The study listed may include approved and non-approved uses, formulations, or treatment regimens. The results reported in any single study may not reflect the overall results obtained on studies of a product. Before prescribing any product mentioned in this Register, healthcare professionals should consult prescribing information for the product approved in their country.

Study No.: Study A4M108119

Title: Study A4M108119: An open-label study of leukocyte counts in the cerebrospinal fluid and blood of subjects with relapsing forms of multiple sclerosis following treatment with firategrast

Rationale: Firategrast is an orally bioavailable selective inhibitor of $\alpha 4\beta 1$ integrins expected to prevent the entry of lymphocytes, including autoreactive lymphocytes, into the central nervous system (CNS), reducing the formation of inflammatory lesions and the frequency of relapses in multiple sclerosis (MS).

Natalizumab, a monoclonal antibody that binds to $\alpha 4\beta 1$ increases the risk of progressive multifocal leukoencephalopathy (PML), an opportunistic infection of the brain caused by the JC virus. Prior to the design of Study A4M108119, it had been demonstrated that natalizumab treatment was associated with significantly lower total leukocyte, Cluster of Differentiation (CD) 4+ and CD8+ T-lymphocyte, CD19+ B-lymphocyte and CD138+ plasma cell counts in cerebrospinal fluid (CSF) compared with patients with other non-inflammatory neurological diseases and MS patients who were not treated with natalizumab. These low counts persisted at 6 months after discontinuation of natalizumab [Stuve, 2006b]. Furthermore, it was observed in the natalizumab-treated patients that there was a decrease in the CSF CD4:CD8 ratio to levels similar to those seen in human immunodeficiency virus-infected patients, a population known to be at increased risk for PML [Stuve, 2006a].

Firategrast acts at the same target as natalizumab and was expected to also reduce lymphocyte and lymphocyte subset counts in the CSF/CNS. However, in the peripheral circulation, the expected elevation in lymphocytes seen during firategrast treatment is reversed within 24 hours of cessation of treatment. This suggests that a reduction of lymphocyte counts in the CSF/CNS produced by firategrast might be more rapidly reversible than has been shown for natalizumab. In the setting of CNS opportunistic infection, this could be of benefit to the patient. Therefore the primary purposes of this study were to determine the extent to which firategrast alters lymphocyte and lymphocyte subset counts in the CSF and, if it lowers them significantly, to determine the extent of their recovery within a specified period of time.

Phase: II

Study Period:18 July 2007 to 12 February 2010 (Last Extended Follow-up Phase Visit)

Study Design: This was a multicenter, open-label, single-group study in subjects with relapsing forms of MS.

Centres: 10 centers in 5 countries (Belgium [2], Czech Republic [2], Denmark [2], Norway [1], Sweden [3])

Indication: Relapsing Remitting Multiple Sclerosis (RMSS)

Treatment: Firategrast was provided as white, round, film coated tablets at a strength of 300 mg. Female subjects took 900 mg (3 tablets) and male subjects took 1200 mg (4 tablets) by mouth twice daily for up to 24 weeks.

Objectives: The most important objectives of this study were to determine: the number and change in number of total leukocytes, total lymphocytes and lymphocyte subsets during and after treatment with firategrast and the CD4:CD8 ratio and change in the CD4:CD8 ratio in the CSF and blood during and after treatment with firategrast.

Other objectives included: to examine blood for mobilization of CD34+ early hematopoietic progenitor cells from the bone marrow during and after treatment with firategrast, to explore the relationship between changes in the number of leukocytes in the CSF and blood, to explore the relationship between systemic exposure to firategrast and the metabolite, GW786375X, and changes in leukocyte counts in the CSF and blood, to determine firategrast and GW786375X levels in CSF, to explore potential relationships between genetic variants and response to firategrast, pharmacokinetic endpoints and bilirubin levels and to investigate the safety and tolerability of up to six months' administration of firategrast.

