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Original Research

Mediation analysis of the testosterone treatment effect to prevent type 2 diabetes in the Testosterone for Prevention of Type 2 Diabetes Mellitus trial

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Abstract

Objective: To determine if testosterone treatment effect on glycaemia is mediated through changes in total fat mass, abdominal fat mass, skeletal muscle mass, non-dominant hand-grip, oestradiol (E2), and sex hormone-binding globulin (SHBG).

Design: Mediation analysis of a randomised placebo-controlled trial of testosterone.

Methods: Six Australian tertiary care centres recruited 1007 males, aged 50-74 years, with waist circumference ≥95 cm, serum total testosterone ≤14 nmol/L (immunoassay), and either impaired glucose tolerance or newly diagnosed type 2 diabetes on an oral glucose tolerance test (OGTT). Participants were enrolled in a lifestyle programme and randomised 1:1 to 3 monthly injections of 1000 mg testosterone undecanoate or placebo for 2 years. Complete data were available for 709 participants (70%). Mediation analyses for the primary outcomes of type 2 diabetes at 2 years (OGTT ≥ 11.1 mmol/L and change in 2-h glucose from baseline), incorporating potential mediators: changes in fat mass, % abdominal fat, skeletal muscle mass, non-dominant hand-grip strength, E2, and SHBG, were performed.

Results: For type 2 diabetes at 2 years, the unadjusted OR for treatment was 0.53 (95% CI:.35-.79), which became 0.48 (95% CI:.30-.76) after adjustment for covariates. Including potential mediators attenuated the treatment effect (OR 0.77; 95% CI:.44-1.35; direct effect) with 65% mediated. Only fat mass remained prognostic in the full model (OR: 1.23; 95% CI: 1.09-1.39; $P < .001$).

Conclusion: At least part of the testosterone treatment effect was found to be mediated by changes in fat mass, abdominal fat, skeletal muscle mass, grip strength, SHBG, and E2, but predominantly by changes in fat mass.

Keywords: mediation analysis, testosterone, diabetes, glucose

Significance

- Testosterone treatment prevents or reverses newly diagnosed type 2 diabetes in high-risk men (aged 50+, waist circumference >94 cm) with testosterone (≤14 nmol/L) but without pathological hypogonadism. However, the mechanism of effect is unresolved.
- We utilised a novel, intuitive method of mediation analysis, applied to our randomised placebo-controlled clinical trial, to decompose the testosterone effect into the effect of testosterone alone and the effect due to changes in other "mediators".
- This analysis highlights the dominant effect of a decrease in fat, and to a lesser extent, skeletal muscle, mass, strength, SHBG and E2, to improve glucose tolerance and prevent type 2 diabetes in response to testosterone treatment.

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Introduction

In randomised controlled trials (RCTs), the mechanisms by which outcomes are induced by health interventions can be investigated by mediation analysis.[1] Mediation analyses have emerged as a powerful tool to disentangle potential causal pathways in data from clinical trials. It has been widely applied in the field of psychology[2] but has also been used in other fields to understand how treatments work in RCTs.[3] They enable researchers to decompose the treatment effect into an indirect component mediated through given variable(s) and the remaining direct effect of treatment or effects of unmeasured mediators. Identifying these mediating variables can help clinicians refine and adapt to improve the effectiveness of interventions and guide their implementation.

The Testosterone for Prevention of Type 2 Diabetes Mellitus (T4DM) trial was a randomised, double blind, placebo-controlled trial on the background of a lifestyle programme, in males aged 50 years or more with a waist circumference of 95 cm or over with a serum testosterone (≤14 nmol/L) and impaired glucose tolerance or newly diagnosed type 2 diabetes (T2D). The trial found that administering testosterone undecanoate 1000 mg injections every 3 months for 2 years significantly lowered the chances of a T2D diagnosis. This was measured by using an oral glucose tolerance test (OGTT) at the end of the 2-year period, with a 40% reduction in likelihood compared with placebo.[4]

The effectiveness of this treatment was linked to beneficial changes in body composition. After 2 years, the fat mass in the placebo group decreased by 1.9 kg, whereas in the testosterone-treated group, it decreased by 4.6 kg. In those given the placebo, skeletal muscle mass and non-dominant hand-grip strength saw a decrease of 1.3 and 0.5 kg, respectively. However, in those treated with testosterone, these measures increased by 0.4 and 1.7 kg, respectively.

