

RISK OF ADENOCARCINOMA OF THE STOMACH AND ESOPHAGUS WITH MEAT COOKING METHOD AND DONENESS PREFERENCE

Mary H. WARD,^{1*} Rashmi SINHA,¹ Ellen F. HEINEMAN,¹ Nathaniel ROTHMAN,¹ Rodney MARKIN,² Dennis D. WEISENBURGER,² Pelayo CORREA³ and Shelia Hoar ZAHM¹

¹Division of Cancer Epidemiology and Genetics, National Cancer Institute, Bethesda, MD

²Department of Pathology and Microbiology, University of Nebraska Medical Center, Omaha, NE

³Department of Pathology, Louisiana State University, New Orleans, LA

Meats cooked at high temperatures (frying, grilling) and for a long duration contain heterocyclic amines (HCAs), which are both mutagens and animal carcinogens. Additionally, barbecuing/grilling of meats produces polycyclic aromatic hydrocarbons (PAHs). Consumption of well-done meat has been associated with an increased risk of colon cancer but has not been evaluated as a risk factor for stomach or esophageal cancers. We conducted a population-based case-control study in 66 counties of eastern Nebraska. Telephone interviews were conducted with white men and women diagnosed with adenocarcinoma of the stomach (n = 176) and esophagus (n = 143) between July 1988 and June 1993 and 502 controls. The dietary assessment included several questions about usual cooking methods for meats and doneness preference for beef. High intake of red meat was associated with increased risks for both stomach and esophageal cancers. Overall, broiling or frying of beef, chicken or pork was not associated with the risk of these tumors. Barbecuing/grilling, reported as the usual cooking method for a small number of study participants, was associated with an elevated risk of stomach and esophageal cancers. After excluding those who reported usually barbecuing/grilling, a source of both PAHs and HCAs, we evaluated doneness level as a surrogate for HCA exposure. Compared to a preference for rare/medium rare beef, odds ratios were 2.4 for medium, 2.4 for medium well and 3.2 for well done, a significant positive trend. Doneness level was not associated with a significant trend in risk of esophageal cancer. *Int. J. Cancer*, 71:14–19, 1997.

© 1997 Wiley-Liss, Inc.†

Dietary risk factors for stomach and esophageal cancers have been evaluated extensively. Low intake of fruits and vegetables increases the risk of stomach and esophageal cancers (Graham *et al.*, 1990; Steinmetz and Potter, 1991; Correa *et al.*, 1985; Risch *et al.*, 1985; Buiatti *et al.*, 1989; Tavani *et al.*, 1994; Ziegler *et al.*, 1981). An increased risk of stomach cancer has also been associated with high intake of salt and processed meats containing nitrite (Graham *et al.*, 1990; Risch *et al.*, 1985; Buiatti *et al.*, 1989, 1990).

Consumption of fried or broiled meats has been investigated as a risk factor for stomach cancer, but results have been equivocal (Kato *et al.*, 1992; Steineck *et al.*, 1993; Knekt *et al.*, 1994; IARC, 1993; Jedrychowski *et al.*, 1992). A few studies of stomach and esophageal cancers have shown elevated risks with high intake of smoked or barbecued meats (Correa *et al.*, 1985; Wu-Williams *et al.*, 1990; Risch *et al.*, 1985; Castelletto *et al.*, 1994). Consumption of “well-done” or “well-browned” meat has been associated with an increased risk of colorectal cancers (Schiffman and Felton, 1990; Lang *et al.*, 1994; Gerhardsson de Verdier *et al.*, 1991) but has not been evaluated for stomach or esophageal cancers. Cooking methods are of interest because frying and grilling/barbecuing (cooking over an open flame, charcoal or ceramic briquets) are high-temperature cooking methods which can produce high levels of heterocyclic amines (HCAs) and other pyrolysis products compared to low or negligible levels found with boiling, baking or roasting (Adamson, 1990; Layton *et al.*, 1995; Sinha *et al.*, 1995). Grilling/barbecuing of meats also produces polycyclic aromatic hydrocarbons (PAHs), which form when fat drips onto the flame or heat surface, pyrolyses and comes up in smoke, which coats the

surface of the meat. HCAs increase with increasing duration of cooking or “doneness” of the meat (Sinha *et al.*, 1995; Knize *et al.*, 1996). Oral administration of HCAs and PAHs has produced gastro-intestinal tract tumors in animal studies (Adamson, 1990; Wattenberg *et al.*, 1979).

We conducted a population-based case-control study of adenocarcinoma of the stomach and esophagus in Nebraska. The primary purpose of our study was to evaluate drinking water nitrate exposure and agricultural exposures as risk factors for these tumors. A secondary purpose of the study was to compare risk factors for adenocarcinomas of the esophagus, stomach cardia and distal stomach. The descriptive epidemiology of adenocarcinomas of the distal stomach and esophagus differ. Stomach cardia tumors share many characteristics with the adjacent esophageal tumors, including an increasing incidence in the United States and in other developed countries (Blot *et al.*, 1991; Powell and McConkey, 1990; Møller, 1992). We now present results for meat and gravy intake, meat cooking methods and doneness preference, information which was ascertained as part of the dietary assessment.

METHODS

A population-based case-control interview study of stomach and lower esophageal cancers was conducted in eastern Nebraska. For efficiency and to reduce the cost of the study, controls were randomly selected from a group of controls from a previous population-based case-control study of hematopoietic cancers in Nebraska (Zahm *et al.*, 1990) and were re-interviewed at the time of the case interviews.

