co investigator: Miss Cerys Jenkins, PhD student

Investigator Site: Swansea University

Contact Details: Email c.jenkins@swansea.ac.uk

Signature and date:.....

Co investigators: Prof John Beynon, Consultant General and Colorectal Surgeon, Honorary Clinical Professor, ABMU LHB and Swansea University

Investigator Site: Singleton Hospital

Contact details: Tel No: 01792 285040

Email: john.beynon@wales.nhs.uk

Signature and Date: …….

Co investigators: Mr Martyn Evans, Consultant General and Colorectal Surgeon, ABMU LHB

Investigator Site: Singleton Hospital

Contact details: Tel No: 01792 703579

Email: martyn.evans@wales.nhs.uk

Signature and Date: …….

Co investigators: Mr Mark Davies, Consultant General and Colorectal Surgeon.

Co Investigator Site: Singleton Hospital, Swansea

Contact details: Tel No: 01792 705209

Email: Mark.Davies8@wales.nhs.uk

Fax:

Signature and Date: …….

Co investigators: Dr. Heather Wilkes, General Practitioner and Macmillan Lead GP ABMU.

Co Investigator Site: Briton Ferry Health Centre, Briton Ferry

Contact details: Tel No: 07834 195509

Email: Heather.Wilkes@wales.nhs.uk

Signature and Date: …….

Glossary of Terms and Abbreviations

AE	Adverse Event
AR	Adverse Reaction
ASR	Annual Safety Report
CI	Chief Investigator
CRF	Case Report Form
CRO	Contract Research Organisation
EC	European Commission
GAfREC	Governance Arrangements for NHS Research Ethics Committees
ICF	Informed Consent Form
JRO	Joint Research and Development Office
MS	Member State
Main REC	Main Research Ethics Committee
NHS R&D	National Health Service Research & Development
PI	Principle Investigator
QA	Quality Assurance
QC	Quality Control
Participant	An individual who takes part in a clinical trial
REC	Research Ethics Committee
SAE	Serious Adverse Event
SDV	Source Document Verification
SOP	Standard Operating Procedure
SSA	Site Specific Assessment

Index **Page Number**

1. Introduction

1.1 Background

Colorectal cancer (CRC) remains one of the leading causes of cancer related death in the Western world. Early diagnosis, the principle behind the UK bowel cancer screening programme, is pivotal to optimise outcomes. Current screening methods are invasive, have low specificity for cancer and do not have widespread patient acceptance. As such there is a pressing need to develop alternative non-invasive acceptable methods of screening for bowel cancer.

There are 14,000 new cases of rectal cancer each year in the UK, many being treated by combined chemotherapy and radiotherapy (RT) to reduce tumour size before surgery. Response to chemoradiotherapy is highly variable and some patients do not respond. Such patients endure the side effects of treatment, including poor bowel function, incontinence, sexual dysfunction and impaired quality of life, without clinical gain.

Whilst high levels of pathological tumour regression in response to RT is desirable and is associated with significantly improved disease free survival (Rödel 2005), in practice a spectrum of response is observed, ranging from complete pathological response to disease progression through treatment resistance. The broad application of RT additionally exposes patients with radiation-resistant disease to long term complications, which includes incontinence, sexual dysfunction and impaired wound healing. An accurate biologically based method is needed to stratify patients in predicting response to RT and rationalising it to those set to experience clinical gain, with alternative treatments for predicted non-responders.

Plasma and faeces of colorectal cancer patients is known to contain various proteomic and metabolomic markers together with circulating tumour cells (CTCs) characterised by specific surface markers. Faecal samples are known to contain shed CRC cells and mutated DNA (Calistri 2010). Equally, tissue biopsies from a variety of human malignancies have been demonstrated as having unique spectra which can be exploited for in-situ diagnosis (Mavarani, 2013).

Raman spectroscopy is an established technology based upon the spectroscopic analysis of inelastically scattered light for changes that reflect the molecular and structural composition of the biological sample under investigation. The resultant Raman spectrum directly reflects the molecular composition of the interrogated sample which may be considered unique for individual disease states, including certain cancer types. Raman therefore has potential not only as a diagnostic tool for the identification of malignancy, but also for personalised cancer medicine, disease monitoring and even nanoparticle-targeted delivery of chemotherapeutics.