In middle-aged and older males, a lower fat mass and increased skeletal muscle mass and strength are known to reduce the risk of T2D.[5-7] Despite these findings, it remains unclear whether, and to what extent, testosterone contributes to preventing T2D by inducing changes in fat mass, and/or skeletal muscle mass and strength. The glucose-lowering effect of testosterone may also be mediated by aromatisation to oestradiol (E2), which increases skeletal muscle insulin sensitivity and glucose utilisation via oestrogen receptor alpha-mediated mechanisms,[8] possibly facilitating insulin transport across the vascular endothelium.[9,10]

Here we present the first mediation analysis into the effect of testosterone, to assess in the T4DM trial whether the treatment effect was mediated through changes in body composition, muscle strength, and E2. Serum sex hormone-binding globulin (SHBG) concentration was included in the analyses because of its role as primary carrier of circulating sex steroids.[11,12]

Materials and methods

Source of data

The Testosterone for Diabetes Mellitus (T4DM) trial was a randomised, double blind, placebo-controlled, 2-year, phase 3b trial performed at six tertiary care centres. The trial design paper[13] and full protocol[4] are previously published. The trial recruitment commenced on February, 5, 2013 and completed on February, 27, 2017, with the final follow-up visit on May, 21, 2019. Briefly, participants provided written informed consent and were included if they met all entry criteria: male, age 50-74 years, waist circumference ≥95 cm, impaired glucose tolerance or newly diagnosed T2D, and a fasting serum testosterone drawn between 8 and 10 AM of ≤14 nmol/L by immunoassay at an accredited pathology provider (Sonic Health Care, Australia). Exclusions included hypothalamic–pituitary–testicular pathology, testosterone treatment in the past 12 months, or history of androgen use at any time. All 1007 participants were given access to a lifestyle programme (WW, formerly Weight Watchers) and randomised (1:1) to testosterone undecanoate (1000 mg) or matched placebo, both administered by deep intramuscular injection every 3 months for 2 years. The T4DM study was approved by the ethics committee at each of the participating centres, with the lead Human Research Ethics Committee (HREC) as Sydney Local Health District HREC—CRGH. The other HRECs were Central Adelaide Local Health Network Human Research Ethics Committee, South Metropolitan Health Service Human Research Ethics Committee and Bellberry Human Research Ethics Committee. The study was registered on the Australia and New Zealand Clinical Trials Registry (ACTRN12612000287831).

Sample size

The sample size for T4DM was performed based on the two primary outcomes and is reported elsewhere.[4,13] Briefly, we determined that 1000 participants would have ≥80% power to detect a change in the proportion of participants with OGTT ≥11.1 mmol/L at 2 years and ≥88% power to detect a change between groups in the reduction of OGTT 2-h glucose from baseline to 2 years. The trial was to be deemed positive if either of the primary outcomes was met.

Outcome

This paper analyses the two primary outcomes from the main trial; (1) 2-h glucose ≥ 11.1 mmol/L and (2) the change in 2-h glucose from baseline, both as measured by OGTT after 2 years of treatment. Two years was defined as an OGTT test performed between 21 and 27 months post randomisation (inclusive) and were assessed blinded to treatment allocation.

