Study Protocol

MS-SMART: Multiple Sclerosis-Secondary Progressive Multi-Arm Randomisation Trial

A MULTI-ARM PHASE IIB RANDOMISED, DOUBLE BLIND PLACEBO-CONTROLLED CLINICAL TRIAL COMPARING THE EFFICACY OF THREE NEUROPROTECTIVE DRUGS IN SECONDARY PROGRESSIVE MULTIPLE SCLEROSIS.

Sponsors	UCL
Funder	EME
Funding Reference Number	11/30/11
Sponsor Number	12/0219
Chief Investigator	Dr Jeremy Chataway
EudraCT Number	2012-005394-31
REC Number	13/SS/0007
ISRCTN Number	ISRCTN28440672
ClinicalTrials.gov Identifier	NCT01910259
Version Number and Date	V7 4th June 2018
Investigational Medicinal Products	Fluoxetine, Riluzole, Amiloride or Placebo
Trial Phase	IIB
Sites	Multi-site

Efficacy and Mechanism Evaluation Programme

MS-SMART Protocol V7 (dated 4th June 2018) supersedes Protocol V6 (dated 5th October 2017) Please note that Protocol V7 was updated due to a non substantial amendment to correct a typing error only.	
Description of changes from Version 6:	***Updated sections (where relevant)***
The ClinicalTrials.gov identifier had a typing error in the number which has been updated.	**Title Page**

MS-SMART Protocol V5 (dated 1st November 2016) supersedes Protocol V4 (dated 25th May June 2015)	
Description of Major Protocol changes from Version 4:	***Updated sections (where relevant)***
1) The protocol has been amended to reflect changes to the SmPC for fluoxetine.	Section 1.2 -Rationale for study- Drug Selection - Fluoxetine Section 6.6 Other medications - Absolute contraindications and use with caution
2) Wording change in line with the Statistical Analysis Plan (SAP)	Study Objectives - Exploratory Objectives to inform mechanism.
3) In the event that a participant misses a visit where questionnaires should be completed, the questionnaires can be sent to the participant for completion.	Section 7 - Study assessments

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"The Chief Investigator and the JRO have discussed this protocol. The investigators agree to perform the investigations and to abide by this protocol.

The investigator agrees to conduct the trial in compliance with the approved protocol, EU GCP and UK Regulations for CTIMPs (SI2004/1031; as amended), the Data Protection Act (1998), the Trust Information Governance Policy (or other local equivalent), the Research Governance Framework (2005), the sponsor's SOPs, and other regulatory requirements as amended.

Signatures

Dr Jeremy Chataway Chief Investigator	Signature	Date
Professor Siddharthan Chandran Co-Chief Investigator	Signature	Date
Professor Nigel Stallard Trial Statistician	Signature	Date
Professor Christopher Weir Trial Statistician	Signature	Date
Gina Cranswick ECTU Representative	Signature	Date
Nick McNally UCL Director of Research Services Sponsor Representative	Signature	Date

list of abbreviations

AE	Adverse Event
ALT	Alanine aminotransferase
ALS	Amyotrophic lateral sclerosis
APR	Annual Progress Report
AR	Adverse Reaction
AST	Aspartate aminotransferase
ASIC	Acid Sensing Ion Channels
ATP	Adenosine TriPhosphate
bd	Bis die (twice daily)
BDI	Beck Depression Index
BDNF	Brain-derived neurotrophic factor
Br	Bilirubin
BPI	Brief Pain Inventory
CS	Clinically Significant
CNS	Central Nervous System
CRF	Case Report Form
CTA	Clinical Trial Authorisation
CTN	Clinical Trials Network
CUPID	Cannabinoid Use in Progressive Inflammatory brain Disease
CSF	Cerebrospinal Fluid
DMC - DMEC	Data Monitoring Committee/Data Monitoring & Ethics Committee
DMT	Disease Modifying Treatment
ECTU	Edinburgh Clinical Trials Unit
EDSS	Expanded Disability Status Scale
EQ-5D	Health Economics questionnaire
EU GCP	European Union Good Clinical Practice
FBC	Full blood count
ɣGT	Gamma-glutamyltransferase
GDNF	Glial cell line-derived neurotrophic factor
GCP	Good Clinical Practice
Hb	Haemoglobin
IMP	Investigational Medicinal Product
ISF	Investigator Site File
JRO	Joint Research Office for the sponsor
LP	Lumbar Puncture
MND	Motor Neurone Disease

MRI	Magnetic Resonance Imaging
MRS	Magnetic Resonance Spectroscopy
MTR	Magnetic Transfer Ratio
MS	Multiple Sclerosis
MSFC	MS Functional Composite Score
MSIS29	Multiple Sclerosis Impact Scale
MSWS	Multiple Sclerosis Walking Scale
NCS	Not Clinically Significant
NF	Neurofilament
NFI	Neurological Fatigue Index
NICE	National Institute for Clinical Excellence
NIHR	National Institute for Health Research
NIMP	Non Investigational Medicinal Product
NGF	Nerve growth factor
NPRS	Numerical Pain Rating Score
NPS	Neuropathic Pain Scale
9HPT	9 Hole Peg Test
OCT	Optical Coherence Tomography
PASAT	Paced Auditory Serial Addition Test
PBVC	Percentage brain volume change
PICS	Participant Identification Centres
PIL	Patient Information Leaflet
PIS	Patient Information Sheets
PPMS	Primary Progressive Multiple Sclerosis
PROMS	Patient Reported Outcome Measures
PwMS	People with MS
PwSPMS	People with secondary progressive Multiple Sclerosis
RGC + IPL	Retinal nerve ganglion cell and inner plexiform layer
RRMS	Relapsing remitting Multiple Sclerosis
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SDMT	Symbol Digit Modalities Test
SIENA	Structural Image Evaluation, using Normalization of Atrophy
SLCVA	Sloan Low contrast visual acuity
SmPC	Summary of Product Characteristics
SOP	Standard Operating Procedure
SPMS	Secondary progressive Multiple Sclerosis
SSRI	Selective Serotonin Re-uptake Inhibitor

SUSAR	Suspected Unexpected Serious Adverse Reaction
T25FW	Timed 25 Foot Walk
TMF	Trial Master File
TMG	Trial Management Group
TSC	Trial Steering Committee
UAR	Unexpected Adverse Reaction
UCL	University College London
WBC	White blood cell count
WOCBP	Women of childbearing potential

LAY summary

Multiple sclerosis (MS) is a disabling and progressive neurological disease that affects approximately 100,000 people in the UK. Many patients with MS experience two phases of disease; early MS (also called relapsing remitting MS, RRMS) and late MS (also called secondary progressive MS (SPMS). Early MS is due to inflammation of the nerves and the insulation (called myelin) that surrounds the nerves. Early MS is often characterised by periods of "attacks" interspersed with periods of "remission" with no or low disease symptoms. Late or progressive MS, which affects the majority of patients and typically emerges after 10-15 years of disease, results from actual nerve death (also called neurodegeneration). The progressive stage of disease results not in individual attacks but slow, cumulative and irreversible disability affecting walking, balance, vision, cognition, pain control, bladder and bowel function. Critically, and unlike early disease, there is no proven treatment for the late stage of MS. This is therefore an urgent and major unmet health need. MS-SMART directly addresses this need and will evaluate in this clinical trial three drugs (fluoxetine, riluzole or amiloride), all of which have shown some promise in MS, and in particular in SPMS. The trial is randomised and blinded. Randomisation means patients can get any one of the three active drugs or the inactive placebo/dummy; blinded means that neither patients nor the doctors will know which drug or placebo patients are receiving. Randomisation and blinding are standard approaches in clinical trials to ensure unbiased testing of drugs. All patients in MS-SMART will have periodic MRI (magnetic resonance imaging) brain scans and after 96 weeks these will be analysed. We will then compare the scans of each drug to the placebo or dummy to see if any of the drugs slow the rate of brain shrinkage that normally occurs in SPMS. This measured change in brain size is the primary (major) outcome of MS-SMART.

STUDY TITLE	MS-SMART: Multiple Sclerosis-Secondary Progressive Multi-Arm Randomisation Trial A MULTI-ARM PHASE IIB RANDOMISED, DOUBLE BLIND PLACEBO-CONTROLLED CLINICAL TRIAL COMPARING THE EFFICACY OF THREE NEUROPROTECTIVE DRUGS IN SECONDARY PROGRESSIVE MULTIPLE SCLEROSIS.
BACKGROUND	Multiple sclerosis (MS) is the commonest acquired disabling neurological disease affecting young adults in temperate latitudes. It is a progressive disorder of the brain and spinal cord that affects approximately 100,000 people in the UK and 2.5 million globally. Most patients with MS experience two phases of disease: (i) early MS (relapsing remitting MS, RRMS) due to bouts of largely reversible inflammation mediated nerve damage and (ii) late MS, secondary progressive MS (SPMS), which affects the majority of patients, usually after 10-15 years from initial MS diagnosis. There is no proven treatment for SPMS - it is therefore a major unmet health need for the NHS.
STUDY OBJECTIVES	**Primary Objective -** To establish whether a drug, from a panel (fluoxetine, riluzole, amiloride) can slow the rate of brain volume loss in SPMS over 96 weeks using MRI-derived Percentage Brain Volume Change (PBVC). **Secondary Objectives -** • To establish that a multi-arm trial strategy is an efficient way of screening drugs in SPMS and can become the template for future work • To explore any anti-inflammatory drug activity using the count of new and enlarging T2 lesions • Examining for evidence of pseudo-atrophy (to ensure reliability of the primary outcome measure) • To examine the clinical effect of neuroprotection as measured by clinician: Expanded Disability Status Scale (EDSS) MS Functional Composite score (MSFC) Symbol Digit Modalities Test (SDMT) Sloan Low contrast visual acuity (SLCVA) relapse rate and patient reported outcome measures: Multiple Sclerosis Impact Scale v2 (MSIS29 v2), Multiple Sclerosis Walking Scale v2 (MSWS v2), Pain - Numerical Pain Rating Score (NPRS) and Brief Pain Inventory (BPI) and Neuropathic Pain Scale (NPS) Fatigue - Neurological Fatigue Index (NFI) • To collect basic health economic data (EQ-5D) to inform future phase III trials **Exploratory Mechanistic Objectives -** • Persistent new T1 hypointense lesion count to assess neuroprotection in new lesions • Grey matter volume change to assess cortical neuroprotection

	• MR spectroscopy (MRS) to measure N-acetyl aspartate (reversal of neuronal mitochondrial dysfunction), myoinositol (prevention of glial cell inflammation) and glutamate (prevention of excitotoxicity) • Magnetic Transfer Ratio (MTR) to evaluate myelination • Cervical cord imaging to assess cord neuroprotection • Composite MRI/disability scores to increase sensitivity and study interaction of treatment mechanisms • Quantification of Cerebrospinal fluid (CSF) neurofilament levels to measure neuroprotection • CSF lymphocyte phenotyping • Optical Coherence Tomography (OCT)-measured Retinal nerve Fibre Layer (RNFL) thickness as a measure of neuroprotection.
STUDY POPULATION	**Secondary Progressive Multiple Sclerosis**
STUDY TREAMENT	Fluoxetine, Riluzole or Amiloride versus placebo
ELIGIBILITY CRITERIA	INCLUSION CRITERIA • Confirmed diagnosis of SPMS. Steady progression rather than relapse must be the major cause of increasing disability in the preceding 2 years. Evidence of progression, either an increase of at least one point in EDSS or clinical documentation of increasing disability in patient notes • Expanded Disability Status Scale (EDSS) 4.0-6.5 • Aged 25 to 65 inclusive • Women and men with partners of childbearing potential must be using an appropriate method of contraception to avoid any unlikely teratogenic effects of the 3 drugs from time of consent, to 6 weeks after treatment inclusive • Women must have a negative pregnancy test within 7 days prior to the baseline visit unless not of child bearing potential (e.g. have undergone a hysterectomy, bilateral tubal ligation or bilateral oophorectomy or they are postmenopausal) • Willing and able to comply with the trial protocol (e.g. can tolerate MRI and fulfils the requirements for MRI, e.g. not fitted with pacemakers or permanent hearing aids), ability to understand and complete questionnaires • Written informed consent provided EXCLUSION CRITERIA • Pregnancy or breast feeding patients • Baseline MRI scan not of adequate quality for analysis (e.g. too much movement artefact) • Significant organ co-morbidity (e.g. malignancy or renal or hepatic failure) • Relapse within **3 months** of baseline visit • Patients who have been treated with iv or oral steroids for an MS relapse/progression within **3 months** of baseline visit (these patients can undergo future screening visits

	once the 3 month window has expired), patients on steroids for another medical condition may enter as long as the steroid prescription is not for multiple sclerosis (relapse/ progression). • Use of Simvastatin at 80mg dose within **3 months** of baseline visit (lower doses of Simvastatin and other statins are permissible) • Commencement of fampridine within **6 months** of baseline visit • Use of immunosuppressants (e.g. azathioprine, methotrexate, cyclosporine) or disease modifying treatments (β-interferons, glatiramer) within **6 months** of baseline visit • Use of fingolimod/fumarate/teriflunomide/laquinomod/or other experimental disease modifying treatment (including research of an investigational medicinal product) within **12 months** of baseline visit • Use of mitoxantrone/ natalizumab/ alemtuzumab/ daclizumab if treated within **12 months** of baseline visit • Primary progressive MS • Relapsing-remitting MS • Known hypersensitivity to the active substances and their excipients to any of the active drugs for this trial • Use of: lithium, chlorpropamide, triamterene and spironolactone within **6 months** of the baseline visit • Current use of potassium supplements • Current use of tamoxifen • Current use of herbal treatments containing St. John's Wort • Significant signs of depression • Use of an SSRI within **6 months** of the baseline visit • Use of monoamine oxidase inhibitors, phenytoin, L-tryptophan) and/or neuroleptic drugs within **6 months** of the baseline visit • A Beck Depression Index score of 19 or higher • Bipolar disorder • Receiving or previously received Electro-Convulsive Therapy • Epilepsy/seizures • Glaucoma • Patients with a history of bleeding disorders or currently on anticoagulants • Routine screening blood values (LFT) >/ 3 x upper limit of normal (ULN) of site reference ranges (ALT/AST, bilirubin, ɣGT) • Potassium <2.8mmol/l or >5.5mmol/l

	• Sodium <125mmol/l • Creatinine >130µmol/l • WBCs <3 x 10^9/l • Lymphocytes <0.8 x 10^9/l • Neutrophil count <1.0 x 10^9 /l • Platelet count <90 x 10^9 /l • Haemoglobin <80g/l
NUMBER OF TRIAL PARTICIPANTS	440
STUDY ASSESSMENTS	Primary Outcome: MRI-derived percentage brain volume change (PBVC) as measured by the SIENA technique. Secondary outcomes: Expanded Disability Status Scale (EDSS) Timed 25 Foot Walk (T25FW) 9 Hole Peg Test (9HPT) Paced Auditory Serial Addition Test (PASAT) MS Functional Composite score (MSFC) (a composite z-score of T25FW, 9HPT, PASAT) Symbol Digit Modalities Test (SDMT) Sloan Low contrast visual acuity (SLCVA) Relapse rate Multiple Sclerosis Impact Scale v2 (MSIS29) Multiple Sclerosis Walking Scale v2 (MSWS) Pain – Numerical Pain Rating Score (NPRS), Brief Pain Inventory (BPI) and Neuropathic Pain Scale (NPS) Fatigue - Neurological Fatigue Index (NFI) Health economics (EQ-5D). Mechanistic outcomes: Persistent new T1 hypointense lesion count, Grey matter volume change, MR spectroscopy, Magnetic Transfer Ratio (MTR), Cervical cord imaging, Composite MRI/disability scores, Quantification of CSF neurofilament levels, CSF lymphocyte phenotyping, OCT-measured RNFL thickness.
TRIAL DURATION PER PARTICIPANT	100 weeks
OVERALL TRIAL DURATION	01.04.2013 – 31.10.2018
PLANNED NUMBER OF SITES	UK Multi-site Trial. 10-15 sites.

1 INTRODUCTION

1.1 BACKGROUND

Multiple sclerosis (MS) is the commonest acquired disabling neurological disease affecting young adults in temperate latitudes. It is a progressive disorder of the brain and spinal cord that affects approximately 100,000 people in the UK and 2.5 million globally.[1] Most patients with MS experience two phases of disease: (i) early MS (relapsing remitting MS, RRMS) due to bouts of largely reversible inflammation mediated nerve damage and (ii) late MS, secondary progressive MS (SPMS), which affects the majority of patients, where patients advance into SPMS usually after 10-15 years from initial MS diagnosis. SPMS results from progressive nerve death or neurodegeneration that causes accumulating and irreversible disability characterised by a range of devastating problems affecting walking, balance, vision, cognition, pain control, bladder and bowel function. Unlike RRMS, where there are an increasing number of advanced treatments, there is no proven treatment for SPMS – it is therefore a major unmet health need for the NHS.

