ARTICLE

Oral taurine but not *N*-acetylcysteine ameliorates NEFA-induced impairment in insulin sensitivity and beta cell function in obese and overweight, non-diabetic men

C. Xiao · A. Giacca · G. F. Lewis

Abstract

Aims/hypothesis Antioxidants have been shown to ameliorate lipid-induced impairment of insulin action and beta cell function, both in vitro and in animal studies. The aim of the present study was to examine the effects of two orally administered antioxidants, *N*-acetylcysteine (NAC) and taurine (TAU), on lipotoxicity in humans.

Methods Nine non-diabetic men, who were either overweight or obese, underwent three studies each, 4–6 weeks apart, in random order: (1) i.v. infusion of saline for 48 h (SAL); (2) i.v. infusion of Intralipid and heparin for 48 h to mimic chronic elevation of plasma NEFA (IH); and (3) IH infusion for 48 h with concurrent oral NAC (IH+NAC). Six men underwent similar studies except for study 3, where instead of NAC they received a 2 week pretreatment with oral TAU (IH+TAU).

Results For both the NAC and TAU studies, a 48 h IH infusion alone without antioxidant impaired insulin sensitivity (S_I, 63% and 62% of SAL in NAC and TAU studies, respectively) and beta cell function, as evidenced by a reduction in disposition index (DI, 55% and 54% of SAL in NAC and TAU studies, respectively). NAC failed to prevent the lipid-induced increase in levels of the plasma oxidative stress marker malondialdehyde and did not prevent the lipid-induced reduction in S_I or DI, whereas TAU completely prevented the rise in malondialdehyde and decreased 4-hydroxynonenal, and significantly improved S_I (91% of SAL) and DI (81% of SAL).

Conclusions/interpretation Oral TAU ameliorates lipid-induced functional beta cell decompensation and insulin resistance in humans, possibly by reducing oxidative stress.

ClinicalTrials.gov ID no. NCT0018873

Abbreviations

Cl_I	insulin clearance
DI	disposition index
G_{inf}	glucose infusion rate
GSIS	glucose-stimulated insulin secretion
4-HNE	4-hydroxynonenal
IH	Intralipid plus heparin
IKKβ	IκB kinase β
ISR	insulin secretion rate
MDA	malondialdehyde
NAC	*N*-acetylcysteine
PKC	protein kinase C
ROS	reactive oxygen species
SAL	saline
S_I	insulin sensitivity index
TAU	taurine

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Introduction

Defects in both insulin action and secretion are hallmarks of type 2 diabetes [1–3]. The defect in insulin secretion is characterised by the selective impairment of glucose-stimulated insulin secretion (GSIS) [4, 5]. The causes of the beta cell defect remain unclear, although prolonged elevation of plasma glucose and NEFAs have been proposed to be detrimental to the beta cells, often referred to as glucotoxicity and lipotoxicity, respectively [6, 7]. We and others have previously demonstrated the impairing effects of chronically elevated plasma NEFAs on GSIS and insulin sensitivity in humans, especially in predisposed individuals [8–12].

Several mechanisms have been proposed to explain lipid-induced insulin resistance and beta cell dysfunction, including but not limited to, increased synthesis of lipid metabolites such as ceramides [13] and diacylglycerides [14], chronic inflammation [15], endoplasmic reticulum stress [16] and oxidative stress [17, 18]. Of note, oxidative stress can activate the JNK [19] and nuclear factor κB [17] proinflammatory pathways and endoplasmic reticulum stress [20]. Lipid-induced insulin resistance in healthy humans is associated with oxidative stress [21].

In animal models and in vitro studies, antioxidants have been shown to improve insulin sensitivity and beta cell function. Infusion of the antioxidant N-acetylcysteine (NAC) or taurine (TAU) abolished hyperglycaemia-induced insulin resistance [22]. NAC prevented diabetes in Zucker diabetic fatty rats and ob/ob mice, partly due to its effects on GSIS and preservation of beta cell mass [23]. TAU was shown to improve insulin sensitivity in diabetic rats [24]. In our preliminary studies, NAC prevented NEFA-induced insulin resistance, and both NAC and TAU prevented a reduction in GSIS in rats [25]. In both healthy subjects and in type 2 diabetic patients, restoration of redox balance by infusion of glutathione improved insulin sensitivity and beta cell function [26]. Infusion of reduced glutathione, an antioxidant, partially prevented lipid-induced insulin resistance in healthy humans [21]. The effects of NAC and TAU in lipid-induced insulin resistance and beta cell dysfunction have not been investigated in humans. The objective of this study was to investigate whether these antioxidants, when administered orally at their currently approved dosages, can prevent or ameliorate the insulin resistance and impairment of beta cell function induced by chronic elevation of NEFA in healthy men.

