SYNOPSIS

Name of The Sponsor / Study Promoter:	Banc de Sang i Teixits. Passeig Taulat 116, PO 08005, Barcelona (Spain) Telephone: +34 93 557 35 00 Fax: + 3493 557 35 03
Finished Product:	XCEL-MC-ALPHA
Name of Active Ingredient:	Autologous adult mesenchymal stem cells from bone marrow expanded and cryopreserved
Title of Study:	Treatment of autologous mesenchymal stem cells derived from bone marrow as a potential therapeutic strategy for the treatment of multiple sclerosis (MS) [Protocol code: XCEL-MS-02; EudraCT: 2012-000734-19]
Principal Investigator / Coordinating Investigator:	Dr. Xavier Montalban. Director del Centre d'Esclerosi Múltiple de Catalunya (CEM-Cat) Hospital Universitari Vall d'Hebron Pg. Vall d'Hebron 119-129, PO 08035, Barcelona (Spain) **Study Centre:** Hospital Universitari Vall d'Hebron Pg. Vall d'Hebron, 119-129 08035 Barcelona
Publication References:	Non applicable
Study Period:	Placebo-Controlled, Crossover Study. Duration: 3 years and 6 moths (initiation date: May 11th, 2015 and completion date November 27th, 2017). The end of the trial was the last visit of the last subject undergoing the trial.
Reporting Period:	• Intermediate data analysis date: From May 11, 2015 to June, 2018 (≥ week 48) • Inclusion date of first patient: May 11, 2015 • End of the study: November, 2018 (week 96)
Phase Development:	Phase I/IIa (Safety and Efficacy)
Background And Rationale For The Study:	Multiple Sclerosis (MS) is a chronic autoimmune disease of the Central Nervous System (CNS) with an unpredictable clinical course causing a slow and progressive disability in most patients. The symptoms usually appear between the ages of 20 and 40. Clinically, MS is a very heterogeneous disease. Most patients present outbreaks that remit partially or totally. Current MS treatment is based on immunomodulatory and non-specific immunosuppressant drugs, but with partial efficacy. Therapy with stem cells is very promising and it is proposed as an alternative therapeutic strategy to repair the neural lesions. Numerous trials confirm that purified hematologic stem cells (HSC) or mesenchymal stem cells (MSC) harvested from blood or bone marrow (BM) can effectively reduce the effects of some autoimmune diseases with no adverse effects attributable to its administration. Pre-clinical and clinical studies with MSC support the expectation that they could modulate the immune response that correlates with the inflammatory activity of MS. Availability of an autologous source of transplantable adult stem cells with relative ease represents a very valuable therapeutic option in the development of regenerative CNS strategies. We initiated this pilot crossover clinical trial (Protocol code: XCEL-MS-02, Nº EudraCT: 2012-000734-19) as a first step to evaluate the therapeutic potential of autologous MSC transplantation as a therapeutic alternative for patients with MS. The present Clinical Study Report (CSR) presents the partial results of safety and efficacy obtained from this trial after 1-year follow-up.
Objectives:	<u>General objective</u> To analyse the treatment of autologous mesenchymal stem cells derived from bone marrow as a potential therapeutic strategy for the treatment of multiple sclerosis (MS)

	Primary objective To evaluate the safety and tolerability of XCEL-MC-ALPHA by: a. Overall incidence of adverse events by treatment. b. Incidence of adverse events by organ system preferred term, by treatment. c. Severity of adverse events per treatment. d. Intensity of adverse events per treatment. e. Causation by treatment. Secondary objective To assess the effectiveness of XCEL-MC-ALPHA determined by the cumulative number of lesions that enhance with gadolinium T1 in sequence Magnetic Resonance Imaging (MRI) in both treatment periods (at week 4, 12, 24, 28, 36 and 48), in the group initially assigned to placebo and the group initially assigned to XCEL-MC-ALPHA. Exploratory objective To assess MSC as immune modulatory treatment in subjects with MS: 1. Evaluate the efficacy of XCEL-MC-ALPHA by the number of outbreaks in both treatment periods. 2. To evaluate the efficacy of administration of XCEL-MC-ALPHA by the proportion of subjects in complete remission (defined as absence of outbreaks, of lesions that are enhanced with gadolinium in the T1 sequence and of new lesions in the T2 sequence MRI) during the first year of treatment. 3. To evaluate the efficacy of the early administration of XCEL-MC-ALPHA by the accumulated number of new lesions in the T2 sequence after one year of treatment (lesions observed at week 48).
Methodology:	Placebo-Controlled, Randomized, Double-Blind, Crossover, Phase I/IIa Study in a single site to evaluate tolerability, safety and efficacy of the investigational product XCEL-MC-ALPHA in 8 subjects with Relapsing-Remitting Multiple Sclerosis (RRMS) or Secondary Progressive Multiple Sclerosis (SPMS) fulfilling our predefined inclusion and exclusion criteria. The trial consisted of 2 arms of 2 consecutive treatments (XCEL-MC-ALPHA/Placebo and Placebo XCEL-MC-ALPHA) without washout period. Subjects were randomly allocated in each arm (4 subjects / arm) to receive the assigned intravenous infusion. Randomization list was generated with Statistical Analysis System (SAS®, *version 9.2*). Arm A consisted of XCEL-MC-ALPHA treatment (24 weeks) followed by placebo (24 weeks) and Arm B consisted of placebo (24 weeks) followed by XCEL-MC-ALPHA treatment (24 weeks) For each subject, the time period to complete the trial was 48 weeks (Figure 1).

