Effects of micronized fenofibrate and vitamin E on in vitro oxidation of lipoproteins in patients with type 1 diabetes mellitus

W Engelen¹, B Manuel-y-Keenoy², J Vertommen², I De Leeuw², L Van Gaal¹, ²

Summary

Objective: The primary objective was to compare the antioxidant activity of micronised fenofibrate 200 mg to 400 IU of vitamin E and placebo, on the LDL and VLDL particles of patients with type 1 diabetes. The secondary objective was to investigate the possible synergy between micronized fenofibrate and vitamin E and to compare the efficacy of these treatments on lipids.

Methods: A double-blind, placebo-controlled trial in which patients were randomised into three treatment groups after a wash-out period of 8 weeks: the placebo group (Pla/Pla-group) in which patients received placebo during two consecutive periods of 8 weeks, the vitamin E group (Vit E/Vit E-group) in which patients received vitamin E during two consecutive periods, and the fenofibrate/vitamin E group (Fen/Fen + Vit E-group) in which patients received fenofibrate during the first period, followed by fenofibrate and vitamin E during the consecutive period. Blood samples taken at each visit, were analysed for routine biochemistry, blood lipids and copper mediated lipid peroxidation in vitro.

Results: The lag time of the non-HDL lipoprotein oxidation increased in the group given fenofibrate. The lag-time increased further when fenofibrate and vitamin E were given in association. (This reached significance in the intention-to-treat population, P = 0.034). The AUC of TBARS formation in the Vit E/Vit E group decreased after the first period, but this effect was not enhanced by continuing vitamin E for another 8 weeks. The AUC of TBARS formation did not change significantly after the administration of fenofibrate. Only after the second period, when vit E was given in association, the AUC of TBARS formation decreased significantly (P = 0.004). Fenofibrate caused a significant decrease in total and LDL-cholesterol and triglycerides (P < 0.05). In contrast, vitamin E had no effect on lipids.

Conclusions: The combination of micronized fenofibrate 200 mg/day and vitamin E 400 IU/day tended to increase the resistance of non-HDL lipoproteins to copper-mediated oxidation, shown by a prolongation of the lag-time. Vitamin E administration only, decreased the oxidation of non-HDL lipoproteins by a reduction of TBARS formation. This protective effect of vitamin E tended to be amplified by micronized fenofibrate.

Key-words: Fenofibrate · Vitamin E · Diabetes type 1 · Lipid peroxidation.

Effects du fénofibrate micronisé et de la vitamine E sur l’oxydation des lipoprotéines in vitro chez des diabétiques de type 1

Objectif: L’objectif primaire était de comparer l’activité anti-oxydante du fénofibrate micronisé 200 mg, de 400 UI de vitamine E et de placebo, sur des particules de LDL et de VLDL de diabétiques de type 1. L’objectif secondaire était d’étudier la possible synergie entre le fénofibrate micronisé et la vitamine E et de comparer l’efficacité de ces traitements sur les lipides.

Méthodes: Un essai en double insu, contrôlé par placebo, a été conduit dans lequel les patients ont été randomisés entre trois groupes thérapeutiques après une période de wash-out de 8 semaines: le groupe placebo (Pla/Pla) dans lequel les patients ont reçu un placebo pendant deux périodes consécutives de 8 semaines, le groupe vitamine E (Vit E/Vit E) dans lequel les patients ont reçu la vitamine E pendant deux périodes consécutives, et le groupe fénofibrate/vitamine E (Fen/Fen + Vit E) dans lequel les patients ont reçu le fénofibrate pendant la première période, puis fénofibrate et vitamine E pendant la seconde période. Les échantillons sanguins pris à chaque visite ont permis l’analyse de la biochimie standard, des lipides sanguins et de la peroxydation lipidique médiée par le cuivre in vitro.

