EXPERT OPINION

Time course of antiproteinuric effect of aliskiren in arterial hypertension associated with type 2 diabetes and microalbuminuria

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Objective: The aim was to compare the antiproteinuric effect of aliskiren and ramipril in hypertensive patients with type 2 diabetes and microalbuminuria.
Research design and methods: A total of 138 patients were treated with aliskiren 300 mg/day or ramipril 10 mg/day for 12 weeks and checked after 1, 2, 4, 8 and 12 weeks and 2 and 4 weeks after treatment withdrawal.
Main outcome measures: Clinic and ambulatory BP, urinary albumin excretion rate (UAER) and plasma aldosterone were measured.
Results: Both aliskiren and ramipril induced a similar lowering in clinic and ambulatory BP ($p < 0.001$ vs baseline). However, such a lowering persisted longer after stopping aliskiren than after stopping ramipril regimen. Both treatments reduced UAER, but the decrease in UAER associated with aliskiren was more pronounced, the difference vs ramipril being maximal at week 12 (-42 vs -15%, $p < 0.01$). Two weeks after stopping therapy, UAER remained below baseline values with aliskiren, but not in the ramipril group. Plasma aldosterone decreased in the aliskiren group, whereas in the ramipril group it decreased until week 8 and thereafter increased toward baseline values.
Conclusions: Aliskiren has a greater and more prolonged antiproteinuric effect than R; it might partly be related to a higher degree of intrarenal renin–angiotensin–aldosterone system blockade.

Keywords: aldosterone, aliskiren, hypertension, microalbuminuria, ramipril

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1. Introduction

Due to the central role of the renin–angiotensin–aldosterone system (RAAS) in the pathogenesis of diabetic nephropathy, RAAS blockade represents the cornerstone treatment for this disease [1,2]. At present, angiotensin-converting-enzyme inhibitors (ACE-Is) and angiotensin receptor blockers (ARBs) are widely used as first-line pharmacological intervention for patients with both incipient and overt diabetic nephropathy after results of landmark trials [3-8]. Their effectiveness can be evaluated in terms of reduction in urinary albumin excretion (UAE), which is widely accepted as a surrogate renal endpoint [9,10]. Indeed, albuminuria, even in the microalbuminuric range (i.e., UAE of 30 – 300 mg/day) is a well-recognized marker for renal function decline and development of cardiovascular diseases in diabetic and non-diabetic individuals [11,12]. Although ACE-Is and ARBs effectively reduce UAE, many patients have residual proteinuria and progress to end-stage renal disease, despite long-term therapy with these drugs, possibly because of insufficient

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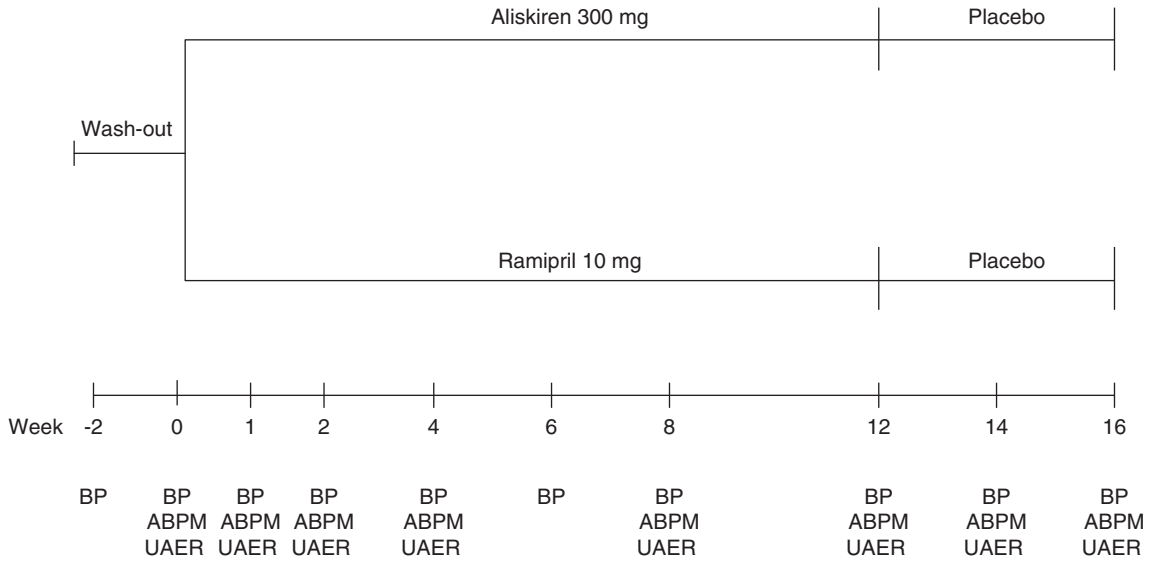