1.1.1. Treatment Failure in Secondary Progressive Multiple Sclerosis

1.1.1.1. Clinical Trial failure in SPMS:

Although immunomodulatory/anti-inflammatory, disease modifying treatments (DMTs), are effective in reducing relapse frequency in RRMS, they have been unsuccessful in slowing disease progression in SPMS.[2] This is the conclusion from trials of over 4500 SPMS patients with DMTs and provides strong evidence that RRMS and SPMS have differential pathological substrates. RRMS reflects focal, largely white matter, immunologically driven inflammation, whilst SPMS is dominated by widespread neurodegeneration. Consequently the absence of effect of anti-inflammatory drugs on the neurodegenerative (SPMS) phase of MS is not unexpected. Despite this identified unmet clinical need for effective neuroprotection, there are comparatively few clinical trials that aim to modify the SPMS disease course. This has been recently underlined by the negative results of the phase III Cannabinoid Use in Progressive Inflammatory brain Disease (CUPID trial). [3]

1.1.2. Mechanistic Processes Underlying Neurodegeneration in SPMS

Overwhelming evidence from human pathological, radiological, clinical and animal based experimental studies have demonstrated that the dominant pathology and key determinant of disability in SPMS is neurodegeneration[4,5,6] MR imaging studies showing progressive and substantial brain volume loss in SPMS along with reduction in neuroaxonal metabolites have been particularly influential in highlighting the significance of neurodegeneration in SPMS. This has led directly to MR based measures of brain atrophy becoming a benchmark outcome criterion for MS neuroprotection studies.[7] The cause of neurodegeneration in SPMS is complex and accumulating evidence from multiple experimental systems including human studies implicate a handful of key mechanistic processes. It is increasingly apparent that some of these neuroaxonal injurious mechanisms not only overlap, but also are common to other diseases characterised by neuronal death such as Alzheimer's disease. A distinctive feature of neurodegeneration in SPMS is that it occurs in the context of chronic demyelination where the resulting loss of oligodendrocyte-myelin mediated support renders the neuron particularly vulnerable to the following inter-linked processes:

1.1.2.1. Oligodendrocyte loss/demyelination:

Demyelinated axons are devoid of the oligodendrocyte-myelin mediated trophic support necessary for neuronal survival and are therefore particularly vulnerable to any form of injury and physiological stress. The most common explanation for this is that the "denuded demyelinated axon" is not only deprived of trophic support, but also responds to demyelination by diffusely redistributing along the axon sodium channels, that were previously restricted to nodes of Ranvier.[8,9,10] This adaptive response comes at a price of "energy failure".

1.1.2.2. **Energy failure:**
The ionic adaptive response leads to an imbalance between energy demand and supply in the demyelinated axon that leads to intra-axonal calcium excess. This in turn reduces Adenosine Tri Phosphate (ATP) production, promotes mitochondrial damage and reactive oxygen species production and triggers calcium mediated injury and death cascades. The recent discovery that lactic acidosis due to energy failure leads to the activation of a class of acid sensing ion channels (ASICs) on neurons and oligodendrocytes - in both human MS derived tissue samples and experimental models of MS provides an additional mechanism whereby intraxonal cation excess leads to neuroaxonal death.[11,12]

1.1.2.3. **Glial (astrocyte/microglial) mediated production of reactive oxygen/nitric oxide species**:
A common feature of progressive MS is the production of reactive oxygen/nitric oxide by activated microglia and reactive astrocytes in SPMS that again sets up a self-perpetuating cycle of injury including mitochondrial damage and altered lipid metabolism.

1.1.2.4. **Excitotoxicity:**
Is another common end-stage outcome of the pathological processes outlined above leading to neuroaxonal injury and death due to excessive stimulation by neurotransmitters such as glutamate.

In summary it is clear that the “maladaptive ionic” response to demyelination sets in train a negative cycle of energy failure and linked processes including reactive oxygen species production by glia, mitochondrial inhibition and ASIC upregulation that converge around a common process of intra-axonal cation excess that initiates secondary calcium mediated injury and ultimately death cascades including excitotoxicity.

MS-SMART is a multi-arm phase IIb trial, which will determine the efficacy, and advance our understanding of the mechanism of action, of three putative neuroprotective (see below) repurposed drugs versus placebo, through standard (core) and advanced MRI scans, disability assessments, visual measurement and cerebrospinal fluid (CSF) analysis.

1.2 RATIONALE FOR STUDY

1.2.1 Drug Selection

Against the background of neurodegenerative processes outlined above, we prospectively sought to identify existing putative neuroprotective drugs that target these pivotal neurodegenerative-causing pathways as the most biologically plausible approach to slowing disease progression in SPMS.

Widespread failure of neuroprotective trials for neurodegenerative diseases as well as acute brain injury (e.g. stroke) along with the prolonged time from target selection to regulatory approval has advocated a new and more systematic approach to drug identification that includes drug rescue and repurposing. Drug rescue (drugs at advanced stage of development but abandoned before approval) and repurposing (already approved drug) by exploiting existing clinical efficacy, safety and regulatory data, is a strategy to reduce cost and time in getting drugs to licensed approval status.[13] *We therefore first undertook an MS-Clinical Trials Network (CTN) commissioned systematic review of animal and human trials of putative neuroprotective drugs.* For the human analysis, in addition to MS, we extended the diseases to include amyotrophic lateral sclerosis [ALS], Huntington’s, Alzheimer’s and Parkinson’s Diseases, because of the existence of shared pivotal pathways in neurodegeneration (see above). Searching Pubmed, EMBASE, ISI (n=27,000) and Cochrane Group MS database (n=2600) returned 120 drugs for which summaries were generated that included analysis of mechanism of action, scores for safety, study quality, efficacy and sample size. Two clinicians then independently reviewed the 120 drugs and excluded 68 as unsuitable using predefined criteria. A specially convened International MS Drug Selection meetings comprising expert representation from Cochrane MS group, neuroscientists (including those from NIH, USA), neurologists, brain imaging, PwMS, trial methodologists and industry considered the remaining 52 drugs.

Structured discussions over three rounds of detailed scrutiny with an emphasis on clinical efficacy led to identification of 7 drugs (ibudilast, riluzole, amiloride, pirfenidone, fluoxetine, oxcarbamazepine and PUFA class dietary supplements) that were further ranked and grouped against class of action/mechanistic plausibility noting we were looking for drugs that target the pivotal neurodegenerative pathways discussed in 1.1.1.[14] From those 7, the original intent (protocol v1) was to use: ibudilast, riluzole and amiloride; however, due to drug supply issues, ibudilast is now replaced with fluoxetine.

1.2.2. **Fluoxetine, Riluzole and Amiloride;**

Three putative neuroprotective drugs for SPMS. *Clinical evidence from trials in patients with MS and drug safety profile* (the latest Summary of Product Characteristics (SmPC) are referenced from the electronic medicines compendium: www.**medicines**.org.uk/).

1.2.2.1. **Fluoxetine**

Is a selective serotonin-reuptake inhibitor (SSRI) widely used for depression. However it also has multiple activities relevant to SPMS including: stimulating glycogenolysis and enhancing the production of brain-derived neurotrophic factor (BDNF) in rodent astrocyte cultures.[15,16] Moreover, after 2 weeks of fluoxetine a significantly improved cerebral white matter NAA/creatine ratio was found on MRI, suggesting an improvement in axonal mitochondrial energy metabolism. [17] It might also suppress the antigen-presenting capacity of glial cells.[18] Further more in a recent Cochrane review, in adults with stroke, SSRIs improved measures of dependence.[19]

Clinical Trial. Phase IIa: two trials of fluoxetine have been carried out in MS. In a cohort of MS (n=40, 10% SPMS)[18] there was a significant reduction in relapse rate incidence to 0.54 (95% CI 0.29-0.98) with a trend towards reduction in new inflammatory lesions. In the second trial, 42 patients with SPMS/PPMS (SPMS=69%) randomised in a 1:1 ratio to 40mg/placebo over 2 years. [20] Whilst no statistical differences were seen in overall progression, grey or white matter volume changes, there were trends in favour of fluoxetine e.g. EDSS progression 25% vs 32% and 9HPT progression 5% vs 14%. The study unfortunately was terminated early due to drug expiry issues and was therefore acknowledged to be underpowered. A phase II trial protocol has been developed by the same group, randomising 120 patients with PP/SPMS to fluoxetine or placebo. [21]The primary outcome is the time to confirmed disease progression either by a ≥20% increase in T25FW or 9HPT. Brain atrophy is a secondary outcome as assessed by the brain parenchymal fraction. Patients can remain on DMT.

Dosing and Safety: The dosing in the above mentioned trials was well tolerated, with minor increases in fatigue and drowsiness over placebo.

Dose justification: The dose used in the trials above was 20mg bd and this will be the dose used in the trial.

Side-effects: Fluoxetine is usually well tolerated, although minor side-effects are reported relatively frequently. The most commonly reported adverse reactions in patients treated with fluoxetine were headache, nausea, insomnia, fatigue and diarrhoea. Undesirable effects may decrease in intensity and frequency with continued treatment and do not generally lead to cessation of therapy.

Rare serious side-effects: Patients with a history of suicide-related events, or those exhibiting a significant degree of suicidal ideation prior to commencement of treatment are known to be at greater risk of suicidal thoughts or suicide attempts, and should receive careful monitoring during treatment.

Mitigation-patients with significant depression are excluded.

Rash and allergic reactions: Rash, anaphylactoid events and progressive systemic events, sometimes serious (involving skin, kidney, liver or lung), have been reported.

Patients will stop medication if there is any evidence of progressive and worsening skin reaction.

Seizures: Seizures are a potential risk with antidepressant drugs. Patients with a history of seizures (epilepsy) are excluded.

Mania: Antidepressants should be used with caution in patients with a history of mania/hypomania. As with all antidepressants, fluoxetine should be discontinued in any patient entering a manic phase.

Hepatic/Renal function: Fluoxetine is extensively metabolised by the liver and excreted by the kidneys.

To enter the trial normal renal and hepatic function is required and is monitored. The IMP is stopped if >3ULN.

Cardiovascular Effects: Cases of QT interval prolongation and ventricular arrhythmia including torsades de pointes have been reported during the post-marketing period. Fluoxetine should be used with caution in patients with conditions such as congenital long QT syndrome, a family history of QT prolongation or other clinical conditions that predispose to arrhythmias (e.g., hypokalemia, hypomagnesemia, bradycardia, acute myocardial infarction or uncompensated heart failure) or increased exposure to Fluoxetine (e.g., hepatic impairment). If patients with stable cardiac disease are treated, an ECG review should be considered before treatment is started. If signs of cardiac arrhythmia occur during treatment with Fluoxetine, the treatment should be withdrawn and an ECG should be performed. As recruitment for this study has now been completed, the eligibility criteria will not be amended to exclude patients with known congenital or family history of long QT syndrome or any clinical conditions that predispose them to arrhythmias.

Diabetes: In patients with diabetes, treatment with an SSRI may alter glycaemic control. Hypoglycaemia has occurred during therapy with fluoxetine and hyperglycaemia has developed following discontinuation. Insulin and/or oral hypoglycaemic dosage may need to be adjusted.

Care will be taken to monitor the glycaemic control as part of normal care and where required any dose adjustments to diabetic medication will be at the discretion of the treating physician.

Akathisia/psychomotor restlessness: The use of fluoxetine has been associated with the development of akathisia, characterised by a subjectively unpleasant or distressing restlessness and need to move, often accompanied by an inability to sit or stand still.

Clinical monitoring of the participant's mental state will occur at study visits throughout trial.

If akathisia/psychomotor restlessness occurs the drug will be discontinued.

Haemorrhage: There have been reports of cutaneous bleeding abnormalities, such as ecchymosis and purpura with SSRIs. Ecchymosis has been reported as an infrequent event during treatment with fluoxetine. Other haemorrhagic manifestations (e.g., gynaecological haemorrhages, gastro-intestinal bleedings and other cutaneous or mucous bleedings) have been reported rarely. Caution is advised in patients taking SSRIs, particularly in concomitant use with oral anticoagulants, drugs known to affect platelet function (e.g., atypical antipsychotics, such as clozapine, phenothiazines, most TCAs, aspirin, NSAIDs), or other drugs that may increase risk of bleeding, as well as in patients with a history of bleeding disorders.

Electroconvulsive therapy (ECT): There have been rare reports of prolonged seizures in patients on fluoxetine receiving ECT treatment. Patients will be excluded from the trial with bipolar disorder/previous ECT. If ECT is needed during the trial, participants will discontinue drug.

St John's Wort: An increase in serotonergic effects, such as serotonin syndrome, may occur when selective serotonin reuptake inhibitors and herbal preparations containing St John's Wort (*Hypericum perforatum*) are used together. St John's Wort is contraindicated during the trial.

CYP2D6 isoenzyme inhibitors: (such as flecanide, encainide, carbamazepine and trycyclic antidepressants), should be initiated at or adjusted to the low end of their dose range.

On rare occasions, development of a serotonin syndrome or neuroleptic malignant syndrome-like events have been reported in association with treatment of fluoxetine, particularly when given in combination with other serotonergic (among others, L-tryptophan) and/or neuroleptic drugs. As these syndromes may result in potentially life-threatening conditions, treatment with fluoxetine should be discontinued if such events (characterised by clusters of symptoms, such as hyperthermia, rigidity, myoclonus, autonomic instability with possible rapid fluctuations of vital signs, mental status changes, including confusion, irritability, extreme agitation, progressing to delirium and coma) occur and supportive symptomatic treatment should be initiated. The risk of this is very low and was not reported in the two phase IIa clinical studies of fluoxetine in multiple sclerosis. [18, 20]

Drug withdrawal: The SPC reports that withdrawal symptoms when treatment is discontinued are common, particularly if discontinuation is abrupt. In previous phase IIa clinical studies of fluoxetine in multiple sclerosis [18 20] they did not report any withdrawal reactions upon discontinuation of long term use of Fluoxetine. Based on these previous studies[18, 20], gradual dose reduction in this protocol will not occur on the last treatment visit, however, participants will be followed by telephone 1 month after terminating the study to ascertain any issues with terminating the medication Participants are also reminded in the Participant Information Leaflet (PIL) to contact the study team if there are any withdrawal symptoms.

Risk minimisation: standard monitoring and treatment of nausea, diarrhoea and headache will be provided as needed. Patients with renal failure or hepatic failure are excluded from the trial. If clinically significant LFT changes occur during the trial, or creatinine>130mmol/l, the steps outlined in section 6.7.3 (Management of Laboratory Abnormalities) will be followed. FBC monitoring (including neutrophil count) is part of the safety blood panel, and the steps outlined in section 6.7.3 will take place if the neutrophil level falls below <1.0 $x10^9$ /l or the platelet level reduces to <50 $x10^9$ /l.

1.2.2.2 **Riluzole**
Is licensed for motor neurone disease/amyotrophic lateral sclerosis (MND/ALS) and has two modes of action of relevance to SPMS: reducing glutamate release and antagonism of voltage dependent sodium channels.

Clinical Trial: Phase IIa: 16 patients with Progressive MS were studied 1 year before treatment, followed by riluzole 50mg bd for 1 year.[22] The primary outcome was the change in cervical spinal cord cross-sectional area. It showed a reduction from -2% (yr 1) to -0.2% (yr 2). In addition, the increase in T1 hypointense lesion load was reduced from 15% in year 1 to 6% in year 2; there was a reduction in whole brain parenchymal/intracranial volume from -1.0% (yr 1) to -0.7% (yr 2).

Dosing and Safety: 50mg bd for 1 year was well tolerated in this study. In other disease areas, a study of 959 ALS patients, demonstrated that it is well tolerated at 100mg/day, with the most frequent drug related events being nausea (12-21% riluzole vs 12% placebo) and fatigue (15-20% vs 12%). Dizziness, diarrhoea, anorexia and paraethesiae were also reported. Increased liver function tests (LFT) were observed in 10% of the 100mg group with an increase of ≥5 upper limit of normal (ULN) leading to withdrawal in 4%. ALTs 3-5 ULN usually decreased to normal within 2-6 months of continuous treatment. In a separate study of 537 patients with Huntington's disease, studied over 3 years, it was well tolerated, with a total drug related drop-out rate of 6%. [23]

Dose justification: 50mg bd was the dose used in the MS trial above; it is also the standard dose in MND/ALS and will therefore be used for this trial.

Side-effects: In phase III clinical studies conducted in ALS patients treated with riluzole, the most commonly reported adverse reactions were asthenia, nausea and abnormal liver function tests. Other common minor side-effects are: headache, dizziness, paraesthesiae, somnolence, diarrhoea,

abdominal pain, vomiting, tachycardia. Rare serious side-effects were: neutropenia and interstitial lung disease.

Risk minimisation: patients with renal failure or hepatic failure are excluded from the trial. If clinically significant LFTs occur during the trial, the steps outlined in section 6.7.3 will be followed. FBC (including neutrophil count) is part of the safety blood panel, and the steps outlined in section 6.7.3 will take place if the neutrophil level falls below <1.0 $x10^9$ /l. The report of a febrile illness should prompt physicians to check white blood cell counts and to discontinue riluzole in case of neutropenia. If respiratory symptoms develop such as dry cough and/or dyspnoea, chest radiography should be performed, and in case of findings suggestive of interstitial lung disease (e.g. bilateral diffuse lung opacities), riluzole should be discontinued immediately. In the majority of the reported cases, symptoms resolved after drug discontinuation and symptomatic treatment.

1.2.2.3. **Amiloride**
Is a widely used diuretic and acid sensing ion channel (ASIC) blocker, with recently recognised myelo and neuroprotective effects in both human and experimental models of progressive MS.

Clinical Trial: Phase IIa**:** 17 patients with Primary Progressive MS (PPMS) participated in a pre-treatment (1 yr) and amiloride treatment phase (1 yr) at 5mg bd.[24,25] In the amiloride-treatment year, there was a significant reduction in the whole brain atrophy rate compared to pre-treatment (p=0.009). Corpus callosal radial diffusivity (myelin integrity) and thalamic mean diffusivity (structural integrity) were also significantly improved.

Dosing and Safety: It has been used as a potassium sparing diuretic since it was first introduced in 1967 and has an extremely good side-effect profile, with the main contraindications related to occasional hyperkalaemia e.g. avoidance of drugs such as triamterene, spironolactone, and usage in renal failure.[26]

Dose justification: the dose used in the trial above was 5mg bd. This dose will therefore be used for this trial.

Side-effects: Amiloride is usually well tolerated, although minor side-effects are reported relatively frequently. Apart from hyperkalaemia, significant adverse reactions have been infrequently reported. Nausea/anorexia, abdominal pain, flatulence and mild skin rash are probably due to amiloride; but other side-effects are generally associated with diuresis or with the underlying disease being treated.

Risk minimisation: patients with renal failure or hepatic failure are excluded from the trial. If the plasma potassium exceeds 5.5mmol/l or plasma sodium <125mmol/l or creatinine>130mmol/l, patients will not be able to enter the trial. If these parameters are breached during the trial, the steps outlined in section 6.7.3 will be followed. Potassium conserving drugs and potassium supplements are prohibited (see above). Vital signs will be observed to ensure significant hypotension or hypovolaemia does not occur during the trial. Patients will be advised to avoid dehydration and the volume status will be carefully monitored of the patients. Other symptoms will be monitored as per routine best medical practice.

