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N-Acetylcysteine Administration Prevents Nonthyroidal Illness Syndrome in Patients With Acute Myocardial Infarction: A Randomized Clinical Trial

Josi Vidart, Simone Magagnin Wajner, Rogério Sarmento Leite, André Manica, Beatriz D. Schaan, P. Reed Larsen, and Ana Luiza Maia

Thyroid Unit (J.V., S.M.W., B.D.S., A.L.M.), Endocrine Division, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul, CEP 90620-000, Porto Alegre, RS, Brasil; Instituto de Cardiologia do RS/Fundação Universitária de Cardiologia (R.S.L., A.M.); and Division of Endocrinology, Diabetes, and Hypertension (P.R.L.), Brigham and Women´s Hospital and Harvard Medical School, Boston, Massachusetts 02115

Context: The acute phase of the nonthyroidal illness syndrome (NTIS) is characterized by low T_3 and high rT_3 levels, affecting up to 75% of critically ill patients. Oxidative stress has been implicated as a causative factor of the disturbed peripheral thyroid hormone metabolism.

Objective: The objective of the study was to investigate whether N-acetylcysteine (NAC), a potent intracellular antioxidant, can prevent NTIS in patients with acute myocardial infarction.

Design: This was a randomized, multicenter clinical trial.

Settings: Consecutive patients admitted to the emergency and intensive care units of two tertiary hospitals in southern Brazil were recruited. Patients and intervention included 67 patients were randomized to receive NAC or placebo during 48 hours. Baseline characteristics and blood samples for thyroid hormones and oxidative parameters were collected.

Main Outcome: Variation of serum T_3 and rT_3 levels was measured.

Results: Baseline characteristics were similar between groups (all $P > .05$). T_3 levels decreased in the placebo group at 12 hours of follow-up ($P = .002$) but not in NAC-treated patients ($P = .10$). Baseline rT_3 levels were elevated in both groups and decreased over the initial 48 hours in the NAC-treated patients ($P = .003$) but not in the control group ($P = .75$). The free T_4 and TSH levels were virtually identical between the groups throughout the study period ($P > .05$). Measurement of total antioxidant status and total carbonyl content demonstrated that oxidative balance was deranged in acute myocardial infarction patients, whereas NAC corrected these alterations ($P < .001$).

Conclusions: NAC administration prevents the derangement in thyroid hormone concentrations commonly occurring in the acute phase of acute myocardial infarction, indicating that oxidative stress is involved in the NTIS pathophysiology. **(*J Clin Endocrinol Metab* 99: 4537–4545, 2014)**

Nonthyroidal illness syndrome (NTIS) refers to characteristic changes in thyroid hormone levels that occur during acute and chronic severe illnesses. The acute phase of the syndrome is characterized by low serum total T_3 and free T_3, as well as high rT_3 concentrations. Serum T_4 may be normal or reduced (1). Although serum TSH remains in the normal range, the nocturnal surge of TSH observed in the normal physiological state is absent (2). In the acute phase of illness, the alterations occur primarily in the peripheral metabolism of thyroid hormones, whereas neuroendocrine abnormalities predominate in prolonged illness (1).

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Abbreviations: AMI, acute myocardial infarction; D, deiodinase; FT4, free T_4; GSH, glutathione; ICU, intensive care unit; NAC, N-acetylcysteine; NTIS, nonthyroidal illness syndrome; ROS, reactive oxygen species; TAS, total antioxidant; TIMI, Thrombolysis In Myocardial Infarction.

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NTIS occurs in 30%–90% of patients with acute myocardial infarction (AMI) (3, 4). Low serum T_3 levels are an independent marker of myocardial damage and poor prognosis during this clinical situation and are associated with increased morbidity and mortality in the short, medium, and long term (4–6). It has been postulated that low T_3 levels in myocardial tissue can produce a state of local hypothyroidism, worsening the tissue damage and cardiac disease (4).

The pathophysiology of NTIS is complex. One of the putative mechanisms for changes in the thyroid hormone levels are derangements in iodothyronine deiodinases function (D1, D2, and D3) (7, 8). These selenoenzymes are a family of oxidoreductases that catalyze peripheral iodothyrosine deiodination. D1 and D2 convert T_4 to the active hormone T_3, whereas it is the source of 80% of the peripheral levels of T_3 (8). D3 inactivates both T_4 and T_3. All three deiodinases require an as-yet-undefined cofactor, probably a thiol or thiol-dependent compound, which acts as a reducing agent releasing iodine from the selenocysteine residue and regenerating the active enzyme (8). It has been recently shown in a cell culture system that changes in the intracellular redox state can impair the thyroid hormone economy by altering the peripheral T_3/T_4 activation/inactivation process (9). These alterations were prevented by the addition to the media of N-acetylcysteine (NAC), an antioxidant that increases the intracellular cysteine and reduced the glutathione (GSH) levels, thus restoring the redox equilibrium. NAC reestablished the activity of the deiodinases by restoring the intracellular cysteine levels and/or replenishing the enzyme thiol cofactor, perhaps GSH.

