



**QUEEN'S
UNIVERSITY
BELFAST**

Blood Biomarkers for Early Detection of Oesophageal Cancer: A Systematic Review

Kunzmann, A. T., McMenamin, U. C., Spence, A. D., Gray, R. T., Murray, L. J., Turkington, R. C., & Coleman, H. G. (2018). Blood Biomarkers for Early Detection of Oesophageal Cancer: A Systematic Review: A systematic review. *European Journal of Gastroenterology and Hepatology*, 30(3), 263-273.
<https://doi.org/10.1097/MEG.0000000000001029>

Published in:
European Journal of Gastroenterology and Hepatology

Document Version:
Peer reviewed version

Queen's University Belfast - Research Portal:
[Link to publication record in Queen's University Belfast Research Portal](#)

Publisher rights
© Wolters Kluwer Health In. All rights reserved.
This work is made available online in accordance with the publisher's policies. Please refer to any applicable terms of use of the publisher.

General rights
Copyright for the publications made accessible via the Queen's University Belfast Research Portal is retained by the author(s) and / or other copyright owners and it is a condition of accessing these publications that users recognise and abide by the legal requirements associated with these rights.

Take down policy
The Research Portal is Queen's institutional repository that provides access to Queen's research output. Every effort has been made to ensure that content in the Research Portal does not infringe any person's rights, or applicable UK laws. If you discover content in the Research Portal that you believe breaches copyright or violates any law, please contact openaccess@qub.ac.uk.

BLOOD BIOMARKERS FOR EARLY DIAGNOSIS OF OESOPHAGEAL CANCER: A SYSTEMATIC REVIEW

Running head: Oesophageal cancer biomarkers review

Authors: Andrew T Kunzmann¹, Úna C McMenamin¹, Andrew D Spence¹, Ronan T Gray¹, Liam J Murray¹, Richard C Turkington² and Helen G Coleman¹

Authors' affiliations: ¹Cancer Epidemiology and Health Services Research Group, Centre for Public Health, Queen's University Belfast, Belfast, Northern Ireland, United Kingdom

²Centre for Cancer Research and Cell Biology, Queen's University Belfast, Belfast, Northern Ireland, United Kingdom

Corresponding author: Dr Andrew Kunzmann. Institute of Clinical Sciences Block B, Queen's University Belfast, Royal Victoria Hospital, Belfast, Northern Ireland, BT12 6BA.

Phone: +44 (0) 28 9063 5009; Fax: +44 (0) 28 90235900; Email: a.kunzmann@qub.ac.uk

Disclosures: The project was kindly funded by **Ochre charity** (Registered charity number: SC032343).

The authors have no conflicts of interest to report.

Word count: 3445

ABSTRACT

Background: Oesophageal cancer prognosis remains poor due to the inability to detect the disease at an early stage. Non-tissue (serum, urinary or salivary) biomarkers potentially offer less invasive methods to aid early detection of oesophageal cancer. We aimed to systematically review studies assessing the relationship between non-tissue biomarkers and subsequent development of oesophageal cancer.

Methods: Using terms for biomarkers and oesophageal cancer, MEDLINE, EMBASE and WEB OF SCIENCE were systematically searched for longitudinal studies, published until April 2016, which assessed the association between non-tissue biomarkers and subsequent oesophageal cancer risk. Random effects meta-analyses were used to calculate pooled relative risk (RR) and 95% confidence intervals (CI), where possible.

Results: A total of 39 studies were included. Lower serum pepsinogen I concentrations were associated with an increased risk of oesophageal squamous cell carcinoma (n=3 studies, pooled RR=2.20, 95% CI: 1.31-3.70). However, the association for the pepsinogen I:II ratio was not statistically significant (n=3 studies, pooled RR=2.22, 95% CI: 0.77-6.40), with a large degree of heterogeneity observed ($I^2=68.0\%$). Higher serum glucose concentrations were associated with a modestly increased risk of total oesophageal cancer (n=3 studies, pooled RR=1.27, 95% CI: 1.02-1.57). No association was observed for total cholesterol and total oesophageal cancer risk (n=3 studies, pooled RR=0.95, 95% CI: 0.58-1.54). Too few studies assessed other biomarkers for meta-analyses.

Conclusions: Serum pepsinogen I concentrations could aid early detection of oesophageal squamous cell carcinoma. More prospective studies are needed to determine the use of other non-tissue biomarkers in the early detection of oesophageal cancer.

Keywords: serum; urinary; oesophageal adenocarcinoma; oesophageal squamous cell carcinoma

INTRODUCTION

Oesophageal cancer has a very poor prognosis, with less than 20% of patients surviving for more than 5 years from diagnosis [1,2]. Poor survival is attributable to late clinical presentation with advanced disease, with less than a quarter of cases diagnosed at stage I or II [3,4].

The two main histological subtypes, oesophageal adenocarcinoma (OAC) and oesophageal squamous cell carcinoma (OSCC), differ by aetiology and geographical incidence, with OAC more common in Western populations (>60% of oesophageal cancers) and OSCC more common (>90%) in Eastern populations, but share a poor prognosis [2,5,6].

Despite the high sensitivity of endoscopy as a diagnostic tool, a national screening programme for oesophageal cancer has not been justified due to the high cost, the invasiveness of the procedure and the psychological burden [7,8]. Cytosponge screening for Barrett's oesophagus and oesophageal cancer is currently being trialled in patients with heartburn symptoms [9,10], but otherwise the potential for viable screening options seems limited. Whilst endoscopy can also identify patients with Barrett's oesophagus (BO), the precursor lesion of OAC, or squamous dysplasia, the precursor lesion of OSCC, risk-stratification tools for identifying individuals at highest risk of neoplastic progression from limited. Surveillance programmes amongst BO patients place a considerable strain on gastroenterology resources and patients due to the limited risk-stratification[7].

Identification of cheaper, less invasive methods to identify individuals at risk of developing oesophageal cancer are needed to improve screening and surveillance options within the general public and patients with precancerous lesions, respectively [7]. Whilst tissue biomarkers offer promise in the early detection of oesophageal cancer, blood, urine or salivary tests could offer a simpler and less invasive alternative.

A large number of epidemiological studies have assessed the role of non-tissue biomarkers in oesophageal cancer risk [11,12]. However, many of the studies have been cross-sectional/retrospective, whereby the biomarker was used as an aid to oesophageal cancer diagnosis. Prospective studies are needed to determine if the changes in biomarkers occur early enough in cancer development to identify high risk individuals and aid early diagnosis.

An examination of biomarkers associated with oesophageal cancer may also provide insight into the mechanisms and aetiology of oesophageal cancer development.

The aim of this systematic review is to summarise evidence from all prospective studies which have assessed the relationship between blood, urinary and salivary biomarkers and risk of oesophageal cancer, in order to identify potential aides for risk stratification and early diagnosis of oesophageal cancer.

