Clinical Trial Results Disclosure Synopsis

Name of Sponsor: Takeda Pharma Vertieb GmbH & Co. KG, Jägerstr.27, 10117 Berlin, Germany

Title of Study: Double Blinded Study of the Effects of Pioglitazone in Combination with Atorvastatin in Comparison to Atorvastatin Treatment Alone on Intima-Media Thickness in Patients at Risk for Vascular Complications

Phase of Development: Phase II

Name of Active Ingredient: [(±)-5-[[4-[2-(5-ethyl-2-pyridinyl) ethoxy] phenyl] methyl]-2,4-] (pioglitazone)

Name of Finished Product: Actos®

Investigators: 2 principal investigators in Germany enrolled subjects into the double-blind treatment period.

Study Sites: 2 sites in Germany enrolled subjects into the study

Study Site 1: Ikfe GmbH, Parcusstr. 8, 55116 Mainz

Study Site 2 :GWT-TUD, Fiedlerstr. 34, 01307 Dresden

Publications Based on the Study (Citations) at Time of Study Completion: None

Study Period:

Date first subject signed informed consent form: 16 June 2005

Date of last subject's last visit/contact (from the Clinical database): 23 October 2006

Objectives:

The aim of this study was to evaluate the effect of Pioglitazone in addition to Atorvastatin compared to therapy with Atorvastatin alone on vascular risk markers and on the intima-media thickness (IMT) in patients with elevated risk for cardiovascular disease.

Methodology: Prospective, double-blind, two-center, randomized, parallel two arm study

Number of Subjects:

Planned: 160 subjects were to be enrolled to achieve 148 evaluable cases (74 per group)

Screened: 251 subjects
Randomized into the double-blind treatment period: 175 subjects
Analyzed: Safety Set: 175; Full-Analysis Set: 148; Per-Protocol Set: 137

Diagnosis and Main Criteria for Inclusion: Male or female patients at risk for vascular complications, with an age between 30 and 70 years, with a body mass index (BMI) $\geq$ 25 kg/m^2 and with a proven intima-media thickness of the common carotid artery (IMT_{CCA}) $\geq$ 0.8 mm at least on one side. Signed written informed consent available.

Duration of Treatment: The treatment phase with test or reference study medication was defined to be 24 weeks.

Test Product, Dose and Mode of Administration, and Lot Number:

Study Medication	Product Dose Strength and Form	Study Dosage	Mode of Administration	Drug Product Lot Number
Pioglitazone	30 mg capsule	30 mg QD	Oral	N/A
Pioglitazone	45 mg capsule	45 mg QD	Oral	N/A
Atorvastatin	20 mg tablet	20 mg QD	Oral	N/A
Atorvastatin	40 mg tablet	40 mg QD	Oral	N/A

Reference Therapy, Dose and Mode of Administration, and Lot Number:

Study Medication	Product Dose Strength	Study Dosage	Mode of Administration	Drug Product Lot Number
Placebo to Pioglitazone 30 mg	Capsule	N/A	Oral	N/A
Placebo to Pioglitazone 45 mg	Capsule	N/A	Oral	N/A

Criteria for Evaluation:

Efficacy:

Primary: The primary efficacy variable was the change of IMT_{CCA} after 24 weeks of treatment compared to baseline. The change was to be calculated as IMT_{CCA} at screening visit (visit 1; week -2±1) minus IMT_{CCA} at the end of study (visit 5; week 24±2).

Secondary: Influence of Pioglitazone in combination with Atorvastatin in comparison to Atorvastatin alone over 24 weeks on IMT of the internal carotid artery (ICA) and the carotid bulbus (CB), over 24 weeks on laboratory parameters of inflammation and vascular function (high-sensitivity C-reactive Peptide (hsCRP); Interleukin-6 (IL-6); Monocyte Chemotactic Protein-1 (MCP-1); Matrix Metalloproteinase-9 (MMP-9); soluble CD40 Ligand (sCD40L); soluble Intercellular Adhesion Molecule (sICAM-1); soluble Vascular Cell Adhesion Molecule (sVCAM-1); p-selectin; Tissue-Plasminogen Activator (t-PA)), over 24 weeks on laboratory

parameters of glucose tolerance and insulin sensitivity (plasma glucose; glycosylated hemoglobin (HbA_{1C}); insulin; intact proinsulin; adiponectin, Homeostatic Model Assessment – Sensitivity (HOMA-S), Homeostatic Model Assessment – beta cell function (HOMA-%B), over 24 weeks on laboratory parameters of lipid metabolism (total cholesterol; high density lipoprotein (HDL); low density lipoprotein (LDL)-triglycerides; LDL-subfractions), and over 24 weeks on vascular function (laser doppler flowmetry; pulse wave velocity).

