Youth Depression Alleviation: Augmentation with an anti-inflammatory agent (YoDA-A): Protocol and rationale for a placebo-controlled randomised trial of rosuvastatin and aspirin

Abstract

Aim: There is growing support for the role of inflammation and oxidative stress in the pathophysiology of Major Depressive Disorder. This has led to the development of novel strategies targeting inflammation in the treatment of depression. Rosuvastatin and aspirin have well-documented anti-inflammatory and antioxidant properties. The aim of the Youth Depression Alleviation: Augmentation with an anti-inflammatory agent (YoDA-A) study is to determine whether individuals receiving adjunctive anti-inflammatory agents, aspirin and rosuvastatin, experience a reduction in the severity of MDD compared with individuals receiving placebo.

Methods: YoDA-A is a 12-week triple-blind, randomised controlled trial, funded by the National Health and Medical Research Council, Australia. Participants aged 15-25, with moderate-to-severe Major Depressive Disorder, are allocated to receive either 10mg/day rosuvastatin, 100 mg/day aspirin, or placebo, in addition to treatment as usual. Participants are assessed at baseline and at weeks 4, 8, 12, and 26. The primary outcome is change in the Montgomery-Åsberg Depression Rating Scale (MADRS) from baseline to week 12.

Results: The study is planned to be completed in 2017. At date of publication, 75 participants have been recruited.

Discussion: Timely and targeted intervention for youth MDD is crucial. Given the paucity of new agents to treat youth MDD, adjunctive trials are not only pragmatic and 'real-world', but additionally aim to target shortfalls in conventional medications. This study has the potential to first provide two new adjunctive treatment options for youth MDD; aspirin and rosuvastatin. Second, this study will serve as proof of principle of the role of inflammation in MDD.

Keywords: aspirin, depression, inflammation, rosuvastatin, youth.

Trial registration: Australian New Zealand Clinical Trials Registry: ACTRN12613000112763

Introduction

Depression is the most prevalent health problem in young people, with up to 24% experiencing major depressive disorder (MDD) before they reach the age of 25.[1] These rates may be increasing.[2] MDD in youth is of particular concern, not only due to the social and developmental impact of the disease[3,4] but also because depression during this period may lead to multiple recurrent major depressive episodes.[5,6]

Depression theory and treatment options have focused on the monoamine hypothesis, postulating that depression is the result of a deficiency of certain monoamine neurotransmitters.[7] While monoamines play a role in the pathophysiology of depression, this approach has neither delivered effective treatments for many individuals, or produced novel agents, beyond the limited treatment options currently available.[8,9] Current conceptualisation of the neurobiology of depression has shifted from monoamines, incorporating changes in inflammatory and oxidative stress into a more comprehensive model of illness neuroprogression.[10]

There are data demonstrating that stress and depression are associated with increased immune activation, impaired immune function, and inflammation.[11,12] Adolescent depression is associated with an inflammatory response, particularly marked in suicidal individuals.[13] Elevated levels of C-reactive protein (a marker of inflammation; CRP) predict the later onset of depression, suggesting that the immune findings are not mere epiphenomena of depression, but contribute to the genesis of the disorder.[14] Some antidepressant treatments reduce inflammatory marker levels in MDD patients[15,16] and reduce the release of inflammatory factors

from stimulated microglia.[17,18] A lack of treatment response to traditional antidepressants is associated with elevated inflammatory marker levels [19,20], suggesting an immunomodulatory role for successful use of antidepressant medication and opening the door to interventions that directly target the pathophysiology of depression.

The relationship between oxidative stress and depression is also supported. Compared with healthy controls, individuals experiencing a Major Depressive Episode have significantly elevated markers of oxidative damage[21], whilst another study demonstrated that oxidative stress was significantly higher in medication-free patients with MDD than in controls.[22] Positive correlations have been shown between oxidative stress index values and depressive severity scores.[22-26] Oxidative parameters return to normal with the resolution of depressive symptoms after selective serotonin re-uptake inhibitor (SSRI) treatment, supporting the contention that oxidative stress has an intrinsic role in the disease process.[23,27]

There is currently minimal evidence to support the effectiveness of any non-psychotherapeutic treatment for depression in young people, with the exception of fluoxetine.[28,29] Although effective, fluoxetine carries an increased risk of suicidal ideation in those aged under 25[30-33], as well as the risk of other side effects that may affect quality of life. There is, therefore, a clear need for the development of interventions with demonstrated effectiveness, which are acceptable to young people experiencing MDD. Statins (3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors), such as rosuvastatin, have well-documented anti-inflammatory

and antioxidant properties, as do aspirin and other non-steroidal anti-inflammatory drugs (NSAID).[34,35]

In vivo statin treatment has been demonstrated to reduce oxidative stress.[36-39] Rosuvastatin has been shown to reduce markers of oxidative stress and inflammation in animal studies[34] and in patients with hypertension and dyslipidaemia.[35] In an adult sample, it has been found that lovastatin, when used as adjunctive treatment, is effective for treating patients with MDD.[40] Both rosuvastatin and lovastatin belong to the class of statins, although the former is hydrophilic and the later lipophilic . Based on its action in reducing oxidative stress and the links with depressive symptoms, rosuvastatin may therefore be useful in reducing symptoms.