METHODS

LITERATURE SEARCH

To investigate blood, urine and salivary based biomarkers in the early diagnosis of oesophageal cancer, three databases, MEDLINE (1947-), EMBASE (1974-) and WEB OF SCIENCE (1980-), were systematically searched from inception to 29th April 2016 for primary studies in humans, published in any language. Databases were searched using exploded subject headings and combinations of keywords for biomarkers and oesophageal cancer (see Supplementary table 1 for full search terms). Reference lists and manual searches were used to identify any additional published studies.

ELIGIBILITY CRITERIA

Titles and abstracts of articles identified through the electronic search were reviewed independently, in duplicate, by two reviewers (from AK, HC, UM, RG, AS). The full text of articles for possible inclusion were then independently reviewed by two investigators (from AK, HC, UM, RG, AS). If there was disagreement between the reviewers at either stage, an independent review was undertaken by a third reviewer and a consensus reached.

Criteria for inclusion in the review included: assessment of a non-tissue (blood, urinary or salivary) based biomarker (including circulating epigenetic markers) in relation to total oesophageal cancer (any histological type), OAC or OSCC risk. Studies reporting only on oesophagogastric junction cancers or high grade dysplasia of the oesophagus, or not separating these from oesophageal tumours were not included. Included studies had to have at least a 6 month follow-up period after biomarker assessment

(to exclude prevalent cancers with diagnostic delays). Cross sectional studies where the biomarkers were measured at, or close to, the time of diagnostic assessment or following diagnosis were excluded. To ensure a narrow scope and to minimise repetition of existing systematic reviews,

oesophageal tissue biomarkers, blood-based genetic polymorphisms and infectious agents were not eligible for inclusion [13,14]. Studies using diagnoses of medical conditions in medical records as a proxy for biomarker status were not included due to potential differences in the exact biomarker measurement used for diagnosis (for example either high total cholesterol or high low-density lipoprotein (LDL) cholesterol could be used to diagnose hypercholesterolemia). Studies of biomarkers and oesophageal cancer mortality were not included. Participants included in individual studies could be healthy at biomarker assessment or have oesophageal conditions other than oesophageal cancer, such as Barrett's oesophagus.

DATA EXTRACTION

Data extraction was performed by an experienced author (AK). Extracted information from each of the individual studies included: author names and date of publication, study setting; study population, participant demographics and baseline characteristics; biomarkers assessed as well as the timing and method of assessment, the sample type; study methodology; recruitment and study completion rates and outcomes of interest. Articles in non-English languages were translated where necessary. The Newcastle-Ottawa scale for Cohort studies was used to assess study quality [15].

STATISTICAL ANALYSIS

Stata 14 software (Stata Corporation, College Station, TX, USA) was used for data analysis. Random effects meta-analyses were used to examine the pooled association between each biomarker and each type of oesophageal cancer (total, OAC or OSCC) if at least 3 eligible studies had assessed the association. Adjusted results were used where possible. A chi-squared test for heterogeneity was calculated and the I^2 statistic determined, to estimate the proportion of variation between study results attributable to heterogeneity rather than chance [16]. Heterogeneity was considered high if I^2 statistic was above 75% [16]. If studies only provided results stratified by participant characteristics (such as gender), the results were combined using preliminary meta-analysis prior to entry into the overall meta-analysis. Due to low study numbers identified for each biomarker,

publication bias could not be formally assessed [17]. Where studies included in meta-analyses used multiple classifications, or cut-offs for biomarkers concentrations (e.g. quartiles, categorical and continuous), we chose results based on similarity of classification to other studies (the majority of studies used tertiles/quartiles).

RESULTS

The searches identified a total of 15,334 study records (Medline=3210, Embase=6414, Web of Science=5710). After removal of duplicates, 6,995 studies remained. After a review of titles and abstracts, 82 articles were chosen for full text review, from which 36 studies were eligible for inclusion. An additional three studies eligible for inclusion were identified by manual searches and review of reference lists [18–20], meaning a total of 39 studies [18–56] were included in the review (Figure 1 and Table 1) and are further described by broad classification of biomarker type.

Fifteen studies were conducted in Eastern or South-East Asia populations, 12 studies were conducted in European populations and eight studies were conducted in North American populations. Seven studies were conducted amongst males only. The studies were mostly conducted within cohorts with a mean BMI below 25, though some studies were conducted amongst cohorts with Barrett's oesophagus at baseline and had average BMI above 25 (Table 1). Most studies were judged to be of a good (n=31 studies) or fair quality (n=6 studies), though two studies assessing exposure to toxic chemical within worker cohorts were judged to be poor quality (Table 2).

Lipid & glucose related biomarkers

We identified seven studies that assessed the association between serum lipid or glucose biomarkers and risk of oesophageal cancer (Supplementary table 2).

A meta-analysis of five studies [18,19,21–23] found no association between total serum cholesterol concentrations and risk of total oesophageal cancer (pooled RR=1.00, 95% CI: 0.79-1.27, $I^2=36.8\%$, Figure 2). Additionally, Lindkvist et al [54] provided a breakdown by type of oesophageal cancer and found no association between high total cholesterol and risk of either OAC or OSCC, (Supplementary table 2).

Wulaningsih et al [23] also tested subtypes of cholesterol related measures, though could not adjust for body mass index. Strong inverse associations with risk of oesophageal cancer were found for LDL

cholesterol, the LDL:HDL ratio and the Apo B:ApoA-1 ratio in that study. No associations were seen for HDL cholesterol, total:HDL cholesterol ratio, the triglyceride:HDL ratio, ApoB, or Apo A concentrations. A positive association was observed for triglyceride concentrations as individuals in the highest quartile of triglyceride concentrations had over double the risk of oesophageal cancer. However, Lindkvist et al [54] found a weaker association for triglycerides, which was not statistically significant for either OAC or OSCC. Ahn et al [21] found a non-significant positive association between HDL cholesterol concentrations and risk of oesophageal cancer.

Wulaningsih et al [23], Stocks et al[20] and Jee et al[52] all assessed the association between glucose concentrations and oesophageal cancer risk. A meta-analysis of these three studies found a 27% increase in risk of total oesophageal cancer in individuals with higher glucose concentrations (pooled RR=1.27, 95% CI: 1.02-1.57) with no evidence of heterogeneity ($I^2=0.0\%$) (Figure 3).

Only Lindkvist et al[54] provided a breakdown by cancer type with the results indicative that glucose concentrations were more strongly associated with OSCC risk than OAC risk.

Duggan et al[55] assessed the association between HOMA (Homeostatic model assessment) scores, calculated by multiplying insulin and glucose concentrations, and risk of oesophageal adenocarcinoma amongst a cohort of individuals with Barrett's oesophagus and found evidence of an association when assessed continuously (RR=1.64, 95% CI: 1.08-2.48).

Nutrition-related biomarkers

We identified 16 studies that assessed the association between nutrition-related biomarkers and oesophageal cancer (Supplementary table 3).

