ORIGINAL ARTICLE

Helicobacter pylori, Homologous-Recombination Genes, and Gastric Cancer

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ABSTRACT

BACKGROUND

Helicobacter pylori infection is a well-known risk factor for gastric cancer. However, the contribution of germline pathogenic variants in cancer-predisposing genes and their effect, when combined with *H. pylori* infection, on the risk of gastric cancer has not been widely evaluated.

METHODS

We evaluated the association between germline pathogenic variants in 27 cancer-predisposing genes and the risk of gastric cancer in a sample of 10,426 patients with gastric cancer and 38,153 controls from BioBank Japan. We also assessed the combined effect of pathogenic variants and *H. pylori* infection status on the risk of gastric cancer and calculated the cumulative risk in 1433 patients with gastric cancer and 5997 controls from the Hospital-based Epidemiologic Research Program at Aichi Cancer Center (HERPACC).

RESULTS

Germline pathogenic variants in nine genes (*APC*, *ATM*, *BRCA1*, *BRCA2*, *CDH1*, *MLH1*, *MSH2*, *MSH6*, and *PALB2*) were associated with the risk of gastric cancer. We found an interaction between *H. pylori* infection and pathogenic variants in homologous-recombination genes with respect to the risk of gastric cancer in the sample from HERPACC (relative excess risk due to the interaction, 16.01; 95% confidence interval [CI], 2.22 to 29.81; P=0.02). At 85 years of age, persons with *H. pylori* infection and a pathogenic variant had a higher cumulative risk of gastric cancer than noncarriers infected with *H. pylori* (45.5% [95% CI, 20.7 to 62.6] vs. 14.4% [95% CI, 12.2 to 16.6]).

CONCLUSIONS

H. pylori infection modified the risk of gastric cancer associated with germline pathogenic variants in homologous-recombination genes. (Funded by the Japan Agency for Medical Research and Development and others.)

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N Engl J Med 2023;388:1181-90.
DOI: 10.1056/NEJMoa2211807

Gastric cancer is the fifth most common neoplasm and the fourth leading cause of death from cancer worldwide.[1] *Helicobacter pylori* has been classified as a group I carcinogen and is an environmental risk factor for gastric cancer.[2] Although *H. pylori* infection affects more than half the world population,[3] the age-standardized global incidence rate of cancer attributable to *H. pylori* infection is highest in East Asia, at 17.6 cases per 100,000 person-years.[4] Tailored surveillance and eradication of *H. pylori* are recommended for controlling the incidence of gastric cancer.[2]

Germline pathogenic variants in cancer-predisposing genes are also essential factors in tailored surveillance and prevention of gastric cancer.[5,6] For example, *CDH1* is a risk gene for hereditary diffuse gastric cancer.[5,6] Families with hereditary diffuse gastric cancer meeting certain criteria are recommended for genetic testing, and carriers of pathogenic variants in *CDH1* are considered for prophylactic gastrectomy.[6] We recently found that germline pathogenic variants in *BRCA1* and *BRCA2* in the homologous-recombination[7] genes substantially increased the risk of gastric cancer.[8] It has also been suggested that other cancer-predisposing genes, such as mismatch-repair genes,[7] are associated with the risk of gastric cancer.[5] However, with the exception of *CDH1*, the identification of variants in these genes are not included in screening recommendations.[9]

A randomized, controlled trial showed that eradication of *H. pylori* reduced the incidence of gastric cancer, with effects seen even in persons with a first-degree family history of gastric cancer.[10] However, because a family history generally involves shared genetic and environmental factors,[11] it is unclear whether eradication was effective primarily in genetically susceptible persons or was effective regardless of genetic status. Because gene–environment interactions are associated with an excess risk of disease,[12] an evaluation of pathogenic-variant carrier status and *H. pylori* infection status may be informative with respect to risk stratification.

We therefore tested for an association between variants in cancer-predisposing genes and the risk of gastric cancer and then evaluated the combined effect of pathogenic variants and *H. pylori* infection status on risk.

