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BRIEF COMMUNICATION

Effects of *Helicobacter pylori* Treatment on Gastric Cancer Incidence and Mortality in Subgroups

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Among 2258 *Helicobacter pylori*–seropositive subjects randomly assigned to receive one-time *H. pylori* treatment with amoxicillin-omeprazole or its placebo, we evaluated the 15-year effect of treatment on gastric cancer incidence and mortality in subgroups defined by age, baseline gastric histopathology, and post-treatment infection status. We used conditional logistic and Cox regressions for covariable adjustments in incidence and mortality analyses, respectively. Treatment was associated with a statistically significant decrease in gastric cancer incidence (odds ratio = 0.36; 95% confidence interval [CI] = 0.17 to 0.79) and mortality (hazard ratio = 0.26; 95% CI = 0.09 to 0.79) at ages 55 years and older and a statistically significant decrease in incidence among those with intestinal metaplasia or dysplasia at baseline (odds ratio = 0.56; 95% CI = 0.34 to 0.91). Treatment benefits for incidence and mortality among those with and without post-treatment infection were similar. Thus *H. pylori* treatment can benefit older members and those with advanced baseline histopathology, and benefits are present even with post-treatment infection, suggesting treatment can benefit an entire population, not just the young or those with mild histopathology.

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The Shandong Intervention Trial (ClinicalTrials.gov: NCT00339768) was initiated in Linqu, China, in 1995 to reduce precancerous gastric histopathology and gastric cancer (GC) incidence (1–4). Fifteen years after a 2-week course of treatment for *Helicobacter pylori* with amoxicillin and omeprazole, there was a statistically significant 39% reduction in GC incidence and a non-statistically significant 33% decrease in GC mortality compared with placebo (4). Here we examine treatment effects in subgroups. Some evidence suggests that *H. pylori* acts at early stages in the progression of precancerous lesions (5,6). Advanced lesions such as intestinal metaplasia (IM) or dysplasia (DYS) are more prevalent at older ages (6,7). Thus it is important to understand whether benefits of treatment extend to older members or those with advanced precancerous lesions, as suggested by a study of metachronous GCs (8). We also assess whether the effects of treatment are mediated entirely through post-treatment infection status because infection after treatment was common in our study (2) and others [eg, (9)], although not in all studies [eg, (10)].

Methods for this trial, including human subjects approval, are reported here and in the Supplementary Materials (available online) and have been reported in previous publications (1,3,4). The original randomized trial and the study extension were approved by the institutional review boards of the Beijing Institute for Cancer Research, the US National Cancer Institute, and Westat, and subjects provided written informed consent. The trial is registered at the National Cancer Institute PDQ database (trial number NCI-OH-95-C-N029; available at http://www.cancer.gov/clinicaltrials). Here we study the 2258 *H. pylori*–seropositive subjects who were randomly assigned to *H. pylori* treatment or its placebo. Infection status after treatment was ascertained by ^{13}C-urea breath tests (CUBT) in 1996 and 2003, except for the few patients with missing CUBT, for whom immunoglobulin G serology was used (Supplementary Figure 1, available online). In a sensitivity analysis, we determined infection only by CUBT. We use post-treatment infection to denote detectable *H. pylori* in 1996 (after retreatment, if any) or 2003, as defined in the Supplementary Material (available online). Such post-treatment infection could represent reinfection or initial failure to eradicate. Baseline histopathology was obtained from seven standard sites at gastroscopy. Associations with cumulative GC incidence were analyzed by conditional logistic regression, and associations with GC mortality were analyzed by Cox regression. All regression models included sex, ever using alcohol, ever smoking, garlic treatment, and vitamin treatment. Additionally the analysis by age subgroup was stratified on baseline histopathology, and the analysis by baseline histopathology subgroup was regression-adjusted for age (see Supplementary Materials, available online, for details) (4). The proportional hazards assumption within strata was tested by including an interaction term between time and treatment ($P > .05$ for all tests). All P values were two-sided, and significance level was defined as P less than .05. Statistical tests were performed using SAS version 9.2 (SAS Institute, Cary, NC). Further detailed methods can be found in the Supplementary Materials (available online).

Of the 2258 randomized subjects, 2172 had information on age, sex, baseline gastric histopathology, smoking, and alcohol intake. Overall, the treatment odds ratio (OR) for GC incidence was 0.61 (95% confidence interval [CI] = 0.38 to 0.96) and

the hazard ratio (HR) for GC mortality was 0.68 (95% CI = 0.36 to 1.29) (Table 1). Among those with IM or DYS at baseline, treatment was associated with a statistically significant reduction in GC incidence (OR = 0.56; 95% CI = 0.34 to 0.91), and a non-statistically significant reduction in GC mortality (HR = 0.63; 95% CI = 0.29 to 1.37) (Table 1). No risk reductions were seen among those with normal or mild gastric lesions, but heterogeneity tests were not statistically significant ($P_{incidence}$ = .34; $P_{mortality}$ = .33). Among those aged 55 years or older, *H. pylori* treatment was statistically significantly inversely associated with GC incidence (OR = 0.36; 95% CI = 0.17 to 0.79) and mortality (HR = 0.26; 95% CI = 0.09 to 0.79) (Table 1). Protective associations were weaker or absent for other age categories, but heterogeneity tests were not statistically significant for incidence (P = .21) and only marginally statistically significant for mortality (P = .048).