Eight studies assessed the role of nutrition-related biomarkers and total oesophageal cancer risk. Kimm et al [35] found that individuals with higher aspartate transaminase: alanine transaminase (AST:ALT) ratios (a biomarker associated with liver damage often caused by alcohol consumption)

had a higher risk of oesophageal cancer. Tsuboya et al [46] found no association between gamma-glutamyltransferase concentrations (associated with excessive alcohol intake and liver disease) and risk of total oesophageal cancer. Knekt et al [56] did not find an association for beta-carotene and the others have not been investigated previously. No associations were observed for alpha-tocopherol [56], selenium [56], retinol [56], tea polyphenols [36], calcium [33] and phosphate [34] and risk of total oesophageal cancer.

Eight studies assessed the role of nutrition-related biomarkers in OSCC risk and three studies assessed the role of nutrition-related biomarkers in OAC risk. Murphy et al [43] found a significant reduction in OSCC risk amongst individuals with higher serum cysteine concentrations. Fanidi et al [42] found a significant increase in OSCC risk amongst individuals with higher serum folate concentrations but not other b-vitamins, or one-carbon products (homocysteine/methionine). Some of the results from studies assessing OSCC risk contrast with the results from separate studies of total oesophageal cancer described earlier, with significant inverse associations seen for selenium [41] and alpha-tocopherol [38], but null results for beta-carotene and beta-cryptoxanthin [39]. Null associations were also seen in individual studies for vitamin C, retinol and lutein [39,40,56]. Chen et al [53] found evidence of an association between serum 25(OH)D and OSCC, however, Abnet et al [32] did not find an association with OSCC or OAC in an analysis of data from 8 prospective cohorts. Takata et al [37] also found no association between serum selenium concentrations and risk of progression to OAC amongst a cohort of individuals with Barrett's oesophagus.

Metabolic/digestive related biomarkers

We identified nine studies identified that assessed the association between biomarkers related to metabolism or inflammation and oesophageal cancer risk (Supplementary table 4).

A random effects meta-analysis of 3 studies [27–29] found that lower pepsinogen I (PGI) concentrations were associated with increased risk of OSCC (pooled RR=2.20, 95% CI: 1.31-3.70), with little heterogeneity observed ($I^2=9.3%$) (Figure 4).

The same studies [27–29] also assessed the association between the PGI: PGII ratio and risk of OSCC but did not find evidence of an association (pooled RR=1.44, 95% CI: 0.42-4.91), with a large degree of heterogeneity ($I^2=75.7\%$) (Figure 5). Sensitivity analyses indicated that heterogeneity was lowered after the exclusion of results from Cook et al [27] (pooled RR=0.91, 95%CI: 0.37-2.20, $I^2=18.3\%$).

Duggan et al [55] found a significant positive linear association between serum leptin concentrations, but not adiponectin, and risk of OAC in individuals with Barrett's oesophagus, including when limiting to cancers diagnosed within 3 years of baseline blood sampling (HR=2.51, 95% CI: 1.09-5.81). However, the association was only statistically significant in continuous analysis and has yet to be tested in other studies. No studies identified assessed leptin and adiponectin and OSCC risk.

De Martel et al [24] found a borderline association between high serum ghrelin concentrations and lower OAC risk in Barrett's oesophagus patients. Similarly, Murphy et al [30] found lower ghrelin levels were associated with an increased risk of OSCC.

Inflammation-related biomarkers

Keeley et al [31] found evidence of strong positive associations between various inflammation-related biomarkers, including Tumour Necrosis Factor (TNF)-alpha, TNF-beta, MCP-3 (Monocyte chemotactic protein), IL-1R, Interleukin (IL)-13 and GM-CSF (granulocyte/macrophage colony-stimulating factor) , and OSCC risk.

Two studies assessed the association between inflammation-related biomarkers in Barrett's oesophagus cohorts. Hardikar et al [25] found no evidence of an association between C-Reactive Protein, IL6, Isoprostane, TNF-alpha or TNF-beta and risk of progression to OAC. Siahpush et al [26] found no evidence of an association between insulin-like growth factor 1 and risk of OAC.

Miscellaneous biomarkers

Results of individual studies examining the association between miscellaneous biomarkers and risk of oesophageal cancer are listed in Supplementary table 5. Haemoglobin was not associated with risk of oesophageal cancer, even when limiting the time from measurement to diagnosis to 3 years, although there was evidence of a trend which suggested a stronger association when limiting to oesophageal cancer diagnoses within a year of biomarker measurement [45].

Leukocyte telomere length was found to have a strong association with risk of progression to oesophageal adenocarcinoma amongst a cohort of individuals with Barrett's oesophagus in one study [49] but requires confirmation in other studies. However, lymphocyte mutagen sensitivity, a marker for the ability to repair DNA damage, was not found to be associated with risk of progression amongst the same Barrett's oesophagus cohort [50].

Biomarkers of smoking behaviours such as total N'-nitrosonornicotine (NNN), a tobacco-specific carcinogen, were found to be associated with risk of oesophageal cancer amongst male smokers in one study [44].

No association was found between sphingosines and risk of OSCC [51].

Exposure to toxins was also investigated, though neither urine levels of lead or trichloroethylene amongst workers in jobs at risk of exposure to these chemicals were found to be associated with oesophageal cancer risk [47,48].

DISCUSSION

This systematic review sought to examine non-tissue biomarkers associated with oesophageal cancer risk, in order to identify biomarkers that could be used to predict oesophageal cancer development. The review identified 39 studies which assessed the role of various biomarkers in oesophageal cancer risk. Meta-analyses revealed an association between serum PGI concentrations and OSCC risk (n=3 studies) and between serum glucose concentrations and total oesophageal cancer risk (n=3 studies). No associations were observed for total cholesterol and total oesophageal cancer risk (n=5 studies). However, there were too few studies to conduct meta-analyses for the majority of biomarkers and biomarkers with promising cross-sectional findings remain to be tested in prospective analyses.

The association between PGI concentrations was consistent across the three studies [27–29] included in the meta-analysis and indicated that individuals with lower PGI concentrations had over double the subsequent risk of OSCC. The association was similar between two studies in China and one in Finland (limited to males) suggesting geographical variation was not indicated. Similar effect sizes were observed in a meta-analysis of the same three studies assessing the association between the PGI:PGII ratio and OSCC risk, though the result was not statistically significant and heterogeneity was high. The results are supported by a study indicating an association between pepsinogen concentrations and risk of esophageal squamous dysplasia [57]. Lower PGI concentrations and a lower PGI:PGII ratio are thought to be indicators of gastric atrophy [58,59]. Lower PGI concentrations could be related to OSCC through bacterial overgrowth resulting from the reduced gastric acid production during gastric atrophy [60–62] or duodenal reflux [27,63]. Gastric atrophy is often caused by *Helicobacter pylori* (*H. pylori*) infections, and *H. pylori* infection has previously been linked to OSCC risk in Eastern populations [64]. However, the association between PGI and OSCC risk appears to be independent of *H. pylori* status as the studies included in the current review either adjusted for *H. Pylori* or did not find similar evidence of an association between *H. pylori* and OSCC

risk in separate analyses. Though serologic amnesia is possible, the results suggest that pepsinogen measurement could potentially aid OSCC risk-stratification independently of *H. pylori* measurement by identifying individuals at higher OSCC risk. PGI and the PGI:PGII ratio have previously been considered as part of gastric cancer screening programs in some countries [58,59,65]. However, questions remained about the cost-effectiveness. Should later studies confirm the results of the current meta-analysis, policy makers could assess the potential additional benefit of early OSCC detection when assessing whether to implement cancer screening programmes measuring pepsinogen concentrations/ratios, particularly in high-risk areas for OSCC [65].

