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The results of two multicenter, open-label studies assessing efficacy, tolerability and safety of protiramer, a high molecular weight synthetic copolymeric mixture, in patients with relapsing–remitting multiple sclerosis

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Objective Two pilot studies were conducted to evaluate safety, tolerability, and efficacy of two doses of Protiramer (TV-5010) in patients with relapsing–remitting multiple sclerosis.

Background Both glatiramer acetate and TV-5010 are synthetic copolymers comprised the same four amino acids in a defined molar ratio. TV-5010 has higher average molecular weight than Glatiramer acetate and might be hypothesized that glatiramoids with higher molecular weight might be more immunoreactive than lower molecular weight peptides, thus increasing therapeutic potential and allowing for less frequent dosing.

Methods In the two separate studies, after a 10 week pretreatment period, TV-5010 was given subcutaneously once weekly at 15 mg and 30 mg for 36 weeks. The primary end point was a reduction in the number of magnetic resonance imaging active lesions (i.e., T1-weigthed gadolinium-enhancing and new T2-weighted lesions) between the pretreatment period and the end of study.

Results Both TV-5010 doses were generally well tolerated. The treatment with TV-5010 at a dose of 15 mg/wk did not show any significant effect. In contrast, in patients treated with at a dose of 30 mg/wk, a significant reduction in the mean number of gadolinium-enhancing (–58.8%; $P = 0.0013$) and new T2-W (–50%; $P = 0.0002$) lesions was observed. However, a large decrease in the mean number of both gadolinium-enhancing (–55%) and new T2-W (–40%) lesions during the pretreatment period made difficult the interpretation of the efficacy assessments.

Conclusions Further studies are needed to confirm these preliminary data on safety and efficacy of TV-5010 at a weekly dose of 30 mg. *Multiple Sclerosis* 2009; **15:** 238–243. http://msj.sagepub.com

Key words: glatiramer acetate; MRI; multiple sclerosis; protiramer; TV-5010

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Introduction

Glatiramer acetate (GA; Copaxone®), which is currently used for the treatment of patients with relapsing–remitting (RR) multiple sclerosis (MS), is a member of the glatiramoid class of compounds. Glatiramoids are a family of synthetic heterogeneous polypeptides mixtures comprising four natural amino acids: L-glutamic acid, L-alanine, L-lysine, and L-tyrosine, in a defined molar ratio. Previous data have shown that GA has an immunomodulatory and, likely, a neuroprotective effect in MS [1,2].

Previous studies in patients with RRMS [3,4] used a substance similar to GA, but with a slightly higher average molecular weight (MW, average 14,000–23,000 Da) than the currently approved formulation of GA (5,000–9,000 Da).

It might be hypothesized that glatiramoids with higher MW might be more immunoreactive than lower MW peptides, thus increasing therapeutic potential and allowing for less frequent dosing (GA is administered once daily). Protiramer (TV-5010), another glatiramoid, is produced (Teva Pharmaceutical Industries Ltd) by making slight changes to the GA manufacturing process. This is a synthetic high MW polypeptide with the same molar ratio as GA and with the average MW of 13,500–18,500 Da. The two molecules, despite some overlap in peptide size distribution, are different in the primary (sequence) and secondary structure of the copolymers.

Two parallel pilot studies were conducted to evaluate the safety, tolerability, and efficacy of TV-5010 in patients with RRMS on magnetic resonance imaging (MRI) disease activity. The first pilot study evaluated subcutaneous (SC) treatment with 15 mg of TV-5010 once weekly and the second one evaluated treatment with 30 mg SC of TV-5010 weekly once. Both the studies were conducted between May 2004 and February 2006.

Methods

Material

Both the protiramer active substance and the finished product were produced by Teva Pharmaceutical Industries Ltd, Kfar Sava, Israel, at a fully GMP manufacturing sites.

The investigational medicinal product was supplied in a single-use prefilled syringe containing 1.0 mL of a clear solution of 15 or 30 mg protiramer and mannitol as an excipient.

