The effects of laropiprant, a selective prostaglandin D₂ receptor 1 antagonist, on the antiplatelet activity of clopidogrel or aspirin.

Aimee Dallob  
*Merck, Whitehouse Station, NJ*

Wen-Lin Luo  
*Merck, Whitehouse Station, NJ*

Julie Mabalot Luk  
*Merck, Whitehouse Station, NJ*

Lisa Ratcliffe  
*Merck, Whitehouse Station, NJ*

Amy O Johnson-Levonas  
*Merck, Whitehouse Station, NJ*

Recommended Citation  
Dallob, Aimee; Luo, Wen-Lin; Luk, Julie Mabalot; Ratcliffe, Lisa; Johnson-Levonas, Amy O; Schwartz, Jules I; Dishy, Victor; Kraft, Walter K.; De Hoon, Jan N; Van Hecken, Anne; De Lepeleire, Inge; Radziszewski, Waldemar; Wagner, John A; and Lai, Eseng, "The effects of laropiprant, a selective prostaglandin D₂ receptor 1 antagonist, on the antiplatelet activity of clopidogrel or aspirin." (2011). *Department of Pharmacology and Experimental Therapeutics Faculty Papers*. Paper 14.  
https://jdc.jefferson.edu/petfp/14

This Article is brought to you for free and open access by the Jefferson Digital Commons. The Jefferson Digital Commons is a service of Thomas Jefferson University's Center for Teaching and Learning (CTL). The Commons is a showcase for Jefferson books and journals, peer-reviewed scholarly publications, unique historical collections from the University archives, and teaching tools. The Jefferson Digital Commons allows researchers and interested readers anywhere in the world to learn about and keep up to date with Jefferson scholarship. This article has been accepted for inclusion in Department of Pharmacology and Experimental Therapeutics Faculty Papers by an authorized administrator of the Jefferson Digital Commons. For more information, please contact: JeffersonDigitalCommons@jefferson.edu.
As submitted to: 

Platelets

And later published as:
The Effects of Laropiprant, a Selective Prostaglandin D\(_2\) Receptor 1 Antagonist, on the Antiplatelet Activity of Clopidogrel or Aspirin

Volume 22, Issue 7, November 2011, Pages 495-503

DOI: 10.3109/09537104.2011.565433

Aimee Dallob\(^1\), Wen-Lin Luo\(^1\), Julie Mabalot Luk\(^1\), Lisa Ratcliffe\(^1\), Amy O. Johnson-Levonas\(^1\), Jules I. Schwartz\(^1\), Victor Dishy\(^2\), Walter K. Kraft\(^3\), Jan N. de Hoon\(^4\), Anne Van Hecken\(^4\), Inge De Lepeleire\(^5\), Waldemar Radziszewski\(^1\), John A. Wagner\(^1\), Eseng Lai\(^1\)

\(^1\)Merck, Whitehouse Station, NJ, USA

\(^2\)Former Merck employee; currently: Daiichi-Sankyo Pharma Development, Edison, NJ, USA

\(^3\)Thomas Jefferson University, Philadelphia, PA, USA

\(^4\)Center for Clinical Pharmacology, University Hospitals Leuven, Campus Gasthuisberg, Leuven, Belgium

\(^5\)Merck, Brussels, Belgium

Reprints and Correspondence: Dr. Eseng Lai

Merck & Co., Inc.
Subheading: Administration of LRPT with clopidogrel or aspirin

Target Journal: Platelets

Pages: 23

Tables: 5

Figures: 4

Abstract word count: 383
Abstract

Laropiprant (LRPT) is being developed in combination with Merck's extended-release niacin (ERN) formulation for the treatment of dyslipidemia. LRPT, an antagonist of the prostaglandin PGD$_2$ receptor DP1, reduces flushing symptoms associated with ERN. LRPT also has affinity for the thromboxane A$_2$ receptor TP (approximately 190-fold less potent at TP compared with DP1). Aspirin and clopidogrel are two frequently used anti-clotting agents with different mechanisms of action. Since LRPT may potentially be co-administered with either one of these agents, these studies were conducted to assess the effects of steady-state LRPT on the antiplatelet activity of steady-state clopidogrel or aspirin. Bleeding time at 24 hr postdose (trough) was prespecified as the primary pharmacodynamic endpoint in both studies. Two, separate, double-blind, randomized, placebo-controlled, crossover studies evaluated the effects of multiple-dose LRPT on the pharmacodynamics of multiple-dose clopidogrel or aspirin. Healthy subjects were randomized to once-daily oral doses of LRPT 40 mg or placebo to LRTP co-administered with clopidogrel 75 mg or aspirin 81 mg for 7 days with at least a 21-day washout between treatments. In both studies, bleeding time and platelet aggregation were assessed 4 and 24 hours post-dose on Day 7. Comparability was declared if the 90% confidence interval for the estimated geometric mean ratio ([LRPT + clopidogrel]/clopidogrel alone or [LRPT + aspirin]/aspirin alone) for bleeding time at 24 hours post-dose on Day 7 was contained within (0.66, 1.50). Concomitant daily administration of LRPT 40 mg with clopidogrel 75 mg or aspirin 81 mg resulted in an approximate 4% to 5% increase in bleeding time at 24 hours after the last dose vs. bleeding time after treatment with clopidogrel or aspirin alone, demonstrating that the treatments had comparable effects on bleeding time. Percent inhibition of platelet aggregation was not significantly different between LRPT co-administered with clopidogrel or aspirin vs clopidogrel
or aspirin alone at 24 hours postdose at steady state. At 4 hours after the last dose, co-
administration of LRPT 40 mg resulted in 3% and 41% increase in bleeding time vs bleeding
time after treatment with aspirin or clopidogrel alone, respectively. Co-administration of LPRT
with clopidogrel or aspirin was generally well tolerated in healthy subjects. Co-administration of
multiple doses of LRPT 40 mg and clopidogrel 75 mg or aspirin 81 mg had no clinically
important effects on bleeding time or platelet aggregation.

