Amoxicillin/clavulanic acid-warfarin drug interaction: a randomized controlled trial

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

- Increased INR was previously observed in patients treated with warfarin and amoxicillin/clavulanic acid (amoxiclav) combination. To date, no prospective study has yet evaluated the effect of amoxiclav on INR in patients on warfarin therapy and consequently, there are no clear-cut conclusions or clear recommendations for clinicians.

WHAT THIS PAPER ADDS

- We provided the first systematic prospective evaluation of the interaction between amoxiclav and warfarin and found that amoxiclav did not modify INR in patients treated with stable warfarin therapy in the absence of any infectious or inflammatory syndrome, suggesting that the previously observed INR increase in these patients may not be attributable to a drug–drug interaction.

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Keywords

amoxicillin/clavulanic acid, drug interaction, infection, inflammation, warfarin

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AIMS

To investigate whether an interaction exists between amoxicillin/clavulanic acid (amoxiclav) and warfarin in patients treated with stable oral anticoagulant therapy.

METHODS

In a double-blind, cross-over, placebo-controlled study, 12 patients on stable warfarin therapy, received a 7 day amoxiclav regimen or placebo.

RESULTS

The mean maximum increase in INR observed was 0.22 ± 0.3 with amoxiclav vs. 0.24 ± 0.6 with placebo ($P = 0.94$). The day 7–day 1 factor II, R(–) and S(–) warfarin plasma concentrations were similar during the amoxiclav and placebo study periods ($P = 0.81$, $P = 0.45$, $P = 0.75$, respectively).

CONCLUSION

Amoxiclav did not modify anticoagulation in patients treated with stable warfarin therapy and without infection.

Introduction

Various antibiotics, e.g. the N-methyl-thio-tetrazole (NMTT) group and beta-lactam antibiotics, have been reported to interact with warfarin [1, 2]. Amoxicillin/clavulanic acid (amoxiclav), a member of the penicillin class, also has been suggested to potentiate the effects of warfarin [3]. Several case reports and retrospective studies stated that amoxiclav increased INR and/or the risk of bleeding with an odds ratio (OR) of 7 in patients treated with warfarin [4–8]. Amoxicillin increased the risk of hospitalization for gastrointestinal bleeding with an OR ranging from 1.20 to 1.64 [4]. Amoxicillin or ampicillin was also associated with an increased risk of over-anticoagulation (OR 1.37, 95% CI 0.92, 2.05) [5]. Two case reports, one patient with microscopic haematuria and another with a rectus sheath haematoma, both with increased INR, were recently published in elderly patients receiving long-term stable warfarin therapy and treated with amoxiclav for otitis media and respiratory tract infection, respectively [7, 8].

The potential amoxiclav-warfarin drug interaction may be explained by either interference with the CYP2C9-dependent liver metabolism of warfarin or alterations in normal gut flora resulting in reduced intestinal vitamin K synthesis, even if it becomes established that the vitamin K produced by the colon flora (essentially K2) does not contribute significantly to the total vitamin K absorbed [9]. Finally, the infection itself (inflammatory syndrome) may alter warfarin metabolism [10] or hepatic clotting factor synthesis [4, 11]. As no prospective study has yet evaluated the effect of amoxiclav on INR in patients on warfarin therapy, there is no clear-cut conclusion or clear recommendations for clinicians. We therefore designed an interaction study between amoxiclav and warfarin in patients without any infectious or inflammatory syndrome.

Methods

This prospective, randomized, double-blind, placebo-controlled, two-phase crossover study was conducted in ambulatory patients, aged 18 years and older, treated with warfarin for more than 1 month with a target INR between 2 and 3, no recent or ongoing infectious or inflammatory disease and who gave their written informed consent. Patients were enrolled as soon as three consecutive INR values (minimum interval of 5 days) were within the target range (between 2 and 3) on the same warfarin dosage. This study was approved by the Ethics Committee of Le Kremlin-Bicêtre Hospital and was registered to the ClinicalTrials.gov website (#NCT00603317).

During each 10 day period, patients received the same warfarin regimen at the same schedule and either a 7 day course of oral amoxiclav (500 mg amoxicillin, 62.5 mg clavulanic acid, Augmentin®, GlaxoSmithKline, Marly-le-Roi,

France) 1 g twice daily or a 7 day course of matching placebo in a randomly assigned order. At each visit, complete physical examination was performed and blood samples were collected at day 0, 3, 5, 6, 7, 10 for INR and day 0 and 7 for the factor II, R(–) and S(–) warfarin (see online supplementary material for details). Blood samples were taken 12 h after warfarin intake in all patients and during either study period. Subjects were asked about any adverse event (bleeding, diarrhoea) or missed treatment dose. The treatment was stopped if an INR value was higher than 3.5.