Primary Outcome/Efficacy Variable: Flow Cytometry: For cell counts and the CD4:CD8 ratio, group summary statistics were evaluated at the end of treatment for the magnitude of change relative to baseline. They were evaluated at follow-up time points for the magnitude of change relative to both baseline and end of treatment.

Secondary Outcome/Efficacy Variable: MRI Efficacy: Exploration of MRI Efficacy was primarily by evaluation of cumulative number of new gadolinium-enhancing lesions on monthly MRI brain scans during the 24-week Treatment Phase. Additional assessments included volume of new, number of persistent and total number of gadolinium-enhancing lesions.

Statistical Methods The basis for this study's sample size of 45 subjects was partly feasibility, and partly assessment of data collected on leukocyte counts approximately six months after discontinuation of treatment with natalizumab. Subjects that received natalizumab had an estimated mean decrease in their CSF white blood cell (WBC) counts of 10,771 cells/milliliter (mL) with a standard deviation of 8,458 cells/mL. Assuming similar results for firategrast, a sample size of 30 evaluable subjects would provide a 95% confidence interval width of approximately 6,200 cells/mL for the mean change in WBC counts in the CSF following 24 weeks of treatment. Note that Stuve reported results for CSF cell counts in cells/mL. Reporting of all results for cell counts in Study A4M108119 use units of cells/microliter (μL).

The Randomized population consisted of all subjects who were "randomized" to receive firategrast and entered the study at Week 0.
The Intent-to-Treat (ITT) population consisted of all subjects who took at least one dose of firategrast and who had at least one post-baseline flow cytometry analysis assessment. This is the primary population used for the cell count endpoints.

The Safety population consisted of all subjects who were "randomized" and took at least one dose of firategrast. This was the primary population used for the safety analysis.

The PGx population consisted of the subset of subjects in the randomized population who provided written informed consent for PGx research. In addition, the subset consists of subjects who provided a blood sample for genotyping, were successfully genotyped for at least one of the genetic markers under study, and who had valid clinical data that passed quality control.

The PK/PD population consisted of all subjects in the ITT population who had both a PK and a PD measurement.
No formal statistical hypothesis testing was performed. For flow cytometry parameters the number of subjects, mean, standard deviation, median, minimum and maximum were determined along with 95% confidence intervals for the within-group mean for each visit, change from baseline and change from end of treatment, as appropriate.

Study Population: Male or female subjects age 18 to 65 inclusive, with a diagnosis of a relapsing form of MS with dissemination in time and space were eligible.

Subjects were required to have an Expanded Disability Status Scale (EDSS) score of between 0 and 6.5 inclusive at the Screening visit, occurrence of at least one relapse in the previous 24 months and a minimum of two T2 lesions on brain MRI at Screening, as determined by the central MRI reader.

Subjects who had a relapse within 4 weeks prior to Screening were excluded, along with subjects who had received specific therapies affecting the immune system or for treatment of MS within specified periods of the Screening visit. Those subjects with a CD4 count <500 cells/cubic millimeter, CD4:CD8 <1.0, JC viremia detected in plasma or white cells, idiopathic CD4 or CD8 lymphopenia or secondary lymphopenia at Screening were also excluded.

	Firategrast 900 mg N=30	**Firategrast 1200 mg N=16**	**Total N=46**
Number of Subjects:			
Planned, N	30	15	45
Randomised, N		16	46
Completed, n (%)			
Completed to Week 24	25 (83)	15 (94)	40 (87)
Completed to Week 36	23 (77)	15 (94)	38 (83)
Total Number Subjects Withdrawn through Week 36, n (%)	7 (23)	1 (6)	8 (17)
Withdrawn due to Adverse Events through Week 36, n (%)	3 (10)	1 (6)	4 (9)
Withdrawn due to Lack of Efficacy through Week 36, n (%)	0	0	0
Withdrawn for other reasons through Week 36, n (%)	4 (13)	0	4 (9)
Entered Extended Follow-Up	27/30 (90%)	16/16 (100%)	43/46 (93%)