Safety: Incidence of adverse events, change of routine and safety laboratory parameters, changes in physical examination and vital signs, electrocardiogram (ECG) recordings and the rate of premature withdrawals.

Statistical Methods:

Data from all clinical assessments whether explicitly referred to in the statistics section or not, were presented in summary tables and in individual patient data listings. Data were summarized with respect to demographic and baseline characteristics, efficacy and safety observations and measurements. Standard descriptive summary statistics were done for continuous variables (i.e. arithmetic mean, standard deviation (SD), minimum/maximum value, lower/median/upper quartile, number of non-missing values). Categorical data were displayed in frequency tables using counts and percentages. Individual patient data listings were presented parameterwise and were sorted by treatment group, center, patient number and visit. Summary tables were displayed by treatment group and for the total of the sample.

Efficacy:

The primary confirmatory analysis of the primary efficacy variable (IMT_{CCA}) for the full analysis set has to be distinguished from supporting exploratory analyses of the primary and secondary parameters. All p-values and confidence levels derived from additional inferential statistical methods are to be interpreted in the exploratory sense only.

SUMMARY OF RESULTS:

Baseline Demographics and Other Relevant Characteristics:

All 175 (88 Pioglitazone + Atorvastatin vs. 87 Atorvastatin) patients in the safety set were of Caucasian origin. When the two treatment groups are compared in this section, the order Pioglitazone + Atorvastatin vs. Atorvastatin alone always applies. The average age overall was 61.8 (6.4) (mean ± standard deviation (SD)) and was comparable across both treatment groups (61.3 (6.7) vs. 62.2 (6.1). Eighty-eight (88) patients were male (47 vs. 41) and 87 were female (41 vs. 46). Height, weight and body mass index (BMI) were also comparable across the two treatment groups (height (cm): 170 (9) vs. 168 (9); weight (kg): 83.7 (12.8) vs. 83.1 (13.5); BMI (kg/m^2: 29.1 (4.0) vs. 29.6 (4.3).

Prior medications were recorded at least once in 165/175 patients with the most frequently listed preparations (in > 10% of the patients) corresponding to the pre-defined study indication of

increased cardiovascular risk and findings in medical history. The most frequently listed single diseases (in > 5%) were hypercholesterolemia in 46/175 patients (26.3%, 21 vs. 25 cases), osteoarthritis in 38/175 patients (21.7%, 14 vs. 24), varicosis in 29/175 patients (16.6%, 15 vs. 14), struma in 27/175 patients (15.4%, 19 vs. 8), arterial sclerosis in 16/175 patients (9.1%, 11 vs. 5), osteoporosis in 15/175 patients (8.6%, 9 vs. 6), prostatic hyperplasia in 15/175 patients (8.6%, 10 vs. 5), hyperuricemia in 13/175 patients (7.4%, 6 vs. 7), coronary artery disease in 13/175 patients (7.4%, 5 vs. 8), back pain in 12/175 patients (6.9%, 6 vs. 6), lipid metabolism disorder in 11/175 patients (6.3%, 5 vs. 6) and seasonal allergy in 9/175 patients (5.1%, 8 vs. 1).

