Effects of clarithromycin on the metabolism of omeprazole in relation to *CYP2C19* genotype status in humans

Background and purpose: A triple therapy with omeprazole, amoxicillin (INN, amoxicilline), and clarithromycin is widely used for the eradication of *Helicobacter pylori*. Omeprazole and clarithromycin are metabolized by CYP2C19 and CYP3A4. This study aimed to elucidate whether clarithromycin affects the metabolism of omeprazole.

Methods: After administration of placebo or 400 mg clarithromycin twice a day for 3 days, 20 mg omeprazole and placebo or 400 mg clarithromycin were administered to 21 healthy volunteers. Plasma concentrations of omeprazole and clarithromycin and their metabolites were determined before and 1, 2, 3, 5, 7, 10, and 24 hours after dosing. CYP2C19 genotype status was determined by a polymerase chain reaction–restriction fragment length polymorphism (PCR-RFLP) method.

Results: Subjects were classified into three groups on the basis of PCR-RFLP analyses for CYP2C19: homozygous extensive metabolizer group (n = 6), heterozygous extensive metabolizer group (n = 11), and poor metabolizer group (n = 4). Mean area under the plasma concentration–time curves from 0 to 24 hours (AUC) of omeprazole in the homozygous extensive metabolizer, heterozygous extensive metabolizer, and poor metabolizer groups were significantly increased by clarithromycin from 383.9 to 813.1, from 1001.9 to 2110.4, and from 5589.7 to 13098.6 ng · h/mL, respectively. There were significant differences in the mean AUC values of clarithromycin among the three groups.

Conclusion: Clarithromycin inhibits the metabolism of omeprazole. Drug interaction between clarithromycin and omeprazole may underlie high eradication rates achieved by triple therapy with omeprazole, amoxicillin, and clarithromycin. (Clin Pharmacol Ther 1999;66:265-74.)

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Ever since the discovery of *Helicobacter pylori,* close relationships between *H pylori* and upper gastrointestinal disorders have been made clear.[1-3] Cure of *H pylori* infection is extremely efficacious in treatment for these diseases.[4,5] Current treatment strategies for the cure of *H pylori* infection have included a triple therapy with a proton pump inhibitor, amoxicillin (INN, amoxicilline), and/or clarithromycin or metronidazole.[6-8] Before use of this proton pump inhibitor–based triple therapy, a dual therapy, such as treatment with omeprazole and amoxicillin, was widely tried. However, the eradication rate achieved by this dual therapy was not necessarily sufficient,[9] although bacterial resistance of *H pylori* to amoxicillin had never been reported. Hence, one more drug such as clarithromycin or metronidazole has been added to this dual therapy. However, development of bacterial resistance to these two drugs has been reported.[10-12]

Omeprazole produces a profound suppression of gastric acid secretion by inhibition of the H^+/K^+-ATPase (proton pump) activity in gastric parietal cells[13,14] and is used as a potent antiulcer agent.[15,16] Omeprazole is mainly metabolized to 5-hydroxyomeprazole by a genetically determined enzyme, *S*-mephenytoin 4′-hydroxylase (CYP2C19), in the liver.[17-21] This pharmacogenetic entity has shown a marked interethnic difference in the incidence of poor or deficient metabolizers: the poor metabolizer frequency is much greater (18% to 23%) in Japanese persons than that (3% to 5%) in American or European white populations.[17-23] At least two different mutation events associated with the poor metabolizer genotypes of CYP2C19 were determined by de Morais et al.[24,25] They concluded that *CYP2C19_{m1}* in exon 5 (m1) and *CYP2C19_{m2}* in exon 4 (m2) account for 100% of the available Japanese poor metabolizers and that comparable detection of m1 and m2 predicts the phenotypes of CYP2C19.[24,25] In individuals with a poor metabolizer phenotype or genotype of CYP2C19, the area under the plasma concentration–time curve (AUC) of omeprazole is markedly increased,[17,20,21] and the effect of omeprazole on acid secretion is enhanced in them.[26] We recently reported that the eradication rate of *H pylori* achieved by a dual omeprazole-amoxicillin therapy in patients with peptic ulcer disease who have a poor metabolizer genotype of CYP2C19 was significantly higher than that in those with an extensive metabolizer genotype of CYP2C19.[27] Higher plasma omeprazole levels in the poor metabolizer patients were assumed to account for the higher eradication rate achieved in them.[27]

On the other hand, omeprazole is also metabolized to omeprazole sulfone by CYP3A4.[28] Clarithromycin is also metabolized by CYP3A4 and is known as a potent inhibitor of CYP3A4.[29] Moreover, it has been suggested that clarithromycin inhibits the CYP2C19 activity to some extent.[29] Therefore a drug interaction is assumed to occur when omeprazole and clarithromycin are coadministered, thereby resulting in an increase in plasma omeprazole levels, which is expected to be related to the higher eradication rate of *H pylori* achieved by a triple omeprazole-amoxicillin-clarithromycin therapy than that by a dual omeprazole-amoxicillin therapy. On the basis of the assumptive backgrounds as described above, we intended to examine whether clarithromycin would really affect the metabolism of omeprazole and to what extent this possible interaction could occur in relation to the CYP2C19 genotype status.

