Supplementary Online Content

Bauchner H, Fontanarosa PB, Golub RM. Evaluation of the Trial to Assess Chelation Therapy: the scientific process, peer review, and editorial scrutiny. *JAMA*. doi:10.1001/jama.2013.2761

eAppendix. Editors' Comments and Author Responses

This supplementary material has been provided by the authors to give readers additional information about their work.

eAppendix. Editors' Comments and Author Responses

Editors' comments:

While we are interested in considering a revised version of your paper, we are wondering whether the reported findings are correctly interpreted, and whether, in fact, the overall findings of the study actually are null. We are concerned about: the possibility that the final alpha level might not be sufficiently stringent (given the multiple interim analyses that were conducted, it seems that the significance level needs further adjustment); the marginally significant p value that was found, and the corresponding upper bound of the CI for the hazard ratio (of 0.99); the detected effect size being less than that on which the study was powered for the main result, and the composite outcome being driven by revascularization rather than other endpoints; the change in anticipated sample size; and questions about the method by which missing data were handled (and how the findings might appear if these data were handled with other accepted methods, eg, multiple imputation, etc).

We understand and appreciate your concern about the interpretation of the overall findings in the trial, particularly in view of the fact that the final p-value falls so close to the required level of significance, and the upper level of the confidence interval for the hazard ratio (0.99) is so close to 1.0. In the revised manuscript, we have attempted to be appropriately circumspect in the interpretation of the results.

We believe that the question you pose is of sufficient importance to the integrity of our results to include the response in the proposed online appendix, and we refer you to page 10 of the online appendix "Interim Monitoring in TACT and the Final Criterion for Significance". As you will see from the explanation, statistical equation, and table, the justification for the final pre-specified level of significance is sound. We hope you will agree with us.

The change in sample size, likewise, was handled in a scientifically rigorous way, with the change proposed by clinical study leadership who had absolutely no knowledge of any treatment-specific outcomes in the trial. It is well known that in long-term follow-up trials such as TACT, the desired level of statistical power depends largely on the number of events that occur rather on the number of patients enrolled. The desired level of power can be achieved by enrolling a specific number of patients and following them for certain period of time, or by enrolling fewer patients but following them for a longer period of time, as long as the desired number of events can be accrued. We have also considered this question sufficiently important to include in the online appendix starting on page 8 "Sample Size Calculations for TACT and Changes in Sample Size". This section provides the practical and scientific bases for the changes in sample size that occurred throughout the course of the trial.

We have also included sections in the proposed online appendix that address your questions regarding missing data, and sensitivity analyses using a variety of different assumptions regarding the outcomes of patients who withdrew consent or were lost-to-follow-up: “Consent Withdrawals and Lost to Follow-up” starting on page 12 of the online appendix, and “Sensitivity Analyses with Imputations of Outcomes”, starting on page 19 of the online appendix.

In these sections, we specifically emphasize two points pertaining to **patients who withdrew consent or were lost to follow-up**. First, there was a sizable number of these patients (52 (18%) of the 289 consent withdrawals) who experienced a documented primary event. Forty-three of the 52 had a primary event prior to consent withdrawal, and 9 patients were found to have died after their consent withdrawal. Those 52 events are all counted and included in the primary analysis. Second, there were significantly more patients in the placebo arm (174) who withdrew consent compared to the active chelation arm (115). Thus, among the patients where we may be missing primary outcome events because of consent withdrawals, there are likely more such events in the placebo arm than in the active treatment arm (because there were more withdrawals in the placebo arm). Even in scenarios unfavorable to EDTA chelation, accounting for these patients does not alter the study conclusions. In many scenarios we examined, accounting for these patients augments the precision of the estimated benefit of chelation therapy.

In order to assess the risk profile of **patients with incomplete follow-up**, we performed extensive analyses comparing baseline characteristics of those patients who withdrew consent or were lost-to-followup with the rest of the patients in the trial. We also compared the baseline characteristics of those **patients with incomplete data** by treatment group assignment to determine whether there were clear differences in risk profile across groups – differences that might have changed the overall interpretation of the trial.

As seen on Table A-2 in the online appendix, **patients who withdrew consent** were more likely to be female, diabetics, and have had anterior MI’s. And, as noted in the manuscript section on Subgroup effects, diabetes and anterior MI were both associated with a larger treatment effect from chelation (Figures 2 and 3 in the manuscript).