This exploratory work will begin to establish a signature based on the molecular composition of different stages of colorectal carcinogenesis. The likely benefits of the proposed research to patients include a more acceptable means of screening for colorectal cancer. Present unmet needs affecting surgical and oncological treatment of CRC patients is the need for improved prognostication to allow personalised treatment of CRC, for example to select patients for adjuvant chemotherapy after surgical resection based on persistent plasma markers of cancer activity, and to predict the response of locally advanced rectal cancer to chemoradiotherapy.

As colorectal cancer is such a high profile condition, positive results will be of enormous public interest, particularly if a simple blood test could predict an individual's risk of having bowel cancer. As such the study will enrol patients from primary care with colorectal symptoms/signs towards reducing the requirement for invasive diagnostic tests.

1.2 Preclinical Data

Early reports have suggested specific Raman parameters in the serum of colorectal cancer patients (Li , Applied Optics 2012), whilst Lin et al (Lin, Optics Express 2011) have had some initial successes at implementing gold nanoparticle enhancement of Raman from blood serum. Circulating stem cells (CSCs) and tumour cells (CTCs) are both present in the peripheral blood of patients at all stages of malignancy, and can be characterised by both specific surface markers (EpCAM, CD44, MUC-1, CK20, CEA) and mutated driver genes (KRAS, BRAF, PIK3CA). It is reported that SERS nanoparticles functionalized with epidermal growth factor receptor (EGFR) affibodies preferentially target EGFR-positive cell line tissues (Jokerst 2011).

Raman scattering suffers from the disadvantage of poor scattering efficiencies due to the small scattering cross-section. This problem can be eased by the use of nanoparticles to act as enhancing agents. The very recent literature is exploiting the effects of nanoparticles at producing enhanced spectra to refine detection sensitivity and specificity.

1.3 Rationale and Risks/Benefits

Blood sampling will involve an invasive blood test with has the attendant risks of pain, local bruising and infection. These risks will be minimised as blood sampling will be done by experienced practitioners using aseptic technique.
A faecal sample will be collected by the patient which may cause intrusion or distress. This will be minimised as the patient can collect the sample in the privacy of their own home.

As this is an exploratory study research participants will not gain personal benefit.

2. Trial Objectives and Design

2.1 Trial Objectives

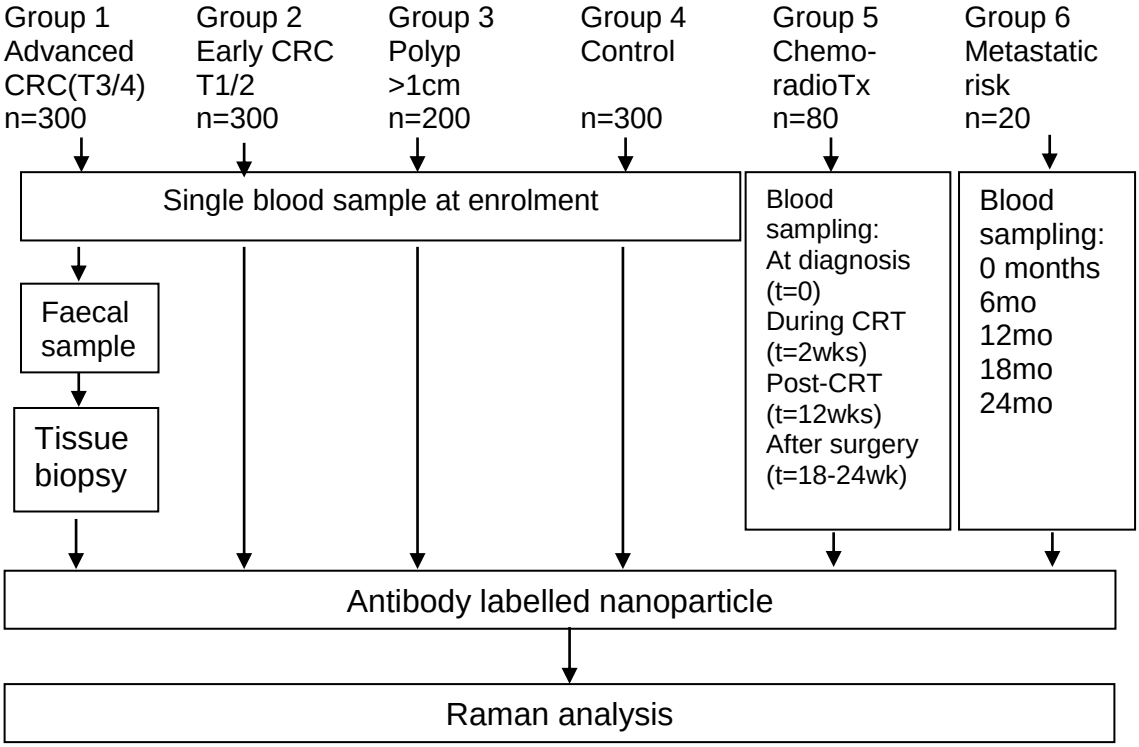
Primary Objective – To determine if there is an association between Raman spectral signature and presence of colorectal malignancy.