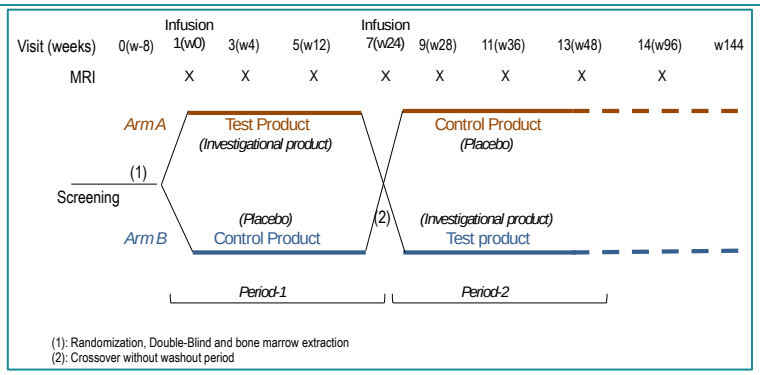

Figure 1. Brief scheme of the Protocol XCEL-MS-02 design (Crossover trial: 2 sequences, 2 treatments, 2 periods)

Infusions were performed at the CEMCAT (*Centre d'Esclorosis Múltiple de Catalunya*) at the Hospital Universitari Vall d'Hebron, Barcelona (Spain). Subjects remained in the Clinical Unit for 2h post-infusion to review tolerability and safety values. Subjects were monitored closely for any adverse event (AE) and safety assessments at week 1 (by telephone) and at week 4, 12 and 24 post-infusion of each intervention at the Clinic Unit. At week 24, treatment crossover was performed in subjects that fulfilled baseline criteria. It was planned to treat intensive outbreaks with corticoids and request subject' informed consent again to carry on the study. When subject' informed consent was not re-signed, two new visits were programed, one at the end of the treatment and another visit at the annual safety follow-up.

Main outcome measures were: adverse events (AEs), intensity of AEs, cause of AEs, clinical laboratory (biochemistry and haematology), vital signs, ECG parameters, pulse oximetry, physical findings, Gadolinium-enhanced T1-weighted (Gd-T1) and T2-weighted brain MRI scans, Expanded Disability Status Scale (EDSS) and outbreaks. After week 48, two visits were schedule to control long-term safety parameters. Subjects completed the study participation 144 weeks after randomization.

Number of Subjects:

Eight adults (age range 18-60 years; 4 women and 4 men) with Relapsing-Remitting Multiple Sclerosis (RRMS) or Secondary Progressive Multiple Sclerosis (SPMS) were planned and analysed.