Methods

Study Population

We included persons from two independent study cohorts in the analysis: BioBank Japan (BBJ) and the Hospital-based Epidemiologic Research Program at Aichi Cancer Center (HERPACC). Our study design is summarized in Figure S1 in the Supplementary Appendix, available with the full text of this article at NEJM.org. The study sample from BBJ included 10,426 patients with gastric cancer and 38,153 controls with nonmalignant diseases. The study sample from HERPACC included 1433 patients with newly diagnosed gastric cancer and 5997 controls without cancer, all of whom had an *H. pylori* infection status indicated. We defined a negative result as negative tests for both anti–*H. pylori* IgG antibodies and atrophic gastritis (as determined with the pepsinogen test according to the "ABC method,"[13] a noninvasive population surveillance tool[14]). We defined a positive result as one in which either test was positive. We used both tests because anti–*H. pylori* IgG serum antibody is known to spontaneously disappear with atrophic gastritis, which occurs with progression of *H. pylori* infection in some persons.[15]

All participants provided written informed consent. The study was approved by the ethics committees of the Institute of Medical Sciences, the University of Tokyo, the RIKEN Center for Integrative Medical Sciences, and Aichi Cancer Center. Details are provided in the Supplementary Methods section of the Supplementary Appendix. The authors vouch for the accuracy and completeness of the data in this report.

Sequencing and Bioinformatics Analysis

We analyzed 27 cancer-predisposing genes, including *CDH1*, homologous-recombination genes (*ATM*, *BARD1*, *BRCA1*, *BRCA2*, *BRIP1*, *CHEK2*, *PALB2*, *RAD51C*, and *RAD51D*),[7] mismatch-repair genes (*MLH1*, *MSH2*, *MSH6*, and *PMS2*),[7] and other genes (*APC*, *BMPR1A*, *CDK4*, *CDKN2A*, *EPCAM*, *HOXB13*, *MUTYH*, *NBN*, *NF1*, *PTEN*, *SMAD4*, *STK11*, and *TP53*), as described previously.[16] In the BBJ cohort, we used the raw data from sequencing of *BRCA1* and *BRCA2* in patients with gastric cancer and unaffected persons[8] and data from sequencing of the other 25 genes in 23,678 unaffected persons[16] from our previous studies. We performed se-

quencing for participants who had not undergone sequencing in the previous studies. With respect to the sequencing data, we implemented quality-control measures that were based on sequencing read depth, call rates, and deviation from Hardy–Weinberg equilibrium. We annotated variants that had a minor-allele frequency of less than 0.1% in the relevant control group. We defined pathogenic variants as those that resulted in loss of function according to SnpEff, version 4.3t,[17] or that were designated as pathogenic or likely pathogenic by ClinVar, version 2021-06-19.[18] Details are provided in Supplementary Methods.

STATISTICAL ANALYSIS

First, we evaluated the association between cancer-predisposing genes and the risk of gastric cancer, using a logistic-regression model with adjustment for age at entry and sex as recorded in BBJ. On the basis of a Bonferroni-corrected threshold of significance, genes with a P value of less than 1.85×10^{-3} (i.e., $0.05\div27$) were defined as gastric cancer risk genes. Second, we evaluated the clinical characteristics of the variant carriers. We also conducted a survival analysis involving patients with newly diagnosed gastric cancer.

Third, we estimated the odds ratios and corresponding 95% confidence intervals to evaluate the combined effect of germline pathogenic variants in the gastric cancer risk genes and *H. pylori* infection status on the risk of gastric cancer, using a logistic-regression model with adjustment for age at entry and sex as recorded in HERPACC. Interactions between pathogenic-variant carrier status and *H. pylori* infection status with respect to the effect on the risk of gastric cancer were estimated as an additive interaction, measured as the relative excess risk due to interaction,[19] and as a multiplicative interaction, measured with the use of interaction terms. For the evaluation of interactions, we used dichotomous categories for pathogenic-variant carrier status (carrier or noncarrier) and *H. pylori* infection status (positive or negative). No values were missing in these two data sets. Finally, we calculated the cumulative risk[20,21] of gastric cancer by using the odds ratio of the combined pathogenic-variant carrier status and *H. pylori* infection status, the prevalence of each status subgroup in our study control group, the age-specific cancer incidence rate in Japan,[22] and the age distribution of the population in Japan.[22]

All statistical tests were two-sided, and a P value of less than 0.05 was considered to indicate statistical significance. A Bonferroni correction was applied where appropriate. All statistical analyses were performed with the use of Stata software, version 16.0 (StataCorp), and R software, version 3.5.2 (R Foundation for Statistical Computing). Details are provided in Supplementary Methods.