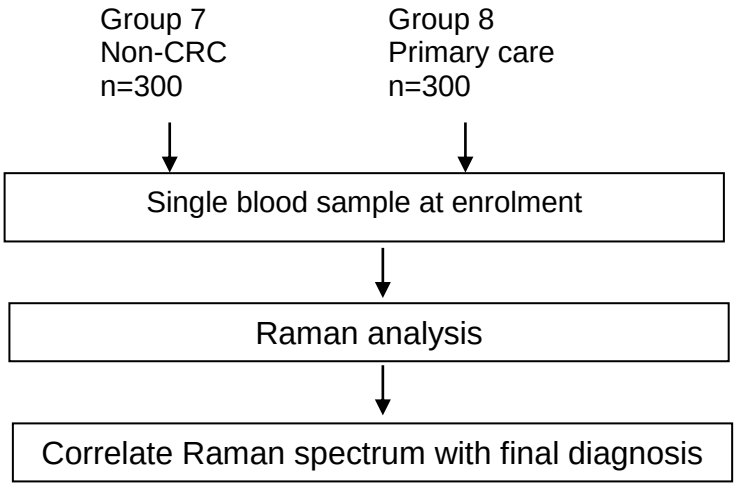
Primary Endpoint-Detection of surface enhanced nanoparticles in samples from colorectal cancer patients

2.2 Trial Design

Cohort observation study

2.3 Study Scheme Diagram





3. Subject Selection

3.1 Number of Subjects and Subject Selection

Paired plasma, faecal samples will be obtained from 300 patients with advanced colorectal cancer (stage T3/4) prior to surgical treatment (group 1). Plasma samples only will be obtained from 300 early colorectal cancers (T1/2) from the Bowel Screening Wales programme (group 2), 200 patients with benign colorectal polyps (>1cm size)(group 3) and 300 matched controls (without history of inflammatory bowel disease or familial bowel cancer)(group 4). The programme of work will be expanded to include 80 patients with locally advanced rectal cancer requiring chemoradiotherapy (group 5) who will have samples taken at diagnosis,during and after chemoradiotherapy and after resection of their tumour to identify changes in the spectra at each stage of treatment. Spectra can be compared with pathological factors, including tumour regression grade as a response to chemoradiotherapy, to establish if spectral analysis before CRT can predict the degree of downstaging seen. A further group of interest will be those at high risk of developing metastatic disease (group 6), for example colorectal cancers of advanced stage demonstrating adverse pathological features such as lymphovascular invasion or lymph node metastases. These twenty patients will have serial blood tests performed at five time-points over a 2 year follow up period, in parallel with standard investigations such as radiological imaging and tumour marker analysis. This aims to detect spectral change which predates development of liver or lung metastases on standard imaging modalities. This would have the potential of diagnosing metastatic disease at a pre-clinical stage affording earlier access to treatment. To establish if the spectral signatures are specific to colorectal cancer, a group of 300 patients diagnosed with stage-matched non-colorectal cancers (for example, lung cancer, pancreatic cancer, prostate cancer, endometrial cancer) will be studied with a single blood test for comparison.
Towards utilising the blood test as a means of discriminating between patients with colorectal cancer and those without we wish to obtain blood samples from 300 patients in primary care with colorectal symptoms and compare the test result (positive, equivocal or negative) with their final diagnosis (colorectal cancer or benign pathology). The blood test results will not be revealed to the GP or the patient so cannot influence the current referral pathway until the blood test is validated for this patient population. We will use this data to plan a subsequent interventional study.

