University of California, San Francisco

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Clinical Research Protocol

A placebo-controlled pilot trial evaluating the effect of a tissue selective estrogen complex on menopausal symptoms in women with multiple sclerosis

Protocol Number:	1
Version Date:	3/29/2016
Investigational Product:	Duavee
IND Number:	We believe this clinical investigation of a *marketed* drug meets all FDA IND exemption criteria, namely: • The drug product is lawfully marketed in the United States. • The investigation is not intended to be reported to FDA as a well-controlled study in support of a new indication and there is no intent to use it to support any other significant change in the labeling of the drug. • The investigation of this prescription drug, is not intended to support a significant change in the advertising for the drug. • The investigation does not involve a route of administration, dose, patient population, or other factor that significantly increases the risk (or decreases the acceptability of the risk) associated with the use of the drug product (21 CFR 312.2(b)(1)(iii)). • The investigation is conducted in compliance with the requirements for review by an IRB (21 CFR part 56) and with the requirements for informed consent (21 CFR part 50). • The investigation is conducted in compliance with the requirements of § 312.7 (i.e., the investigation is not intended to promote or commercialize the drug product).
Development Phase:	Phase II
Sponsor:	Riley Bove Sandler Neurosciences Center, 675 Nelson Rising Lane San Francisco, CA
Funding Organization:	National Multiple Sclerosis Society
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Coordinating Center:	N/A

Approval:

	12/16/2015
PI or Sponsor Signature (Name and Title)	*Date*

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PROTOCOL AGREEMENT

I have read the protocol specified below. In my formal capacity as Investigator, my duties include ensuring the safety of the study subjects enrolled under my supervision and providing the University of California, San Francisco with complete and timely information, as outlined in the protocol. It is understood that all information pertaining to the study will be held strictly confidential and that this confidentiality requirement applies to all study staff at this site. Furthermore, on behalf of the study staff and myself, I agree to maintain the procedures required to carry out the study in accordance with accepted GCP principles and to abide by the terms of this protocol.

Protocol Number: 15-18004

Protocol Title: **A placebo-controlled pilot trial evaluating the effect of a tissue selective estrogen complex on menopausal symptoms in women with multiple sclerosis**

Protocol Date: 12/16/2015

	12/16/2015
Investigator Signature	*Date*

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LIST OF ABBREVIATIONS

AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BUN	blood urea nitrogen
CE	Conjugated estrogen
CFR	Code of Federal Regulations
CRF	case report form
DMC	Data Monitoring Committee
DSMB	Data Safety Monitoring Board
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HIPAA	Health Insurance Portability and Accountability Act of 1996
ICF	informed consent form
ICH	International Conference on Harmonisation
IEC	Independent Ethics Committee
IRB	Institutional Review Board
IV	intravenous
mEq	milliequivalent
MS	Multiple sclerosis
mg	milligram
NAMS	North American Menopause Society
PI	Principal Investigator
SAE	serious adverse experience
SERM	Selective estrogen receptor modulator
SGOT	serum glutamic oxaloacetic transaminase
SGPT	serum glutamate pyruvate transaminase
TSEC	tissue selective estrogen complex

PROTOCOL SYNOPSIS

TITLE	A placebo-controlled pilot trial evaluating the effect of a tissue selective estrogen complex on menopausal symptoms in women with multiple sclerosis
SPONSOR	University of California, San Francisco
FUNDING ORGANIZATION	National Multiple Sclerosis Society
NUMBER OF SITES	1
RATIONALE	Multiple sclerosis (MS) is a chronic neurologic disorder characterized by inflammation and neurodegeneration that affects 3 times more women than men and, in 90% cases, women before the age of 50, i.e. prior to menopause. Given evidence for hormonal regulation of MS, a symptomatic worsening of MS symptoms at menopause due to hot flashes and other phenomena, as well as recent data suggesting that MS disease severity may worsen after menopause, we propose to pilot hormonal therapies in peri-menopausal women. Increasing evidence suggests that estrogen is neuroprotective, and that hormonal modulation after menopause may be an important strategy to combat cognitive decline. Given the potential complications of hormone replacement therapies (HT), tissue selective estrogen complex (TSEC)s have been developed **to combine the beneficial effects of estrogen with the tissue-specific agonistic and antagonistic properties of selective estrogen receptor modulators (SERMs).** Here, we propose a pilot single-center randomized double blind placebo controlled trial to investigate the effectiveness of the TSEC Duavee (conjugated estrogen + SERM bazedoxifene), vs. placebo, for 8 weeks on (1) tolerability, (2) patient-reported symptoms (menopausal: hot flashes, sleep, cognition) and (3) MS-related safety (MS symptoms, MRI). Our team consists of experts in MS translational research, and hormonal therapies.
STUDY DESIGN	A pilot, 2-month, double blind, 1:1 randomized placebo controlled clinical trial investigating the effect of a tissue-selective estrogen modulator (TSEC) (Duavee, which is FDA approved for the treatment of menopausal symptoms in women in the general population) on tolerability and menopausal symptoms in perimenopausal women with MS.
PRIMARY OBJECTIVE	To test the hypothesis that a TSEC (CE+BZA) will be well tolerated in women with MS, and will alleviate their menopausal symptoms. In relieving hot flashes and subsequent sleep and mood disruption, MS symptoms will also be ameliorated.
SECONDARY OBJECTIVES	- To explore the hypothesis that CE+BZA improves menopausal symptoms in MS women. - To explore the hypothesis that CE+BZA results in improvement in MS-related outcomes. - To explore the hypothesis that CE+BZA for 8 weeks is tolerable in women with MS.
NUMBER OF SUBJECTS	24

SUBJECT SELECTION CRITERIA	Inclusion Criteria: Women aged 40-62 who carry a diagnosis of MS that is stable and are perimenopausal (we will use the widely used criterion of 6 months of amenorrhea as this corresponds to either the late menopausal transition or early postmenopause). Exclusion Criteria: Known hypersensitivity or contraindications to estrogen, use of any prescribed therapy that is taken specifically for hot flashes in the past 1 month, steroids in the past 1 month, clinical relapse in last 3 months, evidence of other structural brain disorder, pregnancy, intending pregnancy, or breast feeding, drug or alcohol abuse in the past 1 year, or other considerations to ensure the safety of the patients.
TEST PRODUCT, DOSE, AND ROUTE OF ADMINISTRATION	DUAVEE (0.45mg conjugated estrogens/20 mg bazedoxifene) tablets. Product will be taken orally by patient every day for two months.
CONTROL PRODUCT, DOSE AND ROUTE OF ADMINISTRATION	Microcrystalline cellulose placebo Product will be taken orally by patient every day for two months.
DURATION OF SUBJECT PARTICIPATION AND DURATION OF STUDY	Subjects will be on study for up to 70 days **Screening:** 2 weeks **Treatment:** 2 months
CONCOMMITANT MEDICATIONS	Allowed: MS disease modifying and symptomatic therapies, antidepressants (no adjustments within 2 months of baseline visit) Prohibited: hormone therapies (estrogen, testosterone, progesterone), hot flash therapies; lamotrigine, oxcarbazepine
EFFICACY EVALUATIONS	
PRIMARY ENDPOINT	The primary measure will be the daily number of hot flashes averaged over 3 days (daily diary)
SECONDARY ENDPOINTS	• sleep quality (ISI questionnaire) • Treatment Satisfaction Questionnaire for Medications (TSQM) global score • MS Rating Scale (MSRS) global score • SF36 PCS (physical component score). • MRI: number of new or enhancing lesions
OTHER EVALUATIONS	• Safety labs: LFTs • Exploratory measures: corpus callosum tract integrity on DTI, cognitive function, other QOL outcomes (fatigue, mood) • Blood samples stored for hormone levels (estradiol, testosterone, sex hormone binding globulin) and for immunology.

SAFETY EVALUATIONS	Phone call at week 6 to see if patient is experiencing any unacceptable side effects. When approximately 50% of patients have completed the study through Study Visit 2 (total visit 3), an interim analysis for safety will be conducted by the PI. Serious adverse events will be monitored by the committee on an ongoing basis throughout the study. Incidence of adverse events.
PLANNED INTERIM ANALYSES	When approximately 50% of patients have completed the study through Study Visit 2 (total visit 3), an interim analysis for safety will be conducted by the PI. Serious adverse events will be monitored by the committee on an ongoing basis throughout the study.
STATISTICS **Primary Analysis Plan**	We will perform t-tests comparing the absolute changes in outcomes from the baseline to the 2-month visit in the two treatment groups. Aim 1. Effect of CE+BZA on menopausal symptoms. The primary measure will be the daily number of hot flashes averaged over 3 days (daily diary), and the secondary measure will be sleep quality (ISI questionnaire). Aim 2. Effect of CE+BZA on MS outcomes. The primary measure will be the MS Rating Scale (MSRS) global score, and the secondary measure will be the SF36 PCS (physical component score). Exploratory measures will include corpus callosum tract integrity on DTI, cognitive function (SDMT), and other QOL outcomes (MFIS, SF36 sub-scales, depression scale). Aim 3. Tolerability of CE+BZA. The primary measure will be the percentage of subjects reporting side effects on the Treatment Satisfaction Questionnaire for Medications (TSQM) global score, and the MS-specific secondary measure will be the number of new or enhancing lesions on MRI.
Rationale for Number of Subjects	Given this is a pilot study, we will view the study as a success if the observed mean change in the treatment group is larger by any amount compared to the placebo group ("play the winner" approach). In healthy women treated with estradiol for 8 weeks,(**Joffe et al., 2014**) VMS decreased by 2.2 daily in the placebo group and 4.5 in the estradiol group and the SD for the change was 3.4. Assuming same differences and SD, we will have 80% power to find the treatment arm mean difference to be larger with our proposed sample size.

1 BACKGROUND

Menopause in MS. Multiple sclerosis (MS) affects 3 times more women than men, and before age 50 in about 90% cases, i.e. prior to menopause (Bove et al., 2012). There is broad evidence for hormonal regulation of MS in animal models (Gold & Voskuhl, 2009) and in clinical cohorts (e.g. pregnancy (Bove & Chitnis, 2014)).

