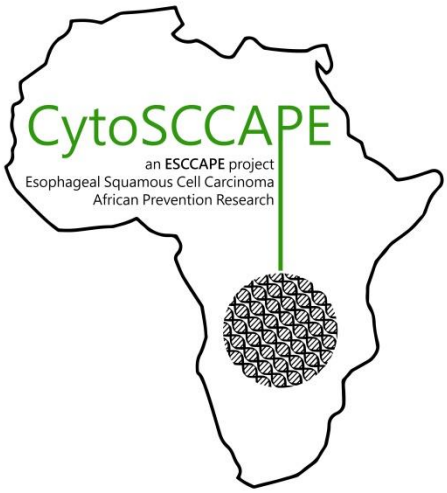
Esophageal Squamous Cell Carcinoma African Prevention Research (ESCCAPE)
Cytosponge™ Feasibility Study

Cytosponge™ Feasibility Study in Tanzania (CytoSCCAPE)



Full study protocol

CYTOSCCAPE_PV3

Date of protocol: 03 March 2019

Contents

1. GENERAL INFORMATION

Protocol title: ESCCAPE Cytosponge™ Feasibility Study – **CytoSCCAPE**

Protocol identifying number: CYTOSCCAPE_PV3

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Person(s) authorized to sign the protocol and amendments for the sponsor:

Dr Valerie McCormack
Scientist
Section of Environment and Radiation
International Agency for Research on Cancer (IARC/WHO)
Email: mccormackv@iarc.fr
Telephone: +33 4 72 73 85 66

Sponsor's medical expert for the trial:

Dr Behnoush Abedi-Ardekani
MD-Pathologist
Genetic Cancer Susceptibility Group
International Agency for Research on Cancer
World Health Organization
150 Cours Albert Thomas
69372 Lyon CEDEX 08, France
Tel. +33472738 504
E-mail: abedib@iarc.fr

Investigators responsible for conducting the trial:

KCRI:
Prof Venance Maro
KCMU-College P.O Box 2240-Moshi
Telephone Number: +255-027-2753616
Cell phone Number: 0754581444
E-mail address: venmaro@ymail.com
Study role: KCRI Principal Investigator

Dr Amos Mwasamwaja MD, M.Med - Int.Medicine
Specialist Physician/Physician Endoscopist (KCMC)
Assistant Lecturer (KCMUCo.)
P.O.BOX 3010, Moshi
Email: a.mwasamwaja@kcri.ac.tz
Mobile +255767386817
Study role: KCRI Co-Principal Investigator

Dr Blandina T. Mmbaga MD, MMed, PhD
Director Kilimanjaro Clinical Research Institute (KCRI)

Box 2236 Moshi
Tanzania
b.mmbaga@kcri.ac.tz; blaymt@gmail.com; blaymt@yahoo.com
Tel. +255272754201 Mobile: +255768435116
Study role: Co-investigator

Dr Alex Mremi, KCRI Pathology Lead, +255 717 083 983, alexmremi@gmail.com

IARC:

Dr Daniel Middleton
Postdoctoral Fellow
Section of Environment and Radiation
International Agency for Research on Cancer (IARC/WHO)
middletond@fellows.iarc.fr
Tel. +33 472 738 310
Study role: IARC Co-Principal Investigator with Dr McCormack; Study Coordinator

Dr Valerie McCormack
Scientist
Section of Environment and Radiation
International Agency for Research on Cancer (IARC/WHO)
Email: mccormackv@iarc.fr
Telephone: +33 4 72 73 85 66
Study role: IARC Co-Principal Investigator with Dr Middleton

Sponsor's monitor:
Dr Partha Basu
Group Head, Screening Group
International Agency for Research on Cancer (IARC/WHO)
Email: basup@iarc.fr
Tel. +33 472 738 167

Dr Behnoush Abedi-Ardekani
MD-Pathologist
Genetic Cancer Susceptibility Group
International Agency for Research on Cancer
World Health Organization
150 Cours Albert Thomas
69372 Lyon CEDEX 08, France
Tel. +33472738 504
E-mail: abedib@iarc.fr
Study role: Co-investigator, Medical Expert

Dr Joachim Schüz, PhD
Head, Section of Environment and Radiation
International Agency for Research on Cancer
World Health Organization
150 cours Albert Thomas
69372 Lyon CEDEX 08, France

Phone +33 472 738 441
E-mail: schuzj@iarc.fr
Study role: Co-investigator
University of Cambridge:

Prof Rebecca Fitzgerald
Professor of Gastroenterology
MRC Cancer Unit
University of Cambridge, UK
rcf29@mrc-cu.cam.ac.uk
Study role: Co-investigator/UK PI

Dr Maria O'Donovan
Consultant Histopathologist and Cytopathologist
Cambridge University Hospitals NHS Foundation Trust
Cambridge Biomedical Campus, Cambridge, UK
maria.o'donovan@addenbrookes.nhs.uk
Study role: Co-investigator/Pathology lead

Irene Debiram-Beecham
Principal Research Nurse
T. +44 1223 761085 / +44 1223 763994
M. +44 7979964818
E-Mail: id213@medschl.cam.ac.uk
Study role: Cytosponge™ procedure training

Martin Bromwich
Biomedical Scientist
University Hospitals NHS Foundation Trust
Tel: +44 1223 596099
E-Mail: martin.bromwich@nhs.net
Study role: Cytosponge™ sample preparation training

Trial site:

Kilimanjaro Clinical Research Institute (KCRI)/KCMC, and its community research facility situated at the Majengo Health Centre, Moshi.
Box 2236 Moshi
Tanzania

Qualified physician responsible for all trial-site related medical decisions:

Dr Gissela Maro

Institutions involved in the trial:

Kilimanjaro Clinical Research Institute (KCRI)/KCMC
Box 2236 Moshi
Tanzania

International Agency for Research on Cancer (IARC)
World Health Organization
150 cours Albert Thomas
69372 Lyon CEDEX 08, France

MRC Cancer Unit (MRC-CU)
University of Cambridge
Cambridge
UK

2. BACKGROUND INFORMATION

2.1. Research rationale:

Oesophageal cancer (EC) is the 8th most common cancer worldwide (1). The histological subtype oesophageal squamous cell carcinoma (ESCC) has one of the steepest spatial gradients of all cancers, including notable "hotpots" in Asia (e.g. Iran and China) and the African EC corridor (2-4). In East Africa, it is the 3rd and 4th most common cancer in men and women, respectively, and occurs from young ages (20+) (5, 6). Tanzania, with an estimated age standardized rate of 12.9 (males and females combined), ranks 8th for ESCC incidence in Africa. Late stage diagnosis and dire prognosis (<6 months (7)) necessitates the identification of risk factors for primary prevention, for which there are several plausible modifiable candidates (8). Although documented in the 1950s (9-12), unlike Chinese and Iranian hotspots (13, 14), African research is lacking other than in South Africa (2, 15). Thus, in 2014 the International Agency for Research on Cancer (IARC) invested in pertinent research, forming **ESCCAPE, the ESCC African Prevention REsearch** network (http://esccape.iarc.fr), which is dedicated to identifying the risk factors of ESCC in East Africa.

