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ORIGINAL ARTICLE

Fish consumption and risk of esophageal cancer and its subtypes: a systematic review and meta-analysis of observational studies

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BACKGROUND/OBJECTIVES: Inconsistent results regarding the association between fish intake and risk of subtypes of esophageal cancer (EC), esophageal squamous cell carcinoma (ESCC) and esophageal adenocarcinoma (EAC), have been reported. To provide a quantitative assessment of this association, we summarized the evidence from observational studies.

SUBJECTS/METHODS: Relevant studies were identified in MEDLINE and EMBASE until 31 May, 2012. Summary relative risks (SRRs) with 95% confidence intervals (CIs) were pooled with a random-effects model. Between-study heterogeneity was assessed using the Cochran's Q and I^2 statistics.

RESULTS: A total of 24 studies (21 case–control and 3 cohort studies) were included in this systematic review and meta-analysis. The SRRs of ESCC were 0.81 (95% CI: 0.66–0.99) for those in the highest fish consumption category compared with those in the lowest consumption category, with significant heterogeneity among studies ($P_{heterogeneity} = 0.007$, $I^2 = 51.9\%$). Subgroup analysis suggested that a weak association between fish consumption and ESCC risk was shown in hospital-based case–control studies, but not in population-based case–control or cohort studies. According to high vs low analysis, fish consumption had no relationship with EAC risk (SRR = 0.86, 95% CI: 0.61–1.22).

CONCLUSIONS: Our results suggest that fish consumption is not appreciably related to risk of both ESCC and EAC.

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Keywords: esophageal squamous cell carcinoma; esophageal adenocarcinoma; meta-analysis; fish

INTRODUCTION

Esophageal cancer (EC) was the eighth most frequently diagnosed cancer worldwide, with an estimated 482 000 new cases in 2008.[1] Among the two major histological forms of EC: esophageal squamous cell carcinoma (ESCC) and esophageal adenocarcinoma (EAC), the incidence rate of EAC have seen a dramatic increase in the United States and Western Europe, along with a decrease in the incidence rate of ESCC.[2,3] However, ESCC is still the dominant histological type in high-incidence regions such as China and Iran, where it accounts for about 90% of the total EC cases.[4,5] The most known risk factors for ESCC in western countries are tobacco smoking and alcohol consumption. For EAC, symptoms of gastro-esophageal reflux (GOR), as well as white race, male gender, obesity and tobacco use, are consistently identified as established risk factors.[6] However, these above risk factors could not fully explain this high incidence, and many individuals who have not been exposed to these risk factors develop EC.[7]

Consumption of fish is suggested to be associated with cancer risks of several sites. Although consumption of fish may reduce colorectal cancer risk through inhibition of the arachidonic acid cascade, and thus carcinogenesis,[8] it was reported not to be associated with pancreatic cancer risk,[9] or even, increased prostate cancer risk.[10] As for the association between fish consumption and risk of EC, divergent results from epidemiological studies have been reported in a few studies.[11–19] In the 1990s, Fernandez *et al.*[11] found that fish consumption could reduce the risk of EC (odds ratios = 0.6 for the highest compared with the lowest level of fish consumption). However, data from a large prospective cohort of United States (NIH-AARP Diet and Health Study) showed that similar incidence of ESCC and a decreased incidence of EAC were observed in those who ate fish at the highest category compared with those at the lowest category.[17]

To our knowledge, there has been no comprehensive quantitative assessment of the association between consumption of fish and EC risk. In response, we preformed the first meta-analysis to assess this association following the meta-analysis of observational studies in epidemiology guidelines.[20]

MATERIALS AND METHODS

Data sources and searches

Two independent investigators (HW and HYJ) comprehensively searched through MEDLINE and EMBASE for all medical literature published up to the end of May, 2012. We searched the relevant studies with the following text word and/or medical subject heading terms: (1) (o)esophag*; (2) cancer OR carcinoma OR neoplasia OR adenocarcinoma; (3) fish OR shellfish OR seafood. Furthermore, we reviewed the reference lists of the relevant articles to identify additional studies. Only articles in English were included.

Study selection

Two authors (HW and HYJ) independently assessed titles and abstracts of potentially eligible studies that investigated fish intake and risk of EC. Our

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inclusion criteria were: (1) published as an observational study; (2) using a case–control, nested case–control or cohort design; and (3) reported point estimates (that is, rate ratios, ORs) and measures of variability (that is, 95% confidence intervals (CIs)) for fish and the risk of EC (ESCC and EAC) at least adjusted for age. Non-peer-reviewed articles, ecologic assessments, cross-sectional studies, correlation studies, experimental animal studies and mechanistic studies were excluded. If data were duplicate in more than one study, the most recent or complete studies were included in this analysis. Studies were excluded if they reported on several cancer sites combined, for example, upper aerodigestive tract cancers or cancers of oral cavity, pharynx and esophagus combined.