Around menopause, many clinical patients report symptom worsening associated with hot flashes, sleep disturbance or mood changes. Longer-term, an age-related decline in gonadal steroids might represent one sex-specific influence (Clayton & Collins, 2014) on the known age-related increases in disability and conversion to progressive course (Tutuncu et al., 2013), which is marked by accelerated brain volume loss and neurodegeneration. Recent data suggest that MS disease severity may worsen after menopause (Bove & al., 2014; Bove et al., 2015).

Hormone therapy (HT). In healthy women, after the abrupt decline in estradiol occurring with early oophorectomy, there is an increased risk for cognitive decline and dementia (Coppus et al., 2010; Corbo et al., 2011; Henderson & Sherwin, 2007; Phung et al., 2010; Rocca et al., 2007; Vearncombe & Pachana, 2009; Zhou et al., 2011) and gray matter (GM) loss in functionally important regions (e.g. hippocampus)(Goto et al., 2011). HT, when taken within a perimenopausal "window of opportunity" (5 years), may protect against neurodegeneration, (Bove et al., 2014b; Henderson, 2013; Rocca et al., 2011; Shao et al., 2012) perhaps due to sparing effects on regional GM atrophy (Boccardi et al., 2006; Lord et al., 2010; Robertson et al., 2009).

Despite the benefits of HT (menopausal symptoms, bone density), very few women (<30% of our cohort) are currently taking HT for menopausal symptoms; this is a result of risks such as (1) breast and endometrial cancer, and (2) stroke in older women in the Women's Health Initiative (NAMS; Langer et al., 2012; Rapp et al., 2003).

INNOVATION: To date, there are no objective assessments of the impact of HT on MS disease course. In an initial pilot designed at providing HT to women with MS, we plan to investigate the tolerability of Duavee in treating menopausal symptoms in women with MS.

Our treatment, Duavee, has been approved by the FDA for the following indications:

1. Treatment of moderate to severe vasomotor symptoms associated with menopause

2. Prevention of postmenopausal osteoporosis

(http://www.accessdata.fda.gov/drugsatfdadocs/label/2006/004782s147lbl.pdf)

Duavee is commercially available.

1.1 Overview of Non-Clinical Studies

SERMs in MS animal models.
Estrogen receptor (ER) α and β and membrane (m)ERs are present on oligodendrocyte lineage cells. In an animal model of MS, experimental autoimmune encephalomyelitis (EAE), SERMs have been explored for their potential in stimulating endogenous myelination and sparing axonal degeneration. Bebo et al found that both tamoxifen and raloxifene (two commercially available SERMs) suppressed myelin antigen specific T-cell proliferation, with tamoxifen being more effective. Tamoxifen treatment reduced the induction of major histocompatibility complex II by

lipopolysaccharide stimulated dendritic cells and decreased their ability to activate myelin specific T-cells. At lower doses, tamoxifen also increased the levels of Th2 transcription factors and induced a Th2 bias in cultures of myelin-specific splenocytes. EAE symptoms and the degree of demyelination were less severe in mice treated with tamoxifen than in control mice.[1]

In subsequent work, the estrogen receptor (ER) β ligand 2,3-bis(4-hydroxyphenyl)-propionitrile (DPN) has been found to reduce clinical disease, be neuroprotective, stimulate endogenous myelination, and improve axon conduction without altering peripheral cytokine production or reducing central nervous system (CNS) inflammation. Both prophylactic and therapeutic treatments with DPN during EAE were found to improve remyelination-induced axon conduction, suggesting adirect effect of treatment on oligodendrocytes. DPN treatment of EAE animals resulted in phosphorylated ERβ and activated the phosphatidylinositol 3-kinase (PI3K)/serine-threonine-specific protein kinase (Akt)/mammalian target of rapamycin (mTOR) signaling pathway, a pathway required for oligodendrocyte survival and axon myelination.[2] Altogether, these studies suggest that SERMs may exert beneficial effects on neuroprotection through ER-beta modulation.

1.2 Overview of Clinical Studies

HT: What is the evidence?

Hormone therapy (HT) during menopause is an area of active inquiry. HT typically consists of estrogen therapy (ET) or combined estrogen-protestogen (EPT) therapy, either administered orally (ET and EPT), vaginally (ET) or transdermally (low dose estrogen). While HT has been proven to alleviate many menopausal symptoms, it is also associated with risks.[68] Currently, the North American Menopause Society (NAMS) recommends individualized HT treatment based on a patient's risk profile, for the shortest possible duration and at the lowest possible dose.[68]

General benefits and Risks.

Benefits. According to the NAMS 2012 consensus statement, HT is the most effective treatment for menopause-related vasomotor symptoms (ET with or without progestogen) and for moderate to severe symptoms of vulvar and vaginal atrophy (local vaginal ET, systemic HT). Additionally, P HT is effective against overactive bladder (local ET; systemic HT may provoke or worsen stress incontinence), reduces the risk of recurrent UTIs (vaginal ET), improves health-related quality of life (HQOL) in symptomatic menopausal women, and may reduce the diagnosis of new onset type 2 diabetes mellitus. At low doses, HT maintains or improves bone mass density (BMD) and at standard doses, reduces postmenopausal osteoporotic fractures even in women without osteoporosis. Additionally, in observational studies at least, HT was associated with lower risks of venous thromboembolism and stroke (transdermal and low-dose ET). There is to date no report of significant improvement in libido associated with HT, although low dose local ET may improve vaginal symptoms such as lubrication, blood flow and sensation.[68]

Risks: Unopposed systemic ET in postmenopausal women with an intact uterus is associated with increased endometrial cancer risk; concomittant progestogen is therefore recommended. In addition, EPT initiated close to the onset of menopause may be associated be an increased risk of breast cancer; this increased risk has not been observed in ET. HT use in breast cancer survivors may be associated with an increased risk of recurrence. EPT use for longer than 5-10 years may be associated with an increased risk of ovarian cancer, although the incidence rate is rare. There may also be increased risk of lung cancer, or of more aggressive lung cancer, particularly in smokers.

Currently, HT is considered primarily in women ages 50-59, within 10 years of menopause. In this age group, HT used for menopausal symptoms is not felt to increase the risk of CHD events, and in fact ET initiated early postmenopausally may reduce coronary artery disease and CHD risk.[68]

HT and neuroprotection.
Observational studies have suggested that HT, when taken within a "window of opportunity" relative to menopause (operationally defined as 5 years), may be protective against cognitive decline including Alzheimer's disease.[69] Beyond this, perhaps due to estrogen receptor down-regulation, HT may even be harmful, leading to increased risk of stroke and dementia. This hypothesis appears to explain findings from the Women's Health Initiative Memory Study (WHIMS), a subset of the Women's Health Initiative, a landmark longitudinal study that included randomized controlled trials evaluating the effects of (1) unopposed estrogen and (2) EPT compared with placebo on a diverse set of health outcomes. In WHIMS, HT when initiated in women aged 65 or above was associated with an increased risk of dementia of any cause and of cognitive decline [70]. Longitudinal placebo-controlled trials of the effects of HT, initiated within the "window of opportunity," on long-term risk of cognitive decline are awaited. HT considerations in MS. Given the initially negative findings from the Women's Health Initiative, very few women who are currently perimenopausal are treated with HT,5 limiting the power of longitudinal observational studies. Of particular importance in MS, are the long-term effects of HT on neuroprotection, altered immune regulation, and osteoporosis, which are more common in women with MS than in healthy women in study populations. Therefore, the benefit:risk ratio in these patients may be different from those in the general population. Additionally, because many women with MS are at increased risk of premature ovarian failure due to their exposure to cyclophosphamide and novantrone, the benefits to them of a longer duration of HT exposure are unknown. Longitudinal studies are required to uncover the effect that HT may have, if any, on the aggregate balance of neuroinflammation and neurodegeneration seen in MS, as well as in patients' experiences and quality of life (QOL).

As a response to the mixture of desirable and undesirable effects of estrogen formulations on various tissues, through a combination of agonistic, neutral and antagonistic effects, selective estrogen receptor modulators (SERMs) have been developed, with a wide range of effects.

TSECs: Focus on CE/BZA. One important consideration of treating human females with SERMs, again, is the impact of these compounds on various tissues. Tamoxifen, despite its promising effects in EAE, induces hot flashes, which may severely limit its immediate tolerability in perimenopausal MS women, who in our experience worsening of symptoms, perhaps through the Uhthoff phenomenon.[3]

More recently, tissue selective estrogen complexes (TSECs) have been developed to combine the benefits of estrogen with SERMs' tissue-specific properties.[4] TSECs provide a progestin-free alternative to traditional estrogen-progestin therapy (EPT) in women with a uterus. Specifically, the effects on breast and endometrial proliferation, as well as venous thromboembolism, observed with estrogens were not noted when estrogen was combined with bazefoxifene, and in fact, BZA appears to mitigate the effects of the estrogen.[4]

The following provides a summary of pre-clinical and clinical data that highlight the safety of the medication, and in contrast to purely estrogen compounds. **[4]**

2.3. Bazedoxifene

2.3.1. Molecular biology

Bazedoxifene is structurally distinct from other SERMs (Table 1) and exhibits a unique tissue-specific profile of ER activity [38]. The core binding domain of bazedoxifene consists of a 2-phenyl-3-methyl indole, in contrast to the benzothiophene core of raloxifene, and the trans-stilbene core of tamoxifen. Bazedoxifene also contains a hexamethylenamine ring at the side chain terminus, unlike raloxifene and tamoxifen, which contain dimethylamine and piperidine rings, respectively [4]. Bazedoxifene exhibits less selectivity for ERα compared with raloxifene, but has an approximately 10-fold lower binding affinity for ERα than does raloxifene [4]. Molecular modeling studies have demonstrated that bazedoxifene binds to ERα in an orientation that is distinct from that of 4-OH tamoxifen, but similar in orientation to raloxifene-ERα binding [39]. Bazedoxifene has been shown to down-regulate ERs by increasing receptor degradation and suppressing cyclin-D1 promoter activity in MCF7 cells [39]. In a transcriptional activation analysis using a hepatic lipase promoter in HepG2 cells, bazedoxifene functioned as an agonist with slightly less potency than 17β-estradiol. These results differed from those of tamoxifen that showed activity only at very high concentrations or of raloxifene and lasofoxifene that produced no detectable response.