**Key words:** Clopidogrel, aspirin, laropiprant, pharmacokinetics
**Introduction**

Niacin, at gram doses, has broad beneficial effects on the overall plasma lipoprotein profile (i.e., increases high-density lipoprotein cholesterol and lowers low-density lipoprotein cholesterol, triglycerides, and lipoprotein (a) levels) and improves cardiovascular outcomes when administered alone or in combination with other lipid-altering agents [1-4]. Niacin is underutilized in clinical practice for the treatment of dyslipidemia primarily due to the bothersome side effect of flushing, which limits niacin dose escalation and frequently leads to the discontinuation of therapy [5-8].

Niacin-induced flushing is mediated primarily by prostaglandin D\(_2\) (PGD\(_2\)), which activates PGD\(_2\) receptor-1 (DP1) in the skin [9-15]. Laropiprant (LRPT) is an orally active, potent, highly selective antagonist of DP1 that has been shown to be effective in suppressing niacin-induced flushing when co-administered with extended-release niacin (ERN) [16-19]. In patients with dyslipidemia, co-administration of LRPT with ERN reduced both the incidence and intensity of niacin-induced flushing symptoms without diminishing the lipid-altering benefits of niacin therapy [20,21]. The concomitant administration of LRPT and ERN has the potential to significantly improve the tolerability and optimize therapeutic dosing of niacin, thereby improving the clinical effectiveness of niacin therapy.

In vitro studies have demonstrated that PGD\(_2\) inhibits platelet aggregation and that activated platelets release PGD\(_2\). Thus, antagonism of DP1 by LRPT has been postulated to inhibit the activity of platelet-derived PGD\(_2\), thereby indirectly enhancing the reactivity of platelets. Activated platelets release thromboxane A\(_2\) (TxA\(_2\)), which, acting through its receptor TxA\(_2\)/prostaglandin H\(_2\) (TP), promotes platelet aggregation. LRPT also has antagonist affinity for the receptor TP, although it is 190-fold less potent at TP compared with DP1. In contrast to the
uncertain role of DP1 in regulating platelet function, the importance of TP and the thromboxane pathway in modulating platelet function has been clearly established [22]. Taken together, these data suggest that LRPT may have the potential to alter platelet function, either by enhancing platelet reactivity through DP1 antagonism or by inhibiting platelet aggregation through TP antagonism. The overall clinical significance of LRPT interactions with receptors DP1 and TP has been assessed with several different assays using both ex vivo and in vivo methods. A recent study demonstrated that LRPT administered alone or in combination with ERN does not enhance platelet reactivity as assessed by collagen-induced platelet aggregation and bleeding time in healthy subjects [23].

Aspirin and clopidogrel are two commonly used antiplatelet agents with different mechanisms of action [21,24]. Since LRPT may potentially be co-administered with either one of these agents, clinical studies were conducted to assess the effects of steady-state LRPT on the antiplatelet activity of steady-state clopidogrel or aspirin. This paper describes the results of two, separate, double-blind, randomized, placebo-controlled, crossover studies designed to evaluate the effects of multiple therapeutic doses of LRPT on the antiplatelet activity of multiple-dose aspirin or clopidogrel as measured by bleeding time and platelet aggregation in healthy subjects.

Methods

Patient Selection Criteria

Both studies enrolled healthy, non-smoking men and women judged to be in good general health based on medical history, physical examination, and laboratory testing. The first (Merck & Co., Inc., Phase I, Protocol No. 049) and second (Merck & Co., Inc., Phase I, Protocol No. 072) studies enrolled subjects 18 to 45 or 18 to 55 years, respectively. Subjects were excluded from
study participation if they had a history of major gastrointestinal abnormalities or disease, peptic ulceration, hematologic, immunologic, genitourinary, cardiovascular, hepatic, renal, pulmonary, psychiatric, or metabolic/endocrine disease. Subjects were not permitted to use aspirin, NSAIDS, or prescription or non-prescription medications or consume excessive amounts of alcohol or caffeinated beverages during the conduct of either study. Protocol 072 specified that subjects with clopidogrel resistance i.e. clopidogrel non-responders (<10% change from baseline platelet aggregation induced by ADP in Treatment B at 4 hours postdose on Day 7) would have been excluded from the primary efficacy analysis, but no subjects met this criterion.