Study design and outcome measures

The 46-week study included a 10-week pretreatment phase followed by a 36-week treatment phase. Patients self-administered weekly SC injections of TV-5010 at a dose of 15 mg (Study 201) or 30 mg (Study 202) and underwent 10 scheduled visits: weeks –10 (screening), –6, 0 (baseline), one week post baseline (visit 1), and at weeks 4, 12, 24, 28, 32, and 36 (termination). Conventional MRI examinations of the brain were acquired in each participating subject at weeks –10, –6, 0, 12, 28, 32, and 36.

Eligible patients had clinically definite RRMS (according to Poser criteria [5]), Expanded Disability Status Scale (EDSS) scores [6] between 0–5 inclusive, experienced at least one relapse in the year before study screening, were relapse-free and steroid-free in the 30 days before screening, and had at least one T1-weighted (T1-W) gadolinium (Gd)-enhancing lesion on one pretreatment MRI scans, which were taken at weeks –10, –6, and 0 (baseline).

Safety and tolerability were based on adverse events (AEs), clinical laboratory tests, vital signs, physical examinations, and ECG measurements. Humoral response to TV-5010 was measured by antibody testing at three sera dilutions: 1/70, 1/700, and 1/7000.

The primary end point was a comparison of the number of T1-W Gd-enhancing lesions present during the 10-week screening period (week –10, –6, and 0) with the number present during the third trimester of the study, weeks 28, 32, and 36. Secondary end points included i) a comparison of the number of new MRI T2-weighted (T2-W) lesions present at the pretreatment period (at weeks –6 and 0 vs week –10) with the number present in the last two treatment visits (at weeks 32 and 36 vs week 28); ii) numbers of on-study relapses, and iii) changes in EDSS scores.

MRI examination and analysis

The Quantitative Neuroimaging Laboratory (Department of Neurological and Behavioral Sciences, University of Siena, Siena, Italy) served as the magnetic resonance image analysis center (MRI-AC) for the trial. Before any clinical site could enroll, study participants were required to image a volunteer patient with clinically definite MS twice, with repositioning according to a strict study imaging protocol using imagers with minimum field strength of 1.0 Tesla. These test images were sent to the MRI-AC as film and electronic data for review to ensure that the site could perform high-quality imaging; 15 MRI sites were approved.

A series of axial, coronal, and sagittal images were obtained to create an axial reference scan for the subsequent careful repositioning of each patient at the follow-up sessions. At each time point, dual-echo, fast spin-echo sequences (TR: 2200–3000, TE: 30–50/60–100, 3 mm slice thickness and 44

contiguous axial slices) yielding proton density-weighted and T2-W) images were acquired. Conventional T1-W images (TR: 600–650, TE: 10–20, 3 mm slice thickness, and 44 axial slices) were also obtained at each time point 5 min after the injection of 0·1 mmol/kg of Gd chelates. Image quality was reviewed centrally according to predetermined criteria. Identification of enhancing lesions, high signal intensity lesions on T2-W images, and hypointense lesions on T1-W enhanced images was done by consensus of two experienced observers. Trained personnel then outlined the lesions using a semi-automated segmentation technique based on local thresholding [7].

Statistical analyses

Each pilot study was planned to include 25 subjects. Using a 2-sided α level of 5%, with the underlying assumption that the change from baseline to termination in the total number of T1 Gd-enhancing lesions would be ≥40%, yielded a power of over 95%.

Safety was assessed in all patients who received ≥1 dose of study drug. To be included in the primary efficacy analysis, patients must have had at least one MRI scan during the last study trimester (weeks 28, 32, or 36). Similarly, to be evaluated for change in number of new T2-W lesions, patients must have undergone MRI at week 32 or 36.

MRI analyses were based on repeated measures Poisson regression with baseline mean number of T1-W Gd-enhancing lesions (or number of new T2-W lesions) as a covariate. Numbers of relapses in the year before study entry, numbers of on-study relapses, and changes in EDSS scores are presented descriptively.

Results

Patient demographics

Patient demographics at baseline are shown in Table 1. A total of 52 patients were screened for each study.

In all, 38 patients were enrolled in the 15 mg/wk study. Mean treatment duration was 247 ± 27.5 days. Overall, 95% patients administered ≥33 TV-5010 doses.

A total 27 patients were enrolled in the 30 mg/wk TV-5010 study. Mean treatment duration was 237 ± 57.3 days. Overall, 89% of patients administered ≥33 TV-5010 doses.