Adults with first-episode depression who received aspirin together with fluoxetine had a greater reduction of oxidative stress parameters than those who received fluoxetine monotherapy.[41] In schizophrenia, a recent study has shown that aspirin reduces core symptoms of the disorder, with a significant decrease in total and positive symptoms. [42] Given the biological similarities between mood and psychotic disorders, this is an intriguing lead.

The above evidence supports a possible role of oxidative and inflammatory processes in depression. It argues for the evaluation of rational and novel pathophysiologically-based therapies that are distinct from conventional antidepressants.

Method

Study Design

The study design is a 12-week, parallel group, triple-blind, randomised-control trial in participants with moderate to severe MDD, allocated to receive either rosuvastatin, or aspirin, or placebo, in addition to treatment as usual (TAU). Assessments are completed at baseline and weeks 4, 8 and 12, with a telephone follow-up assessment at week 26 to determine post-discontinuation effects. Blood samples are collected at baseline and week 12 for the investigation of inflammatory and oxidative stress markers. The most relevant markers, based on current literature at the end of the study, will be explored. The study was approved by the Melbourne Health Human Research Ethics Committee (#HREC/12/MH/148). The study was funded by the National Health and Medical Research Council of Australia and sponsored by Orygen, the National Centre of Excellence in Youth Mental Health, without limitations on publication.

The primary hypothesis is that after 12 weeks of treatment, both the rosuvastatin and aspirin treatment groups will show greater improvement from baseline than the placebo group, on the Montgomery-Åsberg Depression Rating Scale (MADRS).[43]

The secondary hypotheses are that the rosuvastatin and aspirin treatment groups will show greater improvement compared to the placebo group on measures of clinical global status, functioning, quality of life, and symptomatology from baseline to week 12; that these effects, and the reduction in MADRS will be seen at the week 26 follow-up in the rosuvastatin and aspirin groups, compared with placebo; and

that those in the active arms will have reduced levels of inflammatory and oxidative stress levels in serum; and that oxidative stress and inflammation will mediate clinical change.

Tertiary analyses will also be completed to explore mediating factors in the response to rosuvastatin and aspirin treatment.

Study setting

The study is being conducted at the Youth Mood Clinic at Orygen Youth Health, Jigsaw – Barwon Health, and in *headspace* centres in Geelong and Melbourne, Australia.

TAU at these sites includes case management, cognitive behavioural therapy, and pharmacotherapy.

The study commenced recruitment in June 2013 at OYH and *headspace* centres at Glenroy and Sunshine, Victoria. Recruitment at Barwon Health site and Geelong *headspace* will commence in coming months. The study has recruited 75 participants to date and of those, 49 have completed week 12 and 30 have completed week 26.

Inclusion and Exclusion Criteria

To be included in the study participants must: (i) be aged between 15 and 25 years; (ii) have a diagnosis of current MDD using the Structured Clinical Interview for DSM-IV Axis I Disorders, patient version (SCID-I/P)[44]; (iii) have a score on the MADRS[43] of 20 or greater, indicating moderate to severe depression; (iv) have the ability to give informed consent and to comply with standard procedures; (v) use effective contraception if female and sexually active with members of the opposite sex; (vi) have fluency in English; and (vii) have stable pharmacological treatment for at least two weeks prior to enrolment (any changes in medication dose or frequency of therapy is excepted), if currently being treated.

Exclusion criteria are: (i) lifetime or current SCID-I/P diagnosis of a psychotic disorder; (ii) a lifetime SCID-I/P diagnosis of bipolar I or II disorder or alcohol dependence; (iii) an acute or unstable systemic medical disorder; (iv) an inability to comply with the requirements of informed consent or the study protocol; (v) a history of intolerance or allergy to study medications; (vi) those who are currently pregnant or breast feeding (vii) the current regular use of statins, aspirin, NSAID's paracetamol, corticosteroids or any other immunomodulatory agents; (viii) the current or recent use of hypolipidaemics, vitamin K antagonists and other anticoagulants, protease inhibitors, ketoconazole, spironolactone; or cimetidine.