Each subject signed a written informed consent before any study procedures or examinations were performed. The study was conducted in accordance with principles of Good Clinical Practice and was approved by the appropriate institutional review boards and regulatory agencies.

**Study Designs**

Two separate, double-blind, randomized, placebo-controlled, crossover studies were conducted to evaluate the effects of multiple dose LRPT on the antiplatelet effects of multiple-dose aspirin (Aspirin Study; Protocol No. 049) and clopidogrel (Clopidogrel Study; Protocol No. 072). Within each study, eligible subjects were randomized to a sequence of two treatments (Treatments A and B) and each subject received both treatments in crossover fashion, therefore acting as their own control. The order in which subjects received each treatment was randomly assigned by a computer-generated allocation schedule, and there was a minimum 21-day washout between each treatment period.
In the Aspirin Study, eligible subjects were randomized to receive aspirin 81 mg co-administered with LRPT 40 mg once daily for 7 days (Treatment A) and aspirin 81 mg co-administered with placebo once daily for 7 days (Treatment B) in a cross-over fashion. In this study, study medication was administered at approximately the same time each day. Doses on Days 1, 5, 6, and 7 were administered under supervision in the research unit with approximately 240 mL of water following an overnight fast. Doses on Days 2 to 4 were self administered by the subjects outside the clinical research unit. On Day 7 of both treatment periods, water intake was restricted 1 hour prior to and after the administration of study drug. On Day 7 of both treatment periods, a standard meal was provided after the completion of study procedures.

In the Clopidogrel Study, eligible subjects were randomized to receive clopidogrel 75 mg co-administered with LRPT 40 mg once-daily for 7 days (Treatment A) and clopidogrel 75 mg co-administered with placebo once daily for 7 days (Treatment B) in a cross-over fashion. In this study, all doses of study medication were administered with 240 mL of water immediately following the ingestion of a standard low-fat breakfast. On Day 7 of both treatment periods, subjects fasted for 4 hours after dosing. A standard meal was provided after the completion of study procedures on Days 7 and 8.

In both studies, LRPT 40 mg (3 × 5-mg and 1 × 25-mg tablets; Merck & Co., Inc, Rahway, NJ) and placebo (placebo for LRPT 3 × 5-mg and 1 × 25-mg tablets; Merck & Co., Inc, Rahway, NJ) were administered in a double-blind manner according to a randomized allocation schedule, while aspirin 81 mg (1 × 81 mg tablet; Children's Chewable Tablets, Bayer HealthCare LLC, in Leverkusen, Germany) or clopidogrel 75 mg (1 × 75 mg tablet; Plavix® tablets; Bristol-Myers Squibb/Sanofi Pharmaceuticals Partnership, Bridgewater, NJ) were administered open label.
Pharmacodynamic Assessments

Bleeding time measurements were obtained prestudy, 4 hours postdose on Day 7 and 24 hours postdose on day 7. A small incision wound was made with a disposable bleeding-time device (Surgicutt®, International Technidyne Corporation, Edison, NJ). The time of the incision and the time at which bleeding from the incision stopped were recorded. Bleeding times in the Aspirin study greater than 20 minutes and greater than 30 minutes in the Clopidogrel study were truncated and the measurement was reported as >1200 seconds or >1800 seconds, respectively.

The pharmacodynamic goals of both studies were to determine the effect of multiple-dose LRPT 40 mg on bleeding time and platelet aggregation in the presence of either aspirin or clopidogrel over a 24-hour period after dosing on Day 7. To assess the effects of LRPT on platelet aggregation, blood samples (15 mL) were collected at prestudy, predose on Day 1, 4 hours postdose on Day 7, and 24 hours postdose on Day 7. Blood samples were collected into buffered 0.106 M sodium citrate tubes and centrifuged at 400×g for 10 minutes at room temperature to harvest platelet-rich plasma (PRP). The remaining volume was centrifuged at 1500×g for 10 minutes to obtain platelet-poor plasma (PPP). The platelet count of the PRP was adjusted to approximately 200,000 to 300,000 platelets/µL by dilution with PPP. Aliquots of 0.45 mL adjusted for PRP or PPP were added to separate aggregometer cuvettes and warmed to 37°C. The cuvettes were placed in separate wells, with the PPP serving as a blank for light transmission. Once the baseline was stabilized, the aggregometer (2-channel) (Chronolog, Havertown, PA) was calibrated and the increases in light transmission were measured following the addition of 2 µg/mL collagen as the primary agonist. In the Clopidogrel Study 5.0 µM ADP (4 hours and 24 hours postdose on Day 7) also was used as an agonist. All platelet aggregation
studies were performed within 1.5 hours of the blood collection. Aggregation was followed for 5 minutes after the addition of agonist and the maximum percentage of light transmission (i.e., extent of transmission, primary variable) obtained during this period and the instrument-calculated slope (i.e., rate of aggregation, secondary variable) was recorded.