The lack of an association between total cholesterol concentrations and risk of total oesophageal cancer in a meta-analysis of five studies, could be due to the different actions of each type of cholesterol. Wulaningsih et al [23] found stronger inverse association between more specific cholesterol markers, including LDL cholesterol, the ratio of LDL:HDL cholesterol and the ApoB:ApoA ratio, and oesophageal cancer risk but were unable to adjust for BMI which could confound the associations. Therefore, whilst an association between total cholesterol concentrations and oesophageal cancer risk appears unlikely, further detailed investigations into the association between more specific lipid biomarkers and oesophageal cancer risk independently of BMI seem warranted.

The association between serum glucose and total oesophageal cancer risk was modest (27%), but notably consistent across the three studies in the meta-analysis, despite geographical differences (Two studies from Europe and one study from South Korea). Two of the 3 studies included adjustment for BMI [54] or reported no change after adjustment for BMI [52], suggesting the results may be independent of BMI and could add predictive value to potential oesophageal cancer risk stratification models. The findings are consistent with the results of a systematic review which found a 27% increased risk of oesophageal cancer in individuals with diabetes mellitus in a pooled analysis of cohort studies [66]. However, the similarity could arise as the studies included in the current

review used measurements recorded in clinical databases so individuals may have been deemed to be at high risk of diabetes, meaning the results may not be representative of low-risk individuals. The result is also consistent with an observation by Duggan et al [55] of an association between HOMA scores, calculated by multiplying serum insulin and glucose concentrations, and risk of oesophageal adenocarcinoma. The potential mechanism mediating the association between serum glucose concentrations and oesophageal cancer could be oxidative stress related to hyperglycaemia or growth promotion by insulin-like growth factor (IGF) [67]. However, for the latter, it is important to note that no association was observed between IGF and OAC risk in a study by Siahpush et al [26] included in the current review, despite promising findings from non-prospective studies [68]. Only one study in the current review provided a breakdown by histological subtype of oesophageal cancer, therefore more studies will be needed to assess whether the associations with serum glucose are similar for both OAC and OSSC. Nevertheless, the review suggests that serum glucose concentrations could potentially aid risk stratification of individuals for oesophageal cancer risk, especially given the simplicity of blood glucose testing. However, a check of diabetes history may suffice and the modest strength of the association with oesophageal cancer risk means that the potential clinical benefit in terms of identifying high-risk individuals is likely to be limited unless incorporated into a wider panel of biomarkers or risk-stratification measures.

Despite the large number of biomarkers assessed, the majority of biomarkers have been tested by too few studies for strong conclusions to be formed, perhaps due to the difficulty in assessing rarer cancers such as oesophageal cancer in prospective studies. None of the studies identified assessed salivary biomarkers and very few assessed urinary biomarkers, despite promising findings for other gastrointestinal cancers [69]. For other novel blood markers, such as serum anti-p53 and micro RNAs, recent evidence from cross-sectional studies suggests that serum concentrations differ between oesophageal cancer patients and cancer-free controls [12,70,71], but no prospective studies have been conducted so these biomarkers were not included in this review. Further studies

are needed to assess non-tissue biomarkers given the public and clinical demand for blood based biomarkers [72,73].

The review provides a comprehensive assessment of the existing evidence of non-tissue biomarkers in oesophageal cancer risk and has identified promising associations between PGI concentrations and OSCC risk. A limitation was that many studies examined total oesophageal cancer risk without providing separate results for OAC and OSCC, despite the substantial differences between OAC and OSCC [74] in terms of biology, risk factors and patterns of incidence, possibly in order to maximise statistical power. Many of the included studies aimed to identify aetiological rather than early diagnostic biomarkers, which precluded an examination of the association with oesophageal cancers occurring within a period close to biomarker measurement (<3 years) useful for early detection [45]. Similarly, the biomarkers assessed were only measured at one point in time, which could also limit the ability to identify the optimal time-point of biomarker measurement

In summary, some non-tissue biomarkers, such as serum PGI concentrations may offer promise in identifying individuals at risk of oesophageal cancer which could help target screening or surveillance programmes and aid in the early detection of oesophageal cancer. However, too few studies have assessed the same biomarkers, and some promising biomarkers remain to be tested prospectively in order to adequately develop a biomarker panel for risk-stratification or for use in future oesophageal cancer screening programmes.

REFERENCES

1. Cancer research UK. Oesophageal cancer incidence statistics | Cancer Research UK [Internet]. [cited 2016 Oct 27]. Available from: <http://www.cancerresearchuk.org/health-professional/cancer-statistics/statistics-by-cancer-type/oesophageal-cancer/incidence#heading-Three>
2. Gupta B, Kumar N. Worldwide incidence, mortality and time trends for cancer of the oesophagus. *Eur J Cancer Prev.* 2016 Apr;1.
3. Bird-Lieberman EL, Fitzgerald RC. Early diagnosis of oesophageal cancer. *Br J Cancer.* 2009;101(1):1–6.
4. Cancer Research UK. Oesophageal cancer survival statistics [Internet]. [cited 2016 Oct 27]. Available from: <http://www.cancerresearchuk.org/health-professional/cancer-statistics/statistics-by-cancer-type/oesophageal-cancer/survival#heading-Zero>
5. Napier KJ, Scheerer M, Misra S. Esophageal cancer: A Review of epidemiology, pathogenesis, staging workup and treatment modalities. *World J Gastrointest Oncol.* 2014 May 15;6(5):112–20.
6. Arnold M, Soerjomataram I, Ferlay J, Forman D. Global incidence of oesophageal cancer by histological subtype in 2012. *Gut.* 2015 Mar;64(3):381–7.
7. Lao-Sirieix P, Fitzgerald RC. Screening for oesophageal cancer. *Nat Rev Clin Oncol.* 2012;9(5):278–87.
8. Gerson LB, Groeneveld PW, Triadafilopoulos G. Cost-effectiveness model of endoscopic screening and surveillance in patients with gastroesophageal reflux disease. *Clin Gastroenterol Hepatol.* 2004;2(10):868–79.
9. 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 Med.* 2015;12(1):e1001780.
10. MRC Cancer Unit. Barrett's ESophagus Trial 3 (BEST3) — MRC Cancer Unit [Internet]. 2016. Available from: <http://www.mrc-cu.cam.ac.uk/research/rebecca-fitzgerald/clinical-studies/BEST3>
 11. Zhang H, Xia J, Wang K, Zhang J. Serum autoantibodies in the early detection of esophageal cancer: a systematic review. *Tumor Biol.* 2015 Jan 30;36(1):95–109.
 12. Tan C, Qian X, Guan Z, Yang B, Ge Y, Wang F, et al. Potential biomarkers for esophageal cancer. *Springerplus.* 2016;5(1):467.
 13. Kunzmann AT, Graham S, McShane CM, Doyle J, Tommasino M, Johnston B, et al. The prevalence of viral agents in oesophageal adenocarcinoma and Barrett's oesophagus: A systematic review. *Eur J Gastroenterol Hepatol.* 2017;
 14. Li X, Gao C, Yang Y, Zhou F, Li M, Jin Q, et al. Systematic review with meta-analysis: The association between human papillomavirus infection and oesophageal cancer. *Aliment Pharmacol Ther.* 2014;39(3):270–81.
 15. Wells GA, Shea, O'Connell D, Peterson J, Welch V, Losos M, et al. The Newcastle-Ottawa scale (NOS) for assessing the quality of nonrandomized studies in meta-analysis. The Ottawa Health Research Institute. Ottawa, Ontario; 2011. 18-1 p.
 16. Higgins JPT, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. *BMJ.* 2003 Sep 6;327(7414):557–60.
 17. Macaskill P, Walter SD, Irwig L. A comparison of methods to detect publication bias in meta-analysis. *Stat Med.* 2001 Feb 28;20(4):641–54.