With no active cohorts in this setting and a need for timely outcomes, exploring the implementation of alternative epidemiological study designs is a necessity. While the *prevalence* of ESCC is low in the general population in the absolute sense, the prevalence of oesophageal squamous dysplasia (ESD) – an established precursor legion to ESCC – will be appreciably higher. For example, in Bomet, Kenya, one study that endoscoped asymptomatic general population subjects found an ESD prevalence of 14.4% in the local population and as high as 20% in men aged over 50 years (16). This makes ESD a logical, attractive and feasible endpoint to consider when investigating ESCC etiology, assuming that ESCC and its precursor ESC share common risk factors.

Although implemented in the one-off Kenyan study, conventional methods of diagnosing ESD – involving upper-GI endoscopy - are too invasive, timely and expensive to implement in the asymptomatic population in most settings and especially in resource limited settings. A less-invasive alternative has been developed by the MRC Cancer Unit (MRC-CU) at the University of Cambridge, UK - a device called the Cytosponge™. In brief, rather than esophageal tissue being obtained through a biopsy during endoscopy, esophageal cells are obtained by swallowing a Cytosponge™ – a sponge on a string – and pulling this string back up through the mouth during which time esophageal cells are collected on the sponge. This device, originally developed for the diagnosis of Barrett's

oesophagus – the precursor to oesophageal adenocarcinoma - was approved by the UK Medicines and Healthcare Products Regulatory Agency (MHRA) in 2008. It previously received a letter of no objection from the MHRA (MHRA Reference CI/2007/0053) to screen for Barrett's oesophagus. Recent studies (17) have successfully utilised the device for the diagnosis of ESD, a range or benign pathologies (18) and for conducting genomic and epigenetic assays (19), making it an attractive tool for application in the East African setting. However, no studies have been conducted to-date on the acceptability of the Cytosponge™ in this high incidence region. We have designed a feasibility study to examine acceptability and performance of the Cytosponge™ use in general population volunteers in Moshi, Tanzania. This location has high ESCC incidence rates and is also the setting of an ESCC case-control study we are conducting. Findings will be used to inform the design and implementation of future ESCCAPE research studies using the Cytosponge™ to investigate ESCC etiology.

2.2. Name of investigational product:

Cytosponge™

2.3. Description:

The device, developed by Prof Fitzgerald and her research group at the MRC-CU (co-investigators of the present application), consists of a spherical mesh enclosed in a gelatine capsule and attached to a string (Figure 1). The capsule is swallowed with the use of water (approximately 200 mL) and allowed to reach the stomach while remaining attached to the suture which is held onto by the patient or nurse (and which is affixed to a card preventing inadvertent swallowing of the suture). In the stomach the capsule is left for up to 5 minutes where it dissolves allowing the sponge to expand to its full size. It is then withdrawn using the suture, and as it does so collects cells from the lining of the oesophagus. After retrieval, the Cytosponge™, now containing the cytological specimen, is placed in a preservative fluid. The typical cell yield of the procedure is 250,000. These cells are used to prepare a clot which is fixed with alcohol and embedded in paraffin for sectioning and slide preparation. Slides are then examined under a microscope for pathological diagnosis and characterisation. In this regard, the Cytosponge™ is a minimally invasive, cheap and quick procedure which achieves collection of internal oesophageal cells.

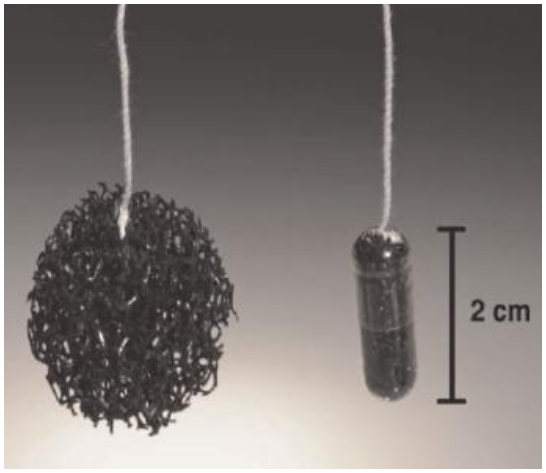


Figure 1: Cytosponge™ within its capsule (right) and expanded after the gelatine capsule has dissolved (left). Image courtesy of University of Cambridge, UK.

2.4. Previous findings relevant to the trial:

Prof Fitzgerald's research group have an extensive track record of successful implementation of the Cytosponge™ in both clinical and research settings. Key milestones from a number of trials successfully completed to-date are summarised below:

Screening for Barrett's oesophagus:

- A proof of principal study (20), consisting of 43 patients with Barrett's oesophagus and 54 healthy volunteers, demonstrated that the Cytosponge™ was acceptable to patients and preferable to endoscopy in 80%. On an acceptability scale of 1 (worst experience) to 10 (very enjoyable), with 5 being neither pleasant nor unpleasant, an average rating of 4 was obtained.
- In the BEST1 prospective cohort study (21) (504 participants), 99% of participants swallowed the device and no serious adverse events occurred. Compared to upper-GI endoscopy, the sensitivity and specificity of detecting Barrett's oesophagus were 73% and 94%, respectively (lesions ≥1 cm circumferential length) and 90% and 94% for lesions ≥2 cm. The majority (82%) of those who swallowed the device reported low levels of anxiety before the test and <4.5% reported psychological stress the following week.
- The performance and acceptability of the Cytosponge™ were further demonstrated in a 1,100 participant case-control study (BEST2) conducted across 11 UK hospitals (22). The sponge was successfully swallowed by 94% of participants and no serious adverse events attributed to the device were reported. The device received a favourable rating from participants compared to standard endoscopy. The overall sensitivity of the test (Cytosponge™ coupled with immunohistochemical staining of Trefoil Factor 3) in diagnosing Barrett's oesophagus was 79.9%.

Screening for oesophageal squamous dysplasia (ESD):

- In a pilot study (23) conducted in the high ESCC incidence Golestan Province in Northern Iran, 301 participants underwent Cytosponge™ collection and endoscopy. Satisfaction with the

Cytosponge™ examination was reported by 93% of participants and no complications were encountered. The sensitivity of sponge cytology to detect high grade ESD was 100% with a specificity of 97%. Accuracy was 100% when p53 immunohistochemistry staining was used in addition to cytological examination.

Assessing DNA methylation:

- Cytosponge™ samples were analysed to compare differentially methylated genes between Barrett's oesophagus and normal squamous epithelium biopsies – identifying potential diagnostic biomarkers (19).

Diagnosis of benign pathologies:

- The Cytosponge™ was administered on 409 UK patients with reflux symptoms and successfully diagnosed a range of benign pathologies: inflammation, candidiasis, eosinophilic oesophagitis and viral inclusions. A 70% agreement was found between the Cytosponge™ and conventional endoscopy (18).

2.5. Potential risks and benefits to human subjects:

Risks:

The potential risks to human subjects swallowing the device are:

- Detachment of sponge and lodgement in either oesophagus or stomach, which has occurred in less than 1 per 1000 uses. In the event of this happening, participants would be transported for emergency endoscopy at KCMC under the care of Dr Gissela Maro.
- Bleeding, though no bleeding complications have occurred to-date with this device.
- Sore throat

The CytoSCCAPE study benefits from the groundwork and knowledge gained by earlier predecessor trials using the device (see section 2.9). A comprehensive risk report for the device, as compiled for the UK OSCAR trial, is provided for reference as a separate document (*FPB-12-0101_D_A*). All measures taken to prevent the above effects are detailed in the risk report (*FPB-12-0101_D_A*).