The baseline characteristics of patients in the chelation arm who withdrew consent, however, are very comparable to the characteristics of the placebo patients who withdrew consent. That is, the risk profiles of the EDTA patients and the placebo patients who withdrew consent are remarkably similar. This observation becomes relevant as we impute the possible outcomes of patients who withdrew consent. We propose to place the entire methodology of the sensitivity analyses in the online appendix that will be available to readers.

Questions regarding the OHRP Inquiry and some study sites-

We have re-ordered the editorial questions to provide a more coherent narrative in the answers. We welcome any requests for more details, as we have tried to be succinct. TACT has been a heavily and carefully monitored NIH trial, organized as a cooperative agreement, in which NHLBI and NCCAM Program Officers were a full part of the investigative team. By virtue of the controversy attached to chelation therapy, the study has attracted attention from multiple quarters.

Please explain in detail, the interruption of the study for an investigation by the OHRP; including actions taken and how interruption affected patients receiving infusions.

In 2008, a self-appointed group comprised of several individuals with a history of aggressive opposition to CAM practices, who had previously voiced strong opposition to chelation therapy and to TACT, wrote to the Office for Human Research Protections with complaints about:

1. the scientific justification for TACT,
2. the TACT consent form,
3. the TACT investigators, and
4. whether patients were properly informed of new developments.

OHRP responded with a letter to the Directors of Research at Mount Sinai Medical Center, the University of Miami, and Duke University on August 25, 2008.

In light of this letter, on August 29, 2008 the TACT study leadership decided to voluntarily suspend all study activities until the OHRP allegations were thoroughly discussed with the TACT Operations Committee, the study sponsors (NHLBI and NCCAM), the TACT DSMB Chair, the main IRBs and the institutions addressed in the OHRP letter.

Following these discussions, on September 7, 2008 a decision was made to resume infusions and all other study activities for currently randomized patients, and voluntarily place a hold on accrual of new patients. Therefore, there was only a 1 week interruption for patients receiving infusions and follow-up. A letter to patients explaining this action was prepared, provided to sites for IRB approval, and then distributed by sites to patients.

The voluntary suspension of new enrollment remained in effect between August 29, 2008 and December 16, 2008. It was lifted after a response to OHRP was sent in December 2008, and changes to the TACT consent form were approved by the University of Miami IRB. At that time, enrollment of new patients resumed.

Given the misinformation disseminated by the anti-chelation groups on the internet and in the media, it is important to emphasize that at no time did NIH or OHRP suspend the study. All the actions taken, including suspension of study activities for a week, hold on enrollment of new patients for 4.5 months, and ultimately full continuation of study activities- were taken by the investigative team in full collaboration with NHLBI, NCCAM, the DSMB, and the IRBs. OHRP, of course was apprised of our decisions.

The specific OHRP questions were answered as follows:

1. Scientific justification: Chelation therapy had been in use by the public for many years and there was, at least, a public health imperative to ascertain risks and benefits. Moreover, TACT was a response to an RFA that had been peer-reviewed by NCCAM and NHLBI Councils, and OHRP recognized this. OHRP did not request additional information on this issue.

2. Consent form: OHRP questioned omission of death from the potential risks, lack of specificity in stating that calcium, not disodium EDTA, was FDA approved for treating lead poisoning, and that disodium EDTA is not known to be useful for treating atherosclerosis.

Death had been omitted from the potential side effects because the currently available literature supported that the drug was safe unless used in high doses in patients with renal failure, or with rapid infusions of high doses. These scenarios were outside the scope of the trial, as we excluded patients with renal failure, and required that infusions administered in the study take at least 3 hours. In fact, the toxicity data in the manuscript bear this out. There were 1% of infusions that were at least 15 minutes too fast, and 0.15% of infusions at least 1 hour too fast. There were no side effects reported from these rapid infusions. There were 2 patients whose deaths, based on independent review, could possibly have been due to a study infusion. One patient was receiving placebo, the other EDTA.

With regards to the specificity of calcium versus disodium EDTA, we had felt that an overemphasis on the specific salt of EDTA used by the trial would be confusing to patients, so the test article was called “EDTA” in the consent form. The original consent form did not imply that EDTA was a known treatment for atherosclerosis.