2.3.2. Preclinical data

Preclinical data showed that bazedoxifene acts as an ER agonist in the bone while having minimal effects on uterine and breast tissue [40]. In studies in MCF7 human breast cancer cells, bazedoxifene has been shown to induce unique transcriptional activity among different genes, unlike those induced by raloxifene or lasofoxifene [41,42]. Although all three SERMs antagonized stimulation of MCF7 cell proliferation by conjugated estrogens (CE), bazedoxifene was a more effective antagonist compared with raloxifene or lasofoxifene [41]. Proliferation and growth-related genes were upregulated by estradiol and down-regulated by bazedoxifene. In another study of gene expression in MCF7 and SKBR3 breast cancer cell lines, bazedoxifene and raloxifene acted as competitive antagonists on most of the genes evaluated, reversing the activity of estradiol; lasofoxifene had a lesser effect [42]. In a study

20 or 40 mg/day was associated with a significantly lower incidence of new vertebral fractures (2.3% with 20 mg, 2.5% with 40 mg) compared with placebo (4.1%; $P < 0.05$), although there was no significant difference in incidence of nonvertebral fractures [51]. In a subgroup of women with a higher risk of fracture, however, nonvertebral fracture rate with combined bazedoxifene doses (5.7%) was significantly lower compared with placebo (9.1%; $P = 0.03$; HR, 0.60; 95% CI, 0.37–0.95), and lower than that observed with raloxifene (8.4%; $P = 0.08$; HR, 0.66; 95% CI, 0.41–1.06) [51]. The efficacy of BZA was sustained through 5 and 7 years of treatment as observed in the extensions of the core trial [52].

Bazedoxifene treatment was not associated with increased risk of breast cancer [52–54]. During the core 3-year trial, rates of breast cancer were 0.3% and 0.2%, among women who received bazedoxifene 20 or 40 mg, respectively, compared with 0.4% and 0.4%, in women given raloxifene 60 mg, or placebo, respectively [53]. Similarly low rates of breast cancer, comparable to those seen with placebo, were observed among women who continued bazedoxifene treatment for up to 5 years (0.5%) [52] and 7 years (0.6%) [54].

Bazedoxifene treatment was also not associated with increases in endometrial thickness or increases in the rate of endometrial hyperplasia or carcinoma [51–53,55]. For example, during 3 years of treatment with bazedoxifene 20 or 40 mg, raloxifene 60 mg, or placebo ($N = 753$), the rate of hyperplasia was similar to that of placebo [55]. However, bazedoxifene was associated with lower rates of endometrial cancer compared with placebo at 5 years (0.2% vs 0.3%; $P = 0.05$) [56] and 7 years (0.1% vs 0.4%; $P = 0.02$) of treatment [54].

Effect of menopause in MS. In individuals with MS onset after age 50 there are fewer sex differences (1) in disease type or (2) in time to an EDSS of 6 than there are in individuals with MS onset prior to age 50. This suggests that changes around age 50 (menopause) may contribute to a narrowing of sex differences (Bove et al., 2012).

In a cohort of postmenopausal women, 124 had been prospectively followed through their menopause during CLIMB. In these women, we noted an inflection point in subjects' disease severity at the time of menopause (Bove et al, MS Journal 2015, in press). The rate of EDSS change increased from 0.047 units per year before, to 0.15 units per year after menopause (difference of 0.10 units; 95% CI: 0.03, 0.17, p=0.005). A similar effect was observed in both the natural and surgical menopause groups. When women with only post-menopausal measurements (n=267) were also included in the model, the estimated increase in the average yearly change was 0.052 units per year (95% CI: 0.005, 0.098, p=0.029). These women were followed for a mean duration of 10 years (SD 2.6, range 0.9-16.6), and have both pre- and post-menopausal serial MRI and blood samples available.

These data were substantiated by results from a patient-powered online research platform, PatientsLikeMe, Inc (PLM).(Bove et al., 2013c) Using results from a reproductive survey deployed to "active users", we compared prospectively collected disease severity scores (MS Rating Score (MSRS), good correlation with EDSS(Bove et al., 2013c)) according to reproductive status. The mean (SD) MSRS at the time of the questionnaire was higher in the surgical menopause group (13.1 (5.4)) than in the natural menopause (9.6 (5.1)) or cycling (8.9 (5.5)) groups (age, disease duration and category-adjusted difference for surgical vs. pre=3.27; p<0.0001; surgical vs. natural=3.44; p<0.0001). A significant effect of menopausal age was observed when this was included in the model (p<0.001). Interestingly, we found associations of menopausal status with MS symptoms for the global MSRS score as well as for individual MSRS

domains not typically overlapping with menopausal symptoms (Bove et al, MS and Related Diseases 2015). Despite the obvious biases in online research, these findings are suggestive because of the biologically plausible associations with early, surgical menopause.

Effect of sex and gonadal steroids in MS neuroimaging. Our first set of preliminary data come from a comparison of BPFs in 4 cohorts: men and women, aged less than (38-46) and greater than (54-62) 50 years, and followed for 2 years. In that study, we found that both the older and male cohorts had significantly greater rates of BPF atrophy than the younger and female cohorts (Table 3). Men had lower BPF in a generalized linear model adjusting for age, disease type and duration ($p<0.0001$). We assessed a sex*age interaction to determine whether there were sex –specific changes occurring after age 50, and this was negative. A very important limitation is that menopausal status in these cohorts was not known at the time, and therefore it is possible that a subset of the women aged under 50 had already experienced early, surgical menopause.(R. Bove et al., 2013b)

In a preliminary analysis related to our neuroimaging measure, the corpus callosum (CC), we measured crosssectional area of the CC (as detailed in Aim 1b) for 228 MRI scans where subject menopausal status was known (RB training set). We found that CC area was significantly associated with timing of menopause (est= -3 mm2 per year since menopause, $p=0.0066$) but not with age at MRI ($p=0.57$), when both were included in a model.

Hormonal trials.
Trials of estrogen treatment in healthy perimenopausal women:
Recent approaches to treating VMS have compared low-dose oral 17β-estradiol (0.5 mg/d) (n=97), with SSRI/SNRIs (E.g. low-dose venlafaxine hydrochloride extended release (75 mg/d) (n=96)) or placebo (n=146) for 8 weeks. Estradiol reduced the frequency of symptoms by 2.3 more per day than placebo ($P<.001$), and venlafaxine reduced the frequency of symptoms by 1.8 more per day than placebo ($P=.005$). Importantly, within 8 weeks, significant reduction in VMS were noted with both therapies (Joffe et al., 2014).

HT in women with MS. Using data from the Nurses Health Study (NHS), the PI and collaborators examined the association between perimenopausal HT use and a physical functioning assessment (PF10; subscale of the Short Form 36 (SF-36) QOL survey) at a timepoint between 3 and 10 years after their final menstrual period (early postmenopause) (Bove et al, under review). We assessed the association between HT use at this timepoint (never vs. at least 12 months of systemic estrogen with/without progestin) and both PF10 and the SF-36 Physical Component Scale (PCS). We employed a linear regression model adjusted for age, MS duration, and menopause type and duration, and further for other significant covariates (ancestry). HT users had average PF10 scores that were 23 points higher than non-HT users (adjusted $p=0.004$), and average PCS scores that were 9.15 points higher in the subset of 59 women with these available (adjusted $p=0.021$). Duration of HT use showed a similar trend for PF10 (adjusted $p=0.057$). While this study cannot establish causality, it does suggest that HT may not result in excess adverse outcomes in women with MS.

2 STUDY RATIONALE

Duavee is an approved medication for menopausal symptoms.
The risks of Duavee are shown to be extremely minimal by FDA reports on previous clinical trials and by their approval of the drug. These minimal risks may be

outweighed by the therapeutic potential of Duavee in menopausal women with MS.

2.1 Risk / Benefit Assessment

Information from this study will benefit others because the medical community will gain more knowledge on hot flashes in MS patients, and responses to the study medication. Due to the low risks associated with participation in this study, the potential benefits of self-monitoring hot flashes outweigh the risks.

3 STUDY OBJECTIVES

3.1 Primary Objective

The goal of this pilot grant is to assess the tolerability of Duavee in a cohort of MS women, and to obtain preliminary data regarding its impact on MS outcomes (advanced neuroimaging, patient-reported measures).

3.2 Secondary Objectives

Aim 1. To explore the hypothesis that CE+BZA improves menopausal symptoms in MS women.

Aim 2. To explore the hypothesis that CE+BZA results in improvement in MS-related outcomes.

Aim 3. To explore the hypothesis that CE+BZA for 8 weeks is tolerable in women with MS.

4 STUDY DESIGN

4.1 Study Overview

Women will be randomized to treatment with Duavee (conjugated estrogens + bazedoxifene) vs. conjugated estrogen vs. placebo. They will undergo MRI with DTI and fMRI both at baseline and again after 2 months of treatment. While our primary comparisons of interest are Duavee vs. placebo, the premarin arm is included in order to provide preliminary data to dissect out an effect of estrogen vs.bazedoxifene.

Participation will include a screening visit, 2 study visits, 2 phone calls, and taking Duavee or the placebo once daily for two months as well as keeping daily hot flash and sleep diaries throughout participation in the study.

The Screening visit will occur at the MS Clinic. Eligible subjects will then be contacted to return to enroll. Enrollement/Study Visit 1 and Study Visit 2 will be separated by 2 months and consist of neuroimaging, PROs, blood draws and neuropsychological testing. Between visits 1 and 2, subjects will complete diaries. They will also be contacted at 1 week and 4 weeks. After Visit 2, subjects will be contacted for follow up and at their 6M regularly scheduled clinic visit.

Screening data will be reviewed to determine subject eligibility. Subjects who meet all inclusion criteria and none of the exclusion criteria will be entered into the study.

Total duration of subject participation will be ten weeks.