MS-SMART will test the efficacy and mechanism of action of three repurposed drugs (Fluoxetine, Riluzole and Amiloride). All three drugs are in human use and have a good safety record. Critically for the purpose of MS-SMART they all have shown promise in early phase human MS clinical trials and target one or more of the pivotal neurodegenerative causing pathways implicated in SPMS. This is a **Type B trial**, as the IMPs are all in human use, have a good safety profile but are not currently used for this patient population.

SPMS develops on average from the age of ~40 years (e.g. MS-STAT trial [27]) therefore an age range of 25-65 has been chosen for this trial. In view of the complexity of the trial (for example with MRI as the primary outcome measure and 3 possible active arms) it does not seem appropriate to include vulnerable adults in trial.

1.3 WHY THIS STUDY IS NEEDED NOW:

The major need for patients with established and progressive MS is neuroprotective or disease modifying treatments that will slow or even stop disease progression. This study will evaluate three highly promising putative neuroprotective drugs as well as comprehensively address several of the current knowledge gaps related to the understanding of neuroprotection and neurodegeneration in SPMS through MRI and CSF examination.

1.4 IMPACT ON DISEASE BURDEN:

MS-SMART has been purposefully designed to address the NHS unmet need in providing treatment options to slow progression in SPMS – there are >100,000 PwMS in the UK, of which the majority have SPMS. This group have no disease modifying therapies available to them, only the prospect of increased disability and premature death.

1.5 ECONOMIC BENEFIT:

MS is associated with a significant health economic burden, mainly through the traditional consequences of the disease (ie progressive disability) noting also that IT afflicts preferentially the younger economically active adult population. The health economic burden of MS is considerable, with total societal costs estimated to be up to £30,000/patient/year (c£3Billion/year in the UK). And as costs correlate with disability, estimates range from £12,000 in RRMS, to £60,000 in SPMS, the impact of successfully repurposing a drug to target SPMS cannot be understated.

1.6 MECHANISMS OF DISEASE:

These are described in section 1.1.2 above.

1.7 MAGNETIC RESONANCE IMAGING IN SPMS

1.7.1. **MRI as a primary outcome in RRMS phase II trials:** MRI has been vital in the development of new disease modifying treatments in RRMS, and potentially has a similar pivotal role in SPMS trial design. In smaller phase II trials in RRMS, reduction in inflammatory activity, inferred by the prevention of new gadolinium enhancing lesions or T2 weighted lesions, has come to be a mandatory step in demonstrating potential efficacy before proceeding to the much larger phase III trials, in which the primary outcome measure is reduction in relapse rate. During the last decade, this strategy has been highly successful e.g. natalizumab, n=213, phase II,[28] n=942 phase III;[29] and fingolimod, n=281 phase II,[30] n=1272 phase III,[31] both of which are licensed and recommended for treatment of highly active RRMS by NICE (National Institute for Health and Clinical Excellence).

1.7.2. **MRI as a primary outcome in SPMS phase II trials:**
In SPMS, whilst there may be some effect on new lesion count and lesion volume, the main MRI metric for investigating neurodegeneration - the substrate of progressive and irreversible disability – is the change (reduction) in brain volume. Compared with age-matched healthy controls, there is a greater decrease in brain volume over time in SPMS, termed atrophy rate, which can be quantified by MRI. On average there is 0.5-1% loss of brain volume per year in SPMS.

1.7.3 **Brain atrophy (MS-SMART Primary MRI Outcome):**
A decrease in brain volume is seen at all stages of MS, and especially in SPMS.[32-35] Neuroaxonal tissue constitutes a large proportion of brain volume [36] and the increased rate of brain atrophy has been interpreted as evidence for neuroaxonal loss. This is supported by finding a significant association between cerebral atrophy and reduced normal appearing white matter N-acetyl aspartate,[37] a neuroaxonal metabolite. Brain atrophy is significantly correlated with disability and cognitive impairment in MS.[33, 38-39] Whereas in RRMS, the immunomodulating drug beta interferon was effective in preventing relapses and new MRI lesions, trials in SPMS reported no reduction in the accumulation of disability [40] or of brain atrophy.[41] A US National MS Society (US MSS)-supported international workshop has recommended brain atrophy as a suitable outcome measure in trials of

neuroprotection in MS.[7] When brain atrophy is measured as the percentage brain volume change (PBVC) using the registration-based SIENA (Structural Image Evaluation, using Normalization, of Atrophy) technique,[42] it provides a less variable (and hence more powerful) measurement of atrophy than segmentation-subtraction methods.[43] SIENA has been extensively used in RRMS trials and modest slowing in brain atrophy, consistent with neuroprotection, has been sometimes been observed, perhaps secondary to the known anti-inflammatory effect of the therapy being studied.[31,44]

Side-effects: MRI is generally well tolerated. There can occasionally be some discomfort and/ or claustrophobia.

Risk minimisation: trial participants will have had MRI as part of the diagnostic pathway and will be familiar with it. During the scan they have real-time contact with the radiographer with any concerns and can halt the scan if needed. Patients fitted with pacemakers or permanent hearing aids are excluded because these are metal containing products and highly unsafe for MRI.

1.8 CEREBROSPINAL FLUID (CSF) IN SPMS

The pathological substrate that results in the acquisition of non-reversible or permanent disability is axonal loss. Neurofilaments (NF) are the structural scaffolding proteins of neurones, axons and dendrites and are composed of light (NFL), medium (NFM) and heavy (NFH) chain subunits. CSF NF levels are a marker of neuroaxonal damage. Raised levels are predictive of the future development of disability.[45] Subjects with low or undetectable CSF NF levels are unlikely to progress in the next 3 to 4 years.[46,47] CSF NF is thus a marker of neurodegeneration in MS and is responsive to disease modulation.[45,48] For example, 6 to 12 months of treatment with natalizumab, in RRMS, reduced NFL levels from a mean value of 1.3 (SD=2.2) to 0.4 (SD=0.27) ng/ml ($p < 0.001$);[49] In addition levels of CSF NF do not return to normal in patients with SPMS.

An effective neuroprotective agent should reduce and normalise CSF NF levels in subjects with SPMS. The inclusion of CSF NF analysis as part of the US NMSS, PROMISE 2010 programme targeting SPMS, highlights the importance of CSF measures as a potential secondary outcome in SPMS trials.

In addition the lymphocytes present in the CSF (which are collected automatically at the same time) will be purified, analysed and phenotyped. Techniques such as deep sequencing will be used to understand lymphocyte repertoires which will enable us to examine the current status of immune activity, as well as to gain further understanding as to the aetiology of MS.

Side-effects: (for more detail see sub-protocol 2, appendix 2)
A leak of cerebrospinal fluid (CSF) can develop after a lumbar puncture which can result in a persistent headache lasting 1 to 2 days. Temporary, minor nerve injury, although rare, can occur. There is a very low risk of infection of the CSF (meningitis), bleeding inside the spinal canal and damage to the cartilage between the vertebrae.

Risk-minimisation:
Atraumatic needles will be used to reduce the incidence of post-lumbar puncture headaches.

1.9 OPTICAL COHERENCE TOMOGRAPHY (OCT) IN SPMS

OCT is a non-invasive imaging technique that uses back-scattered infrared light to detect the retinal layers. Thinning of the retinal nerve fibre layer (RNFL) is seen in progressive MS and the degree of thinning, reflecting axonal loss, is associated with quantitative measures of visual impairment.[50] Although serial OCT-measured RNFL thickness has been proposed as a measure of

neurodegeneration for clinical trials in MS, longitudinal observations are largely confined to relapsing remitting MS.[51] More recently introduced high resolution spectral-domain (SD) OCT can also measure the retinal nerve ganglion cell and inner plexiform layer (RGC+IPL), and thinning, reflecting ganglion cell loss, is seen in MS and is significantly correlated with measures of visual dysfunction.[52] Inclusion of serial SD-OCT in MS-SMART will elucidate the extent and evolution of both RNFL and RGC+IPL loss in secondary progressive MS. It will provide novel information on both axonal and neuronal cell body degeneration in this form of MS. It will also investigate the longitudinal sensitivity and clinical relevance (by correlating with low contrast visual acuity and neurological function measures) of these OCT measures of neurodegeneration and potential neuroprotection that will inform future trial design in secondary progressive MS.

Side-effects: none.

Risk-minimisation: not applicable.

2 STUDY OBJECTIVES

Summary:
The general aim is to determine the efficacy, and advance our understanding of the mechanism of action, of three putative neuroprotective repurposed drugs (flouxetine, riluzole and amiloride) versus placebo, through MRI, disability measurement, OCT and targeted CSF analysis.

Hypothesis:
Drugs that target key neurodegeneration-causing pathways in MS will be neuroprotective as demonstrated by slowing the rate of brain volume loss in people with SPMS.

Aims:
Primary: to test against placebo, the efficacy of fluoxetine, riluzole and amiloride in SPMS over a two year period.

Secondary: to advance our understanding of the mechanisms of efficacy, noting that each drug targets distinct and key disease related mechanistic pathways.

2.1 OBJECTIVES

2.1.1 Primary Objective

- To establish whether a drug, from a panel of 3 leading candidate neuroprotective drugs, slows the rate of brain volume loss in SPMS over 96 weeks using MRI-derived percentage brain volume change (PBVC).

2.1.2 Secondary Objectives

- To establish that a multi-arm trial strategy is an efficient way of screening drugs in SPMS and can become the template for future work
- To explore any anti-inflammatory drug activity using the count of new and enlarging T2 lesions

- Examining for evidence of pseudo-atrophy by MRI (to ensure reliability of the primary outcome measure)
- To examine the clinical effect of neuroprotection as measured by clinician (EDSS, MSFC, SDMT, SLCVA, relapse rate) and patient reported outcome measures (MSIS29v2, MSWSv2, Pain [NPRS, BPI and NPS]) and Fatigue [NF])
- To collect basic health economic data (ED-5D) to inform future phase III trials

2.1.3 Exploratory Objectives to Inform Mechanism

- Persistent new T1 hypointense lesion count to assess neuroprotection in new lesions
- Grey matter volume change to assess cortical neuroprotection
- MR spectroscopy to measure N-acetyl aspartate (reversal of neuronal mitochondrial dysfunction), myoinositol (prevention of glial cell inflammation) and glutamate (prevention of excitotoxicity)
- Magnetic Transfer Ratio (MTR) to evaluate myelination
- Cervical cord imaging to assess cord neuroprotection
- Composite MRI/disability scores to increase sensitivity and study interaction of treatment mechanisms
- Quantification of CSF neurofilament levels to measure neuroprotection
- CSF lymphocyte phenotyping
- OCT-measured RNFL thickness as a measure of neuroprotection

2.2 ENDPOINTS

2.2.1 Primary Endpoint

PBVC measured using the SIENA technique has been chosen as the primary outcome measure in this trial. SIENA is an automated method that registers the follow up scan to the baseline scan and produces an integral of the edge motion existing in each voxel in both scans. It thereby directly calculates the PBVC from those values.

2.2.2 Secondary Endpoints

2.2.2.1 MRI

Count of new and enlarging T2 lesions: This measure has proved sensitive in detecting efficacy of immunomodulatory drugs in preventing new lesion formation in previous trials over 2 years in both RRMS [29,31,53,54] and SPMS.[55,56] Although new and enlarging T2 lesions appear to be less relevant than brain atrophy as a measure of neuroprotection in SPMS, they will be included with brain atrophy as a core outcome measure in order to detect an unanticipated immunomodulatory effect.

Pseudoatrophy: Brain atrophy may be affected by changes in non-neuroaxonal tissue components.[56] There is potential for increased brain tissue volume loss in the early months of introduction of a drug with anti-inflammatory or anti-oedema properties, such as has been reported with intravenous methylprednisolone[57], natalizumab,[58] and beta interferon.[59] A similar "pseudo-atrophy" effect on brain atrophy was observed in the placebo-controlled lamotrigine trial in SPMS,[60] which like MS-SMART, used a measure of cerebral atrophy as the primary efficacy outcome. To specifically examine for this, a baseline scan will be acquired 24 weeks after the start of treatment.

2.2.2.2 Clinical

The current consensus, as codified workshop held in Washington DC in May 2011 and sponsored by the US MS Society and European Committee for Treatment and Research in MS (ECTRIMS), has been published.[61] Whilst recognising the major challenges of measuring disability in a chronic,

unpredictable and multifaceted disease such as MS, it also provides current expert consensus on approaches to MS clinical outcome measurement in trials. We have included outcome measures in MS-SMART that concord with the suggestions and recommendations of the review, namely: (i) expanded disability status scale (EDSS);[62] (ii) MS Functional Composite scale (MSFC);[63] (iii) single digit modality test (SDMT);[64] (iv) Sloan low contrast visual acuity (SLCVA),[65] and (v) patient reported outcomes including Multiple Sclerosis Impact Scale (MSIS29v2), [66] Multiple Sclerosis Walking Scale (MSWSv2), [67] Pain (Numerical Pain Rating Scale [NPRS],[68]Brief Pain Inventory [BPI]),[69] Neuropathic Pain Scale[70] and Fatigue (Neurological Fatigue Index [NFI]).[71]

The classical measurement tool is the Expanded Disability Status Scale (EDSS),[62] introduced over 50 years ago, based largely on neurological examination (with some history) and scoring 7 major domains. EDSS is the standard neurological examination tool in assessing MS. The scoring is complex, though as disease severity increases it is largely ambulatory. This trial, like previous SPMS trials will have EDSS entry criteria 4.0 (walking not unrestricted, but >500m) to 6.5 (patients using bilateral assistance to walk), as highlighted below. The EDSS is widely used and supported by the FDA and other regulatory authorities, though is a non-linear ordinal scale and fails to capture cognition in detail. In keeping with previous studies, we will investigate *time to* and *proportion with* confirmed (48 week or 96 week) EDSS progression.

A variety of attempts have been made to replace EDSS with alternative scoring methods such as the MSFC score,[63] which comprises quantitative tests of walking, arm function and a simple cognitive task (PASAT), with each component converted to a Z-score; or even one of the components of the MSFC, the Timed 25 Foot Walk. This will be acquired, along with a suggested refinement for the PASAT, the SDMT,[64] which measures information processing speed for visually presented stimuli, but is self-paced, with at least equal reliability and sensitivity to the presence of worsening cognitive impairment. The recommended SLCVA correlates well with diseases phase (RRMS vs SPMS), retinal nerve fibre layer (RNFL) thinning on Optical Coherence Tomography, patient reported visual impairment and MRI burden.[65] Whilst SPMS is generally non-relapsing, occasional relapses can occur, and will be recorded. Two commonly used Patient Reported Outcome Measures (PROMS) in MS trials, are the MS Impact Scale-29 v2 (MSIS29v2),[66] which measures physical and psychological well-being over the previous 2 weeks, and the MS Walking Scale v2 (MSWSv2),[67] which looks at walking. The Numerical Pain Rating Score (NPRS), [68] the Brief Pain Inventory (BPI),[69] the Neuropathic Pain Scale (NPS),[70] and the Neurological Fatigue Index (NFI)[71] will assess pain and fatigue respectively. It also seems appropriate to capture some simple health economic metrics for future phase III trials. Therefore the EuroQoL (EQ-5D) will be collected in the total cohort.[72]

In summary the clinical secondary outcome measures are:

- Expanded Disability Status Scale (EDSS)
- Timed 25 Foot Walk (T25FW)
- 9 Hole Peg Test (9HPT)
- Paced Auditory Serial Addition Test (PASAT)
- MS Functional Composite score comprising of the following:
    - Timed 25 Foot Walk (T25FW)
    - 9 Hole Peg Test (9HPT)
    - Paced Auditory Serial Addition Test (PASAT)
- Symbol Digit Modalities Test (SDMT)
- Sloan Low contrast visual acuity (SLCVA)
- Relapse rate
- Multiple Sclerosis Impact Scale v2 (MSIS29v2)
- Multiple Sclerosis Walking Scale v2 (MSWSv2)
- Brief Pain Inventory (BPI)
- Numerical Pain Rating Score (NPRS)
- Neuropathic Pain Scale (NPS)
- Neurological Fatigue Index (NFI)

- Health economics (EQ-5D)

2.2.3 Exploratory Mechanistic Endpoints

2.2.3.1 MRI:

Proportion of new and enlarging T2 lesions at 24 weeks being persistently T1 hypointense at 96 weeks: persistently T1 hypointense lesions exhibit greater axonal loss,[73] and this outcome measures was recommended for neuroprotection trials by the NMSS workshop.[7] The measure provides an indication of the extent of axonal loss associated with new inflammatory-demyelinating white matter lesions and a positive treatment effect was seen with the phase IIa trial of ibudilast.[14] It is therefore an appropriate outcome measure in MS-SMART.

Change in brain grey matter volume: There are several reasons for measuring grey matter atrophy in SPMS. First, cortical demyelinating lesions [74] and cortical neuronal loss [75] are abundant in neuropathology studies of SPMS, and could be expected to result in grey matter volume loss, which MRI studies have indeed identified as a prominent feature in SPMS;[76] thus, reduction in MRI-measured grey matter atrophy would provide supportive evidence of a treatment that prevents cortical demyelination and neurodegeneration. Secondly, there is greater atrophy over time in grey matter than in white matter in SPMS [34,77] suggesting that it will be a sensitive measure of change. Thirdly, as compared with white matter, the volume of grey matter is less affected by inflammation[78] or by treatments that reduce inflammation or tissue fluid volume (as was seen in the results of the recent lamotrigine trial).[4] Fourthly, robust correlations have been found between grey-matter atrophy and disability, both cross-sectionally and longitudinally,[34,56,57] and with cognitive impairment.[79] Grey matter atrophy will therefore be investigated as an additional measure of neuroprotection.