Factor II activity, R(–) and S(–) warfarin plasma concentrations and the genotypes of both *VKORC1* and *CYP2C9* [*CYP2C9**2 (rs1799853) *CYP2C9**3 (rs1057910) and *VKORC1* 1173C>T SNP (rs9934438)] assays were performed all together at the end of study.

Assuming that a significant interaction would lead to a mean maximum INR (INR_{max-D1}) increase of 1.1 and 0.2 unit during the amoxiclav and placebo period, respectively, with a 0.6 standard deviation [15, 16], 12 evaluable patients were required with a 1% type one error and a 90% statistical power. By anticipating that eight patients might prematurely drop out from the study regarding the primary endpoint, we planned to enrol 20 patients.

The order of drug administration was analysed in 12 evaluable subjects using a within-subject design ANOVA in order to verify whether an interaction existed between the order of administration and treatment with warfarin and amoxiclav combination. Either treatment was compared using the non-parametric Wilcoxon signed rank test regarding primary (INR_{max} and INR_{max-D1}) and secondary endpoints (factor II, R(–) and S(–) warfarin). The influence of diarrhoea on INR_{max} and INR_{max-D1} was analysed using the non-parametric Mann–Whitney *U*-test and the effect of *CYP2C9* (*CYP2C9**2 and *CYP2C9**3) and *VKORC1* variants on INR_{max} and INR_{max-D1} was analysed using ANOVA. Results were expressed as mean $\pm$ 95% confidence intervals. Statistical analyses were performed according to the ‘intent-to-treat’ principle using Statview v8.0 (SAS Institute, Cary, NC, USA).

Results

Between October 2008 and October 2009, 13 ambulatory patients were enrolled and 12 patients (six men, six women, mean age 41.3 years, range 22 to 68 years) completed the study. One patient was excluded by the investigator due to renal failure during the placebo period. Mean baseline warfarin dosage was similar between the two treatment periods (6.13 ± 1.67 mg day^{–1}). Baseline INR value, defined as the mean of the last three INR values before treatment or placebo intake, was similar between either study period (2.39 ± 0.24 mg day^{–1} vs. 2.37 ± 0.25 mg day^{–1}, *P* = 0.82).

Figure 1

Change in the mean INR over time with respect to the study period. Amoxicillin/clavulanic acid (■); Placebo (◆)

The mean maximum INR increase from baseline to day 10 [$INR_{(max-D1)}$] did not differ between the amoxiclav (0.22 ± 0.3) and the placebo period (0.24 ± 0.6 , $P = 0.94$). Likewise, the mean maximum INR (INR_{max}) was similar between each period (2.57 ± 0.4 vs. 2.67 ± 0.5 , $P = 0.64$). There was no treatment or order of period interaction for INR_{max} and $INR_{(max-D1)}$ ($P = 0.49$ and $P = 0.95$, respectively). Changes in the mean INR over time are displayed in Figure 1.

No patient was withdrawn from either study period because of an $INR > 3.5$. As shown in Table 1, no difference was observed in factor II, R(-) and S(-) warfarin concentrations between either treatment period and between day 1 and day 7. Among the 12 patients, four (33%) were heterozygous carriers for the *CYP2C9*3* allele and two (16.7%) for the *CYP2C9*2* allele. The allelic frequency of the *VKORC1* 1173C>T polymorphism was 50% (6/12) and 8.3% (1/12) for the CT and TT genotype, respectively. Neither *CYP2C9* nor *VKORC1* genetic variants had an effect on INR_{max} or $INR_{(max-D1)}$. The mean INR_{max} ranged between 2.18 ± 0.02 (*CYP2C9*1/*2*, amoxiclav period) and 2.73 ± 0.40 (*CYP2C9*1/*2*, placebo period, $P > 0.70$) and the mean $INR_{(max-D1)}$ ranged between -0.1 ± 0.06 (*CYP2C9*1/*2*) and 0.31 ± 0.31 (*VKORC1* CT or TT, $P > 0.63$.) Likewise the day 7–day 1 R(-) and S(-) warfarin concentrations did not differ with respect to the *CYP2C9* and *VKORC1* genotype and the study period (data not shown) [17].

Diarrhoea occurred during amoxiclav administration, lasting 1 day (twice or three times a day) in four and 7 days in two patients. The INR_{max} observed in these patients was 2.62 ± 0.25 vs. 2.59 ± 0.49 in those who did not experience diarrhoea ($P = 0.34$). No significant difference in $INR_{(max-D1)}$ between patients who had diarrhoea and those who did not was noted during amoxiclav period (0.29 ± 0.31 vs. 0.23 ± 0.22 , $P = 0.87$, respectively). No bleeding event was reported during the entire study.