3.2 Inclusion Criteria

Age 18 or over
Histological confirmation of colorectal cancer or adenoma (group specific) or in group 8 symptoms raising suspicion of colorectal cancer (anaemia, rectal bleeding, alteration in bowel habit, weight loss, abdominal pain and over the age of 50)
Willingness to consent
Willing to return to enrolling medical site for all study assessments

3.3 Exclusion Criteria

Under age 18
Patients with genetic conditions associated with colorectal cancer (Lynch syndrome and familial adenomatous polyposis)
Patients with inflammatory bowel disease
Unwilling/ unable to consent to trial participation
Patients from vulnerable groups

3.4 Criteria for Premature Withdrawal

If the participant withdraws consent for the study then their samples will be removed from the analysis according to the patient's wishes.

4. Study Procedures

4.1 Informed Consent Procedures

Written informed consent will be obtained by named doctors of the research team after sufficient time is allowed to read and understand the study with assistance from the written patient information sheet. A specific consent form is included for the group 8 primary care patient group.Please refer to appendices 2-6 to see the separate patient information booklets.

Sufficient time will be allowed for the potential participant to decide to take part in the study. It is recognised that in most circumstances patients may prefer to provide the blood sample at the same time as the screening and consent visit, particularly if routine care blood tests are required at the same time. Investigators must ensure that the potential participant does have sufficient time to consider study inclusion however. A copy of the consent form will be kept in the patient's case notes. Patient's ongoing willingness to participate in the study will be recorded each time an additional blood test is taken (groups 5 and 6 only).

4.2 Screening Procedures

Potential participants from groups 1,2,5 and 6 will be identified from colorectal cancer MDT meeting by their usual clinician who will be a named co-investigator. Patients in groups 3, 4 and 7 will be identified from attendance at routine surgical outpatient clinic or ward in-patient encounter by the co-investigators. Patients from group 8 will be identified by their usual GP at the time of first presentation.

Patients' records will be screened by the potential participant's existing clinical care team to establish if inclusion/exclusion criteria are met.

4.3 Schedule of assessment for each visit

Blood sampling- a 5mL blood sample will be obtained from the participant. Serum will be obtained by centrifugation at 3,000 rpm for 10 min and stored at 4°C until analysed. Alternatively the serum may be stored at -80 degrees C until analysis.

Faecal samples will be collected by participants onto the spatula within in a 5mL faecal collection universal container. Samples will be stored at 4°C and will be analysed within 3 days to prevent degradation. Samples will be serially diluted prior to analysis to optimize wavelength detection.

Samples will be subject to Raman analysis with the aim of optimising excitation wavelength and spectral reproducibility. The Raman system (InVia Renishaw) is capable of both 785 nm and 532 nm studies. Comparisons across different patient groups will help determine the best wavelength conditions for reliable data acquisition, sample handling and data reproducibility. Unlike previous published studies, laser power degradation effects on the sample will be tested, as reproducibility is key in identifying the origins of spectral differences from the patient groups. To overcome the inefficiency of Raman scattering it is proposed that through using multiplexed surface-enhanced Raman scattering (SERS) gold nanoparticles functionalised to specific surface markers a unique spectral pattern will be identified. We will analyse blood, faeces and the reference biopsy sample with combinations of functionalised Ag-nanoparticles (labelled with EpCAM, CD44, MUC-1, CK20, CEA) and analyse the resultant spectral peaks and be compared to positive controls. These tests will give unique insight into the potential diagnostic ability of Raman spectroscopy for colorectal cancer.