MR spectroscopy (MRS): Proton MR spectroscopy detects proton containing metabolites in the brain. Several of these metabolites reflect processes relevant to neurodegeneration and their study in our trial may elucidate mechanisms by which the study drugs are neuroprotective. In particular, measures of the following metabolites will be investigated and are expected to be informative in relation to specific drugs:

i) Glutamate: This is an excitatory neurotransmitter and abnormalities of glutamate metabolism have been reported in MS pathological studies.[80] An increase in glutamate has been reported in MS white matter on 3T MRI.[81] High glutamate levels may predispose to neuronal excitotoxicity and this could be modified by the effect of riluzole on glutamatergic neurotransmission.[23,82]

ii) N-Acetyl Aspartate (NAA): this neuroaxonal metabolite synthesised in mitochondria and is often reduced in MS. Partial reversibility of the NAA decrease has also been seen in MS [83] and may reflect an improvement in neuronal mitochondrial function. Measurement of NAA concentrations could assess both neuroprotection and energy metabolism through its association with axonal integrity and mitochondrial function, and could be a responsive indicator of a primary (e.g. with fluoxetine [14]) or secondary (e.g. with riluzole [84]) neuroprotective effect.

iii) Myoinositol: this metabolite comes from glial cells [85] and an elevation of myoinositol is seen in MS white matter lesions [60] and normal appearing white matter. White matter myoinositol levels are correlated with disability in established MS.[86] We will measure myoinositol concentration to assess glial cell proliferation and activation (astrocytes and microglia), which are prominent features of CNS inflammation in SPMS that potentially contribute to on-going neurodegeneration.[87]

MR magnetisation transfer ratio (MTR) to determine demyelination and remyelination: There is an exchange of magnetisation between protons that a freely mobile and those that are bound to macromolecules. The extent of this exchange provides an indication of the amount of macromolecular structure in tissue and can be measured with the MTR. Myelin has a major effect on MTR, and in MS, lower MTR is seen in demyelinated than remyelinated white matter lesions.[88,89] White matter lesion MTR increase may therefore reflect remyelination. In animal models of MS, amiloride has a neuroprotective effect which is associated with a reduction of demyelination and an

increase the trophic factors after inflammation:[14] thus, a positive effect on myelination – and hence MTR - may be detectable with this treatment. In addition, cortical demyelinating lesions are common in SPMS and although it is not possible to directly visualise most cortical lesions with current MRI techniques, the measurement of grey matter MTR may be used to infer cortical demyelination and has been shown to decline over time in SPMS.[90]

Cervical cord imaging: Much of the locomotor disability that occurs in secondary progressive MS is attributable to spinal cord involvement. An often considerable amount of cervical cord atrophy is observed in secondary progressive MS [91] and robust correlations between cervical cord atrophy and locomotor disability are observed in both cross-sectional and longitudinal studies in secondary progressive MS.[92] A method has recently been developed that has a very high intra- and inter-observer and scan-rescan reproducibility in measuring the cross-sectional area of the upper cervical cord.[93] The method, which applies active surface modelling [94] to outline the spinal cord on images acquired using a phase sensitive inversion recovery sequence, will be used to measure cervical cord atrophy in MS-SMART.

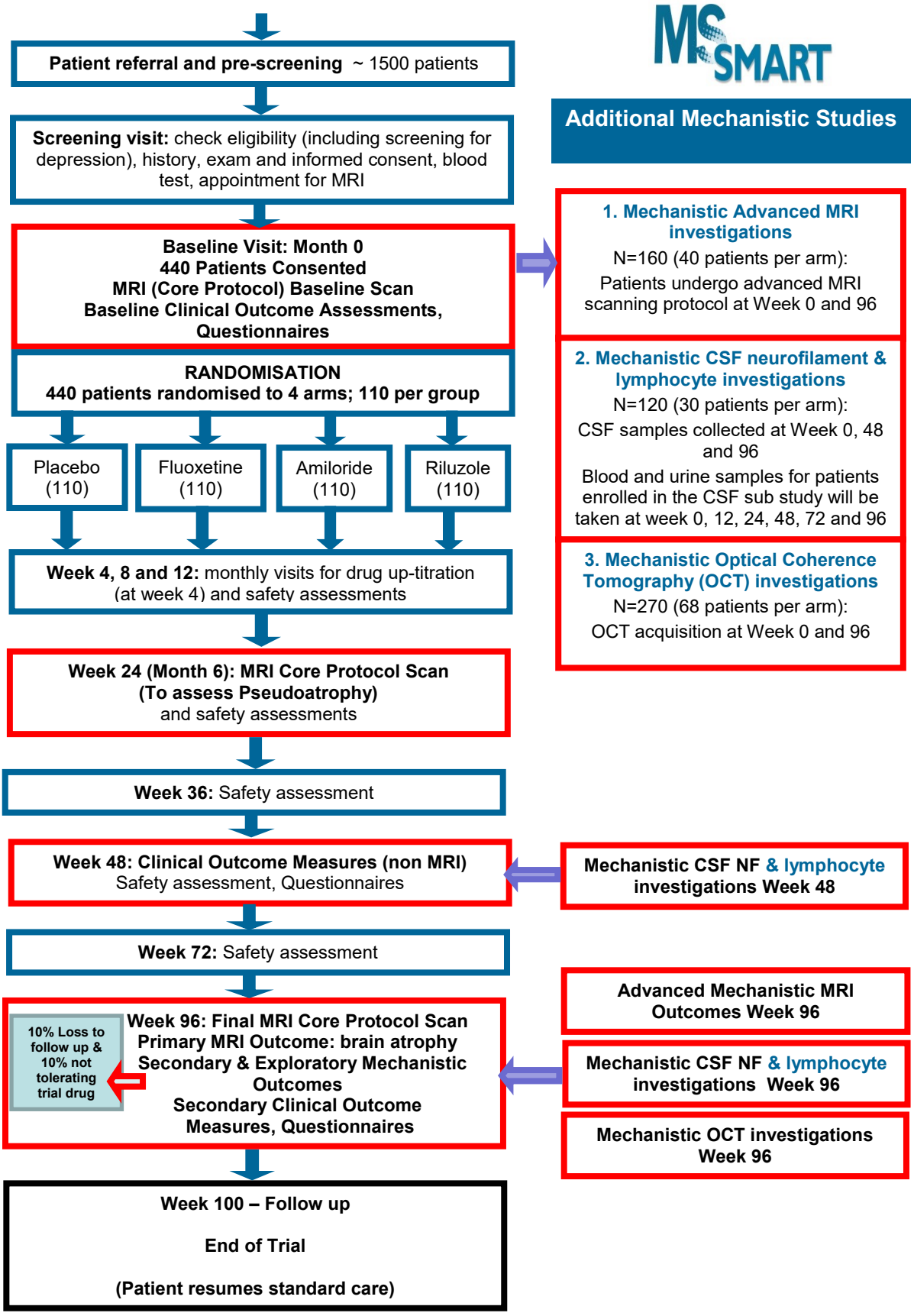
NB. MRS, MTR and cervical cord imaging are acquired with the *advanced* MRI acquisition protocol. Other MR data are acquired from the standard *core* imaging.

2.2.3.2 CSF: measurement of CSF neurofilament levels.

2.2.3.3 OCT: measurement of retinal nerve fibre layer (RNFL) and retinal nerve ganglion cell and inner plexiform layer (RGC+IPL). To determine the extent and time course of layer thinning.

3 STUDY DESIGN

MS-SMART is a multicentre, multi-arm, double blind, placebo-controlled phase IIb randomised controlled trial. A total of 440 patients with SPMS, with an entry criteria of an EDSS score of 4.0-6.5 will be equally randomised to receive placebo or one of the three active agents (amiloride 5mg bd, riluzole 50mg bd and fluoxetine 20mg bd). Patients will be followed up for 96 weeks with outcome-data collected after 0, 24, 48 and 96 weeks (see flowchart below). That is, the duration of the trial for a trial participant is 96 weeks (a telephone assessment at week 100, 4 weeks post completion will be conducted). This is standard practice for phase II trials in SPMS.


Patient referral and pre-screening ~ 1500 patients
Screening visit: check eligibility (including screening for depression), history, exam and informed consent, blood test, appointment for MRI
Baseline Visit: Month 0
440 Patients Consented
MRI (Core Protocol) Baseline Scan
Baseline Clinical Outcome Assessments, Questionnaires
RANDOMISATION
440 patients randomised to 4 arms; 110 per group
Placebo (110)
Fluoxetine (110)
Amiloride (110)
Riluzole (110)
Week 4, 8 and 12: monthly visits for drug up-titration (at week 4) and safety assessments
Week 24 (Month 6): MRI Core Protocol Scan
(To assess Pseudoatrophy)
and safety assessments
Week 36: Safety assessment
Week 48: Clinical Outcome Measures (non MRI)
Safety assessment, Questionnaires
Week 72: Safety assessment
10% Loss to follow up & 10% not tolerating trial drug
Week 96: Final MRI Core Protocol Scan
Primary MRI Outcome: brain atrophy
Secondary & Exploratory Mechanistic Outcomes
Secondary Clinical Outcome Measures, Questionnaires
Week 100 – Follow up
End of Trial
(Patient resumes standard care)
MS SMART
Additional Mechanistic Studies
1. Mechanistic Advanced MRI investigations
N=160 (40 patients per arm):
Patients undergo advanced MRI scanning protocol at Week 0 and 96
2. Mechanistic CSF neurofilament & lymphocyte investigations
N=120 (30 patients per arm):
CSF samples collected at Week 0, 48 and 96
Blood and urine samples for patients enrolled in the CSF sub study will be taken at week 0, 12, 24, 48, 72 and 96
3. Mechanistic Optical Coherence Tomography (OCT) investigations
N=270 (68 patients per arm):
OCT acquisition at Week 0 and 96
Mechanistic CSF NF & lymphocyte investigations Week 48
Advanced Mechanistic MRI Outcomes Week 96
Mechanistic CSF NF & lymphocyte investigations Week 96
Mechanistic OCT investigations Week 96

4 STUDY POPULATION

4.1 NUMBER OF PARTICIPANTS

440 adult patients (male and female) will be recruited into MS-SMART across at least 10 UK sites over a one year recruitment period.

4.2 INCLUSION CRITERIA

- Confirmed diagnosis of SPMS. Steady progression rather than relapse must be the major cause of increasing disability in the preceding 2 years. Progression can be evident from either an increase of at least one point in EDSS or clinical documentation of increasing disability in patients notes
- EDSS 4.0-6.5
- Aged 25 to 65 inclusive
- Women and men with partners of childbearing potential must be using an appropriate method of contraception to avoid any unlikely teratogenic effects of the 3 drugs from time of consent, to 6 weeks after treatment inclusive
- Women must have a negative pregnancy test within 7 days prior to the baseline visit unless not of child bearing potential (e.g. have undergone a hysterectomy, bilateral tubal ligation or bilateral oophorectomy or they are postmenopausal)
- Willing and able to comply with the trial protocol (e.g. can tolerate MRI and fulfils the requirements for MRI, e.g. not fitted with pacemakers or permanent hearing aids) ability to understand and complete questionnaires
- Written informed consent

4.3 EXCLUSION CRITERIA

- Pregnancy or breast feeding females
- Baseline MRI scan not of adequate quality for analysis (e.g. too much movement artefact)
- Significant organ co-morbidity (e.g. malignancy or renal or hepatic failure)
- Relapse within **3 months** of baseline visit
- Patients who have been treated with iv or oral steroids for an MS relapse/progression
- within **3 months** of baseline visit (these patients can undergo future screening visits once the 3 month window has expired) , patients on steroids for another medical condition may enter as long as the steroid prescription is not for multiple sclerosis (relapse/ progression)..
- Use of Simvastatin at 80mg dose within **3 months** of baseline visit (lower doses of Simvastatin and other statins are permissible)
- Commencement of fampridine within **6 months** of baseline visit

- Use of immunosupressants (e.g. azathioprine, methotrexate, cyclosporine) or first generation disease modifying treatments (β-interferons, glatiramer) within **6 months** of baseline visit
- Use of fingolimod/fumarate/teriflunomide/laquinomod/or other experimental disease modifying treatment (including research in an investigational medicinal product) within **12 months** of baseline visit
- Use of mitoxantrone/natalizumab/alemtuzumab/daclizumab if treated within **12 months** of baseline visit
- Primary progressive MS
- Relapsing-remitting MS
- Known hypersensitivity to the active substances and their excipients to any of the active drugs for this trial
- Use of an SSRI within **6 months** of the baseline visit
- Current use of tamoxifen
- Current use of herbal treatments containing St. John's Wort
- Patients with a history of bleeding disorders or currently on anticoagulants
- Use of monoamine oxidase inhibitors, phenytoin, L-tryptophan and/or neuroleptic drugs within **6 months** of the baseline visit
- Use of: lithium, chlorpropamide, triamterene, spironolactone, within **6 months** of the baseline visit
- Current use of potassium supplements
- Significant signs of Depression
- Bipolar disorder
- A Beck Depression Index score of 19 or higher
- Epilepsy/seizures
- Receiving or previously received Electroconvulsive therapy treatment
- Glaucoma
- Routine screening blood values (LFT) >/ 3 x upper limit of normal (ULN) of site reference ranges (AST/ALT, bilirubin, ɣGT)
- Potassium <2.8mmol/l or >5.5mmol/l
- Sodium <125mmol/l
- Creatinine >130µmol/l
- WBCs $<3x10^9/l$
- Lymphocytes $<0.8 \times 10^9/l$
- Neutrophil count $<1.0 \times 10^9/l$
- Platelet count $<90 \times 10^9/l$

- Haemoglobin <80g/l

4.4 Participation in Other Trials

Whilst on study, MS-SMART trial participants cannot take part in other clinical trials of Investigational Medicinal Products (or devices) until 6 weeks after finishing MS-SMART trial medication and/or final assessments. Participation in other research while taking part in MS-SMART is permissible.

5 PARTICIPANT SELECTION AND ENROLMENT

5.1 IDENTIFYING PARTICIPANTS

Initial Identification

There are several potential routes of initial identification:

- In clinics run by PIs or their associate neurologists at participating sites
- In clinics run at other MS centres/neuroscience/hospital centres, where they will be set up as Participant Identification Centres [PICS]. All referrals will require medical history records confirming MS and other relevant medical conditions from referring centres
- Using existing MS Research Databases and other databases such as SHARE, which contain the contact details of people who have expressed a wish, and consented, to be contacted directly when a study they may be suitable for is recruiting
- By General Practitioners (GPs) at routine appointments
- By self-referral from potential participants who may be made aware of the project by people who have already taken part in the study, via trial publicity events/literature or have seen the study on MS charities web-sites or been approached about it

Depending on the route of identification several processes may then be used to follow up their suitability as a participant including:

- Patients may be briefed in clinic about the study directly by a member of the clinical team; and also to ensure that the patient is likely to fulfill the general criteria to enter the trial. Patients will be given a Patient Information Leaflet
- Patients may receive an initial telephone contact from a research nurse, to explain the trial and to ensure that the patient is likely to fulfill the general criteria to enter the trial. Patients will be sent a PIL
- After a period of *at least* 24 hours after receiving the Patient Information Leaflet, the patient will be contacted again and invited for an informed consent, screening visit if they would like to proceed with the study

If required and following documented permission from the patient, their consultant / general practitioner will be contacted to provide written confirmation of the patient's medical status for use in confirming the patient's medical history for assessing eligibility. This will take place prior to the screening visit and will avoid, for example, unnecessarily bringing patients who live some distance from the clinic in for the screening visit.

*– (see below).

*A log of all patients with whom contact has been made (following receipt of the PIL) will be kept (see below).

5.2 CONSENTING PARTICIPANTS

This will occur at the screening visit. All persons involved in obtaining consent at sites must be delegated this task by the Chief Investigator/Principal Investigator and that this delegation must be documented in the delegation log.

Patients will be invited in for an informed consent visit with an appropriately experienced, qualified and Good Clinical Practice (GCP) trained member of the study team who will go through the PIS with them (explaining the aims, methods, anticipated benefits and potential hazards of the study). The patient will be given an opportunity to ask further questions. It will be explained that they are under no obligation to consent to the trial and that they can withdraw at any time during the trial without providing a reason. If they are then still willing to participate they will be asked to give their written consent. This written consent will be witnessed by the investigator who will also sign and date. It is the Principal Investigators responsibility to ensure that written informed consent is obtained from all participants before any study procedures are undertaken. They will then proceed to screening. A copy of the consent form will be given to the participant and the original copy will be retained at the study site.

Patients unable to consent for themselves due to language barriers will have access to the usual translation services provided by the site, to accommodate patient needs.

If new safety information results in significant changes in the risk/benefit assessment, the participant information sheet and consent form will be reviewed and updated if necessary. All participants, including those already being treated, will be informed of any new information, be given a copy of the revised information sheet and will be re-consented as appropriate.

5.3 SCREENING FOR ELIGIBILITY

Only individuals who are NHS employees (substantive or honorary) and who have access permissions will examine hospital and research databases for potentially eligible participants. This will be documented in the delegation log.

Once written informed consent has been obtained, trial participants will follow the trial pathway as outlined in section 7.2.

The patient's consultant / general practitioner will be sent confirmation that their patient has consented to enter the trial and further details about the trial. The consultant/GP will be asked to provide written confirmation of the patient's medical status for use in confirming the patient's medical history for assessing eligibility.

5.4 INELIGIBLE AND NON-RECRUITED PARTICIPANTS

Each MS-SMART recruiting site will be required to maintain an anonymised log of all patients who are ineligible for the trial and all eligible patients who, when approached about the trial, are not randomised because they decline participation. This information will allow generalisation of the trial results in accordance with CONSORT reporting guidelines.

Annoymised information will be collected including:

- age
- gender
- date screened
- AND

- Reason not eligible for trial participation
- OR
- Reason for declining participation despite eligibility
- OR
- Other reason for non-registration

5.5 RANDOMISATION

5.5.1 Randomisation Procedures

MS-SMART is a double blind trial with additional blinded assessment of secondary and exploratory mechanistic outcome measures. Randomisation will be performed by the research nurse via a secure web-based randomisation service (ECTU) on a 1:1:1:1 basis employing the following minimisation variables:

- sex
- age (<≥ 45yrs)
- baseline EDSS (4.0-5.5; 6.0-6.5)
- site

The minimisation algorithm will incorporate a random element to maintain unpredictability of treatment allocation.