RESULTS

CHARACTERISTICS OF STUDY PARTICIPANTS

The characteristics of the study participants are shown in Table 1 and Table S1. The median age at entry into the registry was 69 years among the BBJ patients with gastric cancer, 62 years among the HERPACC patients with gastric cancer, 64 years among the BBJ controls, and 55 years among the HERPACC controls. The percentage of male participants was 74.5% among the BBJ patients with gastric cancer, 74.6% among the HERPACC patients with gastric cancer, 53.1% among the BBJ controls, and 51.1% among the HERPACC controls. The percentage of patients with intestinal cancer was 61.2% among the BBJ patients and 49.3% among the HERPACC patients; this difference may be due to variations in the characteristics of the recruiting hospitals.

ANNOTATION OF GERMLINE VARIANTS

The mean read depths were 1003× among BBJ participants and 800× among HERPACC participants. After quality-control measures were applied, the genetic data from 10,366 BBJ patients with gastric cancer, 37,592 BBJ controls, 1423 HERPACC patients with gastric cancer, and 5990 HERPACC controls were included in the analysis. The read depth was greater than 20× in more than 99.8% of the target region for all participants. We annotated 459 of the 5676 germline variants in BBJ and 104 of the 1572 germline variants in HERPACC as pathogenic (Table S2). In total, we identified 518 pathogenic variants; the ClinVar accession numbers are provided in Table S2.

IDENTIFICATION OF GASTRIC CANCER RISK GENES

The associations between variants in cancer-predisposing genes and the risk of gastric cancer from the BBJ data are shown in Table 2. In ad-

Table 1. Characteristics of the Study Population.*

Characteristic	BBJ		HERPACC	
	Patients with Gastric Cancer (N=10,426)	Controls (N=38,153)	Patients with Gastric Cancer (N=1433)	Controls (N=5997)
Median age at entry (IQR) — yr	69 (62–75)	64 (54–72)	62 (54–69)	55 (45–64)
Sex — no. (%)				
Male	7770 (74.5)	20,242 (53.1)	1069 (74.6)	3063 (51.1)
Female	2656 (25.5)	17,911 (46.9)	364 (25.4)	2934 (48.9)
Gastric cancer subtype — no. (%)				
Intestinal				
Papillary adenocarcinoma	175 (1.7)	—	19 (1.3)	—
Tubular adenocarcinoma	6204 (59.5)	—	688 (48.0)	—
Diffuse				
Poorly differentiated adenocarcinoma	1848 (17.7)	—	550 (38.4)	—
Signet-ring-cell adenocarcinoma	1016 (9.7)	—	125 (8.7)	—
Mucinous adenocarcinoma	148 (1.4)	—	9 (0.6)	—
Gastric cancer not otherwise specified	1035 (9.9)	—	42 (2.9)	—
Helicobacter pylori infection status (%)†				
Positive				
Antibody positive and atrophic gastritis negative	—	—	454 (31.7)	1365 (22.8)
Antibody positive and atrophic gastritis positive	—	—	654 (45.6)	1226 (20.4)
Antibody negative and atrophic gastritis positive	—	—	128 (8.9)	177 (3.0)
Negative				
Antibody negative and atrophic gastritis negative	—	—	197 (13.7)	3229 (53.8)

* Percentages may not total 100 because of rounding. BBJ denotes BioBank Japan, HERPACC Hospital-based Epidemiologic Research Program at Aichi Cancer Center, and IQR interquartile range.

† *H. pylori* infection status was defined as negative for persons who were negative both for anti–*H. pylori* IgG antibody and for atrophic gastritis as defined by pepsinogen testing. Persons were regarded as positive if they had positive results in either of these two tests.

dition to *BRCA1* and *BRCA2*,[8] seven genes that, when variant, confer a predisposition to cancer showed significant associations with the risk of gastric cancer ($P<1.85\times10^{-3}$ for all nine). The same results were observed in sensitivity analysis restricted to persons 60 years of age or older and stratified according to sex (Fig. S2). The locations of the amino acids encoded by the pathogenic variants in these nine genes among the BBJ patients are shown in Figure S3.

