PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Evolution of pain at 3 months by oral resveratrol in knee osteoarthritis (ARTHROL): protocol for a multicentre randomised double-blind placebo-controlled trial
AUTHORS	Nguyen, Christelle ; Boutron, Isabelle; Baron, Gabriel; Coudeyre, Emmanuel; Bernbaum, Francis; Poiraudeau, Serge; Rannou, F

VERSION 1 - REVIEW

REVIEWER	Sarah Kingsbury Leeds Institute of Rheumatic and Musculoskeletal Medicine University of Leeds Leeds UK
REVIEW RETURNED	05-Jun-2017

GENERAL COMMENTS	This manuscript presents the protocol for a randomised clinical trial which aims to examine the efficacy of oral resveratrol for reducing pain in patients with knee osteoarthritis. Overall the manuscript is well written and clearly presents the study methods. The trial is robustly designed to address the research question. I have a few comments relating to trial design for consideration of the authors: Primary outcome: Recent data suggests that WOMAC is a more responsive measure for assessing treatment response than the 11-point numerical rating scale (NRS). Eligibility criteria: 1) Although recent IA corticosteroid/hyaluronic injections are listed as an exclusion criteria, there is no mention of IM/IV/oral steroid use. Given the primary pain outcome, exclusion of patients with recent use of any steroid should be considered due to potential impact on pain scores. A rescreen for patients excluded for temporary reasons such as this could be added into the protocol. 2) Further to point 1, the authors may wish to consider excluding patients with uncontrolled disease states, such as severe asthma, where flares are commonly treated with oral or parenteral corticosteroids, since use of steroids close to outcome measurement could impact on pain scores. Statistics: Sample size: Sample size is based on an 11-point NRS, however measurements for calculation of sample size are defined in mm suggesting these are based on a visual analogue scale (VAS). Whilst the VAS and NRS are similar constructs, they are not inter-changeable and this should be corrected to reflect use of either an NRS or a VAS.

	Analysis: Will covariates included in analysis models be pre-defined? How will treatment compliance, use of rescue medication (particularly use of steroids/NSAIDs close to outcome measurement) and missing data be handled?

REVIEWER	Chandra K. Singh University of Wisconsin, Madison, WI 53706, USA
REVIEW RETURNED	12-Jun-2017

GENERAL COMMENTS	In this manuscript, Nguyen and colleagues have proposed to evaluate the effects of oral resveratrol on pain in knee osteoarthritis in a randomized double-blind placebo-controlled human clinical trial. The outcome of this study may be interesting and may pave the way for the development of resveratrol as a safe alternative strategy for treating painful knee osteoarthritis. Following concerns need attention about this proposed study: 1. Please re-check the 'Experimental group' section (line 138-140) about the resveratrol dose. Is it (two capsules of resveratrol (40 mg) will be administered orally twice a day) 160 mg resveratrol for week 1? This appears to be contradictory to whatever has been mentioned in Abstract section (line 50-51). 2. The basis of selection of resveratrol dose and duration are not justified well. Why higher dose of resveratrol for week 1 followed by lower dose up to 6-month? Please detail resveratrol dose and duration with proper explanation and appropriate references. 3. The study will evaluate the effects of resveratrol at 3 and 6 months. However, this proposed clinical trial has been titled as "Evolution of pain at 3 months by oral resveratrol in knee osteoarthritis ……". Authors are suggested to use appropriate title for the proposed protocol.

VERSION 1 – AUTHOR RESPONSE

Answers to reviewer 1

This manuscript presents the protocol for a randomised clinical trial which aims to examine the efficacy of oral resveratrol for reducing pain in patients with knee osteoarthritis. Overall the manuscript is well written and clearly presents the study methods. The trial is robustly designed to address the research question. I have a few comments relating to trial design for consideration of the authors:

1. Primary outcome: Recent data suggests that WOMAC is a more responsive measure for assessing treatment response than the 11-point numerical rating scale (NRS).
Our primary and secondary efficacy outcomes have been prespecified in accordance with OMERACT recommendations and COMET initiative for phase III clinical trials of knee OA. As recommended, measurements for pain, physical function and patient global assessment have been planned in our protocol. Based on the current body of knowledge on resveratrol efficacy in joint disorders obtained from in vitro and preclinical studies, we hypothesized that our pharmacological intervention at 3 months would improve knee pain, rather than physical function. Therefore, we decided not to use a multiconceptual questionnaire as the WOMAC, but rather prespecified the primary outcome as being

the change from baseline in mean knee pain assessed on an 11-point pain NRS. In a study by Ornetti and colleagues (Ann Rheum Dis 2011), patient-reported NRS demonstrated good psychometric properties, similar those of the WOMAC subscales in knee and hip osteoarthritis. In addition, according to 2015 OARSI recommendations for the design, conduct and reporting of clinical trials in knee osteoarthritis, it seems as appropriate to assess pain using an NRS as the WOMAC pain subscale as « Pain can be recorded on a 5-point Likert scale (e.g., none, mild, moderate, severe, very severe), 11-point (0-10) NRS or on a 100-mm VAS. Single questions about pain can be used […]. Pain measurements as part of instruments with multi-item (e.g.,HAQ), multiconcept (e.g., WOMAC pain subscale) or multidimensional measures can be used as well. » Corresponding reference has been added to the manuscript (Reference 28, line 176).
Finally, the present definition of the primary outcome has been validated by the methodologist (Prof Isabelle Boutron), the principal investigator (Prof François Rannou), the study director (Dr Christelle Nguyen), the funder (French Ministry of Health) and the sponsor (Département de la Recherche Clinique et du Développement of the Assistance Publique-Hôpitaux de Paris) of the trial, as well as by the French authorities (Agence Nationale de Sécurité du Médicament et des produits de santé [French Health Products Safety Agency]) and our institutional review board (Comité de Protection des Personnes Île-de-France III), and has been used by the biostatistician (Dr Gabriel Baron) to calculate the sample size.
2. Eligibility criteria: 1) Although recent IA corticosteroid/hyaluronic injections are listed as an exclusion criteria, there is no mention of IM/IV/oral steroid use. Given the primary pain outcome, exclusion of patients with recent use of any steroid should be considered due to potential impact on pain scores.
We agree and have modified the non-inclusion criteria accordingly (Appendix 1).

3. A rescreen for patients excluded for temporary reasons such as this could be added into the protocol.
We agree and have modified the protocol accordingly (lines 138-139).

4. Further to point 1, the authors may wish to consider excluding patients with uncontrolled disease states, such as severe asthma, where flares are commonly treated with oral or parenteral corticosteroids, since use of steroids close to outcome measurement could impact on pain scores.
We agree and have modified the non-inclusion criteria accordingly (Appendix 1).

5. Sample size: Sample size is based on an 11-point NRS, however measurements for calculation of sample size are defined in mm suggesting these are based on a visual analogue scale (VAS).

Whilst the VAS and NRS are similar constructs, they are not inter-changeable and this should be corrected to reflect use of either an NRS or a VAS.
This is a mistake. Measurements for calculation of sample size should have been defined in points. Corrections have been made accordingly (lines 222-223).

6. Will covariates included in analysis models be predefined? How will treatment compliance, use of rescue medication (particularly use of steroids/NSAIDs close to outcome measurement) and missing data be handled?
Constrained longitudinal model will be predefined. In this model, both the baseline and post-baseline values will be modelled as dependent variables (the constrained longitudinal data analysis model assumes that both the baseline and post-baseline measurements are jointly multivariate normally distributed because the baseline value is treated as part of the response vector). The true baseline means will be constrained to be the same for the 2 treatment groups. This analysis provides an adjustment for the observed baseline difference in estimating the treatment effects. Random effects at patient and centre levels will be added to these models. The constrained longitudinal data analysis model can include all randomized subjects with a baseline or post-baseline value. Such methods based on maximum likelihood are consistent under the missing-at-random assumption. Further

description has been added to the « statistical aspects » section (lines 233-242).

Treatment compliance per group will be described. Although use of steroids and NSAIDs are close to primary outcome, no adjustment is planned because these variables will be collected after randomization. Moreover, steroids and NSAIDs will be assessed as secondary outcomes.