Figure 1. Study design.
ABPM: Ambulatory Blood Pressure Monitoring; BP: Blood pressure; UAER: Urinary albumin excretion rate.

suppression of the intrarenal RAAS [13,14]. In particular, ACE-Is have shown some limits. Thus, for example, in patients with chronic kidney disease, Keilani *et al.* [15] showed that with low dose of ramipril, 1.25 mg/day, there was a significant decrease of UAE as early as after 1 week of administration, but did not decrease any further thereafter, even when the dose was increased eight-fold, up to 10 mg/die. On the other hand, Ruggenenti *et al.* [16] demonstrated that patients with type 2 diabetes mellitus and overt nephropathy developed abnormalities in size-selective function of the glomerular barrier and, at variance with type 1 diabetes mellitus, such changes were not ameliorated either by ACE inhibition or calcium channel blockade.

Direct renin inhibition with aliskiren is a new option to block the RAAS at the first rate-limiting step, with consequent suppression of plasma renin activity (PRA) and reduced Ang II levels [17,18]. Based on some available data [19,20], aliskiren seems to be effective in reducing UAE, independently of its blood pressure (BP) lowering effect, with a better time course of its antiproteinuric effect as compared to other RAAS blockers. Persson *et al.* [19], found that treatment with aliskiren 300 mg/day was associated with a decrease in urinary albumin/creatinine ratio (UACR) by 17% after 2 – 4 days only, a 31% reduction on days 8 – 10 and a maximum reduction of 44% after 28 days of treatment. Interestingly, changes in UACR occurred earlier (days 2 – 4) than changes in 24 h BP (day 7), and UACR was further reduced during the treatment period, whereas the 24 h BP did not change further after day 7. Also in the AVOID study (Aliskiren in the eValuation of prOteinuria In Diabetes) [20], where aliskiren treatment on top of conventional therapy, including losartan 100 mg/day, produced a 20% albuminuria reduction in type 2 diabetes and nephropathy, the maximum reduction in UACR occurred after 28 days of treatment. Both of these studies, however, have the main limitation in the selected population, which included both microalbuminuric and macroalbuminuric patients. Hence there is the need to evaluate the time course of the antiproteinuric effect of aliskiren in a more homogeneous population of hypertensive patients with type 2 diabetes and microalbuminuria, since normalization of proteinuria in microalbuminuric patients has been demonstrated to stop progression of nephropathy [9,10].

Given this background, the aim of the present study was to compare the time course of the antiproteinuric and antihypertensive effect of direct renin inhibitor aliskiren and of ACE-inhibitor ramipril in hypertensive patients with type 2 diabetes and microalbuminuria.

2. Patients and methods

This was a prospective, randomized, open-label, blinded endpoint, parallel-group study. After complete discontinuation of any previous antihypertensive treatment, male and female outpatients aged 25 to 75 years with mild-to-moderate essential hypertension (defined as sitting systolic blood pressure [SBP] ≥ 140 and < 180 mmHg, and sitting diastolic blood pressure [DBP] ≥ 90 and < 105 mmHg), type 2 diabetes mellitus well-controlled by diet or oral hypoglycemic agents (glycated hemoglobin [HbA_{1c}] < 7%, absence of glycosuria, and no change in hypoglycemic drugs in the last 6 months), and in the higher range of microalbuminuria defined as UAE > 200 and < 300 mg/day in two distinct 24 h urine collections, entered a washout run-in period for 2 weeks. At the end of this period, the patients who fulfilled the inclusion criteria were randomized to receive aliskiren 300 mg/day or ramipril 10 mg/day for 12 weeks (**Figure 1**). After 2 weeks

Table 1. Baseline demographic and clinical characteristics of the patients randomized to the two treatment groups*.