The TACT consent evaluated by OHRP had been previously reviewed and approved by:

1) NCCAM and NHLBI
2) A protocol review committee appointed by NHLBI and NCCAM, and investigators from Mount Sinai and the DCRI.
3) The DSMB, which had as a member a respected bioethicist.
4) The Mount Sinai IRB annually
5) Multiple academic IRBs including Hopkins, Mayo, Scripps, and others, annually.
6) A central IRB annually.

Nevertheless, we revised the consent form along the lines desired by OHRP, reconsented patients undergoing infusions, and delivered a Dear Patient letter informing study participants of these changes.

3. TACT investigators: Legal issues with a small group of TACT investigators were brought up by OHRP. All investigators had an unrestricted license to practice medicine in their State, and received:

1) Human subjects training
2) Research training
3) Protocol training in person at study meetings
4) Protocol training on-line
5) IRB approval to enroll and follow patients in the study
6) In person monitoring by DCRI and offsite through the EDC system.

Following approval, all sites had an in-person monitoring visit after 2 patients had been enrolled and annually if additional patients were enrolled. They also underwent off-site electronic monitoring through the EDC system. These training and monitoring plans were reviewed with NHLBI, NCCAM, and the DSMB.

OHRP accepted our investigative sites and enhanced monitoring detailed below.

4. New developments: On June 12, 2008, FDA asked 3 US manufacturers to withdraw their application to manufacture disodium EDTA. They pointed out that there was no indication for the use of this compound, since calcium EDTA was the approved compound for lead poisoning. Patients had not been informed of the FDA action, since disodium EDTA was being used under an FDA IND for an experimental reason- the treatment of atherosclerosis, and the use of disodium EDTA in the trial was not addressed by FDA. The disodium EDTA used by TACT was obtained since the beginning of the trial from Akzo Nobel, a Dutch manufacturer.

Based on the OHRP recommendation, we included this information in the above mentioned Dear Patient letter, and committed to inform patients of any new developments with EDTA.

On October 30, 2009 we received the final letter from OHRP, regarding the TACT inquiry stating that there was no need for further involvement by OHRP in this matter. We are happy to provide more detail, including our detailed responses and the Dear Patient letter.

Explain how the data were handled from sites at which several of the TACT study investigators have been accused of substandard practices by state medical board, involvement in insurance fraud, and three convicted felons.

All data collected from all sites have been entered into the database, analyzed, and form part of the manuscript.

Explain whether those investigators and those sites were able to continue in the study, the amount of data collected from these sites, and whether data from those sites are included in the analyses?

All investigators were licensed to practice medicine in their state, and there was no investigator who was barred from receiving federal funds for conducting research. Thus, these sites continued participation in TACT. Following our receipt of the initial OHRP letter, all IRBs and the DSMB were made aware of any investigators that had, in the past, experienced legal problems.

A past legal problem does not constitute a bar to research participation. In their May 27, 2009 letter, OHRP stated: “while concerning, these things do not automatically preclude an investigator from participating in research and do not automatically indicate a failure of risks to subjects to be appropriately minimized. The details and circumstances surrounding these incidents must be considered by the IRB when ascertaining the ‘acceptability of proposed research in terms of institutional commitments and regulations, applicable law, and standards of professional conduct and practice’ as required by HHS regulations at 45 CFR 46.107(b). Based on the information available to us, we determine that, while true, the alleged facts in themselves do not give rise to a violation of 45 CFR 46.111”.

There were 167 patients enrolled at sites in which the PIs had previously had a variety of legal issues. Of these patients, 106 (63%) completed all 40 infusions, similar to the proportion of patients completing all infusions in the overall study (65%). Moreover, one of the subgroup analyses reported in the manuscript is the interaction of study therapy with site type (chelation site vs conventional cardiology site). There is no evidence of a differential effect of therapy based on site type (Figure 2). All data collected from all sites have been entered into the database and analyzed.

Explain the oversight measures that were instituted to ensure data veracity and study propriety.