Diagnosis and Main Criteria for Inclusion and Exclusion:

Inclusion Criteria:

1. Subjects with MS (McDonald 2010 criteria (Polman et al., 2011)
2. Clinical course of patients with relapsing-remitting MS or secondary progressive (Lublin FD, 1996)
3. Age between 18 and 60
4. Patients in whom is not indicated or are not in a position to begin treatment with drugs that modify disease available for MS, after the researcher has been informed of their benefits and potential adverse events, or who do not respond adequately to standard therapy or cannot tolerate
5. EDSS ≤6.5
6. At least nine T2 lesions
7. Active MS, defined by: At least one outbreak in the last year or At least one Gd-enhancing lesion in the last 6 months
8. Have signed the informed consent for participation in the study

Exclusion Criteria:

1. Patients pre-treated with:

	- Interferon beta or glatiramer acetate 3 months prior to screening 1 - Natalizumab or fingolimod in the 6 months prior to screening - Teriflunomide in the previous 12 months, or in the previous 4 weeks after an accelerated wash-out procedure - Mitoxantrone, cyclophosphamide or other immunosuppressive therapy at any time - Experimental treatment within 3 months prior to screening 2. MS attack in the 4 weeks prior to randomization 3. Serum creatinine > 2.0 mg / dL If serum creatinine is > 1.2 mg / dL, measure glomerular filtration rate was done. Patients were excluded when the filtrate is <60 mL/minute/1, 73 m^2 4. Infectious disease active or uncontrolled 5. Fertile patients who are not using a suitable method of contraception (at the discretion of the investigator). If the patient is postmenopausal or sterile must be documented in the medical record. 6. Pregnancy, intention to become pregnant during the 2 years following the inclusion of the study. 7. Women in breastfeeding period 8. History or signs of immunodeficiency 9. Serious concomitant medical pathology or other disease that in the opinion of the principal investigator may affect the patient's participation in the study 10. History of HIV or positive test of HIV (Anti-HIV-1/2). 11. Positive serology of hepatitis B virus (HBsAg) or C (Anti-HCV-Ab) or syphilis (additional tests may be performed, such as *Treponema Pallidum*, *Trypanosomacruzi*, *Toxoplasma*, EBV, CMV, and / or malaria). The serology must be performed within a month prior to the bone marrow extraction.
Test Product, Dose, Mode of Administration, Batch Number:	XCEL-MC-ALPHA [Autologous adult mesenchymal stem cells from bone marrow expanded and cryopreserved in saline solution balanced with electrolytes (*ViafloPlasmalyte, Baxter, S.L*) and 2% v/v of human serum albumin (*Instituto Grifols, S.A*. Suspension for injection)]. Dose: 10^6±20% cells/kg ($0.8x10^6$ to $1.2x10^6$ cells/kg). Mode of administration: Intravenous use. Infusion duration: 10 minutes. Batch number was not required because a unique and individualized infusion bag of test product was prepared *ad-hoc* for each subject.
Duration of Treatment:	48 weeks to complete the crossover sequence of treatment / Subject
Control Product, Dose, Mode of Administration, Batch Number:	Placebo [saline solution balanced with electrolytes (*ViafloPlasmalyte, Baxter, S.L*) and 2% v/v of human serum albumin (*Instituto Grifols, S.A*)]. Dose: Not available. Mode of administration: Intravenous use. Infusion duration: 10 minutes. Batch number was not required because a unique and individualized infusion bag of control product was prepared *ad-hoc* for each subject.
Endpoints:	Primary end points: Safety and tolerability • Adverse Effects • Abnormal physical findings and vital signs (Heart rate - HR, Respiratory rate – RR, Body temperature – T^a and Blood pressure – BP), clinical laboratory (biochemistry and haematology), Electrocardiography (ECG) parameters and pulse oximetry • Time points of evaluation of this end point: 4, 12, 24, 28, 36 and 48th weeks post-infusion