Answers to reviewer 2

In this manuscript, Nguyen and colleagues have proposed to evaluate the effects of oral resveratrol on pain in knee osteoarthritis in a randomized double-blind placebo-controlled human clinical trial. The outcome of this study may be interesting and may pave the way for the development of resveratrol as a safe alternative strategy for treating painful knee osteoarthritis.

Following concerns need attention about this proposed study:

7. Please re-check the 'Experimental group' section (line 138-140) about the resveratrol dose. Is it (two capsules of resveratrol (40 mg) will be administered orally twice a day) 160 mg resveratrol for week 1? This appears to be contradictory to whatever has been mentioned in Abstract section (line 50-51).
Description of the intervention is consistent between the 2 sections. However, the wording was different between the 2 sections, which may have added some confusion in the doses of resveratrol administered. We have rephrased the 2 sections in order to make them less confusing as follows : « 40 mg (2 caplets) twice a day for 1 week, then 20 mg (1 caplet) twice a day » (Abstract section, lines 52-53) and « 40 mg (2 caplets) of resveratrol will be administered orally twice a day, 30 min before a meal with a glass of water, for 1 week, then 20 mg (1 capsule) twice a day for a total of 6 months (« Experimental group » section, lines 140-141).

8. The basis of selection of resveratrol dose and duration are not justified well. Why higher dose of resveratrol for week 1 followed by lower dose up to 6-month? Please detail resveratrol dose and duration with proper explanation and appropriate references.
Pharmacokinetics, bioavailability and toxicity of trans-resveratrol formulation used in the ARTHROL trial have been previously described in a phase I clinical trial (Reference 24). Briefly, 15 healthy volunteers received a single dose of 40 mg of oral trans-resveratrol in 2 forms (soluble galenic formulation or dry powder). Blood samples were collected at 15 min, 30 min and every hour for 5 h.

Plasma concentrations of trans-resveratrol and its metabolites were analyzed by liquid chromatography and mass spectrometry. The single dose of the soluble trans-resveratrol was well absorbed and elicited biologically efficient blood levels (0.1-6 µM) for several hours. In contrast, trans-resveratrol administered as dry powder barely elicited efficient blood levels for a short duration. The soluble formulation led to 8.8-fold higher trans-resveratrol levels in plasma versus the powder. Trans-resveratrol metabolism was not modified and neither intolerance nor toxicity were observed during the study and the following week.

We have made substantial modifications to the administration scheme as compared to the one tested in the phase I clinical trial:
- because trans-resveratrol is metabolized into glucuronide and sulfate conjugates coupled to renal elimination, we hypothesized that giving a loading dose for 1 week may allow attaining the drug effect more rapidly,
- for the maintenance dose, we chose 40 mg a day as tested in the phase I clinical trial, but in 2 doses, because the half-life of the soluble galenic formulation of trans-resveratrol is 79 min only.
Resveratrol dose and duration have been now detailed with proper explanation and appropriate reference (lines 142-154).

9. The study will evaluate the effects of resveratrol at 3 and 6 months. However, this proposed clinical trial has been titled as “Evolution of pain at 3 months by oral resveratrol in knee osteoarthritis …….”. Authors are suggested to use appropriate title for the proposed protocol.
The effects of resveratrol will be indeed assessed at 3 and 6 months. However, as the prespecified primary outcome (mean change from baseline in mean knee pain) will be assessed at 3 months, we feel that adding the 6-month timepoint in the title may be a bit confusing and that the current title more faithfully reflects the primary objective of the study.

VERSION 2 – REVIEW

REVIEWER	Sarah Kingsbury Leeds Institute of Rheumatic and Musculoskeletal Medicine University of Leeds UK
REVIEW RETURNED	21-Jul-2017

GENERAL COMMENTS	The authors have satisfactorily addressed previous concerns. I am therefore now supportive of this manuscript being accepted for publication.

REVIEWER	CHANDRA SINGH University of Wisconsin-Madison, USA
REVIEW RETURNED	23-Jul-2017

GENERAL COMMENTS	The manuscript has improved since I last reviewed it. The authors have addressed reviewers' concerns sufficiently, which makes this manuscript suitable for publication in its present form.