To ensure that treatment allocation remains concealed to both staff and participants, the following measures will be taken:

- Fluoxetine, amiloride and riluzole will be over-encapsulated so they are identical in appearance and the same placebo for fluoxetine, amiloride and riluzole will be used. The fluoxetine/amiloride/riluzole placebo will be identical in appearance in order to disguise the identity
- The same number of the capsules will be prescribed for participants in each arm
- The drugs supplied from the manufacturers will be re-packaged by an organisation independent of the trial and the same organisation will supply pharmacies at participating sites directly with the trial drugs
- Drug supplies to pharmacies will be coded
- The randomisation list will be held by the ECTU to ensure that treatment allocation is concealed from the investigator's team and the participant, whilst providing provisions for emergency un-blinding. Details of the randomisation process will be available in the Investigator Site File (ISF). The identities of the co-ordinating staff whose roles in the trial require them to have access to treatment allocation codes will be recorded in the trial master file (TMF)

The randomisation system will be web-based and require a personal log-in and password. Only those individuals who have attended the site-initiation visit, are trained and are in the delegation-log can carry out this process.

Once a patient has been randomised they will be given a card to indicate they are on the trial with the emergency contact numbers for medical advice. Participants will be instructed to show this card to any healthcare professional involved in their care who is not involved in the trial. Any trial participant who withdraws from the study will not be replaced.

Emergency Un-blinding Procedures

Un-blinding may take place in situations where the safe management of the participant's medical condition necessitates knowledge of the study medication by the person(s) responsible for the participant's care. Where possible, members of the local research team should remain unblinded.

If unblinding is required the local PI/other medical staff should contact ECTU in the first instance via telephone on 07712 235781 during normal working hours (Monday to Friday 9am to 5pm). The call will then be relayed to one of the co-CIs, Jeremy Chataway or Siddharthan Chandran (or their designated deputy) for medical guidance. Out of hour calls will be diverted to the designated CI (or deputy).

The person requesting the unblinding will provide details including the protocol number and trial name, name of the requester, reason for unblinding, patient name, participant number and timeline to receive the unblinded information. If knowledge of the treatment allocation is required in order to treat the patient, the code break number will be given to the local PI/other medical staff requesting to unblind the patient. The local PI/other medical staff can then use the code break number to reveal the participants treatment allocation. In this way, the treatment will be unblinded at the local site but not to the ECTU member of staff or CIs.

- The local PI/ medical staff will deal with the medical emergency (upon receipt of the treatment allocation revealed by the code break number)
- Details of the code break will be documented on the code break form and filed in the local Site File
- Code breaks will also be documented at the end of the study in the statistical report.
- The CI/ investigator team will notify the Joint Research Office for the UCL sponsor (JRO) of a code break and provide details of the necessity to un-blind
- The CI/PI will also notify trial committees (in accordance with their charter)

The following procedure will be used to un-blind for the submission of a SUSAR report to the regulatory agencies:

- ECTU will hold the code break list during normal working hours (Monday – Friday, 9am to 5pm)
- A member of the JRO sponsor's office will contact the ECTU during normal working hours via telephone on 0131 651 9910 in the first instance, requesting unblinding information from the randomisation list
- ECTU will provide their email address and name for the request to be formalised in an email
- The sponsor will provide in the email the protocol number and trial name, name of the requester, reason for unblinding, patient name, participant number and timeline to receive the unblinded information
- The sponsor will provide the unblinded information on the e-SUSAR website
- This information will not be forwarded to the trial team and will be kept in the JRO sponsor file

5.5.3 Withdrawal of Study Participants

Trial participants are free to withdraw from the trial at any point or can be withdrawn by the investigator. If withdrawal occurs, the primary reason for withdrawal should be documented in the participant's Case Report Form (CRF). Trial participant withdrawals will not be replaced.

If a participant discontinues study drug this does not necessarily constitute withdrawal. In this case all attempts should be made to follow up the participant as per protocol and/or re-commencing treatment (if discontinuation was due to adverse events which have subsequently resolved. Refer to section 10). It is important that all patients (with the exception of those that are withdrawn from the trial) undertake the final MRI scan at 96 weeks.

The IDMC can request trial or trial arm suspension/termination, according to the terms of the IDMC charter, for example unacceptable adverse events.

6 INVESTIGATIONAL MEDICINAL PRODUCT AND PLACEBO

6.1.1 Treatment Allocation

Following randomisation study drug will be dispensed by the site pharmacy.
Patients will be randomised to one of the following arms:

- Fluoxetine 20 mg twice per day (20 mg once per day for first 4 weeks)
- Riluzole 50 mg twice per day (50 mg once per day for first 4 weeks)
- Amiloride 5 mg twice per day (5 mg once per day for first 4 weeks)
- Matched placebo twice per day (1 capsule a day for first 4 weeks)

6.1.2 Name and description of investigational medicinal products

Investigational drugs for this trial: (NB other brands using the same active substances will not be used).

Riluzole (RILUTEK 50 mg film-coated tablets)
Riluzole is a member of the benzothiazole class. Chemically, riluzole is 2-amino-6-(trifluoromethoxy) benzothiazole. Its molecular formula is $C_8H_5F_3N_2OS$ and its molecular weight is 234.2 Riluzole is a white to slightly yellow powder that is very soluble in dimethylformamide, dimethylsulfoxide and methanol, freely soluble in dichloromethane, sparingly soluble in 0.1 N HCl and very slightly soluble in water and in 0.1 N NaOH. RILUTEK (riluzole) is available as a capsule-shaped, white, film-coated tablet for oral administration containing 50 mg of riluzole. Each tablet is engraved with “RPR 202” on one side. The Marketing Authorisation Holder is Aventis MA EU/1/96/010/001. For the purpose of the trial commercial Rilutek will be over-encapsulated by a third party manufacturer holding a MIA IMP in conformance with EU cGMP, in order to achieve the blinding.

Amiloride HCl
Amiloride is an antikaliuretic-diuretic agent, a pyrazine-carbonyl-guanidine that is unrelated chemically to other known antikaliuretic or diuretic agents. It is the salt of a moderately strong base (pKa 8.7). It is designated chemically as 3, 5- diamino-6-chloro- N-(diaminomethylene) pyrazinecarboxamide monohydrochloride, dihydrate and has a molecular weight of 302.12. Its empirical formula is $C_6H_8ClN_7O•HCl•2H_2O$. Each tablet for oral administration contains 5 mg of Amiloride HCl, calculated on the anhydrous basis. For the purpose of the trial commercial Amiloride will be over-encapsulated by a third party manufacturer holding a MIA IMP in conformance with EU cGMP, in order to achieve the blinding.

Fluoxetine
Fluoxetine hydrochloride is a selective serotonin re-upake inhibitor. It is designated (±)-N-methyl-3-phenyl-3-[(a,a,a-trifluoro-p-tolyl)oxy]propylamine hydrochloride and has the molecular formula of C17H18F3NO•HCl. Its molecular weight is 345.79. Fluoxetine hydrochloride is a white to off-white crystalline solid with a solubility of 14 mg/mL in water.
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Fluoxetine 20mg capsules will be over-encapsulated by a third party manufacturer holding a MIA IMP in conformance with EU cGMP, in order to achieve the blinding.

Placebo
Identical placebo to match the 3 active drugs will be manufactured by the same manufacturer over-encapsulating the IMPs. An equivalent amount of inert excipient will be used in place of the active ingredients. The placebo will be packaged for the trial by a MIA IMP holder third party manufacturer in UK.

The blinding will be achieved by over-encapsulation of the IMP and producing an identical placebo. Further detail is given in section 6.3 and 6.4.

6.1.3 Labelling, packaging and storage

The trial drug will be stored below 25°C in a dry place and protected from light. All active and placebo will be packaged by the MIA IMP holder UK third party manufacturer in Polyethylene Bottles containing the same number of capsules.

Labelling will be in a blinded fashion and in accordance to requirements of EU GMP Annex 13. In order to maintain blinding, bottles will be coded and both shelf life and storage conditions will be adjusted to maintain blinding.

6.1.4 Dispensing, handling and drug accountability

The bottles will be dispensed according to site and study-specific SOPs by the local site pharmacy. During the study a subject specific accountability log will be kept to record each dose of the trial drug dispensed for each trial participant. These logs will be monitored. All used returned bottles will be kept for potential reconciliation by the sponsor; they will be discarded by the research staff according to local procedures, upon authorisation from the sponsor.

The **Drug Accountability Log** must be completed to record each dose of the trial drug dispensed for each trial participant.

6.1.5 Summary of Product Characteristics (SmPC) or Investigators Brochure

For fluoxetine, and amiloride, (both IMPs provided by Actavis) and riluzole (Rilutek, provided by Sanofi) the latest SmPC will be used as their reference document and this can be accessed from electronic medicines compendium: www.medicines.org.uk/

6.2 PLACEBO

The MS-SMART placebo comprises a size 00 capsule identical to the over-encapsulated Fluoxetine/Amiloride/Riluzole. The trial medications are described in Section 6.1.2.

6.3 DOSING REGIME

Following randomisation, study drug will be dispensed by the site pharmacy.

Patients are prescribed 1 capsule a day (OD), orally at baseline. At 1 month they will up-titrate to 1 capsule twice a day (bd) orally for the remainder of the study; no relationship to food is required.

That is:

- Fluoxetine 20 mg twice per day (20 mg once per day for first 4 weeks)
- Riluzole 50 mg twice per day (50 mg once per day for first 4 weeks)
- Amiloride 5 mg twice per day (5 mg once per day for first 4 weeks)
- Placebo –1 capsule twice per day (1 capsule once per day for first 4 weeks)

6.4 DOSE MODIFICATION AND STOPPING RULES

The following terms will be used:

- ‘Full dose’ – 1 capsule TWICE a day.
- ‘Half dose’ - 1 capsule ONCE a day.
- ‘Zero dose’ – 0 capsules taken.

As the trial is blinded, participants will not know which treatment they are on.

At Baseline:

Participants will be prescribed the ‘half dose’ for the first 4 weeks.

At 4 Weeks:

Participants will up-titrate to ‘full dose’ for the remainder of the study.

Dose Modification (if required) – Baseline to Week 4:

If a participant cannot tolerate the ‘half dose’ (duration according to the Investigator’s discretion) the dose should be stopped (‘zero dose’). The participant will be re-challenged 2-4 weeks later with the ‘half dose’ at the discretion of the PI. The participant should then take the ‘half dose’ for 4 weeks before up-titrating to the ‘full dose’.

If however, the participant cannot tolerate the ‘half dose’ when re-challenged (duration according to local PI discretion), the participant should be off medication for the remaining duration of the trial. The participant will remain in trial follow-up.

Dose Modification as a result of Adverse Events:

At the discretion of the local PI the participant can down-titrate to ‘half dose’ if Adverse Events (AEs) occur. This does not preclude the participant increasing the dose subsequently (up to the week 24 visit) to the ‘full dose’.

If a participant cannot tolerate the ‘half dose’ (duration according to the Investigator’s discretion) the dose should be stopped (‘zero dose’). The participant can be re-challenged 2-4 weeks later with the ‘half dose’ at the discretion of the PI. If they cannot tolerate the ‘half dose’ again (duration according to local PI discretion) the participant should be off medication for the remaining duration of the trial. The participant will remain in trial follow-up.

If a participant cannot tolerate the ‘full dose’ (duration according to the Investigator’s discretion), the dose should be reduced to ‘half dose’. The participant will be re-challenged at 2-4 weeks later. If they cannot tolerate the ‘full dose’ again (duration according to local PI discretion), they should reduce to ‘half dose’. The participant will be re-challenged a second time 2-4 weeks later with the full dose. If the participant cannot tolerate that dose again (duration according to the Investigator’s discretion) they should reduce to the half dose for the remaining duration of the trial.

At the Investigator’s discretion, participants can be re-challenged with a higher dose a maximum of 2 times. The participant’s dose must be fixed at or by the week 24 visit for the remaining duration of the study (unless in response to managing AEs).

A full record of medication administered will be kept.

6.5 PARTICIPANT COMPLIANCE

At each visit participants will bring back the used and unused study drug and will be asked about compliance. Participants will be asked to detail drug compliance over the past 30 days using a diary card to record the number of capsules taken and to indicate any reason for non-compliance. Compliance will be assessed according to the diary card however a pill count will also take place. This information will be collected on the CRF.

Non-compliance to the protocol study procedures will be documented by the investigator and reported to the sponsor as required in section 14.2. Persistent non-compliance may lead the participant to be withdrawn from the study. Follow up as per the protocol will be attempted for all non-compliant participants.

Primary analyses will be intention-to-treat (ITT) using a dataset of all randomized participants, irrespective of treatment actually taken. A complete case approach will be taken for missing data in analysis of each outcome. Participants will be considered compliant with treatment if they reported taking, on average, at least 90% of their prescribed medication in the 30 days preceding each visit.

6.6 OTHER MEDICATIONS

6.6.1 Non-Investigational Medicinal Products

Participants in the CSF and lymphocyte phenotyping sub study will be administered a local anaesthetic prior to lumbar puncture. The anaesthetic is classed as a NIMP. This trial will not be using any Non Investigational Medicinal Products (NIMPs) other than in the CSF and lymphocyte phenotyping sub study.

6.6.2 **Permitted Medications**

All medications are permitted, apart from those outlined in section 6.3. Where required, for example to treat a relapse, patients on trial treatment and in follow up can receive steroid therapy.

The following should be used with caution as drug interactions are possible:

- Angiotensin-converting inhibitors
- NSAIDs
- Ciclosporin
- Inhibitors of CYP 1A2 (e.g. caffeine, diclofenac, diazepam, nicergoline, clomipramine, imipramine, fluvoxamine, phenacetin, theophylline, amitriptyline and quinolones)
- Inducers of CYP 1A2 (e.g. cigarette smoke, charcoal-broiled food, rifampicin and omeprazole) could increase the rate of riluzole elimination

6.6.3 **Prohibited Medications**

Since the patients will be blinded to the IMP, all individual absolute contraindicated drugs are absolutely contraindicated as a whole. All ‘use with caution’ are ‘use with caution’ as a whole.

Amiloride

Absolute contraindications:

- Lithium
- Chlorpropamide
- Potassium supplements
- Potassium retaining diuretics e.g. triamterene, spironolactone

Riluzole

No absolute contra-indications

Fluoxetine

Absolute contra-indications:

- **Monoamine oxidase inhibitors**
- **Phenytoin**
- **L-tryptophan) and/or neuroleptic drugs**
- **Lithium**
- **SSRI anti-depressants**
- **Metoprolol**

Use with caution:

CYP2D6 isoenzyme inhibitors: (such as flecainide, encainide, carbamazepine and tricyclic antidepressants), should be initiated at or adjusted to the low end of their dose range.

Tamoxifen efficacy maybe reduced

Drugs that prolong the QT interval should be used with caution.

Care with Oral anticoagulants, CYPROHEPTADINE, DRUGS INDUCING HYPONATREMIA AND DRUGS LOWERING THE EPILEPTOGENIC THRESHOLD.

St John's Wort:

If treatment of in-trial depression is needed, the following are allowable (that is can be safely added to fluoxetine): mirtazapine. venlafaxine, duloxetine and agomelatine.

6.6.4 Management of laboratory abnormalities

Potassium

- If Potassium <2.8mmol/l or >5.5mmol/l, the medication will be suspended. Repeat Potassium at 2-4 weeks. If Potassium <2.8mmol/l or ≤ 5.5mmol/l continue to monitor, and consider re-challenge. If not suspend for a further 2-4 weeks.
- If Potassium remains <2.8mmol/l or >5.5mmol/l discontinuation of treatment is recommended.

Sodium

- If Sodium <125mmol/l, the medication will be suspended. Repeat Sodium at 2-4 weeks. If Sodium <125mmol/l continue to monitor, and consider re-challenge. If not suspend for a further 2-4 weeks.
- If Sodium remains <125mmol/l discontinuation of treatment is recommended.

Liver function tests

- If ALT or AST or bilirubin or ɣGT <3 times the ULN, commencement, continuation or advancement of placebo/IMP is reasonable.
- If ALT or AST or bilirubin or ɣGT >3 times the ULN, confirm by repeat measurement within a week. If abnormalities persist, consider dose reduction to 'half dose' (if taking medication bd, i.e. 'full dose') or stopping medication 'zero dose' (if taking 'half dose'). Repeat parameters at 2-4 weeks. If parameters improve towards baseline, continue to monitor, and consider re-challenge.
- If ALT or AST or bilirubin or ɣGT >5 times the ULN discontinuation of treatment is recommended.

Renal function (creatinine)

- If creatinine >130µmol/l, confirm by repeat measurement within a week. If abnormalities persist, reduce to 'half dose' (if taking medication bd, i.e. 'full dose') or stopping medication 'zero dose' (if taking 'half dose'). Repeat parameters at 2-4 weeks. If parameters return to baseline, continue to monitor, and consider re-challenge.
- If creatinine remains >130µmol/l discontinuation of treatment is recommended.

Full blood count (FBC)

- If neutrophil count reduces to $<1.0x10^9$ /l suspend medication. Repeat parameters at 2-4 weeks. If parameters improve towards baseline, continue to monitor, and consider re-challenge. If neutrophil count remains $<1.0x10^9$ /l discontinuation of treatment is recommended.
- If platelet count reduces to $<50\ x10^9$ /l suspend medication. Repeat parameters at 2-4 weeks. If parameters improve towards baseline, continue to monitor, and consider re-challenge. If platelet count remains $<50\ x10^9$ /l discontinuation of treatment is recommended.

Haemoglobin (Hb)

If haemoglobin <80g/l suspend medication and commence local protocol for investigating anaemia.

Repeat parameters at 2-4 weeks. If parameters improve towards baseline, continue to monitor, and consider re-challenge. If haemglobin remains <80g/l discontinuation of treatment is recommended.

7 STUDY ASSESSMENTS

7.1 SAFETY ASSESSMENTS

Safety assessments will include the monitoring of Adverse Events and Serious Adverse Events by the Investigator. For more details on Adverse Events please refer to section 10.

Safety Bloods: A blood sample will be taken at screening and routine blood levels will be checked to identify any values which are more than 3 times the upper limit of normal site reference ranges [for LFT] or below acceptable levels [WBC, haemoglobin, lymphocyte, neutrophil, platelet and sodium levels], above acceptable levels [creatinine] or above/below acceptable levels [potassium]. Those patients will not be eligible for trial entry.