To optimise the nanoparticle functionalisation and SERS detection an initial programme of work will study the blood, tissue and faecal samples from patients with advanced disease and establish the optimal antibody pairing from a potential panel of five antibodies associated with colorectal cancer cells.

4.4 End of Study Definition

Groups 1-4 will be in the study for 1 month to allow a single biological sample to be taken (of blood and/or faeces in group 1, and blood only in groups 2-8), and to allow a precise diagnosis and cancer stage where appropriate to be recorded.

Group 5 will be enrolled for 6 months to permit serial blood sampling (n=4) at diagnosis, during and after completion of chemoradiotherapy, and after subsequent surgery.

Group 6 will be enrolled for 2 years to allow 5 blood samples to be obtained during this standard follow up period. In the event of the death of the participant during the follow up period then only samples collected up to that timepoint will be measured.

4.5 Subject Withdrawal

Subjects will be withdrawn from the study under the following circumstances:

- Withdrawal of consent

4.6 Data Collection and Follow up for Withdrawn Subjects

Patients who withdraw consent may not wish their data to be used. If this is the case then it will be deleted.

5. Laboratories

5.1 Laboratory assessments

Raman analysis- 5mL blood will be required for each Raman analysis. Groups 1,2,3 and 4 will only require one blood sample (5mL total volume). Group 5 will have blood sampling on 4 occasions of 5mL per sample *ie* 20mL total. Group 6 will have 5mL of blood taken on 5 separate occasions *ie* 25mL blood in total.

Blood samples will be transferred to the Centre for NanoHealth laboratory in ILS-2 on ice to maintain the sample at 4 degrees centigrade. Faecal samples wil be transported at room temperature.

5.2 Local laboratories

All analysis will take place at the Centre for NanoHealth laboratory in ILS2 at Swansea University.

5.3 Sample labeling/logging

Samples will be pseudo-anonymised with a unique study number and date of collection.

5.4 Sample receipt/Chain of Custody/ Accountability

All samples received by the laboratory will be assessed on arrival to check their physical integrity. If samples have been compromised in transit the researcher will be notified promptly. On receipt, laboratory staff will ensure that all samples are accounted for and this process will be documented. If samples are poorly labeled, missing or if unexpected samples are received, the study sponsor will be notified. Samples with labeling that does not meet the laboratory's minimum acceptance criteria will be rejected and disposed of according to policy.

Each sample received at the laboratory will be appropriately and uniquely identified. Mechanisms will be in place to track the movement of each sample from arrival

to storage and to analysis. Transportation of samples will be undertaken according to the ABMU Health Board/ Public Health Wales Pathology Department transportation policy Adequate provision will be made to ensure that the laboratory in the Centre for NanoHealth and ILS-2 has sufficient capacity for the storage of refrigerated and frozen samples in the event of malfunction. All refrigerators and freezers will have temperature control logs maintained for QA as routine.

5.5 Sample storage procedures

All samples will either be analysed within 3 days of collection to minimize the chance of degradation, or stored at -80 degrees C until analysis. After analysis, all samples will be destroyed in accordance with the Human Tissue Act.

6. Safety measures

The safety of trial subjects will precedence over any other aspect of the trial. Consequently, prior to the initiation of laboratory work, lines of communication will be established with the study sponsor, to ensure that any issues that may impact on patient safety are reported without delay. These may include, but are not limited to, the reporting of unexpected or out or range results and significant deviations from the clinical protocol or work instructions.

6.1 General Definitions

Instructions for Safety Reporting (Research other than CTIMPs)

In other research other than CTIMPs, a serious adverse event (SAE) is defined as an untoward occurrence that:

(a) Results in death;
(b) Is life-threatening;
(c) Requires hospitalisation or prolongation of existing hospitalisation;
(d) Results in persistent or significant disability or incapacity;
(e) Consists of a congenital anomaly or birth defect; or is otherwise considered medically significant by the investigator.

An SAE occurring to a research participant should be reported to the main REC where in the opinion of the Chief investigator the event was:

- **Related** – that is, it resulted from administration of any of the research procedures, and
- **Unexpected** – that is, the type of event is not listed in the protocol as an expected occurrence.