Subject Disposition:

A total of 251 patients were screened and enrolled by 2 participating German study centers. Thereof, 175 (88 Pioglitazone + Atorvastatin vs. 87 Atorvastatin) patients were randomized and treated with at least one dose of study medication yielding the safety set. Twenty-seven (27) patients were not suitable for the main efficacy analysis since the major entry criteria of IMT_{CCA} $\geq$ 0.8 mm at least on one side or the presence of at least one baseline and one post-baseline IMT measurement were not fulfilled, thus yielding a full-analysis set of 148 (68 Pioglitazone + Atorvastatin vs. 80 Atorvastatin) patients. Moreover, 38 patients with major protocol violations could not be considered for further efficacy analyses, leading to 137 (62 Pioglitazone + Atorvastatin vs. 75 Atorvastatin) patients allocated to the per-protocol analysis set. A total of 34 patients (19.4%; 23 Pioglitazone + Atorvastatin vs. 11 Atorvastatin) discontinued the study prematurely.

Efficacy Results: (full analysis set; n=148 (68 vs. 80))

Primary Efficacy Parameter (full analysis set; n=148 (68 vs. 80)):

The results of the IMT_{CCA}-change between last observation carried forward (LOCF) and V1 were as follows for the different treatment groups and for the study-specific stages of the statistical interim evaluations (stages 1, 2 and total):

Parameter [Unit]	**PIO + Atorvastatin, n=68** Mean Change ± SD (Median; n)	**Atorvastatin + Placebo, n=80** Mean Change ± SD (Median; n)	**p-value**
IMT CCA [mm]; Stage 1, n=80	-0.037 ± 0.037 (-0.030; n=37)	-0.040 ± 0.064 (-0.017; n=43)	0.5908
IMT CCA [mm]; Stage 2, n=68	-0.043 ± 0.044 (-0.041; n=31)	-0.061 ± 0.047 (-0.063; n=37)	0.9511
IMT CCA [mm]; Total, n=148	-0.040 ± 0.040 (-0.037; n=68)	-0.050 ± 0.058 (-0.042; n=80)	0.8957

As shown above, the confirmatory analysis failed to show superior efficacy of treatment with Pioglitazone plus Atorvastatin compared to Atorvastatin alone regarding a change of IMT_{CCA}.

The one-sided confirmatory p-value p_1 obtained from the ANCOVA model based on the original data of the first stage of study analysis resulted in $p_1 = 0.5908$ indicating no statistical significance at the one-sided adjusted significance level $\alpha_1 = 0.01019$.

The exploratory analyses of the second stage and the combined analysis of first and second stage were in accordance to the confirmatory results and trends of the first stage. The one-sided exploratory p-value p_2 obtained from the ANCOVA model based on the original data of the second stage was 0.9511 and the combined p-value ($p_1 x p_2$-product) was 0.5619.

To investigate the robustness of the ANCOVA results in case of serious deviations from the normal distribution assumption, the analysis of covariance was also performed based on normal scores of the ranks using the 'Blom'-transformation. Moreover, the normal distribution assumption of the residuals in the ANCOVA model was examined by the Shapiro-Wilk test and by visual check of plots of the residuals. In this context, the ANCOVA results related to the testing on between-group treatment differences were comparable for the original data and the rank-transformed data, showed no different tendencies and thus supported the primary confirmatory analysis and subsequent exploratory analyses. The one-sided exploratory p-values and their product from the ANCOVA of the rank transformed data were $p_1 = 0.2264$, $p_2 = 0.9593$ and $p_1 x p_2 = 0.2172$.

Moreover, the subgroup of patients who received a previous treatment with statins prior to the study start (n = 22; 8 vs. 14 cases) did not influence the primary study results relevantly.

Secondary Efficacy Parameters (full analysis set; n=148 (68 vs. 80)):

The influence of Pioglitazone in combination with Atorvastatin (Pio+Statin) in comparison to Atorvastatin alone on the different secondary efficacy variables is described in the following for the study period of 24 treatment weeks. A detailed summarizing overview of the secondary efficacy results is shown in the listings below providing the mean change with standard deviation and median between baseline (V1 or V2) and the individual study end (LOCF or V5) as well as the corresponding p-value for the between-group difference calculated by using the F-test for treatment effect on Lest Square (LS)-means of change by ANCOVA (2-sided).