Data extraction

We summarized RRs for ESCC and EAC when the results were presented according to histological subtypes. When the results were reported for all ECs, we assumed that the majority of cases from non-western countries were ESCC.[21] In western countries, we assumed that the majority of cases in the earlier studies that did not report histological subtype had ESCC, as the rise in the incidence of adenocarcinoma in those countries mainly occurred in the most recent decades.[21] Where data were available, two researchers (HW and LJ) independently extracted the following information: the first author's last name, year of publication, geographic locations, design, the source and number of controls for case–control studies, details of exposure, the number of cases, covariables adjusted for and the relative risk estimates with corresponding 95% CIs for the highest vs lowest level. From each study, we extracted the risk estimates that reflected the greatest degree of control for potential confounders. If studies presented data on fresh and other types of fish, respectively, we only extracted the relative risk estimates for fresh fish.

Statistical methods

The meta-analyses were performed by using a random-effects model developed by DerSimonian and Laird,[22] which takes into account both study sample size and between-study variation when weighing study effects. Owing to the discrepancy in the etiology and clinicopathological profiles between EAC and ESCC, we performed meta-analysis of EAC and ESCC risk with fish consumption, respectively. For studies that provided separate RRs for males and females, we pooled the RRs, weighted by the inverse of the variance, within each study.

When possible, we also conducted the linear dose–response analysis of risk estimates for a continuous increase in fish consumption. We calculated the relative risk per unit of one time a week or 20 g/day of fish intake using generalized least-squares trend estimation analysis or variance-weighted least squares regression analysis based on the methods developed by Greenland[23] and Orsini.[24] The two methods, generalized least-squares trend estimation and variance-weighted least squares, require that medians for at least three quantitative exposure categories are known. If medians were not reported, we estimated the midpoint of the upper and lower boundaries in each category as the average intake level. If the highest category was open-ended, the open-ended boundary was calculated using as interval length of the width of the closest interval.

In assessing heterogeneity among studies, we used the Cochran Q and I^2 statistics. For Q statistics, a P-value of <0.1 was considered statistically significant. I^2 value of $>50\%$ was considered to have significant heterogeneity, and I^2 value of $<25\%$ was considered to have no significant heterogeneity.[25] Sources of heterogeneity were explored in subanalyses and by linear meta-regression, according to study design (case–control vs cohort study), geographic locations (USA, Europe, South America and Asia), the number of cases (<200 vs $\geqslant 200$), type of food frequency questionnaires (FFQ) and confounders, including smoking, body mass index (BMI), alcohol use, symptoms of GOR and dietary energy intake. We carried out sensitivity analysis by omitting one study at a time and calculating the summary relative risk for the remainder of studies and by pooling only estimates exclusively for ESCC (not combined with other histological subtypes).

Publication bias was assessed by using funnel plots and the further Begg's-adjusted rank correlation test and Egger's regression test.[26,27] All statistical analyses were performed using STATA, version 11.0 (STATA, College Station, TX, USA). A two-tailed P-value <0.05 was considered to be significant.

RESULTS

Search results and study characteristics

The search strategy generated 408 citations, of which 33 were considered of potential value and the full text was retrieved for detailed evaluation. Additional eight articles were included from the reference review. Seventeen of these 41 articles were subsequently excluded from the meta-analysis for various reasons (Figure 1). The characteristics of the remaining 24 articles (3 prospective cohort, 7 population-based and 14 hospital-based case–control studies), including 6677 subjects with EC, and the items evaluated are summarized in Table 1. The continents or countries where the studies were conducted were: the United States ($n=5$), South America ($n=3$), Europe ($n=8$) and Asia ($n=8$). All studies utilized FFQ to ascertain dietary information pertaining to fish consumption.

Esophageal squamous cell carcinoma

Three studies conducted in the United States and Italy between 1982 and 1996, which reported for all ECs, were excluded in the meta-analysis of ESCC, because we presumed a considerable part were EAC cases among these EC cases.[11,28,29] The three studies were all based on a hospital-based case–control design, and included a total of 559 cases of EC. These three studies reported a non-significantly increased, decreased or significantly decreased risk of EC with fish and shellfish intake for the highest vs lowest analysis, respectively.[11,28,29]

We combined 17 studies that presented results on the highest vs lowest level of fish consumption and ESCC risk. As shown in Figure 2, the summary relative risk (SRRs) of ESCC for the highest group compared with the lowest group of fish intake were significant (SRRs = 0.81, 95% CIs: 0.66–0.99) in a random-effects model, with significant heterogeneity among these studies ($P_{\text{heterogeneity}}=0.007$, $I^2=51.9\%$).