Serious Adverse Events Procedures General Definitions

An adverse event (AE) is any untoward medical occurrence in a participant which does not necessarily have a causal relationship with the study intervention and can include;
-any unintentional, unfavourable clinical sign or symptom
-any new illness or disease or the deterioration of existing disease or illness
-any clinically relevant deterioration in any laboratory assessment or clinical tests

6.2 Notification and reporting Adverse Events or Reactions

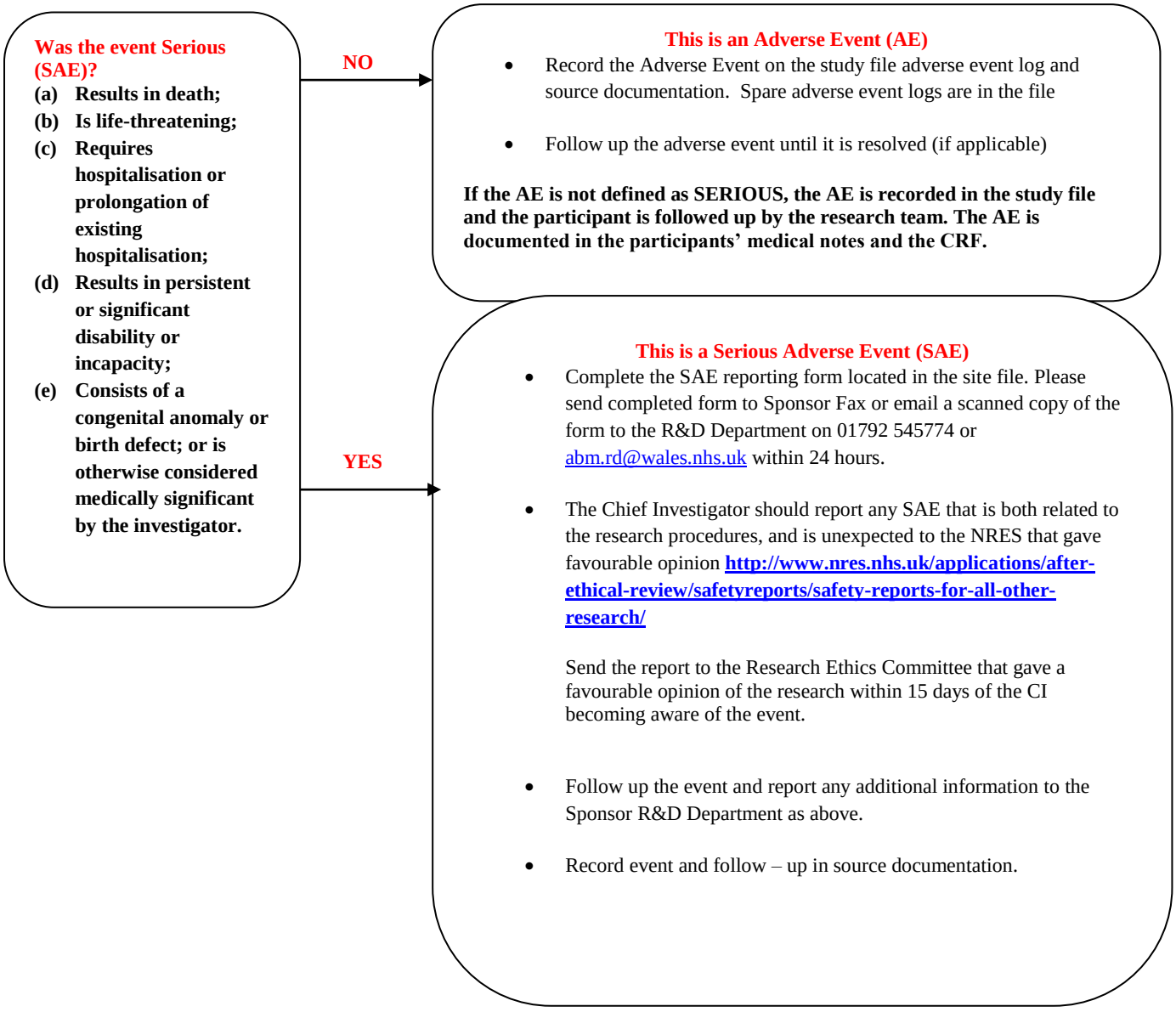
Expected AE AR/SAEs and SARs Not reportable

If the AE is not defined as SERIOUS, the AE is recorded in the study file and the participant is followed up by the research team. The AE is documented in the participants' medical notes (where appropriate) and the CRF.