- The IMT of the internal carotid artery (ICA) was reduced only under Atorvastatin alone whereas the IMT of the carotid bulbus (CB) was reduced in both groups with more favorable results for Pio+Statin:

Parameter [Unit]	**PIO + Atorvastatin, n=68** Mean Change ± SD (Median; n)	**Atorvastatin + Placebo, n=80** Mean Change ± SD (Median; n)	**p-value**
IMT ICA [mm]	0.006 ± 0.138 (0.005; n=36)	-0.050 ± 0.176 (-0.049; n=49)	0.5513
IMT CB [mm]	-0.049 ± 0.214 (-0.039; n=64)	-0.022 ± 0.264 (-0.036; n=74)	0.6641

- As for inflammation and vascular function hsCRP, MCP-1, MMP-9, sICAM-1, p-selectin and t-PA revealed more reduction under Pio+Statin. No clear effects were seen for IL-6, sCD40L and sVCAM-1:

Parameter [Unit]	**PIO + Atorvastatin, n=68** Mean Change ± SD (Median; n)	**Atorvastatin + Placebo, n=80** Mean Change ± SD (Median; n)	**p-value**
hs-CRP [mg/l]	-1.93 ± 5.51 (-0.89; n=65)	-1.20 ± 2.19 (-0.46; n= 79)	0.6339
hs-CRP*) [mg/l]	-1.09 ± 1.30 (-0.80; n= 57)	-0.91 ± 1.34 (-0.45; n= 76)	0.1407
IL-6 [pg/ml]	0.26 ± 2.08 (0.00; n= 65)	-0.03 ± 0.19 (0.00; n= 79)	0.6176
MCP-1 [pg/ml]	-22.2 ± 94.4 (-13.5; n= 65)	-2.8 ± 82.3 (3.4; n= 79)	0.1263
MMP-9 [ng/ml]	-11.8 ± 108.6 (-21.9; n= 65)	13.3 ± 162.2 (17.0; n= 79)	0.3657
sCD40L [pg/ml]	-243 ± 2835 (-409; n= 65)	-392 ± 2828 (-374; n= 79)	0.4566
sICAM-1 [ng/ml]	-0.5 ± 26.7 (-0.5; n= 65)	5.8 ± 25.2 (1.3; n= 79)	0.2351
sVCAM-1 [ng/ml]	48.4 ± 94.7 (50.5; n= 65)	6.4 ± 99.8 (15.8; n= 79)	0.0105
P-selectin [ng/ml]	-5.6 ± 14.9 (-6.0; n= 65)	-1.5 ± 15.6 (-1.8; n= 79)	0.1347
t-PA [ng/ml]	-2.80 ± 3.25 (-2.56; n= 65)	-0.04 ± 3.23 (0.00; n= 79)	<.0001
*): values > 9.9 mg/l eliminated			

- Glucose tolerance and insulin sensitivity were more improved under Pio+Statin for glucose, insulin, intact proinsulin, adiponectin and HOMA-S. No clear effects were seen for HbA1C and HOMA-%B:

Parameter [Unit]	**PIO + Atorvastatin, n=68** Mean Change ± SD (Median; n)	**Atorvastatin + Placebo, n=80** Mean Change ± SD (Median; n)	**p-value**
Glucose [mg/dl]	-2.9 ± 8.9 (-2.3; n= 65)	-0.0 ± 7.9 (0.2; n= 79)	0.0239
HbA_{1C} [%]	0.01 ± 0.12 (0.00; n= 65)	0.09 ± 0.16 (0.10; n= 79)	0.0003
Insulin [µU/ml]	-1.9 ± 7.9 (-1.0; n= 65)	0.1 ± 6.5 (0.7; n= 79)	0.0829
Proinsulin Intact [pmol/l]	-1.0 ± 4.6 (-0.6; n= 65)	-0.5 ± 4.3 (-0.0; n= 79)	0.2231
Adiponectin [µU/ml]	16.3 ± 17.5 (9.9; n= 65)	-0.6 ± 4.5 (-0.5; n= 79)	<.0001
HOMA-S	-0.5 ± 2.2 (-0.3; n= 65)	-0.0 ± 2.1 (0.1; n= 79)	0.1113
HOMA-%B	0.1 ± 131.1 (-6.5; n= 65)	2.0 ± 76.0 (5.7; n= 79)	0.9042

- Concerning lipid metabolism greater effects of Pio+Statin were seen for triglycerides (more decrease) and HDL (more increase) whereas cholesterol and LDL were more reduced under Atorvastatin alone:

Parameter [Unit]	**PIO + Atorvastatin, n=68** Mean Change ± SD (Median; n)	**Atorvastatin + Placebo, n=80** Mean Change ± SD (Median; n)	**p-value**
Cholesterol [mg/dl]	-69.9 ± 40.8 (-69.0; n= 65)	-77.9 ± 40.8 (-82.5; n= 78)	0.8246
HDL [mg/dl]	8.4 ± 11.2 (8.0; n= 65)	3.8 ± 7.8 (5.0; n= 78)	0.0058
LDL [mg/dl]	-65.6 ± 31.9 (-68.0; n= 65)	-73.1 ± 33.5 (-74.5; n= 78)	0.7626
Triglycerides [mg/dl]	-77.5 ± 127.7 (-46.0; n= 65)	-58.5 ± 119.7 (-33.5; n= 78)	<.0001

- In terms of vascular function, measurements of pulse wave velocity (PWV) provided better results for the Atorvastatin-group. For the Laser Doppler Flowmetry (LDF) a consistent trend could not be derived:

Parameter [Unit] analyzed only for study center Mainz	**PIO + Atorvastatin, n=40** Mean Change ± SD (Median; n)	**Atorvastatin + Placebo, n=49** Mean Change ± SD (Median; n)	**p-value**
PWV [mmHg]	-0.3 ± 4.4 (-1.0; n= 40)	-1.5 ± 3.8 (-1.0; n= 48)	0.3475
PWV-Index [%]	-0.9 ± 7.1 (-1.0; n= 40)	-1.7 ± 6.2 (-1.5; n= 48)	0.6414
LDF 37° (arm supine) [AU]	-0.7 ± 10.5 (-2.6; n= 37)	1.6 ± 12.3 (0.8; n= 48)	0.6211
LDF 44°(arm supine) [AU]	-13.9 ± 54.1 (1.3; n= 37)	-6.6 ± 52.9 (-1.4; n= 48)	0.2218
LDF 44°(arm hanging) [AU]	-26.2 ± 87.8 (-29.6; n= 37)	-20.7 ± 90.7 (-25.9; n= 48)	0.3395
LDF abs. change[§)] [AU]	-13.1 ± 52.7 (-11.7; n= 37)	-8.2 ± 48.2 (-3.4; n= 48)	0.2401
LDF rel. change[§)] [%]	-92.2 ± 398.9 (-116.1; n= 37)	-74.8 ± 401.4 (-161.1; n= 48)	0.2583
LDF before ACh [AU]	-8.2 ± 42.7 (-3.1; n= 36)	0.2 ± 41.5 (-0.6; n= 42)	0.8575
LDF after ACh [AU]	-7.3 ± 73.6 (-17.4; n= 36)	-1.8 ± 91.9 (7.7; n= 42)	0.8387
LDF abs. change[$)] [AU]	0.9 ± 77.6 (-18.9; n= 36)	-2.0 ± 92.9 (11.9; n= 42)	0.9602
LDF rel. change[$)] [%]	215.3 ± 982.3 (-2.4; n= 36)	10.4 ± 507.6 (97.0; n= 42)	0.6106
ACh: Acetylcholine; [§)]: comp. 37°vs. 44°supine; [$)]: comp. before vs. after Ach; AU: arbitrary units			

<u>Per-protocol analysis (pp-set; n=137 (62 vs. 75)):</u>

The corresponding statistical analyses of the demographic, primary and secondary efficacy variables for both the per-protocol and center-specific evaluation did not differ relevantly from

the full-analysis results, and therefore supported the confirmatory and exploratory results obtained for the full-analysis set.

Safety Results: (all patient treated set; n=175 (88 vs. 87))

Adverse events were documented in 139/175 (79.4%; 71 vs. 68) treated patients showing 380 (204 vs. 176) individual events classified as treatment emergent adverse events (TEAEs). The most frequently reported events (i.e., in more than 5% of the patients) were nasopharyngitis in 31/175 patients (17.7%; 16 vs. 15 cases), dizziness in 18 patients (10.3%; 9 vs. 9), peripheral edema in 15 patients (8.6%; 10 vs. 5), weight increase in 15 patients (8.6%; 13 vs. 2) and headache in 11 patients (6.3%; 6 vs. 5).