Cases

Cases were white men and women aged 21 years or older, who had been newly diagnosed with adenocarcinoma of the stomach or esophagus (ICD-O codes 150, 151) between July 1, 1988, and June 30, 1993. Cases were limited to whites because the control group excluded other ethnic groups due to expected small numbers. Cases were residents of 66 counties in eastern Nebraska at the time of the interview. Cases who were not resident in Nebraska during 1983–1985 (when controls were identified) were excluded (n = 6) from the analyses so that the same residence criteria applied to cases and re-interviewed controls.

Cases from 1988 through 1990 were identified from the Nebraska Cancer Registry. Cases from 1991 through June 30, 1993, were identified by review of discharge diagnoses and pathology records at the 14 hospitals in Omaha, Lincoln and Grand Island. These 14 hospitals accounted for about 90% of the stomach and esophageal cancer diagnoses in the eastern 66 counties.

*Correspondence to: Division of Cancer Epidemiology and Genetics, National Cancer Institute, 6130 Executive Blvd. EPN-418, Bethesda, MD 20892-7364, USA. Fax: (301) 402-1819.

TABLE I – NUMBERS OF ELIGIBLE INTERVIEWED CASES OF ADENOCARCINOMA OF THE STOMACH AND ESOPHAGUS AND CONTROLS BY GENDER, YEAR OF BIRTH AND VITAL STATUS

Characteristic	Stomach (n = 170)		Esophagus (n = 137)		Controls (n = 502)	
	n	%	n	%	n	%
Gender						
Men	97	57.1	121	88.3	284	56.6
Women	73	42.9	16	11.7	218	43.3
Year of birth						
Before 1915	90	52.9	39	28.5	160	31.9
1915–1924	45	26.5	48	35.0	121	24.1
1925–1939	25	14.7	37	27.0	116	23.1
1940+	10	5.9	13	9.5	105	20.9
Vital status						
Alive	47	27.6	43	31.4	211	42.0
Deceased	123	72.4	94	68.6	291	58.0
Respondent type						
Self	34	20.0	33	24.1	198	39.4
Next of kin	136	80.0	104	75.9	304	60.6
Spouse	48	28.2	64	46.7	135	26.9
Son/daughter	62	36.5	29	21.2	106	21.1
Sibling/parent	10	5.9	11	8.0	34	6.8
Other relative	15	8.8	0	0.0	23	4.6
Friend/other	1	0.6	0	0.0	6	1.2

Only cases confirmed as adenocarcinoma of the stomach and esophagus were retained in the analyses. A gastro-intestinal pathologist (R.M.) reviewed all initial diagnoses of stomach cancer except for lymphomas, leiomyosarcomas and reticulosarcomas and all diagnoses of esophageal cancer except those located in the upper and cervical esophagus (ICD-O codes 150.0, 150.3), which are almost exclusively squamous cell tumors. Cases were classified by anatomical subsite and by histological type (intestinal, diffuse or mixed) using the Lauren (1965) classification. A second gastro-intestinal pathologist (P.C.), who has extensive experience with the Lauren classification, reviewed a subsample (n = 43) of the cases. Agreement was 84%.

Interviews were obtained for 79% of the eligible stomach cancer cases (97 men and 73 women) and for 88% of the esophageal cancer cases (121 men and 16 women). Twenty percent of the interviews with stomach cancer cases and 24% of the interviews with esophageal cancer cases were obtained from the subjects themselves. Next-of-kin were the respondents for those who were deceased or too ill to participate. Surrogate interviews were with the spouse or a son or daughter for 81% of the stomach cancer cases and 89% of the esophageal cancer cases.

Controls

In 1986, the National Cancer Institute (NCI) collaborated with the University of Nebraska Medical Center in conducting a case-control study of non-Hodgkin's lymphoma, multiple myeloma, Hodgkin's disease and chronic lymphocytic leukemia. Population controls were identified from 66 eastern counties of Nebraska and were frequency-matched to the hematopoietic cancer cases by their gender, age (in 5 year groups) and vital status in a 3:1 ratio. Controls under the age of 65 years were selected from the general population (in 1985–1986) by random digit dialing. Subjects aged 65 years and over were identified from Health Care Financing Administration Medicare files. Controls for deceased cases were selected from Nebraska mortality records with the additional matching factor of year of death (1983–1985).

The gender, year of birth and vital status distribution of the controls overlapped adequately with the distribution of stomach and esophageal cases (Table I). A total of 502 eligible controls were re-interviewed. Deceased cases and controls were not matched on year of death. Interviews were conducted with the subjects themselves for 39% of controls. The remaining interviews were obtained from surrogate respondents, the majority of which were interviews with a spouse or a son or daughter (79%).

The response rate in the lymphoma study, which took into account the refusals at stage 1 of the random digit dialing, was 87%. In the current study, the response rate for the re-interview of the sample of controls was 83%, giving an adjusted overall response rate of 72%.

Interviews and dietary assessment

Cases and controls or their next-of-kin were interviewed by telephone during 1992–1994. Interviews included information about agricultural exposures; a life-time occupational, residential and water source history; tobacco and alcohol use; diet; and a medical and familial cancer history.

Dietary information was obtained using a modified version of the Health Habits and History Questionnaire (HHHQ) (Block *et al.*, 1990) that focused on nitrate, nitrite, sodium, vitamin C, carotenes and animal protein. Following the questions about the frequency of intake of beef, pork and chicken, we asked about the usual cooking method (fried/broiled, baked/roasted, boiled/poached, other method to be specified). For beef, we also asked how well cooked the individual usually ate beef (well done, medium well, medium, medium rare, rare). Doneness preference for pork and chicken was not ascertained. We evaluated intake of beef (steaks, roasts, hamburgers), processed meat (bacon, sausage, luncheon meats, hot dogs, ham, home-cured meat) and all red meat (the beef and processed meat groups, pork, liver), and risks were calculated for quartiles of weekly intake using the lowest intake quartile as the reference group. The weekly frequency of consumption of gravy made with meat juices was also evaluated because meat juices can contain high levels of HCAs.