In the current study, we investigated whether early iv NAC administration would prevent the changes in thyroid hormone levels that are observed in patients with AMI.

Subjects and Methods

Eligibility and study design

This was a randomized, prospective, multicenter study (Clinical Trials number NCT01501110) to evaluate whether NAC can prevent the thyroid hormone changes as seen in NTIS after an AMI. Consecutive patients admitted to the emergency and intensive care units of two tertiary hospitals in southern Brazil were recruited. Patients with a diagnosis of AMI within 12 hours of evolution who underwent primary percutaneous coronary intervention were eligible. Myocardial infarction was defined by persistent new electrocardiographic ST elevation at the J point in at least in two contiguous leads of 2.0 mm (men) or 1.5 mm (women) with or without Q-wave formation and subsequent release of biomarkers of myocardial necrosis (10). Patients who met the following criteria were excluded: 1) age younger than 18 years or older than 80 years; 2) history of primary thyroid disease; 3) chronic use of corticosteroids; 4) chronic renal failure requiring hemodialysis; 5) severe hepatic insufficiency, class C category in Child-Pugh score, or a score greater than 15 in the model for end-stage liver disease; 6) severe immunosuppression, defined as marrow or solid organ transplantation, severe leukopenia (white blood cell count $< 1000/mm^3$), hematological malignancy, and immunodeficiency syndromes; and 7) pregnant women or women undergoing postmenopausal hormone replacement. Ten healthy subjects were included for baseline comparison of rT3 levels and oxidative stress and IL-6 measurements.

The Institutional Ethics Committees approved the study protocol and a written informed consent was obtained from all patients before randomization.

Study protocol

Patients were randomly assigned to the use of NAC or placebo on the basis of a random number generator. Patients selected for the intervention group received five doses of 1200 mg (12 mL) of NAC by iv route, the first dose being administered prior to cardiac catheterization and every 12 for 48 hours after the procedure (total dose 6000 mg) (10, 11). Patients randomized to the control group received 0.9% normal saline iv (12 mL), every 12 hours, for 48 hours. Laboratory assessment was performed at baseline (time 0) and at 6, 12, 24, 48, and 120 hours after the first NAC administration. All other aspects of patient care were carried out at the discretion of the treating clinicians.

Participants were recruited during the period of July 2011 to September 2012. Eighty-three patients were assessed for eligibility. Thirteen patients were excluded: four patients had renal failure, seven had thyroid disease, two were receiving immunosuppression therapy, and two patients did not provide written consent. Thus, 68 patients participated in the study (Figure 1).

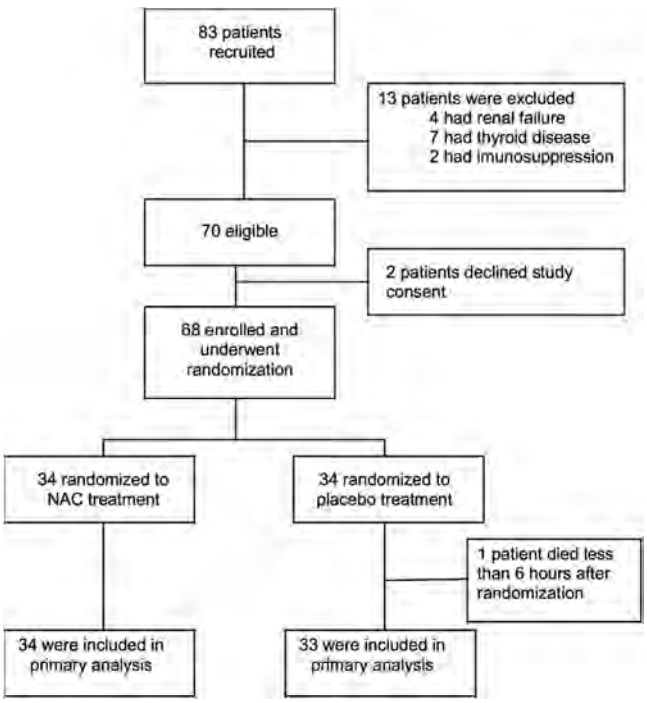

Figure 1. Consolidated Standards of Reporting Trials diagram depicting subject flow.

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Laboratory measurements

Venous blood samples were obtained at admission (baseline), after 6, 12, and 48 hours, and on the fifth day or at the time of hospital discharge if this occurs before the fifth day. Thyroid hormones and oxidative stress parameters were evaluated.

Assays were performed in duplicate on batched serum samples that had been stored at −20°C, pending study completion. Serum total T_3, T_4, free T_4 (FT4), and TSH were measured by electrochemiluminescent immunoassay (ADVIA Centaur XP; Siemens). Serum rT_3 levels were measured by an ELISA kit (USCN Life Science Inc). The interassay coefficients of variation were as follows: T_3, 7.1%; T_4, 10%; FT4, 7%; and rT_3, less than 10%. The intraassay coefficients of variation were as follows: T_3, 6.2%–7%; T_4, 3%–8%; FT4, 3%–6%; and rT_3, 6%–8%. Normal ranges were as follows: serum FT4, 0.7–1.5 ng/dL; T_4, 4.5–12 ng/dL; T_3, 77–180 ng/dL; and rT_3, 10–24 ng/dL.