As a part of the corrective action plan proposed to OHRP, the TACT management team made the decision to institute oversight at a far greater level than what is common in cardiovascular clinical trials. Accordingly, the Clinical Coordinating Center (CCC) instituted biannual State Board license and OIG checks for all investigators. The forms filled out by prospective new investigators were upgraded to capture self-reported Board or other legal issues. We committed to bring any disclosed or undisclosed legal matter to the TACT Operations Committee for review and imposition of corrective actions when necessary. Examples of such corrective actions could include enhanced monitoring, changing the site PI, or closing the site.

How many study sites were closed due to "quality problems" and how many patients had been enrolled at those sites? Some of this appears to be hinted at in the paper on lines 349-350, but these issues need to be discussed in more detail.

During study meetings and online study training, active and prospective sites were told that protocol violations and safety violations would be dealt with swiftly, potentially with site suspension and site closure. There were only 2 sites that were suspended due to safety issues or protocol violations during the course of the trial. One was temporarily suspended early in the trial due to the delivery of over 10 infusions that were under 3 hours in duration. Our investigation showed that this was due to a new coordinator, who was reassigned. Additional training was given, and the site was reopened. Fast infusions did not recur. Another site was suspended from enrolling new patients due to a protocol violation that involved drawing blood studies outside the study protocol that might have unblinded the investigator and patients. The corrective action plan involved permanently suspending new enrollment, protocol training, and quarterly site visits from the Data Coordinating Center (DCC) and CCC with onsite verification of source data.

Specific comments:

Title: please revise, perhaps as: Effect of disodium EDTA chelation on cardiovascular events in patients with previous myocardial infarction: The TACT Randomized Trial

The title has been revised. We prefer the term “EDTA Chelation Regimen” because it implies that the treatment does not involve EDTA only.

Abstract, line 35 [and methods, line 201]: are additional follow-up data available or being collected ? If so, these should be added to the paper.

There are no additional data being collected. All data are in the database and analyzed.

Abstract: Combine Design and Setting paragraphs

Done.

Abstract, Main outcome measure: Please indicate the time point for end of follow up (presumably 5 years). Please add the significance threshold.

Done.

Abstract, Results, lines 58: Please add the actual results.

Done.

Abstract, Results: Please avoid use of the word "minority". If important to the study, include specific terms (white, black, Hispanic). "Nonwhite" is preferred to "minority" (although not ideal)

Changed to “non-white”.

Abstract, Conclusion: Please delete and replace with a single sentence objectively summarizing the key study finding, something like "Among patients with a history of MI, the use of intravenous chelation with disodium EDTA compared with placebo resulted in a lower risk of composite cardiovascular endpoint over 5 years." This would need to be modified if, on reanalysis, the findings are no longer statistically significant.

Done.

Methods, line 132: Although the inclusion criterion was being at least 6 weeks post-MI, median enrollment was 4.6 years post-MI. This seems problematic since (1) there is a lot of variation in duration from MI, and (2) there is no accounting for the differential outcomes that for some patients may have occurred after enrollment, and for others may have occurred between the index MI and enrollment. In some ways, this is parallel to the problem of immortal time bias in observational studies. It would seem that the analyses would need to account for this factor.

The only outcomes reported for the trial are those that occurred following enrollment (i.e., following randomization). Outcomes that occurred between the index MI and enrollment (i.e., after the index MI but before enrollment) were not considered as outcomes in the trial. With regard to possible heterogeneity in the results of the treatment comparison based on the length of time from the index MI to enrollment, we examined this issue in the subgroup analyses reported in the manuscript. The specific time ranges used for the MI were <2 years, 2 to 5 years, and > 5 years. The p-value for testing whether there was an interaction between treatment and time from index MI to enrollment was 0.87. Thus, there was no evidence of heterogeneity in the effect of chelation therapy regardless of time since the index MI.

Methods, line 195: Did the power analysis/sample size calculation allow for the additional sample size need for factorial analysis?

In performing the power analysis for each factor, we did consider the fact that the other factor, if effective, could reduce the event rate in the comparator arm, thereby impacting the statistical power/ sample size requirements. We did account for this possibility in our projection of the 20% 2.5-year control event rate for each factor.

Methods, line 205: Please provide more detail about the intention to treat analysis, and specifically, how missing data were handled. It appears that patients who dropped out / withdrew consent, etc were included in the denominator and contributed person-time until they were lost to follow-up. However, it is unclear whether any attempt to was made to use other techniques to account for missing data on events, such as multiple imputation approaches. The authors should consider conducting these analysis as sensitivity analyses and report the results.