18. Kitahara CM, De González AB, Freedman ND, Huxley R, Mok Y, Jee SH, et al. Total cholesterol and cancer risk in a large prospective study in Korea. *J Clin Oncol*. 2011;29(12):1592–8.
19. Strohmaier S, Edlinger M, Manjer J, Stocks T, Bjørge T, Borena W, et al. Total Serum Cholesterol and Cancer Incidence in the Metabolic Syndrome and Cancer Project (Me-Can). *PLoS One*. 2013;8(1).
20. Stocks T, Rapp K, Bjørge T, Manjer J, Ulmer H, Selmer R, et al. Blood glucose and risk of incident and fatal cancer in the metabolic syndrome and cancer project (Me-Can): Analysis of six prospective cohorts. *PLoS Med*. 2009;6(12).
21. Ahn J, Lim U, Weinstein SJ, Schatzkin A, Hayes RB, Virtamo J, et al. Prediagnostic total and high-density lipoprotein cholesterol and risk of cancer. *Cancer Epidemiol Biomarkers Prev*. 2009;18(11):2814–21.
22. Chyou PH, Nomura AM, Stemmermann GN, Kato I. Prospective study of serum cholesterol and site-specific cancers. *J Clin Epidemiol*. 1992;45(3):287–92.
23. Wulaningsih W, Garmo H, Holmberg L, Hammar N, Jungner I, Walldius G, et al. Serum Lipids and the Risk of Gastrointestinal Malignancies in the Swedish AMORIS Study. *J Cancer Epidemiol*. 2012;2012:792034.
24. de Martel C, Haggerty TD, Corley DA, Vogelman JH, Orentreich N, Parsonnet J. Serum ghrelin levels and risk of subsequent adenocarcinoma of the esophagus. *Am J Gastroenterol*. 2007;102(6):1166–72.
25. Hardikar S, Onstad L, Song X, Wilson AM, Montine TJ, Kratz M, et al. Inflammation and oxidative stress markers and esophageal adenocarcinoma incidence in a Barrett's esophagus cohort. *Cancer Epidemiol Biomarkers Prev*. 2014;23(11):2393–403.
26. Siahpush SH, Vaughan TL, Lampe JN, Freeman R, Lewis S, Odze RD, et al. Longitudinal study of insulin-like growth factor, insulin-like growth factor binding protein-3, and their

- polymorphisms: risk of neoplastic progression in Barrett's esophagus. *Cancer Epidemiol Biomarkers Prev.* 2007;16(11):2387–95.
27. Cook MB, Dawsey SM, Diaw L, Blaser MJ, Perez-Perez GI, Abnet CC, et al. Serum pepsinogens and *Helicobacter pylori* in relation to the risk of esophageal squamous cell carcinoma in the alpha-tocopherol, beta-carotene cancer prevention study. *Cancer Epidemiol Biomarkers Prev.* 2010;19(8):1966–75.
 28. Ren J-S, Kamangar F, Qiao Y-L, Taylor PR, Liang H, Dawsey SM, et al. Serum pepsinogens and risk of gastric and oesophageal cancers in the General Population Nutrition Intervention Trial cohort. *Gut.* 2009;58(5):636–42.
 29. Xue L, Xing L, Wang J, Shen H, Cui J, Mi J, et al. Serum pepsinogens and *Helicobacter pylori* are not associated with esophageal squamous cell carcinoma in a high-risk area in China. *Tumori.* 2013;99(2):134–8.
 30. Murphy G, Kamangar F, Albanes D, Stanczyk FZ, Weinstein SJ, Taylor PR, et al. Serum ghrelin is inversely associated with risk of subsequent oesophageal squamous cell carcinoma. *Gut.* 2012;61(11):1533–7.
 31. Keeley BR, Islami F, Pourshams A, Poustchi H, Pak JS, Brennan P, et al. Prediagnostic serum levels of inflammatory biomarkers are correlated with future development of lung and esophageal cancer. *Cancer Sci.* 2014;105(9):1205–11.
 32. Abnet CC, Chen Y, Chow W-H, Gao Y-T, Helzlsouer KJ, Le Marchand L, et al. Circulating 25-hydroxyvitamin D and risk of esophageal and gastric cancer: Cohort Consortium Vitamin D Pooling Project of Rarer Cancers. *Am J Epidemiol.* 2010;172(1):94–106.
 33. Wulaningsih W, Michaelsson K, Garmo H, Hammar N, Jungner I, Walldius G, et al. Serum calcium and risk of gastrointestinal cancer in the Swedish AMORIS study. *BMC Public Health.* 2013;13:663.

34. Wulaningsih W, Michaelsson K, Garmo H, Hammar N, Jungner I, Walldius G, et al. Inorganic phosphate and the risk of cancer in the Swedish AMORIS study. *BMC Cancer*. 2013;13:257.
35. Kimm H, Kim S, Jee SH. The independent effects of cigarette smoking, alcohol consumption, and serum aspartate aminotransferase on the alanine aminotransferase ratio in Korean men for the risk for esophageal cancer. *Yonsei Med J*. 2010;51(3):310–7.
36. Sun C-L, Yuan J-M, Lee M-J, Yang CS, Gao Y-T, Ross RK, et al. Urinary tea polyphenols in relation to gastric and esophageal cancers: a prospective study of men in Shanghai, China. *Carcinogenesis*. 2002;23(9):1497–503.
37. Takata Y, Kristal AR, Santella RM, King IB, Duggan DJ, Lampe JW, et al. Selenium, selenoenzymes, oxidative stress and risk of neoplastic progression from Barrett's esophagus: results from biomarkers and genetic variants. *PLoS One*. 2012;7(6):e38612.
38. Taylor PR, Qiao Y-L, Abnet CC, Dawsey SM, Yang CS, Gunter EW, et al. Prospective study of serum vitamin E levels and esophageal and gastric cancers. *J Natl Cancer Inst*. 2003;95(18):1414–6.
39. Abnet CC, Qiao Y-L, Dawsey SM, Buckman DW, Yang CS, Blot WJ, et al. Prospective study of serum retinol, beta-carotene, beta-cryptoxanthin, and lutein/zeaxanthin and esophageal and gastric cancers in China. *Cancer Causes Control*. 2003;14(7):645–55.
40. Lam TK, Freedman ND, Fan J-H, Qiao Y-L, Dawsey SM, Taylor PR, et al. Prediagnostic plasma vitamin C and risk of gastric adenocarcinoma and esophageal squamous cell carcinoma in a Chinese population. *Am J Clin Nutr*. 2013;98(5):1289–97.
41. Mark SD, Qiao YL, Dawsey SM, Wu YP, Katki H, Gunter EW, et al. Prospective study of serum selenium levels and incident esophageal and gastric cancers. *J Natl Cancer Inst*. 2000;92(21):1753–63.
42. Fanidi A, Relton C, Ueland PM, Midttun Ø, Vollset SE, Travis RC, et al. A prospective study of