Potential treatment mediators

Body composition was measured at baseline and 2 years by dual x-ray absorptiometry to provide data on skeletal muscle mass (kilograms), total fat mass (kilograms), and abdominal fat mass (percentage; %). Grip strength in the non-dominant hand was measured by hand-grip dynamometer in kilograms.[13] Serum E2 was measured by a validated liquid chromatography–tandem mass spectrometry method.[14] SHBG was measured using platform chemiluminescent-based immunoassay.[4] It was included as a possible mediator given the relationship between sex steroids and SHBG,[11,12] with a sensitivity analysis performed excluding SHBG.

Baseline covariates

Baseline risk factors (as determined previously[4]) were levels of 2-h glucose as measured by OGTT, centre, age, baseline waist circumference, baseline weight, baseline smoking status, baseline serum testosterone, baseline use of selective serotonin reuptake inhibitors, and first-degree family history of T2D.

Additionally, baseline levels of the treatment mediators were included in the relevant models.[15] For the sensitivity analysis excluding SHBG as a mediator, baseline SHBG was included in the model.

Statistical analyses

All analyses were performed according to the intention-to-treat principle, and, other than a sensitivity analysis, included participants with complete data at 2 years. Assessment of treatment mediation was performed using a counterfactual framework that decomposes the causal effect (treatment effect) into a natural direct effect and natural indirect effect, irrespective of the data distribution or scale of the effect.[16-19] This therefore overcomes the flaws in the earlier methodology used in mediation analyses.[20-22] Given that we have parallel mediation (mediators and outcomes measured at the same time-point) with baseline covariates, fitting a single model (joint mediation model) for each outcome with adjustment for baseline covariates was performed.[15] The causal pathway is given in Figure 1, where the mediators are part of the causal pathway and the baseline covariates are other independent variables that may (or may not) predict outcome. Natural direct and indirect effects were estimated using an imputation-based approach as recommended,[19,23] and robust standard errors were used to calculate 95% CI.

Models for 2-h glucose ≥11.1 mmol/L at 2 years were performed with logistic regression, while linear regression was performed for the change in 2-h glucose at 2 years from baseline. Estimates are given as odds ratios (OR) or mean difference in the change from baseline in 2-h glucose (mmol/L) with the relevant 95% CI. Mediated proportions were calculated as the natural indirect effect divided by the total effect for change in glucose, using the log OR or the mean change as the effect for 2-h glucose ≥ 1.1 mmol/L and the change in glucose, respectively.[24]

Assumptions for these mediation analyses included that the control for confounders is sufficient for the relationships between (A) treatment and outcome, (B) treatment and mediators, and (C) mediators and outcome (after adjustment for treatment). Lastly, (D) we assume there are no confounders of the mediator–outcome relationship that are affected by treatment. Given that we have an RCT, assumptions (A) and (B) are met.[19,23] Sensitivity analyses will assess assumption (C) by exploring interactions between treatment and mediators. The joint mediation model given above satisfies assumption (D), given all available mediators have been included.[25] An additional sensitivity analysis was used to explore the impact of missing data by multiple imputation. A total of 709 out of 1007 patients have complete data for baseline covariates, mediators, and 2-h glucose at 2 years. The remaining 298 patients have between 1 and 11 variables missing, with most (126/298; 42%) missing the seven variables collected at 2 years (six mediators and 2-h glucose) (Figure S1). Multiple imputation by chained equations was performed, assuming missing at random, using 30 imputations as 30% of patients had missing data. The imputation model used all possible covariates in the full mediation model, that is, baseline covariates, mediators, and 2-h glucose at 2 years. Distributions of imputed data were checked against observed data. Results from the natural effects models were pooled using Rubin's rules.[26]

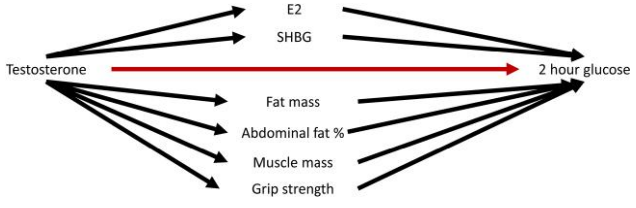

Figure 1. Parallel mediation: relationship between testosterone (treatment), 2-h glucose (outcome), and potential mediators.

