SYNOPSIS OF RESEARCH REPORT [redacted] (PROTOCOL WP21272)

COMPANY: NAME OF FINISHED PRODUCT: NAME OF ACTIVE SUBSTANCE(S):	(FOR NATIONAL AUTHORITY USE ONLY)

TITLE OF THE STUDY / REPORT No. / DATE OF REPORT	An open-label, randomized 2-period crossover study to investigate the pharmacodynamics, pharmacokinetics, safety and tolerability of warfarin in combination with oseltamivir in volunteers stabilized on warfarin therapy. Report No. [redacted] December 2008.		
INVESTIGATORS / CENTERS AND COUNTRIES	[redacted]		
PUBLICATION (REFERENCE)	None		
PERIOD OF TRIAL	February 8, 2008 to July 10, 2008	CLINICAL PHASE	IV
OBJECTIVES	**Primary**: • To investigate the effect of a 5-day treatment course with oseltamivir on the pharmacodynamics of warfarin in volunteers stabilized on warfarin. **Secondary**: • To investigate the effect of a 5-day treatment course with oseltamivir on the steady state pharmacokinetics of warfarin (R and S forms) in volunteers stabilized on warfarin. • To investigate the single dose and steady state pharmacokinetics of oseltamivir and its carboxylate metabolite in the presence of warfarin in volunteers stabilized on warfarin. • To investigate the safety and tolerability of oseltamivir and warfarin when given concomitantly to volunteers stabilized on warfarin.		
STUDY DESIGN	An open-label, randomized 2-period crossover study with at least a 4 day washout between periods in volunteers stabilized on warfarin therapy.		
NUMBER OF SUBJECTS	20 subjects, 10 to each of the two randomized treatment sequences (AB, BA)		
DIAGNOSIS AND MAIN CRITERIA FOR INCLUSION	**Inclusion Criteria:** • Male or female subjects, 18-75 years of age, inclusive. • Subjects must have been receiving warfarin once daily for at least 4 weeks prior to Screening. • Subjects must have regular INR monitoring during warfarin therapy prior to study entry, and willing to be trained in the use of CoaguChek® devices. • INR must fall within a target range of 2.0-3.5 (as determined by CoaguChek® or a clinical laboratory INR test).		

DIAGNOSIS AND MAIN CRITERIA FOR INCLUSION Cont'd	• A BMI between 18-32 kg/m^2 inclusive • Able to participate, and willing to give written informed consent and to comply with the study restrictions • Confirmation by dietician questionnaire of a reasonably balanced diet. **Exclusion Criteria:** • An INR value between screening and Day -1 lower than 2.0 or greater than 3.5. • A change in their prescribed daily warfarin dose between screening and Day -1 • History of any coagulopathy for example von Willebrand's disease. • Consumption of health products or supplements containing Vitamin K. • If capable of reproduction, unwilling to use an effective (barrier) form of contraception (post-menopausal status must be verified by negative hormone panel). • Positive pregnancy test at screening or Day -1 and lactating women. • Confirmed positive urine and/or blood test for drugs of abuse at screening or Day -1. • History of drug or alcohol abuse. • Donation or loss of blood over 500 mL within three months prior to screening. • Participation in an investigational drug or device study within three months prior to screening. • Smokers of > 5 cigarettes or equivalent in tobacco per day (> 3 pipefulls or > 3 cigars per day). • Use of any prescription drug, over the counter medication or herbal product, known to be an inducer or inhibitor of CYP450 enzymes or any drugs included in the list of prohibited medications, taken within 7 days of first dose of study drug, or 5 times the elimination half-life of the medication, whichever is longer, unless the patient/volunteer is stable on the stated medication and it has been taken for a period of ≥ 3 months. • Consistent supine systolic blood pressure (SBP) greater than 160 or less than 80 and Consistent supine diastolic blood pressure (DBP) greater than 90 or less than 60 mm Hg between screening and Day -1 • Consistent supine heart rate (HR) at rest greater than 90 less than 45 beats per minute (bpm) between screening and Day -1. • Any clinically significant abnormalities in laboratory test results in this patient population (including hepatic and renal panels, complete blood count, chemistry panel, serology and urinalysis) at screening or Day -1. • A history of acute clinically significant gastro-intestinal, musculoskeletal, endocrine, hematological, psychiatric, renal, hepatic, bronchopulmonary, neurological disease, acute venous thromboembolism within the last 3 months, a mechanical heart valve, acute stroke, transient ischaemic event (TIA) or acute myocardial infarction.