Methods

Participants Overweight or obese, but otherwise healthy, Caucasian men participated in the studies (n=9 for the NAC study and n=6 for the TAU study, Table 1). None of the participants was taking any medication or had any known systemic illness. Informed, written consent was obtained from all participants in accordance with the guidelines of the Human Subjects Review Committee of the University Health Network, University of Toronto.

Experimental protocol Participants were admitted to the Metabolic Testing Centre of the Toronto General Hospital on three occasions in random order, 4–6 weeks apart, after a 12 h overnight fast. On each occasion, subjects received one of the three following treatments: (1) intravenous infusion of saline for 48 h (SAL); (2) intravenous infusion of Intralipid (Baxter Healthcare, Mississauga, ON, Canada) plus heparin for 48 h to raise plasma NEFAs by approximately twofold, as previously described [8] (IH); (3) IH infusion for 48 h together with either oral NAC (IH+NAC, NAC study) or oral TAU (IH+TAU, TAU study). In the TAU study the subjects were also pretreated with TAU for 2 weeks. Studies with NAC or TAU without lipid infusion were not performed since we have shown in rats that NAC or TAU, even at high intravenous dosages, did not exert any effect on insulin sensitivity or GSIS in the absence of lipid infusion. The dosage of NAC was the same as that recommended for acetaminophen overdose, which is the maximum oral dose for therapeutic use. NAC is not currently approved in Canada for prolonged oral administration at this dose beyond 48–72 h. NAC was administered with a loading dose of 140 mg/kg followed by 70 mg/kg every 4 h during the 48 h IH infusion. In the TAU study, participants ingested TAU (Jarrow Formulas, Los Angeles, CA, USA) at a dosage of 3 g/day divided into three equal doses for 2 weeks prior to and during the IH infusion. Blood samples were taken before and during lipid/saline infusion for assays of plasma glucose, insulin, C-peptide, triacyl-

Table 1 Characteristics of the subjects

Characteristic	Study	
	NAC (n=9)	TAU (n=6)
Age (years)	46.0±2.4 (34–56)	47.0±2.6 (41–56)
BMI (kg/m^2)	30.6±1.3 (28.0–38.8)	30.6±1.5 (27.9–39.1)
Waist circumference (cm)	108.1±4.2 (99.3–136.0)	109.4±5.9 (96.7–137.0)
Fasting plasma concentration		
Glucose (mmol/l)	5.3±0.1 (4.6–5.8)	5.6±0.2 (5.3–6.3)
Insulin (pmol/l)	74.9±8.3 (49.8–110.8)	77.5±10.2 (44.7–113.6)
C-peptide (nmol/l)	0.85±0.13 (0.48–1.57)	0.94±0.12 (0.63–1.26)
NEFA (mmol/l)	0.38±0.05 (0.17–0.55)	0.41±0.03 (0.32–0.54)
Triacylglycerols (mmol/l)	1.68±0.27 (0.77–3.07)	1.51±0.26 (0.89–2.90)

All subjects are male. Values are means±SEM (range)

glycerols and NEFAs. Plasma aldehydes malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE) were analysed in the samples taken on day 3 prior to the hyperglycaemic clamp. The primary [insulin sensitivity (S_I) and beta cell function as indicated by disposition index (DI)] and secondary [insulin secretion rate (ISR), endogenous insulin clearance and oxidative stress markers] outcomes measures were assessed as below.

Assessment of GSIS, insulin sensitivity and insulin clearance In each study, on day 3 of the infusion, a 2 h hyperglycaemic clamp and a 2 h euglycaemic–hyperinsulinaemic clamp were performed to assess GSIS, S_I and insulin clearance (Cl_I). Please refer to the Electronic supplementary material (ESM) for further details.