Discontinuation and withdrawal

Discontinuation of a participant occurs at the discretion of the participant, researcher, or treating physician. Automatic discontinuation occurs if a participant developments a psychotic disorder, bipolar disorder, is pregnant or at risk of

pregnancy, or if they commence rosuvastatin or aspirin treatment. Due to the increased risk of myopathy with rosuvastatin and concurrent heavy alcohol use, a score >20 on the Alcohol Use Disorders Identification Test (AUDIT)[45] necessitates review by the treating physician, and potential discontinuation. Due to the increased risk of bleeding with aspirin, any surgery planned by the participant requires review by the treating physician. Upon discontinuation, the participant is offered the opportunity to continue to contribute to data collection at the follow-up time points.

Interventions

Participants receive either 10mg/day rosuvastatin; or 100 mg/day aspirin; or placebo, in addition to TAU. At each visit, participants are requested to return all unused investigational products. Adherence to medication is assessed by a pill count.

The doses of rosuvastatin and aspirin are derived from the literature regarding the doses at which their targeted actions are effective and the safety profile of each agent. With these factors in mind, 10mg of rosuvastatin reflects the lowest prescribed therapeutic dose.[46] The 100mg dose of aspirin is the same dose used to prevent cardiac events and has been shown to have anti-inflammatory properties.[46] Blinding is maintained by ensuring that the packaging, and appearance of rosuvastatin, aspirin and placebo capsules are identical. All tablets were overcapsulated for blinding purposes, to appear and taste identical.

Outcome measures

The primary outcome measure is the MADRS.[43] The following measures are used to assess effectiveness: change in MADRS; change in the Quick Inventory of

Depression Symptomatology–Self Report (QIDS-SR)[47]; change of rates of remission (defined as MADRS ≤ 7); changes in the Generalized Anxiety Disorder 7-item scale (GAD-7)[48] scores; response, defined as the Clinical Global Impression–Severity scale and the Clinical Global Impression-Improvement scale[49]; and change in self-rated depressive symptoms, assessed using the Patient Global Impression Improvement (PGI)[50]. Quality of life is assessed using the Quality of life Enjoyment and Satisfaction Questionnaire – Short Form (Q-LES-Q-SF)[51] and the Social Adjustment Scale – Self Report (SAS-SR)[52]; The Social and Occupational Functioning Scale (SOFAS)[53] is used to measure psychosocial (social and occupational) functioning.

Predictors, moderators and mediators of treatment response

Potential predictors and moderators of treatment response are assessed using the Dimensional Assessment of Personality Pathology Basic Questionnaire (DAPP)[54], the SCID-I/P substance use module[44], the AUDIT[45] and the Negative Problem Orientation Questionnaire (NPOQ)[55]. Although participants with syndromal bipolar disorder (BD) are excluded from the study, subthreshold bipolar symptoms are characterised in an index which includes the Bipolar Spectrum Diagnostic Scale BSDS[56] and the Young Mania Rating Scale YMRS[57]. Full details of the index and outcomes will be published as a separate sub-study.

Peripheral markers of inflammation and oxidative stress will be explored in consenting participants (blood sampling is optional). Serum, plasma and whole blood are collected from 30ml of blood, taken at baseline and week 12. Samples are stored at -80 degrees Celsius until analysis.

Participants may also optionally consent to baseline brain imaging. These data will be analysed to determine whether brain imaging variables predicts treatment outcome. As this is a sub-study of the main trial, details of this protocol will be published elsewhere.

Safety and adverse events

The trial is monitored by an independent Data and Safety Monitoring Committee (DSMC). Adverse events (AEs) are collected from the time that informed consent has been obtained until the end of the week 12 intervention period using open questions. After the 12-week intervention period AEs are followed up until the AE is resolved or until 7 days after trial medication has ceased. All Serious Adverse Events are reported to all relevant regulatory authorities.

Suicidal thinking is assessed with the Suicidal Ideation Questionnaire (SIQ)[58] and suicidality is assessed with the Columbia Suicide Severity Rating Scale C-SSRS.[59] If a participant scores 5 on intensity of suicidal ideation in the past month, the participant's continuation in the study is reviewed. If a participant scores 20 or above on the AUDIT at any trial visit, their treating physician is informed and the participant is reviewed.

At baseline and week 12, specific blood tests are performed for safety purposes. At baseline, these comprise: a full blood examination, liver function tests (LFT), urea and electrolytes (U&E), C-Reactive protein random glucose, vitamin B12 and folate, creatinine kinase (CK), and thyroid stimulating hormone. At week 12, the LFT, U&E and CK are repeated.