Analyses were conducted in R version 4.1[27] using the gtsummary,[28] medflex[23] and mice[29] packages, and SAS version 9.4 (Cary, USA). The code used to perform these analyses is publicly available at https://github.com/kristyrobledo/T4DM_mediation_paper.

Results

There were 709 (70.4%) participants with complete data for the investigation of testosterone treatment mediation (Figure S1). A comparison of the baseline characteristics between those patients with and without data available is given in Table S1. Briefly, participants were comparable for most characteristics; however, those without data available appeared to be 3 kg heavier, with 3 cm larger waist circumferences, more fat mass (3 kg), and less likely to have a history of T2D. For 2-h glucose ≥11.1 mmol/L at 2 years in those with data available, the unadjusted OR for treatment was 0.53 (95% CI: 0.35-0.79), which became 0.48 (95% CI: 0.30-0.76) after adjustment for baseline covariates.

The effect of treatment was attenuated (OR 0.75 (95% CI: 0.42-1.36)) after inclusion of all six mediators (fat mass, abdominal fat percentage, skeletal muscle mass, grip strength, E2, and SHBG). A comparison of these results is given in Figure 2, showing the attenuation of the OR for 2-h glucose ≥11.1 mmol/L, as well as the attenuation in the change in the 2-h glucose from baseline.

Further exploration of these treatment effects with mediation analyses allowed the estimation of both the direct and indirect effects of testosterone treatment. For both outcomes, the estimated direct effect of testosterone and the estimated indirect effect, mediated through the six mediators, are given in Figure 3. For a subject with given mean baseline covariates, altering treatment from placebo to testosterone while controlling for the given mediators decreases the odds of 2-h glucose ≥11.1 mmol/L by a factor of 0.77 (OR 0.77; 95% CI: 0.44-1.35; direct effect of testosterone treatment compared to placebo). Altering mediator levels as observed in those allocated to placebo to those allocated testosterone, while controlling for baseline covariates, decreases the odds of diabetes at 2 years by a factor of 0.62 (95% CI: 0.45-0.86; indirect effect of testosterone treatment compared to placebo). This gives a proportion mediated of 65.1% for 2-h glucose ≥11.1 mmol/L. Similarly, the direct effect for the change in 2-h glucose is −0.26 mmol/L (95% CI: −0.69-0.17) and the indirect effect is −0.50 mmol/L (95% CI: −0.77 to −0.23), giving a proportion mediated of 65.9%. There was no evidence of interactions between any of the mediators and treatment (all *P*-values >.61).

A sensitivity analysis was performed to explore the effects of missing data, using multiple imputation. The direct and indirect effects in Table S2 show that the effect of testosterone is still partially (but not wholly) mediated by these given six variables, with the proportion mediated slightly attenuated from