METHODS

Subjects. Twenty-one healthy volunteers (mean age ± SE, 29.6 ± 1.0; 16 men and five women) were enrolled in the study. None of them consumed extensive amounts of alcohol or had a smoking habit. None had taken any drug for at least 1 week before and during the study. Written informed consent had been obtained from each of the subjects before participation in the study. The protocol was approved in advance by the Human Institutional Review Board of Hamamatsu University School of Medicine, Hamamatsu, Japan.

Study protocol. This study was conducted according to a double-blind, randomized, crossover two-period design. After administration of 400 mg clarithromycin or placebo twice a day for 3 days, each subject received a single oral dose of 20 mg omeprazole plus 400 mg clarithromycin or placebo at 9 AM. Two standard meals (12 AM and 6 PM), prepared at the hospital, were provided for each of the subjects. Mineral water was allowed ad libitum, but other beverages were not permitted. After more than a 1-week washout interval following the end of the one-period study, subjects underwent the other period study. Venous blood samples for determining plasma concentrations of omeprazole, 5-hydroxyomeprazole, omeprazole sulfone, clarithromycin, and 14-*(R)*-hydroxyclarithromycin were obtained before and 1, 2, 3, 5, 7, 10, and 24 hours after the dose. The samples were centrifuged at 3000 rpm immediately after the collection and stored at –80°C until analyzed.

CYP2C19 genotyping. Deoxyribonucleic acid (DNA) was extracted from each patient's leukocytes with a commercially available kit (Genomix, Talent, Trieste, Italy). Extracted genomic DNA was dissolved in distilled water. Genotyping procedures for identifying *CYP2C19* wild-type (wt) gene and the two mutated alleles, *CYP2C19_{m1}* (m1) and *CYP2C19_{m2}* (m2), were performed by a polymerase chain reaction–restriction fragment length polymorphism (PCR-RFLP) method with use of allele-specific primers, as described by de Morais et al,[24,25] with minor modifications as described by Kubota et al[30]: for exon 4, the forward primer (5′-AACATCAGGATTGTAAGCAC-3′) anneals in exon 4, 33 base pairs (bp) upstream from the exon 4/intron 4 junction, and the reverse primer (5′-TCAGGGCTTG-GTCAATATAG-3′) anneals in intron 4, 85 bp downstream from the exon 4/intron 4 junction.

For detecting *CYP2C19_{m1}* in exon 5 and *CYP2C19_{m2}* in exon 4, genomic DNA (200 ng) was amplified in the PCR buffer containing, in a final volume of 50 μL, 10 mmol/L Tris–hydrochloric acid [pH 8.3], 50 μmol/L potassium chloride, 0.01% gelatin, deoxyribonucleotide triphosphate (dNTP) mix (dATP, dCPT, dGPT, and dTTP; 200 μmol/L each as final concentration, Takara Shuzo Co, Ltd, Shiga, Japan), 0.2 μmol/L con-

centrations of PCR primers, 1.25 units of AmpliTaq DNA polymerase (Hoffmann–La Roche Ltd, Basel, Switzerland), and 1.5 mmol/L magnesium chloride. Amplification was made with an automatic thermal cycler (DNA Thermal Cycler PJ2000, Perkin Elmer, Norwalk, Conn) for 40 cycles that consisted of denaturation at 94°C for 1 minute, annealing at 57°C for 1 minute, and extension at 72°C for 2 minutes. An initial denaturation step at 94°C for 5 minutes and a final extension step at 72°C for 5 minutes were also performed. Restriction enzyme cleavage was conducted at 37°C for 1 hour after the addition of 25 units of *Msp*I for $CYP2C19_{m1}$ and 25 units of *Bam*HI for $CYP2C19_{m2}$. The digested PCR products were analyzed on 3% agarose gels and stained with ethidium bromide.