Safety Measurements

Safety and tolerability were assessed by clinical evaluation of adverse events (AEs) and inspection of safety parameters, including physical examinations, vital signs, routine laboratory safety measurements (hematology, blood chemistry, and urinalysis), and 12-lead electrocardiograms (12-lead ECG). AEs were monitored throughout the study and evaluated in terms of intensity (mild, moderate, severe), duration, severity, outcome, and relationship to study drug.

Statistical Analyses

A mixed-effects model appropriate for a 2-period, crossover study design with terms for sequence, period, treatment as fixed effects and subject within sequence as a random effect was used to evaluate the effect of LRPT on aspirin- or clopidogrel-induced bleeding time. Bleeding time data were analyzed on the log scale, and the data were back transformed for reporting purposes. Ninety percent confidence intervals (CIs) were calculated for the geometric mean ratio (GMR, [LRPT+aspirin]/aspirin or [LRPT+clopidogrel]/clopidogrel) of bleeding time of [aspirin + LRPT] or [clopidogrel + LRPT] vs. aspirin or clopidogrel alone at 24 hours post-dose on Day 7 and compared with the prespecified similarity bounds of (0.66, 1.50). If the 90% CI fell within (0.66, 1.50), it would be concluded that the effects of [aspirin + LRPT] or [clopidogrel + LRPT]
on bleeding time were similar to aspirin or clopidogrel alone at 24 hours postdose on Day 7 (i.e., primary endpoint). Similar analyses were repeated for bleeding time at 4 hours postdose on Day 7 (i.e., exploratory endpoint). In the case of any censored values on bleeding time, the truncated bleeding times were estimated by the maximum observed bleeding time (i.e., 1200 seconds in the Aspirin Study and 1800 seconds in the Clopidogrel Study).

The effects of LRPT on the percent inhibition of platelet aggregation induced by various agonists (i.e., 2 µg/mL collagen [in both studies], 5.0 µM ADP [in the Clopidogrel Study in the presence of aspirin or clopidogrel at 4 hours and 24 hours postdose on Day 7] was analyzed using the same mixed effect model described for the bleeding time analyses. The baseline measurement was defined as the predose measurement performed on Day 1. Percent inhibition of platelet aggregation was calculated as a 1 minus percentage change from the baseline of the extent of platelet aggregation. The analysis was performed on the log scale and final results were reported on the original scale following back-transformation. SAS version 8.2 (SAS Institute, Cary, NC, USA) was used for all statistical analyses.

**Results**

In the Aspirin study, 14 healthy subjects were enrolled and randomized to treatment (Table I). All 14 subjects completed the study per protocol and were included in the pharmacodynamic and safety analyses. In the Clopidogrel study, 22 subjects were enrolled and randomized to treatment (Table I). Twenty-one subjects completed the study per protocol and were included in the pharmacodynamic analyses. One subject discontinued from the study due to increased hepatic enzyme levels (see further details in safety and tolerability section). All 21 subjects were included in the safety analyses.
**Bleeding Times**

The estimated GMRs (90% CIs) for bleeding time measured in the Aspirin and Clopidogrel Studies are shown in Table II.

In the Aspirin Study, the mean bleeding time profiles following concomitant administration of (aspirin + LRPT) or aspirin alone for 7 days are shown in Figure 1. The bleeding time profiles for the two treatments were generally superimposable across the prestudy, 4 hour, and 24 hour postdose time points. The GMRs ([LRPT + aspirin]/aspirin) and corresponding 90% CIs for the GMRs of bleeding times for [aspirin + LRPT] vs. aspirin alone at 4 (i.e., exploratory endpoint) and 24 hours (i.e., primary endpoint) postdose on Day 7 were 1.03 (0.86, 1.22) and 1.05 (0.93, 1.19), respectively. The 90% CIs for these GMRs fell entirely within the prespecified clinical similarity bounds of (0.67, 1.50). Furthermore, there were no statistically significant differences (p>0.200) in bleeding times between [aspirin + LRPT] and aspirin alone at 4 and 24 hours postdose on Day 7. These findings support the conclusion that the bleeding time observed following concomitant administration of aspirin 81 mg and LRPT 40 mg for 7 days was similar to that following the administration of aspirin 81 mg alone for 7 days.