5 CRITERIA FOR EVALUATION

5.1 Primary Efficacy Endpoint

Three primary measurements to assess each of the three aims:

- The number of daily vasomotor symptoms (VMS), obtained from the patient diary. In healthy women, this was shown to significantly decrease with estradiol treatment at 8 weeks (Joffe et al., 2014). Power calculation: Given this is a pilot study, we will view the study as a success if the observed mean change in the treatment group is larger by any amount compared to the placebo group ("play the winner" approach). In healthy women treated with estradiol for 8 weeks (Joffe et al., 2014), VMS decreased by 2.2 daily in the placebo group and 4.5 in the estradiol group and the SD for the change was 3.4. Assuming same differences and SD, we will have 80% power to find the treatment arm mean difference to be larger with our proposed sample size. This will explore the hypothesis that CE+BZA improves menopausal symptoms in MS women.

- The MSRS, a global score capturing patient impressions of MS severity across eight function domains, and which in a tertiary care referral clinic population has shown good correlation with the EDSS. (Bove et al., 2013, Wicks et al., 2012) This will explore the hypothesis that CE+BZA results in improvement in MS-related outcomes.

- The percentage of subjects reporting side effects on the Satisfaction Questionnaire for Medication (TSQM) (Atkinson et al., 2004), which is commonly used in MS treatment studies (e.g. Calkwood et al., 2014). This will explore the hypothesis that CE+BZA for 8 weeks is tolerable in women with MS.

5.2 Secondary Efficacy Endpoints

- Our secondary endpoints to test the hypothesis that CE+BZA improves menopausal symptoms in MS women will be sleep quality from sleep diary.

- Our secondary endpoints to test the hypothesis that CE+BZA results in improvement in MS-related outcomes will include MSQOL54 composite score (primary), measures of mood (CES-D), fatigue (MFIS), cognition (MSNQ), and bladder (MSQLI BLCS).

- Our secondary endpoints to test the hypothesis that CE+BZA for 8 weeks is tolerable in women with MS will be number of missed doses, and the number of new or enhancing lesions on 8-week MRI, to verify that CE+BZA does not yield any marked changes in inflammatory activity.

5.3 Safety Evaluations

We will have 3 formal means of evaluating safety concerns during the course of the study:

(1) All patients receive an interim phone call at week 6 to see if patient is experiencing any unacceptable side effects. We will compare rates in the 2 groups.

(2) When approximately 50% of patients have completed the study through Study Visit 2 (total visit 3), an interim analysis for safety will be conducted by the PI, including clinical measures as well as effects on liver function. Serious adverse events will be monitored by the committee on an ongoing basis throughout the study.

(3) Also, we will compare MRI outcomes in the first 12 patients, in order to ensure that there is no increased MRI signal.

At the end of the study, we will perform a comparison of liver tests, chemistry, hematology, treatment satisfaction, and clinical reported AEs, across the whole group. We will compare uring t-tests.

6 SUBJECT SELECTION

6.1 Study Population

Subjects with a diagnosis of multiple sclerosis who meet the inclusion and exclusion criteria will be eligible for participation in this study.

6.2 Inclusion Criteria

Menopause/HT considerations:

- Women aged 40-62 years.
- Perimenopausal: We will use the widely used criterion of 6 months of amenorrhea (this corresponds to either the late menopausal transition (periods separated by at least 60d) or early postmenopause (LMP within 2Y) (STRAW +10; Figure 1; [**8**]).
    - This can include
        - Women who have had a bi-lateral oöphorectomy;
        - Women without a uterus and who still have one or both ovaries, with FSH level > 20 mIU/mL and estradiol ≤ 50 pg/mL (on at least one of two blood draws two weeks apart);
        - Women with a uterus who have skipped 2 or more menstrual cycles with an amenorrhea interval >=6 months in the past 12 months;
        - Women who are using the Mirena IUD or who have had an endometrial ablation and who still have one or both ovaries, with FSH level > 20 mIU/mL and estradiol ≤ 50 pg/mL (on at least one of two blood draws two weeks apart).
- A mean of two or more hot flashes/night sweats per day (24 hrs) as reported on daily hot flash diaries collected over 2 weeks.
- Hot flashes/night sweats rated as bothersome (‘moderately’ to ‘a lot’) and/or severe (‘moderate’ to ‘severe’) on 4 or more 12 hour (day/night) blocks of times in each of the 2 weeks of diary completion. This criterion must be met on at least one of the 4 scales on the symptom reports (daytime severity, nighttime severity, daytime bother, nighttime bother).
- In general good health as determined by medical history, blood pressure, and heart rate.
- Absence of uncontrolled hypertension (>160/95 mmHg);
- Resting heart rate <110/minute;
- No history of myocardial infarction, angina, or cerebrovascular events;
- No symptoms consistent with or history of thrombo-embolism, deep vein thrombosis, congestive heart failure;
- No history of endometrial, ovarian, or breast cancer;
- No abnormal mammogram in the last 2 years. Women with an abnormal screening mammogram that was found to be benign after additional evaluation are eligible. If a participant has not had a mammogram in the last 2 years, she will be encouraged to schedule one but will not be excluded from study participation if she declines;

- Normal physical and breast exam in the last two years. (If a participant has not had a physical exam in the last 2 years, she will be required to schedule one before randomization.)
- Absence of any current severe or unstable medical illness.
- Signed informed consent.

MS considerations:

- Documentation of a multiple sclerosis diagnosis as evidenced by one or more clinical features consistent with the multiple sclerosis phenotype, and one or more of the following criteria:
    - If using psychotropic medications (SSRIs, SNRIs, MAOIs, and other antidepressants) and anxiolytics, no change in the past 3 months.
    - If on DMT, no change in past 6 months.
    - Normal vitamin D levels (20-50 ng/mL).

6.3 Exclusion Criteria

HT considerations

- BMI >30 kg/m^2 as higher BMI may affect PK/PD
- Use of hormone therapy or hormonal contraceptives (with the exception of the Mirena IUD) during the 2 months before enrollment. Vaginal postmenopausal estrogens allowed, with the exception of vaginal creams used >3 times a week.
- Use of any prescribed therapy that is taken specifically for hot flashes in the past 1 month.
- Use of any over-the-counter or herbal therapies that are taken specifically for hot flashes in the past 2 weeks.
- Use of selective estrogen receptor modulators (SERMs) or aromatase inhibitors during the 2 months before enrollment.
- Known hypersensitivity or contraindications to estrogen.
- Not using a medically approved method of birth control, if sexually active and not 12 or more months since last menstrual period.
- Drug or alcohol abuse in the past 1 year.
- Depression: moderate or severe (HAD score > 8)
- Other psychiatric disease meeting DSM-IV criteria (bipolar disorder, obsessive compulsive disorder, or schizophrenia
- Lifetime diagnosis of psychosis or bipolar disorder.
- Pregnancy, intending pregnancy, or breast feeding.
- Bazedoxifene undergoes metabolism by UGT enzymes in the intestinal tract and liver; coadministration with UGT inducers (lamotrigine, oxcarbazepine) may reduce bazedoxifene exposure/efficacy and therefore increase risk of endometrial hyperplasia
- History of any of the following, as determined by clinician review of the potential participant's medical history:
    - Pre-breast cancer or high-risk breast cancer condition;
    - Abnormal bleeding suggestive of endometrial pre-cancer;
    - Endometrial hyperplasia.
    - Asthma, diabetes mellitus, epilepsy, and migraine disorders that are not stable or under medical management
    - Active or past history of venous or arterial thromboembolism
    - History of gallstones IF gallbladder intact

  - Known or suspected estrogen-dependent neoplasia
  - History of coronary artery disease
  - Hypersensitivity (angioedema, anaphylaxis) to estrogens, bazedoxifene, or any ingredients
  - Known hepatic impairment or disease
  - Thyroid dysfunction on thyroid medications
  - Known hypoparathyroidism
- Blood test results indicating:
  - Liver function tests: AST >2.5 times upper limit of normal; ALT >2.5 times upper limit of normal; total bilirubin 1.5 times upper limit of normal;
  - Kidney test: creatinine >1.5 mg/dL;
  - Blood count: hematocrit <30%;
  - Hemoglobin <8 g/dL.
- Current participation in another drug trial or intervention study.
- Inability or unwillingness to complete the study procedures.

MS considerations:

- Clinical relapse within the last three months (to ensure disease stability)
- Steroid treatment in prior 1 month
- Evidence of other structural brain disease (e.g. prior stroke)

MRI considerations:

- Left handed
- Metal implants
- Prior head trauma
- Claustrophobia requiring anxiolytic or sedation, or other contraindication to MRI.

7 CONCURRENT MEDICATIONS

All subjects should be maintained on the same medications throughout the entire study period, as medically feasible, with no introduction of new chronic therapies.

7.1 Allowed Medications and Treatments

Standard therapy for Multiple Sclerosis is allowed except for treatments noted in the exclusion criteria described above and as noted in the prohibited medications section below.

Prohibited Medications and Treatments

The following medications are prohibited during the study and administration will be considered a protocol violation: other hormone therapies, other hot flash therapies, UGT inducers (lamotrigine, oxcarbazepine).

8 STUDY TREATMENTS

8.1 Method of Assigning Subjects to Treatment Groups

Up to 24 eligible patients will be randomly assigned to DUAVEE or placebo treatment groups in a 1:1 ratio using a SAS-based computer-generated randomization scheme developed by the study

data management provider. The CTSI statistician advising the PI will complete a randomization worksheet.

8.2 Blinding

Due to the objectives of the study, the identity of test and control treatments will not be known to investigators, research staff, or patients. The following study procedures will be in place to ensure double-blind administration of study treatments.

- Access to the randomization code will be strictly controlled.

- The CTSI statistician advising the PI will perform the randomization/blinding chart and provide this to the Compounding Pharmacy. They will perform interim safety analysis without umblinding the study, and will only unblind the study to the investigators at the end of study.

- Packaging and labeling of test and control treatments will be identical to maintain the blind.

- Packaging and labeling will be done by pharmacy personnel who are not involved in analysis of study data.

The study blind will be broken on completion of the clinical study and after the study database has been locked. The investigators will be given the randomization worksheet from the CTSI statistician after the last participant has completed their study.

During the study, the blind may be broken **only** in emergencies when knowledge of the patient's treatment group is necessary for further patient management. When possible, the Investigator should discuss the emergency with Dr. Ari Green prior to unblinding.

