Haem iron intake and risk of lung cancer in the European Prospective Investigation into Cancer and Nutrition (EPIC) cohort

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Abstract

Background: Epidemiological studies suggest that haem iron, which is found predominantly in red meat and increases endogenous formation of carcinogenic *N*-nitroso compounds, may be positively associated with lung cancer. The objective was to examine the relationship between haem iron intake and lung cancer risk, using detailed smoking history data and serum cotinine to control for potential confounding.

Methods: In the European Prospective Investigation into Cancer and Nutrition (EPIC), 416 746 individuals from ten countries completed demographic and dietary questionnaires at recruitment. Cox proportional hazards models were used to estimate hazard ratios (HR) and 95% confidence intervals (CI) for incident lung cancer (n=3 731) risk relative to haem iron, non-haem iron, and total dietary iron intake. A corresponding analysis was conducted among a nested subset of 800 lung cancer cases and 1,489 matched controls for whom serum cotinine was available.

Results: Haem iron was associated with lung cancer risk, including after adjustment for details of smoking history (time since quitting, number of cigarettes per day): as a continuous variable (HR per 0.3 mg/1000 kcal 1.03, 95% CI 1.00 – 1.07), and in the highest versus lowest quintile (HR 1.16, 95%CI 1.02 – 1.32; trend across quintiles: $P = 0.035$). In contrast, non-haem iron intake was related inversely with lung cancer risk; however, this association attenuated after adjustment for smoking history. Additional adjustment for serum cotinine did not considerably alter the associations detected in the nested case-control subset.

Conclusions: Greater haem iron intake may be modestly associated with lung cancer risk.

Running title: Haem iron and lung cancer risk in EPIC

Keywords (5-10): Haem iron, non-haem iron, dietary iron, lung cancer, cohort, EPIC, cotinine

Introduction

Lung cancer is the most common cancer in the world, both in terms of incidence (an estimated 1.8 million cases in 2012) and mortality (1.6 million deaths in 2012), owing to the high case fatality. [1] Smoking is the major determinant of lung cancer, estimated to be responsible for 85% of all cases, [2] and accordingly is the primary target for public health interventions to reduce lung cancer incidence. However, diet is also a potentially modifiable risk factor for lung cancer [3]. Red meat is one such dietary component of interest: individuals with the highest red meat consumption were at 34% greater risk of lung cancer compared to the lowest consumers in a meta-analysis of 18 cohort studies. [4] One of the proposed mechanisms for the carcinogenicity of red meat is haem iron, a subtype of dietary iron that is found in animal products (primarily red meat). Other dietary sources of iron include non-haem iron, present mainly in cereals, legumes, and some vegetables. [5] Consumption of haem iron through diet appears to lead to the formation of endogenous *N*-nitroso compounds (NOCs), [6] which may increase the risk of some common cancers. [7] For lung cancer specifically, there is evidence from molecular biological studies that haem availability is significantly increased in cancer cells and tumours, resulting in elevated production of haemoproteins and support for cancer cell progression through intensified oxygen consumption and cellular energy production. [8]

To date, studies of haem iron in relation to lung cancer risk are limited to four cohort studies [9–12] and one case-control study. [13] A 2014 meta-analysis of three of the prospective studies [9,11,12] reported a pooled relative risk (RR) of 1.12 (95% confidence interval [CI] 0.98-1.29) per 1 mg/day difference in haem iron. [7] The pooled studies were large cohorts from the United States with similar dietary

assessment methods; however, the studies varied in haem calculation methods (use of a measured values database [11,12] vs. applying a single value for red and white meat products [9]) and in approaches to address smoking as a potential confounder. The magnitude of the association between smoking and lung cancer, along with established dietary variability by smoking status (e.g. current smokers tend to report lower fruit and vegetable intake and higher meat intake than non-smokers [14–16]), requires extensive efforts to control for potential confounding by smoking in diet-lung cancer analyses. Ideally, such associations can be examined separately among never smokers to reduce the likelihood of smoking as a source of confounding. However, to date only the National Institutes of Health-American Association for Retired Persons Diet and Health study (NIH-AARP) has been large enough to do such an analysis, reporting similar positive effect sizes among smokers and non-smokers. [11] More recently, a smaller (n=211 cases) European cohort study reported an inverse association between haem and lung cancer risk, but this association was dependent upon adjustment for red meat in the model and could not be examined separately by smoking status. [10] In light of these unclear associations, we sought to further examine the relationship between haem intake and lung cancer risk in a large European cohort, using detailed smoking history data and serum cotinine as a biomarker for tobacco exposure to control for confounding by smoking status.