- one-carbon metabolism biomarkers and cancer of the head and neck and esophagus. *Int J Cancer*. 2015;136(4):915–27.
43. Murphy G, Fan J-H, Mark SD, Dawsey SM, Selhub J, Wang J, et al. Prospective study of serum cysteine levels and oesophageal and gastric cancers in China. *Gut*. 2011;60(5):618–23.
 44. Hecht SS, Murphy SE, Stepanov I, Nelson HH, Yuan J-M. Tobacco smoke biomarkers and cancer risk among male smokers in the Shanghai cohort study. *Cancer Lett*. 2013;334(1):34–8.
 45. Edgren G, Bagnardi V, Bellocco R, Hjalgrim H, Rostgaard K, Melbye M, et al. Pattern of declining hemoglobin concentration before cancer diagnosis. *Int J cancer*. 2010;127(6):1429–36.
 46. Tsuboya T, Kuriyama S, Nagai M, Hozawa A, Sugawara Y, Tomata Y, et al. Gamma-Glutamyltransferase and Cancer Incidence: The Ohsaki Cohort Study. *J Epidemiol*. 2012;22(2):144–50.
 47. Hansen J, Sallmen M, Selden AI, Anttila A, Pukkala E, Andersson K, et al. Risk of cancer among workers exposed to trichloroethylene: analysis of three Nordic cohort studies. *J Natl Cancer Inst*. 2013;105(12):869–77.
 48. Gwini S, Macfarlane E, Del Monaco A, McLean D, Pisaniello D, Benke GP, et al. Cancer Incidence, Mortality, and Blood Lead Levels Among Workers Exposed to Inorganic Lead. *Ann Epidemiol*. 2012;22(4):270–6.
 49. Risques RA, Vaughan TL, Li X, Odze RD, Blount PL, Ayub K, et al. Leukocyte telomere length predicts cancer risk in Barrett’s esophagus. *Cancer Epidemiol Biomarkers Prev*. 2007;16(12):2649–55.
 50. Chao DL, Maley CC, Wu X, Farrow DC, Galipeau PC, Sanchez CA, et al. Mutagen sensitivity and neoplastic progression in patients with Barrett’s esophagus: a prospective analysis. *Cancer*

- Epidemiol Biomarkers Prev. 2006;15(10):1935–40.
51. Abnet CC, Borkowf CB, Qiao YL, Albert PS, Wang E, Merrill AHJ, et al. Sphingolipids as biomarkers of fumonisin exposure and risk of esophageal squamous cell carcinoma in china. *Cancer Causes Control*. 2001;12(9):821–8.
 52. Jee SH, Ohrr H, Sull JW, Yun JE, Ji M, Samet JM. Fasting serum glucose level and cancer risk in Korean men and women. *JAMA*. 2005;293(2):194–202.
 53. Chen W, Dawsey SM, Qiao Y-L, Mark SD, Dong Z-W, Taylor PR, et al. Prospective study of serum 25(OH)-vitamin D concentration and risk of oesophageal and gastric cancers. *Br J Cancer*. 2007;97(1):123–8.
 54. Lindkvist B, Johansen D, Stocks T, Concin H, Bjorge T, Almquist M, et al. Metabolic risk factors for esophageal squamous cell carcinoma and adenocarcinoma: a prospective study of 580,000 subjects within the Me-Can project. *BMC Cancer*. 2014;14:103.
 55. Duggan C, Onstad L, Hardikar S, Blount PL, Reid BJ, Vaughan TL. Association between markers of obesity and progression from Barrett’s esophagus to esophageal adenocarcinoma. *Clin Gastroenterol Hepatol*. 2013;11(8):934–43.
 56. Knekt P, Aromaa A, Maatela J, Alfthan G, Aaran RK, Nikkari T, et al. Serum micronutrients and risk of cancers of low incidence in Finland. *Am J Epidemiol*. 1991;134(4):356–61.
 57. F. K, L. D, W.-Q. W, C.C. A, G.-Q. W, M.J. R, et al. Serum pepsinogens and risk of esophageal squamous dysplasia. Vol. 124, *International Journal of Cancer*. F. Kamangar, Division of Cancer Epidemiology and Genetics, NCI, 6120 Executive Blvd., Bethesda, MD 20892-7232, United States. E-mail: kamangaf@mail.nih.gov: Wiley-Liss Inc. (111 River Street, Hoboken NJ 07030-5774, United States); 2009. p. 456–60.
 58. Correa P. Serum pepsinogens in gastric cancer screening. *Dig Dis Sci*. 2010 Aug;55(8):2123–5.

59. Kim N, Jung HC. The role of serum pepsinogen in the detection of gastric cancer. *Gut Liver*. 2010 Sep;4(3):307–19.
60. Yu G, Gail MH, Shi J, Klepac-Ceraj V, Paster BJ, Dye BA, et al. Association between upper digestive tract microbiota and cancer-predisposing states in the esophagus and stomach. *Cancer Epidemiol Biomarkers Prev*. 2014;23(5):735–41.
61. Chen X, Winckler B, Lu M, Cheng H, Yuan Z, Yang Y, et al. Oral Microbiota and Risk for Esophageal Squamous Cell Carcinoma in a High-Risk Area of China. *PLoS One*. 2015;10(12).
62. Rosen R, Amirault J, Liu H, Mitchell P, Hu L, Khatwa U, et al. Changes in Gastric and Lung Microflora With Acid Suppression. *JAMA Pediatr*. 2014 Oct 1;168(10):932.
63. Nehra D, Howell P, Williams CP, Pye JK, Beynon J. Toxic bile acids in gastro-oesophageal reflux disease: influence of gastric acidity. *Gut*. 1999 May;44(5):598–602.
64. Xie F-J, Zhang Y-P, Zheng Q-Q, Jin H-C, Wang F-L, Chen M, et al. Helicobacter pylori infection and esophageal cancer risk: an updated meta-analysis. *World J Gastroenterol*. 2013 Sep 28;19(36):6098–107.
65. Gao Q-Y, Fang J-Y. Early esophageal cancer screening in China. *Best Pract Res Clin Gastroenterol*. 2015;29(6):885–93.
66. Huang W, Ren H, Ben Q, Cai Q, Zhu W, Li Z. Risk of esophageal cancer in diabetes mellitus: a meta-analysis of observational studies. *Cancer Causes Control*. 2012 Feb 18;23(2):263–72.
67. Cowey S, Hardy RW. The Metabolic Syndrome: A High-Risk State for Cancer? *Am J Pathol*. 2006;169(5):1505–22.
68. Huang XP, Zhou WH, Zhang YF. Genetic variations in the IGF-IGFR-IGFBP axis confer susceptibility to lung and esophageal cancer. *Genet Mol Res*. 2014;13(1):2107–19.
69. Altobelli E, Angeletti PM, Latella G. Role of Urinary Biomarkers in the Diagnosis of Adenoma