Total antioxidant capacity (TAS) and carbonyl content and were measured in serum for assessment of oxidative stress and response to treatment with NAC. The quantitative determination of the total antioxidant status (TAS) was determined using the TAS kit (Randox, UK). ABTS 92,2′-azino diethyl-benzothiazoline sulfonic acid) was incubated with a peroxidase and H_2O_2 to generate the cation 2,2′-azino diethyl-benzothiazoline sulfonic acid, a relatively stable blue-green compound measurable at 600 nm. The antioxidants present in the sample inhibit this reaction, producing a decrease in the color intensity, which is proportional to the total antioxidant concentration (12). Samples were run in triplicate and were expressed as percentage variation in the color intensity compared with control. For carbonyl measurement, duplicate aliquots of plasma (containing ~0.3 mg of protein) were incubated with 500 μL of 10 mM 2.4-dinitrophenylhydrazine or 1.0 mL of 2 M HCl (blank tube). After 30 minutes, 250 μL of 50% trichloroacetic acid was added. The samples were centrifuged at 8000 × *g* for 30 minutes to obtain the protein pellets, which were immediately washed with ethanol-ethyl acetate 1:1 (vol/vol). The final protein pellets were diluted in 500 μL of 8 M urea buffer and incubated at 50°C for 90 minutes. The difference between the 2.4-dinitrophenylhydrazine -treated and HCl-treated samples (blank) was used to calculate the carbonyl content determined at 370 nm. Carbonyl (CO) content was calculated using the millimolar absorption coefficient of hydrazone (e370 nm = 2.1×10^4 M^{-1}/cm^{-1}), and the results were expressed in nanomoles of carbonyl per milligram protein measured by the Bradford method.

IL-6 measurements were performed according to the manufacturer's instructions on a Luminex 200 System (Luminex Corp). The Luminex protocol is a sandwich immunoassay system using magnetic beads. This method allows the detection of cytokines in a dual-laser flow analyzer. Human cytokine analysis kits were custom ordered (Invitrogen by Life Science) and included all necessary reagents for analysis. Briefly, cytokine antibody-conjugated beads were added to each well of a flat-bottom, 96-well plate. Plasma samples were thawed completely at room temperature, mixed well by vortexing, and centrifuged to remove precipitated material. Serum samples were diluted 1:2 with the provided diluents and pipetted into the wells, incubated, and washed appropriately. After final incubation and washing, fluorochrome bound to magnetic beads was quantified by the Luminex-200, calibrated using calibration microspheres. The median fluorescence intensity of fluorochrome-conjugated antibody bound to individual microspheres was derived from flow analysis of 50 microspheres per region. Cytokine quantification was plotted via standard curves. The intensity of the fluorescence was directly proportional to the concentration of cytokine. Quality control was performed between the plates by using the controls provided in the kit. The assay was completed on the same day by the same person. The plate variation was 6.94% in this assay. IL-6 levels were calculated using the xPonent version 3.1 software package (Luminex Corp) and expressed as nanograms per liter. The detection limit for IL-6 measurement was 0.3 pg/mL.

Outcome measures

The primary outcome measure was the change in plasma thyroid hormones. A secondary measure was the reduction in markers of oxidative stress.

Statistical analysis

The study was originally designed to enroll 68 patients. On the basis of previous data (13), the sample size was calculated to provide a statistical power of 90% to detect an absolute difference between the 2 groups of 20% in T_3 levels (assuming a two sided level $\alpha < .05$) using EpiInfo StatCalc (version 7.1.3). Categorical data are presented as frequencies and their differences were analyzed using the χ^2 or Fisher's exact test. Quantitative data with normal distribution are presented as mean ± SD, and their differences were analyzed using the Student's *t* test. Nonparametric variables are presented as median ± interquartile range and analyzed by Mann-Whitney's *U* test or repeated-measures ANOVA. Outcomes were analyzed according to the intention-to-treat principle. Within each group, changes (δ) in oxidative markers were calculated by subtracting the baseline values from the values measured after the intervention. Between-group differences were calculated by subtracting the change observed in the NAC group from the change observed in the placebo group. A value of $P < .05$ was considered significant. The analyses were performed by PASW statistics version 18.0.

Results

Patients

Sixty-eight patients were randomly assigned to receive NAC or placebo. One patient in the placebo group died during the initial evaluation. Therefore, the study data included 67 patients (Figure 1). The mean age was 57.2 ± 9 years, and 78% of the patients were male. The mean time from symptom onset was 6.3 ± 3 hours, and most patients were classified as low risk, according to the Thrombolysis In Myocardial Infarction (TIMI) risk score (ST elevation ≤ 3 in 53% of patients). There were no significant differences between the groups with respect to any of the characteristics listed (Table 1).