8.3 Formulation of Test and Control Products

8.3.1 Formulation of Test Product

DUAVEE (conjugated estrogens/bazedoxifene) tablets, 0.45 mg/20 mg will be manufactured by Pfizer, Inc.

Duavee is an oral, biconvex, pink tablet branded with "0.45/20" in black ink on one side.

For blinding, it will be made into encapsulated into a gelatin capsule and fill remaining space with microcrystalline cellulose, by the Safeway Compounding Pharmacy (Dr. Jack Castaldo) at:

Compounding Pharmacist/BHRT Consultant
Safeway Compounding Pharmacy
6100 Hellyer Ave #100
San Jose, Ca. 95138.

Table X: Formulation and Measured pH of Duavee and Placebo

	Duavee	**Placebo**
Active Ingredient, mg/mL	Conjugated estrogens, 0.45 mg (mixture of sodium estrone sulfate and sodium equilin sulfate and other components, including sodium sulfate conjugates, 17α-dihydroequilin, 17α-estradiol, and 17β-dihydroequilin.)	

	Bazedoxifene, 20 mg	
Other ingredient, mg/mL	calcium phosphate tribasic, hydroxypropyl cellulose, microcrystalline cellulose, powdered cellulose, hypromellose, lactose monohydrate, magnesium stearate, polyethylene glycol, sucrose, ascorbic acid, sucrose palmitic acid ester, hydroxyethylcellulose, titanium dioxide, red iron oxide, yellow iron oxide, black iron oxide, povidone, polydextrose, maltitol, poloxamer 188, propylene glycol, isopropyl alcohol.	microcrystalline cellulose
pH		

8.3.2 Formulation of Control Product

Placebo is: Microcrystalline cellulose, encapsulated into a gelatin capsule, taken daily.

8.3.3 Packaging and Labeling

Both the drug and the placebo will be single use daily tablets packaged into monthly blister packs. Each carton (kit) of study drug will be labeled with the required FDA warning statement, the protocol number, a treatment number, the name of the sponsors, and directions for patient use and storage.

8.4 Supply of Study Drug at the Site

The Sponsor (or designee) will ship Study Drug to the investigational pharmacy. The initial study drug shipment will be shipped after site activation (i.e., all required regulatory documentation has been received by the Sponsor and a contract has been executed). Subsequent study drug shipments will be made after site request for resupply.

The randomization and blinding chart will be provided by CTSI statistician to the pharmacy personnel. The Safeway Compounding Pharmacy then ship the study drug to the PI, and the research team (PI or research coordinator) will give the 8-week package to participants at their Visit 1.

8.4.1 Dosage/Dosage Regimen

The medication (Duavee or placebo) will be taken once daily, in the morning, for 8 weeks. The Duavee dosing is: 0.45mg conjugated estrogens/20 mg bazedoxifene.

8.4.2 Dispensing

The Safeway Compounding pharmacy will ship the drugs to study personnel, who will then give the samples to the study participants at their Visit 1.

8.4.3 Administration Instructions

Patients will take this tablet orally once a day.

8.5 Supply of Study Drug at the Site

Study personnel will receive the Study Drug/placebo shipment from the Safeway Compounding Pharmacy and maintain it under locked conditions in the Sandler Neurosciences Center. Subjects will be randomized and provided the study drug on the same day.

8.5.1 Storage

Study drug should be stored by the study site at controlled room temperature, 15 to 30ºC (59 to 86ºF). Subjects will be instructed to store the medication in blister packaging at room temperature according to the instructions outlined on the Drug Administration Instructions.

8.6 Study Drug Accountability

An accurate and current accounting of the dispensing and return of study drug for each subject will be maintained on an ongoing basis by a member of the study site staff. The number of study drug dispensed and returned by the subject will be recorded on the Investigational Drug Accountability Record. The study monitor will verify these documents throughout the course of the study.

8.7 Measures of Treatment Compliance

Subjects will be asked to keep a patient diary noting the day and date they take their study drug and any adverse events. They will be asked to bring their patient diary to each study visit along with all used and unused study drug containers.

9 STUDY PROCEDURES AND GUIDELINES

A Schedule of Events representing the required testing procedures to be performed for the duration of the study is diagrammed in Appendix 1.

Prior to conducting any study-related activities, written informed consent and the Health Insurance Portability and Accountability Act (HIPAA) authorization must be signed and dated by the subject. If appropriate, assent must also be obtained prior to conducting any study-related activities.

9.1 Clinical Assessments

9.1.1 Concomitant Medications

All concomitant medication and concurrent therapies will be documented at Screening and at the first Study Visit on Week 2, at final Visit on Week 10, and at early termination when applicable. Dose, route, unit frequency of administration, and indication for administration and dates of medication will be captured.

9.1.2 Demographics

Demographic information (date of birth, gender, race) will be recorded at Screening.

9.1.3 Medical History

Relevant medical history, including history of current disease, other pertinent respiratory history, and information regarding underlying diseases will be recorded at Screening.

9.1.4 Physical Examination

A complete physical examination will be performed by either the investigator or a sub-investigator who is a physician at the Screening Visit. Qualified staff (MD, NP, RN, and PA) may complete the abbreviated physical exam at all other visits. New abnormal physical exam findings must be documented and will be followed by a physician or other qualified staff at the next scheduled visit.

9.1.5 Vital Signs

Body temperature, blood pressure, pulse and respirations will be performed after resting for 5 minutes at all Study Visits.

9.1.6 Other Clinical Procedures

Neurological examination:
Expanded Disability Status Scale (EDSS): EDSS Score will be determined, based on neurological examination, by a trained certified EDSS rater at scheduled visits according to the Schedule Table, using the validated Neurostatus scoring system. Individual patients will have EDSS performed by same rater on each visit. The EDSS is an ordinal scale used for assessing neurological impairment of MS based on a neurological examination. It consists of scores in each of seven functional systems (FS) and an ambulation score that are then combined to determine the EDSS [ranging from 0 (normal) to 10 (death due to MS)]. The FSs are the Visual, Brain Stem, Pyramidal, Cerebellar, Sensory, Bowel & Bladder, and Cerebral functions. The FSs and EDSS steps will be assessed in a standardized manner. EDSS is a widely used and accepted instrument to evaluate disability status at a given time and longitudinally, to assess disability progression in clinical studies in MS.

T25W: The Timed 25-foot Walk will be assessed according to the schedule in the Table. The Timed Walk is an objective quantitative test of neurological function, and is one of three components of the Multiple Sclerosis Functional Composite (MSFC), a composite measure assessing upper extremity function, ambulation and cognitive function (Fisher et al. 1999). The Timed Walk is an ambulation measurement: a walk of 25 feet (7.62 meters): time taken in seconds. The Timed Walk test will be administered either by the Independent EDSS Rater or by another qualified individual not involved in the treatment of the patients.

Patient-reported Outcomes:
MSQOL 54: The Multiple Sclerosis Quality of Life- 54 (MSQOL-54) was the first MS-specific QOL measure.(1, 2) It includes a 36 generic instrument as well as 18 additional items, for a total of 54 items. (3) It is graded on 12 subscales and 2 summary scores: the Physical Health and Mental Health Composite Summaries.

MFIS: The Modified Fatigue Impact Scale (MFIS)(4) is a 21-item fatigue scale that measures the physical, cognitive, and psychosocial aspects of fatigue.

HAD: Hospital Anxiety and Depression Scale (HADS)(5) cutoff is 8 for anxiety, depression.

MS Rating Score-Revised: (MSRS, free under Creative Commons License), patients rate their disability on a 0-4 scale in 8 areas (walking, use of upper extremities, speech disturbance, vision, dysphagia, cognitive or affective disturbance, sensory disturbance and bowel/bladder). Composite scores range from 0-32. The patient-reported MSRS has shown high internal consistency,

concordant validity with the NARCOMS Patient-Determined Disease Step, and demonstrated adequate known-groups validity (6). In a clinic population, the previous version (MSRS) has demonstrated good correlations with physician-derived EDSS (MSRS composite score rs=0.61, MSRS walking domain rs =0.74)(7).

Bladder Control (MSQLI version): This questionnaire assesses bladder symptoms and has been validated in MS (8).

MSNQ: MS Neuropsychological Screening questionnaire (9).

Neuropsychological Tests:
Symbol Digit Modalities Test (SDMT):
The SDMT is a sensitive and specific test to assess processing speed and working memory which are typically affected domains in cognitively impaired MS patients. The SDMT has been found to accurately categorize 72% of MS subjects as cognitively impaired based on a longer, comprehensive test battery.
During the administration of the SDMT, only the examiner and the patient should be in the testing room. The examiner can be the study coordinator, independent EDSS Rater or a qualified individual not involved in the treatment of the patients who has been instructed in the appropriate administration of the test. Patients are presented with a test instrument at the top of which there is a row of nine numbers paired with unique symbols. Below this part of the test instrument is an array of symbols paired with empty spaces, the patient's task is to verbally match the number for each symbol as quickly as possible. The test takes approximately 5 minutes to administer. The test scoring is calculated based on the number of correct answers in 90 seconds. (Partmenter et al, Mult Scler 2007;13:52-57).

Letter Number Sequencing Subtest: LNS, from the Wechsler Memory Scale (verbal working memory). This test, of 5 minute duration, will be used to assess verbal working memory. We will use the recall score.

9.1.7 Adverse Events

Information regarding occurrence of adverse events will be captured throughout the study. Duration (start and stop dates and times), severity/grade, outcome, treatment and relation to study drug will be recorded on the case report form (CRF).

9.2 Clinical Laboratory Measurements

9.2.1 Hematology

Blood will be obtained and sent to each site's clinical hematology lab for a complete blood count (hemoglobin, hematocrit, red blood cell count, white blood cell count, white blood cell differential, and platelet count).

9.2.2 Blood Chemistry Profile

Blood will be obtained and sent to each site's clinical chemistry lab for determination of serum sodium, potassium, chloride, bicarbonate, random glucose, BUN, creatinine, aspartate aminotransferase (AST/SGOT), alanine aminotransferase (ALT/SGPT), alkaline phosphatase, total bilirubin, direct bilirubin.