A repeat blood sample may be taken at baseline to re-test one or more values out of range as necessary.

Routine safety bloods will be collected at trial assessments and processed at local site laboratories in accordance with local policy and guidelines. Putative Serum biomarkers such as lactate

dehydrogenase (LDH) and haptoglobin will be monitored at the UCL site only *to monitor progression of the disease.*

Women of childbearing potential (WOCBP, defined as women who are not postmenopausal (12 months since last menses or permanently sterilised) will require a negative urine pregnancy test prior to each MRI scan compliant with National Guidelines for MRI and prior to study inclusion (within 7 days of baseline visit). WOCBP will be required to use adequate contraception during the trial.

Safety assessments will include the monitoring of adverse events and serious adverse events by the site investigator. For more details on adverse events please refer to Section 10.

Depression: Patients with a score of 19 or higher on the BDI at the screening visit will not be eligible for trial entry.

CSF Neurofilament and CSF lymphocyte phenotyping **Sub Study** ***(This section only applicable to centres participating in this sub-study):***

Those participants on the CSF neurofilament and CSF lymphocyte phenotyping sub study will have consented to this sub study. These samples will be processed in a central laboratory that holds an appropriate licence to hold, process and analyse tissue samples. Details on sample handling, processing, storage and shipment are in the CSF neurofilament and CSF lymphocyte phenotyping Sub study protocol 2.

7.2 STUDY ASSESSMENTS

The Schedule of Events below provides details of all assessments.

	MS-SMART Schedule of Events													
Clinic Visit Number	**0**		**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**		**9**		
Visit Type	**Screening**	**Baseline MRI Visit**	**Baseline**	**Titration**							**End of Study MRI {must be done while on IMP]**	**End of study**	**Follow-up (Tel.)**	**Unscheduled visit**
			Wk 0	**Wk 4**	**Wk 8**	**Wk 12**	**Wk 24**	**Wk 36**	**Wk 48**	**Wk 72**	**Wk 96**	**Wk 96**	**Wk 100**	
Window				**+/- 1wks**	**+/- 1wks**	**+/- 1wks**	**+/- 2wks**	**+/- 2wks**	**+/- 2wks**	**+/- 2wks**		**+2 weeks**	**+/- 4 days**	
Informed consent	**x**													
Inclusion/exclusion criteria review	**x**		**x**											
Demography	**x**													
Review of medical and MS History	**x**													**x**
EDSS - Review [Treating physician]	**x**													
EDSS – [Assessing physician]			**x**						**x**			**x**		
Physical Examination	**x**								**x**			**x**		**x**
Vital signs	**x**								**x**			**x**		**x**
Safety bloods CR&E, FBC, LFT (inc.GammaGT)†	**x**			**x**	**x**	**x**	**x**	**x**	**x**	**x**		**x**		**xd**
Urine pregnancy test†		**xa**	**xa**				**xa,e**					**xa,e**		
Book MRI	**x**					**x**				**x**				
Core MRI		**xb**					**x**				**xc**			
Compliance assessment				**x**	**x**	**x**	**x**	**x**	**x**	**x**		**x**		
Relapse assessment (count / grade)	**x**		**x**	**x**	**x**	**x**	**x**	**x**	**x**	**x**		**x**		**x**
Adverse events			**x**	**x**	**x**	**x**	**x**	**x**	**x**	**x**		**x**	**x**	**x**
Concomitant Medication	**x**		**x**	**x**	**x**	**x**	**x**	**x**	**x**	**x**		**x**		**x**
Randomisation			**x**											
Prescription issued			**xf**	**x**	**x**	**x**	**x**	**x**	**x**	**x**				
Drug up-titration				**x**										
Follow-up													**x**	
Blinding Questionnaire												**x**		
MSFC (9HTP, 25TFW, PASAT)			**x**						**x**			**x**		
SDMT			**x**						**x**			**x**		
SLCVA			**x**						**x**			**x**		

MSIS-29v2			x						x			x		
MSWSv2			x						x			x		
NFI - PRO			x						x			x		
NPRS, NPS and BPI			x						x			x		
EQ-5D			x						x			x		
BDI	x													

Clinic Visit Number	**0**		**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**		**9**		
Visit Type	**Screening**	**Baseline MRI Visit**	**Baseline**	**Titration**							**End of study MRI**	**End of study**	**Follow-up (Tel.)**	**Unsheduled visit**
			Wk 0	**Wk 4**	**Wk 8**	**Wk 12**	**Wk 24**	**Wk 36**	**Wk 48**	**Wk 72**		**Wk 96**	**Wk 100**	
Window				**+/- 1wks**	**+/- 1wks**	**+/- 1wks**	**+/- 2wks**	**+/- 2wks**	**+/- 2wks**	**+/- 2wks**		**+2wks**	**+/- 4 days**	
OCT (subgroup)			**xg**									**xc**		
CSF Neurofilament/lymphocytes (subgroup) Lumbar puncture††			**xg**						**x**			**xc**		
CSF Neurofilament/lymphocytes (subgroup) Blood & urine samples††			**xg**			**x**	**x**		**x**	**x**		**xc**		
Advanced MRI (subgroup)		**x**										**x**		**x**
Staff Present														**x**
Treating Neurologist	y								y			y		**x**
Independent Assessing Neurologist			y						y			y		
Study Nurse	y		y	y	y	y	y	y	y	y		y		**xd**

Notes
a= must occur prior to MRI and at baseline visit, within 1 week (-7 days) Applies to WOCBP. Repeat testing will be dependent on timing of assessments.
b= must occur before baseline visit
c= must occur within 7 days prior to the final clinic visit or at the final clinic visit (visit 9)
d= at PI discretion
e= pregnancy test prior to MRI (within 7 days prior)
f= Prescription after randomisation
g= Must occur before or on day of baseline visit (i.e. procedures conducted before trial treatment commences)
† Samples processed at local site laboratories
†† samples processed at central laboratory

Screening Visit (Visit 0):

Screening (- 30 days)
Patients will be identified as indicated above (see section 5.1 for further clarification) and if deemed suitable to enter the trial will be informed about the trial and provided with a patient information leaflet. They will be asked to contact the co-ordinating centre if they would like to attend for a screening assessment or the recruiting centre will contact the patient to arrange a screening assessment.

At the screening visit, patients will have further opportunities to discuss the trial with members of the clinical team and if they wish to proceed, will provide written, *informed consent*.

The following will be assessed and recorded:

- inclusion/exclusion criteria
- demography
- review of medical and MS history
- review EDSS [treating physician]
- physical examination
- vital signs
- safety bloods (Cr&E,FBC, Hb, LFT inc. ɣGT)
- relapse assessment (count and grade)
- concomitant medication
- BDI-II

Anticipated visit duration = 60 minutes

If any of the safety bloods are clinically significant (CS), they can be repeated at Baseline and the repeat safety blood result(s) at Baseline will be used to assess eligibility.

If a patient is ineligible at Screening (apart from CS safety bloods), they can be re-screened (where appropriate) after a minimum period of 1 month.

Core Brain MRI (Completed by Week 0)
WOCBP will have a urine pregnancy test within 7 days prior to their MRI, compliant with National Guidelines for MRI.

Sub-studies (Completed by Week 0)
Those participants who have consented to additional MRI imaging (advanced MRI) will have this procedure at the time of the baseline MRI visit. Participants taking part in CSF neurofilament and CSF lymphocyte phenotyping testing (lumbar puncture, blood and urine) &/or Optical Coherence Tomography (OCT) will have these procedures completed either before the baseline visit or during the baseline visit (before trial treatment commences).

Visit Number 1:
Baseline/Randomisation (Week 0)
At baseline, the following assessments will be completed:
inclusion/exclusion criteria confirmation
EDSS [assessing physician]
urinary pregnancy test (within 7 days of baseline visit, where applicable)
MS Functional Composite (MSFC: 9HPT,25TFW,PASAT)
Symbol Digit Modalities Test (SDMT)
Sloan Low contrast visual acuity (SLCVA)
Multiple Sclerosis Impact Scale v2 (MSIS29)
Multiple Sclerosis Walking Scale v2 (MSWS)
NFI
NPRS, BPI and NPS
Health economic questionnaire (EQ-5D)

Repeat safety bloods (if required)
patients will be assessed to confirm they have not had a relapse (count/grade) between screening and baseline
adverse events
concomitant medication

Randomisation will occur once all baseline assessments have been done and the participant has been confirmed as eligible. Trial participants will then receive their baseline (Week 0) drug prescription. Participants will take the 'half dose' for the first 4 weeks, starting treatment the morning after the Baseline/Randomisation visit.

Anticipated visit duration = 60 minutes

Visit Number 2:
Week 4 (+/- 1 week)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC, Hb, LFT inc. ɣGT)
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication

The dose will be up-titrated to the maximum 'full dose'.

Trial participants will receive their next prescription.

Anticipated visit duration = 20 minutes

Visit Number 3:
Week 8 (+/- 1 week)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC, Hb, LFT inc. ɣGT)
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication

Trial participants will receive their next prescription.

Anticipated visit duration = 20 minutes

Visit Number 4:
Week 12 (+/- 1 week)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC,Hb, LFT inc. ɣGT)
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication

The next MRI appointment is booked.

Trial participants will receive their next prescription.

Anticipated visit duration = 20 minutes

Those patients consented to the CSF neurofilament and CSF lymphocyte phenotyping sub study will have their blood and urine sampled +/- 14 days of visit 4.

Visit Number 5:
Week 24 (+/- 2 weeks)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC,Hb, LFT inc. ɣGT)
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication

Trial participants will receive their next prescription.

Anticipated visit duration = 20 minutes

Core Brain MRI and CSF neurofilament and CSF lymphocyte phenotyping **sub study**
All patients will undergo the 24 week (6 month) brain core MRI scan +/- 14 days of visit 5.
WOCBP will have a urine pregnancy test prior to their MRI, compliant with National Guidelines for MRI.

Those patients consented to the CSF neurofilament and CSF lymphocyte phenotyping sub study will have their blood and urine sampled +/- 14 days of visit 5.

Visit Number 6:
Week 36 (+/- 2 weeks)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC,Hb, LFT inc. ɣGT, serum biomarkers [UCL])
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication

Trial participants will receive their next prescription.

Anticipated visit duration = 20 minutes

Visit Number 7:
Week 48 (+/- 2 weeks)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC,Hb, LFT inc. ɣGT,serum biomarkers [UCL])
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication
physical examination
vital signs

They will undertake the following assessments
*EDSS [assessing physician]
MS Functional Composite (MSFC - 9HPT,25TFW,PASAT)
Symbol Digit Modalities Test (SDMT)
Sloan Low contrast visual acuity (SLCVA)
Multiple Sclerosis Impact Scale v2 (MSIS29)
Multiple Sclerosis Walking Scale v2 (MSWS)
NFI

NPRS, BPI and NPS
Health economic questionnaire (EQ-5D)

*A telephone EDSS assessment (assessing physician or nurse) is permissible if the participant is unable to attend clinic, however the participant should attend their appointments wherever possible and the EDSS be conducted face-to-face.

Trial participants will receive their next prescription.

Anticipated visit duration = 60 minutes.

CSF Neurofilament and CSF lymphocyte phenotyping **Sub-study**
Patients consented to the CSF neurofilament and CSF lymphocyte phenotyping sub study will have their **CSF**, blood and urine sampled +/- 14 days of visit 7.

Visit Number 8:
Week 72 (+/- 2 weeks)
Trial participants return to the research clinic and the following assessments will be completed:

safety bloods (Cr&E,FBC, Hb, LFT inc. ɣGT, serum biomarkers [UCL])
compliance assessment
relapse assessment (count/grade)
adverse events
concomitant medication

The next MRI appointment is booked.

Trial participants will receive their next prescription.

Anticipated visit duration = 20 minutes.

CSF Neurofilament Sub-study
Patients consented to the CSF neurofilament and CSF lymphocyte phenotyping sub study will have their blood and urine sampled +/- 14 days of visit 8.

Visit Number 9:
Week 96 (+2 weeks)

End of Study MRI:

Core brain MRI
Core brain MRI will be completed at this visit (or within 7days prior to visit 9). WOCBP will have a urine pregnancy test prior to their MRI, compliant with National Guidelines for MRI.

Advanced MRI, CSF Neurofilament and CSF lymphocyte phenotyping **and OCT Sub-studies**
Those participants that have consented to additional MRI imaging (advanced MRI) and/or CSF, blood and urine Neurofilament and CSF lymphocyte phenotyping and/or Optical Coherence Tomography (OCT) will have these assessments completed at this visit (or within 7 days prior to visit 9).

END OF TREATMENT SCHEDULE
Trial participants finish their study medication and the following assessments will be completed:

- safety bloods (Cr&E,FBC,Hb, LFT inc. ɣGT, serum biomarkers [UCL])
- compliance assessment
- relapse assessment (count/grade)

- adverse events
- concomitant medication
- physical examination
- vital signs
- blinding questionnaire

They will undertake the following assessments
*EDSS [assessing physician]
MS Functional Composite (MSFC - 9HPT,25TFW,PASAT)
Symbol Digit Modalities Test (SDMT)
Sloan Low contrast visual acuity (SLCVA)
Multiple Sclerosis Impact Scale v2 (MSIS29)
Multiple Sclerosis Walking Scale v2 (MSWS)
NFI
NPRS and BPI
Health economic questionnaire (EQ-5D)

*A telephone EDSS assessment (assessing physician or nurse) is permissible if the participant is unable to attend clinic, however the participant should attend their appointments wherever possible and the EDSS be conducted face-to-face.

Anticipated visit duration = 60 minutes.

Telephone Follow up:
Week 100 (+/- 4 days)
Trial participants will be contacted by telephone to monitor for any AEs occurring within 30 days following completion of trial medication. AEs occurring in the follow up period are to be reported as outlined in section 10.

The patients will then exit the study and their usual best medical care will resume.

Unscheduled visits
Participants will be instructed to contact the local study team between scheduled visits should they suspect a relapse. The proforma shown in appendix 4 will be completed and recorded in the CRF. An unscheduled visit (either for or not for a relapse) can occur at the local study team's discretion and the following will be assessed and recorded:
review of current medical and MS history
physical examination
vital signs
safety bloods at PI discretion
relapse assessment (as per appendix 4)
concomitant medication
adverse events

Missed visits
It is vital that participants attend all clinic visits as scheduled. In the event that a visit cannot be re-scheduled the following measures may be taken:

- Concomitant medication and adverse event assessment by telephone
- Questionnaires are due at Visit 7 and 9 and may be sent to the participant for completion (please note this does not apply to the baseline questionnaires. Baseline questionnaires must be completed in clinic)
- Visit 7 and 9 – telephone EDSS assessment as outlined above

  Please note that these measures apply to participants on treatment and those off treatment and in follow up.

8 DATA COLLECTION

8.1 Data Entry

Research staff at sites will enter data onto an eCRF via a secure, web-based portal. Access will be password protected and limited to nominated staff recorded on the delegation log. Members of staff will be identifiable by a unique username and password.

Site staff are responsible for recording full and accurate data onto the database. Anonymised data only will be recorded on trial paperwork and the eCRF. Designated staff at ECTU will follow ECTU SOPs to obtain missing data and resolve queries with site staff and to ensure data quality and completeness of data across sites. Data management will comply with ECTU SOPs TM 06, 07 and 09.

The Data Analysis will be conducted independently of data entry. ECTU statisticians will be responsible for the Data Analysis.

The trial database is a bespoke, ECTU-developed web-interface. Each component of the interface is built and maintained according to ECTU Standard Operating Procedures (ECTU SOPs I.T. 02-09). The trial database includes in-built systems to ensure the validity and quality of the data, and to generate queries. Cross validation will be employed and data entry will be single entry. The trial data will be held on a secure server at Edinburgh Clinical Trials Unit (ECTU), protected by network firewall and antivirus software.

The database servers and file servers are backed up on a daily basis. LTO (linear tape open) and DLT (digital linear tape) tape backup devices with autoloaders write the data to the backup tapes. Specialist software is used to manage the catalogues and automate the scheduled execution of the data backup processes. Full backups occur over the weekend and incremental backups occur during the week. Backup tapes are stored in data grade fire safes capable of withstanding intense temperatures of fire for over two hours. The integrity of backups is tested with sampled restores of data to alternative non-production locations. Data storage media used in the backup processes is controlled and decommissioned appropriately. Network attached storage devices are also used to store replicated copies of large file stores. These devices are protected by strict firewalls.

Data snapshots of databases are stored on access controlled fileservers whenever a statistician performs analysis for data monitoring committees, interim analysis or final analysis. This ensures that their findings can be reproduced. Data that is stored within databases associated with CTIMP studies will have full audit logs enabled so that a history is maintained of who did what and when.

Physical security - Access to the server room is controlled and is limited to essential personnel only.

Logical security 1 - Access to the web-interface will be via the encrypted Secure Socket Layer protocol. Authorisation will be via a unique username/password combination.

Logical Security 2 - Read-only access to the data repository will be provided to analysts on a named basis for a fixed period of time to allow analyses to be undertaken.

8.2 Data transfer

Data transfer where will be covered by agreements. All transfer of data will be in accordance with the Data Protection Act 1998 and the UCL Information Security Policy and Trust Information Governance Policy.

Source Data (in medical notes)	Trial Data (Paper source)	Trial Data (Electronic source)
Medical and MS history Diagnostic investigations Blood results Annotations in notes Correspondence (e.g. GP letters)	Relevant results/observations documented in the notes, transcribed onto source document record (copy to be kept at site)	Details transcribed on source document record inputted onto eCRF held on secure trial website (anonymised electronic data held – ECTU)
	Patient questionnaires/trial specific paperwork completed during the trial will form **Source Data** and be kept at site	Details transcribed on questionnaires/trial specific paperwork inputted onto eCRF (as above)

8.3 Source Data

Data recorded by designated trial staff on the trial specific eCRF will be obtained from the source document, i.e. the medical notes. The source data includes medical and MS history, diagnostic investigations, safety blood results, medical assessments and annotations in the notes. Data completed on paper questionnaires by the trial participants (aided by the research staff) will be source data which is then transcribed onto the CRF (the eCRF held on the secure trial website). As the paper questionnaires are trial specific data, they will be filed in trial specific files. The original paper copy of the questionnaires is source data and will be retained by the site, along with all paper records and trial specific data.