Furthermore, we observed a lower relative risk of ESCC in hospital-based case–control studies (SRRs = 0.66, 95% CIs: 0.55–0.80; 11 studies), but not in population-based case–control studies (SRRs = 1.58, 95% CIs: 1.09–2.29; 3 studies) or cohort studies (SRRs = 0.87, 95% CIs: 0.60–1.27; 3 studies).

The SRRs were not significant for studies conducted in USA (SRRs = 1.30, 95% CIs: 0.71–2.39; $P_{\text{heterogeneity}}=0.115$, $I^2=59.6\%$), South America (SRRs = 0.74, 95% CIs: 0.43–1.28; $P_{\text{heterogeneity}}=0.180$, $I^2=41.6\%$) and Asia (SRRs = 0.86, 95% CIs: 0.65–1.13; $P_{\text{heterogeneity}}=0.09$, $I^2=43.3\%$). However, the SRRs were significant for studies conducted in Europe (SRRs = 0.60, 95% CIs: 0.42–0.86; $P_{\text{heterogeneity}}=0.166$, $I^2=40.9\%$). In subgroup analyses by number of cases (<200 vs $\geqslant 200$), type of FFQ (validated vs not validated), and adjustments for confounders, such as BMI, smoking, alcohol use, symptoms of GOR and dietary energy

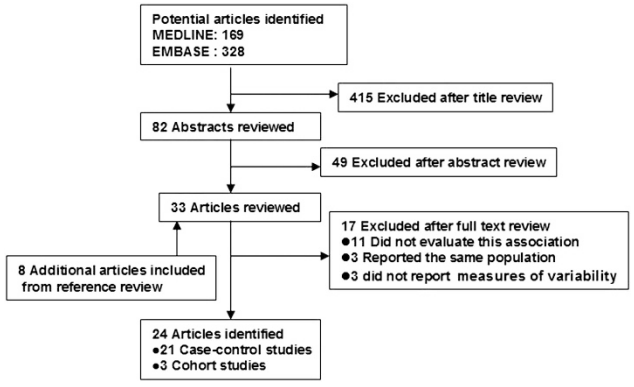

Figure 1. Flow diagram of systematic literature search on fish intake and the risk of esophageal cancer.

Table 1. Characteristics of studies of fish consumption and subtypes of esophageal cancer risk