Additionally, the following hormones will be measured:

- antimullerian hormone (AMH)
- follicle stimulating hormone (FSH)
- luteinizing hormone (LH)
- estradiol (E2)
- progesterone (P)
- testosterone (total and free)
- sex hormone binding globulin (SHBG)
- vitamin D.
- estrone (E1)

9.2.3 Pregnancy Test

A serum pregnancy test will be obtained from female subjects who are of childbearing age prior to their participation in the study.

9.3 Research Laboratory Measurements (include sections as appropriate)

9.3.1 Peripheral blood mononuclear cells (PBMCs). These will be stored for future use. Should there be any significant hormonal changes observed during the study, then we will pursue immunology testing to probe
correlations between hormonal and immunological changes.

9.3.2. MRI

There will be an MRI exam at Visit 1 and 2, for a subset of the first 12 participants.
Prior to the MRI, each participant will be asked a number of questions concerning his or health, lack of metal implants and absence of claustrophobia.
They will then be asked to lie down on a narrow bed that will then be moved into a tunnel that is 6 feet by 2.5 feet. Each person will need to lie there quietly for about one hour during which time machine-like banging noises will occur. Earplugs will be provided to reduce the sound level. Communication with the technologists outside will be possible at all times by a microphone and loudspeaker. If the participant wishes to be removed from the magnet, this will be done immediately. Towards the end of the exam, an injection of gadolinium contrast will be performed by a registered nurse or MR technologist, just as is done for many routine clinical exams. If necessary, an I.V. (intravenous) line may be set up during or prior to the exam to allow the injection of this compound during the MRI acquisition. I.V. access is the placing of a small Teflon catheter or small needle into one of the veins, preferably in the arm. A small amount of fluid, usually normal saline (salt water) or dextrose (sugar) 5% in water is dripped in to prevent the catheter from clogging.
The MRI scan performed for the study will be reviewed by a neuroradiologist. The Primary Treating Physician must be contacted in case of unexpected findings (not consistent with MS) detected on the MRI scan for safety actions and Adverse Event reporting. During the study the scan will be assessed by the blinded trained physician from the NIC MS MRI team. Upon acquisition MRI will be evaluated for quality, completeness, and adherence to the manual. Confirmation of MRI quality or a description of the quality problems, if detected, will be communicated to the investigators. If scan is incomplete or incorrectly performed, the study center will be asked to repeat it as soon as possible. After completion of the quality check, all scans will be analyzed according to a standardized procedure.

10 EVALUATIONS BY VISIT

10.1 Visit 1 (Week 0) - SCREENING

1. Review the study with the subject and obtain written informed consent and HIPAA authorization and assent, if appropriate.
2. Assign the subject a unique screening number.
3. Record demographics data.
4. Record medical history, including a history of MS, diagnosis date, and prior MS treatments, as well as reproductive and menopausal history.
5. Record concomitant medications.
6. Perform a complete physical examination.
7. Perform and record vital signs.
8. Perform and record results of blood pressure testing.
9. Collect blood for clinical laboratory tests (chemistry, hematology, pregnancy test, hormones).
10. Initiate subject diary
11. Schedule subject for Visit 2 in 14 days +/-2.

10.2 Visit 2 (Day 14)

1. Concomitant medications review.
2. Perform abbreviated physical examination.
3. Perform neurological examination.
4. Obtain patient reported outcomes
5. Perform neuropsychological testing
6. Perform and record vital signs.
7. Randomize subject, Dispense study drug
8. Collect blood for research laboratory tests
9. MRI in ½ subjects.

10.3 Visit 3 (Week 8)

1. Record any Adverse Experiences and/or Review subject diary for adverse experiences and dosing compliance.
2. Record changes to concomitant medications.
3. Perform abbreviated physical examination.
4. Perform neurological examination.
5. Obtain patient reported outcomes
6. Perform neuropsychological testing

7. Perform and record vital signs.
8. Randomize subject, Dispense study drug
9. Collect blood for research laboratory tests
10. MRI in first ½ subjects.

10.4 Early Withdrawal Visit

1. Record any Adverse Experiences and/or Review subject diary for adverse experiences and exclusionary medication use.
2. Record changes to concomitant medications.
3. Perform complete physical examination.
4. Perform and record vital signs.
5. Collect blood for clinical laboratory tests: Chemistry, Hematology, Urinalysis, hormones.

11 ADVERSE EXPERIENCE REPORTING AND DOCUMENTATION

11.1 Adverse Events

An adverse event (AE) is any untoward medical occurrence in a clinical investigation of a patient administered a pharmaceutical product and that does not necessarily have a causal relationship with the treatment. An AE is therefore any unfavorable and unintended sign (including an abnormal laboratory finding), symptom or disease temporally associated with the administration of an investigational product, whether or not related to that investigational product. An unexpected AE is one of a type not identified in nature, severity, or frequency in the current Investigator's Brochure or of greater severity or frequency than expected based on the information in the Investigator's Brochure.

The Investigator will probe, via discussion with the subject, for the occurrence of AEs during each subject visit and record the information in the site's source documents. Adverse events will be recorded in the patient CRF. Adverse events will be described by duration (start and stop dates and times), severity, outcome, treatment and relation to study drug, or if unrelated, the cause.

AE Severity

The National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE) Version 3.0 should be used to assess and grade AE severity, including laboratory abnormalities judged to be clinically significant. The modified criteria can be found in the study manual. If the experience is not covered in the modified criteria, the guidelines shown in Table 1 below should be used to grade severity. It should be pointed out that the term "severe" is a measure of intensity and that a severe AE is not necessarily serious.

Table 1. AE Severity Grading

Severity (Toxicity Grade)	Description
Mild (1)	Transient or mild discomfort; no limitation in activity; no medical intervention or therapy required. The subject may be aware of the sign or symptom but tolerates it reasonably well.
Moderate (2)	Mild to moderate limitation in activity, no or minimal medical intervention/therapy required.
Severe (3)	Marked limitation in activity, medical intervention/therapy required, hospitalizations possible.
Life-threatening (4)	The subject is at risk of death due to the adverse experience as it occurred. This does not refer to an experience that hypothetically might have caused death if it were more severe.

AE Relationship to Study Drug

The relationship of an AE to the study drug should be assessed using the following the guidelines in Table 2.

Table 2. AE Relationship to Study Drug

Relationship to Drug	Comment
Definitely	Previously known toxicity of agent; or an event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to the suspected drug; that is confirmed by stopping or reducing the dosage of the drug; and that is not explained by any other reasonable hypothesis.
Probably	An event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to the suspected drug; that is confirmed by stopping or reducing the dosage of the drug; and that is unlikely to be explained by the known characteristics of the subject's clinical state or by other interventions.
Possibly	An event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to that suspected drug; but that could readily have been produced by a number of other factors.
Unrelated	An event that can be determined with certainty to have no relationship to the study drug.

11.2 Serious Adverse Experiences (SAE)

An SAE is defined as any AE occurring at any dose that results in any of the following outcomes:

- death
- a life-threatening adverse experience
- inpatient hospitalization or prolongation of existing hospitalization
- a persistent or significant disability/incapacity

- a congenital anomaly/birth defect

Other important medical events may also be considered an SAE when, based on appropriate medical judgment, they jeopardize the subject or require intervention to prevent one of the outcomes listed.

11.2.1 Serious Adverse Experience Reporting

Study sites will document all SAEs that occur (whether or not related to study drug) per UCSF CHR Guidelines. The collection period for all SAEs will begin after informed consent is obtained and end after procedures for the final study visit have been completed.

All adverse reporting guidelines will be followed carefully. There are no procedures in this protocol that are considered to be high risk or pose physical harm to subjects. Nevertheless, unforeseen adverse events may occur. Any adverse event will be reported in writing to the IRB within 24 hours of the event. We will then provide a full written report to the Partners Human Studies Committees within 10 working days/14 calendar days. Study staff members will meet together weekly in a private room with the Principal Investigator to review subject-related issues or data problems. The Study Coordinator takes notes from the weekly meetings and records them into minutes which are kept on file. All information gathered in this study is reviewed and cross-checked for accuracy and completeness by the PI. Study staff will also ensure the adherence to the IRB-approved protocol and integrity of the data by monitoring active case reports for missing data or errors, including the completeness of consent forms, on a weekly basis. If any information is missing or errors are found, study staff will notify the Principal Investigator and work with her to develop a plan of corrective action.
All Human Subjects Committees will be notified in writing if missing data or errors are determined to be a protocol deviation/violation within 24 hours.
For more urgent and severe adverse events, the PI will be notified immediately and a plan of action will be developed according to the policies and protocols developed over many years. The IRB will be notified as described above.

In accordance with the standard operating procedures and policies of the local Institutional Review Board (IRB)/Independent Ethics Committee (IEC), the site investigator will report SAEs to the IRB/IEC.

11.3 Medical Monitoring

Dr. Riley Bove should be contacted directly at these numbers to report medical concerns or questions regarding safety.

Phone: (415) 595 2795
Pager: (415) 443 0871

12 DISCONTINUATION AND REPLACEMENT OF SUBJECTS

12.1 Early Discontinuation of Study Drug

A subject may be discontinued from study treatment at any time if the subject, the investigator, or the Sponsor feels that it is not in the subject's best interest to continue. The following is a list of possible reasons for study treatment discontinuation:

- Subject withdrawal of consent (or assent)
- Subject is not compliant with study procedures
- Adverse event that in the opinion of the investigator would be in the best interest of the subject to discontinue study treatment:
    - **Uterine bleeding, venous thromboembolic event**
- Protocol violation requiring discontinuation of study treatment
- Lost to follow-up
- Sponsor request for early termination of study
- Positive pregnancy test

If a subject is withdrawn from treatment due to an adverse event, the subject will be followed and treated by the Investigator until the abnormal parameter or symptom has resolved or stabilized.

All subjects who discontinue study treatment should come in for an early discontinuation visit as soon as possible and then should be encouraged to complete all remaining scheduled visits and procedures.

All subjects are free to withdraw from participation at any time, for any reason, specified or unspecified, and without prejudice.