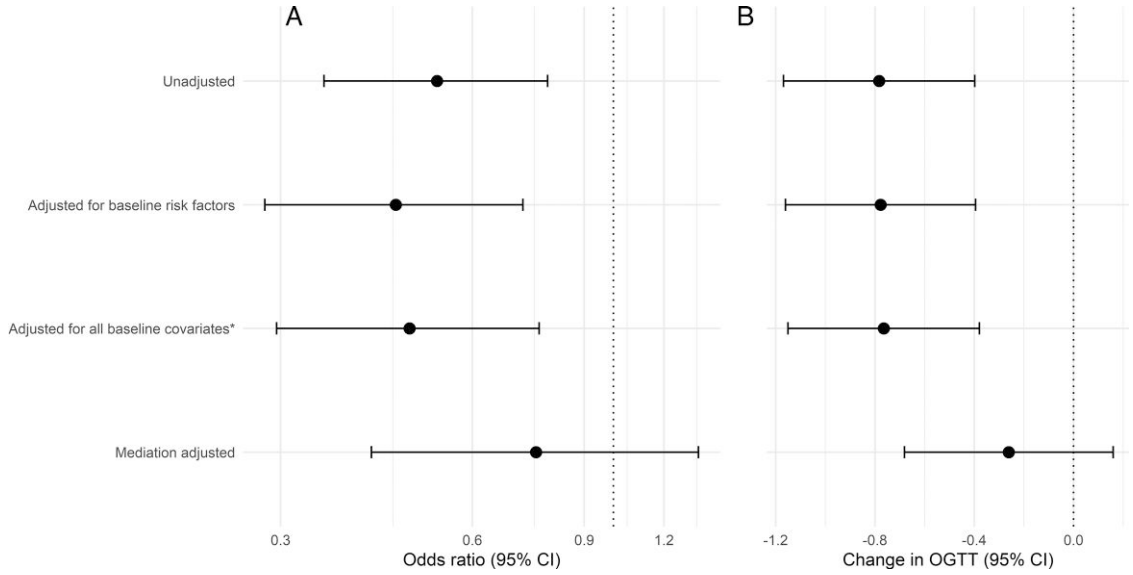

Figure 2. Comparison of testosterone treatment effects from unadjusted and adjusted models for (A) 2-h glucose ≥11.1 mmol/L and (B) changes in oral glucose tolerance test (OGTT) levels at 2 years.

65% to ~50%. Another sensitivity analysis explored the exclusion of SHBG as a possible mediator, instead including SHBG as a baseline risk factor only. This gave 56% and 63% of the effect of testosterone mediated (for 2-h glucose ≥11.1 mmol/L and change in glucose respectively) compared to the ~65% seen previously (Table S3).

While we have shown that the effects of testosterone are partially mediated through changes in fat mass, skeletal muscle mass, abdominal fat percentage, grip strength, E2, and SHBG, it is unclear which of these (if any) is the primary mediator. However, if we explore the effects of the mediators in the full model (Table 1), we can see that the change in fat mass retains a relationship with outcome for 2-h glucose ≥11.1 mmol/L, after accounting for the effect of testosterone. Similar results are seen for change in 2-h glucose, with changes in fat mass the only mediator prognostic for outcome. A 1-kg decrease in fat mass at 2 years from baseline gives a mean decrease in 2-h glucose of 0.20 mmol/L (95% CI: 0.12 to 0.28) or decreases the odds by 0.81 (OR: 1/1.23 = 0.81; 95% CI: 0.72 to 0.92) for 2-h glucose ≥11.1 mmol/L, after adjustment for baseline covariates, treatment, and other mediators. A 1 pmol/L increase in E2 at 2 years compared to baseline corresponds to an OR of 1.002 (95% CI: 1.000 to 1.004) for 2-h glucose ≥11.1 mmol/L, after adjustment for baseline covariates, treatment, and other mediators. Meanwhile, a 1-kg increase in skeletal muscle mass at 2 years compared to baseline corresponds to an OR of 1.01 (95% CI: 0.91 to 1.13) for 2-h glucose ≥11.1 mmol/L, after adjustment for baseline covariates, treatment, and other mediators. Almost negligible effects are seen for SHBG, grip strength, and abdominal fat mass after adjustment for other factors.

Discussion

We found in this mediation analysis that the effect of testosterone on 2-h glucose at 2 years was attenuated with the inclusion of treatment mediators over the 2 years indicating that around 65% of the treatment effect may be explained through the combined effects of changes in body composition, grip strength, E2, and SHBG.

While other trials have established an improvement in glucose tolerance in parallel to a decrease in fat mass, mediation analyses have not been performed to ascertain the relative effects of fat mass and other possible mediators. This novel mediation analysis shows that a decrease in fat mass is the predominant (but not entire) mechanism by which testosterone mediates the beneficial effects on glucose tolerance and diabetes risk. The remaining effect is the direct effect of testosterone and/or the effects of any unmeasured mediators.