Subjects and Methods

Study population

The European Prospective Investigation into Cancer and Nutrition (EPIC) is a multi-centre prospective cohort to study the relationship between lifestyle, nutrition and cancer. Over 520 000 participants were recruited from 23 centres in

10 European countries between 1992 and 2000: Denmark (Aarhus and Copenhagen), France, Germany (Heidelberg and Potsdam), Greece, Italy (Florence, Naples, Ragusa, Turin, and Varese), the Netherlands (Bilthoven and Utrecht), Norway, Spain (Asturias, Granada, Murcia, Navarra, and San Sebastian), Sweden (Malmö and Umeå), and the United Kingdom (Cambridge and Oxford). Participants were recruited from the general population of their respective countries, with the following exceptions: the French cohort were teacher health insurance programme members; the Italian and Spanish cohort included members of blood donor associations and the general population; the Utrecht and Florence cohorts contained participants from mammographic screening programs; the Oxford cohort included a large proportion of vegetarians, vegans, and low meat eaters; finally, only women participated in the cohorts of France, Norway, Utrecht and Naples. Additional details of the design and methods used in the EPIC study has been published elsewhere.[17] The study was approved by all relevant ethical review boards, and all participants provided consent for the retention of acquired data and follow-up for incidence of cancer and death.

In the present study, we excluded participants with prevalent cancer at baseline (except non-melanoma skin cancer, *n*=25 185), participants missing information on diet (n=6 205) or smoking (*n*=11 696), and participants within the extreme percentiles of the ratio of energy intake to estimated energy requirement (*n*=9 573) or body mass index (BMI) (≤18.11 kg/m^2, *n*=4 920; ≥ 38.54 kg/m^2, *n*=4 932). Additionally, we excluded participants whose recorded date of loss to follow up or death was on the same date as recruitment (*n*=25), completion of lifestyle questionnaires (*n*=402) or completion of the dietary questionnaires

(*n*=46 440). In total, there were 416 746 participants included in the present study.

Assessment of diet, lifestyle, and anthropometry

At baseline, participants reported dietary intake using country-specific validated questionnaires. In most centres, a self-administered food frequency questionnaire (FFQ) was used to assess intake over the past 12 months (88 to 266 food items). In Denmark, Norway, Naples (Italy), and Umeå (Sweden), semi-quantitative FFQs were administered. A combination of dietary methods (semi-quantitative FFQ and diet record) was adopted in Malmö (Sweden) and the United Kingdom. In order to standardise the dietary information received from all centres, 24-hour dietary recall data was taken in 5-12% of participants in each sub-cohort to correct for over- or under-estimations between centres.[18] Usual intake of total iron was assessed by multiplying the iron content per food source according to the EPIC Nutrient Database (ENDB) with the individual mean daily intake of related food sources. To obtain product-specific estimates of haem iron intake, published data on percentages of haem iron to total iron content in different animal products were applied to total iron (65% for cooked beef, 39% for pork and 26% for chicken or fish), and then summed to obtain individual's total haem-iron intake. [19,20] Further details on methodology have been published previously.[21] Non-haem iron was calculated by subtracting haem iron estimates from total dietary iron.

Non-dietary information was also collected on variables related to dietary status, likely or potential risk factors for cancer. A standardised set of questions was agreed between the original seven EPIC countries (France, Germany, Greece, Italy, the Netherlands, Spain and United Kingdom), which included those on education, health history, smoking history (smoking status: current, former, never,

number of cigarettes currently smoked, and duration of smoking), alcohol consumption patterns, physical activity, hormone replacement therapy use, contraception use and any exposure to previous carcinogens. [17] Questionnaires from centres that joined the study later (those in Denmark, Sweden, Norway, and Naples) were re-coded and standardised to original EPIC questions.