Calculations of ISR, S_I, Cl_I and DI ISR was calculated from deconvolution of plasma C-peptide levels during the last 30 min of the hyperglycaemic clamp [27]. S_I was calculated as $S_I = Cl_{glc}/(Ins_{clamp} - Ins_{base})$ from the last 30 min of the euglycaemic clamp, where Cl_{glc} is the glucose clearance estimated as glucose infusion rate (G_{inf}) divided by glucose concentration; Ins_{clamp} is the insulin concentration during the clamp; Ins_{base} is the basal insulin level. Cl_I was calculated as $Cl_I = ISR/(Ins_{clamp} - Ins_{base})$ during the last 30 min of the hyperglycaemic clamp. DI was calculated as $DI = ISR \times S_I$ using the ISR and S_I estimated as above.

Laboratory methods Plasma glucose, insulin, C-peptide, triacylglycerols and NEFAs were assayed as previously described [8]. Plasma MDA and 4-HNE were analysed by a modification of a previously described method [28, 29], on a Hewlett-Packard 5973N MSD GC-MS equipped with an HP-5 MS column and performed in negative-ion chemical ionisation.

Statistical analysis Plasma glucose, insulin, C-peptide, triacylglycerols and NEFAs during the 48 h infusion period were analysed by two-way ANOVA for repeated measures with Tukey's multiple comparison test to detect differences between treatments at each time and between times within treatment. Plasma aldehyde concentrations were analysed by two-way ANOVA. Two-way ANOVA for repeated measures with Tukey's multiple comparison test was performed to detect differences between treatments during the last 30 min of the clamps. A p value of <0.05 was considered significant. All statistical analyses were performed with SAS software (version 8.0; Cary, NC, USA).

Results

NAC study Plasma triacylglycerols and NEFAs were elevated by IH infusion by approximately two- to fourfold from pre-infusion levels and maintained at levels approximately twofold higher than pre-infusion levels prior to the hyperglycaemic clamp. Both triacylglycerols ($p<0.01$ IH and IH+NAC vs SAL) and NEFAs ($p<0.01$ IH vs SAL, $p<0.05$ IH+NAC vs SAL) were higher in IH and IH+NAC than in SAL after 48 h IH infusion (Table 2).

Plasma glucose, insulin and C-peptide levels and G_{inf} during the last 30 min of the hyperglycaemic clamp are

Table 2 Fasting plasma glucose, triacylglycerol and NEFA levels for 48 h before the hyperglycaemic clamp in the NAC study

Day/time (24 h clock)	Glucose (mmol/l)			Triacylglycerol (mmol/l)			NEFA (mmol/l)		
	SAL	IH	IH+NAC	SAL	IH	IH+NAC	SAL	IH	IH+NAC
Day 1									
08:00 hours	4.9±0.1	5.2±0.2	5.1±0.1	2.02±0.32	1.64±0.30	1.57±0.15	0.36±0.04	0.35±0.05	0.47±0.07
12:00 hours	5.4±0.3	5.3±0.3	5.7±0.2	2.40±0.39	4.04±0.90*	3.55±0.34*,†	0.34±0.11	0.97±0.19*,†	0.85±0.14
16:00 hours	5.2±0.3	5.5±0.3	5.5±0.3	2.66±0.49	4.73±0.66**,‡	5.20±0.37**,‡	0.31±0.09	1.02±0.11**,‡	1.33±0.21**,‡
Day 2									
08:00 hours	5.3±0.1	5.3±0.2	5.2±0.1	1.96±0.25	4.85±0.70**,‡	5.36±0.74**,‡	0.36±0.08	1.54±0.18**,‡	1.52±0.22**,‡
12:00 hours	5.0±0.2	5.2±0.4	5.0±0.1	2.18±0.27	5.53±0.84**,‡	5.71±0.87**,‡	0.28±0.07	1.58±0.22**,‡	1.45±0.17**,‡
16:00 hours	5.5±0.1	5.7±0.3	5.8±0.4	2.27±0.33	5.80±0.99**,‡	5.16±0.68**,‡	0.24±0.05	1.62±0.25**,‡	1.33±0.20**,‡
Day 3									
08:00 hours	5.3±0.2	5.8±0.1	5.9±0.3	1.70±0.27	3.96±0.31**,‡	3.55±0.37**,†	0.41±0.05	0.84±0.08*,‡	0.68±0.08*,†

SAL, saline infusion; IH, Intralipid plus heparin infusion; IH+NAC, Intralipid plus heparin infusion plus oral NAC. Infusions of saline or Intralipid plus heparin and oral administration of NAC were started immediately after the blood sample at 08:00 hours on day 1 and continued throughout the hyperglycaemic and euglycaemic–hyperinsulinaemic clamp studies, which were conducted immediately after the sample at 08:00 hours was drawn on day 3. Subjects were fasted for 12 h overnight prior to the sample at 0800 hours on days 1 and 3 but ingested three meals per day during the remainder of the 48 h infusion studies.