The intention to treat analysis included all patients in the arm to which they were randomized and used all the follow-up information that was available on each patient. The patients who withdrew consent or were lost to follow-up were included in the analysis with as much follow-up (person-time) as was available until they withdrew consent or were lost to follow-up, at which time the patient became a censored observation. Not all of these patients were censored observations, however. There were 52 of the 289 patients who withdrew consent, and 3 of the 22 patients who were lost to follow-up who experienced one of the primary events, and those events were all included in the analysis. Thus the primary analysis consisted of all the data that were available. We used the outcomes that were known, without any imputation of outcomes in the patients for whom we did not have complete follow-up. We have also performed the imputations suggested by the Editor and the reviewers and they are included in the suggested online appendix material.

Methods, line 213 - 217: Given there were 11 interim analyses plus the additional final analysis, it seems that the alpha level adjustment is insufficient. The author should provide a clear, detailed, and thorough justification for this, including the calculations used to arrive at the 0.036 significance level [as part of the response letter]. For instance, is it possible that not all of these analyses were for efficacy, but only for safety?

We have provided a response in an earlier section, directed to the Editor. Details of the alpha-level adjustment using the well-established alpha spending function approach are also included in the online appendix. We hope this will answer the reviewer's questions and allay the concerns.

Methods, line 219: Please indicate that significance testing was 2-sided.

Significance testing was 2-sided. This is now made clear in the text.

Methods: Did follow-up time begin with the first infusion? It seems that it would require a minimum of about 1 year to complete the intervention protocol. Is that correct?

Follow-up time began upon randomization, not at the time of the initial infusion. All events that occurred after randomization were counted and included in the primary analysis. The infusion protocol, if followed precisely according to protocol, would take a minimum of 30 weeks for the weekly infusions + 10 maintenance infusions 2 weeks apart = 50 weeks, or just under a year. However, the protocol allowed for the 10 maintenance infusions to be 2-8 weeks apart, so the entire 40 infusions could have extended beyond 1 year.

Methods: How much time could elapse between infusions and have the participant still considered to be in treatment?

The trial focused on delivering the total of 40 infusions of the blinded therapy if patients were unable to be fully compliant with the protocol-mandated schedule (due to vacations, for example). Therefore, there was no time limit for the patients to receive the maximum number of infusions.

Methods: An individual could contribute only 1 event to the composite primary and secondary endpoints, correct?

This is correct. Individual patients were only counted once even if they experienced more than one of the events comprising the primary endpoint.

Methods: Mention statistical software used, including version

All analyses were performed using SAS software, version 9.2 (SAS Institute, Cary, North Carolina). This information has been added to the Methods section.

Methods: Please add sentence explaining how and why race/ethnicity information was collected.

As explained in an earlier response to the editors, race/ethnicity information was self-reported by the patients and reported in the electronic case report form by the enrolling site. As a federally funded clinical trial, we were required to collect and report this information.

This was added in the “Study Population” section.

Results: Please report the median (IQR) duration of follow-up in each group; Please report the median (IQR) duration of the intervention phase in each group.

The median duration of follow-up was 55 months (IQR 26,60) overall. Active treatment patients were followed 56 (27, 60) months, while placebo patients were followed 53 (24, 60) months. This is now reported in the manuscript.

The slightly longer follow-up in the chelation arm is due to the fact that there were more consent withdrawals in the placebo arm. We mention again that, as there were more consent withdrawals and hence a shorter follow-up on average in the placebo arm, if events were missed because of consent withdrawals, it is likely that there were more missed in the placebo arm.

Results section and Discussion (Comment) section: Please present comparative absolute rates with all relative differences or percentage changes.

These changes have been made.

Results, beginning line 234: Please report treatment compliance data by study group. If 65% completed all 40 infusions and 30% discontinued study fusions, what accounts for the other ~5%?

This 5% consists of patients who died during the course of their infusion treatment regimen (they were not counted among the discontinuations), and the relatively small number of patients enrolled late in the trial who had not yet completed all the infusions prior to the end of the follow-up phase of the trial. This is now reported in the Results section.