We anticipated a more significant effect of muscle mass and strength in mediating the testosterone effect on glycaemia. It is possible that either (or both) the increase in muscle mass or strength was insufficient or (and) alterations in muscle fibre composition and function following treatment with testosterone are different compared to the effects of physical activity or differ in the context of concomitant diet-induced weight loss. In the T4DM study, the mean increase in skeletal muscle mass in response to testosterone was modest at 0.4 kg, but contrasts with the 1.3-kg decrease in skeletal muscle mass in the placebo group. Other studies of testosterone treatment show significant greater effects of testosterone treatment on skeletal muscle mass. For example, in a group of men who were on average older than those in the current study, topical testosterone increased skeletal muscle mass by ~1 kg[30] and 1.9 kg[31] after 6 months and 3 years treatment, respectively. However, unlike T4DM, none of these studies had a diet-induced weight loss component. Two prior studies of testosterone treatment in the context of dietary induced weight loss, one in a population of men with obesity and mean age just over 50 years where treatment was with 3 monthly testosterone undecanoate over 12 months[32] and the other using topical testosterone gel in in men with obesity aged 65 and over[33] showed that treatment with testosterone preserved but did not increase lean body mass. Alternatively, in line with our findings, another view is that diet-induced weight loss attenuated the effects of testosterone on muscle mass.

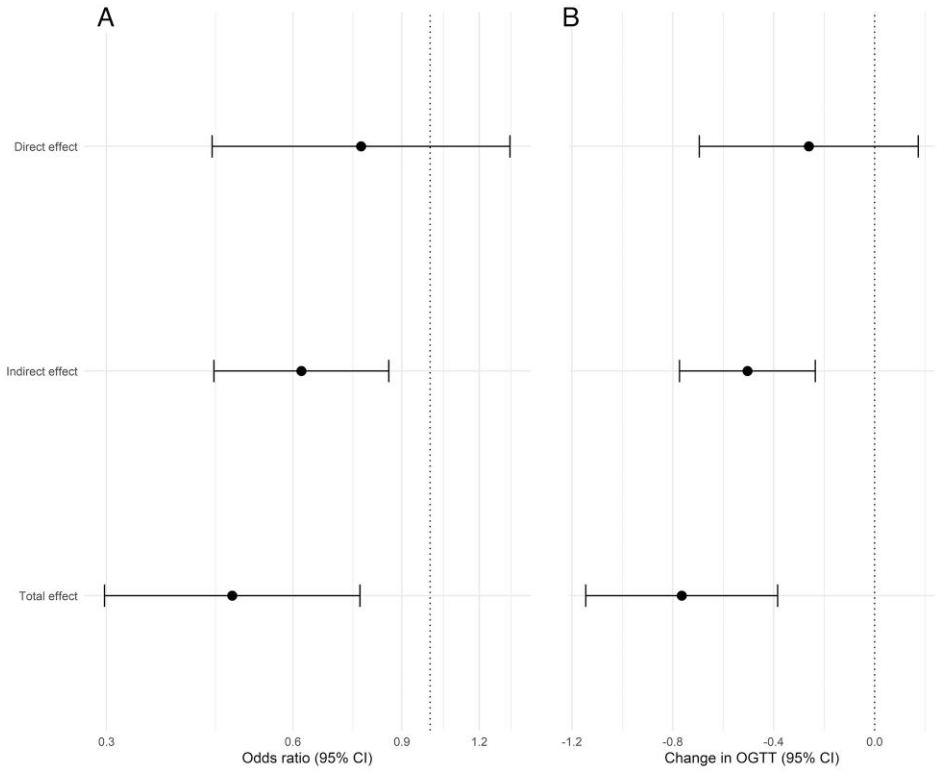

Figure 3. Decomposition of the testosterone treatment effects into direct and indirect effects for (A) 2-h glucose ≥ 11.1 mmol/L and (B) changes in oral glucose tolerance test (OGTT) levels at 2 years.

Table 1. Treatment mediation: estimates of effects from models of 2-h glucose ≥11.1 mmol/L and change in OGTT at 2 years.