Data analysis

We excluded cases and controls who had unknown intakes for 20% or more of the food questions. A total of 154 (91%) of the stomach cancer cases, 124 (91%) of the esophageal cancer cases and 449 (89%) controls were included in the dietary analyses. Maximum likelihood estimates of the year-of-birth and gender-adjusted odds ratios (ORs) and 95% confidence intervals (95% CI) were calculated using stratified analysis (Gart, 1970). Multiple logistic regression analysis was used to evaluate the effects of multiple factors simultaneously and to evaluate confounding. Adjustment for total calories (as a continuous variable) and non-dietary risk factors, including education level, parental origin and familial history of cancer (see "Results"), did not change the ORs substantially. All ORs were adjusted for the matching factors of year of birth and gender. ORs for meat doneness, cooking method and gravy intake were adjusted for red meat intake, which changed the ORs somewhat. Trend tests for categorized exposure variables were performed by assigning ordinal scores to the categories and testing for a non-zero slope. We evaluated the combined effect of doneness preference and beef intake.

When the numbers of cases were adequate, ORs were calculated separately for stomach cardia (n = 30) and distal stomach tumors (n = 124) and by the Lauren (1965) histologic types (intestinal n = 79, diffuse n = 53, mixed n = 16).

RESULTS

Risk factors for stomach cancer in this study population were similar to those seen in some other studies. For stomach cancer, ORs were significant or marginally significant for parental origin (one or more parent born outside the United States OR = 2.1, 95% CI = 1.4–3.3), family history of cancer (any family history of gastro-intestinal cancer compared with no family history OR = 2.4, 95% CI = 1.4–4.2) and vitamin use for 1 year or more (OR = 0.6, 95% CI = 0.4–0.9). In contrast to many other stomach cancer studies, there was not a significant association with education level (high school or greater compared to less than high school OR = 0.9, 95% CI 0.6–1.3), an indicator of socio-economic status. Dietary risk factors which were associated with stomach cancer risk, comparing the highest quartile of intake to the lowest quartile

TABLE II – ODDS RATIOS FOR STOMACH AND ESOPHAGEAL ADENOCARCINOMAS WITH FREQUENCY OF INTAKE OF MEAT AND GRAVY

Meat group (times/week)	Stomach cancer			Esophageal cancer	
	Number of cases	Number of controls	OR ¹ (95% CI)	Number of cases	OR ¹ (95% CI)
Total red meat ²					
<8	18	99	1.0	16	1.0
8–12	29	113	1.4 (0.7–2.9)	25	1.3 (0.6–2.9)
13–18	43	111	2.1 (1.1–4.2)	32	1.4 (0.7–2.9)
19+	64	126	2.4 (1.3–4.8)	51	2.0 (1.0–4.0)
<i>p</i> for trend:		<0.001			0.02
Processed meats ³					
<4	32	125	1.0	25	1.0
4–5	35	102	1.4 (0.8–2.7)	22	1.3 (0.6–2.6)
6–8	40	118	1.3 (0.7–2.4)	31	1.1 (0.6–2.1)
8+	47	104	1.6 (0.9–2.9)	46	1.7 (0.9–3.3)
<i>p</i> for trend:		0.06			0.04
Beef (steaks/roasts, hamburgers)					
<3	30	115	1.0	26	1.0
3–4	65	179	1.5 (0.9–2.6)	58	1.4 (0.8–2.6)
5	22	54	1.8 (0.9–3.7)	14	1.0 (0.4–2.3)
6+	37	101	1.6 (0.9–3.0)	26	1.1 (0.6–2.1)
<i>p</i> for trend:		0.06			0.37
Gravy made with meat juices ⁴					
<0.7	17	86	1.0	18	1.0
0.7–1	32	126	1.2 (0.6–2.3)	27	1.0 (0.5–2.0)
2–3	56	138	1.5 (0.8–2.9)	35	1.0 (0.5–2.1)
4+	48	91	1.6 (0.8–3.3)	43	2.3 (1.0–5.0)
<i>p</i> for trend:		0.01			0.01

¹Adjusted for gender and year of birth. ²Includes beef group, beef stew/pot pie, processed meats, fresh ham/pork, liver. ³Includes bacon, sausage, processed ham, home-cured meats, sandwich meats such as bologna or salami, hot dogs. ⁴ORs adjusted for gender, year of birth and red meat intake.

(other than the meat and cooking-related ORs) included sodium (OR = 1.7, 95% CI = 0.9–3.4), protein (OR = 1.9, 95% CI = 1.1–3.4), saturated fat (OR = 2.4, 95% CI = 1.3–4.4), total fat (OR = 1.8, 95% CI = 1.0–3.1) and total calories (OR = 1.6, 95% CI = 0.9–2.9). Risk factors for adenocarcinoma of the esophagus were smoking (ever smoked cigarettes 3 months or more compared to never used tobacco OR = 2.2, 95% CI = 1.2–4.1), alcohol consumption (>30 years compared to never for >1 year OR = 1.6, 95% CI = 0.9–3.0) and body mass index (highest quartile of Quetelet's index compared with the lowest quartile OR = 1.9, 95% CI = 1.0–3.6). Dietary risk factors (highest vs. lowest quartile of intake) included vitamin C (OR = 0.5, 95% CI = 0.3–0.9) and beta-carotene (OR = 0.4, 95% CI = 0.2–0.8).