	Aliskiren	Ramipril
n	69	69
Male/Female	33/36	32/37
Age (year)	61.6 ± 8.5	62.1 ± 8.9
BMI (kg/m^2)	27.4 ± 3.7	27.6 ± 3.9
SBP (mmHg)	158.4 ± 9.3	158.6 ± 9.1
DBP (mmHg)	94.8 ± 4.7	94.1 ± 4.5
HR (beats/min)	77.4 ± 6.5	75.2 ± 7.4
FPG (mg/dL)	135.3 ± 25.1	136.2 ± 24.5
HbA_{1c} (%)	6.7 ± 0.6	6.6 ± 0.8
UAER (mg/day)	256.8 ± 31.3	259.2 ± 27.2
Creatinine (mg/dL)	1.04 ± 0.18	1.05 ± 0.18
Serum K^+ (mEq/L)	4.3 ± 0.33	4.5 ± 0.34
Aldosterone (ng/dL)	77.4 ± 53.6	76.2 ± 52.1
PRA (ng/mL/h)	1.34 ± 0.37	1.33 ± 0.39
PRC (μU/mL)	21.9 ± 9.1	21.4 ± 8.9
Smokers (%)	14 (20.3)	15 (21.7)
Duration of hypertension (years)	6.4 ± 5.5	6.97 ± 5.8
Duration of diabetes (years)	6.8 ± 4.2	6.2 ± 4.7

Data are means ± SD.

*No significant difference was found between the two groups with regard to the considered parameters.

BMI: Body mass index; DBP: Diastolic blood pressure; FPG: Fasting plasma glucose; HR: Heart rate; HbA_{1c}: Glycated hemoglobin; PRA: Plasma renin activity; PRC: Plasma renin concentration; SBP: Systolic blood pressure; UAER: Urinary albumin excretion rate.

therapy, if clinic BP was > 130/80 mmHg, amlodipine 2.5 mg was added; after 4 weeks, if BP was yet uncontrolled, amlodipine was doubled to 5 mg and, after 6 weeks, it was again doubled to 10 mg. At week 8, the patients with BP > 130/80 mmHg were withdrawn.

Exclusion criteria included secondary hypertension, myocardial infarction or stroke within 6 months before the beginning of the study, obesity (body mass index ≥ 30 kg/m^2), congestive heart failure, major non-cardiovascular diseases, pregnancy, lactation, and known or suspected hypersensitivity to the trial medications. The study protocol was approved by the local ethics committee, and informed consent was obtained from each patient before enrolment. Clinic BP, ambulatory BP monitoring (ABPM), UAE rate (UAER) as well as RAS biomarkers (PRA, plasma renin concentration [PRC] and plasma aldosterone), serum creatinine and potassium were evaluated at the end of the washout run-in period (baseline), after 1, 2, 4, 8 and 12 weeks of treatment and after 2 and 4 weeks of treatment withdrawal. BP was measured in the morning, immediately before the daily dose of medication (i.e., at trough, 24 h after dosing) by the same clinician, using the same calibrated mercury sphygmomanometer (Korotkoff I and V) on the same patients in the dominant arm, after the subject had been sitting for 5 min in a quiet room. Three BP measurements were made with a 2 min interval between them and averaged. ABPM was performed over 24 h with the use of a clinically validated device (Spacelabs 90207, Spacelabs, Inc., Redmond, Washington, WA, USA) [21] that was programmed to measure BP every 15 min during the entire course of the recording. Each recording was started in the morning, immediately after clinic BP assessment and drug administration. Patients were instructed to remain motionless each time a reading was taken. Analysis of 24 h BP recordings was preceded by removal of artifacts, according to previously described editing criteria [21,22]. Recordings were considered valid when no more than two non-consecutive hours were missing over 24 h. For each patient, the following data related to SBP, DBP and heart rate (HR) were obtained through analysis of the recordings: 24 h mean values, daytime (07.00 – 23.00), and nighttime (23.00 – 07.00), mean values.