Completed Extended Follow-Up (having entered Extended Follow-Up)			27/27 (100%)	16/16 (100%)	43/43 (100%)
Total Number of Withdrawn during Extended Follow-Up			0	0	0
Demographics			**Firategrast 900 mg N=30**	**Firategrast 1200 mg N=16**	**Total N=46**
N (ITT)				16	46
Females: Males				0:16	30:16
Mean Age, years (SD)				39.1 (9.66)	39.5 (10.09)
White, n (%)				15 (94)	45 (98)
Primary Efficacy Results: **Median Change in Total Lymphocyte Count in Cerebrospinal Fluid by Visit**					
Visit:		**Baseline**	**Week 24**	**Week 28**	**Week 36**
Count at Visit (cells/μL)	n Median Range	44 5.26 0.3-70.2	31 3.25 0.0-99.0	32 2.96 0.0-58.2	29 3.52 0.0-274.8
Median change from Baseline	n Median Range	NA	31 -1.08 -10.5-70.5	30 0.21 -13.7-30.4	27 -0.72 -10.9-259.0
Median change from Week 24	n Median Range	NA	NA	27 0.02 -47.016.9	26 0.57 -75.2-188.5
Median Change and Mean CD4:CD8 Ratio in Cerebrospinal Fluid by Visit					
Visit:		**Baseline**	**Week 24**	**Week 28**	**Week 36**
Median Ratio:	n Median Range	41 2.945 1.10-10.87	29 2.248 0.64-5.95	28 3.797 1.62-9.01	21 3.804 2.11-9.43
Median change from Baseline:	n Median Range	NA	28 -0.697 -6.76-2.02	25 0.372 -3.01-2.57	19 0.527 -2.02-2.20
Median change from Week 24:	n Median Range	NA	NA	23 1.057 -0.54-4.35	19 1.578 -0.70-3.45
Mean Ratio	n Mean SD	41 3.608 2.1225	29 2.773 1.5007	28 3.907 1.6064	21 4.213 1.8859
Median Change in Total Lymphocyte Count in Peripheral Blood by Visit					
Visit:		**Baseline**	**Week 4**	**Week 24**	**Week 28**	**Week 36**
Count at Visit (cells/μL):	n Median Range	43 **1513.18** 593.7-3642.5	41 **2093.24** 1238.1-5129.2	32 **2046.07** 1260.9-4145.0	35 **1419.88** 982.1-2376.4	30 **1592.39** 756.5-2918.5
Median change from Baseline:	n Median Range	NA	38 **615.60** -1430.2-1844.6	30 **603.55** -414.9-1935.1	33 **15.14** -1577.8-1410.8	29 **142.20** -1218.0-741.4
Median change from Week 24:	n Median Range	NA	NA	NA	28 **-561.04** -2672.6-341.6	26 **-679.06** -2412.0-314.1
Median Change CD4:CD8 Ratio in Peripheral Blood by Visit						

Visit:		Baseline	Week 4	Week 24	Week 28	Week 36
Median Ratio:	n Median Range	42 2.294 1.30-7.62	41 2.230 1.20-13.95	32 2.120 1.09-6.61	35 2.550 1.30-13.32	30 2.338 0.82-7.44
Median change from Baseline:	n Median Range	NA	37 -0.056 -1.30-12.04	30 -0.122 -1.40-0.32	32 0.174 -1.04-10.94	29 0.014 -1.22-0.73
Median change from Week 24:	n Median Range	NA	NA	NA	28 0.342 -0.14-10.81	26 0.218 -0.95-0.98

Secondary Outcome Variables at Week 24:

		Week 24
Number of New Gadolinium-Enhancing Lesions	n Mean Range	46 2.8 0-30
Cumulative Volume of New Gadolinium-Enhancing Lesions	n Mean Range	46 405.7 0-4264
Cumulative Number of Persistent Gadolinium-Enhancing Lesions	n Mean Range	46 0.8 0-12
Cumulative Total Number of Gadolinium-Enhancing Lesions	n Mean Range	46 3.6 0-42

If the AE or SAE onset date is any date from the date of initiation of investigational product to the date investigational product was permanently discontinued + 3 days (inclusively) then it was considered 'On-treatment'.