Intake of processed meats, beef and total red meat was categorized into quartiles, and year-of-birth- and gender-adjusted ORs were calculated using the lowest intake quartile as the reference group. There were significantly increasing risks of stomach and esophageal cancers with increasing red meat intake (Table II). The major components of the red meat group were processed meats and beef. High intake of processed meats was associated with an elevated risk of stomach and esophageal cancers, while high beef intake was associated with an increased risk of stomach cancer but not esophageal cancer. Total red meat intake showed the strongest relationship with risk. The upper quartile of red meat intake was associated with about a 2-fold increased risk of stomach and esophageal cancers compared with the lowest quartile of <8 times per week (stomach OR = 2.4, 95% CI 1.3–4.8; esophagus OR = 2.0, 95% CI 1.0–4.0). Next-of-kin and self-respondents exhibited similar risks. Saturated fat, total fat and protein intake were highly correlated with red meat intake (Pearson correlation coefficients, $r > 0.8$); therefore, we did not adjust for intake of these dietary components. Adjustment for total calorie intake did not change the ORs substantially.

Beef that is roasted in the oven contains few HCAs even when it is cooked well done. However, the fat drippings and meat juices contain HCAs, and gravy made from juices of well-done beef contains high levels (R. Sinha, Bethesda, personal communication). We categorized the weekly intake of gravy made with meat

juices into approximate quartiles and calculated ORs adjusted for year of birth, gender and red meat intake. Consumption of gravy 4 or more times per week was associated with a 60% increased risk of stomach cancer and a greater than 2-fold increased risk of esophageal cancer. The association differed by respondent type. Among next-of-kin respondents, there was no association between stomach cancer and gravy intake (adjusted OR = 1.0, 95% CI 0.4–2.3) and a weaker association with esophageal cancer (adjusted OR = 1.5, 95% CI 0.6–3.7). In contrast, among self-respondents, the adjusted ORs for the highest quartile of gravy consumption were 4.2 (95% CI 0.9–18.9) for stomach cancer (12 cases, 23 controls) and 7.9 (95% CI 1.3–46.5) for esophageal cancer (14 cases, 23 controls). Adjustment for red meat intake decreased the ORs for gravy use slightly compared with the year-of-birth and gender-adjusted ORs.

Frying and broiling were the most commonly reported cooking techniques for beef. Table III shows the ORs by usual cooking method for beef, comparing the higher temperature cooking methods of frying/broiling and barbecuing/grilling to the lower temperature methods of baking/roasting. Frying or broiling was not associated with risk of stomach or esophageal cancer. However, within the frying/broiling group, increasing doneness preference was associated with an increasing risk of stomach cancer but not esophageal cancer. Compared with a preference for fried/broiled rare or medium rare beef, ORs were 2.4, 2.1 and 3.2 for medium, medium well and well done. The ORs for doneness preference decreased when the referent group included those whose usual cooking method was baking or roasting (all doneness preferences combined).

Grilling/barbecuing, reported as the usual cooking technique by a small number of subjects, was associated with non-significant 2-fold increased risk of stomach cancer and a 50% non-significantly elevated risk of esophageal cancer (Table III). The ORs are adjusted for red meat intake, which slightly increased the magnitude of the associations for barbecuing. Adjustment for other dietary factors did not change the ORs substantially.

TABLE III – ODDS RATIOS FOR STOMACH AND ESOPHAGEAL ADENOCARCINOMAS WITH BEEF COOKING METHOD

Method	Stomach cancer			Esophagus cancer		
	Number of cases	Number of controls	OR ¹ (95% CI)	Number of cases	Number of controls	OR ¹ (95% CI)
Baked/roasted/boiled	14	38	1.0	10	38	1.0
Fried/broiled	128	379	1.1 (0.6–2.1)	101	379	1.0 (0.4–2.1)
Grilled/barbecued	8	21	1.9 (0.6–5.6)	9	21	1.5 (0.5–4.8)

¹Adjusted for gender, year of birth and weekly red meat intake.

TABLE IV – ODDS RATIOS FOR ADENOCARCINOMA OF THE STOMACH AND ESOPHAGUS WITH DONENESS PREFERENCE FOR BEEF¹

Doneness level	Stomach			Esophagus	
	Cases	Controls	OR ² (95% CI)	Cases	OR ² (95% CI)
Rare/medium rare	7	60	1.0 ³	14	1.0 ³
Medium	21	76	2.4 (0.9–6.2)	16	1.0 (0.4–2.3)
Medium well	25	84	2.4 (0.9–6.1)	30	1.8 (0.9–3.9)
Well	93	206	3.2 (1.4–7.6)	53	1.5 (0.7–2.9)

¹Excludes those who usually barbecued/grilled beef. ²Adjusted for gender, year of birth and weekly red meat intake. ³*p* for trend: stomach cancer *p* = 0.004, esophageal cancer *p* = 0.35.

The ORs for frying/broiling were similar for self- and next-of-kin respondents. Among next-of-kin respondents, the ORs for barbecuing were 2.5 for stomach cancer and 3.1 for esophageal cancer. Among cases reporting for themselves, only 2 stomach cases and no esophageal cases reported barbecuing as their usual cooking method. Broiling/frying of pork and chicken was not associated with risk of stomach and esophageal tumors (data not shown). The numbers of individuals reporting barbecuing as their usual cooking technique for these meats were too few to evaluate.