In the Clopidogrel study, the mean bleeding time profiles following concomitant administration of [clopidogrel + LRPT] or clopidogrel alone for 7 days are shown in Figure 2. The bleeding time profiles for the two treatments were generally superimposable across the prestudy and 24-hour postdose time points but not the 4-hour postdose time point. The GMR ([LRPT+clopidogrel]/clopidogrel) 90% CI for the GMR of bleeding time for [clopidogrel + LRPT] vs. clopidogrel alone at 24 hours (i.e., primary endpoint) postdose on Day 7 was 1.04 (0.80, 1.34) which fell entirely within the prespecified clinical similarity bounds of (0.67, 1.50).
(Table II). The GMR and 90% CI for bleeding time at 4 hours (i.e., exploratory endpoint) postdose on Day 7 was 1.41 (1.09, 1.83). A statistically significant increase (p = 0.028) in bleeding time was seen with [clopidogrel + LRPT] vs. clopidogrel alone at 4 hours postdose on Day 7, whereas no significant between-treatment difference was seen at 24 hours postdose. These findings support the conclusion that bleeding time at 24 hours postdose on Day 7 following concomitant administration of clopidogrel 75 mg and LRPT 40 mg was similar to that seen following the administration of clopidogrel 75 mg alone for 7 days; however, a transient increase in bleeding time was observed following treatment with [clopidogrel + LRPT] at 4 hours postdose.

**Platelet Aggregation**

The effects of LRPT on platelet aggregation induced by various agonists (i.e., 2 µg/mL collagen [in both studies], 5.0 µM ADP [in the Clopidogrel Study], in the presence of aspirin or clopidogrel at 4 hours and 24 hours postdose on Day 7 are summarized in Tables III and IV, respectively. The time profile plots of percent inhibition of platelet aggregation as measured by percent amplitude in the Aspirin and Clopidogrel studies are depicted in Figures 3 and 4, respectively.

In the Aspirin Study, no statistically significant differences in collagen-induced platelet aggregation were seen at 4 and 24 hours postdose on Day 7 following treatment with [aspirin + LRPT] vs. aspirin alone (p=0.362 and p=0.987, respectively) (Table III). The time profile plots of percent inhibition in platelet aggregation were generally superimposable across the predose on Day 1, and 4 hour and 24 hour postdose time points (Figure 3). In the Clopidogrel Study, the percent inhibition in platelet aggregation induced by collagen and ADP were not statistically
significantly different (p>0.050) following treatment with [clopidogrel + LRPT] vs. clopidogrel alone at 24 hours postdose on Day 7 (Table IV). At 4 hours postdose on Day 7, statistically significant between-treatment differences (p<0.050) in the percent inhibition of collagen and ADP-induced platelet aggregation were observed (Table IV). Specifically, treatment with [clopidogrel + LRPT] vs. LRPT alone led to increases in inhibition of collagen- and ADP-induced platelet aggregation by 25% and 7%, respectively, at 4 hours postdose on Day 7 (Figure 4).

**Safety and Tolerability**

Co-administration of multiple-dose aspirin 81 mg or clopidogrel 75 mg with multiple-dose LRPT 40 mg was generally well-tolerated in healthy adult subjects. No serious clinical or serious laboratory adverse experiences were reported in either study and there were no consistent treatment-related changes in laboratory, 12-lead electrocardiogram or vital sign safety parameters.

In the Aspirin study, 5 subjects reported a total of 12 clinical adverse experiences of which 9 (i.e., nausea, loose stools, fatigue, abdominal cramp, general body pain, throat irritation, dry throat, scalp laceration and ecchymosis) occurred in subjects receiving [aspirin + LRPT] and 3 (i.e., cough, nasal congestion, rhinitis) occurred in subjects receiving aspirin alone. All of these adverse experiences were considered by the investigator to be mild in intensity, did not require action regarding study drugs, and all experiences resolved by the end of the study. One subject reported 1 adverse experience (i.e., abdominal cramp) while receiving [aspirin + LRPT], which was considered by the investigator to be possibly drug-related. There were no laboratory adverse experiences in this study.
In the Clopidogrel study, 16 subjects reported a total of 34 clinical adverse experiences of which 20 occurred in subjects receiving [clopidogrel + LRPT] and 14 occurred in subjects receiving clopidogrel alone (Table V). The most common clinical adverse experiences reported were headache, abdominal discomfort, nausea and vomiting. Two subjects experienced laboratory adverse experiences of white blood cells in urine while taking [clopidogrel + LRPT] which the investigator considered definitely not related to study drug. One subject was discontinued from the study prior to dosing [placebo + LRPT] on Day 2 of Period 1 due to increased hepatic enzyme levels already present prior to dosing. The investigator did not consider this discontinuation to be related to an adverse experience or to be drug related because the elevated enzyme was present before the study drug was administered. All 21 subjects were included in the safety analyses.

Discussion

The primary objectives of these studies were to evaluate the effects of LRPT, administered at the therapeutic dose of 40 mg, on the anti-platelet activity of aspirin or clopidogrel as measured by bleeding time and platelet responsiveness to inducers of platelet aggregation (i.e., collagen and ADP). Aspirin and clopidogrel are widely used antiplatelet drugs that lower morbidity and mortality in patients with cardiovascular risk [25]. Because many patients with dyslipidemia take aspirin and a significant number also take clopidogrel to reduce the risk of atherothrombotic events, these studies were performed to investigate the potential pharmacodynamic interaction between LRPT and either aspirin or clopidogrel.