DIAGNOSIS AND MAIN CRITERIA FOR INCLUSION Cont'd	• Creatinine Clearance of < 60 mL/min (as estimated using Cockroft-Gault formula). This may be confirmed by urine creatinine clearance, the results of which will override the Cockroft-Gault estimated results. • Any other concomitant disease or condition that could interfere with, or for which the treatment of might interfere with, the conduct of the study, or that would, in the opinion of the investigator, pose an unacceptable risk to the subject in this study. • Known history of clinically significant allergic hypersensitivity reactions or drug hypersensitivity (non-active hay fever is acceptable), including a known allergy to the study drug or to any of its components.
TRIAL DRUG / STROKE (BATCH) No.	Material no. RO 64-0796-V01, batch no. [redacted]
DOSE / ROUTE / REGIMEN / DURATION	• Tamiflu® (Ro 64-0796) • Warfarin Eligible subjects were already receiving warfarin once daily, at a dose determined through titration by their usual hematologist (to maintain an INR target value of 2.0-3.5). There were two treatment periods each of 5 days duration, and the order in which these treatments were received was randomized. **Treatment A**: The subject's usual once daily maintenance dose of warfarin (as determined by previous titration). Treatment was administered with a glass of water. **Treatment B**: In addition to their once daily maintenance dose of warfarin (as determined by previous titration), subjects received Tamiflu® (Ro 64-0796), dosed as oseltamivir 75 mg capsules b.i.d. p.o. for 4 days, with a single dose on Day 5. Oseltamivir was administered a.m. on Day 1 irrespectively of the time of the day at which the subjects took their regular warfarin QD dose (a.m. or p.m.). Oseltamivir was administered with water and at approximately 12-hourly intervals. Oseltamivir was given with some food. N.B. Subjects continued to receive their prescribed usual dose of warfarin throughout the study (unless safety findings indicated that dose adjustment was warranted). **If dose adjustment of warfarin was judged warranted, administration of oseltamivir was discontinued,** and their safety monitored to resolution.
CRITERIA FOR EVALUATION	
PHARMACODYNAMICS:	• INR: AUEC (Day 1-5), maximum INR and time to reach maximum INR. • Factor VII activity: AUEC (Day 1-5), minimum factor VII activity and time to reach minimum factor VII activity. • Plasma concentrations of vitamin K_1 before and after treatment with oseltamivir.

PHARMACOKINETICS:	**Warfarin** R- and S-warfarin: AUC_{0-12h}, AUC_{0-24h}, C_{max}, $t_{½}$, CL/F. **RO640796 and RO640802** Day 1 - RO640796: AUC_{0-12h}, C_{max}, t_{max}, $t_{½}$, CL/F Day 1 - RO640802: AUC_{0-12h}, C_{max}, t_{max}, apparent $t_{½}$ Day 5 - RO640796: AUC_{0-12h}, $AUC_{0-24,}$ C_{max}, t_{max}, $t_{½}$, CL/F Day 5 – RO640802: AUC_{0-12h}, $AUC_{0-24,}$ C_{max}, t_{max}, apparent $t_{½}$
SAFETY:	A complete physical examination was performed at screening and follow up. Adverse events were monitored throughout the entire study (screening through follow up). <u>Vital Signs</u> Systolic (SBP) and diastolic (DBP) blood pressure (BP), body temperature and pulse rate (PR). BP and PR were always taken after remaining in the supine position for at least 5 minutes. <u>ECG:</u> Machine-read semi-automated ECGs were recorded and along with information on T- and U-waves as normal/abnormal and post dose ECG significant changes from baseline were noted. ECGs were taken after remaining in the supine position for at least 5 minutes. <u>Laboratory tests</u> : 1. Hematology: red blood cell count, white blood cell count, hemoglobin, hematocrit, MCV, MCH, MCHC, platelet count, total and differential WBCs (neutrophils, eosinophils, basophils, lymphocytes, monocytes,), PT, Thrombin time, activated partial thromboplastin time (aPTT), INR (derived) 2. Biochemistry: sodium, potassium, chloride, bicarbonate, creatinine, glucose, urea, albumin, calcium, magnesium, inorganic phosphorus, alkaline phosphatase (ALP), aspartate amino transferase (SGOT/AST), alanine amino transferase (SGPT/ALT), Gamma-glutamyl transferase (GGT), total bilirubin, total protein, total cholesterol, HDL, LDL, triglycerides, uric acid, serum folate, amylase, thyroid-stimulating hormone (TSH) and T4. 3. Urinalysis: A midstream, clean-catch urine specimen was collected for dipstick analysis for blood, glucose, leukocytes and pH. Microscopic analysis was only performed if results were positive or strong positive for blood, protein or leukocytes. 4. Drugs of Abuse (urine): Cannabinoids, amphetamines, opiates, methadone, cocaine, benzodiazepines, and barbiturates 5. Virology: Anti-HAV IgM, HBsAg, anti-HBc IgM, anti-HCV antibody, and anti-HIV antibody 6. Alcohol: Breath or saliva test 7. Pregnancy Test: Urine specimen was collected and analyzed (β-hCG). 8. Post-menopausal status: FSH (screening only)