Treatment comparisons in strata defined by post-treatment infection status may be confounded (11). Nonetheless, we analyzed treatment effects within such strata to assess whether treatment has a beneficial effect independent of postinfection status. Furthermore, absence of a treatment effect among patients with post-treatment infection may imply a need for retreatment. We adjusted for potential confounders (see footnotes to Table 2; Supplementary Table 1, available online).

Among patients with any proven infection at or after 1996, the odds ratio for GC incidence was 0.66 (95% CI = 0.38 to 1.15), compared with an odds ratio of 0.44 (95% CI = 0.11 to 1.73) for those without proven infection (Table 2, stratified analysis 1). The preventive effect of treatment appears somewhat greater among those without post-treatment infection, but the difference was not statistically significant ($P_{heterogeneity}$ = .59). A more refined classification of post-treatment infection status yielded an odds ratio of 0.28 (95% CI = 0.05 to 1.57) for those uninfected in 1996 and 2003 (Table 2, stratified analysis 2), but again there was no statistically significant evidence of heterogeneity. Similar comments apply to GC mortality. Similar results were obtained with infection status after treatment determined only by CUBT (Supplementary Table 1, available online).

Gastric carcinogenesis is marked by progressive precancerous histopathologic changes with progressively increasing risk of gastric cancer incidence (6). *H. pylori* infection leads to superficial gastritis and chronic atrophic gastritis and is therefore thought to act early in carcinogenesis (5), a hypothesis also supported by stronger associations with GC among subjects infected many years earlier (12). One trial reported a reduction in GC incidence from *H. pylori* treatment only among patients without atrophy, IM, or DYS (13), but only six GCs occurred in this subgroup. Our finding that *H. pylori* treatment is at least as effective in those with IM or DYS as in those with less severe baseline histopathology suggests that *H. pylori* also promotes later stages of carcinogenesis. Another possibility is that amoxicillin or omeprazole retards later stages of carcinogenesis through mechanisms unrelated to *H. pylori* infection. A study of second GC after an initial GC diagnosis also indicates that *H. pylori* treatment retards GC incidence in patients with advanced precancerous lesions (8).

Although the strongest reductions in GC incidence and mortality occurred in those who tested negative for *H. pylori* both in 1996 and 2003, risk reductions were also seen in those with evidence of infection after treatment. Thus, the entire effect of treatment is not mediated by post-treatment infections status. It is possible that even short-term interruption in infection can have long-term beneficial effects. Short-term treatment of *H. pylori* resulted in a long-term statistically significant reduction in GC incidence and a

Table 1. Effects of *Helicobacter pylori* treatment on gastric cancer incidence and mortality stratified by gastric histopathology and by age at baseline*

Subgroup	Incidence of GC			Death due to GC		
	Placebo, No./No.†	Active, No./No.†	OR‡ (95% CI)	Placebo, No./No.†	Active, No./No. †	HR‡ (95% CI)
Overall	51/1086	33/1086	**0.61 (0.38 to 0.96)**	23/1086	16/1086	0.68 (0.36 to 1.29)
According to baseline gastric lesions						
Normal or SG or CAG	5/441	5/446	1.08 (0.31 to 3.81)	3/441	4/446	1.47 (0.33 to 6.65)
IM or DYS	46/645	28/640	**0.56 (0.34 to 0.91)**	20/645	12/640	0.63 (0.29 to 1.37)
$P_{heterogeneity}$§			.34			.33
According to baseline age						
<45 years	13/556	13/558	0.96 (0.43 to 2.11)	4/556	8/558	1.94 (0.58 to 6.47)
45–54 years	14/285	10/293	0.72 (0.31 to 1.67)	4/285	4/293	1.04 (0.26 to 4.16)
55–71 years	24/245	10/235	**0.36 (0.17 to 0.79)**	15/245	4/235	**0.26 (0.09 to 0.79)**
$P_{heterogeneity}$§			.21			**.048**

* CAG = chronic atrophic gastritis; CI = confidence interval; DYS = dysplasia; GC = gastric cancer; HR = hazard ratio; IM = intestinal metaplasia; OR = odds ratio; SG = superficial gastritis.

† Number of subjects with an event divided by total number of subjects.

‡ Associations with GC incidence were based on conditional logistic regression and those with GC mortality on Cox regression. Odds ratios and hazard ratios use the placebo group as referent. All models were regression-adjusted for sex, ever using alcohol, ever smoking, garlic treatment, and vitamin treatment. Additionally the analysis by age subgroup was stratified on baseline histopathology and the analysis by baseline histopathology subgroups was regression-adjusted for age (see Supplementary Materials, available online, for details). Number of subjects may not be equal to the number in the intervention trial because of missing values of baseline histology, smoking, and alcohol intake status. Two-sided tests were performed, and results in bold are statistically significant (P < .05).