Benefits:

Analysis of cells collected with the sponge may help to diagnose an otherwise undetected condition, such as ESD – which may progress to ESCC – or an early stage, asymptomatic ESCC, as well as other benign undiagnosed pathologies.

2.6. Route of administration, dosage, dosage regimen and treatment period:

The device is transient and will be administered on a single occasion. The procedure is expected to take no more than 10 minutes – 2-3 minutes for the study researcher to un-package and prepare the device, which is then swallowed by the participant; 5 minutes waiting time for the capsule to dissolve and release the sponge followed by <1 minute to withdraw the device. Only a single sample

will be taken during this study, as the administration of multiple devices has not been tried previously.

Additional study components:

- Informed consent process (15 minutes)
- Exclusion criteria confirmation
- Questionnaire (10 minutes)

2.7. Statement:

The investigators declare that the study will be conducted in compliance with the protocol, GCP and TFDA requirements.

2.8. Population to be studied:

The study population will consist of healthy (i.e. asymptomatic for esophageal conditions) adults (≥30 years old) residing in Kilimanjaro District, Tanzania, who will be invited to visit the Majengo Healthcare Centre, Moshi, where the Cytosponge™ will be administered. Potential participants will be approached during routine visits to the facilities. This geographic area encompasses the catchment area of an ongoing case-control study of the risk factors of ESCC, which is being conducted at KCRI. The target age and gender distribution of the study population is shown in Table 1. This distribution was informed by (i) the prevalence of ESD found in a comparable population (16) from Bomet, Kenya (i.e. <6% under age 30, a prevalence too low to be meaningful compared to 15-20% above) and (ii) the age and gender distribution of ESCC cases in the ESCCAPE case-control study being conducted at KCRI, which has zero patients under age 30 among the first 200 recruited. Those individuals invited to participate in the study will be recruited during visits to the Majengo Healthcare Facility, or will be invited to attend the facility, and be asked to complete a short lifestyle questionnaire.

Table 1: Target age and gender distribution of participants

Age	Male	Female
30-39	10	10
40-49	10	10
50-59	15	15
60+	15	15
Total	50	50

2.9. Literature and data references with relevance to the trial:

Selected literature (as summarised in Section 2.4.):

Lao-Sirieix P, Rous B, O'donovan M, Hardwick RH, Debiram I, Fitzgerald RC. Non-endoscopic immunocytological screening test for Barrett's oesophagus. *Gut*. 2007; **56**(7):1033-4.

Kadri SR, Lao-Sirieix P, O'Donovan M, Debiram I, Das M, Blazeby JM, et al. Acceptability and accuracy of a non-endoscopic screening test for Barrett's oesophagus in primary care: cohort study. *BMJ*. 2010; **341**: c4372.

Ross-Innes CS, Debiram-Beecham I, O'Donovan M, Walker E, Varghese S, Lao-Sirieix P, et al. Evaluation of a minimally invasive cell sampling device coupled with assessment of trefoil factor 3 expression for diagnosing Barrett's esophagus: a multi-center case–control study. *PloS Medicine*. 2015; **12**(1): e1001780.

Roshandel G, Merat S, Sotoudeh M, Khoshnia M, Poustchi H, Lao-Sirieix P, et al. Pilot study of cytological testing for oesophageal squamous cell dysplasia in a high-risk area in Northern Iran. *Br J Cancer*. 2014; **111**(12): 2235-41.

Chettouh H, Mowforth O, Galeano-Dalmau N, Bezawada N, Ross-Innes C, MacRae S, et al. Methylation panel is a diagnostic biomarker for Barrett's oesophagus in endoscopic biopsies and non-endoscopic cytology specimens. *Gut*. 2017: gutjnl-2017-314026.

Paterson AL, Lao-Sirieix P, O'donovan M, Debiram-Beecham I, Pietro M, Miremadi A, et al. Range of pathologies diagnosed using a minimally invasive capsule sponge to evaluate patients with reflux symptoms. *Histopathology*. 2017; **70**(2):203-10.

3. TRIAL OBJECTIVES AND PURPOSE

3.1. Purpose:

Assess the feasibility of successful Cytosponge™ use in Tanzania for etiological research on oesophageal tissue health/pathology.

3.2. Core Objectives:

1) Assess the recruitment response rate and post-use acceptability of the device using a previously employed acceptability scale.
2) Assess implementation success of the device – determined as the proportion of participants who successfully swallowed the device in 3 attempts or less.
3) Evaluate the local, site-specific implementation of Cytosponge™ sample collection/processing protocols.
4) Investigate the prevalence and determinants of ESD, and document other esophageal pathologies observed, in the study population.
5) Biobank samples for future investigation of the prevalence and determinants of immunohistochemistry markers for specific ESCC-related exposures in collected cells (e.g. polycyclic aromatic hydrocarbons; inflammatory cytokines; proliferation markers).

3.3. Extended objective:

6) Evaluate DNA yield and quality for use in genomic and epigenetic studies.

4. TRIAL DESIGN

4.1. Endpoints to be measured during the trial:

Core:

- Response rate of participants invited to swallow device.
- Proportion of participants able to swallow the device within three attempts and number of attempts taken (frequency distribution of 1, 2 and 3 attempts).
- Post-swallow acceptability of device as per a previously employed acceptability scale (graded 1-10). Figure 2 shows the results from a similar study conducted in Northern Iran (17).

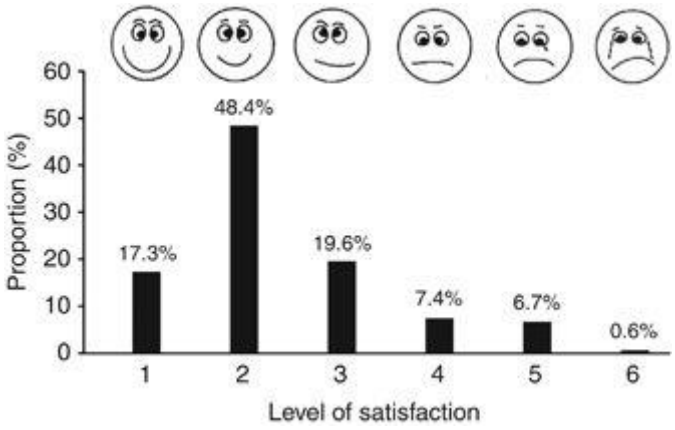


Figure 2: Acceptability of the procedure in a North Iranian population. Source: Roshandel et al. (2014)

- Proportion of collected sponges successfully processed into paraffin blocks.
- Prevalence of ESD as determined by examination by a trained pathologist.
- Prevalence of benign oesophageal pathologies (inflammation, candidiasis, eosinophilic oesophagitis).

Extended (subject to future funding):

- Prevalence of positivity for antibodies against chemical exposures and proliferation markers.
- DNA yield from cells collected from the device.
- DNA methylation profile on a subset of 30 sponges.
- Genetic mutations in candidate cancer-relevant gene panel.