Bleeding time at 24 hr postdose (trough) was prespecified as the primary pharmacodynamic endpoint in both studies. When properly conducted, capillary bleeding time is
considered a reproducible and sensitive method to measure the effects of treatment on overall hemostasis [26]. The bleeding time assesses platelet function in the context of damaged skin and microvasculature as well as the interaction of platelets with the coagulation and fibrinolytic system. Ex vivo platelet aggregometry assays also were conducted in both studies in an exploratory manner to evaluate platelet reactivity in response to inducers of platelet aggregation (i.e., agonists) in platelet rich plasma. Collagen was used as an agonist in both studies to induce the aggregation of platelets ex vivo. Aspirin inhibits collagen-induced platelet aggregation by blocking TxA2 synthesis and preventing the activation of TP [21]. For the Clopidogrel study, ADP also was used as an agonist because clopidogrel inhibits platelet aggregation by inhibiting the binding of ADP to its platelet receptor and the subsequent ADP-mediated activation of the glycoprotein complex governing platelet adhesion to the substratum [27]. Clopidogrel also suppresses platelet aggregation induced by agonists other than ADP by inhibiting the amplification of platelet activation by released ADP [28].

The co-administration of LRPT 40 mg and aspirin 81 mg for 7 days did not alter the known effects of aspirin on the prolongation bleeding time or inhibition of collagen-induced platelet aggregation compared with aspirin alone. At 24 hours post dose on Day 7, there were neither clinically meaningful nor statistically significant changes in bleeding time or platelet aggregation with [aspirin + LRPT] vs. aspirin alone. Similar findings were also observed at 4 hours postdose on Day 7. These findings indicate that LRPT neither reduces nor enhances the antiplatelet activity of aspirin in healthy adult subjects. The dose of aspirin used in this study was expected to suppress COX-1 derived prostanoids from platelets, including TxA2 and PDG2, but not from other cellular sources. The lack of an effect of LRPT on the antiplatelet effects of aspirin was not surprising given that the activities of these two drugs are believed to be mediated
by a common pathway. Aspirin 81 mg has been shown to inhibit production of TxA2 by >95% which prevents the activation of the TP receptor and thus would be expected to abrogate modest inhibitory effects of LRPT on TP.

The co-administration of LRPT 40 mg and clopidogrel 75 mg for 7 days resulted in modestly longer bleeding times and greater inhibition of collagen-induced platelet aggregation compared with clopidogrel alone at 4 hours post dose. These transient effects likely reflect the off-target activity of LRPT on the TP receptor. Inhibition of ADP-induced platelet aggregation also was significantly increased at 4 hours postdose but to a lesser degree compared with collagen. This finding was not surprising given that ADP-induced platelet aggregation is known to be less dependent on TP activation compared with that seen in response to collagen. At 24 hours post dose on Day 7, there were no clinically meaningful or statistically significant changes in bleeding time or platelet aggregation with [clopidogrel + LRPT] vs. clopidogrel alone. Aspirin is effective in reducing the incidence of atherothrombotic events at doses that inhibit collagen-induced platelet aggregation and prolong bleeding time throughout the entire dosing interval. Other NSAIDs that are reversible inhibitors of COX-1 and transiently inhibit platelet function have generally not been demonstrated to be effective at reducing atherothrombotic events. Based on these findings in the literature, it is concluded that transient effects on platelets which do not result in prolongation of bleeding time throughout the dosing interval are not likely to be clinically important. Taken together, these findings suggest that LRPT produces a transient, moderate, effect on the antiplatelet activity of clopidogrel at 4 hours postdose but did not result in any clinically important effects at the 24 hours postdose time point.

The co-administration of LRPT 40 mg with either aspirin 81 mg or clopidogrel 75 mg was generally well tolerated in healthy adult subjects. There were no serious clinical or
laboratory adverse experiences reported in these studies. No subjects discontinued from these studies due to adverse experiences. There were no clinically significant effects of the concomitant administration of LRPT and aspirin or clopidogrel on blood chemistry tests, hematology parameters, vital signs and ECG parameters. Overall, clinical adverse experiences associated with the concomitant administration of LRPT with aspirin or clopidogrel were mild, transient, and self-limited in nature.

Acetylsalicylic acid (ASA) and clopidogrel are used together in patients with increased cardiovascular risk. The effect of LRPT in patients taking both antiplatelet agents cannot be determined from the results reported here. Studies evaluating the effect of LRPT on the antiplatelet activity of ASA and clopidogrel taken together will inform about the use of LRPT in these patients. Pending these results, clinicians should be aware of the potential increased antiplatelet effect of ASA and clopidogrel in combination with LRPT.