CARRIERS OF PATHOGENIC VARIANTS IN GASTRIC CANCER RISK GENES

We investigated the clinical and demographic characteristics of pathogenic-variant carriers in the nine gastric cancer risk genes among the BBJ patients (Fig. S4). The ages of the carriers at diagnosis are shown in Figure 1. The proportion of carriers with a pathogenic variant decreased with advancing age at diagnosis (Fig. 1A). As shown in Figure S5, carriers of variants in *BRCA1*, *BRCA2* and *ATM* accounted for more than half the variant carriers in any age group, *CDH1* variants were present in 28.6% of carriers younger than 40 years of age, and *APC* and *MLH1* variants were each detected in 16.7% of carriers 40 to 49 years of age. The median age at diagnosis among patients who carried a pathogenic variant in one of these genes was more than 10 years younger than that among noncarriers (18.0 years for *APC*, 20.5 years for *CDH1*, and 11.5 years for *MLH1*) (Fig. 1B).

Table 2. Associations between Cancer-Predisposing Genes and Risk of Gastric Cancer in BBJ.*

Gene	Patients with Gastric Cancer (N = 10,366)	Controls (N = 37,592)	Odds Ratio (95% CI)	P Value†
	no. of carriers of pathogenic variants (%)			
BRCA2	104 (1.00)	79 (0.21)	5.08 (3.72–6.94)	9.83×10^{-23}
ATM	76 (0.73)	56 (0.15)	5.50 (3.82–7.90)	6.91×10^{-19}
BRCA1	32 (0.31)	27 (0.07)	4.81 (2.80–8.27)	1.37×10^{-8}
MLH1	13 (0.13)	4 (0.01)	14.45 (4.49–46.45)	7.39×10^{-6}
PALB2	16 (0.15)	20 (0.05)	3.71 (1.85–7.43)	2.19×10^{-4}
APC	6 (0.06)	3 (0.01)	14.17 (3.28–61.25)	3.85×10^{-4}
MSH6	19 (0.18)	16 (0.04)	3.48 (1.75–6.94)	3.97×10^{-4}
MSH2	11 (0.11)	8 (0.02)	4.85 (1.85–12.73)	1.33×10^{-3}
CDH1	6 (0.06)	5 (0.01)	8.04 (2.23–28.93)	1.42×10^{-3}
CHEK2	15 (0.14)	35 (0.09)	1.57 (0.83–2.96)	0.17
BARD1	8 (0.08)	19 (0.05)	1.71 (0.72–4.04)	0.23
BRIP1	9 (0.09)	50 (0.13)	0.66 (0.32–1.37)	0.27
RAD51C	2 (0.02)	14 (0.04)	0.44 (0.10–2.05)	0.30
TP53	7 (0.07)	13 (0.03)	1.53 (0.59–3.98)	0.38
NBN	9 (0.09)	50 (0.13)	0.73 (0.35–1.52)	0.40
PMS2	6 (0.06)	17 (0.05)	1.50 (0.56–4.02)	0.42
EPCAM	1 (0.01)	8 (0.02)	0.54 (0.06–4.78)	0.58
NF1	3 (0.03)	17 (0.05)	0.72 (0.20–2.59)	0.62
PTEN	1 (0.01)	8 (0.02)	0.61 (0.07–4.96)	0.64
HOXB13	2 (0.02)	9 (0.02)	0.74 (0.15–3.54)	0.70
RAD51D	35 (0.34)	122 (0.32)	1.06 (0.72–1.56)	0.78
BMPR1A	0	0	NA	NA
CDK4	0	1 (<0.01)	NA	NA
CDKN2A	0	3 (0.01)	NA	NA
MUTYH	0	0	NA	NA
SMAD4	0	3 (0.01)	NA	NA
STK11	0	2 (0.01)	NA	NA

* Results are for genetic data from BBJ after the application of quality-control measures. Carriers of pathogenic variants were defined in the recessive model for *MUTYH* and in the dominant model for the other genes. The associations between pathogenic variants in cancer-predisposing genes and the risk of gastric cancer were evaluated with the use of a logistic-regression model with adjustment for sex and age at entry. NA denotes not available.

† On the basis of a Bonferroni-corrected threshold of significance, genes with a P value of less than 1.85×10^{-3} (i.e., $0.05\div27$) were defined as gastric cancer risk genes.

With regard to other characteristics of carriers of pathogenic variants, a family history of related cancer types (breast, colorectal, and gastric cancer) was more common among carriers of pathogenic variants than among noncarriers (Table S3). Those with a pathogenic variant in *APC* or *CDH1* had an increased risk of intestinal and diffuse types of cancer, respectively, a finding consistent with recent guidelines[9] (Fig. S6). Among the 1558 patients in BBJ who were included in the survival analysis, the median follow-up time was 7.5 years (interquartile range, 2.2 to 10.4). Overall survival did not differ significantly between variant carriers and noncarriers (P = 0.58 by log-rank analysis; hazard ratio from multivariable analysis, 1.27; 95% confidence interval [CI], 0.82 to 1.97; P = 0.28) (Fig. S7).