A schematic of the abovementioned endpoints as measured at different time points throughout the study is shown in Figure 3.

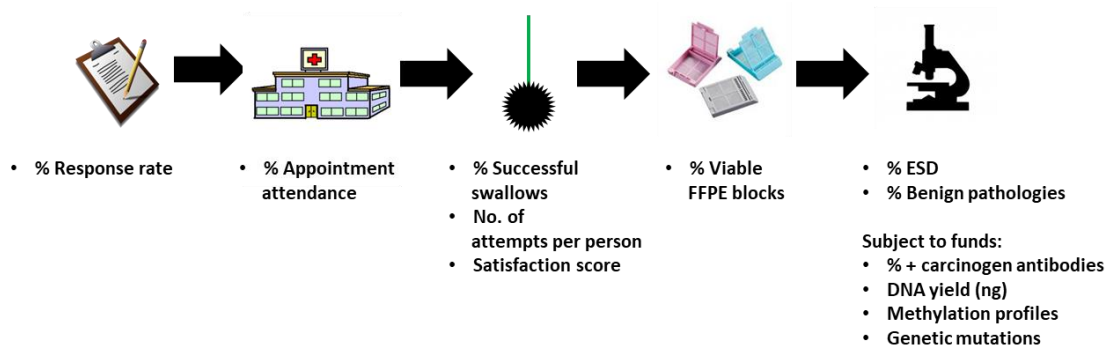


Figure 3: Study endpoints to be measured.

4.2. Type/design of trial:

Recruitment:

The study will consist of a general population cross-sectional design. Healthy participants will be recruited during routine visits to the KCRI Majengo Healthcare Facility or nearby residents will be invited to attend, where a full description of the study and Cytosponge™ procedure will be given. Individuals (≥30 years old), will be invited to Majengo to participate in the Cytosponge™ study and, if they agree, they will be asked to return to Majengo the following day to complete a questionnaire, administered using a mobile-based application (RedCap), to collect socio-demographic variables and information on putative risk factors for ESCC. They will then undergo the Cytosponge™ procedure, having not eating in the four hours prior to the appointment.

4.3. Measures taken to minimize/avoid bias:

Determining which people are more or less likely to be willing to undergo Cytosponge™ examination will be a crucial determinant of future research efforts. To maintain adherence to the target age and gender distribution specified in Section 2.8, age and gender frequency distribution grids will be used to track recruitment. Once a participant has successfully swallowed the device, a cross will be marked in a box corresponding to their age and gender. When all boxes in the grid are filled, study recruitment will be complete. The age and gender of all non-respondents, including of large groups approached, appointment attendance failures and swallow failures will be recorded.

4.4. Trial treatment and dosage of investigational product:

The intervention will consist of a single administration of a single Cytosponge™ device.

4.5. Packaging and labelling:

A unique identifier will be added by the manufacturer to aid device tracking and accountability. Devices will be provided in sealed securitainers whose function is to contain the device and provide a degree of tamper evidence with the pull-off seal before use. It has been selected on the basis that this type of package is routinely used for packaging of pharmaceutical capsule products. A forensic grade has been selected for cleanliness.

The labelled package will read: "**CytoSCCAPE Study — Principal Investigator Prof Venance Maro**" It will have the model number and lot numbers as per CE- marked requirement.

We will also add to this "This device is only to be used for the ESCCAPE Cytosponge™ Feasibility Study". The device will also be marked "non-sterile".

4.6. Expected duration of subject participation

For the Cytosponge™ component (i.e. excluding the recruitment process) participation of an individual will take no longer than the duration of an average hospital visit. However, in this setting, such visits can take up a participant's full day, depending on travel circumstances. Appointments themselves are anticipated to take approximately 30 minutes, with the administration procedure of the Cytosponge™ taking approximately 15 minutes. A dedicated staff member will be stationed to greet participants as they arrive and escort them to the designated study room to minimise time. Hosting the study at the Majengo Healthcare Facility, situated close to residential areas, will not interfere with normal hospital procedures and keep waiting times to a minimum. Participants will then take their journey home by public transport (maximum 1 hour) for which the cost will be covered by the study. If follow-up is not required, this will be the full duration of participation for a given individual. For individuals whose samples display ESD, their ongoing participation will consist of the duration consistent with the normal endoscopy service of the hospital. Treatment for those with high grade ESD will be organised with the cooperation of Dr Mike Mwachiro from Tenwek Hospital and subject to local waiting times.

4.7. Stopping rules/discontinuation criteria:

As this study only consists of a single visit/administration, discontinuation for an individual participant will occur if they fail to swallow the Cytosponge™ after three attempts. Full training in use of the device will be provided and, in the unlikely event of more than two adverse events (see Section 8.1) in a single week, the study will be discontinued pending investigation.

4.8. Accountability procedures for investigational products:

For all issues relating to the design of the Cytosponge™, the UK Medical Research Council is accountable. For issues relating to the manufacture of the device, EUROPLAZ – the manufacture – is accountable.

4.9. Maintenance of trial treatment randomization codes:

While not a randomized trial design, all participants will receive an anonymous, unique ID number with a corresponding QR code. During recruitment, participants will be assigned a paper label containing their unique ID and QR code, these labels will be (i) attached to consent forms and scanned into the mobile app questionnaire; (ii) placed on the Cytosponge™ collection container and (iii) an additional label with the same ID and code will later be placed on sample bags used to contain the parafin blocks prepared for each Cytosponge™ sample. This system is designed to eliminate transcription related errors commonly encountered when hand writing ID codes.

4.10. Data to be recorded directly on CRFs:

As with questionnaires, non-identifiable CRFs will be hosted on the RedCap application and will collect the following information:

- ID (generated by scanning QR code)
- Appointment time and date
- Date of birth
- Age (generated from previous or estimated if not known)
- Gender
- HIV Status
- Number of attempts taken to swallow sponge (between 1 and 3)
- Acceptability
- Adverse events
- Severity of adverse events
- Comments from nurse administering the device

A separate hard copy CRF containing identifiable and contact information will be recorded separately and stored at KCRI.

CRFs will later be linked by unique ID to their corresponding questionnaires and laboratory generated outcomes. The full questionnaire is in Appendix P1. Please note: the content of the questionnaire may be subject to change following IRB approval of any additions.