Table 1 Patient demographics at baseline

Baseline Characteristics	TV-5010 15 mg/wk Study 201 (*N* = 38)	TV-5010 30 mg/wk Study 202 (*N* = 27)
Sex		
Female; *n* (%)	29 (76.3)	18 (66.7)
Male; *n* (%)	9 (23.7)	9 (33.3)
Age (years) mean (SD)	33.8 (7.9)	33.6 (8.2)
MS duration; (years) mean (SD)	4.6 (5.3)	2.4 (3.3)
EDSS Score; mean (SD)	2.1 (1.4)	2.0 (1.5)
Prior 1-year relapse rate; mean (SD)	1.4 (0.6)	1.6 (0.7)
Number of T1-enhancing lesions[a]; mean (SD)	3.5 (4.6)	1.3 (1.3)

MS, multiple sclerosis; EDSS, Expanded Disability Status Scale.
[a]Measured at week 0.

Efficacy

MRI data

Changes in numbers of T1 Gd-enhancing lesions and new T2-W lesions during treatment periods of both studies are shown in Table 2.

MRI changes in Study 201

One patient was ineligible for the primary end point analysis and two patients were ineligible for the secondary end point analysis.

After treatment with 15 mg/wk TV-5010, there was no significant decrease in the number of T1-W Gd-enhancing lesions (–30.1%; $P = 0.0616$). Similarly, there were no significant changes in number of new T2-W lesions after the treatment period (mean change –0.1 ± 1.2, –9%; $P = 0.7471$).

MRI changes in Study 202

Two patients were ineligible for the primary end point analysis and three patients were ineligible for the secondary end point analysis.

In this group, the mean number of Gd-enhancing lesions dropped sharply during the pretreatment period: 2.9 ± 6.0 Gd-enhancing lesions at week –10 versus 1.3 ± 1.3 at week 0 (–55%) (Figure 1). During the treatment period, the mean number of T1-W Gd-enhancing lesions decreased significantly from the mean number during pretreatment periods (mean change –1.2 ± 2.8, –58.8%; $P = 0.0013$).

As data from a single patient with an unusually high number of Gd-enhancing lesions, which decreased markedly by week 0, could have biased this result, the analysis was performed without this patient's data. The marked decrease in these lesions during the pretreatment period, however, did not alter the final results (T1-W Gd-enhancing lesions decreased of 45.4%, $P = 0.0012$) (Table 2).

Table 2 Number of T1 Gd-enhancing and new T2-W lesions during study

	15 mg/wk TV-5010 Study 201 *N* = 37[a]	30 mg/wk TV-5010 Study 202 *N* = 24[b]
Number of T1 Gd-enhancing lesions (pretreatment)		
Mean ± SD	3.3 ± 3.3	1.4 ± 1.1
Median	2.0	1.0
Range (min, max)	0.3, 13.0	0.3, 3.7
Number of T1 Gd-enhancing lesions (third trimester)		
Mean ± SD	2.3 ± 2.9	0.7 ± 0.9
Median	0.7	0.3
Range	0, 12.3	0, 2.7
Change in number of T1 Gd-enhancing lesions between pretreatment-treatment periods		
Mean ± SD	−1.0 ± 3.2	−0.7 ± 0.9
Median	−0.7	−0.7
Range	−12.7, 7.3	−3.0, 0.7
P value[c]	0.0616	0.0012
Number of new T2-W lesions (pretreatment)		
Mean ± SD	0.7 ± 1.0	0.8 ± 1.1
Median	0.5	0.5
Range (min, max)	0,4.5	0,4.0
Number of new T2-W lesions (third Trimester)		
Mean ± SD	0.6 ± 0.9	0.4 ± 0.5
Median	0.0	0.0
Range	0, 3.5	0, 1.5
Change in number of new T2-W lesions between pretreatment-treatment periods		
Mean ± SD	−0.1 ± 1.2	−0.4 ± 0.8
Median	0.0	0.0
Range (min, max)	−4.5, 3.5	−3.0, 0.5
P value	0.7471	0.0002

[a]Excludes one patient missing third trimester MRI evaluations.
[b]Excludes two patients missing third trimester MRI evaluations and one patient whose pretreatment number of Gd-enhancing lesions was a statistical outlier.
[c]Principal analysis was based on repeated measures Poisson Regression with baseline mean number of Gd-enhancing lesions as a covariate.