Assays of omeprazole and its two metabolites in plasma. The plasma concentrations of omeprazole and its two primary metabolites, 5-hydroxyomeprazole and omeprazole sulfone, were determined by HPLC as described by Kobayashi et al.[31] In brief, the assay consisted of the following procedures: after addition of 100 µL phenacetin (10 µg/mL in methanol) as an internal standard, 0.25 g sodium chloride, and 500 µL of 0.5 mol/L phosphate buffer (pH 8.0) to 0.5 mL of the samples, extraction was conducted by shaking with 5 mL chloroform. After centrifuging, the organic layer was transferred to a glass tube and evaporated at 40°C. The residue was reconstituted with 300 µL of the mobile phase as described below and passed through a 0.5-µm filter (Nihon Millipore Ltd, Tokyo, Japan). One hundred microliters of the filtrate was injected into the HPLC apparatus. The HPLC system consisted of a model L-7100 pump, a model L-7400 ultraviolet absorbance detector set at a wavelength of 302 nm, a model L-7200 autosampler, a model D-7500 integrator (Hitachi Ltd, Tokyo, Japan) and a 4.6 mm × 25 cm Capcell Pak C_{18} UG 120 column (Shiseido Co Ltd, Tokyo, Japan). The mobile phase consisted of 0.05 mol/L phosphate buffer (pH 8.5) and acetonitrile (75:25, vol/vol), and was delivered at a flow rate of 0.8 mL/min. The detection limits were 3 ng/mL for omeprazole and its two metabolites.

Assays of clarithromycin and 14-(R)-hydroxyclarithromycin in plasma. Plasma concentrations of clarithromycin and 14-*(R)*-hydroxyclarithromycin were also determined by HPLC with electrochemical detection. A constant aliquot (0.5 mL) of plasma was combined with 125 µL internal standard solution (erythromycin B; 2 µg/mL in acetonitrile) and 10 µL saturated sodium carbonate in a 10-mL test tube and vortexed vigorously for 10 seconds. Samples were extracted with 2.5 mL ethylacetate by shaking for 10 minutes. After centrifugation

for 5 minutes at 3000 rpm (4°C), the supernatant was transferred to another test tube, followed by addition of 2.5 mL of 1.0 mmol/L hydrochloric acid. Reconstituted samples were back-extracted and centrifuged as above. The upper organic layer was aspirated to waste. After the addition of 60 µL saturated sodium carbonate, the aqueous layer was extracted with 2.5 mL ethylacetate. After centrifugation, ethylacetate layer was put into a test tube and the solvent was evaporated at 40°C. The residue was dissolved in 0.25 mL of the mobile phase as described below, and a 20-µL aliquot was injected onto the HPLC.

The HPLC electrochemical detection system consisted of a model 880-PU pump (Nihonbunko, Co, Tokyo, Japan), a model SIL-6B automatic injector (Shimadzu Co, Kyoto, Japan), an electrochemical detector (Coulochem 5100A, ESA, Bedford, Mass), and a model C-R4A integrator (Shimadzu Co). Chromatographic separation occurred on a 4 mm internal diameter × 250 mm Nucleosil 100-5 C_{18} column (M Nagel Co, Duren, Germany) with use of a mobile phase composed of acetonitrile, 0.05 mol/L phosphate buffer (pH 6.5), and methanol (5:3:2, vol/vol/vol) at a flow rate of 0.8 mL/min. The column effluent was monitored by use of the electrochemical detector with the electric potentials of the screening and working electrodes set at +0.65 V and +0.90 V, respectively.

Quantification was performed with use of the standards of clarithromycin and 14-*(R)*-hydroxyclarithromycin spiked into blank plasma over the range from 0.05 to 10 µg/mL. In each case, the peak height ratio of the compound of interest to the internal standard versus the spiked concentration was linear (correlation coefficient >0.999) over the concentration ranges observed in the study. The lowest limit of quantification was 0.05 µg/mL with a 0.5-mL plasma sample. Quality control samples that contained clarithromycin and 14-*(R)*-hydroxyclarithromycin at concentrations of 0.05, 0.5, and 10 µg/mL, respectively, were included in each assay to monitor the performance of the quantitative analysis.