NAC prevents the decrease in serum T_3 and promotes decreases in rT_3 levels

In the placebo group, we observed a decrease in serum T_3 at 6 hours (98.7–88.6 ng/dL; $P = .001$) and 12 hours (98.7–86.8 ng/dL; $P = .001$, Figure 2A). Serum T_3 re-

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Table 1. Clinical and Baseline Laboratory Characteristics of the Study Population

	NAC (n = 34)	Placebo (n = 33)	*P* Value
Age, y	56.9 ± 9.4	57.4 ± 8.4	.80
Sex, % male	80	75	.56
Body mass index, kg/m^2	26. 8 ± 4.4	27.1 ± 3.5	.88
Hypertension, %	66	64	.60
Diabetes, %	39	32	.61
Heart failure, %	6.6	9	.67
Angina pectoris, %	50	42	.62
Time of presentation, h	5.7 ± 2.9	6.5 ± 2.9	.29
Troponin T, pg/mL	3029.8 ± 3122.3	3139.7 ± 2368.3	.87
KILLIP 1–2, %	100	93	.27
TIMI 0–3, %	53	53	.99
Median arterial pressure, mm Hg	96.6	95.2	.76
TSH, μIU/mL	1.7 ± 1.2	1.6 ± 1.1	.82
T_4, μg/dL	7.4 ± 1.9	7.4 ± 1.4	.21
T_3, ng/dL	100.4 ± 16.5	98.7 ± 20.7	.70
FT4, ng/dL	1.1 ± 0.2	1.1 ± 0.3	.82
rT_3, ng/dL	54.7 ± 8.11	55.2 ± 6.1	.57

Reference ranges are as follows: serum FT4, 0.7–1.5 ng/dL; T_4, 4.5–12 ng/dL; T_3, 77–180 ng/dL; and rT_3, 10–24 ng/dL.

turned to baseline levels at 48 hours (96.5 ng/dL) and did not change between 48 and 120 hours (96.5 to 94.2 ng/dL, $P > .05$). In the group treated with NAC, there was no significant change in serum T_3 levels (100.4 to 94.2 to 97 to 93.5 to 92.0 ng/dL; $P > .05$; Figure 2A). Compared with the NAC-treated patients, the serum T_3 levels in the placebo group were lower at 6 hours (94.2 vs 88.6 ng/dL $P < .001$) and 12 hours (97 vs 86.8 ng/dL; $P < .001$; Figure 2A). Serum T_3 was similar between groups at 48 hours and on the fifth day of follow-up.