Procedure

Written informed consent is obtained by the investigator or research assistant. If a participant is <18 years, consent is obtained from both the parent/legal guardian and the participant. If eligibility is confirmed the baseline assessment is conducted (see Figure 1 for participant flow chart). The participant is randomised to the rosuvastatin, aspirin, or placebo groups. Investigators, clinicians, research assistants, and statisticians are blind to treatment allocation for data collection and analysis.

Following baseline assessment and randomisation, participants are subsequently assessed at weeks 4, 8 and 12 on psychopathology, functioning, adverse events and side effects. Instruments and procedures implemented at each study visit are outlined in Table 1. A follow up assessment is also conducted over the telephone at week 26. For engagement and safety purposes, the participant is also telephoned at week 2. Formal inter-rater reliability assessments are carried out at various stages throughout the study.

Participants are reviewed by a treating doctor at baseline, and are seen again one week after commencing medication. The doctor then reviews the participant at weeks 4, 8, and 12.

Randomisation

Participants are randomised to the three groups in a 1:1:1 ratio. Randomisation is enabled by computer-generated numbers programmed into the electronic case report form (eCRF) and conducted according to the International Conference on Harmonisation (ICH) Guideline. The randomisation is stratified by

gender and age (< 18 vs. ≥ 18 years) and participants are allocated to treatment groups using randomly permuted blocks of varying size within each stratum.

Online unblinding is available for emergency situations. If unblinded, participants discontinue treatment in the study but continue to be assessed at the scheduled times, provided they have not withdrawn consent.

Statistical analyses

Primary analyses will be undertaken on an intent-to-treat basis, including all participants as randomised, regardless of treatment actually received or withdrawal from the study. Mixed-model repeated measures (MMRM) analyses will be used to analyse change in the primary outcome measure (MADRS).[60] *A priori* planned comparisons of change from baseline to the week 12 endpoint will be used to test the primary hypotheses. The most suitable, parsimonious variance-covariance structure for the relationships between observations at different occasions will be determined using information measures. Stratification variables and other variables found to be substantially imbalanced between groups post randomisation will be tentatively included in these models and retain if statistically significant and influential on outcomes. Similar analyses of scaled secondary measures will assess differential change on clinical global status, symptom burden, quality of life and functioning. Mathematical transformation or categorisation of raw scores may be undertaken to meet distributional assumptions and include outliers. For dichotomous outcomes such as remission, a comparable mixed modelling approach will be used. Relative and reduction in risk of remission based on MADRS score

status will be estimated at the trial endpoint and follow-up. Number needed to treat[61] will be derived from these values.

In addition to ITT estimates of the effectiveness of each treatment, alternative estimates of effects will be calculated using subgroups identified as having complied with treatment schedules to an extent judged necessary to induce clinical change. Complier, per protocol and complier average causal effect[62] estimates may be examined.

The differential effectiveness of aspirin and rosuvastatin will be examined on outcome variables. As differences between the two active treatments are likely to be small, it is acknowledged that these analyses are exploratory. Safety data will be compared between treatment arms using Fisher's exact test. All tests of treatment effects will be conducted using a two-sided alpha level of 0.05 and 95% confidence intervals. Changes in biological markers will be explored between baseline and endpoint and the role of oxidative stress and inflammatory markers as potential mediators of depressive symptom outcome will be investigated using structural equation models.[63]

Determination of sample size

Based on previous work[42] we estimate baseline-endpoint effect sizes for the aspirin treatment to be between 0.26 and 0.47 standard deviations. Larger effect sizes for statins have been observed in epidemiological studies, with data indicating a 79% reduction in risk of developing depression.[64] An effect size of this magnitude is unlikely to be observed in a treatment study. Power calculations for the primary

outcome were conservatively based on those smaller estimated effect sizes for aspirin. To sufficiently power the study (80% power) to detect differences in change from baseline of approximately 0.4 standard deviations in *a priori* contrasts of treatment arms conducted within the framework of omnibus test of condition-by-time mixed model repeated measures (MMRM) analysis, and assuming a correlation of 0.5 between baseline and endpoint measurements, a total sample size of 200 is required. A sample of 270 is therefore proposed, allowing attrition rates up to 25%.

Data management

Data collection and entry is conducted according to GCP guidelines.[65,66] Monitoring of the data is conducted by the DSMC, the Project Manager and the Sponsor-appointed Clinical Research Associate.