STATISTICAL METHODS	**Pharmacodynamics** To assess the effect of multiple doses or oseltamivir on the warfarin INR, the $AUEC_{0\text{-}96h}$ (baseline corrected) was compared between treatment arms A and B and 90% confidence intervals for the difference (B-A) were derived. Similar analyses were performed for INR E_{max}, Factor VII $AUEC_{0\text{-}96h}$ and E_{min}, and for Vitamin K_1 change from baseline. **Pharmacokinetics** For assessment of the effect of oseltamivir on the PK of warfarin following multiple dosing with oseltamivir and warfarin (Treatment B) were compared to those following multiple dosing of warfarin alone (Treatment A). Safety The safety and adverse event data will be presented in individual listings, summary tables and graphs as appropriate. Adverse events will be listed and summarized at onset of adverse event. Body system and preferred terms within each body system will be summarized. All adverse events with all occurrences will be listed by subject. Clinical laboratory data will be listed by subject. Values outside the reference ranges will be highlighted and clinical significance stated. Vital sign measurements will be listed by subject. Plots of vital signs data will be provided.

METHODOLOGY:

Screening (Days -43 to -15)

After written informed consent had been given, a screening examination took place. Screening was performed between 43 and 15 days (inclusive) before the subject's first dosing day and involved demographics, physical examination including height and weight measurement for calculation of BMI, a complete medical history, clinical laboratory tests (including hematology, biochemistry, urinalysis and serology), a urine and/or blood drugs of abuse test and Alcohol Breath Test (ABT), vital signs (supine blood pressure, pulse rate and body temperature) and a 12-lead ECG recording. A pregnancy test was performed for all females of child-bearing potential. Hormone levels (FSH) for females were also measured to confirm postmenopausal status, as appropriate. All females of child-bearing potential undertook a pregnancy test prior to the first treatment period. Following these investigations, subjects who had no clinically relevant findings and fulfilled all the inclusion and exclusion criteria were accepted to the study conditional on INR results within a stable range, as defined in the inclusion/exclusion criteria, between Day -14 to -1. Consequently, all subjects were supplied a CoaguChek® device for the purposes of measuring the INR during the conduct of the study, and which were provided by the site. Tests strips were also provided by the site for the duration of the study (screening through to follow up).

Throughout screening, baseline and during the course of the study, subjects continued to take their usual warfarin therapy (i.e. with their regularly prescribed dose and brand of warfarin), and also any other regularly administered concomitant medications.

Day -14 to -4

The following assessments were performed:

Daily INR checks took place for two weeks prior to Day 1 of study as determined by CoaguChek® whenever at home or alternatively a clinical laboratory INR test if present at the Clinical Unit. Subjects were provided with a 'patient record card' to self-record the INR results from Day -14 to Follow-up.

Day -3 to -2

The following assessments were performed at the Clinical Unit:

Vital signs (supine blood pressure, pulse rate and body temperature) and a 12-lead ECG. Daily INR checks took place as determined by CoaguChek®, urine and/or blood drugs of abuse test, alcohol breath test.

Subjects were admitted into the Unit, and resided on site until the morning of Day 6, after the 24-hour pharmacokinetic sample of each treatment period had been taken and all assessments had been completed. When residing in the Unit subjects received standardized meals.

After Period 1, subjects could then leave the Unit, and return on Day -3 prior to Period 2.

Note: Whenever subjects were receiving Treatment A, they were treated the same as their counterparts in the opposite sequence. Consequently, they stayed in the Unit until all Treatment B assessments had been completed.