Data are means±SEM; $n=9$. *$p<0.05$ vs 08:00 hours on day 1, **$p<0.01$ vs 08:00 hours on day 1, †$p<0.05$ vs SAL, ‡$p<0.01$ vs SAL
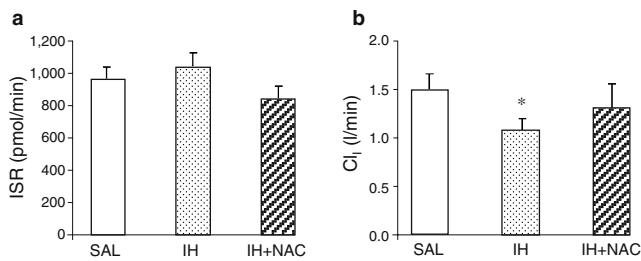
Fig. 1 ISR (**a**) and Cl_I (**b**) calculated from the last 30 min of the hyperglycaemic clamp. *$p < 0.05$ vs saline

shown in ESM Fig. 1. The calculated ISR did not differ between treatments (Fig. 1a). Cl_I in IH was decreased as compared with SAL ($p < 0.05$), and Cl_I in IH+NAC was not significantly different from either SAL or IH (Fig. 1b).

Plasma glucose and insulin levels and G_{inf} during the last 30 min of the euglycaemic–hyperinsulinaemic clamp are presented in ESM Fig. 2. The calculated S_I was significantly lower in IH and IH+NAC than in SAL ($p < 0.0001$; Fig. 2a). No significant difference in S_I was observed between IH and IH+NAC. DI was reduced by 33% by IH infusion alone and by 41% by IH+NAC ($p < 0.0001$). DI in IH+NAC was not significantly different from that in IH (Fig. 2b). Consistent with the lack of effects of NAC on S_I and DI, NAC did not prevent the rise of plasma oxidative stress markers induced by IH infusion. After 48 h IH infusion, plasma MDA concentrations were elevated ($p < 0.05$ for IH and IH+NAC vs SAL). Plasma 4-HNE concentrations in IH were not significantly different from SAL. Both 4-HNE and MDA concentrations in IH+NAC were similar to those in IH (Fig. 2c).

TAU study IH infusion for 48 h increased plasma NEFA and triacylglycerols from pre-infusion levels ($p < 0.05$), while NEFA and triacylglycerol levels in SAL remained relatively unchanged, which resulted in an approximately twofold elevation in plasma NEFA and triacylglycerols in IH and IH+TAU compared with SAL ($p < 0.05$) prior to the hyperglycaemic clamp (Table 3).

Plasma glucose, insulin and C-peptide levels and G_{inf} during the last 30 min of the hyperglycaemic clamp are shown in ESM Fig. 3. To maintain plasma glucose at ~20 mmol/l, slightly higher G_{inf} were required in IH than in SAL and IH+TAU ($p < 0.05$), whereas G_{inf} in IH+TAU were similar to those in SAL. Insulin levels were higher in IH than in SAL ($p < 0.05$) and IH+TAU ($p < 0.05$) but not different between IH+TAU and SAL. The calculated ISR did not differ between protocols (Fig. 3a). The calculated Cl_I was significantly lower in IH ($p < 0.05$) than in IH+TAU or SAL (Fig. 3b).

Plasma glucose, insulin levels and G_{inf} during the last 30 min of the euglycaemic–hyperinsulinemic clamp are presented in ESM Fig. 4. To maintain plasma glucose at ~5.6 mmol/l, a lower G_{inf} was required in IH ($p < 0.01$) and IH+TAU ($p < 0.05$) than in SAL. G_{inf} in IH+TAU was higher than that in IH ($p < 0.05$). Insulin concentrations in IH+TAU were not different from SAL but were significantly lower than in IH ($p < 0.05$).