Author/year/country	*Study design*	*Case and control or participants, n*	*Type of questionnaire*	*Exposure details*	*RR (95% CI) (highest vs lowest)*	*Adjustments*
Brown/1988/USA[28]	H-B	207 EC, 422 controls	FFQ 65 item NA	Fish and shellfish Q3 vs Q1	1.2 (0.6–2.1)	Age, sex, race, smoking, alcohol use
Tavani/1994/Italy[29]	H-B	46 EC, 230 controls	FFQ NA	Fish No or occasionally, 1, >1	0.5 (0.2–1.2)	Age, sex, education, smoking, alcohol use
Rolon/1995/Paraguay[53]	H-B	131 EC, 381 controls	FFQ 50 items Validated	Fish Q4 vs Q1	1.5 (0.6–4.0)	Age, sex, race, education, cigarettes, alcohol, fats and milk
Kinjo/1998/Japan[54]	Cohort	440 EC, 220 272 total *N*	Questionnaire NA	Fish ⩽1–3 times/month, ⩽1–3 times/week, ⩾4 times/week	1.11 (0.71–1.75)	Age, prefecture, occupation, sex
Launoy/1998/France[31]	H-B	208 ESCC, 399 controls	FFQ Validated	Fresh fish 0–10, 10–20, 20–30, >30 g/day	0.35 (0.18–0.67)	Age, interviewer, smoking, aniseed aperitifs, hot calvados, whisky, total alcohol, total energy intake and food groups
Fernandez/1999/Italy[11]	H-B	316 EC, 7990 controls	FFQ 37 items NA	Fish <1, 1, ⩾2 servings/week	0.6 (0.4–0.9)	Age, sex, residence, education, smoking, alcohol consumption and BMI
De Stafani/1999/Uruguay[55]	H-B	82 ESCC, 248 controls	FFQ 64 items NA	Fish Q3 vs Q1	0.70 (0.34–1.43)	Age, sex, residence, urban/rural status, hospital, family history of esophageal cancer, BMI, smoking, alcohol intake, mate consumption, vegetables, fruits intake
Levi/2000/Switzerland[12]	H-B	101 EC (92 ESCC and 9 EAC), 327 controls	FFQ 79 items Validated	Fish <1.2, 1.2–1.7, ⩾1.7 times/week	0.90 (0.55–1.80)	Age, sex, smoking, education, alcohol, energy intake
Boseti/2000/Italy[32]	H-B	304 ESCC, 743 controls	FFQ 78 items Validated	Fish <0.9, 0.9–1.4, 1.4–1.9, >1.9 times/week	0.57 (0.33–0.97)	Age, sex, area of residence, education, tobacco smoking, alcohol drinking, non-alcohol energy
Takezaki/2001/China[56]	P-B	199 EC, 235 controls	FFQ 152 items Validated	Fish, salted fish Q4 vs Q1	1.42 (0.67–3.04)	Age, sex, smoking, alcohol
Chen/2002/USA[33]	P-B	124 EAC, 449 controls	FFQ 54 items Validated	Fish Q4 vs Q1	0.14 (0.04–0.48)	Age, sex, energy intake, respondent type, BMI, alcohol use, tobacco use, education, family history, vitamin supplement use
Phukan/2001/India[57]	H-B	502 EC, 1004 controls	Questionnaire NA	Fish Never, occasionally, 1–4/week, daily	0.82 (0.03–3.9)	Age, sex
De Stafani/2003/Uruguay[13]	H-B	166 ESCC, 664 controls	FFQ 64 items Validated	Fish 0, 1–12,13–52, ⩾52 servings/year	0.53 (0.30–0.92)	Age, sex, residence, urban/rural status, education, BMI, tobacco, smoking, alcohol drinking, mate drinking and total energy intake
Wu/2006/China[58]	P-B	531 EC, 531 control	Questionnaire Validated	Fish and seafood Q4 vs Q1 Q4 vs Q1	1.91 (1.00–3.64) 1.04 (0.46–2.33)	Age, sex, education, economic status, smoking, alcohol drinking, BMI group, cancer family history, eating speed and food temperature
Wu/2007/USA[59]	P-B	206 EAC, 1308 controls	FFQ 124 items Validated	Fish and seafood Q4 vs Q1	0.85 (0.5–1.4)	Age, sex, race, birthplace, education, smoking, alcohol, BMI, reflux, use of vitamins and total calories
Fan/2008/China[60]	Cohort	101 EC, 18 244 total *N*	FFQ NA	Fish and seafood Q3 vs Q1	0.59 (0.36, 0.97)	Age, year of interview, neighborhood of residence, education, BMI, smoking, drinks
Sapkota/2008/Europe[15]	H-B	187 ESCC, 1288 controls	FFQ-23 items Validated	Fish Q3 vs Q1	0.70 (0.42–1.15)	Age, country, sex, tobacco, education, BMI, alcohol consumption, total vegetable and total fruit consumption
Ganesh/2009/India[16]	H-B	442 EC, 1628 controls	FFQ NA	Fresh-fish Yes/no	0.8 (0.5–0.9)	Age, place of residence, religion and occupation, smoking, alcohol and diet
Hajizadeh/2011/Iran[18]	H-B	47 ESCC, 96 controls	FFQ 168 items Validated	Fish 0–1.00, 1.01–3.00, 3.01–17.00 times/month	0.67 (0.25–1.83)	Age, sex, symptomatic GOR, BMI

Table 1. (Continued)

Author/year/country	*Study design*	*Case and control or participants, n*	*Type of questionnaire*	*Exposure details*	*RR (95% CI) (highest vs lowest)*	*Adjustments*
Mulholland/2011/UK[61]	P-B	218 EAC, 252 controls	FFQ-101 Validated	Oily fish 0, 0.5, ⩾1	1.20 (0.63–2.28)	Age, sex, energy intake, smoking, BMI, education, occupation, alcohol, NSAID location, *Helicobacter pylori* infection, saturated fat intake, glycaemic index intake, GOR symptoms
Silvera/2011/USA[30]	P-B	282 EA, 206 ESCC, 687 controls	FFQ 104 Validated	Fish Q4 vs Q1 Q4 vs Q1	EAC: 0.94 (0.59–1.480) ESCC: 1.83 (0.99–3.37)	Age, sex, site, race, income, education, proxy status, energy intake
Gao/2011/China[62]	H-B	600 ESCC, 1514 controls	Questionnaire NA	Fish and seafood Never, seldom, monthly, daily/weekly	0.50 (0.28–0.88)	Age, gender, geographic region
O'Doherty/2011/UK[19]	P-B	227 EAC, 260 controls	FFQ 101 items Validated	Total fish 9.3, 21.7, 31.2, 57.0 g/day	1.49 (0.72–3.10)	Age, sex, smoking, BMI, job type, education, energy intake, fruit, vegetable alcohol intake, *H. pylori* infection, NSAID, symptoms of GOR, location
Daniel/2011/USA[17]	Cohort	185 ESCC, 553 EAC, 492 186 total *N*	FFQ 124 items Validated	Total fish 3.6, 7.0, 9.9, 13.4, 21.4 g/1000 kcal/day	ESCC: 0.69 (0.42–1.13) EAC: 0.95 (0.72–1.26)	Age, sex, family history of cancer, race, BMI, smoking status, physical activity, alcohol, fruit, vegetables, energy intake