The UAER was assessed by means of radioimmunoassay; each reported value represents the mean of two distinct 24 h urine collections within 1 week. Blood sample for PRC, PRA and aldosterone levels were taken always at the same hour in the morning after 30 min of supine rest and after centrifugation plasma was frozen (-80°C). PRC was measured with a radioimmunometric kit (Renin III, CisBio, Gif-sur-Yvette, France). PRA was measured by radioimmunoassay of Ang I formed during incubation of plasma for 1 h at 37°C (Incstar, Stillwater, MN, USA). Aldosterone was measured with a radioimmunoassay kit (Coat-A-Count, Diagnostic Products Corp., Los Angeles, CA, USA). At each visit, adverse events (AEs) spontaneously reported by patients or elicited by the investigators were recorded. Serum creatinine and potassium were measured using commercial kits employing routinely used clinical chemistry procedures.

2.1 Sample size

The primary endpoint was change in UAER measured from baseline to 1, 2, 4, 8 and 12 weeks after the start of treatment. The population so far studied was very heterogeneous, with a great variability in proteinuria. After a log-transformation of the data, we assume that the mean of ln (albuminuria) in this study population is 5.5 mg/day (considering that only patients with albuminuria in the range 200 – 300 mg/day are included in the study) and the standard deviation is 0.3 mg/day. We also assume a very low effect of ramipril (10% albuminuria reduction) and an effect of aliskiren of at least 25% in this specific study population. On the log scale, this means a difference in means of about 0.2 mg/day. A sample size of 49 patients per group will have a 90% power to detect a difference in means of 0.2 mg (15% difference) with a standard deviation of 0.3 mg/day, using a two groups *t*-test with a 0.05 two-sided significant level.

2.2 Statistical analysis

Data are expressed as means ± SD. The homogeneity check of patient distribution between treatment groups was performed using the χ^2 test. The results were statistically analyzed, using analysis of variance and the Student's *t*-test for paired and unpaired data as appropriate. Treatment comparison between

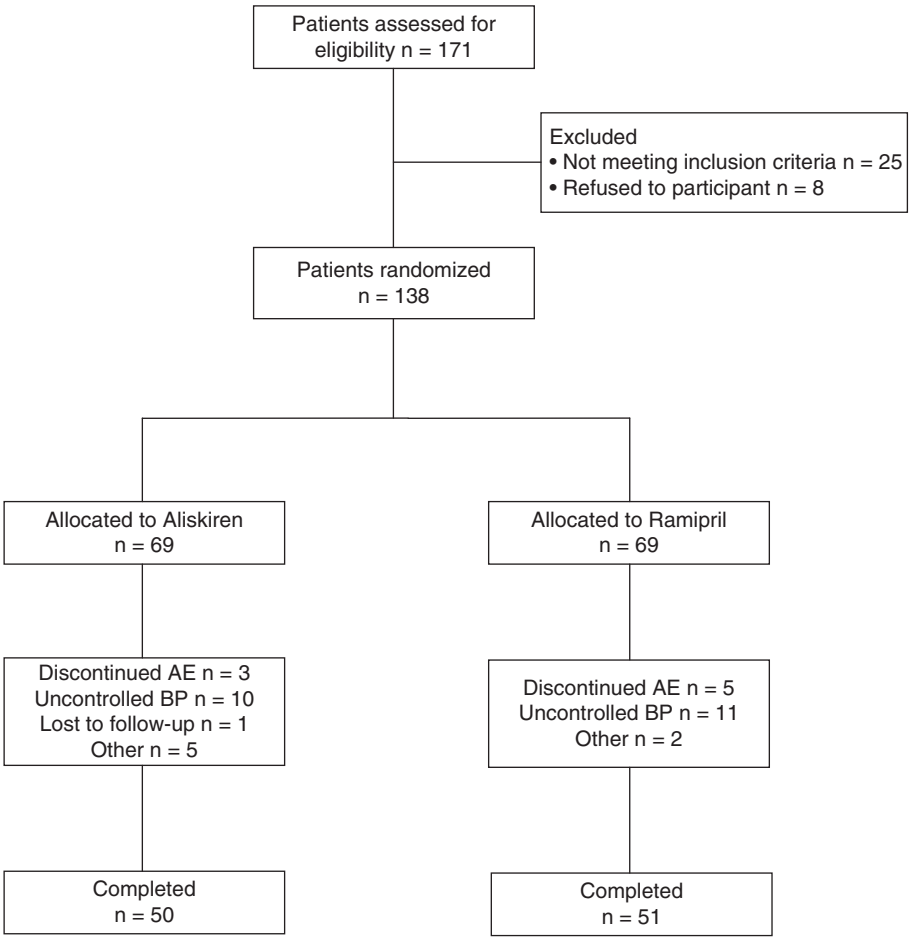

Figure 2. Flow diagram of the study.