5. SELECTION AND WITHDRAWAL OF STUDY PARTICIPANTS

5.1. Participant inclusion criteria (self-reported):

- 30 years or older
- Able to provide informed consent
- Resident of the study area (Kilimanjaro Region)

5.2. Participant exclusion criteria (self-reported):

- Individuals with symptoms of dysphagia
- Current history of active oro-pharyngeal cancer
- Oesophageal varices, stricture or requiring dilatation of the oesophagus
- Recent history of vomiting blood
- Taking anticoagulation therapy/medication (warfarin, clopidogrel, heparin, tinzaparin or enoxiparin) for high risk conditions
- Use of anti-thrombotic medication
- Individuals who have had a myocardial infarction or any cardiac event less than six months prior to recruitment into the study
- Individuals who have had a cerebrovascular event less than 6 months ago where their swallowing has been affected
- Eaten and drank within the previous 4 hours
- Received prior surgical intervention to the oesophagus
- Known current pregnancy

- Lacking capacity to provide informed consent

5.3. Participant withdrawal:

- Participants are free to withdraw from the study at any time and for any reason. After recruitment appointments, participants will be provided with relevant contact details for the investigators so that they can notify the study team of their desire to withdraw, while no sponge will be collected, their previously corrected data will still be analysed and used to report withdrawal statistics.
- The target age and gender distribution of the study population will be monitored with the use of frequency grids. These grids will be updated should participants choose to withdraw, so that a suitable replacement participant can be recruited.
- In the case of an abnormal cytology result, if a participant withdraws prior to the study team informing them of this result, they will still be informed so that a follow-up endoscopy can be recommended as per the duty of care of the investigators.
- The following scenarios should not result in participants being withdrawn from the study:

  1. Unable to swallow Cytosponge™: Their data may continue to be collected and analysed.
  2. Inadequate Cytosponge™ test: Their data may continue to be collected and analysed.
  3. Subsequently deemed ineligible for the Cytosponge™: Based on further review following written consent, participants found to not be eligible will not be withdrawn. Unless specifically requested, their data may continue to be collected and analysed.

6. TREATMENT OF STUDY PARTICIPANTS

Intervention will consist of a single Cytosponge™ administration in a private study room under the supervision of a dedicated nurse. Written consent will be taken at the beginning of the Cytosponge™ appointment. Participants who meet the inclusion crietria will then be asked to swallow the capsule. If a participant fails to swallow the capsule, they will be asked to try again. Participants will be able to try up to three times before they are classified as a “failed to swallow”. Samples will be refrigerated and processed in the local KCMC pathology laboratory. In the event of a Cytosponge™ detachment or an obvious bleed the research nurse will immediately inform the endoscopist (Dr G. Maro) as the participant falls under their duty of care.

For participant’s whose cells exhibit ESD (any grade) follow-up chromoendoscopy with Lugol’s iodine will be performed by Dr G. Maro. Those found to have high-grade ESD during this procedure will undergo appropriate treatment by either endoscopic mucosal resection or radiofrequency ablation, under the supervision of Dr Michael Mwachiro from Tenwek Hospital, who is experienced in this procedure.

6.1. Medications/treatments not permitted before and during the trial:

Anticoagulation therapy/medication: warfarin, clopidogrel, heparin, tinzaparin or enoxiparin and anti-thrombotic medication are NOT permitted.

6.2. Procedures for monitoring participant's compliance:

Participants will be asked to verify, prior to Cytosponge™ administration, that they have not eaten for 4 hours prior to the appointment.

7. ASSESSMENT OF EFFICACY

7.1. Efficacy parameters:

- 1. Successful device swallow
- 2. Successful sponge deployment
- 3. Successful cell harvesting
- 4. Successful block preparation

7.2. Methods and timing for assessing, recording and analysing efficacy parameters:

- 1. Three attempts will be made to swallow the device, visual inspection of the back of the tongue and length of cord will be made by the nurse administering the device. The number of attempts will be recorded and failures will be recorded for those not able to swallow after three attempts.
- 2. A digital timer will be set to 5 minutes once the patient is confident that the device has been successfully and completely swallowed. The device will not be withdrawn until the timer has reached zero.
- 3. Visual inspection of the sponge on retrieval will determine if it was effectively deployed from the capsule and thus optimal in harvesting cells. Any observations of improperly deployed sponges will be noted in the "Comments from the nurse" fields in CRFs.

8. ASSESSMENT OF SAFETY

8.1. Safety parameters:

Definitions:

- An adverse device event (ADE), as it relates to the use of the Cytosponge™ is defined as an untoward medical occurrence resulting from: insufficiencies or inadequacies in the instructions for use, deployment, implantation, installation, operation, or any malfunction, a use error or intentional misuse. Possible ADEs are shown in Table 2.

Table 2: Possible adverse device events occurring as a result of Cytosponge™ administration.

Possible adverse device events
Cytosponge™ detached from the string while in the patient's oesophagus/stomach
Inability or difficulty to remove the Cytosponge™

Laceration at the back of the throat
Obvious bleeding from the oesophagus
Perforation or tear of the oesophagus
Obstruction on breathing or airway as a result of the Cytosponge™

- A serious adverse device event (SADE), as it relates to the use of the Cytosponge™, is defined as any adverse event that has resulted in any of the characteristics of an SADE, that:

- led to a death

- led to a serious deterioration in health

- resulted in a life threatening illness or injury, (a Life-threatening refers to an event where the participant was at risk of death at the time of the event. It does not refer to an event which hypothetically might have caused death if it were more severe or might have resulted in a permanent impairment of a body structure or a body function).

- required in-patient hospitalisation or prolongation of existing hospitalisation, (Any hospitalisation that was planned prior to recruitment will not meet SADE criteria. Any hospitalisation that is planned post recruitment as a result of the study procedures, will meet the SADE criteria.)

- resulted in medical or surgical intervention to prevent life threatening illness or injury or resulted in persistent or significant disability or incapacity

This includes device deficiencies that might have led to a serious adverse event if:

- suitable action had not been taken

- intervention had not been made

- circumstances had been less fortunate

A planned hospitalisation for pre-existing condition, or a procedure required by the protocol without a serious deterioration in health, is not considered to be a SADE.

8.2. Methods and timing for assessing, recording and analysing safety parameters:

Timing: those ADEs arising at the time of device administration will be recorded on the RedCap-based CRF. Events that are reported post-visit will be recorded in a separate log.

Assessment of adverse events: Each ADE must be assessed for causality, seriousness, severity and expectedness by the site PI and nurse.

Assessment of relatedness: An adverse event should be categorised as unrelated or possibly related. Where an adverse event is deemed to be possibly related this will indicate that the nature of the

event, the underlying medical conditions, concomitant medication or temporal relationship make it possible that the ADE has a causal relationship to the research procedure.

Assessment of severity: The assessment of severity will be recorded according to the following categories:

- Mild: an event that is easily tolerated by the participant causing minimal discomfort and not interfering with every day activities.
- Moderate: an event that is sufficiently discomforting to interfere with normal every day activities.
- Severe: an event that prevents normal everyday activities.

Assessment of expectedness: The investigator must make an assessment of the expectedness of the ADE based on knowledge of the effect and any relevant product information. Where the effect is not expected, the SADE will be deemed to be an unanticipated serious adverse event (USADE). In the Trial, a USADE, as it relates to the use of the Cytosponge™, is defined as a SADE that, by its nature, incidence, severity of outcome, has not been identified in the current version of the protocol or risk assessment.

8.3. Procedures for eliciting reports and recording/reporting adverse events and incurrent illnesses:

Mode of reporting:

The KCRI PI (with delegation to research nurses) will report SADEs within 24 hours of becoming aware of the incident to IARC.

A paper SADE CRF will be completed and signed by the PI or delegate and emailed to IARC and MRC-CU. KCRI will ensure that any patient identifiable information is not contained in any paper CRFs or documents when transferring to IARC/MRC-CU.