Résultats: Le lag time de l’oxydation des lipoprotéines non-HDL a augmenté dans le groupe sous fénofibrate. Le lag-time a encore augmenté en cas de prise combinée de fénofibrate et de vitamine E. (Cela a atteint la signification statistique dans la population en intention de traiter, P = 0.004). L’aire sous la courbe de la formation des TBARS dans le groupe vit E/Vit E a diminué après la première période, mais cet effet n’a pas été renforcé par la poursuite de la vitamine E pendant une seconde période de 8 semaines. L’aire sous la courbe de la formation des TBARS n’a pas été significativement affectée par l’administration de fénofibrate. Ce n’est qu’après la seconde période, lorsque la vitamine E a été associée que l’aire sous la courbe de la formation des TBARS a diminué significativement (P = 0.004). Le fénofibrate a induit une diminution significative du cholestérol total et LDL et des triglycérides (P < 0.05). À l’inverse, la vitamine E n’a pas eu d’effets sur les lipides.

Conclusions: L’association de fénofibrate micronisé 200 mg/jour et de vitamine E 400 UI/jour tend à augmenter la résistance des lipoprotéines non-HDL à l’oxydation médiée par le cuivre, ce qui est illustré par une prolongation du lag-time. L’administration de vitamine E seul lement a permis de réduire l’oxydation des lipoprotéines non-HDL ce qui est mis en évidence par la formation de TBARS. Cet effet protecteur de la vitamine E tend à être amplifié par le fénofibrate micronisé.

Mots-clés: Fénofibrate · Vitamine E · Diabète de type 1 · Peroxydation lipidique.

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Received: July 12nd, 2004; revised: December 31st, 2004

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In diabetic patients, vascular complications are the main causes of morbidity and mortality and the risk of atherosclerosis is increased 2- to 4-fold. Clustering of classical cardiovascular risk factors, such as high cholesterol, arterial hypertension, positive family history, nicotine abuse and obesity only account for 25 to 50\% of this increased risk [1, 2]. Diabetes yields an additional burden that induces an accelerated progression of atherosclerosis. This burden can be ascribed to two main abnormalities, namely dyslipidemia and oxidative stress.

The metabolic changes, characteristic of the diabetic state, initiate dramatic alterations in lipids and lipoprotein profiles: chylomicrons and VLDL accumulate, thereby augmenting triglyceride levels. This hypertriglyceridemia leads to triglyceride enrichment of both HDL and LDL particles, thereby increasing the number of small, dense, highly atherogenic LDL particles and reducing the levels of HDL-cholesterol [3, 4].

The high oxidative stress found in diabetes, can be mainly ascribed to increased free radical production and defective antioxidant defences. Hyperglycemia, by enhancing auto-oxidation of glucose and non-enzymatic glycation, gives rise to an accelerated release of free radicals [5, 6]. Hyperglycemia also leads to changes in the levels of inflammatory mediators, abnormal activation of leukocytes and vascular dysfunction, which can cause ischemia-reperfusion and localised tissue destruction, contributing to the enhanced free radical release in diabetes mellitus [7].

An increased flux of glucose through the polyol-pathway leads to insufficient anti-oxidants through the following mechanisms: the increased flux of glucose through the polyol-pathway due to hyperglycemia, induces an increased consumption of NADPH during the reduction of glucose to sorbitol, and thus impairs the regeneration of natural anti-oxidants, such as vitamin E, vitamin C and glutathione. Oxidative and glyative damage to anti-oxidative enzymes and increased consumption of anti-oxidants, will additionally contribute to the decreased anti-oxidant defences in diabetes [8-12].

Any intervention which can improve these diabetes-related alterations, could attenuate LDL-oxidation and thereby contribute to the prevention or delay of atherosclerosis in these patients.

With respect to oxidative stress, vitamin E is probably one of the most studied anti-oxidants present in lipoproteins. Its potential role in the prevention of cardiovascular disease is supported by several epidemiological studies [13-15].