Day -1

On Day -1 of each treatment period, the following assessments were performed:

Clinical laboratory tests (including hematology, biochemisty INR and urinalysis), vital signs (supine blood pressure, pulse rate and body temperature) and a 12-lead ECG. All females of child-bearing potential undertook a pregnancy test prior to the first treatment period.

Treatment Periods (Days 1 to 5 of Periods 1 and 2)

Prior to first dosing in the morning of Day 1 (Period 1 only), subjects were randomized to one of the two treatment sequences (AB, BA). Thus, oseltamivir (Treatment B) was only administered either in Period 1 or Period 2. Between Period 1 and Period 2 there was at least a 4 day washout period.

Whilst resident in the Clinical Unit, subjects had standardized breakfast, lunch, dinner, and snacks were offered at set times. Fluids were permitted ad libitum, except following morning dosing on Day 1 and Day 5, where subjects were not allowed to eat or drink anything additional to the standardized meals or snacks for two hours after their Treatment A or B dose(s). During these two hours subjects were requested to remain in an upright position. All study medication was administered with approximately 200 mL of water.

On Day 1 to 5 of each treatment period, the following assessments were performed:

Clinical laboratory tests (including hematology, biochemistry and urinalysis), vital signs (supine blood pressure, pulse rate and body temperature) and a 12-lead ECG. On Days 1, 4 and 5 only, PK assessments took place. Once all the Day 6 assessments were completed, subjects were allowed to leave the unit.

Adverse events were monitored throughout the study (from after the screening visit to follow-up). Any adverse events noted were entered in the eCRF for clinical safety assessment and monitoring. Subjects were encouraged to report promptly any adverse event that happened during the course of the study.

Follow up

A follow up examination was performed **4-12** days after the last dose in Period 2. Samples for clinical laboratory tests (hematology, biochemistry, urinalysis). The follow up assessment also included a physical examination, 12-lead ECG and vital sign.

PHARMACODYNAMIC RESULTS:

The computed INR and Factor VIIa parameters were similar between treatments. The change in plasma concentrations of vitamin K_1 was similar between treatments.

PHARMACOKINETIC RESULTS:

There were no marked differences between the normalized primary pharmacokinetic parameters of warfarin enantiomers (total and unbound to plasma proteins) when given with or without oseltamivir. Similarly, there were no patent differences between any of the secondary parameters either alone or in combination wit oseltamivir. There were no marked differences in the pharmacokinetics of oseltamivir when given with or without warfarin either after a single dose or at steady state. The results also indicate that the data from the present study is broadly comparable to that from the historic study NP15717, but also that the possibility of drug-drug interaction of warfarin on oseltamivir is unlikely.

SAFETY RESULTS:

There was no treatment-related increase in the severity of reported adverse events and no adverse events indicative of a toxic effect on a given organ system. There were no deaths occurring in this study. During the washout period prior to follow-up there was a serious adverse event where Subject [redacted] presented 'angina pectoris'. This type of event was foreseeable given the baseline diseases of the population required of this study. The adverse event was moderate in intensity and judged by the Investigator to be unrelated to the trial medication. The event resolved with no sequels. No subjects who received study drug were withdrawn early from the study due to an adverse event. There were no clinically significant or treatment related changes from baseline in vital signs or electrocardiogram following the administration of the subject's usual warfarin dose regimen with or without oseltamivir. A number of isolated out-of-rage laboratory parameters were identified, but these were considered normal findings taking into account the demographics of the studied population with a median (range) age of 60 (54-75) years. There were no relevant trends, either increase or decrease, in any of the laboratory parameters, and there were no clinically significant changes from baseline noted.

CONCLUSIONS: The results of this study confirm that oseltamivir does not enhance the effects of warfarin when both are given in combination in subjects who are stable on warfarin. This is supported by the absence of any signs of increased anticoagulation, as shown by the studies pharmacodynamic parameters: INR, Factor VIIa and Vitamin $K_{1.}$

There were no marked differences in the pharmacokinetics of oseltamivir when given with or without warfarin either after a single dose or at steady state.

Furthermore, the study confirms that oseltamivir was well tolerated and that it can be safely administered to subjects receiving concomitant warfarin therapy, as demonstrated by the low number of adverse events and the lack of effects on any of the studied clinical laboratory parameters, vital signs and 12-lead ECGs.