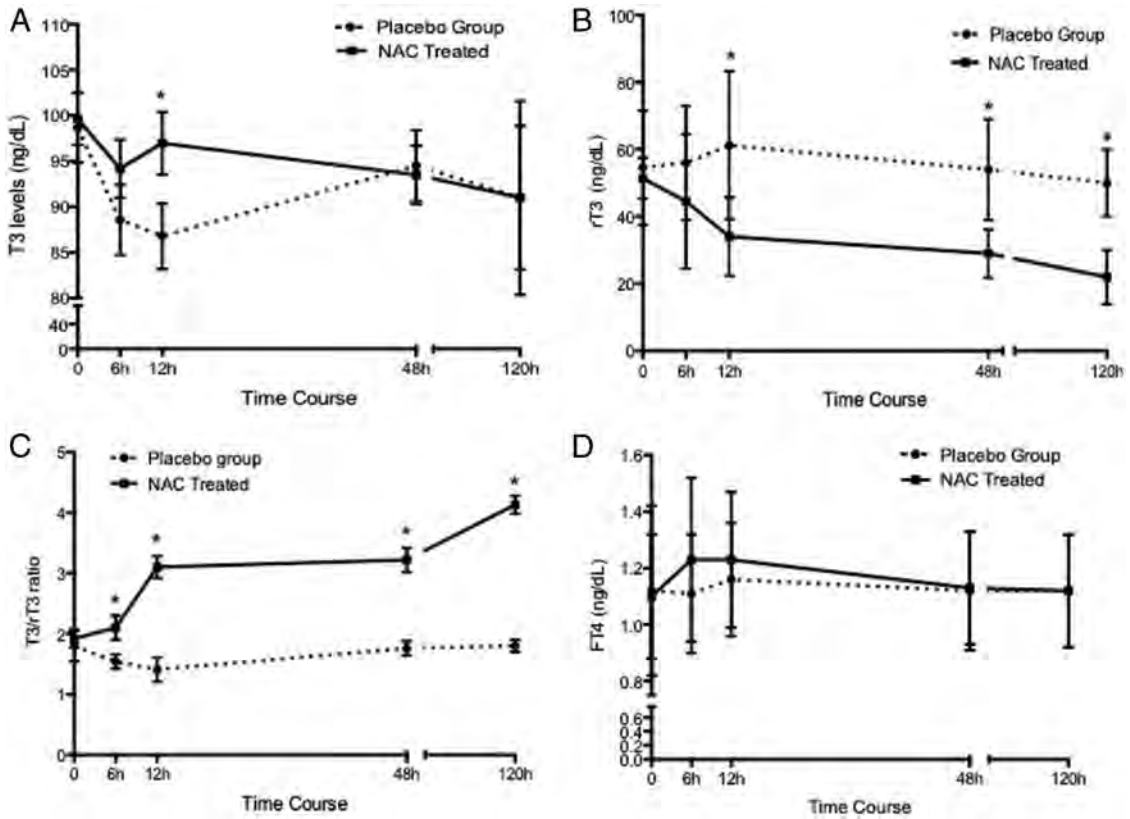

Figure 2. Changes in thyroid hormone levels in patients with acute myocardial infarction who received NAC treatment or placebo. Serum T_3 levels decreases at 6 and 12 hours in the placebo group but not in patients who received NAC (A). No significant changes were observed in serum rT_3 levels in the placebo group, whereas rT_3 decreased in a time-dependent fashion in NAC-treated patients (B). The T_3 to rT_3 ratio increases progressively in NAC-treated patients, whereas it remains stable in the placebo group (C). Neither group showed significant changes in serum FT4 levels (D). *, $P < .001$.

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Considering that the decreases in serum T_3 correlate with disease severity, we sought also to investigate the NAC effect on a subgroup of sickest patients. Thus, patients were grouped as moderate to high (TIMI score ≥ 4) or low (TIMI score ≤ 3) risk. Indeed, the average decrease in serum T_3 levels at 12 hours in the moderate- to high-risk placebo subgroup (n = 16) was more pronounced as compared with the low-risk subgroup (n = 17; 19.9 vs 10.9%; 99.3 to 75.5 vs 98.1 to 88.2 ng/dL; $P = .003$). Importantly, in the subgroup of moderate- to high-risk NAC-treated patients (n = 16), there was no significant changes in serum T_3 levels (103.9 to 92.1 to 93.8 to 85.6 to 88.5 ng/dL, $P = .537$), demonstrating that NAC administration was able to prevent the T_3 decreases.

The baseline serum rT_3 levels were 3- to 4-fold elevated in both groups, relative to healthy subjects (14.2 vs 55.2 vs 52.7 ng/dL; $P < .001$; Figure 2B). No significant changes were observed in the serum rT_3 levels in the placebo group during the follow-up period (55.2 to 57.5 to 58.4 to 48.5 to 50 ng/dL; $P = .75$; Figure 2B). In contrast, in the group treated with NAC, serum rT_3 levels decreased in a time-dependent fashion, reaching the lowest value at 120 hours (52.7 to 43.8 to 33.7 to 29 to 22 ng/dL; $P = .003$; Figure 2B). Accordingly, as compared with the placebo group, serum rT_3 levels were lower in NAC-treated patients at 12 (50.7 vs 33.7 ng/dL, $P = .003$) and 48 hours (48.5 vs 29 ng/dL, $P = .05$).

Accordingly, although the T_3 to rT_3 ratio remained low and stable in the placebo group (1.8 to 1.5 to 1.4 to 1.7 to 1.8; $P = .6$; Figure 2C), it increased over time in the NAC-treated patients (1.9 to 2.1 to 3.1 to 3.2 to 4.1; $P < .001$; Figure 2C). Compared with the NAC-treated patients, the T_3 to rT_3 ratio in the placebo group were lower at all times evaluated ($P < .001$).

No significant changes were observed in serum FT4 levels in the placebo group throughout the follow-up period (1.12 to 1.11 to 1.16 to 1.12 ng/dL, at baseline, 6, 12, and 48 h, respectively; $P = .84$; Figure 2D). Interestingly, although not statistically significant upon post hoc testing, we observed an increase at 6 and 12 hours in serum FT4 in patients who received NAC (1.10 to 1.23 to 1.23 to 1.13 ng/dL; $P = .06$; Figure 2D). There were no differences in serum FT4 levels between the groups ($P = .11$).

NAC treatment does not alter the pituitary-thyroid feedback mechanism

In the placebo group, we observed a progressive increase in the mean serum TSH levels during the follow-up period (1.6 to 2.5 to 3.9 μIU/mL; at baseline, 48 h, and on the fifth day, respectively; $P < .001$). Similar increases in serum TSH were observed in patients who received NAC (1.7 to 2.3 to 4.0 μIU/mL; at baseline, 48 h, and on the fifth day, respectively; $P < .001$). Serum TSH values were virtually identical between the groups ($P = .82$).

NAC administration normalizes the oxidative parameters in patients with AMI

Next, we evaluated the oxidative status of the patients. As compared with healthy subjects, the initial total amount of intra- and extracellular antioxidant molecules (TAS) was reduced in placebo and NAC-treated groups (2.90 vs 1.75 vs 1.64 mmol/mg · protein respectively, $P = < .002$; Figure 3A). No significant changes occurred in serum TAS levels in the placebo group (1.75 to 1.78 to 1.70 mmol/mg · protein, $P = .75$; Figure 3A). In contrast, in the NAC group, the serum TAS increased significantly, reaching the levels observed in healthy controls (1.64 to 2.93 to 2.5 mmol/mg · protein, $P < .001$; Figure 3A). The TAS concentrations were significantly higher in the NAC-treated patients at 6 hours (1.78 vs 2.93 mmol/mg · protein; $P = .008$) and 12 hours (1.70 vs 2.50 mmol/mg · protein; $P = .03$) of follow-up.