For the purpose of the research study, adverse events related to the study are limited to those involved with sample collection. Blood sampling will involve an invasive blood test which has the attendant risks of pain, local bruising and infection. These risks will be minimised as blood sampling will be done by experienced practitioners using aseptic technique.

6.3 Notification and Reporting of Serious Adverse Events

Instructions for Safety Reporting (Research other than CTIMPs)



6.4 Urgent Safety Measures

The CI may take urgent safety measures to ensure the safety and protection of the clinical trial subjects from any immediate hazard to their health and safety, in

accordance with Regulation 30. The measures should be taken immediately. In this instance, the approval of the Licensing Authority Approval prior to implementing these safety measures is not required. However, it is the responsibility of the CI to inform the sponsor and Main Research Ethics Committee (via telephone) of this event **immediately**.

The CI has an obligation to inform both the Main Ethics Committee **in writing within 3 days**, in the form of a substantial amendment. The sponsor (ABM University LHB through its Research and Development Office) must be sent a copy of the correspondence with regards to this matter.

6.5 Annual Safety Reporting

The CI will send the Annual Progress Report to the main REC using the NRES template (the anniversary date is the date on the MREC "favourable opinion" letter from the MREC) and to the sponsor.

6.6 Overview of the Safety Reporting Process responsibilities

The CI/PI has the overall safety reporting responsibility. The CI/PI has a duty to ensure that monitoring and reporting is conducted in accordance with the sponsor's requirements.

7. Statistical Considerations

7.1 Sample Size

1,800 patients will be included in the study to provide a representative number in each subgroup for the purposes of this study.

Based on analysis from our pilot data (currently at n=200) the 300 samples per subgroup are predicted to provide the required independent validation test set to achieve over 85% sensitivity and specificity with 80% power at the 95% confidence interval.

7.2 Statistical Analysis

Statistical analysis and feasibility: The spectral analysis will be conducted using in-house principle component analysis (PCA) tools. This type of analysis is excellent for discriminating subtle spectral differences and determining commonality within datasets. The key aspect here will also include using the principle component loadings to ascertain the origin of the spectral differences, i.e. linking the spectral changes to macromolecular changes in the samples. Specially, by combining principal component analysis and hierarchical cluster analysis, the statistical difference between Raman spectra of different patient groups can be revealed effectively leading to specific accuracy, sensitivity and specificity measures. The research will also consider the development of PCA-based algorithms to establish calibration PCA loadings which can be run against 'unknown' samples. These new tools will enable the development of a

diagnosis platform and hence establish the route towards early diagnostics and personalised medicine.

8. Data Handling & Record Keeping

8.1 Confidentiality

Samples will be pseudoanonymised with a unique participant study number for the purposes of analysis. This number will be used on all CRFs and study forms.
A file listing the unique participant study number and patient hospital number will be maintained on a password protected NHS computer accessible only by the Principal Investigator. The study sponsor (NHS R&D department) will have access to study data for monitoring purposes. Participants consent will be sought to permit this access.

8.2 Study Documents

- A signed protocol and any subsequent amendments
- Current/Superseded Patient Information Sheets (as applicable)
- Current/Superseded Consent Forms (as applicable)
- Indemnity documentation from sponsor
- Conditions of Sponsorship from sponsor
- Conditional/Final R&D Approval
- Signed site agreement
- Ethics submissions/approvals/correspondence
- CVs of CI and site staff
- Delegation log
- Staff training log
- Patient identification log
- Enrolment log
- Protocol training log
- Correspondence relating to the trial
- Communication Plan between the CI/PI and members of the study team

8.3 Record Retention and Archiving

During the course of research, all records are the responsibility of the Chief Investigator and will be kept in secure conditions in a pseudoanonymised manner in either a locked filing cabinet in a locked office in the case of paper records, or on a password protected secure computer in the case of digital records. When the research trial is complete, records will be kept for a further 5 years as per local R&D guidelines. Archiving is provided by Transmedia, ABMU's archive facility. The R&D department keeps a log of all archiving activity.