Regarding interventions aimed at correcting the dyslipidemia of diabetes, in particular hypertriglyceridemia, fibrates are the drugs of choice. Besides their triglyceride-lowering effects and the subsequent improvement of lipoprotein composition, fibrates may have additional anti-atherogenic properties [16, 17]. We could demonstrate that fenofibrate can enhance the resistance of LDL and VLDL to oxidation. In diabetic patients treated with fenofibrate, incubation of their LDL and VLDL with copper was followed by a significantly lower production of thiobarbituric acid reactive substances (TBARS), a marker of lipid peroxidation, and a longer lagtime phase which precedes the formation of peroxidation products [18]. The exact mechanisms behind this effect of fenofibrate on lipoprotein oxidation are not known. Micronised fenofibrate was chosen because of its enhanced absorption.

In the present study, the primary objective was to compare the anti-oxidant activity of micronised fenofibrate in its recommended dose of 200 mg/day to \( \alpha \)-tocopherol and placebo in type 1 diabetic patients. The evaluation of the efficacy of the treatment on lipids and the investigation of a synergy between fenofibrate and vitamin E were secondary study-objectives.

**Methodology**

**Patients and methods**

This study is a mono-centric, randomised, double blind, double-dummy, placebo-controlled, phase II trial. Non-smoking type 1 diabetics, aged 18 to 60 years, with a HbA1c < 10% and a BMI < 28 kg/m\(^2\) were randomly assigned to three different treatment groups. Exclusion criteria were: hypertension (systolic blood pressure > 140 mmHg or diastolic blood pressure > 90 mmHg after 5 minutes in sitting position), creatinine level \( \geq \) 15 mg/l or positive microalbuminuria, total cholesterol > 300 mg/dl or triglycerides > 500 mg/dl, pregnancy, breast feeding, women of child bearing age without adequate contraception, recent or unstable cardiovascular or cerebrovascular disease. Unauthorised medications including high dose contraceptive pills with oestradiol > 30 \( \mu \)g, hormone replacement therapy per os, ACE inhibitors with SH groups, \( \alpha \)- and \( \beta \)-blockers, vitamins, retinoids, ciclosporin, anti-vitamins K and lipid lowering drugs. All patients were consuming a standard diabetes diet. Dietary and lifestyle habits remained stable during the study period.
As shown in Figure 1, all patient groups started with an 8 weeks placebo run-in period (Period A). The first group (Vit E/Vit E) received 400 IU RRR α-tocopherol acetate once daily for 2 successive periods of 8 weeks each (Period B and C). The second group (Fen/Fen + Vit E) received micronized fenofibrate 200 mg for 8 weeks (Period B) and afterwards the combination of 400 IU RRR α-tocopherol acetate and 200 mg micronized fenofibrate for 8 weeks (Period C). The third group (Pla/Pla) received placebo for 16 weeks (Period B and C). The total trial time for all patients was 24 weeks. Visits to the polyclinic were scheduled every 8 ± 1 weeks.

Micronized fenofibrate was presented as an orange coloured capsule containing the active substance. α-tocopherol was presented as a soft transparent capsule, with a clear yellow content. The fenofibrate placebo and α-tocopherol placebo were presented in capsules, identical in appearance as the active medication. All medication has been supplied by Fournier Laboratories. Compliance was assessed by counting the number of returned capsules and empty blisters. Those patients taking less than 70% of the total prescribed number of capsules per period were to be considered non-compliant. Medication was supplied at visit 1, 2 and 3. At Visit 2, 3 and 4, patients were to return their treatment packs. The number of capsules in each returned pack was to be counted and the number of remaining capsules recorded in the drug accountability form. At each of the visits, the occurrence and nature of adverse events were recorded. Each adverse event was assessed for its severity on a three point ordinal scale (slight, moderate, severe) and for its relationship to the study drug on a four point ordinal scale (excluded, doubtful, possible, probable). For the statistical analysis only the treatment-emergent adverse event, with onset in periods B and C were considered.

The protocol was submitted to an independent ethics committee and its approval was obtained. Informed written consent was obtained from each patient before being enrolled in the study. The study was conducted in compliance with the requirements of the declaration adopted by the World Medical Assemblies and in compliance with the current Good Clinical Practices.