Data analysis. The area under the plasma concentration–time curves (AUC) from 0 to 24 hours of omeprazole, omeprazole sulfone, 5-hydroxyomeprazole, clarithromycin, and 14-*(R)*-hydroxyclarithromycin were calculated with the linear trapezoidal method. The paired *t* test was used to determine whether AUC values for omeprazole, omeprazole sulfone, and 5-hydroxyomeprazole were affected by clarithromycin administration. Statistically significant differences in AUC values for omeprazole, omeprazole sulfone, 5-hydroxyomeprazole, clarithromycin, and 14-*(R)*-hydroxyclarithromycin
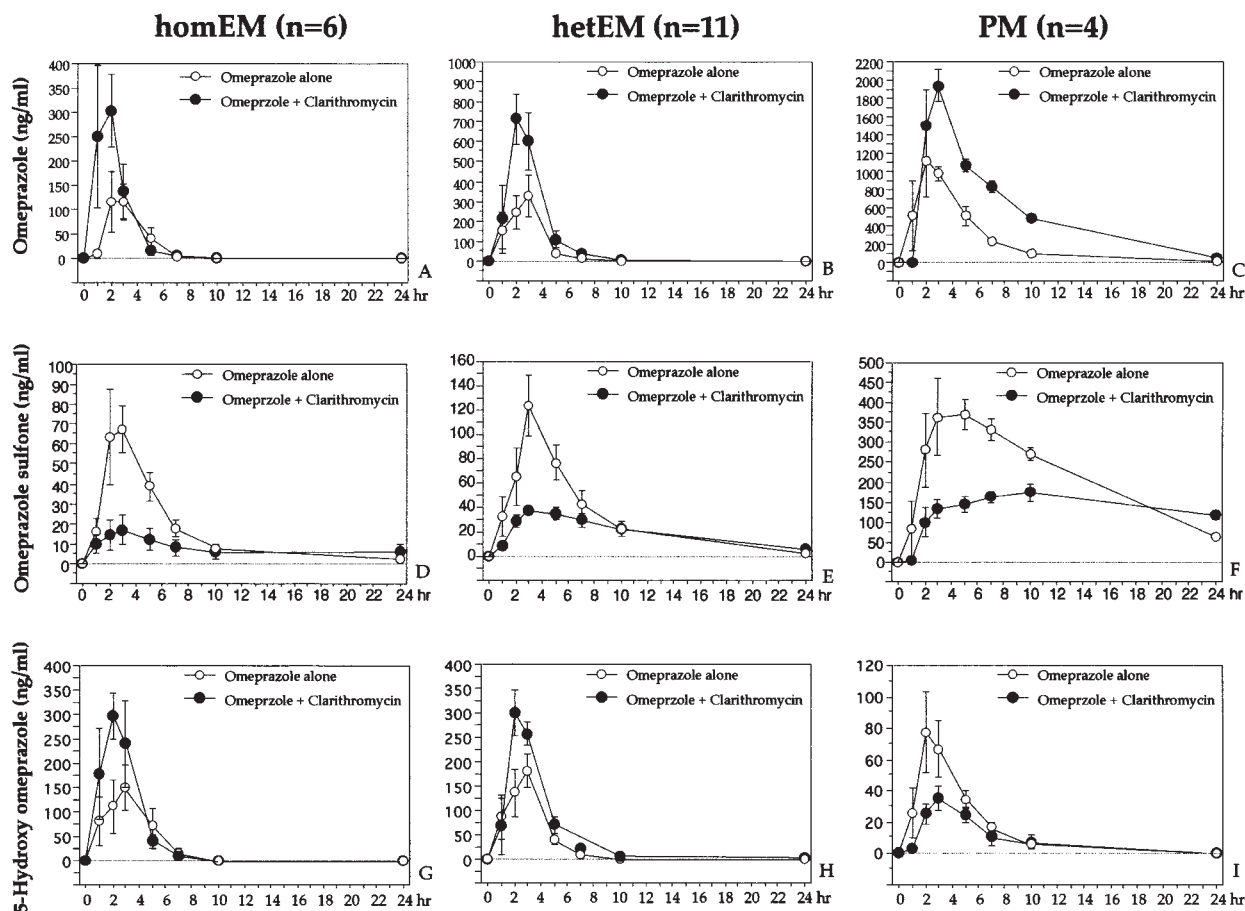
Fig 1. Mean ± SE plasma levels of omeprazole, omeprazole sulfone, and 5-hydroxyomeprazole in the homozygous extensive metabolizer (homEM), heterozygous extensive metabolizer (hetEM), and poor metabolizer (PM) groups with and without clarithromycin. Plasma concentrations of omeprazole were significantly increased by clarithromycin in the three different CYP2C19 genotype groups **(upper panel).** Plasma concentrations of omeprazole sulfone were significantly decreased by clarithromycin in the three different CYP2C19 genotype groups **(middle panel).** Plasma concentrations of 5-hydroxyomeprazole were significantly increased in the homozygous extensive metabolizer and heterozygous extensive metabolizer group, whereas those in the poor metabolizer group were decreased by clarithromycin, but not significantly **(lower panel).**

Table I. Demographic characteristics of subjects enrolled in the study as a function of CYP2C19 status