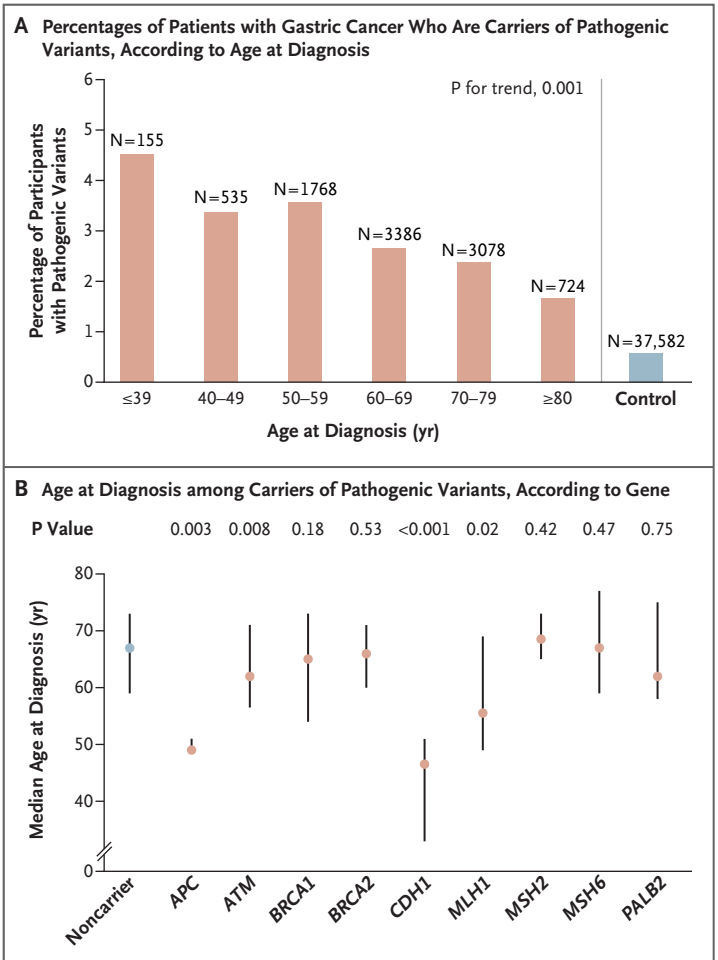

Figure 1. Age at Diagnosis of Gastric Cancer among Carriers of Germline Pathogenic Variants in Gastric Cancer Risk Genes in BioBank Japan.

Panel A shows percentages of patients with germline pathogenic variants in gastric cancer risk genes according to age at diagnosis; the percentage of controls with germline pathogenic variants is shown for comparison. The trend among patients with gastric cancer was evaluated with the use of the Cochran–Armitage test. Panel B shows the median age at diagnosis among carriers of pathogenic variants in gastric cancer risk genes according to gene; the median age at diagnosis among noncarriers of pathogenic variants in gastric cancer risk genes is shown for comparison. Error bars indicate the interquartile range. Differences were evaluated with the use of the Mann–Whitney U test. Information on age at diagnosis was unavailable for 7.0% of the patients with gastric cancer (720 of 10,366). The gastric cancer risk genes identified in this study were *APC*, *ATM*, *BRCA1*, *BRCA2*, *CDH1*, *MLH1*, *MSH2*, *MSH6*, and *PALB2*.

COMBINED EFFECT OF GENETIC VARIATION AND *H. PYLORI* INFECTION

In HERPACC, we identified 74 different pathogenic variants in the nine gastric cancer risk genes in 36 patients with gastric cancer (2.5%; 36 of 1423) and 72 controls (1.2%; 72 of 5990). The prevalence of pathogenic-variant carriers among patients with gastric cancer was similar in HERPACC and BBJ (HERPACC, 2.5%; BBJ, 2.7%), and 88.9% of carriers and 86.4% of noncarriers had evidence of *H. pylori* infection. *H. pylori* infection was associated with gastric cancer risk regardless of the cancer subtype ($P<0.001$ for both intestinal type and diffuse type).