Analytical methods

Routine blood tests (blood count, urea nitrogen, creatinin, uric acid, glucose, liver enzymes, protein, albumin, total and HDL-cholesterol, triglycerides, sodium, potassium, calcium, C-reactive protein and alkaline phosphatase) were analysed in the routine laboratory of the clinic. Total analytical variability, expressed as coefficient of variation CV, was 2%, 1.9% and 0.9% for total cholesterol, HDL-cholesterol and triacylglycerol, respectively. LDL-cholesterol was calculated according to the Friedewald formula [19]. Metabolic control of the diabetic patients was evaluated by means of the glycated hemoglobin HbA1c, measured by the Modular Diabetic Monitoring System (BIO-RAD, California, USA) using a HPLC cation exchange column. The reference values are between 4.8 and 6%, CV is 1.5%. The susceptibility of low-density (LDL) and very low-density lipoproteins (VLDL) to copper catalysed oxidation was measured according to the method of A Zhang, as was previously described [20].

Statistical analysis

The three treatment groups were compared for the primary efficacy criterion and the secondary efficacy criteria at the end of each period (V3 and V4). For the analysis of the different description variables the chi-square test was used, or in case of expected frequencies < 5, the Fisher Exact test was used. For the continuous descriptive variables the comparison was performed by an one way analysis of variance (ANOVA). As far as the lipids are concerned, a one-way analysis of the difference of the results compared to visit 2 was performed and expressed as a ratio, for example (V3 - V2)/V2. Non-HDL oxidizability was analysed on the basis of AUC of the TBARS curve between 30 and 180 min. In case of a globally statistically significant difference, each treatment group was compared with the placebo group using Dunnett’s test. If any of the conditions for applying analysis of variance was not satisfied, the treatment groups were compared by a non-parametric method (Kruskal-Wallis). Exploratory analyses included a global repeated measures ANOVA and within groups paired — t tests on the ITT population.
Results

Study Population

Out of 65 patients screened, 44 patients (27 males, 17 females, aged 36.5 ± 9.8 years) were randomised in a double blind manner (intention to treat population — ITT). During the course of the study, 22 patients did not fully comply with all the protocol conditions (prohibited medication at inclusion, variables missing, non-compliance). The 22 patients who fully complied with all conditions composed the per protocol population (PP). Baseline characteristics are summarized in Table I.

Serum lipids

Total cholesterol changed from 201 ± 33 mg/dl at baseline to 163 ± 27 mg/dl after 8 weeks of treatment with fenofibrate (P < 0.05, Dunnett’s test vs placebo), and LDL-cholesterol changed from 127 ± 28 mg/dl to 96 ± 27 mg/dl (P < 0.05, Dunnett’s test vs placebo). Combination with vitamin E for a period of 8 weeks caused no further decrease in cholesterol, nor LDL-cholesterol levels. Total cholesterol and LDL-cholesterol did not change in the patients who received vitamin E (Tab II and III). HDL-cholesterol remained unchanged in all groups (Tab IV). Triglycerides declined from 82 ± 38 mg/dl at baseline, to 58 ± 23 mg/dl after 8 weeks of treatment with fenofibrate (P < 0.05, Dunnett’s test vs placebo). In the same patients, triglycerides were 63 ± 28 mg/dl after the second 8 weeks treatment period with fenofibrate and vitamin E given in association. Vitamin E given alone had no effect on triglycerides (Tab V). The differences found in the PP population reached similar statistical significances.

Lipoprotein Peroxidation

The lag time of the oxidation of non-HDL lipoproteins did not change statistically in the patients who received vitamin E (Tab II and III). HDL-cholesterol remained unchanged in all groups (Tab IV). Triglycerides declined from 82 ± 38 mg/dl at baseline, to 58 ± 23 mg/dl after 8 weeks of treatment with fenofibrate (P < 0.05, Dunnett’s test vs placebo). In the same patients, triglycerides were 63 ± 28 mg/dl after the second 8 weeks treatment period with fenofibrate and vitamin E given in association. Vitamin E given alone had no effect on triglycerides (Tab V). The differences found in the PP population reached similar statistical significances.