Similarly, there was a decrease of 40% in new T2-W lesions during the pretreatment period (Figure 2). Also, the number of new T2-W lesions were significantly decreased after the treatment period (mean change −0.4 ± 0.8, −50%; $P = 0.0002$) (Table 2 and Figure 2).

Clinical data

The majority of patients remained relapse free during both studies: 73.7% with 15 mg/wk and 74.1% with 30 mg/wk TV-5010. Proportions of patients who experienced one on-study relapse were 23.7% and 22.2% and one patient in each study experienced three confirmed relapses. There was no change in screening EDSS score with either dose of TV-5010.

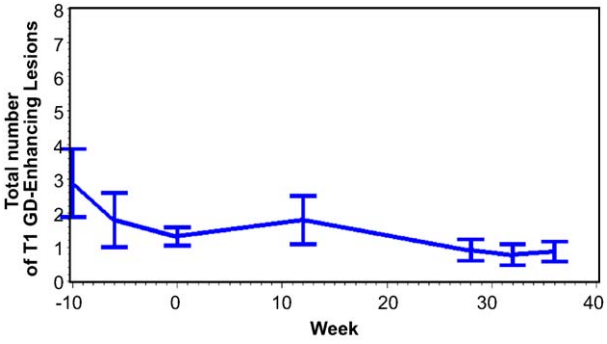

Figure 1 Changes of the total number of T1-weighted gadolinium-enhancing lesions over time (Study 202).

Safety

The most common AEs (Table 3) in both studies were injection-site reactions and symptoms consistent with immediate post-injection reaction (IPIR). Erythema, pain, and induration were the most common injection-site reactions; there were no reports of skin necrosis. Laboratory measures, vital signs, and ECG findings remained within normal limits at most testing intervals. No deaths occurred during either study.

Three patients withdrew prematurely from the 15 mg/wk study, one withdrew consent, and two due to AEs (symptoms consistent with IPIR and mild hypersensitivity reaction, both were considered possibly related to study drug).

Four patients withdrew from the 30 mg/wk study, all due to AEs (idiopathic thrombocytopenic purpura, pneumothorax, hypersensitivity reaction, and seizure; the latter 2 AEs were considered possibly related to study drug).

All patients developed anti-TV-5010-specific IgG antibodies, which were detectable at 4 weeks, peaked at approximately 3 months, and remained at constant levels over the rest of the 9-month studies. Two patients receiving 15 mg/wk and one patient receiving 30 mg/wk developed anti-TV-5010 IgE antibodies that were slightly above

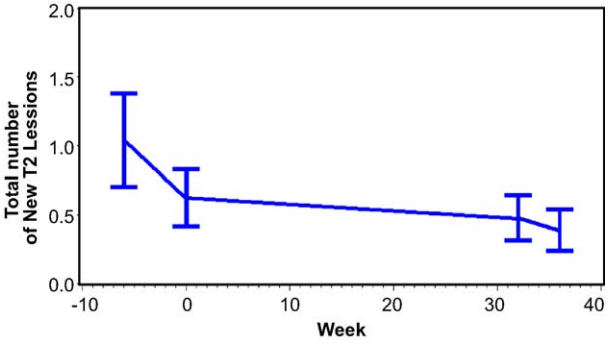

Figure 2 Changes of the total number of new T2-weighted lesions over time (Study 202).

Table 3 Most common (≥10%) adverse events

	15 mg/wk TV-5010 Study 201 *N* = 38, *n* (%)	30 mg/wk TV-5010 Study 202 *N* = 27, *n* (%)
Any adverse event, number of patients reporting (%)	32 (84.2)	27 (100)
Injection-site reactions	19 (50.0)	24 (88.9)
IPIR symptoms[a]	11 (28.9)	6 (22.2)
Upper respiratory tract infection	11 (28.9)	5 (18.5)
Pyrexia	8 (21.1)	4 (14.8)
Headache	7 (18.4)	6 (22.2)
Diarrhea	5 (13.2)	
Nausea	4 (10.5)	
Anxiety	4 (10.5)	

[a]Immediate post-injection reaction; symptoms may have included vasodilation, chest pain, dyspnea, palpitations, and/or tachycardia.

the limit of detection (LOD). One patient had IgE antibodies just above LOD pretreatment and at week 4; no additional samples were obtained (the patient withdrew from the study for reasons unrelated to study drug). Another patient had IgE antibodies just above the LOD at visit 2, but tested negative at all subsequent time points. The third patient had IgE antibodies at week 36 (end of study, follow-up not available). None of these patients has developed allergic hypersensitivity reaction.