	Homozygous extensive metabolizers (n = 6)	*Heterozygous extensive metabolizers (n = 11)*	*Poor metabolizers (n = 4)*	*P Values*
CYP2C19 genotype	wt/wt	wt/m1 or wt/m2	m1/m2	—
Age (y)	27.8 ± 2.1	31.1 ± 2.4	28.3 ± 2.6	0.5939
Body weight (kg)	63.5 ± 2.7	62.9 ± 2.9	53.8 ± 4.6	0.1851
Male/female	5/1	9/2	2/2	0.3919

wt, Wild-type; m1, *CYP2C19* mutation in exon 5; m2, *CYP2C19* mutation in exon 4; wt/wt, homozygous for the wt alleles in both exon 5 and exon 4 (homozygous extensive metabolizer); wt/m1, heterozygous for the *CYP2C19$_{m1}$* mutation without the *CYP2C19$_{m2}$* mutation (heterozygous extensive metabolizer); wt/m2, heterozygous for the *CYP2C19$_{m2}$* mutation without *CYP2C19$_{m1}$* mutation (heterozygous extensive metabolizer); m1/m2, heterozygous for both the *CYP2C19$_{m1}$* mutation and the *CYP2C19$_{m2}$* mutation (poor metabolizer).
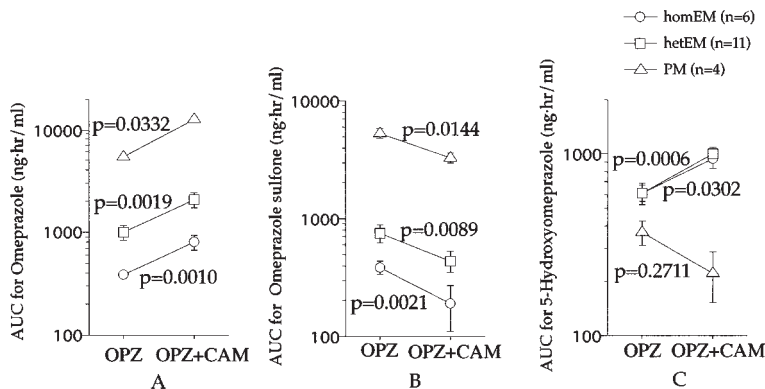
Fig 2. Effects of clarithromycin on mean area under the plasma concentration–time curves from 0 to 24 hours (AUC) for omeprazole **(A),** omeprazole sulfone **(B),** and 5-hydroxyomeprazole **(C)** as a function of CYP2C19 genotype status. AUC values for omeprazole were significantly increased by clarithromycin in the three different genotype groups, whereas AUC values for omeprazole sulfone were significantly decreased by clarithromycin in the three different genotype groups. AUC values for 5-hydroxyomeprazole were significantly increased by clarithromycin in the homozygous extensive metabolizer (homEM) and heterozygous extensive metabolizer (hetEM) group, whereas that in the poor metabolizer (PM) group was decreased by clarithromycin (but not significantly).

among the three different *CYP2C19* genotype groups were determined by one-way ANOVA with Scheffe's multiple-comparison test. All numerical data are given as mean values ± SE. Values of $P < .05$ were taken to indicate statistical significance.

RESULTS

No clinically undesirable signs or symptoms attributable to the study drugs administered were recognized at any time during the study period. All subjects completed the study following the protocol.

Four different allelic band patterns were observed in the 21 volunteers enrolled in this study. Of the 21 subjects, six were homozygous for the wt alleles in both exon 5 and exon 4 (wt/wt), seven were heterozygous for the $CYP2C19_{m1}$ mutation without the $CYP2C19_{m2}$ mutation (wt/m1), four were heterozygous for the $CYP2C19_{m2}$ mutation without the $CYP2C19_{m1}$ mutation (wt/m2), and four were heterozygous for both the $CYP2C19_{m1}$ mutation and the $CYP2C19_{m2}$ mutation (m1/m2). The subjects were arbitrarily classified into the three genotype groups as follows: The homozygous extensive metabolizer group (nonmutation group, wt/wt, n = 6), heterozygous extensive metabolizer group (one-mutation group, wt/m1 or wt/m2, n = 11), and poor metabolizer group (two-mutation group, m1/m2, n = 4). There were no statistical differences in mean age, mean body weight, or male/female ratios among the three different genotype groups (Table I).

Mean (±SE) plasma concentration–time profiles for omeprazole with and without clarithromycin in the three different genotype groups are shown in the *upper panel* of Fig 1. Plasma omeprazole levels were significantly increased by clarithromycin in the three different groups. Mean plasma concentration–time profiles for omeprazole sulfone and 5-hydroxyomeprazole with and without clarithromycin in the three different genotype groups are shown in the *middle* and *lower panels* of Fig 1, respectively. Plasma omeprazole sulfone levels were significantly decreased by clarithromycin in the three different groups. Mean plasma 5-hydroxyomeprazole levels were significantly increased by clarithromycin in the homozygous extensive metabolizer and heterozygous extensive metabolizer groups, whereas those in the poor metabolizer group decreased, but not significantly.