- and Colorectal Cancer: A Systematic Review and Meta-Analysis. *J Cancer*. 2016;7(14):1984–2004.
70. Zhang H, Xia J, Wang K, Zhang J. Serum autoantibodies in the early detection of esophageal cancer: a systematic review. *Tumour Biol*. 2015;36(1):95–109.
71. Shah AK, Saunders NA, Barbour AP, Hill MM. Early Diagnostic Biomarkers for Esophageal Adenocarcinoma-The Current State of Play. *Cancer Epidemiol Biomarkers Prev*. 2013;22(7):1185–209.
72. Anker P, Mulcahy H, Qi Chen X, Stroun M. Detection of Circulating Tumour DNA in the Blood (Plasma/Serum) of Cancer Patients. *Cancer Metastasis Rev*. 1999;18(1):65–73.
73. TIME. A Blood Test for Cancer Gets Closer. *TIME*. 2016;
74. Holmes RS, Vaughan TL. Epidemiology and pathogenesis of esophageal cancer. *Semin Radiat Oncol*. 2007 Jan;17(1):2–9.

Tables:

Table 1. Characteristics of studies included in review

| Author (year) | Cohort name | Country | Study | Type | Ag | Sex | BMI (average, | Follow- | Biomarkers measured | Bioma | Oesophageal cancer | | |
|-----------------|------------------------------|-------------|-------|------|----|-----|------------------|-----------|----------------------------|--------|--------------------|-----|-----|
| | | | | | | | | | | | Total | OAC | OSC |
| Abnet (2001) | General population trial | China | NCC | OSCC | 40 | All | n/s | 5.25 | Sphingolipids | Serum | | | Yes |
| Abnet (2003) | General population trial | China | NCC | OSCC | 40 | All | 22.1 (2.5) | 5.25 | Carotenoids | Serum | | | Yes |
| Abnet (2010) | Vitamin D pooling project | Multiple | NCC | All | 18 | All | 25.3 (n/s) | 1.7-11.8† | 25(OH)D | Serum | Yes | Yes | Yes |
| Ahn (2009) | ATBC | Finland | Cohor | All | 50 | Men | n/s | 14.9 | Cholesterol | Serum | Yes | | |
| Chao (2006) | Seattle Barretts esophagus | USA | Cohor | OAC | 18 | All | n/s | 6.4† | Mutagen sensitivity | Serum | | Yes | |
| Chen (2007) | General population trial | China | NCC | OSCC | 40 | All | n/s | 5.25 | 25(OH)D | Serum | | | Yes |
| Chyou (1992) | Honolulu heart program | USA | Cohor | All | 28 | All | n/s | 22 max | Cholesterol | Serum | Yes | | |
| Cook (2010) | ATBC | Finland | NCC | OSCC | 50 | Men | 26.4 (3.9) | 14.9 | Pepsinogens | Serum | | | Yes |
| De Martel | Kaiser Permanente | USA | NCC | OAC | 18 | All | n/s | 19.7 | Ghrelin | Serum | | Yes | |
| Duggan (2013) | Seattle Barretts esophagus | USA | Cohor | OAC | 18 | All | 29.4 (4.3) | 6.7 † | Leptin, Adiponectin & | Serum | | Yes | |
| Edgren (2010) | Scandinavian blood donors | Scandinavia | NCC | All | 18 | All | n/s | 5 max | Haemoglobin | Serum | Yes | | |
| Fanidi (2015) | EPIC | European | NCC | All | 25 | All | n/s | 6.4 † | One carbon metabolites | Plasm | | Yes | Yes |
| Gwini (2012) | Lead workers | Australia | Cohor | All | 18 | Men | n/s | 16.2 | Lead | Blood | Yes | | |
| Hansen (2013) | Trichloroethylene workers | Scandinavia | Cohor | All | 18 | All | n/s | 45 max | Trichloroethylene | Urinar | Yes | Yes | |
| Hardikar (2014) | Seattle Barretts esophagus | USA | Cohor | OAC | 18 | All | n/s | 6.14† | Cytokines | Serum | | Yes | |
| Hecht (2013) | Shanghai mens cohort | China | NCC | All | 45 | Men | n/s | 12 max | Smoking biomarkers | Urinar | Yes | | |
| Jee (2005) | National Health Insurance | South | Cohor | All | 30 | All | 23.2 (2.6) | 10 max | Glucose | Serum | Yes | | |
| Keeley (2014) | Golestan cohort study | Iran | NCC | OSCC | 40 | All | n/s | 9 max | Cytokines | Serum | | | Yes |
| Kimm (2010) | National Health Insurance | South | Cohor | All | 30 | Men | 23.2 (2.7) | 14 † | Alcohol biomarkers | Serum | Yes | | |
| Kitahara (2011) | National Health Insurance | South | Cohor | All | 30 | All | 23.2 (2.6) | 12.7 | Cholesterol | Serum | Yes | | |
| Knekt (1991) | Social Insurance Institution | Finland | NCC | All | 15 | All | n/s | 9 max | Micronutrients | Serum | Yes | | |
| Lam (2013) | General population trial | China | NCC | OSCC | 40 | All | n/s | 5.25 | Vitamin C | Plasm | | | Yes |
| Lindkvist | Me-Can | Multiple | Cohor | Both | 18 | All | 25.3 (4.0) | 12 | Cholesterol, glucose | Serum | | Yes | Yes |
| Mark (2000) | General population trial | China | NCC | OSCC | 40 | All | n/s | 5.25 | Selenium | Serum | | | Yes |
| Murphy (2011) | General population trial | China | NCC | OSCC | 40 | All | 21.8 (2.6) | 5.25 | Cysteine | Serum | | | Yes |
| Murphy (2012) | ATBC | Finland | NCC | OSCC | 50 | Men | 26.6 (4.0) | 14.9 | Ghrelin | Serum | | | Yes |
| Ren (2009) | General population trial | China | Cohor | OSCC | 40 | All | 22 (n/s) | 5.25 | Pepsinogens | Serum | | | Yes |
| Risques (2007) | Seattle Barretts esophagus | USA | Cohor | OAC | 18 | All | n/s | 5.8 | Leukocyte telomere length | Serum | | Yes | |
| Siahpush | Seattle Barretts esophagus | USA | Cohor | OAC | 18 | All | 28.7 (n/s) | 5.8 | Insulin-like Growth Factor | Serum | | Yes | |
| Sun (2002) | Shanghai Mens cohort | China | NCC | All | 45 | Men | n/s | 5.05 | Tea polyphenols | Urinar | Yes | | |
| Stocks (2009) | Me-Can | Multiple | Cohor | All | 18 | All | n/s | 12.2 | Glucose | Serum | Yes | | |
| Strohmaier | Me-Can | Multiple | Cohor | All | 18 | All | 25.3 (4.0) | 11.7 | Cholesterol | Serum | Yes | | |
| Takata (2012) | Seattle Barretts esophagus | USA | Cohor | OAC | 18 | All | n/s | 7.3 † | Selenium | Serum | | Yes | |
| Taylor (2003) | General population trial | China | NCC | OSCC | 40 | All | n/s | 5.25 | Tocopherols | Serum | | | Yes |
| Tsuboya (2012) | Ohsaki cohort | Japan | Cohor | All | 40 | All | 23.6 (3.0) | 10 max | Gamma- | Serum | Yes | | |
| Wulaningsih | AMORIS | Sweden | Cohor | All | 20 | All | n/s | 12 | Lipids & glucose | Serum | Yes | | |
| Wulaningsih | AMORIS | Sweden | Cohor | All | 20 | All | n/s | 12.75 | Phosphate (inorganic) | Serum | Yes | | |
| Wulaningsih | AMORIS | Sweden | Cohor | All | 20 | All | n/s | 12.75 | Calcium | Serum | Yes | | |
| Xue (2013) | Hebei province | China | Cohor | OSCC | 18 | All | n/s | 15 max | Pepsinogens | Serum | | | Yes |