the patients who received aliskiren and those who received ramipril were performed with the use of a two-sided test with a significance level of 0.05. Correlation between changes from baseline in BP and changes from baseline in the UAER were assessed by linear regression analysis and the Pearson correlation coefficient *r* was used.

3. Results

In total, 171 patients were screened in the washout phase of the trial and 138 of them (65 males and 73 females, mean age 61.85 years) were randomly assigned to aliskiren 300 mg/day (n = 69) or to ramipril 10 mg/day (n = 69). As shown in **Table 1** the distribution of patient between the two treatment groups was satisfactory with regard to the main demographic and clinic characteristics.

A total of 37 patients dropped out after randomization (19 in the aliskiren group and 18 in the ramipril group), 21 patients because of uncontrolled BP, 8 due to AEs, 7 because of consent withdrawn and 1 patient was lost at follow-up. Thereafter, 101 patients completed the study period (**Figure 2**). The number of patients who needed the addition of amlodipine at week 2 was 45 in the aliskiren group and 46 in the ramipril group.

3.1 Blood pressure

Clinic BP results are shown in **Figure 3**. Both aliskiren- and ramipril-based therapy provided significant reductions in SBP and DBP mean values at week 12 endpoint, with no significant difference between the two treatments (-24.1/18.6 mmHg with aliskiren, $p < 0.001$ vs baseline and -24.4/17.1 mmHg with ramipril, $p < 0.001$ vs baseline). However, a greater number of patients given ramipril-based therapy than aliskiren-based therapy were receiving additional add-on therapy with amlodipine (42 [84%] vs 37 [74%]) at week 12 endpoint. Besides, at week 2 (end of monotherapy with each agent) aliskiren produced greater mean reduction in SBP compared with ramipril (-13.5 vs -9.8 mmHg), the difference between treatments being statistically significant ($p < 0.05$). Clinic BP reductions persisted longer after stopping aliskiren-based therapy than after stopping ramipril-based therapy. Most of the posttreatment increase in BP occurred 2 weeks after stopping ramipril, whereas BP increased more gradually after stopping aliskiren, so that at week 14 SBP and DBP values