Abbreviations: BMI, body mass index; CI, confidence interval; EAC, esophageal adenocarcinoma; EC, esophageal cancer; ESCC, esophageal squamous cell carcinoma; FFQ, food frequency questionnaire; GOR, gastro-esophageal reflux; H-B, hospital-based case–control study; NA, data not available; NSAIDs, nonsteroid anti-inflammatory drugs; P-B, population-based case–control study; RR, relative risk.

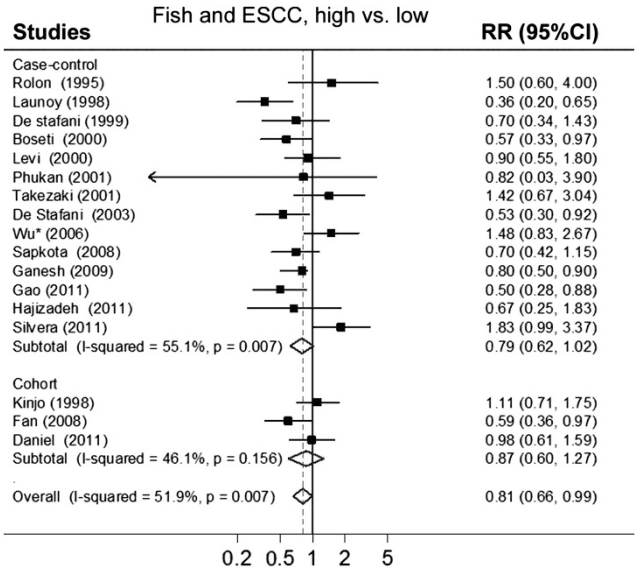

Figure 2. High vs low meta-analyses of fish intake and the risk of esophageal squamous cell carcinoma. Studies are subgrouped according to design.

intake, most of the SRRs for ESCC with fish intake were not significant (Table 2).

In univariate meta-regression analysis, all the above variables were not found to be significant factors for the association between fish intake and ESCC risk. When the overall homogeneity and effect size were calculated by removing one study at a time, we found that pooled RRs from the sensitivity analysis ranged from 0.82 (95% CIs: 0.67–0.98) after excluding study from Silvera *et al.*[30] to 0.92 (95% CIs: 0.73–1.11) after excluding study from Launoy *et al.*[31] When pooling only estimates exclusively for ESCC, we found fish consumption might reduce ESCC risk, with SRR (95% CI) of 0.63 (0.49–0.81) (data not shown).

The shape of the funnel plots for studies on the association between fish consumption and ESCC risk seemed symmetrical, and the *P*-value of Begg's test and Egger's test were 0.318 and 0.773, respectively, for high vs low analysis (Figure 3).

Six studies could be used in the meta-analysis of per unit of one time per week of fish consumption and risk of ESCC.[12–15,18,32] The SRRs were 0.81 (95% CIs: 0.73–0.89) for each extra time per week that fish was consumed. There was no significant heterogeneity among these studies ($P_{heterogeneity} = 0.589$, $I^2 = 0$). Three studies could be used in the meta-analysis of cancer risk per 20 g of fish consumption a day,[17,19,31] and the SRRs were 0.86 (95% CIs: 0.58–1.27; see Figure 4).

Esophageal adenocarcinoma

A total of six studies, including 1610 cases of EAC, presented results on EAC risk with fish consumption. The SRRs for the highest group compared with the lowest group of fish intake were 0.86 (95% CIs: 0.61–1.22), with severe heterogeneity among studies ($P_{heterogeneity} = 0.035$, $I^2 = 58.4\%$; see Figure 5). When excluding study from Chen *et al.*,[33] combining the remaining studies showed a non-significantly decreased risk of EAC with no significant heterogeneity (SRRs 0.89, 95% CI: 0.73–1.09, $P_{heterogeneity} = 0.450$, $I^2 = 0$). As shown in Table 2, studies on EAC with fish intake were exclusively conducted in the USA and Europe, and used a validated FFQ. Further analyses showed that there was no association between fish consumption and EAC risk in the five population-based case–control studies combined (SRRs = 0.86, 95% CI: 0.53–1.41). Only one cohort study presented results on fish intake and EAC risk, and a borderline