IH infusion significantly decreased S_I ($p < 0.05$ vs SAL), whereas S_I in IH+TAU did not differ from SAL (Fig. 4a). DI (Fig. 4b) was reduced by 46% by IH infusion ($p < 0.05$). Compared with IH, ingestion of oral TAU for 2 weeks improved DI by 51% ($p < 0.05$) to a value not significantly different from SAL. IH infusion for 48 h elevated plasma MDA as compared with SAL ($p < 0.05$). Consistent with the effects of TAU on S_I and DI, plasma MDA levels in IH+TAU were lower than in IH ($p < 0.05$) but similar to SAL (Fig. 4c). Plasma 4-HNE levels in IH+TAU were lower than in both SAL and IH ($p < 0.05$).
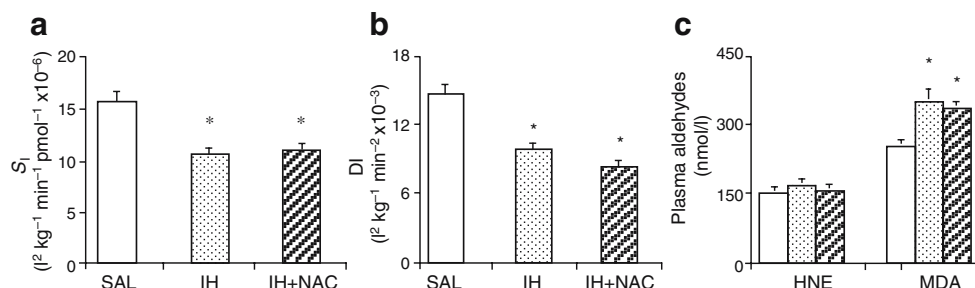
Fig. 2 **a** Insulin sensitivity index (S_I) calculated from the last 30 min of the euglycaemic–hyperinsulinaemic clamp in the NAC study, which was performed after 48 h of infusion of either saline (SAL), Intralipid plus heparin (IH), or Intralipid plus heparin plus oral NAC (IH+NAC). **b** Disposition index (DI) was calculated as the product of S_I derived from the euglycaemic–hyperinsulinaemic clamp and ISR derived from the hyperglycaemic clamp. **c** Plasma aldehyde concentrations were measured after 48 h infusion of either saline (white bars), Intralipid plus heparin (dotted bars), or Intralipid plus heparin plus oral NAC (hatched bars). *$p < 0.05$ vs saline

Table 3 Fasting plasma glucose, triacylglycerol and NEFA levels for 48 h before the hyperglycaemic clamp in the TAU study

Day/time (24 h clock)	Glucose (mmol/l)			Triacylglycerols (mmol/l)			NEFA (mmol/l)		
	SAL	IH	IH+TAU	SAL	IH	IH+TAU	SAL	IH	IH+TAU
Day 1									
08:00 hours	5.1±0.2	5.3±0.1	5.2±0.1	1.7±0.39	1.61±0.38	1.32±0.12	0.43±0.05	0.36±0.06	0.44±0.06
12:00 hours	5.4±0.5	5.8±0.3	6.1±0.6	2.16±0.51	3.78±1.14*	3.35±0.40*,‡	0.25±0.11	0.94±0.26*,†	0.96±0.18
16:00 hours	5.9±0.6	6.3±0.2	6.5±0.4	2.48±0.48	4.78±0.91**,‡	4.79±0.53**,‡	0.28±0.09	1.16±0.16**,‡	1.27±0.18**,‡
Day 2									
08:00 hours	5.2±0.2	5.5±0.1	5.4±0.2	1.75±0.28	4.63±0.86**,‡	4.05±0.61**,‡	0.41±0.09	1.49±0.20**,‡	1.45±0.18**,‡
12:00 hours	5.1±0.2	5.6±0.3	5.9±0.3	2.17±0.31	5.03±1.23**,‡	4.37±0.65**,‡	0.23±0.06	1.26±0.14 **,‡	1.36±0.19 **,‡
16:00 hours	5.6±0.2	6.2±0.4	6.2±0.3	2.31±0.37	5.39±1.24 **,‡	4.04±0.77**,‡	0.25±0.06	1.34±0.14**,‡	1.67±0.20**,‡
Day 3									
08:00 hours	5.3±0.2	6.0±0.3	5.8±0.2	1.77±0.32	3.06±0.42**,‡	3.26±0.36**,†	0.44±0.05	0.71±0.09*,‡	0.77±0.07*,†

SAL, saline infusion; IH, Intralipid plus heparin infusion; IH+TAU, Intralipid plus heparin infusion plus oral TAU. Infusions of saline or Intralipid plus heparin were started immediately after the blood sample at 08:00 hours on day 1 and continued throughout the hyperglycaemic and euglycaemic–hyperinsulinaemic clamp studies, which were conducted immediately after the sample at 08:00 hours was drawn on day 3. Oral administration of TAU was started 2 weeks before day 1 and continued throughout the studies. Subjects were fasted for 12 h overnight prior to the sample at 08:00 hours on days 1 and 3 but ingested three meals per day during the remainder of the 48 h infusion studies.