Table I

Baseline characteristics of the patient groups.

<table>
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<tr>
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<th>All Patients</th>
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<th>Pla/Pla</th>
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<tr>
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<td>ITT (N = 44)</td>
<td>PP (N = 22)</td>
<td>ITT (N = 14)</td>
<td>PP (N = 6)</td>
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<tr>
<td>Age (years)</td>
<td>35 (19 - 58)</td>
<td>39 (20 - 58)</td>
<td>34 (19 - 51)</td>
<td>35 (31 - 49)</td>
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<tr>
<td>Gender (n)</td>
<td>27/17</td>
<td>13/9</td>
<td>4/10</td>
<td>5/1</td>
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<td>BMI (kg/m²)</td>
<td>24.4 (2.2)</td>
<td>24.3 (2)</td>
<td>24.1 (2.3)</td>
<td>23.5 (2)</td>
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<td>HbA1c (%)</td>
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<td>7,5 (0,7)</td>
<td>7,6 (0,7)</td>
<td>7,9 (0,4)</td>
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<tr>
<td>Time since diagnosis</td>
<td>16 (9)</td>
<td>17 (10)</td>
<td>16 (11)</td>
<td>19 (11)</td>
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<tr>
<td>Systolic BP (mmHg)</td>
<td>126 (10)</td>
<td>131 (8)</td>
<td>127 (12)</td>
<td>134 (6)</td>
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<tr>
<td>Diastolic BP (mmHg)</td>
<td>78 (7)</td>
<td>79 (7)</td>
<td>79 (9)</td>
<td>84 (5)</td>
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</table>

ITT: Intention to treat Population; PP: Per Protocol Population. Results are expressed as mean (SD), as median (minimum-maximum) or as number of cases. *P < 0.05, One-way ANOVA.

Adverse events

Adverse events were reported for 7 patients (50%) in the Vit E/Vit E group, for 10 patients (62.5%) in the Fen/Fen + Vit E group and for 8 patients (57.1%) in the Pla/Pla group. The difference between the groups is not statistically significant (Fisher Exact test). None of the events were considered to be serious.
### Table II
Total cholesterol.

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<th>Vit E/Vit E</th>
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<td>Visit 3</td>
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<td>Visit 4</td>
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</table>

Results are expressed in mg/dl as mean (SD).

* Dunnett’s test for comparison of Fen/fen + Vit E vs Pla/Pla.

### Table III
LDL-cholesterol.

<table>
<thead>
<tr>
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<th>Vit E/Vit E</th>
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Results are expressed in mg/dl as mean (SD).

* Dunnett’s test for comparison of Fen/Fen + Vit E vs Pla/Pla.

### Table IV
HDL-cholesterol.

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Results are expressed in mg/dl as mean (SD).

### Table V
Triglycerides.

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</table>

Results are expressed in mg/dl as mean (SD).

* Dunnett’s test for comparison between Fen/Fen + Vit E vs Pla/Pla.
Discussion

After eight weeks of treatment with fenofibrate (200 mg/day), total cholesterol decreased by 18%, triglycerides by 26% and LDL-cholesterol decreased by 24%. HDL-cholesterol levels remained unchanged. Vitamin E given alone had no effect on serum lipids and the lipid lowering capacity of fenofibrate was not affected when vitamin E (400 IU/day) was given in association [21, 22].