The levels of HCAs produced in meat increase with increasing duration of cooking (doneness). The cooking method of barbecuing/grilling produces PAHs in addition to HCAs, so we excluded those who usually barbecued beef to better evaluate doneness level as a surrogate for HCA exposure (Table IV). We observed an increasing risk of stomach cancer with increasing doneness. Among those reporting for themselves, there was a stronger association with well-done beef (OR = 9.9, 95% CI 1.1–87) based on 17 cases and 65 controls compared to those with next-of-kin respondents (OR = 2.1, 95% CI 0.8–5.7) based on 63 cases and 109 controls. The confidence intervals are wide due partly to a small number of cases in the reference categories (1 case among self-respondents, 6 cases among next-of-kin respondents). Doneness preference was not strongly or monotonically associated with esophageal cancer overall (Table IV) nor among self- and next-of-kin respondents.

We evaluated the combined effect of doneness preference and intake of beef, excluding those who usually barbecued beef. Table V shows ORs for stomach cancer with doneness level stratified by beef intakes below the median and at or above the median. There was no statistical interaction between beef intake and doneness level. Risk increased with doneness level in both low- and high-intake categories. The OR for high intake of well-done beef compared with low intake of rare or medium rare beef was 5.3 (95% CI 1.5–18.9). For esophageal cancer, there was no significant association for any combination of doneness preference and beef intake and no evidence of an interaction (data not shown).

Stomach cardia tumors have been hypothesized to have risk factors similar to adenocarcinomas of the esophagus due to the close proximity of the tumor sites and recent increases in incidence of both tumors. We compared the risks for stomach cardia with the risks for distal stomach tumors. The ORs for stomach cardia did not differ substantially from the ORs for the distal stomach tumors for red meat intake, beef intake, beef cooking method and doneness preference. The association with high gravy intake (≥ 4 times per week vs. < 0.7 times per week) was stronger among stomach cardia cases (OR = 3.2, 95% CI 1.0–9.8) and closer to the risk for

esophageal tumors compared to the risk for distal stomach tumors (OR = 1.4, 95% CI 0.6–3.1).

We compared risks for the intestinal and diffuse morphologic types of stomach cancer as classified by Lauren (1965). The association with high gravy intake was slightly stronger for the intestinal type of stomach cancer (OR = 1.8, 95% CI 0.7–4.8) compared with the diffuse type (OR = 1.2, 95% CI 0.4–3.4). Red meat intake, cooking method and doneness preference showed similar associations for both histologic types. A small number of stomach tumors were of the mixed histologic type (*n* = 16) and were too few to evaluate.

DISCUSSION

In this study, we observed that a high intake of red meat was associated with a significant 2.4-fold risk of stomach cancer and a 2-fold risk of esophageal cancer. Processed meats and beef, the major components of the red meat group, showed a positive association with risk of stomach cancer, while only processed meat intake was associated with esophageal cancer risk. Red meat intake was highly correlated with the intake of total fat, saturated fat and protein; therefore, it was not possible to assess the independent effects of each factor.

The cooking methods of frying and broiling were not associated with an increased risk of stomach or esophageal cancers compared to baking or roasting. Grilling/barbecuing of beef was associated with about a 2-fold non-significantly increased risk of stomach cancer and a smaller non-significant excess of esophageal cancer. Few people reported grilling or barbecuing as their usual cooking technique, probably due to the seasonal nature of this activity, and the associations were based on small numbers. The level of doneness preference for beef was associated with a significantly increasing risk of stomach cancer. A preference for well-done beef was associated with a 3.2-fold increased risk of stomach cancer compared to a preference for rare or medium rare beef. Doneness preference was not strongly associated with esophageal cancer risk.

High intake of gravy made with meat juices was associated with an increasing risk of stomach cancer and an elevated risk of esophageal cancer in the highest-intake quartile. Gravy made from meats cooked well done can contain high levels of HCAs (R. Sinha, personal communication).

Most previous studies of stomach cancer have not shown an association with fresh meat intake, though positive associations with processed meat have been observed (Boeing *et al.*, 1991; Gonzalez *et al.*, 1991). High intakes of fresh meat have generally been protective in epidemiologic studies of esophageal cancer in developed countries (Ziegler *et al.*, 1981; Franceschi, 1993). Some previous studies of stomach cancer have shown an increased risk with high intake of fried or broiled meats (Kato *et al.*, 1992; Jedrychowski *et al.*, 1992). However, other studies have shown no association (Graham *et al.*, 1972; Knekt *et al.*, 1994; Hansson *et al.*, 1993). Barbecuing/grilling was not associated with stomach cancer in the few studies which have evaluated it (Correa *et al.*, 1985; Hansson *et al.*, 1993), but consumption of smoked foods or meats has been associated with an increased risk of stomach cancer in most studies (Boeing *et al.*, 1991; Correa *et al.*, 1985; Risch *et al.*, 1985; Lee *et al.*, 1990; Falcao *et al.*, 1994). Eating barbecued meat more than once per week was associated with a 2-fold increased

TABLE V – ODDS RATIOS FOR STOMACH CANCER BY PREFERRED BEEF DONENESS AND FREQUENCY OF BEEF INTAKE¹

Doneness level	Beef intake					
	<4/week			≥4/week		
	Cases	Controls	OR ² (95% CI)	Cases	Controls	OR ² (95% CI)
Rare/medium rare	3	38	1.0	4	22	2.0 (0.4–10.3)
Medium/medium well	28	101	3.0 (0.8–10.9)	18	59	3.5 (0.9–13.0)
Well	59	138	3.8 (1.1–13.3)	34	68	5.3 (1.5–18.9)

¹Excludes those who usually barbecued/grilled beef. ²Adjusted for gender and year of birth.

risk of squamous cell esophageal cancer in a study in Argentina (Castelletto *et al.*, 1994), but few studies of esophageal cancer have evaluated meat cooking techniques.