Data are means±SEM; n=6. *p<0.05 vs 08:00 hours on day 1, **p<0.01 vs 08:00 hours on day 1, †p<0.05 vs SAL, ‡p<0.01 vs SAL
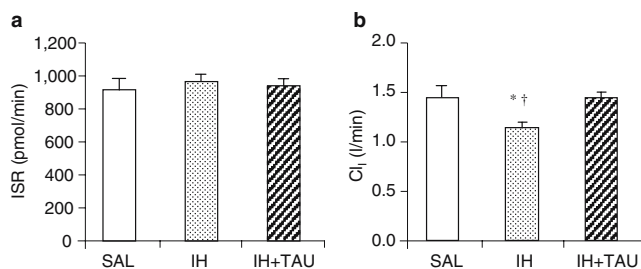
Discussion

In the present study we report that TAU, but not NAC, administered orally at currently recommended dosages, ameliorated lipid-induced impairments in insulin sensitivity and beta cell function in overweight and obese, non-diabetic men. To our knowledge, the effects of these two agents, which have antioxidant activity, on lipid-induced insulin resistance and beta cell dysfunction have not previously been examined in humans.

As we and others have previously shown, chronic elevation of plasma NEFAs by Intralipid plus heparin infusion induces insulin resistance [6, 8, 12]. This effect of NEFAs was partially prevented by 2 weeks of oral TAU pretreatment. TAU is an aldehyde scavenger, and by reducing cytotoxic aldehydes, producing reactive oxygen species (ROS), it serves as an antioxidant in vivo. TAU is

Fig. 3 ISR (**a**) and insulin clearance (Cl$_l$) (**b**) calculated from the last 30 min of the hyperglycaemic clamp. *p<0.05 vs saline, †p<0.05 vs Intralipid plus heparin plus oral TAU

also synthesised from cysteine; thus, TAU supplementation may preserve cysteine for synthesis of glutathione, a natural intracellular antioxidant [30, 31]. In the current study, 2 weeks of oral TAU prevented lipid infusion-induced elevation of MDA in plasma, suggesting that TAU had an antioxidant effect. Both NAC and TAU, when given i.v. to rats, prevented hyperglycaemia-induced insulin resistance—which was also thought to be mediated through oxidative stress—and increases in lipid peroxidation products [22]. In humans, lipid-induced insulin resistance correlated with oxidative stress [32–34]. Co-infusion of reduced glutathione with Intralipid for 6 h improved whole-body glucose disposal as compared with lipid infusion alone [21]. Lipid-induced muscle insulin resistance in humans is associated with increased protein kinase C (PKC)-δ and -βII membrane translocation and decreased inhibitor of nuclear factor κB-α, evidence of IκB kinase β (IKKβ) activation [35]. In our recent studies in rats, lipid-induced PKC-δ translocation induced oxidative stress, which in turn activated IKKβ/JNK1 and increased serine phosphorylation of insulin receptor substrates 1 and 2, resulting in insulin resistance (unpublished results). It is therefore tempting to postulate that TAU ameliorated lipid-induced insulin resistance through prevention of oxidative stress induced by PKC activation and subsequent events.