	Secondary end points: Efficacy • Lesions (spatial and temporal dissemination of demyelinating plaques) by means of serial Gadolinium-enhanced T1-weighted (Gd-T1) brain MRI scans • Time points of evaluation of this end point: 4, 12, 24, 28, 36 and 48th weeks post-infusion Exploratory end points: Clinical efficacy • Number of MS outbreaks at week 48 post-infusion • New lesions by means of serial T2-weighted brain MRI scans at week 48 post-infusion • Complete remission: absence of outbreaks, absence of lesions in Gd-T1 brain MRI scans (at weeks 4, 12, 24, 28, 36 and 48 post-infusion) and absence of new lesions in T2-weighted brain MRI scans (at week 48 post-infusion)
Statistical Methods:	Security. Descriptive statistics Efficacy. Analysis includes all the subjects in the safety population who have not incurred any significant protocol violation and have at least one evaluation of efficacy variable at baseline and another later. Parameters: • Cumulative number of lesions that enhance with gadolinium T1 in sequence MRI: Generalized Linear Model (GLM) for repeated measures using Proc GLM-Mix, SAS® *version 9.2* • Comparison of MS outbreak rates (XCEL-MC-ALPHA vs placebo and Arm A vs Arm B): GLM for counts • Proportion of subjects with complete remission: Conditional logistic regression (paired data) adjusted by treatment, treatment period before crossover, and treatment sequence
Summary of Results:	No protocol deviation, but changes in the planned analysis of the results were required due to the small size and the homogeneity of the descriptive results observed. The analysis deviation did not affect the performance of the study. Descriptive analysis of the endpoints is presented by sequence of treatment, by treatment and by treatment period. **Subject Disposition:** A total of 8 eligibly subjects were randomized in 2 arms of 2 consecutive treatments (4 subjects in Arm A of XCEL-MC-ALPHA/Placebo and 4 subjects in Arm B of Placebo/XCEL-MC-ALPHA). All 8 total subjects compliant the treatment (a unique dose of XCEL-MC-ALPHA plus a unique dose of Placebo). Duration of the i.v. infusions were 10 min. No subjects required hospital stay after the intervention. The period of time between infusions was 24 weeks without washout. All subjects completed the 48 weeks crossover treatment sequence and 4 out of 8 total subjects (2 subjects/Arm) completed the 96 weeks of follow-up for intention-to-treat analysis. All 8 total subjects will complete the follow-up by November 2018. **Demography and Baseline Characteristics**: Mean (SD) age = 43.3 (5.6), 50% men, mean (SD) of years since the first outbreak = 15.7 (8.3), mean (SD) of outbreaks in the last year = 1.0 (0.5) and mean (SD) of baseline EDSS score = 3.0 (0.7). No significant differences were observed on demography and baseline characteristics, medical history and concurrent illnesses between the two groups at randomisation (U Mann-Whitney test, $p>0.05$ and Fisher's exact test, $p>0.05$). **Safety Results:** A total of 20 AEs were observed during the crossover clinical trial, but most of them were mild. No AEs were related with the infusion of XCEL-MC-ALPHA or Placebo. No withdrawal from the study was required. The order of treatment sequence (Placebo / XCEL-MC-ALPHA or XCEL-MC-ALPHA / Placebo) did not affect the onset of AEs (Skillings-Mack test, $p > 0.05$). No significant increases of the proportion of subjects with AEs changes due to the infusion of XCEL-MC-ALPHA were observed in both groups (Simmetry test, $p>0.05$). And, the proportion of subjects with AEs who received XCEL-MC-ALPHA was similar to those who received placebo at any period (Fisher's exact test, $p>0.05$). No carry out bias was observed.

	Regarding other safety parameters, the order of treatment sequence did not affect nether the vital signs (Skillings-Mack test, $p > 0.05$) nor ECG (Skillings-Mack test, $p > 0.05$). No significant differences of vital signs and ECG were observed between pre-post infusion of XCEL-MC-ALPHA or Placebo at period-1 (U Mann-Whitney test, $p>0.05$) or period-2 (U Mann-Whitney test, $p>0.05$). **Efficacy Results:** Subjects did not show new Gd-T1 brain lesions within-subject conditions in Arm A or in Arm B nor MS disease progression. The order of treatment sequence (Placebo / XCEL-MC-ALPHA or XCEL-MC-ALPHA / Placebo) did not affect the mean (SD) of within-subject Gd-T1 lesions (Skillings-Mack test, $p > 0.05$). After the infusion of XCEL-MC-ALPHA, no subject developed Gd-T1 brain MRI changes neither at week 12 in Arm A nor at week 48 in Arm B of treatment. The proportion of subjects with Gd-T1 brain lesions analysed between Arms at each period of treatment did not change. No carry out bias was observed. Regarding other secondary efficacy parameters such as Gd-T2 brain lesions, MS outbreaks and EDSS scores, no significant changes were observed at sequence of treatment, treatment and treatment period level. The minor changes observed in EDSS scores were not clinically relevant.
Conclusion:	The crossover design used was appropriated for this study and no carry out bias was observed. The infusion XCEL-MC-ALPHA (autologous MSC based) and Placebo in 8 subjects with RRMS or SPMS was feasible, safe and well tolerated. Not significant efficacy effects were detected. A new methodological approach (dose, administration route and mediators) must be study to elicit the efficacy XCEL-MC-ALPHA on MS.
Date and Version of this Report:	DRAFT CSR XCEL-MS-02 version 2.0. Date March 18th, 2019