A histogram showing number of study infusions received in each treatment group as a supplemental figure could be informative and relevant to the discussion of treatment compliance.

The histogram is provided on page 24 of the proposed online appendix. We have also included below a more detailed numeric table of infusions received that we believe may be more informative to the reviewer.

Patient Infusion Completion			
	EDTA Chelation (N= 839)	**Placebo (N= 869)**	**All Patients (N=1708)**
Number of patients who have completed:			
All 40 infusions	565	552	1117
35-39 infusions	47	42	89
30-34 infusions	35	54	89
25-29 infusions	20	19	39
20-24 infusions	17	28	45
15-19 infusions	24	34	58
10-14 infusions	29	41	70
5-9 infusions	30	28	58
1-4 infusions	45	37	82
0 infusions	27	34	61
Total no. of infusions completed	27,382	27,840	55,222

Survival plots of time to discontinuation of intervention could be informative.

The plot is included on page 23 of the proposed online appendix.

Results, lines 244-245: Please report the 95% CIs for the Kaplan-Meier 5-year event rates.

We have now reported the confidence interval for each randomized arm in the text.

Comment section, lines 279-281: Please delete.

Done.

Comment section, line 346: please add a more detailed discussion of the study limitations

We have added a more robust “Limitations” section, and deleted the section describing clinical application of this therapy.

Acknowledgments: Add formal Role of the Sponsor statement using JAMA standard language

Done in Acknowledgements as requested.

Table 1: Specify the race/ethnicity categories instead of lumping as "minority." Break down into white, black, Hispanic, and any others. What are continuous data reported as--median (IQR)? Please specify. Please list all data that are No. (%) in descending order of prevalence. Please clarify whether the rates of the individual component endpoints are all rates and not "first occurrence" rates?

We have provided the detail requested for the race/ethnicity categories in Table 1. Using the categories required by the NIH, we have categorized the patients into the ethnicity categories of Hispanic/Latino versus others, and then the racial categories of white, black, Asian, and other.

Figure 1: Please confirm that time 0 is time of randomization. When was the first infusion administered relative to the time of randomization?

Time 0 is the time of randomization. This is now stated in the Methods. The median time (IQR) from randomization to first infusion was 8 days (6, 12). By individual treatment arm, the median (IQR) in the chelation arm was 8 (6, 12), and in the placebo arm, it was 7 (6, 12). This is stated in the Results section.

Is the P value shown from the log-rank test? Please specify.

Yes. This is now stated in the figure.

Figure 2: Are these data for the primary endpoint? Please specify. Please report the sample sizes and number of events in each subgroup by study intervention group. Are the "minority" categories white vs non-white? The time from MI to enrollment categories overlap at 5 years. Please revise.

The data in this figure are for the primary endpoint. The figure has been revised based on your recommendations. We have provided the sample sizes and numbers of events in the subgroups as a supplementary table in the online appendix, pages 28 and 29.

Figure 3: Please report the numbers at risk for each study group at intervals along the horizontal axes in each graph. Note that point estimates become less precise as the sample sizes get smaller; suggest truncating these analyses when the sample sizes are less than 10% of those at time 0, if applicable. Is the start of follow-up time for these graphs the same as in Figure 1? The horizontal axis labels differ.

The figure has been revised based on your recommendations. The start of follow-up is the same for all the figures and making the horizontal axis labels uniform throughout has made this clearer.

Are the P values shown from the log-rank test? Please specify.

Yes. This is now stated in the figure.

Figures 1 and 3: Please save or export these graphs directly out of the software application used to create them in a vector file format such as .wmf or .eps. The curves should be shown as thin, solid lines in different colors. The numbers at risk may be provided in a separate document, if easier.

The figures have been provided in their original format in order to avoid further delay. The re-formatted figures will be provided.

Study Flow Diagram: Please provide the numbers for each of the reasons for the exclusion of 147 screened patients including the 2-3 specific major reasons for study ineligibility.

Unfortunately, once excluded, the database did not store reasons for exclusion. More detail is not available.

Please report the distribution of study infusions received by non-overlapping categories. Please add the numbers for each of the reasons for not receiving the randomized treatment.

This has been done.

Please add the numbers for each of the reasons for discontinuation of the intervention to this diagram.

The CONSORT diagram has been modified to include the reasons for discontinuation.