	Firategrast 900 mg N=30	Firategrast 1200 mg N=16	Total N=46
Most Frequent Adverse Events – On-Therapy	**n (%)**	**n (%)**	
Subjects with any AE(s), n(%)	27 (90)	13 (81)	40 (87)
Headache	15 (50)	3 (19)	18 (39)
Nasopharyngitis	6 (20)	7 (44)	13 (28)
Nausea	7 (23)	3 (19)	10 (22)
Fatigue	6 (20)	1 (6)	7 (15)
Back pain	5 (17)	1 (6)	6 (13)
Urinary Tract Infection	5 (17)	0	5 (11)
Viral Infection	4 (13)	1 (6)	5 (11)
Gastroenteritis	3 (10)	1 (6)	4 (9)
Upper Respiratory Tract Infection	3 (10)	1 (6)	4 (9)
Cough	2 (7)	1 (6)	3 (7)
Depression	2 (7)	1 (6)	3 (7)
Diarrhea	1 (3)	2 (13)	3 (7)
Oropharyngeal Pain	3 (10)	0	3 (7)
Post lumbar puncture syndrome	2 (7)	1 (6)	3 (7)
Pyrexia	3 (10)	0	3 (7)
Oral herpes	2 (7)	0	2 (4)
Pharyngitis	2 (7)	0	2 (4)
Rhinitis	0	2 (13)	2 (4)
Sinusitis	1 (3)	1 (6)	2 (4)
Viral upper respiratory tract infection	2 (7)	0	2 (4)
Dizziness	2 (7)	0	2 (4)
Abdominal pain upper	2 (7)	0	2 (4)
Dyspepsia	2 (7)	0	2 (4)
Sleep disorder	1 (3)	1 (6)	2 (4)

Eczema	0	2 (13)	2 (4)
Lymphadenopathy	1 (3)	1 (6)	2 (4)

Serious Adverse Events - On-Therapy
n (%) [n considered by the investigator to be related to study medication]

	Firategrast 900 mg bid (N=30)	**Firategrast 1200 mg bid (N=16)**	**Total (N=46)**
Subjects with non-fatal SAEs, n (%)	**n (%) [related]**	**n (%) [related]**	**n (%) [related]**
ANY EVENT	4 (13) [0]	0	4 (9) [0]
Gastroenteritis	1 (3) [1]	0	1 (2) [1]
Urinary Tract Infection	1 (3) [0]	0	1 (2) [0]
Hip Fracture	1 (3) [0]	0	1 (2) [0]
Alanine aminotransferase increased	1 (3) {1]	0	1 (2) [1]
Back Pain	1 (3) [0]	0	1 (2) [0]
Radicular Syndrome	1 (3) [0]	0	1 (2) [0]
Subjects with fatal SAEs, n (%)	**n (%) [related]**	**n (%) [related]**	**n (%) [related]**
Any death	0	0	0

Conclusion: In this open-label study, subjects with relapsing forms of MS and in subjects with baseline total leukocyte counts in the CSF of $\leq$15 cells/μL, treatment with firategrast for up to 24 weeks appears to be associated with a decrease in CSF total lymphocytes, CD4+ and CD8+ T-lymphocytes and CD19+ B-lymphocytes in most subjects. This decrease is small, and may mean that sufficient numbers of lymphocytes can access the subarachnoid space where they have an important role in CNS immune surveillance. Treatment with firategrast for up to 24 weeks does not appear to be associated with a substantial alteration in the CD4:CD8 ratio in the CSF or peripheral blood. . A total of 40 (87%) of all patients reported a treatment-emergent AE. Most commonly, patients reported AEs categorized as "infections and infestations." A total of 27 (59%) patients reported at least 1 infection/infestation. Four (13%) patients in the 900mg group reported an SAE while 0 patients in the 1200mg group reported SAEs. There were no deaths reported in this study. There was no evidence of renal abnormalities, reactivation of JC virus or PML in follow-up to one year after the last dose of firategrast.

Publications: None