We then assessed the risk of gastric cancer in persons with or without pathogenic variants and with or without *H. pylori* infection. As shown in Table 3, pathogenic variants and *H. pylori* infection showed a significant additive interaction with respect to the risk of gastric cancer ($P=0.02$). (The test for multiplicative interaction did not yield a significant result [$P=0.09$].) The additive interaction was robust in a bootstrap analysis with 10,000 resamplings and in a sensitivity analysis that excluded participants who were negative for anti–*H. pylori* IgG antibody and positive for atrophic gastritis (Tables S4 and S5). Participants with both a pathogenic variant and *H. pylori* infection had excess disease risk as compared with those who had either factor alone.

Four of the nine gastric cancer risk genes encode proteins that mediate homologous recombination: *ATM*, *BRCA1*, *BRCA2*, and *PALB2*. Because gastric carcinogenesis is related to genome instability caused by *H. pylori* infection,[23-26] we performed the same test of association but restricted it to variants in the four homologous-recombination genes and again observed a significant association (Table 3). The effect that the interaction between germline pathogenic variants in homologous-recombination genes and *H. pylori* infection had on gastric cancer risk was much larger than the effects of interactions between these variants and other environmental factors (smoking, drinking, obesity, and sodium intake); moreover, the interaction was independent of the other environmental factors (Figs. S8 and S9). In contrast, we did not observe an interaction between variants in mismatch-repair genes and *H. pylori* infection with respect to risk (Table 3). We could not evaluate interactions with variants in *APC* or *CDH1* owing to the numbers of carriers (no patients for *APC* and one patient for *CDH1*). The cumulative risk of gastric cancer through 85 years of age in each group is shown in Figure 2. The cumulative risk of gastric cancer in persons 85 years of age without infection was less than 5%, regardless of carrier status. In contrast, in

persons with *H. pylori* infection, the cumulative risk was higher among carriers of pathogenic variants in homologous-recombination genes than among noncarriers (45.5% [95% CI, 20.7 to 62.6] vs. 14.4% [95% CI, 12.2 to 16.6]).

DISCUSSION

We evaluated the prevalence of germline pathogenic variants in cancer-predisposing genes and their interaction with *H. pylori* infection with respect to the risk of gastric cancer. In addition to *BRCA1* and *BRCA2*,[8] seven genes (*APC*, *ATM*, *CDH1*, *MLH1*, *MSH2*, *MSH6*, and *PALB2*) had pathogenic variants associated with the risk of gastric cancer, and variant carriers had different clinical and demographic characteristics. Furthermore, pathogenic variants and *H. pylori* infection interacted to markedly increase the risk of gastric cancer in persons with both risk factors.

CDH1 is a well-known hereditary gastric cancer risk gene.[5,6,27] We observed that *CDH1* was associated with a risk of gastric cancer, particularly the diffuse type, and the age of patients at diagnosis among pathogenic-variant carriers was younger than that among noncarriers. In addition to variants in *CDH1*, variants in eight other genes were more common among patients with gastric cancer than among controls. It is notable that the prevalence of variants in the eight genes was equal to or greater than that observed for *CDH1*. Furthermore, variant carriers differed in age at diagnosis according to gene. These results indicate that, in addition to variants in *CDH1*, variants in at least eight other genes are associated with hereditary gastric cancer, with per-gene differences in disease risk, carrier frequency, and age at presentation. Gastric cancer in some patients may be detected through surveillance of carriers of pathogenic variants in these genes, and these patients may benefit from specific treatment, such as with a poly ADP-ribose polymerase inhibitor, as is the case with other neoplasms with homologous-recombination deficiency.[28]

Persons with both a germline pathogenic variant in a homologous-recombination gene and *H. pylori* infection had an excess disease risk as compared with persons who had either factor alone. We observed a significant additive interaction, although the multiplicative interaction was not significant. These results indicate

Table 3. Combined Effect of Germline Pathogenic Variants in Gastric Cancer Risk Genes and *H. pylori* Infection Status on the Risk of Gastric Cancer in HERPACC.*