Table 2. Stratified meta-analyses of fish consumption and risk of esophageal squamous cell carcinoma and esophageal adenocarcinoma

Subgroup	*ESCC*				*EAC*			
	Studies, reference no.	*Cases, n*	*SRR (95% CI)*	*P; I^2(%)*	*Studies, reference no.*	*Case, n*	*SRR (95% CI)*	*P; I^2(%)*
Design								
Cohort	17, 54, 60	726	0.87 (0.60–1.27)	0.156; 46.1	17	553	0.78 (0.59–1.03)	—
Case–control	12, 13, 15, 16, 18, 30–32, 53, 55–58, 62	3782	0.79 (0.62–1.02)	0.007; 55.1	19, 30, 33, 59, 61	1057	0.86 (0.53–1.41)	0.024; 64.4
Population based	30, 56, 58	1012	1.58 (1.09–2.29)	0.84; 0	19, 30, 33, 59, 61	1057	0.86 (0.53–1.41)	0.024; 64.4
Hospital based	12, 13, 15, 16, 31, 32, 53, 55, 57, 62	2770	0.66 (0.55–0.80)	0.321; 12.9	—		—	—
Geographic locations								
Europe	12, 15, 31, 32	800	0.60 (0.42–0.86)	0.166; 40.9	19, 61	445	1.32 (0.81–2.14)	0.663; 0
USA	17, 30	467	1.30 (0.71–2.39)	0.115; 59.6	17, 30, 33, 59	1165	0.72 (0.48–1.09)	0.044; 62.9
South America	13, 53, 55	379	0.74 (0.43–1.28)	0.180; 41.6	—			
Asia	16, 18, 54, 56–58, 60, 62	2862	0.86 (0.65–1.13)	0.09; 43.3	—			
Number of cases								
<200	12, 13, 15, 17, 18, 53, 55, 56, 60	1199	0.78 (0.63–0.97)	0.365; 8.4	33	124	0.14 (0.04–0.49)	—
$\geqslant 200$	16, 30–32, 54, 57, 58, 62	3309	0.82 (0.57–1.18)	0.001; 71.3	17, 19, 30, 59, 61	1486	0.89 (0.73–1.09)	0.450; 0
Validated FFQ								
Yes	12, 13, 15, 17, 18, 30–32, 53, 56, 58	2257	0.86 (0.64–1.17)	0.003; 62.7	17, 19, 30, 33, 59, 61	1610	0.86 (0.61–1.22)	0.035; 58.4
No	16, 54, 55, 57, 60, 62	2251	0.75 (0.60–0.94)	0.312; 15.8	—		—	—
Adjustments								
BMI								
Yes	13, 15, 17, 18, 55, 58, 60	1299	0.77 (0.59–1.01)	0.181; 32.4	17, 19, 33, 59, 61	1328	0.83 (0.53–1.30)	0.019; 66.1
No	12, 16, 30–32, 53, 54, 56, 57, 62	3209	0.85 (0.62–1.15)	0.004; 62.4	30	282	0.94 (0.59–1.48)	—
Smoking/alcohol								
Yes	12, 13, 15–17, 32, 53, 55, 56, 58, 60	2637	0.77 (0.62–0.96)	0.032; 47.9	17, 19, 33, 59, 61	1328	0.83 (0.53–1.30)	0.019; 66.1
No	18, 30, 54, 57, 62	1871	0.93 (0.55–1.58)	0.038; 60.6	30	282	0.94 (0.59–1.48)	—
Dietary energy intake								
Yes	12, 13, 17, 30–32, 55	1328	0.74 (0.51–1.08)	0.006; 67.0	17, 19, 30, 33, 59, 61	1610	0.86 (0.61–1.22)	0.035; 58.4
No	15, 16, 18, 53, 54, 56–58, 60, 62	3180	0.86 (0.68–1.09)	0.113; 37.0	—		—	—
Symptoms of GOR								
Yes	17	185	0.98 (0.61–1.59)	—	19, 59, 61	651	1.07 (0.76–1.53)	0.432; 0
No	12, 13, 15–18, 30–32, 53–58, 60, 62	4323	0.75 (0.60–0.94)	0.005; 53.9	17, 30, 33	959	0.63 (0.34–1.17)	0.019; 74.9

Abbreviations: BMI, body mass index; CI, confidence interval; EAC, esophageal adenocarcinoma; EC, esophageal cancer; ESCC, esophageal squamous cell carcinoma; FFQ, food frequency questionnaire; GOR, gastro-esophageal reflux; SRR, summary relative risk.

significant reduction in risk in the highest group compared with the lowest group of fish intake was reported (SRRs = 0.78, 95% CIs: 0.59–1.03). In subgroup analyses by number of cases (<200 vs $\geqslant 200$), and adjustments for confounders, such as BMI, smoking, alcohol use, symptoms of GOR and dietary energy intake, most of the SRRs for EAC risk with fish intake were not significant. (Table 2) We did not assess publication bias due to the small number of studies included.