Discussion

Efficacy data reported here were limited by the study design (i.e., absence of placebo group). However, they do possibly suggest that short-term treatment with 30 mg/wk TV-5010 of patients with RRMS significantly reduced the number of both T1-W Gd-enhancing and T2-W lesions compared with pretreatment, meeting the primary and secondary end points of the pilot study. In contrast, the 15-mg/wk TV-5010 did not demonstrate a clear beneficial effect. Nevertheless, in interpreting the data, it must be stressed that the large reduction of the number of both Gd-enhancing lesions and new T2 lesions during the pretreatment period may have biased the results. However, Gd-enhancing lesion counts also presented a large further reduction between baseline (week 0) and end of treatment with the 30 mg dose (37.4%, data not shown), suggesting that our cohort of patients with MS might have had a beneficial effect of taking a high dose of TV-5010. Clearly, however this study does not provide unequivocal proof for efficacy, and more data are needed to fully establish whether this product is effective or not.

TV-5010 was generally well tolerated. There was a greater incidence of injection-site reactions with the higher TV-5010 dose; no other dose relationship for AEs was observed. Three patients developed anti-TV-5010 IgE antibodies just above the LOD; however, the significance of these results is questionable (e.g, anti-TV-5010 IgE antibodies were detected in a patient pretreatment) and requires confirmatory testing.

The use of GA has shown to have therapeutic effects on patients with MS [8,9]. It is reasonable, therefore, to conjecture that other copolymeric mixtures comprising the same amino acids as GA, but with different molecular structures, might also have therapeutic properties. However, as small changes in the manufacturing process may lead to significantly different product with different protein structure, it cannot be assumed that minor changes in molecular structures between GA and TV-5010 will produce similar therapeutic effects. Data from this pilot study suggest that short-term treatment of patients with RRMS with TV-5010 30-mg/wk is safe and may have a beneficial effect on brain tissue damage. More studies are required to confirm these preliminary data and determine the long-term safety and effectiveness of TV-5010 in patients with RRMS.

Acknowledgements

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References

1. Blanchette, F, Neuhaus, O. Glatiramer acetate: evidence for a dual mechanism of action. *J Neurol* 2008; **255**(Suppl. 1): 26–36.
2. Ziemssen, T, Schrempf, W. Glatiramer acetate: mechanisms of action in multiple sclerosis. *Int Rev Neurobiol* 2007; **79**: 537–570.
3. Bornstein, MB, Miller, A, Slagle, S, *et al*. A pilot trial of Cop 1 in exacerbating-remitting multiple sclerosis. *N Engl J Med* 1987; **317**: 408–414.
4. Bornstein, MB, Miller, A, Slagle, S, *et al*. Clinical experience with COP-1 in multiple sclerosis. *Neurology* 1988; **38**: 66–69.
5. Poser, CM, Paty, DW, Scheinberg, L, *et al*. New diagnostic criteria for multiple sclerosis: guidelines for research protocols. *Ann Neurol* 1983; **13**: 227–231.
6. Kurtzke, JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). *Neurology* 1983; **33**: 1444–1452.
7. Rovaris, M, Inglese, M, van Schijndel, RA, *et al*. Sensitivity and reproducibility of volume change measurements of different brain portions on magnetic resonance imaging in patients with multiple sclerosis. *J Neurol* 2000; **247**: 960–965.
8. Perumal, J, Filippi, M, Ford, C, *et al*. Glatiramer acetate therapy for multiple sclerosis: a review. *Expert Opin Drug Metab Toxicol* 2006; **2**: 1019–1029.
9. Wolinsky, JS. The use of glatiramer acetate in the treatment of multiple sclerosis. *Adv Neurol* 2006; **98**: 273–292.

Appendix

TV5010 study group

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