Synopsis of study report: 22/2003
Location in Module 5:

Report Version
Version 1.0 (dated 3 July 2003)

Title of the study:

Determination of cytochrome P450 3A4 activity in case subject AL10 of Study BY217/FHP027 - a case control study

Study center(s):

Swiss Pharma Contract Ltd., Allschwil, Switzerland

Publication (reference):

Not applicable

Studied period (years):

27 March 2003 – 04 April 2003

Clinical phase:

Phase I

Objectives:

The objective of this study is to determine the cytochrome P450 3A4 activity in subject AL10 of Study BY217/FHP027 *in vivo* and to compare this activity with the results of historic data of control subjects.

Methodology:

This study was conducted as a monocenter case control study. The study consisted of a pre-study medical examination (including blood pressure, heart rate, and ECG) within one week prior to study start, one treatment period of 12h on Day 1 (subject was hospitalized the afternoon before study start, Day -1 and remained in hospital until conclusion of the study, Day 2), and a post study medical examination conducted on Day 2 after study termination (including blood pressure, heart rate, and ECG). Blood samples for determination of

midazolam and its metabolites were collected predose (before intravenous administration of midazolam) and during the study period (after administration of midazolam) for up to 12h.
Blood pressure, heart rate, and ECG were recorded at predose, 5 minutes, 15 min, 0.5h, 1h, 2h, 4h, 6h, 8h, 10h, and 12h after administration of study drug. Adverse events were monitored throughout.

No. of subjects (total and for each treatment):

Case: 1 case subject (subject No. 10 from Study BY217/FHP027, Clinical Study Report Number 208/2001).
Control: Historical control group of healthy subjects taken from Floyd et al.2003.
Genotype - phenotype associations for common CYP 3A4 and CYP 3A5 variants in the basal and induced metaoblism of midazolam in European- and African-American men and women. Pharmacogenetics, in press 2003.

Diagnosis and criteria for inclusion:

The case subject was included in this study after showing high levels of roflumilast and roflumilast N-oxide in the plasma during Study No. BY217/FHP027, as compared with other participants in that study. The subject gave her written consent to participate in the study.

Test product:

Midazolam 5 mg/5 ml injection solution

Dose:

Midazolam 1 mg, administered as a single dose over 1 min
(Dormicum® V 5mg/5ml Roche, Grenzach Wyhlen, Germany)

Mode of administration:

Intravenous injection

Batch No.:

006103

Duration of treatment:

The study period lasted 12 h.
At the start of the study period, a single intravenous dose (1 mg administered over 1 min) was administered in the morning.

Reference product:

None

Dose:

Not applicable

Mode of administration:

Not applicable

Batch No.:

Not applicable

Criteria for evaluation:

Pharmacokinetics:

The primary variable of this study was to determine the enzyme activity of cytochrome P450 3A4. Midazolam has been widely used as a cytochrome P450 3A phenotyping probe. A dose of 1 mg midazolam administered intravenously over 1 min has been used in several clinical studies with good tolerability.

The primary routes of midazolam metabolism are 1-hydroxylation and 4-hydroxylation via CYP3A4 and CYP3A5. Conjugated 1-hydroxymidazolam (1-OHMDZ) is the primary metabolite found in plasma and urine, minor metabolites include 4-hydroxymidazolam (4-OHMDZ) and 1,4-dihydroxymidazolam. Several studies have demonstrated that midazolam weight normalized systemic clearance provides an accurate reflection of CYP 3A4 activity

Safety and tolerability:

Physical examinations, including vital signs such as blood pressure and heart rate, use of 12-lead ECG, and clinical laboratory examinations (blood chemistry, hematology, coagulation and urinalysis) were used as safety variables during this trial. In addition, adverse events were monitored during the entire study.

Statistical methods:

The primary variable of the study was the midazolam weight normalized systemic clearance [dose/(AUC x weight)] of subject AL10.

The secondary variables were:

- the ratio of $AUC_{(0\text{-inf.})}$ of 1-hydroxymidazolam/$AUC_{(0\text{-inf.})}$ of midazolam;
- the ratio of $AUC_{(0\text{-inf.})}$ of 4-hydroxymidazolam/$AUC_{(0\text{-inf.})}$ of midazolam;

- $AUC_{(0\text{-inf.})}$ of midazolam;
- $t_{1/2}$ of midazolam;
- safety variables.

SUMMARY - CONCLUSIONS

Summary:

Pharmacokinetic results:

The weight normalized clearance determined for case subject AL10 of Study BY217/FHP027 after administration of 1 mg of midazolam intravenously was 2.99 mL/(min × kg). Comparison with reference data showed that the subjects weight normalized clearance falls in the lower percentile (range: 2.61 - 3.88 mL/(min × kg) of the reference data set. Therefore, it can be concluded that the low clearance of the CYP 3A4 substrate midazolam observed in case subject AL10 is indicative that the case subject has reduced activity of the enzyme CYP 3A4.

Safety results:

During the treatment period one adverse event (tiredness) was reported by the case subject. The adverse event was assessed by the investigator as mild and as 'likely' related to the study drug (due to the sedative effect of midazolam). No serious adverse events were reported and no deaths occurred in this study.

Laboratory values did not show any clinically relevant changes between the screening and post-study examination. After intake of the study drug, no clinically relevant alterations were observed during physical examination (including ECG and vital signs).

Overall, the safety data obtained in the present study indicate that intake of a single intravenous dose of midazolam 1 mg was safe and well tolerated.

Conclusions:

The objective of this study was to determine the cytochrome P450 3A4 activity in case subject No. 10 of Study BY217/FHP027 (case subject AL) and to compare the enzyme activity of this subject with those of reference data obtained from the literature. The study was conducted as a case control study in case subject No. 10 of study BY217/FHP027. Midazolam was used as the marker compound to study the activity of cytochrome P450 3A4. Enzyme activities were determined by using a weight normalized systemic clearance of midazolam after one single intravenous dose of 1 mg midazolam.

The results of the present study show that the activity of the cytochrome P450 3A4 of the case subject was reduced when compared with reference data from (historical) control group

(Floyd et al. 2003). Because cytochrome P450 3A4 contributes considerably to the metabolism of roflumilast, such a decrease of CYP 3A4 activity could be responsible for decreased rate of metabolism of roflumilast and consequently may contribute to the increased plasma levels of roflumilast and roflumilast N-oxide found in the case subject.

Laboratory values (including blood chemistry, hematology, and urinalysis) and measurements of blood pressure, pulse rate and ECG parameters did not reveal any clinically significant alterations after administration of the study medication. The present study does not raise any safety issues regarding administration of a single dose of intravenous midazolam 1 mg. The adverse event (tiredness) reported by the case subject during the study period is in keeping with the relatively high midazolam exposure (resulting from reduced metabolism of midazolam) caused by the reduced activity of CYP 3A4 in the case subject.