Anthropometric measurements varied by centre: height, weight, and waist and hip circumference were measured in all EPIC centres excluding France, Oxford and Norway. In France and Oxford, this information was obtained through either self-reporting or on-site measurement. The Cambridge index of physical activity was derived by combining occupational activity level with recreational activity, as assessed by the amount of time in hours per week during winter and summer spent cycling and in other physical exercises (e.g. jogging, swimming). [22]

Blood was taken from 385 747 of EPIC participants, most of which is stored and managed at the International Agency for Research on Cancer (IARC) central biological bank. Filled syringes were kept at 5°C to 10°C, protected from light, and transferred to a local laboratory for further processing. Blood fractions (serum, citrate plasma, red cells, and buffy coat) were aliquoted into 0.5-mL straws that were subsequently heat sealed and stored in liquid nitrogen tanks at the IARC, Lyon, France, at −196°C, except in Umeå, Sweden, where samples were stored in 1.8-mL plastic tubes in −80°C freezers. All biochemical analyses, including measurements of serum cotinine, were performed at Bevital A/S (http://www.bevital.no), Bergen, Norway.

Cotinine nested case-control subset

The association between haem iron intake and lung cancer was examined in an existing nested case-control dataset for which serum cotinine, a biomarker of

tobacco exposure, was available. [23] In brief, two control participants per lung cancer case were chosen at random from appropriate risk sets consisting of all cohort members alive and cancer free (except non-melanoma skin cancer) at the time of diagnosis of the index case. Matching criteria were country, sex, date of blood collection (±1 month, relaxed to ±5 months for sets without available controls), and date of birth (±1 year, relaxed to ±5 years for sets without available control participants). The nested case-control subset for the present analysis included 800 cases and 1 489 controls.

Endpoint definition

In seven study countries (Denmark, Italy, the Netherlands, Norway, Spain, Sweden and United Kingdom), information on incident cancer cases was obtained through population cancer registries. Health insurance records, cancer and pathology registries and active follow-up of participants and next of kin were used as available in the remaining three countries (France, Germany and Greece). The last date of follow-up varied by EPIC centre, and ranged from June 2008 to December 2013.

Outcomes for the purposes of this analysis were first primary, incident lung cancer cases using the International Classification of Diseases for Oncology (ICD-O-2) site code C34. Furthermore, we conducted analyses by histologic sub-types of lung cancer according to the following ICD-O morphology codes: squamous-cell cancer (codes 8070, 8071, 8072, 8073, 8075, 8083, 8094, and 8123), small-cell cancer (codes 8041, 8042, 8043, 8044, 8045, and 8246), large-cell cancer (codes 8012, 8020, and 8021), adenocarcinoma (codes 8140, 8200, 8211, 8230, 8250, 8251, 8253, 8260, 8310, 8470, 8480, 8481, 8490, and 8550), and ‘unclassified’ (codes 8000, 8001, 8003, 8010, 8011, 8022, 8030, 8031, 8032, 8046, 8240,

8560, 8710, 8800, 8801, 8990, 9120, 9133, and 9699). Among the 416 746 individuals with a mean of 13.9 follow-up years, 3 731 incident, first primary lung cancers were diagnosed and included in this analysis; of these, 1 335 were adenocarcinomas, 735 were squamous cell carcinomas, 595 were small cell cancers and 213 were large cell.

Statistics

Cox proportional hazards models were used to estimate hazard ratios (HR) and 95% CIs. P-values reported are two-sided and associations with *P* values <0.05 were considered statistically significant. Age was used as the underlying time metric for all Cox models. When constructing the models, time of entry into the study was participants' age at recruitment, and time of exit was the age at which the first lung cancer was recorded, the time of death, loss to follow-up, or censoring. Schoenfeld residuals were used to test the proportional hazards assumption for all variables in the model. Where variables violated this assumption – as was the case with smoking status – stratification was performed to adjust the model.

The dose-response relationship was examined by fitting Cox proportional hazards models with restricted cubic splines for haem iron, non-haem iron, and total iron as continuous variables, adjusted for the covariates in model 2 (described below). Knots were placed at the 5th, 25th, 75th and 95th percentiles of intake followed by corresponding likelihood ratio tests comparing the goodness-of-fit of the models with and without the spline terms. [24,25] The nutritional exposures of interest (haem iron, total iron, and non-haem iron) were entered into the models as continuous variables per 1000 kcal per day, re-scaled into units of

approximately one standard deviation; as sex-specific quintiles, and as a trend variable (quintile sex-specific midpoints assigned).