9 STATISTICS AND DATA ANALYSIS

The statistician involved in trial design is Professor Chris Weir (Edinburgh Clinical Trial Unit, University of Edinburgh). Data analysis will be conducted by ECTU statisticians.

9.1 SAMPLE SIZE CALCULATION

9.1.1 Primary MRI Outcome, PBVC

A total of 440 patients will be randomised equally (1:1:1:1) between the three active treatments and the placebo to give 110 per treatment arm. The primary analysis will be intention-to-treat (ITT) on the whole study cohort (n=110/arm). Based on two UK phase II trials (Lamotrigine [60] and MS-STAT [27]), we expect 10% of the total cohort to drop out of the trial before Year 2, and a further 10% of the total cohort to come for their Year 2 visit, but be off medication. Thus we anticipate 90 patients per arm completing the study.

From the calculations reported in by Altmann et al [43] for measurement of PBVC using the SIENA registration-based method, and further data provided by Dr Altmann summarised in the table below (Table A), 90 patients per arm would give over 90% power to detect a 40% reduction in PBVC on any active arm compared to placebo and 80% power to detect a 35% reduction (Bonferroni adjustment for multiple comparisons of three 1.67% two-sided tests, giving 5% overall two-sided significance level). While these sample sizes are based on annual measurement time points and comparison of mean rates of change using longitudinal linear mixed models, Altmann et al also confirm that they are almost identical to the sample size requirements under analysis of covariance of PBVC adjusting for baseline value, the proposed method of analysis in this trial.[43]

MRI measurement times		Baseline, 96 weeks			
2-sided significance level		0.0167		0.05	
Statistical power		80%	90%	80%	90%
Treatment effect	30%	123	158	92	123
	35%	90	116	68	91
	40%	69	89	52	70

Table A - Required sample size per arm by treatment effect size, significance level and statistical power; treatment effect expressed as relative mean difference in PBVC under treatment. PBVC assessed using SIENA registration-based method.

For a more exploratory analysis without adjusting for multiple comparisons, this sample size will give almost 90% power to detect a 35% reduction in atrophy. The study is not powered to detect differences between the three active treatment arms.

Notably, sample sizes for using percentage (SIENA) and absolute (SIENAX subtraction) measures of brain volume change have been calculated for a treatment trial in SPMS using data from the placebo arm of a previous trial. The mean/standard deviation ratios of PBVC were much higher than those of absolute BVC after 1 and 2 years of follow up (table 1,[43]) and the sample size required to show a specified treatment effect were almost 10 times higher when absolute volume change measures were used. Therefore, PBVC is the preferred primary outcome measure for the trial.

9.2 PROPOSED ANALYSES

9.2.1 **Recruitment and Baseline**

A CONSORT flow diagram will be reported. Exploratory summary methods will be used to describe baseline characteristics: continuous variables will be summarized using summary statistics (mean, standard deviation, median, minimum, and maximum) by treatment group, and categorical variables will be presented using frequency distributions by treatment group. Proportions of patients with missing 96 week MRI data in each treatment group will also be compared, as will baseline data for patients with missing and non-missing 96 week follow-up data.

9.2.2 **Primary MRI Outcome Measure**

The primary endpoint will be the percentage brain volume change (PBVC) measured using the Structural Image Evaluation, using Normalization, of Atrophy (SIENA) method between baseline and 96 weeks. A normal linear model will be used to compare the three active treatment group arms with placebo adjusting for baseline normalised brain volume and minimisation variables: age, gender, treatment centre (as a fixed effect) and baseline EDSS. The efficacy measure for each active treatment will be the mean difference in PBVC change versus placebo. All patients for whom baseline and 96 week brain volume data are available will be included in the analysis according to the treatment group to which they were randomised irrespective of which treatment(s) they may have received. Dunnett's method will be used to adjust for the multiple pairwise comparisons versus a common placebo group. No formal comparison of the active treatments will be undertaken.

9.2.3 **Secondary MRI Outcome**

9.2.3.1 **Pseudoatrophy**:

Using the same methods as for the primary MRI outcome, the mean difference in PBVC from baseline to 6 months between the placebo group and each of the active treatment groups will also be assessed. If the reduction in PBVC is significantly greater in any treatment group a secondary analysis will compare PBVC from week 24 to week 96 between that treatment group and the placebo group using normal linear modelling as in section 9.2.2. As this is an exploratory secondary analysis, no formal adjustment for multiplicity will be made.

9.2.3.2 **Counts of new and enlarging T2 lesions:**

Each active treatment group will be compared with placebo in terms of the number of new and enlarging T2 lesions between the baseline and 96 week MRI. Over-dispersed Poisson regression models will be used to estimate the percentage difference in new/enlarging lesion count after adjusting for baseline T2 lesion volume and the minimisation variables: age, gender, treatment centre and baseline EDSS.

9.2.4 Clinical Secondary Outcome Measures

When the change over time in continuous outcomes (EDSS, 9 hole peg test, PASAT, MS functional composite score, Symbol Digit Modalities Test (SDMT), Sloan Low contrast visual acuity (SLCVA), Multiple Sclerosis Impact Scale v2 (MSIS29), Multiple Sclerosis Walking Scale v2 (MSWS), NFI, NPRS, BPI, NPS and health-related quality of life (EQ-5D)) is found to be reasonably normally distributed, perhaps following transformation, these will be compared for active treatments and the placebo groups using normal linear models as in section 9.2.2. If normality cannot be assumed, non-parametric comparison tests will be used. Cox proportional hazard models (adjusting for the minimisation variables) will be used for time to first relapse, timed 25 foot walk and time to progress to a given EDSS score, with the difference between each active treatment and placebo being expressed in terms of a hazard ratio. In exploratory analyses, additional statistical modelling will assess whether composites of imaging and disability measures can be used to predict temporal evolution of SPMS and response to treatment.

9.2.5 Interim analyses

Unblinded safety data will be monitored by a Data Monitoring Committee to ensure the ongoing safety of patients in the study. No formal interim analyses will be conducted.

9.2.6 Statistical Analysis Plan

A full statistical analysis plan (SAP) will prepared which will include details of methods for calculating derived variables, methods for handling missing data and withdrawals, any sensitivity analyses and approaches to testing the assumptions in the statistical analyses. The SAP will summarise the plan for validation of the statistical analysis. The SAP will list the tables, data listings and figures to be reported.

10 ADVERSE EVENTS

10.1 DEFINITIONS

Term	Definition
Adverse Event (AE)	Any untoward medical occurrence in a patient or clinical trial subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment.
Adverse Reaction (AR)	Any untoward and unintended response in a subject to an investigational medicinal product which **is related** to any dose administered to that subject.

	This includes medication errors, uses outside of protocol (including misuse and abuse of product)
Serious adverse event (SAE), serious adverse reaction (SAR) or unexpected serious adverse reaction	Any adverse event, adverse reaction or unexpected adverse reaction, respectively, that: • results in death, • is life-threatening, • requires hospitalisation* or prolongation of existing hospitalisation, • results in persistent or significant disability or incapacity, or • consists of a congenital anomaly or birth defect
Important Medical Event	These events may jeopardise the subject or may require an intervention to prevent one of the above characteristics/consequences. Such events should also be considered 'serious'.
Unexpected adverse reaction	An adverse reaction the nature and severity of which is not consistent with the information about the medicinal product in question set out: (a) in the case of a product with a marketing authorization, in the summary of product characteristics for that product, (b) in the case of any other investigational medicinal product, in the investigator's brochure relating to the trial in question.
SUSAR	Suspected Unexpected Serious Adverse Reaction

***Expected events relating to SPMS resulting in hospitalisation including Grade 3 relapses will not meet the criteria of an SAE in this trial. See sections 10.4.6 and 10.4.6.1.**

10.2 DETECTING AEs AND SAEs

All AEs will be recorded in the medical records from the time a participant signs the consent form to take part in the study until study exit (Week 100).

Participants will be asked about the occurrence of AEs/SAEs at every visit during the study. Open-ended and non-leading verbal questioning of the participant will be used to enquire about AE/SAE occurrence. Participants will also be asked if they have been admitted to hospital, had any accidents, used any new medicines or changed concomitant medication regimens. AE data is also available from information written by the participant on the participant diary (this is considered source data). If there is any doubt as to whether a clinical observation is an AE, the event will be recorded.

AEs and SAEs may also be identified by support departments e.g. laboratory values.

10.3 RECORDING AEs AND SAEs

Collection, recording and reporting of adverse events (including serious and non-serious events and reactions) to the sponsor will be completed according to the sponsor's SOP(INV/S05).

When an AE/SAE occurs, it is the responsibility of the Investigator to review all documentation (e.g. hospital notes, laboratory and diagnostic reports) related to the event. The Investigator will then record all relevant information on the AE log in the CRF and on the SAE form (if the AE meets the criteria of serious). However, if the AE is non–serious (see section 10.1 for serious criteria definition) or MS related (see section 10.4.6.1 for MS expected adverse events) these will remain in the source data (Medical records and Participant diary cards) only. Information to be

collected includes dose, type of event, onset date, Investigator assessment of severity and causality, date of resolution as well as treatment required, investigations needed and outcome.

Clinically significant abnormalities in the results of objective tests (e.g. laboratory variables) will also be recorded as adverse events on the AE log in the CRF, and if are not expected as part of disease or IMP, these will be recorded as unexpected.

All adverse events will be recorded with clinical symptoms and accompanied with a simple, brief description of the event, including dates as appropriate.

All adverse events will be recorded until 4 weeks after the last clinical visit (study exit, visit 10) All Serious Adverse Events will be reportable to the sponsor up to 30 days post last IMP administration.

10.4 ASSESSMENT OF AEs AND SAEs

Seriousness, causality, severity and expectedness will be assessed as though the participant is taking active IMP. Cases that are considered serious, possibly, probably or definitely related to IMP and unexpected (i.e. SUSARs) will be unblinded.

The Investigator is responsible for assessing each AE.

The Chief Investigator (CI) may not downgrade an event that has been assessed by an Investigator as an SAE or SUSAR, but can upgrade an AE to an SAE, SAR or SUSAR if appropriate.

10.4.1 **Assessment Of Seriousness**

The Investigator will make an assessment of seriousness as defined in Section 10.1.

10.4.2 **Assessment Of Causality**

Refer to Table B.

The assessment of relationship of adverse events to the administration of IMP is a clinical decision based on all available information at the time of the completion of the case report form. The categories described in Table B will be used to define the causality of the adverse event.

TABLE B: ASSESSMENT OF CAUSALITY The assessment of relationship of adverse events to the administration of IMP is a clinical decision. The following categories will be used to define the causality of the adverse event:	
Category	**Definition**
Definitely:	There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out.
Probably:	There is evidence to suggest a causal relationship, and the influence of other factors is unlikely
Possibly	There is some evidence to suggest a causal relationship (e.g. the event occurred within a reasonable time after administration of the trial medication). However, the influence of other factors may have contributed to the event (e.g. the patient's clinical condition, other concomitant events).
Unlikely	There is little evidence to suggest there is a causal relationship (e.g. the event did not occur within a reasonable time after administration of the trial medication). There is another reasonable explanation for the event (e.g. the patient's clinical condition, other concomitant treatments).
Not related	There is no evidence of any causal relationship.
Not Assessable	Unable to assess on information available.

10.4.3 **Assessment of Severity**

The Investigator will make an assessment of severity for each AE/SAE and record this on the CRF or AE form according to one of the categories described in Table C:

TABLE C: ASSESSMENT OF SEVERITY OF AE/SAE The Investigator will make an assessment of severity for each AE/SAE and record this on the CRF or AE form according to one of the following categories:	
Category	**Definition**
Mild	The adverse event does not interfere with the volunteer's daily routine, and does not require intervention; it causes slight discomfort
Moderate	The adverse event interferes with some aspects of the volunteer's routine, or requires intervention, but is not damaging to health; it causes moderate discomfort
Severe	The adverse event results in alteration, discomfort or disability which is clearly damaging to health

Note: the term 'severe', used to describe the intensity, should not be confused with 'serious' which is a regulatory definition based on participant/event outcome or action criteria. For example, a headache may be severe but not serious, while a minor stroke is serious but may not be severe.

10.4.4 Assessment Of Expectedness

If an event is judged to be an AR/SAR, the evaluation of expectedness will be made based on knowledge of the reaction and the relevant product information documented in the SmPC refer to Table D.

TABLE D: ASSESSMENT OF EXPECTEDNESS OF AR/SAR	
Category	**Definition**
Expected	An adverse event that is classed in nature as serious and which is consistent with the information about the IMP listed in the Investigator Brochure (or SmPC if Licensed IMP) **or clearly defined in this protocol.**
Unexpected	An adverse event that is classed in nature as serious and which is not consistent with the information about the IMP listed in the Investigator Brochure (or SmPC if Licensed IMP)

The reference document to be used to assess expectedness against the IMPs are:

Latest version of SmPC for fluoxetine and amiloride (both IMP supplied by Actavis) and riluzole (rilutek supplied by Sanofi) available from electronic medicines compendium: www.medicines.org.uk/

10.4.5 **Seriousness**

Seriousness as defined for an SAE in section 10.1

10.4.6 **Non Reportable AEs/SAEs/SARs**

The following will not be reported on the AE log in the CRF or to the sponsor as AEs/SAEs/SARs

10.4.6.1 **Expected events related to SPMS**:
SPMS is a progressive neurological condition and as such deterioration in neurological symptoms is expected. Therefore natural changes in motor, sensory, balance, sphincter (including urinary tract infections), visual, cognitive and fatigue levels are excluded as AEs/SAEs/SARs and will not be reported as such. In addition, relapses will not be counted as AEs/SAEs/SARs, but will be collated separately. They will be graded as follows (and see appendix 4):

Grade 1 relapse: relapse not treated with corticosteroids
Grade 2 relapse: relapse treated with corticosteroids, but not requiring hospitalisation
Grade 3 relapse: relapse treated with corticosteroids and requiring in-patient hospitalisation; or relapse not treated with corticosteroids but requiring in-patient hospitalisation

If the participant feels they are experiencing a relapse, they should contact their local MS team (nurse/consultant) or General Practitioner as per routine practice, so that appropriate management can occur. They should also inform the MS-SMART team at their next scheduled visit, so that the relapse can be documented as per appendix 4. At the Investigator's/nurse's discretion the patient can attend for an unscheduled visit, see section 7.

If however, the investigators suspects that the disease has progressed faster due to the administration of the IMP, then they will report this as an unexpected adverse event (important medical event) to the sponsor.

All laboratory values will be recorded on the appropriate pages of the CRF. When there is a shift of a solitary, or group of values during the course of the study, which in the opinion of the Investigator may be classed as serious (life-threatening) and may or may not be deemed

attributable to the medication, this finding must be notified to the sponsor immediately. In these instances, the patient will be treated appropriately without regard to the confines of the protocol.

Full details of contraindications and side effects that have been reported following administration of the IMP can be found in the relevant Summary of Product Characteristics (SmPC).

Participants will be instructed to contact their Investigator at any time after consenting to join the trial if any symptoms develop. All adverse events (AE) that occur after joining the trial must be reported in detail in the Case Report Form (CRF) or AE form. In the case of an AE, the Investigator should initiate the appropriate treatment according to their medical judgement. Participants with AEs present at the last visit must be followed up until resolution of the event.

10.5 REPORTING OF SAEs/SARs/SUSARs

Reporting to the sponsor will be completed as per UCL sponsor's SOP and using the UCL SAE forms (INV/S05).

All serious adverse events will be recorded in the hospital notes and the CRF, and the sponsor's AE log. The AE log will be reported to the sponsor at least once per year. All serious adverse events will need to be reported to the sponsor on a SAE form unless stated in section 10.4.6 whereby it lists expected SAEs that will not be reported to the sponsor, with justifications as to why they will not be reported. SAEs are reportable for 30 days after IMP administration – only SARs thereafter.

For reportable SAEs (those that are not described in section 10.4.6) the Chief/ Principal Investigator or delegated individual will complete the sponsor's serious adverse event form and the form will be faxed to the sponsor on 020 3108 2312, or preferably emailed to sae@ucl.ac.uk within 24 hours of his / her becoming aware of the event. A copy will be sent in tandem to the ECTU for notification. The Chief/ Principal Investigator or delegated individual will respond to any SAE queries raised by the sponsor as soon as possible.

All SUSARs must be notified to the sponsor immediately (or at least within 24 hours) according to the sponsor's written SOP.

The sponsor will notify the main REC and MHRA of all SUSARs. SUSARs that are fatal or life-threatening must be notified to the MHRA and REC within 7 days after the sponsor has learned of them. Other SUSARs must be reported to the REC and MHRA within 15 days after the sponsor has learned of them.

The procedure for unblinding in the event of a SUSAR is described in section 5.5.2 of the protocol.

10.6 FOLLOW UP PROCEDURES

After initially recording an AE or recording and reporting an SAE, the Investigator will follow each participant until resolution or death of the participant. Follow up information on an SAE will be reported to the JRO office.

AEs still present in participants at the last study visit or reported in 30 day follow up period after completion of trial medication will be monitored until resolution of the event, or until no longer medically indicated.

Any SUSAR related to the IMP will need to be reported to the sponsor irrespective of how long after IMP administration the reaction has occurred.

10.7 DEVELOPMENT SAFETY UPDATE REPORTS

The sponsor will provide the main REC and the MHRA with Development Safety Update Reports (DSUR) which will be written in conjunction with the trial team and the sponsor's office. The report

will be submitted within 60 days of the Developmental International Birth Date (DIBD) of the trial each year until the trial is declared ended. This will be done in accordance with the sponsor's SOP (SPON/S17).

10.8 NOTIFICATION OF DEATH

All deaths will be reportable to the Sponsor. This report will be immediate.