HCAs are formed by the pyrolysis of creatinine and amino acids in the meat juices during high-temperature cooking (Adamson, 1990; Skog, 1993; Sugimura *et al.*, 1988). These compounds are among the most potent mutagens tested by the Ames/Salmonella bioassay and are carcinogenic in mice, rats and non-human primates (Felton and Knize, 1991; Ohgaki *et al.*, 1986, 1991; Ito *et al.*, 1991). In addition to producing HCAs, grilling or barbecuing of meats produces PAHs, and smoked meats also contain PAHs. A possible role for PAHs in the carcinogenesis of upper gastrointestinal tract tumors has been suggested from animal studies (Wattenberg *et al.*, 1979; F. Beland, personal communication) and epidemiologic studies evaluating smoked foods, tobacco and occupational exposures (Blot, 1994). To evaluate a possible link between stomach cancer and PAH exposure separately from HCA exposure, it would be necessary to obtain more detailed information about barbecuing, consumption of smoked foods and occupational exposures.

Overall, we found no association between the cooking methods of frying/broiling and stomach cancer risk. At face value this does not support a role for HCAs in stomach cancer risk. However, limitations of the questions about meat cooking method may have led to substantial misclassification and may partly explain the lack of an association (Sinha and Rothman, 1997). Specifically, HCA levels are different for frying and broiling and for different cuts of beef (R. Sinha, personal communication). Non-differential misclassification usually attenuates risk estimates (Flegal *et al.*, 1986; Correa *et al.*, 1995).

Doneness preference may be a better surrogate for exposure to HCAs or other pyrolysis products than a question about usual cooking method since there may be a uniform preference across cuts of meat and cooking methods. Indeed, we did observe an association with doneness preference within the group who usually fried or broiled beef. The strength of the association between doneness level and stomach cancer in spite of the limitations of the questionnaire data was striking.

ORs for doneness preference and gravy intake were stronger among self-respondents and may have been due to differences in

reported intake between these 2 types of controls. Median gravy intake for deceased controls was 2 times per week compared to self-respondent reports of 0.7 times per week. Higher reported intakes among deceased controls would reduce the risk estimates. Other studies have shown that controls chosen from mortality files have a higher intake of meat, a lower fruit and vegetable intake and a higher proportion of heavy drinking and smoking compared with living persons of the same age and gender (McLaughlin *et al.*, 1985). Differences in risk estimates by respondent type may also be due to differences in the quality of the information. Next-of-kin respondents may give less accurate information than subjects about dietary intakes, which would lead to non-differential misclassification and attenuation of the risk estimates. Surrogate dietary information provided by spouses, particularly from wives reporting on their husbands' dietary intakes, has been shown to have good agreement with subject reports (Samet, 1990; Marshall *et al.*, 1980) but poorer agreement with other types of respondent (Herrmann, 1985).

A high intake of red meat was associated with elevated risks of both stomach and esophageal cancers, independent of doneness preference. The association with well-done beef consumption and stomach cancer risk suggests that dietary HCAs or some other component of well-done beef may play a role in human stomach cancer risk. Risk of esophageal adenocarcinomas did not show the strong association with doneness preference observed for stomach cancer. Our data suggest that the degree of doneness of meat (a surrogate for HCAs) may be an aspect of a high red meat diet that further increases the risk of stomach cancer. Since most of the information was based on next-of-kin responses, studies with more detailed questions about dietary exposure to HCAs and PAHs are needed to confirm or refute these suggestive findings.

ACKNOWLEDGEMENTS

The authors acknowledge Mr. T. Brooker and Ms. S. Keehn of IMS for programming support. We thank Ms. C. Russell, Mr. B. Saal of Westat, Inc., and Ms. C. Boudreau of Survey Research Associates for their contribution to data collection and management.

REFERENCES

- ADAMSON, R.H., Mutagens and carcinogens formed during cooking of food and methods to minimize their formation. In: V.T. DeVita, S. Hellman and S.A. Rosenberg (eds.), *Cancer prevention*, pp. 1–7, Lippincott, Philadelphia (1990).
- BLOCK, G., HARTMAN, A.M. and NAUGHTON, D., A reduced dietary questionnaire: development and validation. *Epidemiology*, **1**, 58–64 (1990).
- BLOT, W.J., Esophageal cancer trends and risk factors. *Semin. Oncol.*, **21**, 403–410 (1994).
- BLOT, W.J., DEVESA, S.S., KNELLER, R.W. and FRAUMENI, J.F., JR., Rising incidence of adenocarcinoma of the esophagus and gastric cardia. *J. Amer. Med. Ass.*, **265**, 1287–1289 (1991).
- BOEING, H. and 15 OTHERS, Case-control study on stomach cancer in Germany. *Int. J. Cancer*, **47**, 858–864 (1991).
- BUIATTI, E. and 16 OTHERS, A case-control study of gastric cancer and diet in Italy: II. Association with nutrients. *Int. J. Cancer*, **45**, 896–901 (1990).
- BUIATTI, E. and 14 OTHERS, A case-control study of gastric cancer and diet in Italy. *Int. J. Cancer*, **44**, 611–616 (1989).
- CASTELLETTO, R., CASTELLSAGUÉ, X., MUÑOZ, N., ISCOVICH, J., CHOPITA, N. and JMELNITSKY, A., Alcohol, tobacco, diet, mate drinking, and esophageal cancer in Argentina. *Cancer Epidemiol. Biomarkers Prevent.*, **3**, 557–564 (1994).
- CORREA, A., STEWART, W.F., FRANCO-MARINA, F. and SEACAT, H., Bias from nondifferential misclassification in case-control studies with three exposure levels. *Epidemiology*, **6**, 276–281 (1995).
- CORREA, P., FONTHAM, E., PICKLE, L.W., CHEN, V., LIN, Y. and HAENSZEL,