In 9/175 patients (5.1%; 7 vs. 2) a total number of 15 (11 vs. 4) coded signs or symptoms referring to TEAEs were documented as serious adverse events (SAEs) mainly due to hospitalization. Thus, 365 (193 vs. 172) events were non-serious. All SAEs were described as different single episodes mainly characterized as gastrointestinal disorders (3 patients; 1 vs. 2) and as infections, injuries and nervous system disorders, respectively (2 patients each). In all cases but one the respective SAEs were rated as unlikely related to study drug administration (possibly related severe dizziness and moderate nausea/vomiting in the Atorvastatin-group). The only SAE classified as non-TEAE was a severe syncope in one patient from the Pioglitazone-group. Premature discontinuation of the study due to an adverse event according to the entries in the appropriate adverse events (AE)-form occurred in 23/175 patients (13.1%; 15 vs. 8 cases) reporting 46 (29 vs. 17) single events. Cases of death did not occur during the entire study period.

The course of the single events were determined as unique for 51 (26 vs. 25), as intermittent for 115 (61 vs. 54) and as continuous for 214 (117 vs. 97) events. Regarding severity 274 (135 vs. 139) events were assessed as mild, 95 (63 vs. 32) as moderate and 11 (6 vs. 5) as severe. Relationship to study drug administration was rated as unlikely/not related in 246 (118 vs. 128), as possibly related in 90 (58 vs. 32) and as probably related in 44 (28 vs. 16) single events. The vast majority of events were classified as recovered during study (343; 179 vs. 164) whereas 30 (21 vs. 9) events did not yet recover at study end or recovered with sequelae (6; 4 vs. 2).

Regarding laboratory results a clear trend towards a study therapy related influence on specific parameters can not be derived from the sum of changes assessed as clinically significant by the investigators. Among the evaluation of vital signs clinically relevant changes both during the study course and between the treatment groups did not occur. Nevertheless, obvious changes during study treatment (i.e., V1-V5) were seen for systolic and diastolic blood pressure showing a slight consistent decrease equally in both treatment groups and for body weight with a slight increase only under treatment with Pioglitazone.

Overall Conclusions:

The addition of Pioglitazone to Atorvastatin in this study failed to statistically confirm a superior efficacy of a 24-week treatment compared to Atorvastatin alone regarding the possible potential for a reduction of intima-media thickness (IMT) in non-diabetic patients at risk for vascular complications. However, the combination offers clearly positive results in terms of several established clinical and laboratory markers for cardiovascular diseases and risk factors. Obvious beneficial influences in the sense of multiple pleiotrophic effects were seen for various parameters of inflammation and vascular function (hsCRP, MCP-1, MMP-9, sICAM-1, p-selectin and t-PA), glucose tolerance and insulin sensitivity (glucose, insulin, intact proinsulin, adiponectin and HOMA-S) and in parts for the microvascular function (LDFAch).

In terms of safety issues the study did not reveal any potential unknown risks or unexpected or new signs and symptoms allocated to the study drugs in comparison to the known range of thiazolidinedione- and/or statin-specific adverse reactions. Observations like dizziness, peripheral edema, weight increase, headache, nausea, dyspnoea, fatigue, myalgia and gastrointestinal problems are consistent with the expected safety profile of the used study drugs. For the evaluation of laboratory tests and vital signs a clear trend towards a study therapy related pathologic influence on specific parameters can not be derived.

Significant Changes During Study:

There were no official changes to the study protocol or the conduct of the clinical trial. There was one modification in the final statistical analysis plan which enhanced the definition of the full analysis set with the addition of specific measurement of $IMT_{CCA} \geq 0.8$ mm (at least on one side) at baseline and at least one measurement post baseline.

Study ID Number:

ATS K015

Other Study ID Number(s):

2004-004463-30 [EudraCT Number]

D-PIO-106 [Takeda ID]

U1111-1115-9124 [Registry ID: WHO]

DATE OF DISCLOSURE SYNOPSIS: 13 June 2012