Vitamin E administered in monotherapy, in its natural form (RRR-\(\alpha\)-tocopherol), at a dose of 400 IU/day, decreased the oxidizability of non-HDL lipoproteins, as expressed by the generation of TBARS, but did not affect resistance against oxidation since the lag time of the formation of fluorescent products remained unchanged at a dose of 400 IU/day. There is no general agreement on the optimal dose, and one could presume that the dose used, was not sufficient to increase the lag-time significantly in this group of type 1 diabetes mellitus patients. In a placebo-controlled study in 79 healthy volunteers, protection against copper catalysed LDL oxidation could only be evidenced at a dose of 400 IU/day RRR-\(\alpha\)-tocopherol and higher (doses of 100, 200, 400 and 800 IU/day were compared) [23]. Again in healthy subjects, supplementation with synthetic dl-\(\alpha\)-tocopherol at a dose of 1200 IU proved to be significantly more active in decreasing LDL-lipoprotein oxidative susceptibility than at doses of 400 IU/day [24]. Similarly to our study, the administration of the synthetic dl-\(\alpha\)-tocopherol isomer at a dose of 1200 IU proved to be significantly more active in decreasing LDL-lipoprotein oxidative susceptibility than at doses of 400 IU/day [24]. Similarly to our study, the administration of the synthetic dl-\(\alpha\)-tocopherol isomer at 400 IU/day for eight weeks in healthy men was not associated with a statistically significant increase in lag time. In contrast, this dose was sufficient to induce a decrease in TBARS formation [25]. However, in long term observational studies, the supplemental intake of 100 IU/day seemed to be the threshold intake of vitamin E to decrease cardiovascular morbidity and mortality. The prospective Nurses’ Health Study showed a 40% reduction of coronary heart diseases in middle-aged women, without cardiovascular history nor cancer, who used vitamin E supplements for at least 2 years [26]. A comparable reduction was demonstrated in middle-aged men using vitamin E supplements in the Health Professionals Follow Up Study [27]. Nevertheless, the results of intervention studies remain controversial, as proven by the Heart Outcomes Prevention Evaluation (HOPE) Study, where 9237 women and men, aged 55 years or more and at high risk for cardiovascular events were randomly assigned to receive 400 IU/day of vitamin E from natural sources or placebo for a mean of 4.5 years, without apparent effect on cardiovascular outcome [28]. Probably diabetic patients require higher doses of vitamin E than non-diabetic subjects in order to achieve a protective effect against LDL oxidation. In a double blind placebo controlled study, RRR-\(\alpha\)-tocopherol 400 IU/day given for eight weeks did not increase the lag time in normolipidemic diabetic patients with type 1 diabetes, whereas it did in matched non-diabetic controls [29].

Fenofibrate treatment alone was not significantly associated with an increase in resistance of non-HDL lipoproteins to oxidation. This is probably due to a lack of statistical power since the lag time clearly tended to increase. When vitamin E was associated with fenofibrate, a further increase in lag time was observed suggesting that the possible effect of fenofibrate therapy could be amplified by vitamin E. Accordingly, the mean difference in the lag time from baseline was clearly higher after the combined treatment than after fenofibrate alone or after vitamin E alone. The between-group differences were not statistically significant due to insufficient statistical power.

The AUC of TBARS formation decreased after eight weeks on vitamin E alone, remaining stable after a further eight weeks of vitamin E supplementation. Fenofibrate had a slight effect by itself. This effect was intensified when given in combination with vitamin E. Vitamin E and fenofibrate given in combination tended to be more efficient in increasing the lag time and in decreasing the AUC of TBARS formation than the two treatments separately, suggesting that the two compounds act in synergy. The same synergistic interaction of fenofibrate with \(\alpha\)-tocopherol was also described in rats [30]. The explanation for this synergy
could be related to the effect of fenofibrate on the relative amounts of poly-unsaturated fatty acids (PUFA) and mono-unsaturated fatty acids creating a favourable ratio of vitamin E/PUFA with a reduction in free radical propagating PUFA. Moreover the activation of PPARα has been shown to be associated with decreased tissue oxidative stress [31]. In the same study with rats, the combined administration of α-tocopherol with fenofibrate was compared to the combination with bezofibrate or gemfibrozil. The combination with fenofibrate proved to be more effective than the other combinations. Bezafibrate and fenofibrate lowered serum cholesterol to a comparable degree and gemfibrozil with α-tocopherol was the least effective in increasing the lag-phase.