DISCUSSION

The present meta-analysis, carried out on 24 studies and 6677 cases, showed no appreciable overall association between fish consumption and risk of both ESCC and EAC, although we observed a weak association between fish consumption and ESCC risk in hospital-based case–control studies. The lack of association is further supported by the consistency of the results across strata of geographic region, type of FFQ and several covariates.

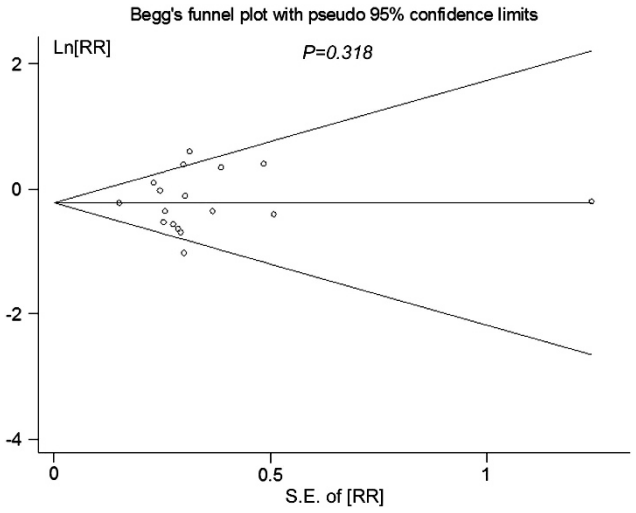

Figure 3. Begg's funnel plot of fish consumption and risk of esophageal squamous cell carcinoma ($P = 0.318$).

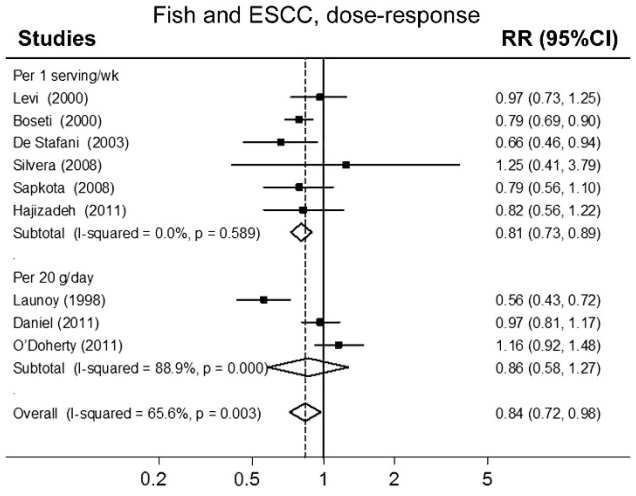

Figure 4. Linear dose–response meta-analyses of fish intake and the risk of esophageal squamous cell carcinoma. Studies are subgrouped according to unit of consumption.

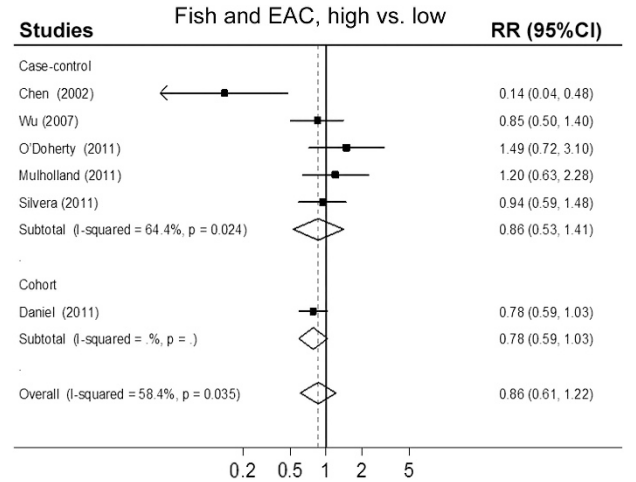

Figure 5. High vs low meta-analyses of fish intake and the risk of esophageal adenocarcinoma. Studies are subgrouped according to design.

Significant heterogeneity was detected among these studies, which may be partially explained by differences in study design.