We also measured total carbonyl content, a parameter of protein oxidation. This was initially elevated in placebo and NAC groups, as compared with healthy subjects (0.46 vs 1.32 vs 1.4 nmol/mg · protein, respectively, $P < .001$) but did not differ between the placebo and NAC groups (Figure 3B). In the placebo group, no significant changes were observed in the total carbonyl content during the observational period (1.32 to 1.31 to 1.51 to 0.85 nmol/mg · protein, at baseline, 6 h, 12 h, and 48 h, respectively; $P = .11$; Figure 3B). However, in patients receiving NAC, the total carbonyl content decreased significantly, reaching normal levels by 48 hours (1.4 to 0.6 to 0.8 to 0.5 nmol/mg · protein, at baseline, 6 h, 12 h, and 48 h, respectively; $P < .001$; Figure 3B). The total carbonyl content was lower all times after initiating NAC treatment (Figure 3B).

NAC administration does not alter the acute phase response in AMI patients

Next, we measured the serum IL-6 levels to evaluate the NAC effect in the acute phase response of AMI patients. The baseline serum IL-6 levels were elevated in both groups, as compared with healthy subjects (3.4 vs 49.3 vs 48.8 ng/L, respectively, $P < .001$). We observed a progressive increase in IL-6 levels from baseline to 12 hours in both placebo and NAC-treated groups (49.3 to 50.1 to 74 ng/L, $P = .02$; 48.8 to 58.6 to 81 ng/L, $P < .02$, respectively, Figure 4). In both groups, a subsequent decrease in mean serum IL-6 levels occurred at 48 hours and continued to drop reaching the baseline values at 120 hours (49.3 to 50.1 to 74 to 42.8 to 31 and 48.8 to 58.5 to 81 to 58.3 to 40 ng/L, respectively, Figure 4). No significant differ-

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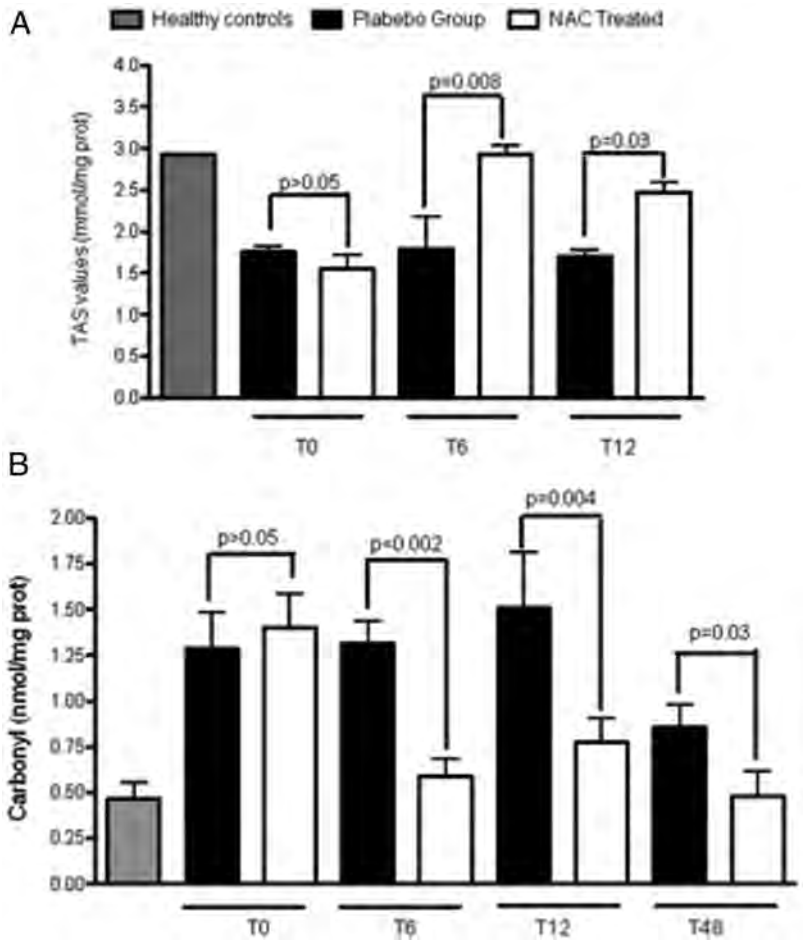

Figure 3. Oxidative stress biomarkers in patients with AMI who received NAC treatment or placebo. No significant changes occurred in serum TAS levels in the placebo group, whereas in the NAC group, TAS increased significantly, reaching the levels observed in healthy individuals (A). No significant changes were observed in the total carbonyl content in the placebo group, whereas the total carbonyl content decreased significantly and reached the levels observed in healthy subjects in NAC-treated patients (B).

ences were observed between the groups ($P = .6$). Interestingly, however, the IL-6 levels were inversely correlated with T_3 levels in the placebo group ($r = -054$, $P = .001$) but not in the NAC-treated patients ($r = -0.04$, $P = .86$).