10.9 OVERDOSE

Sites will record details of reported overdoses on the deviation log and inform the trial manager at ECTU as soon as possible after being made aware of the information. Sources of information can include patient-reported, pill counts, diary cards and drug charts. ECTU will notify the sponsor that an overdose has occurred.

In the event that an SAE is associated with the overdose the SAE reporting procedure in addition should be followed (section 10). Details of the overdose will documented in the SAE form.

An overdose is defined as taking double the normal medication/day for 1 day or more. If an overdose has taken place, the patient will suspend medication for at least 48 hours, and resume the previous dose as soon as possible afterwards, at the discretion of the PI. The patient will continue in trial.

The individual IMP data is given below. It is clear that if clinically significant symptoms occur, the trial participant should be unblinded to aid medical management.

Amiloride: no data are available; it is not known whether the drug is dialysable. No specific antidote is available. The most likely signs and symptoms are dehydration and electrolyte imbalance which should be treated by established methods. Therapy should be discontinued and the patient closely observed. Emesis should be induced or gastric lavage performed if ingestion is recent. Treatment is symptomatic and supportive. If hyperkalaemia occurs, active measures should be taken to reduce plasma potassium levels.

Riluzole: Neurological and psychiatric symptoms, acute toxic encephalopathy with stupor, coma, and methemoglobinemia have been observed in isolated cases. In case of overdose, treatment is symptomatic and supportive.

Fluoxetine: Cases of overdose of fluoxetine alone usually have a mild course. Symptoms of overdose have included nausea, vomiting, seizures, cardiovascular dysfunction ranging from asymptomatic arrhythmias to cardiac arrest (including very rare cases of Torsades de Pointes), pulmonary dysfunction, and signs of altered CNS status ranging from excitation to coma. Fatality attributed to overdose of fluoxetine alone has been extremely rare. Cardiac and vital signs monitoring are recommended, along with general symptomatic and supportive measures. No specific antidote is known.

Forced diuresis, dialysis, haemoperfusion, and exchange transfusion are unlikely to be of benefit. Activated charcoal, which may be used with sorbitol, may be as or more effective than emesis or lavage. In managing overdosage, consider the possibility of multiple drug involvement. An extended time for close medical observation may be needed in patients who have taken excessive quantities of a tricyclic antidepressant if they are also taking, or have recently taken, fluoxetine.

11. PREGNANCY

Pregnancy is not considered an AE or SAE; however, the Investigator will collect pregnancy information for any female participants or female partners of male participants who become pregnant while participating in the study.

Reporting to the sponsor will be completed as per the sponsor's SOP and using the UCL pregnancy notification forms (INV/S05). The Investigator will record the information on the sponsors Pregnancy Notification Form and fax this to the sponsor on 020 3108 2312 or preferably emailed to sae@ucl.ac.uk, within 24 hours of being made aware of the pregnancy.

All pregnant female participants and partners of male participants will be followed up until following the outcome of the pregnancy.

12 TRIAL MANAGEMENT AND OVERSIGHT ARRANGEMENTS

12.1 TRIAL MANAGEMENT GROUP

The Central Trial Office is based in the Edinburgh Clinical Trials Unit (ECTU) and will provide support to each site. The office will be responsible for randomisation, collection of data in collaboration with the research nurses, data processing and analysis.

Publication and dissemination of the study results will be coordinated by ECTU in collaboration with the Chief Investigator and Investigators as per the publication policy.

The trial will be coordinated by a Project Management Group, consisting of the Chief Investigators, lead research staff from selected sites, selected grantholders, sponsor representatives and ECTU trial staff.

The Trial Manager will oversee the study and will be accountable to the Chief Investigator. The Trial Manager will be responsible for checking the CRFs for completeness, plausibility and consistency. Any queries will be resolved by the Investigator or delegated member of the trial team.

A Delegation Log will be prepared for each site, detailing the responsibilities of each member of staff working on the trial.

12.2 TRIAL STEERING COMMITTEE

A Trial Steering Committee (TSC) will be established to oversee the conduct and progress of the trial. The terms of reference of the Trial Steering Committee will be documented in a charter that will be held in the TMF at ECTU.

12.3 DATA MONITORING COMMITTEE

An independent Data Monitoring Committee (DMC) will be established to oversee the safety of participants in the trial. The terms of reference of the Data Monitoring Committee will be documented in a charter that will be held in the TMF at ECTU.

12.4 INSPECTION OF RECORDS

Investigators and institutions involved in the study will permit trial related monitoring and audits on behalf of the sponsor, REC review, and regulatory inspection(s). In the event of an audit or monitoring, the Investigator agrees to allow the representatives of the sponsor, REC and Regulatory Authority direct access to all study records and source documentation. In the event of regulatory inspection, the Investigator agrees to allow inspectors direct access to all study records and source documentation.Trial participants are informed of this during the informed consent discussion and participants will consent to provide access to their medical records.

12.5 RISK ASSESSMENT

An independent risk assessment will also be carried out by the sponsor to determine monitoring requirement and if an audit should be performed before/during/after the study and if so, at what locations and at what frequency.

12.6 STUDY MONITORING

A Trial specific monitoring plan will be established for the trial in accordance with the sponsors SOPs. An appointed monitor will visit the Investigator site prior to the start of the study and during the course of the study and in accordance with the monitoring plan.

13 GOOD CLINICAL PRACTICE

The sponsor will ensure that the trial protocol, patient information sheet, consent form, GP letter and submitted supporting documents have been approved by the appropriate regulatory body (MHRA in UK) and a main research ethics committee, prior to any patient recruitment.

13.1 ETHICAL CONDUCT

The trial will be conducted in compliance with the protocol, the UK Regulations, EU GCP and applicable regulatory requirement(s).

A favorable ethical opinion will be obtained from the appropriate REC and local R&D approval will be obtained prior to commencement of the study.

Before a site can enrol patients into the trial, the Chief Investigator or designee must apply for NHS permission from their Trust Research & Development (R&D) and be granted written permission. It is the responsibility of the Chief Investigator or designee at each site to ensure that all subsequent amendments gain the necessary approval. This does not affect the individual clinician's responsibility to take immediate action if thought necessary to protect the health and interest of individual patients

An annual progress report (APR) will be submitted to the REC within 30 days of the anniversary date on which the favourable opinion was given, and annually until the trial is declared ended. The chief investigator and ECTU will prepare the APR.

13.2 REGULATORY COMPLIANCE

The study will not commence until a Clinical Trial Authorisation (CTA) is obtained from the appropriate Regulatory Authority. The protocol and study conduct will comply with the Medicines for Human Use (Clinical Trials) Regulations 2004, and any relevant amendments.

13.3 INVESTIGATOR RESPONSIBILITIES

The Investigator is responsible for the overall conduct of the study at the site and compliance with the protocol and any protocol amendments. As outlined in the principles of EU GCP, the following areas listed in this section are also the responsibility of the Investigator. Responsibilities may be delegated to an appropriate member of study site staff.

13.3.1 Informed Consent

The Investigator is responsible for ensuring informed consent is obtained before any protocol specific procedures are carried out. The decision of a participant to participate in clinical research is voluntary and should be based on a clear understanding of what is involved.

Participants must receive adequate oral and written information – appropriate Participant Information and Informed Consent Forms will be provided. The oral explanation to the participant will be performed by the Investigator or qualified delegated person, and must cover all the elements specified in the Participant Information Sheet and Consent Form.

The participant must be given every opportunity to clarify any points they do not understand and, if necessary, ask for more information. The participant must be given sufficient time to consider the information provided. It should be emphasized that the participant may withdraw their consent to participate at any time without loss of benefits to which they otherwise would be entitled.

The participant will be informed and agree to their medical records being inspected by regulatory authorities and representatives of the sponsor(s) but understand that their name will not be disclosed outside the hospital.

The Investigator or delegated member of the trial team and the participant will sign and date the Informed Consent Form(s) to confirm that consent has been obtained. The participant will receive a copy of this document and a copy will be filed in the Investigator Site File (ISF) and participant's medical notes.

13.3.2 **Study Site Staff**

The Investigator must be familiar with the IMPs, protocol and the study requirements. It is the Investigator's responsibility to ensure that all staff assisting with the study are adequately informed about the IMPs, protocol and their trial related duties.

13.3.3 **Data Recording**

The Principle Investigator is responsible for the quality of the data recorded in the CRF at each Investigator Site. Any delegation of tasks must be clearly documented on the delegation log.

13.3.4 **Investigator Documentation**

Prior to beginning the study, each Investigator will be asked to provide particular essential documents to ECTU, including but not limited to:

- An original signed Investigator's Declaration (as part of the Clinical Trial Agreement documents);
- Curriculum vitae (CV) signed and dated by the Investigator indicating that it is accurate and current.

ECTU will ensure all other documents required by EU GCP are retained in a Trial Master File (TMF), where required, and that appropriate documentation is available in local ISFs.

13.3.5 **GCP Training**

All study staff must hold evidence of appropriate GCP training.

13.3.6 **Confidentiality**

All laboratory specimens, evaluation forms, reports, and other records must be identified in a manner designed to maintain participant confidentiality. All records must be kept in a secure storage area with limited access. Clinical information will not be released without the written permission of the participant. The Investigator and study site staff involved with this study may not disclose or use for any purpose other than performance of the study, any data, record, or other unpublished, confidential information disclosed to those individuals for the purpose of the study. Prior written agreement from the sponsor or its designee must be obtained for the disclosure of any said confidential information to other parties.

The Case Report Forms (CRFs) will not bear the subject's name or other personal identifiable data and only the subject's initials, date of birth and trial identification number, will be used for identification.

13.3.7 **Data Protection**

All Investigators and study site staff involved with this study must comply with the requirements of the Data Protection Act 1998 with regard to the collection, storage, processing and disclosure of personal information and will uphold the Act's core principles. Access to collated participant data will be restricted to those clinicians treating the participants.

Computers used to collate the data will have limited access measures via user names and passwords.

Published results will not contain any personal data that could allow identification of individual participants.

14 STUDY CONDUCT RESPONSIBILITIES

14.1 PROTOCOL AMENDMENTS

Any changes in research activity, except those necessary to remove an apparent, immediate hazard to the participant in the case of an urgent safety measure, must be reviewed and approved by the Chief Investigator, Sponsor and ECTU.

If any urgent safety measures are taken, the PI/CI shall immediately and in any event no later than 3 days from the date the measures are taken, give written notice to the sponsor, MHRA and the relevant REC of the measures taken and the circumstances giving rise to those measures. Substantial amendments to the protocol as assessed by the sponsor must be submitted in writing to the appropriate REC, Regulatory Authority and local R&D for approval prior to participants being enrolled into an amended protocol.

14.2 PROTOCOL VIOLATIONS AND DEVIATIONS

MS SMART is a pragmatic trial and it is recognized that clinical practice will vary across participating sites. All such variations are acceptable and will not constitute a protocol deviation from the protocol (with the exception of the inclusion and exclusion criteria and safety reporting). Investigators will not implement any deviation from the protocol without agreement from the Chief Investigator and appropriate REC, Regulatory Authority and R&D approval except where necessary to eliminate an immediate hazard to trial participants.

In the event that an Investigator needs to significantly deviate from the protocol, the nature of and reasons for the deviation will be recorded in the CRF. If this necessitates a subsequent protocol amendment, this should be submitted to the REC, Regulatory Authority and local R&D for review and approval if appropriate.

14.3 SERIOUS BREACH REQUIREMENTS

A serious breach is a breach which is likely to effect to a significant degree:

(a) the safety or physical or mental integrity of the participants of the trial; or

(b) the scientific value of the trial.

If a potential serious breach is identified by the Chief investigator, Principal Investigator or delegates, the sponsor and ECTU must be notified within 24 hours. It is the responsibility of the sponsor to assess the impact of the breach on the scientific value of the trial, to determine whether the incident constitutes a serious breach and take the appropriate action.

Not every violation from the protocol needs to be reported to the regulatory authority as a serious breach. If the sponsor deems the incident to be a violation that does not constitute a serious breach from the protocol when identified, corrective and preventative actions will be taken where appropriate and they will be recorded in file notes, held within the TMF and ISF.

14.4 STUDY RECORD RETENTION

All study documentation will be kept for a minimum of 5 years from the protocol defined end of study point. Chief Investigators and Local PI are responsible for the secure archiving of trial site documents and database as per their trust policy. All essential documents will be archived for a minimum of 5 years after completion of the trial. Archiving will be authorised by the sponsor following submission of the end of study report. Other essential documents, including source data, consent forms, and regulatory documents, will be archived by or for the Investigator in an appropriate archive facility in line with current regulatory requirements and made available for monitoring, audit and regulatory inspection. The trial master file and database will be held by ECTU for a minimum of 5 years from the defined end of study point.

When the minimum retention period has elapsed, study documentation will not be destroyed without permission from the sponsor.

14.5 END OF STUDY

The end of study is defined as the last patient, last visit.

The Investigators and/or the trial steering committee and/or the sponsor have the right at any time to terminate the study for clinical or administrative reasons.

The end of the study will be reported to the REC and Regulatory Authority within 90 days, or 15 days if the study is terminated prematurely. The Investigators will inform participants of the premature study closure and ensure that the appropriate follow up is arranged for all participants involved.

A summary report of the study will be provided to the REC and Regulatory Authority within 1 year of the end of the study.

14.6 CONTINUATION OF DRUG FOLLOWING THE END OF STUDY

Trial medication will be discontinued after the final clinic assessment for each participant.

14.7 INSURANCE AND INDEMNITY

University College London (UCL) holds insurance against claims from participants for injury caused by their participation in the clinical trial. Participants may be able to claim compensation if they can prove that UCL has been negligent. However, as this clinical trial is being carried out in a hospital, the hospital continues to have a duty of care to the participant of the clinical trial. University College London does not accept liability for any breach in the hospital's duty of care, or any negligence on the part of hospital employees. This applies whether the hospital is an NHS Trust or otherwise.

Participants may also be able to claim compensation for injury caused by participation in this clinical trial without the need to prove negligence on the part of University College London or another party. Participants who sustain injury and wish to make a claim for compensation should

do so in writing in the first instance to the Chief Investigator, who will pass the claim to the sponsor's Insurers, via the sponsor's office.

Hospitals selected to participate in this clinical trial shall provide clinical negligence insurance cover for harm caused by their employees and a copy of the relevant insurance policy or summary shall be provided to University College London, upon request."

15 REPORTING, PUBLICATIONS AND NOTIFICATION OF RESULTS

The trial is registered with an authorised registry (ISRCTN to be confirmed), according to the ICMJE Guidelines.

15.1 AUTHORSHIP POLICY

The success of the study depends upon the collaboration of all participants. For this reason, credit for the main results will be given to all those who have collaborated in the study, through authorship and by contribution.

Uniform requirements for authorship for manuscripts submitted to medical journals will guide authorship decisions. These state that authorship credit should be based only on substantial contribution to:

- Conception and design, or acquisition of data, or analysis and interpretation of data
- Drafting the article or revising it critically for important intellectual content
- Final approval of the version to be published

and that all these conditions must be met (www.icmje.org).

In light of this, the Chief Investigators, Co-Applicants and senior ECTU staff will be named as authors in any publication, and an appropriate first author agreed through discussion amongst the Trial Management Group (TMG) members. The MS-SMART team should be acknowledged in all publications, as should UCL and NIHR EME (as detailed in Section 15.2.3. below). Other key individuals will be included as authors or contributors as appropriate and at the discretion of the TMG. We will include collaborators who will be listed as contributors for the main study publication, and for those principle investigators we will endeavour to name them as co-authors on the primary publication subject to this being acceptable inclusion criteria by the target journal. Any disputes relating to authorship will be resolved by the CIs in consultation with the TMG and if need be the Trial Steering Committee (TSC).

The MS-SMART team should be acknowledged in all publications, as should UCL and MRC/EME. The Chairs and Independent members of the TSC and Data Monitoring and Ethics Committee (DMEC) will be acknowledged, but will not qualify for full authorship, in order to maintain their independence.

Relevant NIHR Clinical Research Networks (e.g. CCRN) support should be acknowledged appropriately in trial publications.

15.2 PUBLICATION

All proposed publications and presentations must be discussed with the sponsor, co-CIs and ECTU prior to their release.
The clinical study report will be used for publication and presentation at scientific meetings.
Summaries of results will also be made available to Investigators for dissemination within their clinics (where appropriate and according to their discretion).

15.2.1 **Data release for publication**

To maintain the scientific integrity of the study, data will not be released prior to the first publication of the results of the primary endpoint analysis, either for study publication or oral presentation purposes, without the permission of the DMEC and the TSC.

The TSC will agree a publication plan and must be consulted prior to release or publication of any study data.

Individual collaborators must not publish data concerning their participants which is directly relevant to the questions posed in the study until the main results of the study have been published. Local collaborators may not have access to study data until after publication of the main study results.

15.2.2. **Data Source**

Data from the ECTU database in Edinburgh must be used for data analyses for all abstracts and publications relating to the questions posed within the trial protocol.

15.2.3. **Processes For The Drafting, Review And Submission Of Abstracts And Manuscripts**

The agreed first author of abstracts is responsible for circulating these to the other members of the TMG and the Sponsor for review at least 15 days prior to the deadline for submission.
The agreed first author of manuscripts is responsible for ensuring:

- Timely circulation of all drafts to all co-authors during manuscript development and prior to submission
- Timely (and appropriate) circulation of reviewers comments to all co-authors incorporation of comments into subsequent drafts
- Communication with the TSC (i.e. ensuring submission is in line with TSC publication plan, and ensuring TSC receive the final draft prior to submission)

The first author is responsible for submission of the publication and must keep the TMG and all authors informed of the abstract's or manuscript's status. The TSC will be kept informed of rejections and publications as these occur. On publication, the first author should send copies of the abstract or manuscript to the TSC, the TMG, the sponsor and to all co-authors, and ensure communication with the National Institute for Health Research Efficacy and Mechanism Evaluation (NIHR EME).

15.3 PEER REVIEW

This study has been extensively peer reviewed by NIHR EME.

15.4 Finances

This trial is funded by the Medical Research Council/ Evaluation and Mechanism Evaluation Programme grant. The IMP, riluzole is provided by the drug company Sanofi.

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