All Cox regression models were stratified by sex, centre, age at recruitment (one-year groupings) and smoking status (current, former, or never). Adjustment for potential confounders was conducted in three steps. First, model 1 was adjusted for total caloric intake, as per the multivariate nutrient density method for energy adjustment. Second, model 2 was adjusted additionally for socioeconomic and lifestyle confounders identified from cancer-related meta-analyses [26] and an earlier EPIC study: [27] BMI (< 18.5 kg/m^2, 18.5-24.9 kg/m^2, 25-29.9 kg/m^2, >30 kg/m^2); education (none/primary school, technical/professional, secondary, longer education, or missing), height (cm), physical activity (Cambridge index categories: inactive, moderately inactive, moderately active, active, or missing), total fat (g/1000kcal). Third, model 3 was further adjusted for time since quitting (years) and number of cigarettes per day. Due to a high proportion of missing data (n = 47 555 for number cigarettes per day, 4 787 for time since quitting), multiple imputation was used for this analysis (SAS PROC MI and MIANALYZE, number of iterations = 20). The predictor variables for the multiple imputation of time since quitting and number of cigarettes per day were the primary dietary variables of interest (total iron, haem iron and non-haem iron), all covariates listed for model 2 above, plus sex, age and total person years of follow-up (the latter was log-transformed).

The analyses above were repeated separately by smoking status and by tumour histologic subtype. In addition, sex-stratified results are presented in online supplementary information table for comparison with the results from other cohorts.

In analyses of the nested case-control subset with serum cotinine available, conditional logistic regression analyses (matched) were conducted to estimate odds ratios (OR) and 95% CIs for lung cancer risk by iron intake on a continuous scale. Adjustment for confounders was conducted in a multi-step process in parallel to the analysis of the full EPIC cohort, described above. As in the main analysis, multiple imputation was used in the adjustment for time since quitting smoking and number of cigarettes per day due to a high proportion of missing data.

Sensitivity analyses included i) restriction of the analysis to those with two or more years of follow-up to reduce the potential influence of undiagnosed prevalent cancer cases at baseline; ii) adjustment for alcohol, fruit, vegetables, vitamin C, calcium, and beta-carotene (related to non-haem iron absorption); iii) adjustment for central adiposity (waist circumference, waist to height ratio); and iv) running model 3 from Table 2 as a complete case analysis rather than imputing missing data. Lastly, tests for interaction between haem iron, non-haem iron, and total iron by dichotomized intake of fruit, vegetables, and vitamin C (based on median intake in the cohort) were conducted using the Wald test.

Analyses were performed using SAS version 9.3 (SAS Institute, Inc., Cary, North Carolina).

Results

Descriptive statistics and evaluation of linearity of associations

Descriptive statistics of the cohort according to quintile of haem intake are presented in Table 1. Those in the highest quintile of haem intake had relatively higher BMI values, were more likely to be current smokers, and to report lower levels of education and vitamin C intake than those in the lower quintiles of intake

(Table 1). The cubic spline analysis indicated there was no evidence of non-linearity for haem iron (*P* = 0.13), non-haem iron (*P* = 0.14) or total iron (*P* = 0.089) (Supplementary Figures 1- 3).

Cox regression analysis

The data from this study showed that higher intake of haem iron was positively associated with the risk of lung cancer. After adjusting for potential confounders, including details of smoking behaviour (model 3), the risk of lung cancer was 16% higher in the highest quintile of haem intake compared to the lowest, with a significant test for trend across quintiles (Table 2), and a modest but significant association for haem as a continuous variable (HR per 0.3 mg/1000 kcal 1.03, 95% CI 1.00 – 1.07). In contrast, there was a suggestive inverse association non-haem iron intake and lung cancer risk in EPIC. Prior to adjusting for time since quitting and number of cigarettes per day, the risk of lung cancer was significantly lower in all quintiles of non-haem relative to the lowest group, with a significant trend across quintiles and an inverse association when analysed as a continuous variable (HR per 1.2 g/1000 kcal 0.92, 95% CI 0.88 – 0.96) (Table 2). Adjustment for details of smoking history (model 3) attenuated the associations in each quintile of non-haem iron, the trend test and for non-haem as a continuous variable. For total iron, adjustment for details of smoking history also attenuated the formerly significant trend across quintiles and the analysis of continuous intake (HR per 1.3 g/1000 kcal 0.98, 95% CI 0.94 – 1.02). These results are presented separately for men and women in Supplementary Table 1; however, there was no evidence of effect modification by sex in relation to haem iron (*P* = 0.11), non-haem (*P* = 0.47) or total iron (*P* = 0.66).