Recording of SADE:

The study information will be recorded in the participant's notes. Each patient will be provided with contact details on their information sheet indicating clearly whom to contact in the event of an ADE. Only ADEs (including SADE) occurring within 7 days of the Cytosponge™ procedure will be recorded and investigated. For each SADE, the following information will be collected:

- Full details in medical terms and case description
- Event duration (start and end dates, if applicable)
- Action taken
- Outcome
- Seriousness criteria

- Causality (i.e. relatedness to study procedures), in the opinion of the investigator
- Whether the event would be considered anticipated or unanticipated.

Electronic management of SADEs:

PIs, delegated to research nurses, will record SADEs within 24 hours of becoming aware of the event/effect. They will additionally send an email to IARC. Details provided within the SADE CRF will include ESCCAPE ID, type of SADE, severity, outcome and remedial actions taken.

The team will request an update about outcomes and remedial actions taken directly from the KCRI research nurse/Principal Investigator and the SADE record reviewed by IARC/MRC-CU as complete (fully investigated, remedial actions taken and REC informed where relevant).

All reports of ADE and SADEs will be kept in the local KCRI file.

8.4. Type and duration of the follow-up of subjects after adverse events:

ADEs and SADEs reported up to 7 days after the administration of the Cytosponge™ will be recorded and reviewed. All participants receiving the Cytosponge™ will receive a telephone call at 7 days to assess any ADEs.

All SADEs occurring from the time of Cytosponge™ administration (up to 7 days post) will be reported within 24 hours of the PI becoming aware.

9. STATISTICS

9.1. Statistical methods to be employed:

The following outcomes arise from binary responses:

- Proportion of approached subjects who agree to the study in principle;
- Proportion of approached subjects who attempt Cytosponge™ swallow;
- Proportion of participants with swallow attempts who successfully swallow;
- Dysplasia proportion

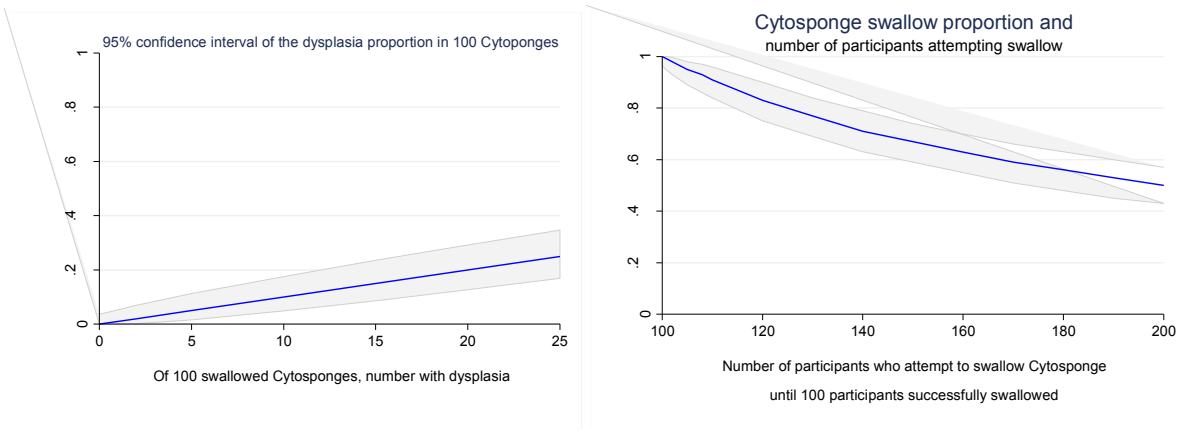
For all the above the above, the full distribution will be provided and exact 95% confidence intervals on the binomial proportion will be calculated. Differences in proportions will be tested using chi-squared tests between different groups (e.g. males and females, drinkers of beverages above and below median temperatures, age (<50, 50+) and HIV status.

The post-device swallowing satisfaction score is an ordinal variable (scale of 1 to 10). The distribution of this score will be provided and compared to previous distributions using chi-squared significance

tests. We will also analyse differences in the score by the following factors, age (under or over 50 years), gender and HIV status, using ordinal logistic regression models for this ordinal outcome.

9.2. Number of subjects and justification:

We aim to estimate the proportion of Cytosponge™-detected dysplasia with a 95% confidence interval of ± 10%, which will be achieved through 100 participants (e.g. a 15% dysplasia prevalence will have a 95% CI of 9% to 24%). In the plot below (left) the confidence interval around the dysplasia prevalence is shown and is < ± 10% in the prevalence range expected, which was set to be at or below the findings in a previous study conducted in Bomet, Kenya with 14.4% dysplasia (20% in men over 50). In order to achieve 100 successfully swallowed sponges, the number of participants approached will exceed this number by up to 2-fold and the confidence limits around this successful swallow proportion are narrower (below (right)).



9.3. Level of significance:

A two-sided P-value <0.05 will denote statistical significance.

9.4. Criteria for termination of trial:

The trial will be terminated when the participant age and gender frequency grid has been filled, and thus a sufficient number of sponge samples (100) have being collected.

9.5. Procedure for accounting for missing, unused and spurious data:

The use of the RedCap platform for data collection will limit the opportunity missing data. The questionnaire and CRF will be programmed so that progress to the next question cannot be made if a required field is incomplete. Similarly, constraints will be programmed to prevent nonsensical entries being made. All data will be monitored periodically as it is uploaded to servers so that erroneous entries can be investigated in a timely fashion. The joint consultation on pathological findings will help to ensure their validity.

9.6. Procedures for reporting deviations from statistical plan:

No deviation is anticipated for this feasibility study, for which the statistical methods are straightforward. All statistical methodologies will be reported in detail in journal publications.

9.7. Selection of study participants to be included in analyses:

For this feasibility study, all participants will be included in the analysis. Stratified analyses will only be performed by gender and broad age range. The study will not be sufficiently powered for a more detailed analysis.

10. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

The investigators and institutions hereby confirm that the TFDA will be permitted to conduct inspections and be granted direct access source data/documents.

11. QUALITY CONTROL AND QUALITY ASSURANCE

11.1. Training:

A dedicated research nurse from the MRC-CU (Irene Debiram-Beecham) with extensive experience in administering the Cytosponge™ and histopathologist Dr Maria O'Donovan, who has developed and disseminated SOPs for preparing FFPE blocks from cells harvested from the device, will travel to KCRI to conduct an intensive on-site training program of two primary components:

1) Safely and effectively administering the Cytosponge™: intensive on-site training to be delivered by Irene Debiram-Beecham to the local KCRI nurse responsible for using the device in the local trial.

2) Processing Cytosponge™ cells: training to be delivered by Martin Bromwich and Dr Maria O'Donovan, who has developed and optimised the SOPs and been involved with Cytosponge since its inception. Local scientific and technical staff will learn how to release sponge cells into solution and form a cellular clot to be formalin fixed and embedded in paraffin.

11.2. Pathology examination:

Cytological examination of slides will be conducted independently by three pathologists – one from each partner institution. Disagreements will then be resolved by joint consultation. This component will benefit substantially from the involvement of Dr Maria O'Donovan who has developed and optimised the SOPs and been involved with Cytosponge™ since its inception. Dr Maria O'Donovan will carry out training of pathologists to read these sample types and help develop the histopathology reporting format. All slides will be scanned at high resolution and stored on an IARC-hosted slidepath viewing system and shared between the pathology reviewers.