- W., Dietary determinants of gastric cancer in south Louisiana inhabitants. *J. Nat. Cancer Inst.*, **75**, 645–653 (1985).
- FALCAO, J.M., DIAS, J.A., MIRANDA, A.C., LEITAO, C.N., LACERDA, M.M. and DA MOTTA, L.C., Red wine consumption and gastric cancer in Portugal: a case-control study. *Europ. J. Cancer Prev.*, **3**, 269–276 (1994).
- FELTON, J.S. and KNIZE, M.G., Occurrence, identification, and bacterial mutagenicity of heterocyclic amines in cooked food. *Mutation Res.*, **259**, 205–217 (1991).
- FLEGAL, K.M., BROWNIE, C. and HAAS, J.D., The effects of exposure misclassification on estimates of relative risk. *Amer. J. Epidemiol.*, **123**, 736–751 (1986).
- FRANCESCHI, S., Role of nutrition in the aetiology of oesophageal cancer in developed countries. *Endoscopy*, **25**(Suppl.), 613–616 (1993).
- GART, J.J., Point and interval estimates of the common odds ratio in the combination of 2×2 tables with fixed marginals. *Biometrika*, **57**, 471–475 (1970).
- GERHARDSSON DE VERDIER, M., HAGMAN, U., PETERS, R.K., STEINECK, G. and ÖVERVIK, E., Meat, cooking methods and colorectal cancer: a case-referent study in Stockholm. *Int. J. Cancer*, **49**, 520–525 (1991).
- GONZALEZ, C.A., SANZ, J.M., MARCOS, G., PITA, S., BRULLET, E., SAIGI, E., BADIA, A. and RIBOLI, E., Dietary factors and stomach cancer in Spain: a multi-centre case-control study. *Int. J. Cancer*, **49**, 513–519 (1991).
- GRAHAM, S., HAUGHEY, B., MARSHALL, J., BRASURE, J., ZIELEZNY, M., FREUDENHEIM, J., WEST, D., NOLAN, J. and WILKINSON, G., Diet in the epidemiology of gastric cancer. *Nutr. Cancer*, **13**, 19–34 (1990).
- GRAHAM, S., SCHOTZ, W. and MARTIN, P., Alimentary factors in the epidemiology of gastric cancer. *Cancer*, **32**, 927–938 (1972).
- HANSSON, L.-E., NYRÉN, O., BERGSTRÖM, R., WOLK, A., LINDGREN, A., BARON, J. and ADAMI, H.-O., Diet and risk of gastric cancer. A population-based case-control study in Sweden. *Int. J. Cancer*, **55**, 181–189 (1993).
- HERRMANN, N., Retrospective information from questionnaire. I. Comparability of primary respondents and their next-of-kin. *Amer. J. Epidemiol.*, **121**, 937–947 (1985).
- IARC, *Monographs on the evaluation of carcinogenic risks to humans. Some naturally occurring substances. Food items and constituents, heterocyclic aromatic amines and mycotoxins*, Lyon, IARC (1993).
- ITO, N., HASEGAWA, R., SANO, M., TAMANO, S., ESUMI, H., TAKAYAMA, S. and SUGIMURA, T., A new colon and mammary carcinogen in cooked food, 2-amino-1-methyl-6-phenylimidazo[4,5-b]pyridine (PhIP). *Carcinogenesis*, **12**, 1503–1506 (1991).
- JEDRYCHOWSKI, W., BOEING, H., POPIELA, T., WAHRENDORF, J., TOBIASZ-ADAMCZYK, B. and KULIG, J., Dietary practices in households as risk factors for stomach cancer: a familial study in Poland. *Europ. J. Cancer Prev.*, **1**, 297–304 (1992).
- KATO, I., TOMINAGA, S. and MATSUMOTO, K., A prospective study of stomach cancer among a rural Japanese population: a 6-year survey. *Jpn. J. Cancer Res.*, **83**, 568–575 (1992).
- KNEKT, P., STEINECK, G., JÄRVINEN, R., HAKULINEN, T. and AROMAA, A., Intake of fried meat and risk of cancer: a follow-up study in Finland. *Int. J. Cancer*, **59**, 756–760 (1994).
- KNIZE, M.G., SINHA, R., SALMON, C.P., MEHTA, S.S., DEWHIRST, K.P. and FELTON, J.S., Formation of heterocyclic amine mutagen/carcinogens during cooking of muscle meat. *J. Muscle Foods*, **7**, 271–279 (1996).
- LANG, N.P., BUTLER, M.A., MASSENGILL, J., LAWSON, M., STOTTS, R.C., HAUSER-JENSEN, M. and KADLUBAR, F.F., Rapid metabolic phenotypes for acetyltransferase and cytochrome P4501A2 and putative exposure to food-borne heterocyclic amines increases the risk for colorectal cancer or polyps. *Cancer Epidemiol. Biomarkers Prevent.*, **3**, 675–682 (1994).
- LAUREN, P., The two histological types of gastric carcinoma: diffuse and so-called intestinal type carcinoma: an attempt at a histochemical classification. *Acta Pathol. Microbiol. Scand.*, **64**, 31–49 (1965).