Discussion

In this randomized, double-blind, placebo-controlled study, amoxiclav did not increase INR in stable patients treated with warfarin. The mechanism of increased INR and risk of bleeding observed in previous case reports and observational studies is unclear [4–8]. This interaction could theoretically be explained by two distinct mechanisms. Inhibition of *CYP2C9*-dependent warfarin metabolism, leading to higher S(-) warfarin plasma concentrations and therefore higher INR was ruled out in the current study. Alternatively, amoxiclav is known to alter the intestinal flora that produces vitamin K, and this decrease in vitamin K is believed to decrease synthesis of vitamin K-dependent clotting factors II, VI, IX, and X, consistent with a previously published study showing that broad-spectrum antibiotics (e.g. amoxiclav) alter the normal gut flora, leading to vitamin K deficiency [9]. We did not analyse the intestinal flora before and after amoxiclav or placebo treatment in the current study. More recently, it has been shown that the intestinal flora produce mainly vitamin K2 and this flora was localized in the colon from which the absorption is more than limited so major effects on this flora were not supposed to significantly alter the total vitamin K absorption. Hence, further studies are needed to clarify the role of any antibiotic-induced change in the intestinal flora in INR changes in patients treated with warfarin.

While previous observations of INR increase with amoxiclav were reported in patients taking amoxiclav for various infections, it is difficult to determine whether amoxiclav or the infection itself resulted in an increase in INR. Diarrhoea, the major side effect of amoxiclav [18], might have altered warfarin absorption but half of our patients experienced diarrhoea without any warfarin pharmacokinetic or pharmacodynamic alteration.

Alternatively, several studies have shown that *CYP* expression and activity may be decreased in humans during infection or inflammation, resulting in lowered drug clearance, increased toxicity and altered physiological function [10, 19]. In the absence of any infection or inflammation, we were unable to detect any effect of amoxiclav on INR, clotting factor II and R(-) and S(-) warfarin plasma concentrations. This suggests that increased INR and bleeding events observed in patients treated with amoxiclav and warfarin during infectious episodes may be consecutive to the inflammatory and/or infectious syndrome interfering with warfarin pharmacokinetics and/or pharmacodynamics [20, 21].

Our study has several limitations. First, a mean maximum INR increase of 1.1 was used in the power calculation during the amoxiclav period. This is high considering that warfarin users need to stay within 1 INR unit (i.e. 2.0–3.0 or 2.5–3.5). Therefore, our study with only 12 patients was only powered to measure large differences in INR and we may have missed smaller, less clinically relevant

Table 1Variation (Δ) in factor II, R(-) and S(-) warfarin plasma concentrations over time*

Parameters	Amoxiclav period (n = 12)	Placebo period (n = 12)	P value**
Factor II (%)			
Day 1	28 (24.3, 31.9)	28 (22.9, 32.6)	0.91
Day 7	27 (21.7, 32.3)	27 (22.3, 32.6)	0.88
Δ FI (D ₇ -D ₁)	-1 (-5.6, 3.4)	-0.3 (-5.3, 4.6)	0.81
R(-)-warfarin (ng ml⁻¹)			
Day 1	854 (563, 1146)	1014 (748, 1281)	0.39
Day 7	910 (712, 1107)	963 (689, 1237)	0.56
Δ R(-)-warfarin (D ₇ -D ₁)	55 (-145, 256)	-51 (-191, 88)	0.45
S(-)-warfarin (ng ml⁻¹)			
Day 1	701 (382, 1020)	714 (493, 935)	0.92
Day 7	702 (468, 936)	756 (582, 929)	0.44
Δ S(-)-warfarin (D ₇ -D ₁)	1 (-178, 181)	42 (-78, 161)	0.75

*Mean (95% CI); **Between treatments comparison.

differences in INR. Additionally, too many biases (e.g. food, drug, genetic factors) and confounding factors may result in the lack of a drug-drug interaction from the small sample size. A second limitation was the lack of measurement of vitamin K plasma concentrations, since one possible hypothesis was an effect of amoxiclav on the gut bacterial flora that might have affected vitamin K concentrations. Although factor II was measured as a surrogate of this effect, it may also depend on the status of vitamin K epoxide reductase. Finally, our results can be only applied to patients younger than 68 years.

In conclusion, amoxiclav did not modify INR in patients treated with stable warfarin therapy in the absence of any infection or inflammation, suggesting that the previously observed INR increase in these patients may not be attributable to a drug-drug interaction. Therefore, anticoagulation therapy in patients with infection should be managed independent of amoxiclav administration.

Competing Interests

There are no competing interests to declare.

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