Studies were within cohorts not selected on basis of presence of precancerous condition (such as Barrett's oesophagus) unless indicated by †.

Study follow-ups reported as means unless stated otherwise, or medians if indicated by ‡

Abbreviations: 25(OH)D: 25-hydroxyvitamin D; AMORIS: Swedish Apolipoprotein Mortality Risk study; ATBC: Alpha-tocopherol, Beta-caroten trial; EPIC: European Prospective investigation into Cancer and Nutrition; HOMA: Homeostatic model assessment; max: maximum; Me-Can: Metabolic Syndrome and Cancer cohort; NCC: Nested case-control; OAC: Oesophageal adenocarcinoma; OSCC: Oesophageal squamous cell carcinoma.

Table 2. Quality assessment using the Newcastle-Ottawa scale for cohort studies

| Author (year) | Selection | | | | Comparability | Outcome | | | Total | Grade ^c |
|-----------------|----------------|------------------------|------------------------|---------------------|----------------------|--------------------|-------------------|-------------------|-------|--------------------|
| | Representative | Selection of non-cases | Exposure ascertainment | Outcome not present | Adjustment/matching* | Outcome assessment | Follow-up length† | Loss to follow-up | | |
| Abnet (2001) | 0 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 6 | Fair |
| Abnet (2003) | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 7 | Good |
| Abnet (2010) | 0 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 8 | Good |
| Ahn (2009) | 0 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 8 | Good |
| Chao (2006) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 8 | Good |
| Chen (2007) | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 7 | Good |
| Chyou (1992) | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 8 | Good |
| Cook (2010) | 0 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 8 | Good |
| De Martel | 0 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Duggan (2013) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Edgren (2010) | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 8 | Good |
| Fanidi (2015) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Gwini (2012) | 0 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 3 | Poor |
| Hansen (2013) | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 1 | 5 | Poor |
| Hardikar | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Hecht (2013) | 0 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 8 | Good |
| Jee (2005) | 0 | 0 | 1 | 1 | 2 | 1 | 1 | 1 | 7 | Fair |
| Keeley (2014) | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 1 | 8 | Fair |
| Kimm (2010) | 0 | 0 | 1 | 1 | 2 | 1 | 1 | 1 | 7 | Fair |
| Kitahara (2011) | 0 | 0 | 1 | 1 | 2 | 1 | 1 | 1 | 7 | Fair |
| Knekt (1991) | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 8 | Good |
| Lam (2013) | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 7 | Good |
| Lindkvist | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Mark (2000) | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 7 | Good |
| Murphy (2011) | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 7 | Good |
| Murphy (2012) | 0 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 8 | Good |
| Ren (2009) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Risques (2007) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Siahpush | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Stocks (2009) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Strohmaier | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Sun (2002) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Takata (2012) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Taylor (2003) | 1 | 1 | 1 | 0 | 1 | 1 | 1 | 1 | 7 | Good |
| Tsuboya (2012) | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 | Good |
| Wulaningsih | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 7 | Good |
| Wulaningsih | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 7 | Good |
| Wulaningsih | 0 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 7 | Good |
| Xue (2013) | 1 | 1 | 1 | 1 | 1 | 0 | 1 | 1 | 6 | Fair |

^a Age and sex=1, BMI/WHR for OAC/Smoking for OSCC=2

^b 5 years+

^c **Good quality:** 3 or 4 stars in selection domain AND 1 or 2 stars in comparability domain AND 2 or 3 stars in outcome/exposure domain

Fair quality: 2 stars in selection domain AND 1 or 2 stars in comparability domain AND 2 or 3 stars in outcome/exposure domain

Poor quality: 0 or 1 star in selection domain OR 0 stars in comparability domain OR 0 or 1 stars in outcome/exposure domain

FIGURE LEGENDS:

Figure 1. Numbers at each stage of the study selection process.

Figure 2. Forest plots from random effects meta-analysis for the association between total serum cholesterol concentrations and risk of oesophageal cancer (total). Columns indicate the number of cases and non-cases with low (reference category) and high total serum cholesterol, relative risk (95% confidence intervals) and weighting. Abbreviations: ES: Relative risk; CI: Confidence intervals.

Figure 3. Forest plots from random effects meta-analysis of the association between serum glucose concentrations and risk of oesophageal cancer (total). Columns indicate the number of cases and non-cases with low (reference category) and high total serum glucose concentrations, relative risk (95% confidence intervals) and weighting. Abbreviations: ES: Relative risk; CI: Confidence intervals.

Figure 4. Forest plots from random effects meta-analysis of the association between serum PGI concentrations and risk of oesophageal squamous cell carcinoma. Columns indicate the number of cases and non-cases with high (reference category) and low serum PGI concentrations, relative risk (95% confidence intervals) and weighting. Abbreviations: ES: Relative risk; CI: Confidence intervals; PG: pepsinogen.

Figure 5. Forest plots from random effects meta-analysis of the association between serum PGI:PGII ratio and risk of oesophageal squamous cell carcinoma. Columns indicate the number of cases and non-cases with high (reference category) and low serum PGI:PGII ratio, relative risk (95% confidence intervals) and weighting. Abbreviations: ES: Relative risk; CI: Confidence intervals; PG: pepsinogen.