8.4 Compliance

The CI will ensure that the trial is conducted in compliance with the principles of the Declaration of Helsinki (1996), and in accordance with all applicable regulatory requirements including but not limited to the Research Governance Framework, Trust and Research Office policies and procedures and any subsequent amendments. Similarly all procedures, handling and disposal of human tissue will be in accordance with the principles laid out in the Human Tissue Act.

8.5 Clinical Governance Issues

8.5.1 Ethical Considerations
This protocol and any subsequent amendments, along with any accompanying material provided to the patient in addition to any advertising material will be submitted by the Investigator to the South West Wales Research Ethics Committee.

8.6 Quality Control and Quality Assurance

7.6.1 Summary Monitoring Plan
The Co investigators will establish quality control. The co-investigators will monitor protocol deviations, with the Chief investigator assuming overall responsibility.

8.7 Non-Compliance
(A noted systematic lack of both the CI and the study staff adhering to SOPs/protocol/ICH-GCP, which leads to prolonged collection of deviations, breaches or suspected fraud.)

These non-compliances may be captured from a variety of different sources including monitoring visits, CRFs, communications and updates. The sponsor will maintain a log of the non-compliances to ascertain if there are any trends developing which to be escalated. The sponsor will assess the non-compliances and action a timeframe in which they need to be dealt with. Each action will be given a different timeframe dependent on the severity. If the actions are not dealt with accordingly, the R&D department will agree an appropriate action, including an on-site audit.

9. Publication Policy

The findings of this research project will be disseminated at local, regional and national levels, and used in future funding applications. This will be done through

presentation of results at departmental and regional meetings. We plan for the results of this study to be published in peer reviewed journals and thus disseminated throughout the relevant scientific community.

10. References

Rödel C, Martus P, Papadoupolos T, Füzesi Letal. Prognostic significance of tumor regression after preoperative chemoradiotherapy for rectal cancer. J Clin Oncol. 2005 Dec 1;23(34):8688-96

Li X, Yang T, Li S. Discrimination of serum Raman spectroscopy between normal and colorectal cancer using selected parameters and regression-discriminant analysis. Appl Opt. 2012; 51(21): 5038-43.

Lin D, Feng S, Pan J, Chen Y, Lin J, Chen G, Xie S, Zeng H, Chen R. Colorectal cancer detection by gold nanoparticle based surface-enhanced Raman spectroscopy of blood serum and statistical analysis. Opt Express. 2011; 19(14): 13565-77.

Di Lena M, Travaglio E, Altomare DF. New strategies for colorectal cancer screening. World J Gastroenterol. 2013; 19(12): 1855-60.

Mavarani L, Petersen D, El-Mashtoly,SF, Mosig A, Tannapfel A, Kotting C, Gerwert K. Spectral histopathology of colon cancer tissue sections by Raman imaging with 532 nm excitation provides label free annotation of lymphocytes, erythrocytes and proliferating nuclei of cancer cells. Analyst, 2013, 138, 4035

Jokerst JV, Miao Z, Zavaleta C, Cheng Z, Gambhir SS (2011) Affibody-functionalized gold-silica nanoparticles for Raman molecular imaging of the epidermal growth factor receptor. Small 7(5):625–633.

Calistri D, Rengucci C, Casadei Gardini A, Frassineti GL, Scarpi E, Zoli W, Falcini F, Silvestrini R, Amadori D. Fecal DNA for noninvasive diagnosis of colorectal cancer in immunochemical fecal occult blood test-positive individuals. Cancer Epidemiol Biomarkers Prev. 2010; 19(10): 2647-54.

11. Appendices

Consent form v2

Patient Information Sheets v2

Faecal collection method