§ For each stratified analysis, heterogeneity of odds ratio or hazard ratio among strata was evaluated using Q statistics for meta-analysis. These tests are two-sided for two strata and ominbus tests for more than two strata.

Table 2. Association of gastric cancer with amoxicillin/omeprazole treatment in strata defined by *Helicobacter pylori* infection status at or after 1996*

Strata	Incidence of gastric cancer			Deaths due to gastric cancer		
	Placebo, No./No.†	Treatment, No./No.†	OR (95% CI)	Placebo, No./No.†	Treatment, No./No.†	HR (95% CI)
Stratified analysis 1‡						
Any proven infection at or after 1996	48/1022	19/576	0.66 (0.38 to 1.15)	22/1022	7/576	0.56 (0.24 to 1.32)
No evidence of infection at or after 1996: *H. pylori* negative or missing in 1996 and negative or missing in 2003	3/64	14/510	0.44 (0.11 to 1.73)	1/64	9/510	0.88 (0.11 to 7.23)
$P_{heterogeneity}$§			0.59			0.54
Overall OR/HR combining two strata			0.62 (0.37 to 1.06)			0.61 (0.28 to 1.30)
Stratified analysis 2§						
H.pylori positive in 1996	45/923	10/273	0.68 (0.33 to 1.40)	20/923	4/273	0.74 (0.25 to 2.21)
H.pylori negative or missing in 1996, positive in 2003	3/99	9/303	0.95 (0.25 to 3.66)	2/99	3/303	0.43 (0.07 to 2.77)
H.pylori negative in 1996 and negative in 2003	3/52	9/442	0.28 (0.05 to 1.57)	1/52	4/442	0.46 (0.04 to 5.24)
$P_{heterogeneity}$§			0.69			0.86
Overall OR/HR combining three strata			0.64 (0.35 to 1.17)			0.60 (0.24 to 1.49)

* CI = confidence interval; HR = hazard ratio; OR = odds ratio.

† Number of subjects with an event divided by total number of subjects.

‡ *H. pylori* positive and negative infection determined by ^{13}C-urea breath test (CUBT), or, for those with missing CUBT, from immunoglobulin G serology. Details on how infection status was determined and on the analysis are given in the Supplementary Material (available online). The analyses within strata defined by infection status are based on conditional logistic regression (or Cox regression), stratified on baseline histopathology, and regression-adjusted for age, sex, ever using alcohol, ever smoking, garlic treatment, and vitamin treatment. Numbers of subjects are less than the 2258 randomized because of missing data for baseline histopathology, smoking, or alcohol intake. The overall odds ratio (hazard ratio) was calculated using conditional logistic regression (or Cox regression) stratified by post-treatment infection status, with regression adjustment for the factors above and for baseline histopathology in the categories above. Odds ratios and hazard ratios use the placebo group as referent.

§ For each stratified analysis, heterogeneity of odds ratio or hazard ratio among strata was evaluated using *Q* statistics for meta-analysis.

non-statistically significant reduction in GC mortality in our study, despite post-treatment infection in 586 of 1130 treated subjects. Another hypothesis that might account for beneficial treatment effects among those with post-treatment infection is that treatment initially eradicated *H. pylori* strains with higher carcinogenic activity than those present after treatment.

Among the strengths of this study were the randomized masked placebo–controlled design, excellent treatment compliance, and excellent follow-up (1,3,4). The main limitation is the small numbers of incident GC and deaths from GC, which limits power to detect treatment effects in subgroups and to demonstrate effect heterogeneity. Failure to find an effect of *H. pylori* treatment among younger subjects or those with mild gastric lesions may be because of small numbers of events. Treatment comparisons in strata defined by post-treatment infection status are not protected by the randomization and may be confounded, despite adjustments. An alternative framework for interpreting such data has been proposed but requires unverifiable assumptions (14). Stratification by newer staging systems, such as the corpus-predominant gastritis index (15), operative link on gastritis assessment (16), or operative link on intestinal metaplasia (17) systems, might provide additional insights, but our data were insufficient for those systems. Because treatment comparisons within strata based on age and baseline histology were protected by randomization, it is unlikely that using other staging systems would affect results in Table 1.

Short-term *H. pylori* treatment statistically significantly reduced GC incidence overall, in those with IM or DYS at baseline, and in those aged 55 years or older. Non-statistically significant reductions in GC incidence were found not only in those without post-treatment infection but also in those with proven post-treatment infection, although there was a non-statistically significant tendency for stronger treatment effects in those without post-treatment infection. These results suggest that prevention efforts can be applied widely and need not be restricted to the young or those with mild histopathology. However, more data are needed to define the full range of risks and benefits from antibiotic treatment of *H. pylori* in various regions, the best treatment regimens in various regions, and whether periodic retreatment is beneficial.

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Notes

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