We performed analysis specific for ESCC and EAC, which was considered appropriate, because the two forms of EC may be different in the etiology, clinicopathological characteristics. To our knowledge, this is the first meta-analysis of fish intake and ESCC and EAC risk based on high vs low analysis and linear dose–response meta-analysis. In addition, all of the included studies evaluated multiple confounders including BMI, total energy intake, alcohol use, symptoms of GOR and smoking and so on, and the relationships in each study were derived from regression after adjustment at least for age and sex.

Several plausible mechanisms have been proposed to explain the hypothesis that fish intake may protect against the development of cancers, including EC. Fish are rich in the long-chain *n*-3 polyunsaturated fatty acids (PUFAs), which are important components of cell membranes. The long-chain *n*-3 PUFAs have been reported to suppress mutations, reduce inflammatory processes, inhibit cell growth and enhance cell apoptosis, thus reduce the risk of cancer.[34,35] Animal studies and clinical observational studies have shown that supplementing the diet with *n*-3 fatty acids has suppressed the growth or development of various types of cancers.[36–39] In addition, *n*-3 PUFAs have a direct role in the inhibition of cyclooxygenase-2.[40,41] Cyclooxygenase-2 is an enzyme involved in the production of prostaglandins, which is also implicated in the cancer process.

However, the present meta-analysis has several limitations. First, of these 24 included studies, 21 studies used a case–control design, which is susceptible to recall biases. In addition, of these 21 case–control studies, 14 studies used a hospital-based case–control design, which is subject to selection bias from the selection of cases and controls. Indeed, we found a significant inverse association between ESCC risk and fish intake among hospital-based case–control studies, whereas this was not made for cohort and a population-based designs. We should treat this result with caution.

Second, the ranges and units of fish intake varied considerably. Indeed, results based on portions per week, servings per month, grams per day and servings per year were reported. Some studies reported data based on tertiles or quartiles of consumption without demarcating the cut-points of exposure. In addition, six studies used non-validated FFQ to assess dietary exposure. All of these may lead to measurement errors in the assessment of dietary intake, which can lead to overestimation of the range of intakes and underestimation of the magnitude of the relationship between dietary intake and cancer risk.[42]

Third, severe heterogeneity is found across included studies ($I^2 = 51.9\%$ for ESCC and $I^2 = 58.4\%$ for EAC). The great heterogeneity across studies may be partially explained by varying definitions of fish in different studies, because different fish have different nutritional profiles and biological effects, one obvious example being fresh, fried and salted fish. Many studies have reported increased cancer risks associated with consumption of salted fish.[43,44] Compared with marine fish, fresh-water fish contain lower levels of *n*-3 but higher levels of *n*-6 fatty acids, which are reported to have no significant association with cancer risk.[37,45] Frying is found to considerably increase the *n*-6 PUFA/ *n*-3 PUFA ratio in fish,[46] and deep-frying could generate oxidized lipids, trans-fatty acids or food mutagens such as heterocyclic amines, which may promote carcinogenesis and are found to be associated with elevated EC risk.[47,48] However, we only assessed total fish consumption because most of these studies were not primarily designed to investigate the effect of fish consumption on EC risk, and did not focus on different species or preparation methods of fish. This limitation might contribute to the null or marginal findings in this meta-analysis. Further studies are clearly warranted to focus on the different types and

preparation methods of fish and their potentially different associations with EC risk.

Fourth, residual confounders are always of concern in observational studies. Tobacco smoking is an established risk factor for ESCC and EAC development. Alcohol abuse is a known risk factor for ESCC,[49] although may not be associated with EAC risk.[50] Symptoms of GOR are known risk factors for EAC, but have no relationship with ESCC development. BMI (or obesity) is suggested to be a risk factor for EAC,[51] but a protecting factor for ESCC.[4] In addition, adjustment for total energy intake is of importance in nutritional epidemiological studies to account for potential confounding by dietary correlates.[52] When we conducted separate subgroup analyses according to studies controlled for these variables, the null or inverse association between fish intake and EC risk was not significantly changed.

Fifth, small studies with null results tend not to be published, only articles published in the English language were included, and we did not search for unpublished studies or original data. So, a degree of publication bias is unavoidable, although no publication bias is indicated visually or in formal statistical testing for studies on ESCC.

In summary, our findings suggest that fish consumption might not have an appreciable role in the prevention of both ESCC and EAC. It might be inferred that mixing all fish species and preparation methods may have masked the potential inverse association of fish intake with EC risk. We, therefore, believe that an extensive analysis of fish subgroups (for example, non-fried fish, fried fish, fresh fish, salted fish) for EC (especially for EAC) risk is warranted in the future.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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