- LAYTON, D.W., BOGEN, T., KNIZE, M.G., HATCH, F.T., JOHNSON, V.M. and FELTON, J.S., Cancer risk of heterocyclic amines in cooked foods: an analysis and implications for research. *Carcinogenesis*, **16**, 39–52 (1995).
- LEE, H.-H., WU, H.-Y., CHUANG, Y.-C., CHANG, A.-S., CHAO, H.-H., CHEN, K.-Y., CHEN, H.-K., LAI, G.-M., HUANG, H.-H. and CHEN, C.-J., Epidemiologic characteristics and multiple risk factors of stomach cancer in Taiwan. *Anticancer Res.*, **10**, 875–882 (1990).
- MARSHALL, J., PRIORE, R., HAUGHEY, B., RZEPKA, T. and GRAHAM, S., Spouse-subject interviews and the reliability of diet studies. *Amer. J. Epidemiol.*, **112**, 675–683 (1980).
- MCLAUGHLIN, J.K., BLOT, W.J., MEHL, E.S. and MANDEL, J.S., Problems in the use of dead controls in case-control studies. I. General results. *Amer. J. Epidemiol.*, **121**, 131–139 (1985).
- MØLLER, H., Incidence of cancer of oesophagus, cardia and stomach in Denmark. *Europ. J. Cancer Prev.*, **1**, 159–164 (1992).
- OHGAKI, H., HASEGAWA, H., KATO, T., SUENAGA, M., UBUKATA, M., SATO, S., TAKAYAMA, S. and SUGIMURA, T., Carcinogenicity in mice and rats of heterocyclic amines in cooked foods. *Environ. Health Perspect.*, **67**, 129–134 (1986).
- OHGAKI, H., TAKAYAMA, S. and SUGIMURA, T., Carcinogenicities of heterocyclic amines in cooked food. *Mutation Res.*, **259**, 399–410 (1991).
- POWELL, J. and MCCONKEY, C.C., Increasing incidence of adenocarcinoma of the gastric cardia and adjacent sites. *Cancer*, **62**, 440–443 (1990).
- RISCH, H.A., JAIN, M., WON CHOI, N., FODOR, G.J., PFEIFFER, C.J., HOWE, G.R., HARRISON, L.W., CRAIB, K.J.P. and MILLER, A.B., Dietary factors and the incidence of cancer of the stomach. *Amer. J. Epidemiol.*, **122**, 947–959 (1985).
- SAMET, J.M., Surrogate sources of dietary information. In: W. Willett (ed). *Nutritional epidemiology*, pp. 133–142, Oxford University Press, New York (1990).
- SCHIFFMAN, M.H. and FELTON, J.S., Re: Fried foods and the risk of colon cancer. *Amer. J. Epidemiol.*, **131**, 376–378 (1990).
- SINHA, R., ROTHMAN, N., BROWN, E.D., SALMON, C.P., KNIZE, M.G., ROSSI, S., LEVANDER, O.A. and FELTON, J.S., High concentrations of carcinogen PHIP occur in chicken but are dependent on the cooking method. *Cancer Res.*, **55**, 4516–4519 (1995).
- SINHA, R. and ROTHMAN, N., Heterocyclic aromatic amines exposure assessment in epidemiologic studies. *Mutation Res.* (1997) (in press).
- SKOG, K., Cooking procedures and food mutagens: a literature review. *Food Chem. Toxicol.*, **31**, 655–675 (1993).
- STEINECK, G., GERHARDSSON DE VERDIER, M. and ÖVERVIK, E., The epidemiological evidence concerning intake of mutagenic activity from the fried surface and the risk of cancer cannot justify preventive measures. *Europ. J. Cancer Prev.*, **2**, 293–300 (1993).
- STEINMETZ, K.A. and POTTER, J.D., Vegetables, fruit and cancer. I. Epidemiology. *Cancer Causes Control*, **2**, 325–357 (1991).
- SUGIMURA, T., SATO, S. and WAKABAYASHI, K., Mutagens/carcinogens in pyrolysate of amino acids and proteins and in cooked foods: heterocyclic aromatic amines. In: Y.T. Woo, D.Y. Lai, J.C. Arcos and M.F. Argue (eds.), *Chemical induction of cancer, structural bases and biological mechanisms*, pp. 681–710, Academic Press, New York (1988).
- TAVANI, A., NEGRI, E., FRANCESCHI, S. and LA VECCHIA, C., Risk factors for esophageal cancer in lifelong nonsmokers. *Cancer Epidemiol. Biomarkers Prevent.*, **3**, 387–392 (1994).
- WATTENBERG, L.W., JERINA, D.M., LAM, L.K.T. and YAGI, H., Neoplastic effects of oral administration of (\pm)-trans-7,8-dihydroxy-7,8-dihydrobenzo(a)pyrene and their inhibition by butylated hydroxyanisole. *J. Nat. Cancer Inst.*, **62**, 1103–1106 (1979).
- WU-WILLIAMS, A.H., YU, M.C. and MACK, T.M., Life-style, workplace, and stomach cancer by subsite in young men of Los Angeles County. *Cancer Res.*, **50**, 2569–2576 (1990).
- ZIEGLER, R.G., MORRIS, L.E., BLOT, W.J., POTTERN, L.M., HOOVER, R. and FRAUMENI, J.F., JR., Esophageal cancer among black men in Washington, D.C. II. Role of nutrition. *J. Nat. Cancer Inst.*, **67**, 1199–1206 (1981).