	2-h glucose ≥ 11.1			2-h glucose change from baseline		
Characteristic	OR[a,b]	95% CI[b]	*P*-value	Mean change[c]	95% CI[b]	*P*-value
Treatment with testosterone	0.75	0.42, 1.36	.4	−0.26	−0.68, 0.16	.2
Change in skeletal muscle mass (kg)	1.01	0.91, 1.13	.8	0.04	−0.04, 0.11	.3
Change in fat mass (kg)	1.23	1.09, 1.39	<.001	0.20	0.12, 0.28	<.001
Change in abdominal fat (%)	1.02	0.90, 1.15	.8	0.00	−0.09, 0.08	>.9
Change in non-dominant grip strength (kg)	1.01	0.97, 1.06	.5	0.00	−0.03, 0.03	.8
Change in E2	1.00	1.00, 1.00	.066	0.00	0.00, 0.00	.2
Change in SHBG	1.00	0.97, 1.03	.8	0.02	−0.01, 0.04	.14

[a]Models are adjusted for all baseline covariates (baseline risk factors and baseline mediators).
[b]OR, Odds Ratio; CI, Confidence Interval.
[c]Change is calculated as 2 years minus baseline, with positive values indicating increases from baseline and negative as decreases from baseline.
Abbreviations: E2, oestradiol; SHBG, sex hormone-binding globulin.

While the increase in muscle mass may not have been sufficient to independently mediate part of the effect of testosterone on glucose metabolism, functional changes in muscle may occur independent of trophic effects. For example, aerobic training improves fatty acid oxidation in skeletal muscle but a 1.4-kg increase in muscle mass induced by resistance training did not.[34]

Another potential mechanism is differential effects on muscle fibre type, as T2D is associated with reduced and functionally different type I (slow twitch myosin heavy chain) and type II (fast twitch myosin heavy chain) muscle fibres.[35] Type I muscle fibres appear to be more important than type II fibres for whole body insulin sensitivity,[36] and are more responsive to testosterone.[37,38] By contrast, the benefit of the diet and exercise intervention in the Finnish Diabetes Prevention Study was associated with an improvement glucose metabolism in type II fibres.[39]

There are accumulating data on the role of E2 in mediating insulin sensitivity and glucose metabolism in males.[40] In this study, after adjustment for all other mediators, there was no significant effect of E2. The E2 receptor mutations and aromatase deficiency in males are associated with increased adipose tissue, insulin resistance, impaired glucose metabolism, and dyslipidaemia.[41] Short-term treatment with E2 regulates lipid metabolism pathways in skeletal muscle of men.[42] Recent experimental data in animals demonstrate that E2 induces, via ERα dependent mechanisms, an increase GLUT 4 expression and activity[8] and insulin transport across vascular endothelium, in skeletal muscle.[9] E2 increases overnight pulsatile growth hormone (GH) secretion from the pituitary while inhibiting the hepatic IGF-1 response to GH,[43] and therefore increasing skeletal muscle glucose metabolism independent of any trophic effects.

Further, beyond effects in skeletal muscle, E2 via ERα may also improve glucose metabolism in males by improving pancreatic beta cell function and central nervous system regulation of insulin production.[44] There are a few possible reasons why we did not see a significant mediating effect of E2. Testosterone is converted to E2 by the enzyme aromatase in a tissue-specific manner, and while serum E2 concentrations increased in response to treatment with testosterone in T4DM, this provides only a limited approximation of tissue E2 exposure. Further, serum E2 concentration was measured in trough samples taken just prior to the next injection and it is likely the proportion of E2 derived from aromatisation of testosterone in adipose tissue decreased as fat mass decreased. The absence of a significant mediating effect of SHBG is consistent with published data showing that men with obesity and the metabolic syndrome testosterone but not SHBG is an independent determinant of incident T2D.[45]

The strength of this study is the randomised design of the T4DM trial, which allows assumptions of mediation analyses to be met. As participants are randomly allocated to testosterone or placebo treatment, the relationships between both treatment and outcome, and treatment and mediators, are therefore not confounded.[19,23] Further, the use of natural effects models in a counterfactual framework enabled the testosterone effect to be decomposed into direct and indirect effects in an easily interpretable fashion. While 30% of the patients had missing data for the analyses, predominately due to not returning for the 2-year assessments, our findings in complete case analyses were confirmed with sensitivity analyses using multiple imputation.