Gene Category and *H. pylori* Status	Noncarriers†		Carriers		Relative Excess Risk Due to Interaction		Odds Ratio	
	No. of Patients/No. of Controls	Odds Ratio (95% CI)	No. of Patients/No. of Controls	Odds Ratio (95% CI)	Estimate (95% CI)	P Value for Additive Interaction	Estimate (95% CI)	P Value for Multiplicative Interaction
Overall gastric cancer risk genes‡								
H. pylori–negative	189/3173	1.00 (reference)	4/51	1.27 (0.45 to 3.59)	14.22 (2.50 to 25.93)	0.02	2.76 (0.84 to 9.04)	0.09
H. pylori–positive	1198/2745	5.76 (4.88 to 6.80)	32/21	20.25 (11.28 to 36.37)				
Homologous-recombination genes§								
H. pylori–negative	189/3173	1.00 (reference)	4/39	1.68 (0.59 to 4.83)	16.01 (2.22 to 29.81)	0.02	2.32 (0.69 to 7.81)	0.18
H. pylori–positive	1198/2745	5.76 (4.88 to 6.80)	30/18	22.45 (12.09 to 41.70)				
Mismatch-repair genes¶								
H. pylori–negative	189/3173	1.00 (reference)	0/7	NA	–0.67 (–10.55 to 9.22)	0.90	NA	NA
H. pylori–positive	1198/2745	5.76 (4.88 to 6.80)	1/2	4.09 (0.36 to 45.99)				

* Results are for genetic data from HERPACC after the application of quality-control measures. The effect of interactions between germline pathogenic-variant carrier status and *H. pylori* infection status on the risk of gastric cancer was evaluated with the use of a logistic-regression model with adjustment for sex and age at entry in HERPACC. The evaluation of interactions included the combination of the dichotomous status of carrier status (carrier or noncarrier) and *H. pylori* infection status (positive or negative).
† Noncarriers were defined as persons without pathogenic variants in gastric cancer risk genes.
‡ The gastric cancer risk genes were *APC*, *ATM*, *BRCA1*, *BRCA2*, *CDH1*, *MLH1*, *MSH2*, *MSH6*, and *PALB2*.
§ The homologous-recombination genes were *ATM*, *BRCA1*, *BRCA2*, and *PALB2*.
¶ The mismatch-repair genes were *MLH1*, *MSH2*, and *MSH6*.

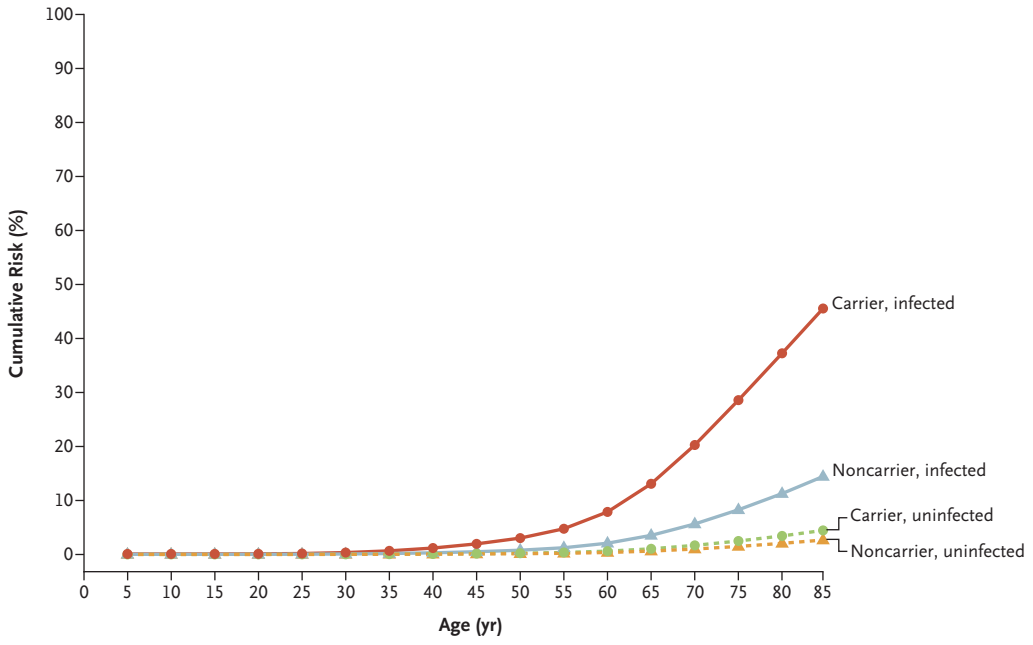

Figure 2. Cumulative Risk of Gastric Cancer through 85 Years of Age According to Germline Pathogenic-Variant Carrier Status and *Helicobacter pylori* Infection Status.