Conclusion

In summary, co-administration of multiple doses of LRPT 40 mg and aspirin 81 mg or clopidogrel 75 mg had no clinically relevant effects on bleeding time or platelet aggregation.

Declarations of Interest

The funding for this study was provided by Merck Research Laboratories, Merck & Co., Inc. Financial Disclosures: Authors A. Dallob, W.L. Luo, J.A. Mabalot Luk, L. Ratcliffe, A.O. Johnson-Levonas, J. Schwartz, V. Dishy, I. De Lepeleire, W. Radziszewski, J.A. Wagner, and E. Lai are current or former employees of Merck & Co., Inc. and may own stock or hold stock
options in the company. Grants were provided by Merck to the institutions of W. Kraft, A. Van Hecken, and J.N. de Hoon for this study.
References


**Table I.** Baseline demographics for the randomized subjects

<table>
<thead>
<tr>
<th>Demographic</th>
<th>Aspirin Study PN047</th>
<th>Clopidogrel Study PN072</th>
</tr>
</thead>
<tbody>
<tr>
<td>Race, n (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Black</td>
<td>6 (42.9%)</td>
<td>0</td>
</tr>
<tr>
<td>Hispanic</td>
<td>1 (7.1%)</td>
<td>0</td>
</tr>
<tr>
<td>Caucasian</td>
<td>7 (50.0%)</td>
<td>22 (100%)</td>
</tr>
<tr>
<td>Gender, n (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>10 (71.4%)</td>
<td>12 (54.5%)</td>
</tr>
<tr>
<td>Female</td>
<td>4 (28.6%)</td>
<td>10 (45.5%)</td>
</tr>
<tr>
<td>Age (yr), mean (range)</td>
<td>31.1 (20-45)</td>
<td>37.0 (21-53)</td>
</tr>
<tr>
<td>Height (cm), mean (range)</td>
<td>172.8 (151-190)</td>
<td>174.4 (159-189)</td>
</tr>
<tr>
<td>Weight (kg), mean, (range)</td>
<td>77.3 (59.9-74.3)</td>
<td>74.6 (54.0-101.8)</td>
</tr>
</tbody>
</table>
**Table II.** Bleeding time (seconds) at 4 and 24 hours postdose on Day 7 following treatment with aspirin 81 mg or clopidogrel 75 mg with or without laropiprant 40 mg in healthy subjects

<table>
<thead>
<tr>
<th>Time (Day 7)</th>
<th>Aspirin N</th>
<th>LS mean&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Aspirin+ Laropiprant N</th>
<th>LS mean&lt;sup&gt;a&lt;/sup&gt;</th>
<th>[Aspirin+ Laropiprant]/Aspirin GMR&lt;sup&gt;b&lt;/sup&gt; (90% CI)</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 hr</td>
<td>14</td>
<td>572</td>
<td>14</td>
<td>587</td>
<td>1.03 (0.86, 1.22)</td>
<td>0.790</td>
</tr>
<tr>
<td>24 hr</td>
<td>14</td>
<td>519</td>
<td>14</td>
<td>546</td>
<td>1.05 (0.93, 1.19)</td>
<td>0.461</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Time (Day 7)</th>
<th>Clopidogrel N</th>
<th>LS mean&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Clopidogrel+ Laropiprant N</th>
<th>LS mean&lt;sup&gt;a&lt;/sup&gt;</th>
<th>(Clopidogrel+ Laropiprant)/Clopidogrel GMR&lt;sup&gt;c&lt;/sup&gt; (90% CI)</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 hr</td>
<td>20</td>
<td>622</td>
<td>20</td>
<td>879</td>
<td>1.41 (1.09, 1.83)</td>
<td>0.028</td>
</tr>
<tr>
<td>24 hr</td>
<td>20</td>
<td>572</td>
<td>20</td>
<td>594</td>
<td>1.04 (0.80, 1.34)</td>
<td>0.803</td>
</tr>
</tbody>
</table>

<sup>a</sup>LS mean = least squares mean; back-transformed from log scale
<sup>b</sup>GMR = geometric least-squares mean ratio [aspirin + laropiprant]/aspirin
<sup>c</sup>GMR = geometric least-squares mean ratio [clopidogrel + laropiprant]/clopidogrel
CI = confidence interval
Table III. Summary statistics for percent inhibition of collagen-induced platelet aggregation at 4 and 24 hours postdose on Day 7 following treatment with aspirin 81 mg with or without laropiprant 40 mg in healthy subjects

<table>
<thead>
<tr>
<th>N</th>
<th>Time (Day 7)</th>
<th>Aspirin LS mean</th>
<th>Aspirin + Laropiprant LS mean</th>
<th>[Aspirin + Laropiprant]-Aspirin Difference (90%CI)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>14</td>
<td>4 hr</td>
<td>86.0</td>
<td>84.1</td>
<td>-1.9 (-5.5, 1.6)</td>
<td>0.362</td>
</tr>
<tr>
<td>14</td>
<td>24 hr</td>
<td>81.3</td>
<td>81.2</td>
<td>-0.1 (-7.2, 7.1)</td>
<td>0.987</td>
</tr>
</tbody>
</table>