In conclusion, this study in type 1 diabetic patients shows that the combination of micronized fenofibrate 200 mg/day and vitamin E 400 IU/day, tended to increase the resistance of non-HDL lipoproteins to copper-mediated oxidation. Vitamin E alone decreased the extent of oxidation of non-HDL lipoproteins as shown by the reduction of TBARS formation. This protective effect of vitamin E tended to be amplified by micronized fenofibrate. Vitamin E had no effect on lipids and did not alter the lipid-lowering effect of micronized fenofibrate. The two treatments given separately or in combination were well tolerated.

Further studies with the fenofibrate/vitamin E combination are needed to confirm these preliminary observations.

**Acknowledgements** – The study was financed by Fournier Laboratories S.A., Daix, France. The study could be done thanks to the staff of the laboratory of the Department of Diabetology, Nutrition and Metabolic Diseases and the nurses of the department. The study has been performed in compliance with the rules and principles of Good Clinical Practice.

### Table VII
Slope of the propagation curve of non-HDL copper-mediated oxidation.

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</tr>
<tr>
<td>Visit 2</td>
<td>0.92 (0.28)</td>
<td>0.96 (0.38)</td>
<td>0.8 (0.17)</td>
<td>0.84 (0.16)</td>
<td>0.93 (0.2)</td>
<td>0.89 (0.2)</td>
</tr>
<tr>
<td>Visit 3</td>
<td>0.72 (0.28)</td>
<td>0.65 (0.23)</td>
<td>0.74 (0.23)</td>
<td>0.75 (0.24)</td>
<td>0.76 (0.13)</td>
<td>0.73 (0.07)</td>
</tr>
<tr>
<td>Visit 4</td>
<td>0.68 (0.18) *</td>
<td>0.73 (0.1)</td>
<td>0.65 (0.22) **</td>
<td>0.68 (0.2)</td>
<td>0.65 (0.24)</td>
<td>0.68 (0.24)</td>
</tr>
</tbody>
</table>

Results are expressed in Fluorescence units/min as mean (SD)

* : paired t-test between V2 and V4 (P = 0.017)

** : paired t-test between V2 and V4 (P = 0.022).

### Table VIII
AUC of TBARS formation.

<table>
<thead>
<tr>
<th></th>
<th>ITT</th>
<th>PP</th>
<th>ITT</th>
<th>PP</th>
<th>ITT</th>
<th>PP</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(N = 14)</td>
<td>(N = 6)</td>
<td>(N = 16)</td>
<td>(N = 9)</td>
<td>(N = 14)</td>
<td>(N = 7)</td>
<td>one-way ANOVA</td>
</tr>
<tr>
<td>Visit 2</td>
<td>5338 (1499)</td>
<td>5255 (2151)</td>
<td>6433 (2064)</td>
<td>6866 (2569)</td>
<td>5726 (1676)</td>
<td>5474 (1744)</td>
<td>P = 0.053 ITT, P = 0.004 PP</td>
</tr>
<tr>
<td>Visit 3</td>
<td>4255 (2254)</td>
<td>3107 (1046)</td>
<td>6123 (1969)</td>
<td>6479 (2070)</td>
<td>5409 (1474)</td>
<td>4841 (1409)</td>
<td>P = 0.053 ITT, P = 0.004 PP</td>
</tr>
<tr>
<td>Visit 4</td>
<td>4433 (1701)</td>
<td>4435 (1764)</td>
<td>4572 (1591) *</td>
<td>4321 (1137)</td>
<td>5786 (1378)</td>
<td>5440 (1005)</td>
<td>P = 0.179 ITT, P = 0.314 PP</td>
</tr>
</tbody>
</table>

Results are expressed in nmol MDA/ mg non-HDL cholesterol x hours as mean (SD).

* : paired t-test, uncorrected between V2 and V4 (P = 0.003).

*: one-way ANOVA at each visit showed no significant difference between the various treatment groups at visit 2 and 4. At visit 3 there was a significant difference in the PP group (P = 0.004) and a tendency in the ITT group (P = 0.053).
References