The mean changes in the AUC values for omeprazole, 5-hydroxyomeprazole, and omeprazole sulfone by clarithromycin obtained from the three genotype groups are shown in Fig 2, *A, B,* and *C,* respectively. Clarithromycin significantly increased the AUC for omeprazole (Fig 2, *A*) and decreased the AUC for omeprazole sulfone (Fig 2, *B*) in the three genotype groups, whereas clarithromycin significantly increased the AUC for 5-hydroxyomeprazole in the homozygous extensive metabolizer and heterozygous extensive metabolizer groups and tended to decrease the AUC for 5-hydroxyomeprazole in the poor metabolizer group (Fig 2, *C*).

Table II. AUC values for omeprazole, omeprazole sulfone, and 5-hydroxyomeprazole with and without clarithromycin as a function of CYP2C19 status

CYP2C19	AUC for omeprzole (ng · h/mL)		AUC for omeprazole sulfone (ng · h/mL)		AUC for 5-hydroxyomeprazole (ng · h/mL)	
	Omeprazole + placebo	Omeprazole + clarithromycin	Omeprazole + placebo	Omeprazole + clarithromycin	Omeprazole + placebo	Omeprazole + clarithromycin
Homozygous extensive metabolizers (n = 6)	383.9 ± 26.3	813.1 ± 141.8	385.9 ± 47.9	188.6 ± 77.6	606.4 ± 81.1	946.0 ± 105.3
Heterozygous extensive metabolizers (n = 11)	1,001.9 ± 160.3	2,110.4 ± 351.9	756.6 ± 133.6	435.3 ± 90.6	613.03 ± 62.2	1,016.8 ± 76.0
Poor metabolizers (n = 4)	5,589.7 ± 146.8	13,098.6 ± 512.7	5,245.7 ± 487.8	3,304.2 ± 342.4	370.5 ± 59.6	220.5 ± 69.5
P Values	<.0001*	<.0001†	<.0001‡	<.0001§	.1075‖	<.0001¶

AUC, Area under the plasma concentration–time curve from 0 to 24 hours.

P Values of post hoc test by Scheffe's multiple comparison test are as follows: *P = .0298, P < .0001, and P < .0001; †P = .0566, P < .0001, and P < .0001; ‡P = .3936, P < .0001, and P < .0001; §P = .4422, P < .0001, and P < .0001; ‖P = .9975, P = .1929, and P = .1252; ¶P = .8446, P = .0007, and P < .0001 (homozygous extensive metabolizer versus heterozygous extensive metabolizer, homozygous extensive metabolizer versus poor metabolizer, and heterozygous extensive metabolizer versus poor metabolizer, respectively).

In addition, there were significant differences in the mean AUC values for omeprazole, omeprazole sulfone, and 5-hydroxyomeprazole among the three different genotype groups irrespective of clarithromycin dosing (Table II). The mean AUC for omeprazole when coadministered with clarithromycin in the poor metabolizer group was 34 times as high as that when coadministered with placebo in the homozygous extensive metabolizer group.

Mean plasma concentration–time profiles for clarithromycin and 14-*(R)*-hydroxyclarithromycin and AUC values for clarithromycin and 5-hydroxyomeprazole as a function of CYP2C19 genotype status are shown in Fig 3, *A, B, C,* and *D,* respectively. The mean plasma clarithromycin level in the poor metabolizer group was the highest, that in heterozygous extensive metabolizer subjects was the second highest, and that in homozygous extensive metabolizer group was the lowest of the three groups. The similar trend was observed in the mean plasma 14-*(R)*-hydroxyclarithromycin levels among the three genotype groups (Fig 3, *D*).

DISCUSSION

The main current therapeutic strategy for cure of *H pylori* infection consists of a variety of concomitant treatments with a proton pump inhibitor and one or two antibacterial agents, such as amoxicillin, clarithromycin, and metronidazole.[5-8] The role of proton pump inhibitor in these treatment strategies is to maintain the availability of antibiotics by raising the intragastric pH.[32] It has been reported that as the dose of proton pump inhibitor increases, eradication rates by the dual treatment with proton pump inhibitor and amoxicillin increase.[33] Bayerdorffer et al[34] reported

that an eradication rate higher than 90% is able to be achieved by the daily doses of 120 mg omeprazole and 2250 mg amoxicillin for 2 weeks without clarithromycin or metronidazole. However, the dose of 120 mg omeprazole is not often prescribed as an initial dosing in clinical practice. Therefore the recent regimens for cure of *H pylori* infection include a proton pump inhibitor and amoxicillin, and/or clarithromycin, and/or metronidazole.[5-8] However, metronidazole has been reported to exhibit a risk of the later development of lung cancer.[35] For this reason, clarithromycin is used more frequently than metronidazole in Japan.