*aLS mean = least squares mean; back-transformed from log scale
CI = confidence interval
Table IV. Summary statistics for percent inhibition of collagen- and ADP-induced platelet aggregation at 4 and 24 hours postdose on Day 7 following treatment with clopidogrel 75 mg with or without laropiprant 40 mg in healthy subjects

<table>
<thead>
<tr>
<th>N</th>
<th>Time (Day 7)</th>
<th>LS Mean</th>
<th>Difference</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Clopidogrel</td>
<td>Clopidogrel + Laropiprant</td>
<td>[Clopidogrel + Laropiprant] - Clopidogrel</td>
</tr>
<tr>
<td></td>
<td></td>
<td>% Inhibition of Platelet Aggregation Induced by Collagen: % Amplitude</td>
<td></td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>4 hr</td>
<td>14.2</td>
<td>38.9</td>
<td>24.6 (11.8, 37.6)</td>
</tr>
<tr>
<td>20</td>
<td>24 hr</td>
<td>13.4</td>
<td>28.9</td>
<td>15.4 (1.7, 29.3)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>% Inhibition of Platelet Aggregation Induced by ADP: % Amplitude</td>
<td></td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>Day 7 - 4 hr</td>
<td>59.8</td>
<td>66.7</td>
<td>6.9 (2.9, 10.9)</td>
</tr>
<tr>
<td>20</td>
<td>Day 7 - 24 hr</td>
<td>56.1</td>
<td>60.3</td>
<td>4.2 (-0.3, 8.7)</td>
</tr>
</tbody>
</table>

---

**Notes:**

- LS mean = least-squares mean; back-transformed from log scale.
- Difference = difference of percent inhibition between [clopidogrel + laropiprant] - clopidogrel back-transformed from log percent scale.
- CI = confidence interval.
Table V. Number (%) of subjects with specific clinical adverse experiences

<table>
<thead>
<tr>
<th>Adverse Experiences</th>
<th>Placebo + Clopidogrel 75 mg N = 21</th>
<th>Laropiprant 40 mg + Clopidogrel 75 mg N = 22</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n (%) a</td>
<td>n (%) a</td>
</tr>
<tr>
<td>Gastrointestinal Disorders</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Abdominal discomfort</td>
<td>1 (4.8)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Abdominal pain</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>1 (4.8)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Dyspepsia</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Nausea</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Stomach discomfort</td>
<td>2 (9.5)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Vomiting</td>
<td>1 (4.8)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>General Disorders/Treatment Site Conditions</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fatigue</td>
<td>1 (4.8)</td>
<td>2 (9.1)</td>
</tr>
<tr>
<td>Sensation of pressure</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Vessel puncture site hematoma</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Vessel puncture site pain</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Infections and Infestations</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Influenza</td>
<td>0 (0.0)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Nasopharyngitis</td>
<td>1 (4.8)</td>
<td>1 (4.5)</td>
</tr>
<tr>
<td>Nervous System Disorders</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Headache</td>
<td>4 (19.0)</td>
<td>3 (13.6)</td>
</tr>
<tr>
<td>Respiratory, Thoracic, and Mediastinal Disorders</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cough</td>
<td>1 (4.8)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Pharyngolaryngeal pain</td>
<td>0 (0.0)</td>
<td>2 (9.1)</td>
</tr>
<tr>
<td>Rhinorrhea</td>
<td>1 (4.8)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Skin and Subcutaneous Tissue Disorders</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rash papular</td>
<td>1 (4.8)</td>
<td>0 (0.0)</td>
</tr>
</tbody>
</table>

aAlthough a subject may have had two or more clinical adverse experiences, the subject is counted only once within a category. The same subject may appear in more than one category.
Figure Legends

**Figure 1.** Time profile (mean ± SE) of bleeding time at 4 and 24 hours postdose on Day 7 following treatment with aspirin 81 mg (Study 1) with or without laropiprant 40 mg.

**Figure 2.** Time profile (mean ± SE) of bleeding time at 4 and 24 hours postdose on Day 7 following treatment with clopidogrel 75 mg (Study 2) with or without laropiprant 40 mg.

**Figure 3.** Time profile (mean ± SE) of percent inhibition from baseline in collagen-induced platelet aggregation at 4 and 24 hours postdose on Day 7 following treatment with aspirin 81 mg (Study 1) with or without laropiprant 40 mg.

**Figure 4.** Time profile (mean ± SE) of percent inhibition from baseline in collagen- and ADP-induced platelet aggregation at 4 and 24 hours postdose on Day 7 following treatment with clopidogrel 75 mg (Study 2) with or without laropiprant 40 mg.
Figure 1.
Figure 2.
Figure 3.
Figure 4.