Subgroup and sensitivity analyses

In the analysis by histological types, the effect sizes yielded for adenocarcinoma in relation to haem, non-haem, and total iron intakes were broadly similar to those seen for all lung cancers, although not statistically significant in the fully adjusted models (Table 3). In contrast, haem iron intake was positively associated with the risk of small cell lung cancer (HR per 0.3 mg/1000 kcal 1.13, 95% CI 1.04– 1.21), and modestly associated with total iron intake (HR per 1.3g/1000 kcal 1.11, 95% CI 1.00 – 1.22) after adjustment for details of smoking history (model 3, Table 3). Stratification by smoking status yielded results among current smokers that were broadly similar to those detected at group level (Table 4). For haem iron and non-haem iron, there was no evidence of an interaction across smoking groups and lung cancer risk. For total iron there was a borderline significant interaction detected (*P* 0.05, Table 4); among former smokers, an inverse association with lung cancer risk was detected (HR per 1 SD 0.90, 95% CI 0.83 – 0.97).

Further adjustment for serum cotinine did not substantially modify the observed effect sizes (Table 5). For non-haem iron, the corresponding adjustment for serum cotinine modestly attenuated the results observed relative to models without serum cotinine (HRs and 95% CI on a continuous scale: 0.94 (0.81 – 1.09) and 0.90 (0.78 – 1.03), respectively, Table 5). For total iron, the attenuation was similar to that observed for non-haem iron (Table 5).

Additional sensitivity analyses yielded results that were not materially different to those presented in Table 2: the exclusion of the first two years of follow-up, adjustment for alcohol, fruits, vegetables, vitamin C, calcium, and beta-carotene, or adjustment for central adiposity (waist circumference and waist to height ratio). Restricting the adjustment for details of smoking history (Table 2, model 3) to those with complete data on details of smoking history yielded similar results to

those obtained in the imputed models (Supplementary Table 2). There was no evidence of interactions between haem iron intake and fruit, vegetables, or vitamin C (P = 0.17, 0.29, and 0.60 respectively) and lung cancer risk; similarly, the corresponding tests for interaction were null for non-haem iron (P= 0.68, 0.72, and 0.43, respectively) and total iron (P= 0.40, 0.47, and 0.47 respectively).

Discussion

The present analysis comprises the largest analysis of dietary haem iron and lung cancer risk in a European cohort, with a modest positive association between haem iron intake and lung cancer risk detected. There was no evidence of an interaction between smoking status, haem iron intake and the risk of lung cancer, and adjustment for serum cotinine had a minimal impact on the observed haem iron-lung cancer association. The association between haem iron and lung cancer appeared to be restricted to the small-cell histologic subtype. In contrast, non-haem iron was inversely associated with lung cancer risk, though the attenuation after adjustment for details of smoking history and after adjustment for serum cotinine in the nested case-control subset suggest that this association may be due to confounding.

The suggested positive association between haem iron and lung cancer risk in EPIC is of a similar magnitude to that detected in the largest study on the association to date, the US NIH-AARP study, which included 6 361 incident cases of lung cancer.[10] In a comparably adjusted model, effect sizes were slightly stronger in NIH-AARP than in EPIC, and were statistically significant among both men and women. In addition to greater statistical power, the estimates of haem iron in NIH-AARP were calculated from a database of haem in specific food items rather than broader food groups as was the case in EPIC; this may have also

contributed to the stronger effects seen in the former study. Our observation of no material differences for the association between haem iron and lung cancer risk by smoking status were consistent with the NIH-AARP conclusion of no difference among current, former, or never smokers and after sensitivity analyses controlling for smoking status, smoking intensity, and time since quitting. Analysis of another US cohort, the Prostate, Lung, Colorectal, and Ovarian (PLCO) Cancer Screening Trial, used the same haem-content database as NIH-AARP but reported no association between haem and lung cancer; however, that was a notably smaller cohort (n=782 lung cancer cases).[12] Other smaller studies found no association between haem intake and lung cancer risk overall [9,10] but one detected a positive association among users of vitamin C supplements.