Reasonable attempts will be made by the investigator to provide a reason for subject withdrawals. The reason for the subject's withdrawal from the study will be specified in the subject's source documents Refer to Section 10 for early termination procedures.

12.3 Withdrawal of Subjects from the Study

A subject may be withdrawn from the study at any time if the subject, the investigator, or the Sponsor feels that it is not in the subject's best interest to continue.

All subjects are free to withdraw from participation at any time, for any reason, specified or unspecified, and without prejudice.

Reasonable attempts will be made by the investigator to provide a reason for subject withdrawals. The reason for the subject's withdrawal from the study will be specified in the subject's source documents. As noted above, subjects who discontinue study treatment early (i.e., they withdraw prior to Visit 3) should have an early discontinuation visit. Refer to Section 10 for early termination procedures.

12.4 Replacement of Subjects

Subjects who withdraw from the study treatment prior to Visit 3 will be replaced.

Subjects who withdraw from the study will be replaced.

13 PROTOCOL VIOLATIONS

A protocol violation occurs when the subject or investigator fails to adhere to significant protocol requirements affecting the inclusion, exclusion, subject safety and primary endpoint criteria. Protocol violations for this study include, but are not limited to, the following:

Failure to meet inclusion/exclusion criteria

Use of a prohibited concomitant medication

Failure to comply with Good Clinical Practice (GCP) guidelines will also result in a protocol violation. The PI will determine if a protocol violation will result in withdrawal of a subject.

When a protocol violation occurs, it will be discussed with the investigator and a Protocol Violation Form detailing the violation will be generated. This form will be signed by a Sponsor representative and the Investigator. A copy of the form will be filed in the site's regulatory binder and in the Sponsor's files.

14 DATA SAFETY MONITORING

Safety Data Monitoring throughout the trial: Since this trial is a single-site trial and the overseeing PI will be present throughout the trial, a Data and Safety Monitoring Board is not deemed necessary.

The study investigator will be responsible for monitoring safety data and adverse events recorded in the patient file and will be responsible for reporting such events to the CHR.

- In addition to the PI, a staff neurologist (Dr. Ari Green, a senior clinician with expertise in MS clinical trials) and a staff obstetrician/gynecologist (Dr. Robin Lamar) will review together the interim safety analyses to be conducted after (1) 12 patients have completed the entire trial, including MRI (2) each patient has completed the week 6 phone call, and (3) any AEs are reported.
    - **STOPPING RULE:**
        - **MRI: 2-fold increase in new T2 or gado enhancing lesions in treatment compared to placebo group**
    - **PATIENT WITHDRAWAL RULE:**
        - **Abnormal uterine bleeding**
        - **Thromboembolic event (DVT, PE, other)**
- Further details regarding the timing and content of the interim reviews is included in the statistical section below.

Safety assessments will be throughout patient visits to ensure patient welfare and safety in daily activities. Such assessments will include:

- Physical Examination
- Vital Signs
- Height and Weight measurements
- Pregnancy and Assessments of Fertility
- Fatigue Assessment
- Health Status Questionnaire
- Blood tests to assess potential abnormal increase of liver enzymes and serum triglycerides
- Lab tests to assess hormone levels.
- MRIs

The Investigator will meet with the study staff biweekly to review safety as well as efficacy data collected from patient visits and assess the safety of the visits and study outcomes.

Patient withdrawal from the study / Stopping rules: Study treatment must be discontinued for any patient if the investigator determines that continuing would result in a significant risk for that patient. The following conditions/events may be considered sufficient to support a decision about the study drug discontinuation in individual cases:

- Adverse event/serious adverse event
- Abnormal laboratory value(s) or abnormal test result(s)
- Withdrawal of informed consent
- Pregnancy
- Use of prohibited other investigational medications
- Deviations from the prescribed dose regimen for the study drug
- Any other protocol deviation that results in a significant risk to the patient's safety

Discontinuation of the study drug must be recorded along with the patient information in REDCap, giving the date and primary and secondary reasons for stopping study treatment. The investigator must also contact the Committee on Human Research at the study site (University of California at San Francisco Multiple Sclerosis Center) to register the patient's discontinuation from randomized study drug. Patients may voluntarily withdraw from the study for any reason at any time. They may be considered withdrawn if they state an intention to withdraw, fail to return for visits, or become lost to follow-up for any other reason. For patients who are lost to follow-up (i.e. those patients whose status is unclear because they fail to appear for study visits without stating an intention to withdraw), the investigator should show "due diligence" by documenting in the source documents steps taken to contact the patient, e.g. dates of telephone calls, registered letters, etc. If premature withdrawal occurs for any reason, the investigator must make every effort to determine the primary reason for a patient's premature withdrawal from the study and record this information with the patient's research data in REDCap as well as report this withdrawal to the Committee on Human Research if deemed necessary by the committee.

Patients will be considered to have completed the study if they attend all protocol specified visits and complete all assessments. A patient who discontinues the study medication for any reason, including an adverse event must complete a Safety Follow-Up visit. Any patient still in screening at the time 24 patients have been enrolled will be allowed to continue screening to determine if they will be eligible to enter the study, provided all screening assessments are complete and the randomization visit can occur on or before the designated end of subject recruitment date. The investigator must provide follow-up medical care for all patients who are prematurely withdrawn from the study or must refer them for appropriate ongoing care. The study can be terminated at any time for any reason by the investigator. Should this be necessary, each patient should be seen as soon as possible and treated as described for a prematurely withdrawn patient. The investigator may be informed of additional procedures to be followed in order to ensure that adequate consideration is given to the protection of the patient's interests. The investigator will be responsible for informing the Committee on Human Research of the trial's early termination.

15 STATISTICAL METHODS AND CONSIDERATIONS

Prior to the analysis of the final study data, a detailed Statistical Analysis Plan (SAP) will be written describing all analyses that will be performed. The SAP will contain any modifications to the analysis plan described below.

15.1 Data Sets Analyzed

All eligible patients who are randomized into the study and receive at least one dose of the study drug (the Safety Population) will be included in the safety analysis.

15.2 Demographic and Baseline Characteristics

The following demographic variables at screening will be summarized by treatment assignment: race, age, height and weight.

15.3 Analysis of Primary Endpoint

We will compare outcomes at baseline and at 8 weeks, using t-tests, between women receiving TSEC vs. placebo. Then, regressions will be run to adjust for subject age, disease duration, and DMT (disease modifying therapy).

Primary endpoints:

Aim 1. To explore the hypothesis that CE+BZA improves menopausal symptoms in MS women. Outcomes: Our primary measure will be the number of daily vasomotor symptoms (VMS), obtained from the patient diary. In healthy women, this was shown to significantly decrease with estradiol treatment at 8 weeks).(Joffe et al., 2014)

Power calculation: Given this is a pilot study, we will view the study as a success if the observed mean change in the treatment group is larger by any amount compared to the placebo group ("play the winner" approach). In healthy women treated with estradiol for 8 weeks,(Joffe et al., 2014) VMS decreased by 2.2 daily in the placebo group and 4.5 in the estradiol group and the SD for the change was 3.4. Assuming same differences and SD, we will have 80% power to find the treatment arm mean difference to be larger with our proposed sample size.

Aim 2. To explore the hypothesis that CE+BZA results in improvement in MS-related outcomes. Our primary outcome will be MSQOL54 composite score (primary).

Aim 3. To explore the hypothesis that CE+BZA for 8 weeks is tolerable in women with MS. Our primary measure will be the percentage of subjects reporting side effects on the Satisfaction Questionnaire for Medication (TSQM)(Atkinson et al., 2004), which is commonly used in MS treatment studies (e.g.(Calkwood et al., 2014)).

15.4 Analysis of Secondary Endpoints

These methods will be the same as per primary endpoints.
Secondary endpoints are:

Aim 1. To explore the hypothesis that CE+BZA improves menopausal symptoms in MS women. Our secondary measure will be sleep quality from sleep diary.

Aim 2. To explore the hypothesis that CE+BZA results in improvement in MS-related outcomes. Our secondary outcomes will be: measures of mood (CES-D), fatigue (MFIS), cognition (MSNQ), and bladder (MSQLI BLCS), and our primary outcome is the MSRS, a global score capturing patient impressions of MS severity across eight function domains.

Aim 3. To explore the hypothesis that CE+BZA for 8 weeks is tolerable in women with MS. Our secondary measures will be number of missed doses, and the number of new or enhancing lesions on 8-week MRI, to verify that CE+BZA does not yield any marked changes in inflammatory activity.

Safety and tolerability data will be summarized by treatment group.

Adverse event rates will be coded by body system and MedDra classification term. Adverse events will be tabulated by treatment group and will include the number of patients for whom the event occurred, the rate of occurrence, and the severity and relationship to study drug.

15.5 Interim Analysis

After each patient completes the Week 6 phone call, the side effects reported will be analyzed.

When 50% of patients have completed the study through Study Visit 2 (total visit 3), an interim analysis for safety will be conducted by the PI, with the collaboration of Drs. Green (neurology) and Dr. Lamar (Ob/Gyn).

Serious adverse events will be monitored by the committee on an ongoing basis throughout the study.

15.6 Sample Size and Randomization

The sample size for this protocol was determined by the following. Given this is a pilot study, we will view the study as a success if the observed mean change in the treatment group is larger by any amount compared to the placebo group ("play the winner" approach). In healthy women treated with estradiol for 8 weeks,(Joffe et al., 2014) VMS decreased by 2.2 daily in the placebo group and 4.5 in the estradiol group and the SD for the change was 3.4. Assuming same differences and SD, we will have 80% power to find the treatment arm mean difference to be larger with our proposed sample size.

Subjects will be 1:1 randomized.

16 DATA COLLECTION, RETENTION AND MONITORING

16.1 Data Collection Instruments

The Investigator will prepare and maintain adequate and accurate source documents designed to record all observations and other pertinent data for each subject treated with the study drug.

Study personnel at each site will enter data from source documents corresponding to a subject's visit into the protocol-specific electronic Case Report Form (eCRF) when the information corresponding to that visit is available. Subjects will not be identified by name in the study database or on any study documents to be collected by the Sponsor (or designee), but will be identified by a site number, subject number and initials.