Adverse events and safety

The average length of hospital stay was 4.94 days in the placebo group and 4.24 days in the NAC-treated patients ($P = .13$). Two patients in the control group but none in the NAC group required vasopressors. There were no deaths during the follow-up period.

Discussion

The NTIS affects approximately 70% of patients with AMI and the serum T_3 concentration correlates inversely with mortality (3, 5). Increased reactive oxygen species (ROS) generation, as observed in many diseases, disrupts deiodinase function and may well play a central role in the derangement of peripheral thyroid hormone metabolism. Here we have demonstrated that the administration of NAC, a potent intracellular antioxidant, prevents the characteristic changes in serum T_3 and rT_3 during NTIS by the correction of the oxidative stress imbalance.

NTIS encompasses a number of changes in thyroid hormone physiology. The most striking alterations are the decreases in serum T_3 and increases in rT_3 levels observed in a variety of illness situations (14). These alterations are observed in the first hours of the disease and are among the last to recover (1, 15). The degree of reduction in thyroid hormone levels in sick patients is correlated with prognosis and survival (3, 16). A prospective observational study involving 480 unselected intensive care unit (ICU) patients has demonstrated that free T_3 was the only independent predictor of ICU mortality (16).

In the present study, we planned to determine whether NAC administration could prevent the characteristic thyroid hormonal changes seen in individuals with AMI. We show that patients who received NAC virtually eliminated the decrease in serum T_3 levels observed in the placebo group. Moreover, in the NAC-treated group, we also observed a prevention of the increase in serum rT_3, which have declined to nearly normal levels (Figure 2). Both the fall in T_3 and the increase in rT_3, which occur uniformly in sick patients as a mark of NTIS, can be attributed to changes in the peripheral metabolism of thyroid hormones. Indeed, early reports have demonstrated that decreased T_3 production and increased rT_3 concentrations developed rapidly in AMI patients (17), whereas studies performed in critically ill patients demonstrated a down-regulation of the hepatic D1 activity as well as an induction in liver and skeletal muscle D3 activity (18).

One puzzling observation in this study was the persistence of high rT3 levels in the placebo group, notwithstanding there was a near normalization of T_3 levels starting at 48 hours (Figure 2, A and B). There are several

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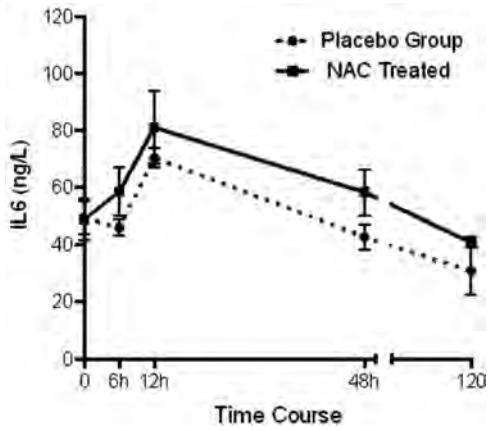

Figure 4. Serum IL-6 levels in patients with AMI who received NAC treatment or placebo. In both groups, the levels of IL-6 increases from baseline to 12 hours. A subsequent decrease in occurred at 48 hours and continued to drop, reaching the baseline values at 120 hours. No significant differences were observed in serum IL-6 levels between the groups.

potential possibilities for this finding. The D2 enzyme catalyzes only outer-ring deiodination of T_4 to T_3, but D1 deiodinates T_4 approximately equally at the outer and inner ring in vitro (19). On the other hand, rT_3 is favored as a substrate for D1, and an increase in its activity will accelerate rT_3 clearance. Assuming a recovery of activity of both D1 and D2, this will increase the T_3 to rT_3 ratio. On the other hand, D3 activity might also be increased in liver, which would generate more rT_3 than degrade T_3. The balance between all of these interweaving iodothyronine degradation pathways in vivo is admittedly difficult to predict, but the best index of the whole process should be the ratio of circulating T_3 to rT_3. Indeed, the T_3 to rT_3 ratio in the placebo group was virtually identical at 12, 48, and 120 hours and remains much lower than in the NAC patients, demonstrating that the peripheral hormone ratio abnormalities are still not better without NAC out to 120 hours (Figure 2C). Of interest, recent data from animal models also showed high D3 levels associated with increased cardiomyocyte thyroid hormone inactivation after myocardial infarction, but this is likely to have a greater effect on the local myocardial intracellular T_3 concentration than that in the circulation (20).