Clarithromycin shows an antibacterial effect by inhibiting the bacterial protein synthesis.[36] Although *H pylori* often acquires bacterial resistance to clarithromycin[12] and the eradication rate by a dual omeprazole-clarithromycin therapy was sometimes insufficient,[37] eradication rates achieved by the triple omeprazole-amoxicillin-clarithromycin treatment[8] are sufficiently better than those achieved with the dual omeprazole-amoxicillin treatment,[9] suggesting that clarithromycin has an effect other than that as an antibiotic.

Clarithromycin is metabolized by CYP3A4 in the liver, which is a CYP isoform.[29] This enzyme is also associated with the metabolism of omeprazole—omeprazole and 5-hydroxyomeprazole are metabolized by CYP3A4 to omeprazole sulfone and 5-hydroxyomeprazole sulfone, respectively.[28] In addition, omeprazole is mainly metabolized by CYP2C19, which is also a CYP isoform in the liver, to 5-hydroxyomeprazole.[17-21] In this study, plasma concentrations of omeprazole, 5-hydroxyomeprazole, and omeprazole sulfone differed among the three different *CYP2C19* genotype groups.
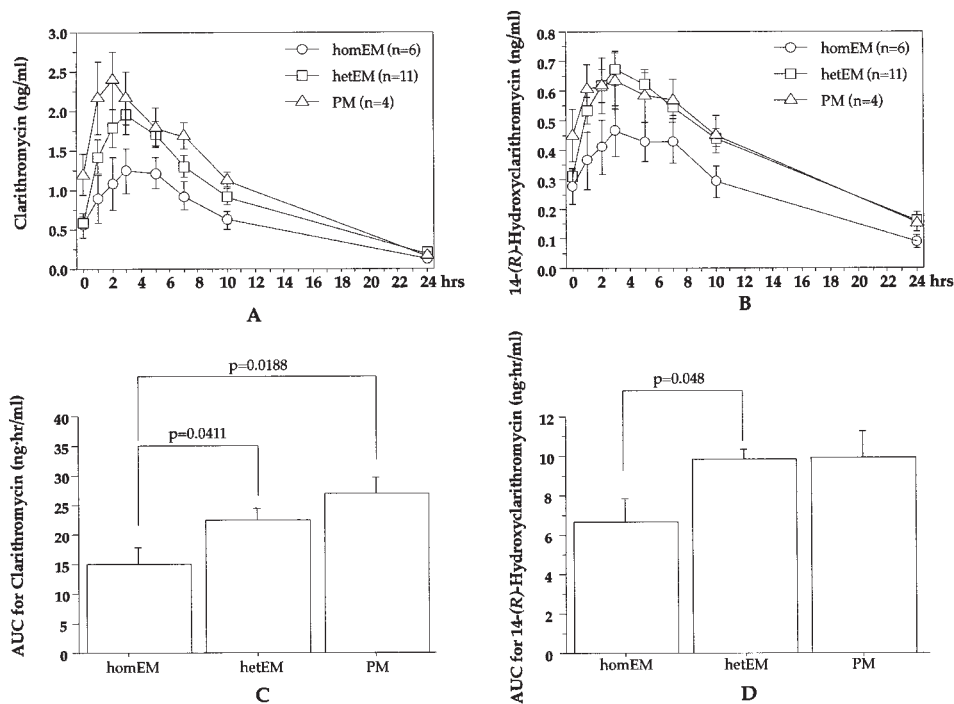
Fig 3. Mean ± SE plasma concentration–time curves of clarithromycin **(A)** and 14-*(R)*-hydroxyclarithromycin **(B)** and AUC values for clarithromycin **(C)** and 14-*(R)*-hydroxyclarithromycin **(D)** in the three different genotype groups. Significant differences in plasma clarithromycin levels were observed among the three different CYP2C19 genotype groups.

Omeprazole and 5-hydroxyomeprazole are, however, metabolized to omeprazole sulfone and 5-hydroxyomeprazole sulfone, respectively, by CYP3A4, which also metabolizes clarithromycin as noted above. Clarithromycin is also known as a potent inhibitor of CYP3A4.[29,38] Moreover, CYP2C19 is also related to the metabolism of clarithromycin.[29] Therefore the metabolism of omeprazole and 5-hydroxyomeprazole is assumed to be affected by clarithromycin. Our observation that plasma omeprazole sulfone levels were decreased by clarithromycin and that plasma omeprazole levels were increased by clarithromycin indicates that clarithromycin inhibited the sulfoxidation of omeprazole mediated by CYP3A4. Furthermore, plasma 5-hydroxyomeprazole levels were increased in the homozygous and heterozygous extensive metabolizer groups, suggesting that the sulfoxidation of 5-hydroxyomeprazole to 5-hydroxyomeprazole sulfone by CYP3A4 was also inhibited by clarithromycin in the homozygous and heterozygous extensive metabolizer subjects. Moreover, because of an inhibition of the CYP3A4 activity by clarithromycin, omeprazole has to be much more dominantly metabolized to 5-hydroxyomeprazole by CYP2C19, which would also contribute