If a correction is required for an eCRF, the time and date stamps track the person entering or updating eCRF data and creates an electronic audit trail.

The Investigator is responsible for all information collected on subjects enrolled in this study. All data collected during the course of this study must be reviewed and verified for completeness and

accuracy by the Investigator. A copy of the CRF will remain at the Investigator's site at the completion of the study.

16.2 Data Management Procedures

The data will be entered into a validated database. The Data Management group will be responsible for data processing, in accordance with procedural documentation. Database lock will occur once quality assurance procedures have been completed.

All procedures for the handling and analysis of data will be conducted using good computing practices meeting FDA guidelines for the handling and analysis of data for clinical trials.

16.3 Data Quality Control and Reporting

After data have been entered into the study database, a system of computerized data validation checks will be implemented and applied to the database on a regular basis. Queries are entered, tracked, and resolved through the EDC system directly. The study database will be updated in accordance with the resolved queries. All changes to the study database will be documented.

16.4 Archival of Data

The database is safeguarded against unauthorized access by established security procedures; appropriate backup copies of the database and related software files will be maintained. Databases are backed up by the database administrator in conjunction with any updates or changes to the database.

At critical junctures of the protocol (e.g., production of interim reports and final reports), data for analysis is locked and cleaned per established procedures.

16.5 Availability and Retention of Investigational Records

The Investigator must make study data accessible to the monitor, other authorized representatives of the Sponsor (or designee), IRB/IEC, and Regulatory Agency (e.g., FDA) inspectors upon request. A file for each subject must be maintained that includes the signed Informed Consent, HIPAA Authorization and Assent Form and copies of all source documentation related to that subject. The Investigator must ensure the reliability and availability of source documents from which the information on the CRF was derived.

All study documents (patient files, signed informed consent forms, copies of CRFs, Study File Notebook, etc.) must be kept secured for a period of two years following marketing of the investigational product or for two years after centers have been notified that the IND has been discontinued. There may be other circumstances for which the Sponsor is required to maintain study records and, therefore, the Sponsor should be contacted prior to removing study records for any reason.

16.6 Monitoring

Monitoring visits will be conducted by representatives of the Sponsor according to the U.S. CFR Title 21 Parts 50, 56, and 312 and ICH Guidelines for GCP (E6). By signing this protocol, the Investigator grants permission to the Sponsor (or designee), and appropriate regulatory authorities to conduct on-site monitoring and/or auditing of all appropriate study documentation.

16.7 Subject Confidentiality

In order to maintain subject confidentiality, only a subject number and subject initials will identify all study subjects on CRFs and other documentation submitted to the Sponsor. Additional subject confidentiality issues (if applicable) are covered in the Clinical Study Agreement.

17 ADMINISTRATIVE, ETHICAL, REGULATORY CONSIDERATIONS

The study will be conducted according to the Declaration of Helsinki, Protection of Human Volunteers (21 CFR 50), Institutional Review Boards (21 CFR 56), and Obligations of Clinical Investigators (21 CFR 312).

To maintain confidentiality, all laboratory specimens, evaluation forms, reports and other records will be identified by a coded number and initials only. All study records will be kept in a locked file cabinet and code sheets linking a patient's name to a patient identification number will be stored separately in another locked file cabinet. Clinical information will not be released without written permission of the subject, except as necessary for monitoring by the FDA. The Investigator must also comply with all applicable privacy regulations (e.g., Health Insurance Portability and Accountability Act of 1996, EU Data Protection Directive 95/46/EC).

17.1 Protocol Amendments

Any amendment to the protocol will be written by the PI. Protocol amendments cannot be implemented without prior written IRB/IEC approval except as necessary to eliminate immediate safety hazards to patients. A protocol amendment intended to eliminate an apparent immediate hazard to patients may be implemented immediately, provided the IRBs are notified within five working days.

17.2 Institutional Review Boards and Independent Ethics Committees

The protocol and consent form will be reviewed and approved by the IRB/IEC of each participating center prior to study initiation. Serious adverse experiences regardless of causality will be reported to the IRB/IEC in accordance with the standard operating procedures and policies of the IRB/IEC, and the Investigator will keep the IRB/IEC informed as to the progress of the study. The Investigator will obtain assurance of IRB/IEC compliance with regulations.

Any documents that the IRB/IEC may need to fulfill its responsibilities (such as protocol, protocol amendments, Investigator's Brochure, consent forms, information concerning patient recruitment, payment or compensation procedures, or other pertinent information) will be submitted to the IRB/IEC. The IRB/IECs written unconditional approval of the study protocol and the informed consent form will be in the possession of the Investigator before the study is initiated. The IRB/IECs unconditional approval statement will be transmitted by the Investigator to the Sponsor prior to the shipment of study supplies to the site. This approval must refer to the study by exact protocol title and number and should identify the documents reviewed and the date of review.

Protocol and/or informed consent modifications or changes may not be initiated without prior written IRB/IEC approval except when necessary to eliminate immediate hazards to the patients or when the change(s) involves only logistical or administrative aspects of the study. Such modifications will be submitted to the IRB/IEC and written verification that the modification was submitted and subsequently approved should be obtained.

The IRB/IEC must be informed of revisions to other documents originally submitted for review; serious and/or unexpected adverse experiences occurring during the study in accordance with the standard operating procedures and policies of the IRB; new information that may affect adversely

the safety of the patients of the conduct of the study; an annual update and/or request for re-approval; and when the study has been completed.

17.3 Informed Consent Form

Informed consent will be obtained in accordance with the Declaration of Helsinki, ICH GCP, US Code of Federal Regulations for Protection of Human Subjects (21 CFR 50.25[a,b], CFR 50.27, and CFR Part 56, Subpart A), the Health Insurance Portability and Accountability Act (HIPAA, if applicable), and local regulations.

The Investigator will prepare the informed consent form, assent and HIPAA authorization and provide the documents to the Sponsor or designee for approval prior to submission to the IRB/IEC. The consent form generated by the Investigator must be acceptable to the Sponsor and be approved by the IRB/IEC. The written consent document will embody the elements of informed consent as described in the International Conference on Harmonisation and will also comply with local regulations. The Investigator will send an IRB/IEC-approved copy of the Informed Consent Form to the Sponsor (or designee) for the study file.

A properly executed, written, informed consent will be obtained from each subject prior to entering the subject into the trial. Information should be given in both oral and written form and subjects must be given ample opportunity to inquire about details of the study. If appropriate and required by the local IRB/IEC, assent from the subject will also be obtained. If a subject is unable to sign the informed consent form (ICF) and the HIPAA authorization, a legal representative may sign for the subject. A copy of the signed consent form (and assent) will be given to the subject and the original will be maintained with the subject's records.

17.4 Publications

The preparation and submittal for publication of manuscripts containing the study results shall be in accordance with a process determined by mutual written agreement among the study Sponsor and participating institutions. The publication or presentation of any study results shall comply with all applicable privacy laws, including, but not limited to, the Health Insurance Portability and Accountability Act of 1996.

17.5 Investigator Responsibilities

By signing the Agreement of Investigator form, the Investigator agrees to:

1. Conduct the study in accordance with the protocol and only make changes after notifying the Sponsor (or designee), except when to protect the safety, rights or welfare of subjects.
2. Personally conduct or supervise the study (or investigation).
3. Ensure that the requirements relating to obtaining informed consent and IRB review and approval meet federal guidelines, as stated in § 21 CFR, parts 50 and 56.
4. Report to the Sponsor or designee any AEs that occur in the course of the study, in accordance with §21 CFR 312.64.
5. Ensure that all associates, colleagues and employees assisting in the conduct of the study are informed about their obligations in meeting the above commitments.
6. Maintain adequate and accurate records in accordance with §21 CFR 312.62 and to make those records available for inspection with the Sponsor (or designee).

7. Ensure that an IRB that complies with the requirements of §21 CFR part 56 will be responsible for initial and continuing review and approval of the clinical study.
8. Promptly report to the IRB and the Sponsor (or designee) all changes in the research activity and all unanticipated problems involving risks to subjects or others (to include amendments and IND safety reports).
9. Seek IRB approval before any changes are made in the research study, except when necessary to eliminate hazards to the patients/subjects.
10. Comply with all other requirements regarding the obligations of clinical investigators and all other pertinent requirements listed in § 21 CFR part 312.

APPENDIX 1. SCHEDULE OF STUDY VISITS

	SCREENING VISIT 1 (Week 0)[a]	STUDY1 VISIT 2 (Week 2)[a]	STUDY2 VISIT 3 (Week 10)[a]
Informed Consent	X		
Medical History	X		
Complete Physical Exam	X		
Abbreviated Physical Exam		X	X
Height	X		
Weight	X	X	X
Vital Signs	X	X	X
Chemistry	X		X
Pregnancy Test (Serum)	X		
Hematology	X		
Hormone levels	X		
Research blood stored	X		X
Neurological Exam			
Questionnaires, testing		X	X
MRI (subset)		X	X
Randomization	X		
Dispensing or Administration of Study Drug		X	
Counting of Returned Study Drug			X
Initiate Subject Diary	X		
Subject Diary Review		X	X
Concomitant Medication Review	X	X	X
Adverse Experiences			

[a] ±2 day

Safety Phone call at Week 6.

1. Bebo BF, Jr., Dehghani B, Foster S, Kurniawan A, Lopez FJ, Sherman LS. Treatment with selective estrogen receptor modulators regulates myelin specific T-cells and suppresses experimental autoimmune encephalomyelitis. Glia 2009;57:777-790.

2. Kumar S, Patel R, Moore S, et al. Estrogen receptor beta ligand therapy activates PI3K/Akt/mTOR signaling in oligodendrocytes and promotes remyelination in a mouse model of multiple sclerosis. Neurobiology of disease 2013;56:131-144.

3. Bove R, Chitnis T, Houtchens M. Menopause in multiple sclerosis: therapeutic considerations. J Neurol 2014;261:1257-1268.

4. Komm BS, Mirkin S, Jenkins SN. Development of conjugated estrogens/bazedoxifene, the first tissue selective estrogen complex (TSEC) for management of menopausal hot flashes and postmenopausal bone loss. Steroids 2014.