Due to the importance of differentiating between reduction of T_4 transport into cells to be activated into T_3 vs a derangement in the T_4-to T_3 activation process, we also evaluated serum FT4 concentrations in these patients. No significant differences in serum FT4 levels were observed between the groups, although we have detected a slightly increase FT4 in patients who received NAC. These observations support the idea that the most probable mechanism responsible for the acute NTIS is indeed the impaired peripheral T_4-to-T_3 conversion and rT_3 clearance and not a reduction in T_4 availability (21). Consistent with the concept that the regulation of neuroendocrine feedback is a marker of recovery from acute illness, although not suppressed on admission, the progressive increase in TSH from the baseline to the fifth day of follow-up indicates that the predominant derangement in this moderately stressful illness is in peripheral thyroid hormone metabolism. The contrast used for the coronary angiography procedure did not appear to have a major effect, although it was received by both groups (22).

Oxidative stress, due to augmented ROS or reactive nitrogen species generation is observed in many diseases that are associated with NTIS (23). Patients usually have reduced plasma and intracellular levels of antioxidant scavenging molecules, including GSH, as well as decreased activity of the antioxidant enzymatic system involved in ROS detoxification (24). We have recently shown that the changes in the intracellular redox state, as observed in critically ill patients, reduces the serum T_3 to T_4 ratio due to the inhibition of deiodinase function, reducing D1- and D2-mediated T_4-to-T_3 conversion as well as increasing D3-mediated T_3 (and T_4) inactivation, thus mimicking events during illness (9).

To further demonstrate the correlation between the derangement in the redox status and changes in thyroid hormone levels, we measured biomarkers of oxidative stress. In addition, we measured the levels of IL-6, to evaluate the NAC effect on the overall acute-phase response in AMI patients. As expected, total carbonyl content was elevated and TAS was diminished at baseline, relative to normals, in all patients. However, those patients who received NAC had a reduction in the carbonyl content and return of TAS to levels in healthy volunteers. No differences were observed in serum IL-6 values between the placebo and NAC-treated groups (Figure 4). Taken together, these results demonstrate that oxidative stress occurred early in the course of AMI, and the administration of NAC was able to protect proteins from oxidative damage as well as correct the impaired total body antioxidant capacity. Notably, these results were paralleled, in a timely fashion, with serum thyroid hormone adjustment, suggesting that the T_3 decrease is mainly due to oxidative stress-induced impairment of thyroid hormone activation (9). Although a potential effect of the cellular redox imbalance on T_4 transport into cells could also contribute to the effect, previous studies in critically ill patients did not show a correlation between monocarboxylate transporter 8 expression and the ratio of the serum to tissue concentration of the different iodothyronines (25, 26).

There are several mechanisms by which low thyroid function might alter myocardial cells. Apart from the tissue consequences of decreased cardiac energetic efficiency, a low serum T_3 may alter cardiac contractility and

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heart rhythm. More recently reduced T_3 has been implicated in the process of myocardial hypertrophy and fibrosis as well as in the altered vasoactive properties of vessels (27, 28). Despite the importance of thyroid hormones homeostasis for cardiac function and the poor prognosis associated with NTIS in patients with ischemic heart disease, a treatment for NTIS has never been tested in patients with AMI. Long-term thyroid hormone administration has been shown to improve chronic cardiac function in post-myocardial infarction heart failure in rats (29, 30), but experience with hormone replacement in humans is limited. Studies in patients undergoing cardiac surgery and in patients with severe congestive heart failure have demonstrated hemodynamic benefit, with a reduced need for inotropic agents (31–33). The lack of benefit in the long-term outcomes and concerns about adverse effects, such as increased myocardial demand, arrhythmias and suppression of the hypothalamic-pituitary-thyroid axis, have reduced enthusiasm for T_3 administration in acute settings.

This study was limited to the assessment to the effect of NAC on the thyroid hormone economy during illness and not designed to evaluate the cardiac response to the use of NAC or whether NAC administration had effects on patient outcome. Nevertheless, it is worth mentioning that in vitro studies have shown that ROS mediates myocardial injury secondary to ischemia and reperfusion and activates fibrogenic pathways that favor adverse ventricular remodeling and cardiac dysfunction (34–37). However, antioxidant therapy for ischemic cardiovascular disease has produced controversial results to date. Arstall et al (38) showed that adjunctive therapy with NAC reduces oxidative stress markers and improved left ventricular function. In contrast, other studies failed to demonstrate clinical benefit with respect to myocardial reperfusion injury, despite the NAC-induced decreases in oxidative stress markers (10).

In conclusion, we demonstrate that NAC administration will rapidly reverse the acute derangements in thyroid hormone levels produced by the oxidative stress in patients with acute myocardial infarction. These findings have potential clinical relevance because low T_3 concentrations are associated with a poor outcome in acute and long-term cardiovascular events.

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Address all correspondence and requests for reprints to: Ana Luiza Maia, MD, PhD, Serviço de Endocrinologia, Hospital de Clínicas de Porto Alegre, Rua Ramiro Barcelos, 2350, CEP 90035-003 Porto Alegre, RS, Brasil. E-mail: almaia@ufrgs.br.

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