12. ETHICS

12.1. Existing approval:

This study has been approved by the IARC Ethics Committee and the National Institute for Medical Research (NIMR). Please see appendix P2-P3 for copies of the approval letters. Please note: due to changes in the study design, amendments to these approvals will be required in the following areas:

- Information leaflet and consent form

- Recruitment strategy
- Questionnaire and CRF content
- Participant reimbursement
- Participant follow-up

12.2. Choice of investigators:

Investigators from the three collaborating institutions were selected on the basis of their successful track record in clinical and epidemiological research and project expertise. Drs Mwasamwaja and Mmbaga both have essential experience in the fields of clinical research and cancer epidemiology and Dr Mwasamwaja is a highly experienced endoscopist in addition to possessing vital knowledge of the local research setting. Drs Mwasamwaja and Mmbaga will be supported by a local team of fieldwork staff, endoscopy nurses and a pathologist (Dr Alex Mremi).
Drs McCormack and Schüz share a wealth of knowledge and experience in conducting international cancer epidemiology research, particularly in East Africa. Dr Middleton has relevant project management and scientific expertise and extensive fieldwork experience including numerous overseas visits. Drs McCormack, Middleton and Schüz will receive the support of a pathologist (Dr Behnoush Abedi-Ardekani) and a team of genetic (Drs Jiri Zavadil and Michael Korenjak) and epigenetic (Drs Zdenko Herceg and Fazlur Rahman Talukdar) experts.
Prof Fitzgerald, having designed and developed the device, has a wealth of experience in using the Cytosponge™ for research and has seen a number of successfully implemented trials reach their conclusion. Prof Fitzgerald has years of experience as a gastroenterologist and international expert in cancers of the upper-digestive tract. Prof Fitzgerald will also have the support of a Research Governance and Integrity Coordinator (Dr. Jennifer Furman), a histopathologist (Dr Maria O'Donovan, who has been involved in the development of Cytosponge™, since its inception) and an interdisciplinary team of scientists, laboratory and technical experts and research nurses.

12.3. Monitors and monitoring plan:

Monitoring responsibilities will be met locally by Dr Sokoine Kivuyo, and remotely/internationally by Dr Partha Basu. In brief, Dr Kivuyo will satisfy local monitoring responsibilities with frequent visits to the Majengo Healthcare Centre during recruitment and Cytosponge™ collections. Dr Basu will maintain regular contact with Dr Kivuyo and undertake periodic checks of data stored on online servers. The specific monitoring responsibilities are listed below and followed by a proposed monitoring plan.

- Ensuring the eligibility of enrolled participants
- Auditing the informed consent procedure
- Ensuring that research staff are following protocol procedures.
- Monitoring the supply, storage and shelf life of Cytosponge™ devices
- Ensuring that the device is being administered correctly to eligible participants only
- Maintaining logs and inventories of the devices used in the study
- Returning of unused devices, or those suspected to be faulty to the MRC-CU
- Supplying research staff with the correct versions of the Investigator's Brochure and Protocol
- Checking the accuracy, consistency, and completeness of entries on study questionnaires and CRFs

- Overseeing the accurate and timely reporting of adverse events in concordance with the Protocol and Investigator's Brochure
- Checking the CRFs for the correct reporting of missed Cytosponge™ appointments, failures to swallow and withdrawals
- Alerting the research staff of deviations from the protocol or good clinical practice and initiating the necessary training to prevent recurrences
- Notifying the wider study team of noncompliance that significantly affects or has the potential to significantly affect human subject protection or reliability of study results/outcomes

Study launch/site initiation visit:

Following the approval of the study by the TFDA, and the approval of the necessary protocol amendments by the IEC and NIMR, a study launch visit will be hosted in Moshi by KCRI and attended by PIs and Co-Investigators from IARC and MRC-CU. The purpose of this visit will be to consolidate the familiarisation of local research staff with the protocol and study procedures and initiate participant recruitment under the supervision of MRC-CU researchers, who have a wealth of experience in conducting similar trials. Specific activities will be undertaken as follows:

- Tour of local facilities: Cytosponge™ supplies, recruitment areas, interview rooms, private Cytosponge™ collection rooms, sample processing labs and sample storage spaces
- Study group meeting including presentations of previous trial experiences from MRC-CU and final group-wide training
- Local community visits
- Initial recruitment of participants and observed interviews
- Initial Cytosponge™ collections under the training and supervision of MRC-CU research nurse Irene Debarim-Beecham as per the specific SOP (see Investigator's Brochure Appendix IB1).
- Intensive laboratory training and preparation of the first FFPE blocks with collected Cytosponge™ sample sunder the supervision of Martin Bromwich and Maria O'Donovan

Interim/routine monitoring visits:

Dr Kivuyo will undertake local monitoring activities to ensure that: (i) the conduct of the trial is in compliance with the currently approved protocol, GCP guidelines, and IRB regulatory requirements; (ii) data (including source documents and consent forms) are correctly input, maintained and original and (iii) that the site has the adequate resources to continue the study. Regular contact with Dr Basu at IARC and period meetings with the wider study team will enable ample troubleshooting opportunities.

Closeout:

After the completion of recruitment, a closeout visit will be undertaken to dispose of remaining Cytosponge™ devices, ensure the correct filing for longer term storage of paper records and package/ship FFPE Cytosponge™ samples for analysis in the UK and France. Any equipment loaned to KCRI will be returned to the sponsor. A review meeting will be held to inform future studies. After

this initial pilot, and subject to funding, a larger recruitment of 500-1000 participants will be undertaken as per a protocol submitted to the TFDA as an amendment.

12.4. How additional staff will maintain patient confidentiality, follow protocol and abide by ethical and TFDA requirements:

By using the secure RedCap data collection platform, all information related to participants will be immediately uploaded to a secure server. Downstream laboratory analysis will be conducted on samples marked only with the unique ID and QR code and technicians will be blind to additional information. By receiving comprehensive on-site training and having access to the Investigator's Brochure, staff will have all information available to adhere to protocol procedures. Contact details for trial investigators will be available for remote support at all times during the trial.

12.5. Insurance and indemnity measures:

The study has insurance coverage for research related injuries, disabilities and malpractice; with the policy as shown in the attached note from the NIC. All patients will have insurance in case of evens. In case of any illness associated with the procedure the patient will be taken for treatment at a tertiary hospital. In case of death while on study and related to the study the insurance coverage will pay compensation to the family.

12.6. Patient information leaflets and informed consent forms:

All participants will be given a detailed information leaflet and be required to sign an informed consent form for them to be eligible for the study. These documents are presented in Appendices P4-P5.

12.7. Treatment and/or management of participants and their disease conditions after completion of trial:

For patients found to have any degree of ESD in Cytosponge™ cells, the study will pay for initial clinical investigation – standard endoscopy procedure at KCMC endoscopy unit (under the supervision of Dr G. Maro) - and histopathology. If no abnormalities are found, these individuals will be called back 6 months later for a repeat of this procedure.

12.8. Institutional ethics committee capacity to monitor site:

The KCMUCo CRERC, which is the local IRB, has capacity to monitor and evaluate safety, ADEs and unexpected events involving risks to subjects or others as well as ethics complaints or ADE reports submitted by the PI. Additionally, the National Health Research Ethics Review committee from the NIMR secretariat conducts regular monitoring and evaluation of studies.