to an increase in plasma 5-hydroxyomeprazole levels in the homozygous extensive metabolizer and heterozygous extensive metabolizer groups. However, the CYP2C19 activity is slightly inhibited by clarithromycin,[29] as noted above, which would also contribute to the increased levels of plasma omeprazole in the homozygous and heterozygous extensive metabolizer groups. In contrast, in the poor metabolizer group plasma 5-hydroxyomeprazole levels were decreased by clarithromycin. This observation is hardly explainable, because the poor metabolizer subjects lack CYP2C19 in the liver. In this respect, we are tempted to assume that the hydroxylation of omeprazole to 5-hydroxyomeprazole in the poor metabolizer subjects might be mediated, to certain extent, through CYP3A4.

Amoxicillin is unstable and its antibacterial activity is decreased under low pH conditions. Therefore an increase in the gastric juice pH to neutral levels with use of acid-inhibitory agents such as omeprazole is recommended for the cure of *H pylori* infection.[39] Thus an increase in the plasma concentration of omeprazole by coadministration with clarithromycin is assumed to make amoxicillin more stable in the stomach. Moreover, because omeprazole per se has an anti–*H pylori*

effect,[40] the increased plasma concentration of omeprazole by coadministration with clarithromycin is expected to enhance the bactericidal effect of omeprazole. In our recent report, in patients with peptic ulcer disease who have a poor metabolizer genotype of CYP2C19, the cure rate for *H pylori* infection by a dual omeprazole-amoxicillin therapy was perfect (100.0% [95% CI, 66.4% to 100.0%]; n = 9), whereas that for homozygous extensive metabolizer patients was less than 30% (28.6% [CI, 13.1% to 48.7%]; n = 28) and that for heterozygous extensive metabolizer patients (60.0% [CI, 38.6% to 83.0%]; n = 25) was intermediate between the former two groups.[27] As shown in this study, the AUC values for omeprazole after coadministration with clarithromycin were increased to the level about two times as high as that after administration of omeprazole alone in the homozygous extensive metabolizer group and in the heterozygous extensive metabolizer and poor metabolizer groups. This increase in plasma omeprazole levels by clarithromycin appears to underlie a higher eradication rate achieved by the triple omeprazole-amoxicillin-clarithromycin therapy than those by the dual omeprazole-amoxicillin therapy.

Plasma concentrations of clarithromycin and 14-*(R)*-hydroxyclarithromycin differed among the three different genotype groups in this study. The mean plasma omeprazole levels in the poor metabolizer group were the highest; therefore omeprazole would interfere more strongly with the metabolism of clarithromycin by CYP3A4 in the poor metabolizer group. Moreover, the possibility cannot totally be negated that CYP2C19 might be associated with the metabolism of clarithromycin to a certain extent[29] and omeprazole might have inhibited the metabolism of clarithromycin mediated through both CYP2C19 and CYP3A4 in the homozygous and heterozygous extensive metabolizer groups.

Therefore the coadministration of omeprazole with clarithromycin appears to increase the plasma concentrations of each other (eg, a mutual drug-drug interaction). Drug interaction between clarithromycin and omeprazole is assumed to contribute, on a theoretical basis, to a high eradication rate achieved by therapeutic regimens that include omeprazole and clarithromycin, such as a triple omeprazole-clarithromycin-amoxicillin therapy.

Finally, the results of this study need to be interpreted with the context of its limitations. This study was performed in healthy volunteers with a single administration of omeprazole. Therefore it must be assessed whether the drug interaction we observed in the healthy individuals receiving therapy with oral omeprazole and clarithromycin occurs similarly in *H pylori*–positive patients receiving multiple-dose therapy with omeprazole and clarithromycin. Although we cannot offer any reasonable explanation for this critical question based on the results of this study, we assume that multiple dosings of omeprazole and clarithromycin would, on a theoretical basis, produce a more profound drug interaction than that observed after the single dosing. As shown in this study, the mean AUC of omeprazole in the poor metabolizer group after coadministration of omeprazole and clarithromycin was more than 30 times as high as that in the homozygous extensive metabolizer group after the administration of omeprazole alone. It is therefore clinically important to take differences in *CYP2C19* genotype patterns into account when omeprazole is administered, as well as to consider a drug interaction mediated through CYP3A4 and CYP2C19 when omeprazole and clarithromycin are coadministered. These considerations should also be taken into account in clinical practice when a patient receives other substrate drugs associated with CYP2C19 and CYP3A4 concurrently because a variety of drugs are metabolized by these two CYP isoforms.[41]

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