The main limitation is the difficulty of decomposition of the indirect effect further into the contribution of each mediator, as mediation analyses require that all potential confounders are concurrently present in the model. Thus (formal) investigation of the individual contributions currently cannot be conducted with certainty.[17] Nevertheless, we found that only change in fat mass was significantly associated with glucose at 2 years after adjustment for all other mediators, treatment, and baseline variables. This observation is consistent with the well-established relationship between decrease in adipose tissue and risk of T2D.[46] For example, in the diabetes prevention programme, weight loss was the dominant predictor of reduced diabetes incidence with a hazard ratio per 5-kg weight loss of 0.42.[47]

In conclusion, we found that at least part of the effect of testosterone to reduce diabetes risk was mediated through changes in fat mass, abdominal fat, skeletal muscle mass, grip strength, SHBG, and E2. Of the mediators investigated, the largest contributor to mediating the testosterone effect on glucose was change in fat mass. We could not rule out effects of muscle that may have been sensitive to diet-induced weight loss and were not dependent on mass or strength. It is likely that there are other pathways that mediate the effects of testosterone requiring further investigation.

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Supplementary material

Supplementary material is available at *European Journal of Endocrinology* online.

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Contributors

All authors conceived the initial T4DM trial. K.R., G.W., and I.M. proposed the treatment mediation analyses. K.R. performed the analyses and all authors interpreted the analyses. K.R. and G.W. drafted the paper and all authors revised and approved the final paper.

Conflicts of interest: G.W. has received research funding from Bayer, Lilly, Lawley Pharmaceuticals, and WW, and speaker honoraria from Bayer, Lilly and Besins Healthcare. C.A. has received honoraria from Besins Healthcare and is an advisory board member for Ferring. M.G. is on the editorial board of EJE. They were not involved in the review or editorial process for this paper, on which they are listed as an author. M.G. has received research funding from Bayer, Novartis, WW, Lilly and speaker's honoraria from Besins Healthcare and Otsuka. D.J.H. has received institutional grants for investigator-initiated studies of testosterone pharmacology (Lawley, Besins Healthcare) but no personal income and has provided expert testimony to antidoping and professional standards tribunals and testosterone litigation. B.B.Y. has received speaker honoraria and conference support from Bayer, Lilly and Besins Healthcare, and research support from Bayer, Lilly and Lawley Pharmaceuticals, and has held advisory roles with Lilly, Besins Healthcare, Ferring and Lawley Pharmaceuticals. D.J. has received honoraria for speaking at GP education sessions for Lilly, Amgen and Boehringer Ingelheim. B.G.A.S. has received speaker's honoraria from Besins Healthcare. K.R., K.B., W.I., and B.S. declare no relevant conflicts of interest.

Data availability

T4DM data are available on request to the trial management committee. Please contact karen.bracken@sydney.edu.au to submit a proposal to request access.

Ethics approval and consent to participate

The T4DM study was approved by the ethics committee at each of the participating centres, with the lead HREC as Sydney Local Health District HREC—CRGH. The other HRECs were Central Adelaide Local Health Network Human Research Ethics Committee, South Metropolitan Health Service Human Research Ethics Committee and Bellberry Human Research Ethics Committee. All patients gave written informed consent for the trial and separately for further clinic or remote follow-up. The study was performed in accordance with the Declaration of Helsinki, and

registered with Australia and New Zealand Clinical Trials Registry (ACTRN12612000287831).

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