No significant alleviating effect of NAC on lipid-induced insulin resistance was observed. As an antioxidant, NAC not only serves as a precursor for glutathione synthesis but also directly reduces free-radical species and other oxidants [36]. In the rodent studies discussed above [22, 25], NAC was infused intravenously. Unlike TAU,
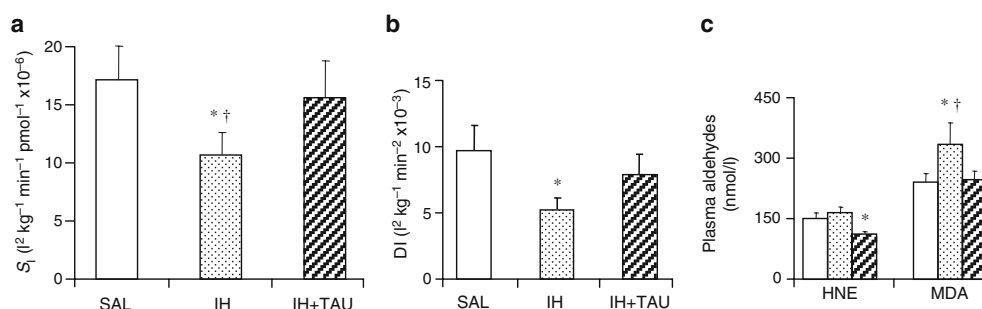
Fig. 4 **a** Insulin sensitivity index (S_I) calculated from the last 30 min of the euglycaemic–hyperinsulinemic clamp in the TAU study, which was performed after 48 h of infusion of either saline (SAL), Intralipid plus heparin (IH) or Intralipid plus heparin following 2 weeks of oral TAU (IH+TAU). **b** Disposition index (DI) was calculated as the product of S_I derived from the euglycaemic–hyperinsulinaemic clamp and ISR derived from the hyperglycaemic clamp. **c** Plasma aldehyde concentrations were measured after 48 h infusion of either saline (white bars), Intralipid plus heparin (dotted bars), or Intralipid plus heparin following 2 week oral TAU (hatched bars). *$p<0.05$ vs saline, †$p<0.05$ vs Intralipid plus heparin plus oral TAU

NAC undergoes extensive first-pass hepatic metabolism when administered orally, resulting in low systemic bioavailability [37, 38]. The partial effect of NAC on endogenous insulin clearance suggests that effective NAC concentrations were reached in the liver. Therefore, the lack of effect of NAC in improving insulin sensitivity in this study might be due to insufficient NAC delivered to peripheral tissues, thus limiting the direct effect of NAC. Consistent with this, oral NAC did not prevent the rise in the lipid peroxidation product MDA in the plasma stimulated by chronic IH infusion, which suggests a lack of adequate systemic antioxidant effect of NAC administered in this fashion. In this study, the power to detect a difference for both S_I and DI were greater than 78%; thus, the lack of effect of NAC does not seem to be due to type II errors. Future studies are required to examine the effects of i.v. NAC or oral NAC over a prolonged period.

IH, NAC and TAU did not affect absolute GSIS in this study. Although GSIS was not reduced by IH infusion, beta cells failed to compensate for the lipid-induced insulin resistance by appropriately increasing insulin secretion, as evidenced by a reduction in the DI, as previously shown [8–10]. In healthy individuals there is a hyperbolic relationship between insulin secretion and sensitivity [39, 40]. Two weeks of oral TAU restored DI with IH infusion to the level with SAL infusion, which indicates that this hyperbolic relationship was restored even in chronically elevated plasma NEFAs. Since improvement of DI in IH+TAU was similar to that of insulin sensitivity, the secretory capacity of beta cells was now 'adequate', with improved insulin sensitivity. Very few studies have directly evaluated the effects of TAU on beta cell function, and no previous study has examined its effect on lipid-induced beta cell dysfunction in humans. Brons et al. [41] did not find any effect of TAU (1.5 g/day for two crossover periods of

8 weeks) alone (without lipid infusion) on insulin secretion in overweight first-degree relatives of individuals with type 2 diabetes. In addition to higher TAU dosages, our study was designed to specifically examine the effects of TAU on insulin resistance and beta cell dysfunction induced by a prolonged elevation of plasma NEFAs. In our studies in rats, NAC or TAU administered intravenously prevented lipid-induced impairment of GSIS, in accordance with their ability to prevent the lipid-induced increase in islet ROS [25], but had no effect in the absence of lipid infusion. The direct effects of antioxidants on lipid-induced beta cell dysfunction have not yet been examined in human islets, although compounds with antioxidant properties were effective [42].

In conclusion, oral administration of TAU, but not NAC, at dosages currently approved in Canada, ameliorated lipid-induced impairment in insulin sensitivity and beta cell function in non-diabetic men who were either overweight or obese. This study suggests, but does not provide definitive proof, that the beneficial effect of TAU was mediated by its antioxidant activity, and we acknowledge that mechanisms independent of oxidative stress cannot be ruled out. We also acknowledge that this short-term study was done in limited numbers of male participants only under experimental conditions of lipid infusion. Further studies are needed to provide data on the safety and long-term impact of TAU on glucose homeostasis.

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