There has been very limited study of dietary iron and non-haem iron in relation to lung cancer in cohort studies. In the Rotterdam study, no association between non-haem iron and risk of lung cancer was reported, but as noted previously that analysis included only a small number of lung cancer cases.[10] In the NIH-AARP study, total iron intake was inversely associated with lung cancer risk, with significantly lower risks in the highest versus lowest quintile (HR 0.87, 95% CI 0.79-0.97) and an inverse trend across quintiles. [28] In the present analysis, total iron was inversely associated with lung cancer risk among former smokers only. The covariates included in the analysis of the NIH-AARP cohort were similar to those used in the present analysis, including details of smoking history. [28]

Non-haem comprises the majority of dietary iron, therefore the relative consistency of results for total iron and non-haem iron in the present result are unsurprising. An apparent protective effect of iron in relation to lung cancer is somewhat unexpected in the context of the oxidative potential of iron, including the Fenton reaction, a

process that causes the conversion of hydrogen peroxide and superoxide to free radicals. [29] The imbalance in redox reactions brought about by iron excess may lead to premature cell aging and death.[30] We are unaware of any proposed biological pathways for a protective effect of iron or non-haem iron on cancer risk; however, haem iron is absorbed two to three times more readily than non-haem iron, and also increases absorption of the latter when eaten together thus haem iron poses a much greater risk of overload than non-haem iron. [30] More importantly, differences in dietary sources of haem and non-haem iron may have contributed to the divergent associations detected in the present analysis: sources of haem iron include red meats, poultry, and fish, whereas non-haem iron is found in many plant products and in dairy³⁰⁽³⁰⁾, as well as iron-fortified foods such as cereals and grains. [30] Therefore, the inverse association detected for non-haem iron may reflect other anti-carcinogenic properties of food sources high in non-haem iron (e.g. antioxidants in fruits and vegetables, a food group associated with lower risk of lung cancer) [31] rather than a specific biological pathway for non-haem iron. Sensitivity analyses included the addition of fruit and vegetable intake to the models, which did not affect our findings but the possibility of uncontrolled confounding cannot be ruled out.

In analyses by histologic subtype, the association between haem iron intake and lung cancer risk was only significant for small-cell carcinomas, and the effect size was larger than that estimated for the other types of lung cancer under study.

The underlying causes of these differences are unclear. Small-cell carcinoma is a comparatively fast-growing form of cancer that is highly metastatic, and is rare in non-smokers. It is possible that the associations detected between haem iron, total iron, and small-cell carcinoma in the present study reflects uncontrolled

confounding due to smoking; however, such confounding would also have been expected to yield associations for squamous cell carcinoma, as both histological types are strongly related to tobacco exposure. [32]

Strengths of the present study include a large sample size, long follow-up time, and detailed information collected on diet and a wide range of potentially confounding covariates, including tobacco exposure. We endeavoured to control for confounding by smoking through adjustment for details of smoking history, examining associations separately by smoking status, and adjusting for serum cotinine values in a nested subset of participants. However, it is impossible to fully exclude the possibility of confounding by smoking or other factors (such as carcinogenic advanced glycation end products, yielded when meat is cooked at high temperatures [33]), particularly in the context of the modest effect size detected. Never smokers comprised only 9% of lung cancer cases in the present analysis; therefore, there was limited power to examine this subgroup. Adjustment for serum cotinine measurements would have provided some control for second-hand smoke exposure at baseline,[34] although information on longer-term exposure would have been valuable. Similarly, detailed information on vitamin and mineral supplement use may have been informative, both for examining supplementary iron intake and further exploring the interaction between haem and supplementary vitamin C previously reported. [9] In EPIC, standardised questions on supplement use were only included in a calibration sub-study of participants (n= 36 994); [35] otherwise, study centres varied in the nature of supplement data collected and harmonized variables are not available. It is possible that the use of a more detailed database of haem content, rather than applying a constant value per meat type, could have yielded different results. Lastly, in 2015 the World

Health Organization issued an update to their guidelines for the classification of lung tumours, which included notable changes to the classification of large cell carcinomas;[36] the present analysis by histological subtype in EPIC would not have reflected the current guidelines and therefore may include some misclassification, particularly for large cell carcinomas.

Implications and future research

The results from EPIC are suggestive of a moderately positive association between haem intake and lung cancer; this observation is consistent with evidence from the largest study to date, conducted among US adults. Further study of populations within Europe and internationally will help determine the consistency of this association. The possible protective effect of non-haem iron in the current study has not been reported previously and warrants further study to determine the strength and reliability of this association and, if found to be robust, to understand the underlying mechanisms. Continued research on dietary risk factors for lung cancer may yield insight that informs preventive measures complementary to anti-tobacco strategies.

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Data sharing: For information on how to submit an application for gaining access to EPIC data and/or biospecimens, please follow the instructions at http://epic.iarc.fr/access/index.php

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