Cumulative risks of gastric cancer were estimated for carriers and noncarriers of germline pathogenic variants and persons who were positive and negative for *H. pylori* infection in the Hospital-based Epidemiologic Research Program at Aichi Cancer Center. Noncarriers were defined as persons without pathogenic variants in gastric cancer risk genes (*APC*, *ATM*, *BRCA1*, *BRCA2*, *CDH1*, *MLH1*, *MSH2*, *MSH6*, and *PALB2*). Only carriers of pathogenic variants in homologous-recombination (HR) genes (*ATM*, *BRCA1*, *BRCA2*, and *PALB2*) are shown in this figure because of the limited number of participants with variants in non-HR genes (*APC*, *CDH1*, *MLH1*, *MSH2*, and *MSH6*).

that *H. pylori* infection modifies the risk of gastric cancer when germline pathogenic variants are present in homologous-recombination genes.

A possible mechanism underlying this excess risk is genome instability caused by *H. pylori* infection that contributes to gastric carcinogenesis.[23-26] *H. pylori* CagA is a multifunctional effector protein.[29] Most *H. pylori* strains isolated in East Asia (up to 96.3%) are CagA-positive.[30] Imai et al. showed that *H. pylori* CagA, both Western and East Asian types, elicits features similar to those seen in *BRCA*-mutated cells: DNA double-strand breaks and a disabling of error-free DNA repair mediated by homologous recombination, which contribute to gastric carcinogenesis.[23] We hypothesize that the gastric carcinogenesis–related DNA damage due to *H. pylori* infection is enhanced in persons with a reduced DNA damage-repair capacity due to damaging variants in the homologous-recombination genes. The cumulative risk of gastric cancer at 85 years of age among carriers of a pathogenic variant who did not have *H. pylori* infection was not high (<5.0%), a finding consistent with the absolute risk reported for carriers of damaging *BRCA1* or *BRCA2* variants in countries with lower rates of infection with *H. pylori* of the East Asian type.[31] These results suggest that the eradication of *H. pylori* infection in persons with a pathogenic variant would mitigate the risk of gastric cancer.

This study has several limitations. First, it is retrospective. Randomized, controlled trials would be optimal but challenging to perform because of ethical issues stemming from the efficacy of *H. pylori* eradication in preventing gastric cancer.[2] Second, we analyzed only single-nucleotide variants and small insertion–deletions because we could not reliably detect copy-number variants from our amplicon-target sequencing data or from the available single-nucleotide polymorphism array data (data not shown), al-

though copy-number variants may be associated with cancer-predisposing syndromes.[32] Therefore, we may have underestimated the prevalence of germline pathogenic variants. However, the effect of such underestimation would apply to both patients and controls. Third, although the patients with gastric cancer in this study are representative of the broader population of patients with gastric cancer encountered by clinicians in practice in East Asia, there are biologic differences between the East Asian and Western types of *H. pylori* (particularly with respect to the CagA protein[33]) that may limit the extent to which the results of this study are relevant to other populations. Further evaluations of the Western type of *H. pylori* are warranted.

We identified nine gastric cancer risk genes in a large-scale evaluation of patients with different clinical and demographic characteristics. *H. pylori* infection modified the risk of gastric cancer associated with germline pathogenic variants in homologous-recombination genes. Our results suggest that in persons known to carry a pathogenic variant in a homologous-recombination gene, evaluation and eradication of *H. pylori* infection may be particularly important.

Supported by grants (JP15ck0106177, JP18km0605001, JP19kk0305010, and JP21ck0106553) from the Japan Agency for Medical Research and Development; Grants-in-Aid for Scientific Research (17015018, 221S0001, JP16H06277 [CoBiA], JP18H03045, and JP22H04923) from the Ministry of Education, Science, Sports, Culture, and Technology of Japan; a Grant-in-Aid for the Third Term Comprehensive 10-year Strategy for Cancer Control from the Ministry of Health, Labor, and Welfare of Japan; and a grant (ID177524) from the Australian National Health and Medical Research Council.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

We thank all the participants in the study, the Laboratory for Genotyping Development in RIKEN, the RIKEN-IMS Genome Platform, the BioBank Japan project, the Hospital-based Epidemiologic Research Program at Aichi Cancer Center (HERPACC) study, and the Aichi Cancer Center Joint Research Project on Priority Areas and Cancer BioBank Aichi.

APPENDIX

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