12.9. Explanation if minimum compensation is not being provided:

Not applicable – participants will receive reimbursement (20,000 TZS) for the cost of travel/their time.

12.10. Follow-up of participants after trial:

In the event of finding high or low grade ESD with the Cytosponge™, participants will be informed and invited to undergo chromoendoscopy at KCMC. The costs of transport, endoscopy and any subsequent treatment required will be provided from the research budget following international ethical guidelines. Confirmed cases of low grade ESD will be advised to undergo repeat endoscopy 12 months later. Cases of high grade ESD will be invited to undergo treatment under the supervision of Dr Michael Mwachiro from Tenwek hospital in Bomet, Kenya. Treatment for high grade ESD will consist of either endoscopic mucosal resection or radiofrequency ablation, depending on the extent and severity of legions. This will either take place at KCMC under Dr Mwachiro's supervision or, if transport to Tenwek is necessary, participants will be covered for travel and treatment by the sponsor.

12.11. Material Transfer Agreement (MTA):

The transfer of oesophageal paraffin embedded biopsies to IARC from KCRI is covered by an existing MTA as per the existing overarching ESCCAPE research project. This MTA is in attached as a separate document (*MTA_IARC_KCRI*). An MTA for the transfer of samples is currently being prepared to cover transfer between KCRI and MRC-CU. The TFDA will be sent a copy of this MTA prior to the transfer of samples and the necessary permits will be obtained.

13. DATA HANDLING AND RECORD KEEPING

Data will be collected on a mobile application developed for this study and uploaded to a secure server in real-time. Participants will be assigned an anonymous ID number and no personally-identifiable information will be stored on the server. For adults to complete the Cytosponge™, locally we will record the name, physical address and telephone number of the participant and a next-of-kin so that we could re-contact participants if the need arises. For the purpose of participant follow-up, if necessary, names and contact details will be stored securely at KCRI.

14. PUBLICATION POLICY

Findings from this study will be published in international peer-reviewed academic journals with authorship contributions from all partner institutions.

References cited:

1. Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, et al. Cancer incidence and mortality worldwide: Sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer. 2015;136(5):E359-E86.
2. Sewram V, Sitas F, O'Connell D, Myers J. Diet and esophageal cancer risk in the Eastern Cape Province of South Africa. Nutr Cancer. 2014;66(5):791-9.
3. Somdyala NI, Parkin DM, Sithole N, Bradshaw D. Trends in cancer incidence in rural Eastern Cape Province; South Africa, 1998-2012. Int J Cancer. 2014.
4. Ahmed AA. The surgical management and outcome of oesophageal cancer in Addis Ababa. Ethiop Med J. 2000;38(3):147-52.

5. Middleton DR, Bouaoun L, Hanisch R, Bray F, Dzamalala C, Chasimpha S, et al. Esophageal cancer male to female incidence ratios in Africa: a systematic review and meta-analysis of geographic, time and age trends. Cancer Epidemiol. 2018.
6. Dawsey SP, Tonui S, Parker RK, Fitzwater JW, Dawsey SM, White RE, et al. Esophageal cancer in young people: a case series of 109 cases and review of the literature. PLoS One. 2010;5(11):e14080.
7. White RE, Parker RK, Fitzwater JW, Kasepoi Z, Topazian M. Stents as sole therapy for oesophageal cancer: a prospective analysis of outcomes after placement. Lancet Oncol. 2009;10(3):240-6.
8. McCormack VA, Menya D, Munishi MO, Dzamalala C, Gasmelseed N, Leon Roux M, et al. Informing etiologic research priorities for squamous cell esophageal cancer in Africa: A review of setting-specific exposures to known and putative risk factors. International Journal of Cancer. 2016.
9. BURRELL RJ. Esophageal cancer among Bantu in the Transkei. J Natl Cancer Inst. 1962;28:495-514.
10. Coetzee C. Carcinoma of the Oesophagus. South Africa Journal of Surgery. 1966;4(3).
11. Linsell CA. Cancer incidence in Kenya 1957-63. Br J Cancer. 1967;21(3):465-73.
12. McGlashan ND. Oesophageal cancer and alcoholic spirits in central Africa. Gut. 1969;10(8):643-50.
13. Islami F, Kamangar F, Nasrollahzadeh D, Moller H, Boffetta P, Malekzadeh R. Oesophageal cancer in Golestan Province, a high-incidence area in northern Iran - a review. Eur J Cancer. 2009;45(18):3156-65.
14. Qiao YL, Dawsey SM, Kamangar F, Fan JH, Abnet CC, Sun XD, et al. Total and cancer mortality after supplementation with vitamins and minerals: follow-up of the Linxian General Population Nutrition Intervention Trial. J Natl Cancer Inst. 2009;101(7):507-18.
15. Sewram V, Sitas F, O'Connell D, Myers J. Tobacco and alcohol as risk factors for oesophageal cancer in a high incidence area in South Africa. Cancer Epidemiol. 2016;41:113-21.
16. Mwachiro MM, Burgert SL, Lando J, Chepkwony R, Bett C, Bosire C, et al. Esophageal squamous dysplasia is common in asymptomatic Kenyans: a prospective, community-based, cross-sectional study. The American journal of gastroenterology. 2016;111(4):500.
17. Roshandel G, Merat S, Sotoudeh M, Khoshnia M, Poustchi H, Lao-Sirieix P, et al. Pilot study of cytological testing for oesophageal squamous cell dysplasia in a high-risk area in Northern Iran. Br J Cancer. 2014;111(12):2235-41.
18. Paterson AL, Lao‐Sirieix P, O'donovan M, Debiram‐Beecham I, Pietro M, Miremadi A, et al. Range of pathologies diagnosed using a minimally invasive capsule sponge to evaluate patients with reflux symptoms. Histopathology. 2017;70(2):203-10.
19. Chettouh H, Mowforth O, Galeano-Dalmau N, Bezawada N, Ross-Innes C, MacRae S, et al. Methylation panel is a diagnostic biomarker for Barrett's oesophagus in endoscopic biopsies and non-endoscopic cytology specimens. Gut. 2017:gutjnl-2017-314026.
20. Lao-Sirieix P, Rous B, O'donovan M, Hardwick RH, Debiram I, Fitzgerald RC. Non-endoscopic immunocytological screening test for Barrett's oesophagus. Gut. 2007;56(7):1033-4.
21. Kadri SR, Lao-Sirieix P, O'Donovan M, Debiram I, Das M, Blazeby JM, et al. Acceptability and accuracy of a non-endoscopic screening test for Barrett's oesophagus in primary care: cohort study. Bmj. 2010;341:c4372.
22. Ross-Innes CS, Debiram-Beecham I, O'Donovan M, Walker E, Varghese S, Lao-Sirieix P, et al. Evaluation of a minimally invasive cell sampling device coupled with assessment of trefoil factor 3 expression for diagnosing Barrett's esophagus: a multi-center case–control study. PLoS medicine. 2015;12(1):e1001780.
23. Roshandel G, Merat S, Sotoudeh M, Khoshnia M, Poustchi H, Lao-Sirieix P, et al. Pilot study of cytological testing for oesophageal squamous cell dysplasia in a high-risk area in Northern Iran. Br J Cancer. 2014;111(12):2235-41.