Discussion

Despite the high burden of youth depression, there is limited evidence supporting current pharmacological and psychosocial treatments in this area, leading to a call for new treatments to be developed. The current trial aims to investigate the effectiveness of add-on doses of aspirin or rosuvastatin in the treatment of youth depression.

If the study shows effectiveness in either of the two treatment arms, a novel approach to treating youth depression will be supported. Additionally, should the study demonstrate positive findings, targeted treatments might be further developed by linking the positive results seen in clinical symptoms with biological markers. This study will additionally provide information regarding the biological

underpinnings of youth depression. As samples are taken at baseline and at the end of the treatment phase, this allows us to explore the pathophysiology of depression in this cohort.

Apart from the effect of individual treatment arms, participant characteristics might moderate or mediate treatment response. Smoking, for example, has a differential effect on anti-oxidant systems such as paraoxonases among persons with bipolar and unipolar affective disorders[67], indicating a variable impact of anti-inflammatory agents. Personality factors have been associated with both poor outcome among depressed persons.[68] Diagnostic conversion from MDD to BD can also be examined within this study, in the 6-month prospective follow up of participants included in the trial. Given the association between lower age of onset of MDD and higher risk of conversion to BD[69-71] the rate of conversion to BD may be moderate to high.

This study will explore the effectiveness of aspirin and rosuvastatin as novel therapies and provide meaningful data related to youth depression and the trajectory of illness in this cohort.

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Table 1: Instruments and procedures that are implemented at each study visit.

VISIT	0 Screening Phase	1 Baseline	2 Phone call	3 Interim assessment	4 Interim assessment	5 Primary Endpoint	6 Follow-up phone interview
WEEK	**-3 (weeks)**	**0**	**2**	**4**	**8**	**12**	**26**
SCREENING AND CONSENT							
Informed consent	X						
Inclusion/exclusion criteria	X						
Medical history	X						
Clinical blood test	X					X	
Pregnancy test (females only) †	X						
SCID-I/P	X						X ‡
YMRS	X					X	
Drug dispensation		X		X	X		
SAFETY							
Adverse events recording sheet			X	X	X	X	X
SIQ		X		X	X	X	
AUDIT		X		X	X	X	
EFFECTIVENESS							
MADRS		X		X	X	X	X
QIDS-SR		X		X	X	X	
GAD-7		X		X	X	X	
CGI-S		X		X	X	X	
CGI-I				X	X	X	
PGI						X	
FUNCTIONING AND QUALITY OF LIFE							
Q-LES-Q-SF		X				X	
SOFAS		X				X	X

VISIT	0 Screening Phase	1 Baseline	2 Phone call	3 Interim assessment	4 Interim assessment	5 Primary Endpoint	6 Follow-up phone interview
WEEK	**-3 (weeks)**	**0**	**2**	**4**	**8**	**12**	**26**
SAS-SR - 64		X				X	
ASSESSMENT OF PREDICTORS, MODERATORS AND MEDIATORS							
DAPP-BQ				X			
NPOQ		X				X	
BSDS				X			
Bipolarity Index				X			
- Affective Disorders Evaluation				X			
- Family History Questionnaire					X		
- Antidepressant associated mood changes						X	
- Subthreshold mania questions		X					
- **OTHER**							
Demographics		X					
Psychiatric and family history		X			X		
Concomitant medication		X		X	X	X	X
Research blood sample - Biomarkers		X				X	

† If participant is not sexually active or the treating clinician feels it is not warranted, this is not compulsory.

‡ Only the mood modules from the SCID are conducted at week 26 as part of the bipolarity index.

Table Legend:

SCID I/P (Structured Clinical Interview for DSM-IV Axis I Disorders); YMRS (Young Mania Rating Scale); SIQ (Suicidal Ideation Questionnaire); AUDIT (Alcohol Use Disorders Identification Test); MADRS (Montgomery Åsberg Depression Rating Scale); QIDS-SR (Quick Inventory of Depressive Symptamology); GAD-7 (Generalized Anxiety Disorder 7-item scale); CGI-S (Clinical Global Impression-Severity scale); CGI-I (Clinical Global Impression-Improvement scale); PGI (Patient Global Impression Improvement); Q-LES-Q-SF (Quality of life Enjoyment and Satisfaction Questionnaire – Short Form); SOFAS (Social and Occupational Functioning Scale), SAS-SR-64 (Social Adjustment Scale – Self Report); DAPP-BQ (Dimensional Assessment of Personality Pathology - Basic Questionnaire); NPOQ (Negative Problem Orientation Questionnaire); BSDS (Bipolar Spectrum Diagnostic Scale).

Figure 1 – Flowchart of study design.