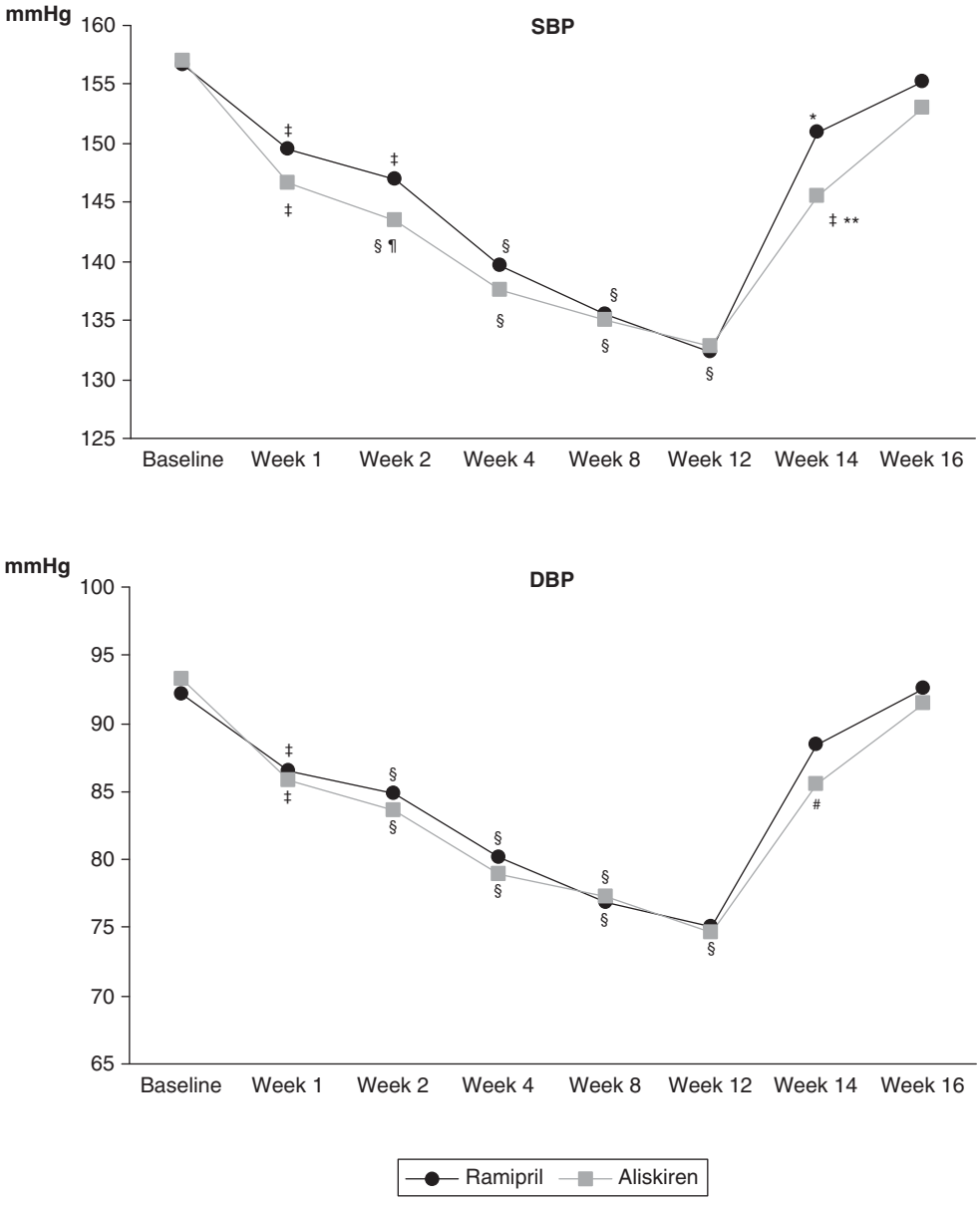

Figure 3. Clinic BP.

* $p < 0.05$; ‡ $p < 0.01$; § $p < 0.001$ vs baseline.

¶ $p < 0.05$; # $p < 0.01$; ** $p = 0.002$ vs ramipril.

BP: Blood pressure; DBP: Diastolic blood pressure; SBP: Systolic blood pressure.

were significantly lower in the aliskiren than in the ramipril group (145.6 ± 8.7 vs 150.9 ± 8.29 mmHg for SBP, $p = 0.002$ and 84.1 ± 5.4 vs 88.4 ± 4.1 mmHg for DBP, $p < 0.01$). Four weeks after stopping treatment (week 16), clinic BP returned to baseline values in both treatment groups, without difference between them. Clinic HR values were unaffected by both treatments at any time of the study.

Ambulatory BP results are shown in **Figure 4**. Both aliskiren- and ramipril-based therapy produced similar reductions in 24 h, daytime and nighttime SBP and DBP mean values at week 12 endpoint, with no significant difference between the two treatments. The mean decrease in 24 h SBP/DBP was of 22.3/17 mmHg ($p < 0.001$ vs baseline) with aliskiren and of 21.3/16.4 mmHg ($p < 0.001$ vs baseline) with ramipril. Daytime SBP/DBP were decreased by a mean of 24.6/18.1 mmHg ($p < 0.001$ vs baseline) by aliskiren and by a mean of 23.7/17.6 mmHg ($p < 0.001$ vs baseline) by ramipril. Similar reductions were observed in nighttime SBP/DBP mean values (-17.6/14.7, $p < 0.001$ vs baseline with aliskiren and -16.5/13.8 mmHg, $p < 0.001$ vs baseline with ramipril). As already observed with clinic BP, at week 2 (end of monotherapy), the mean reduction in ambulatory SBP values was

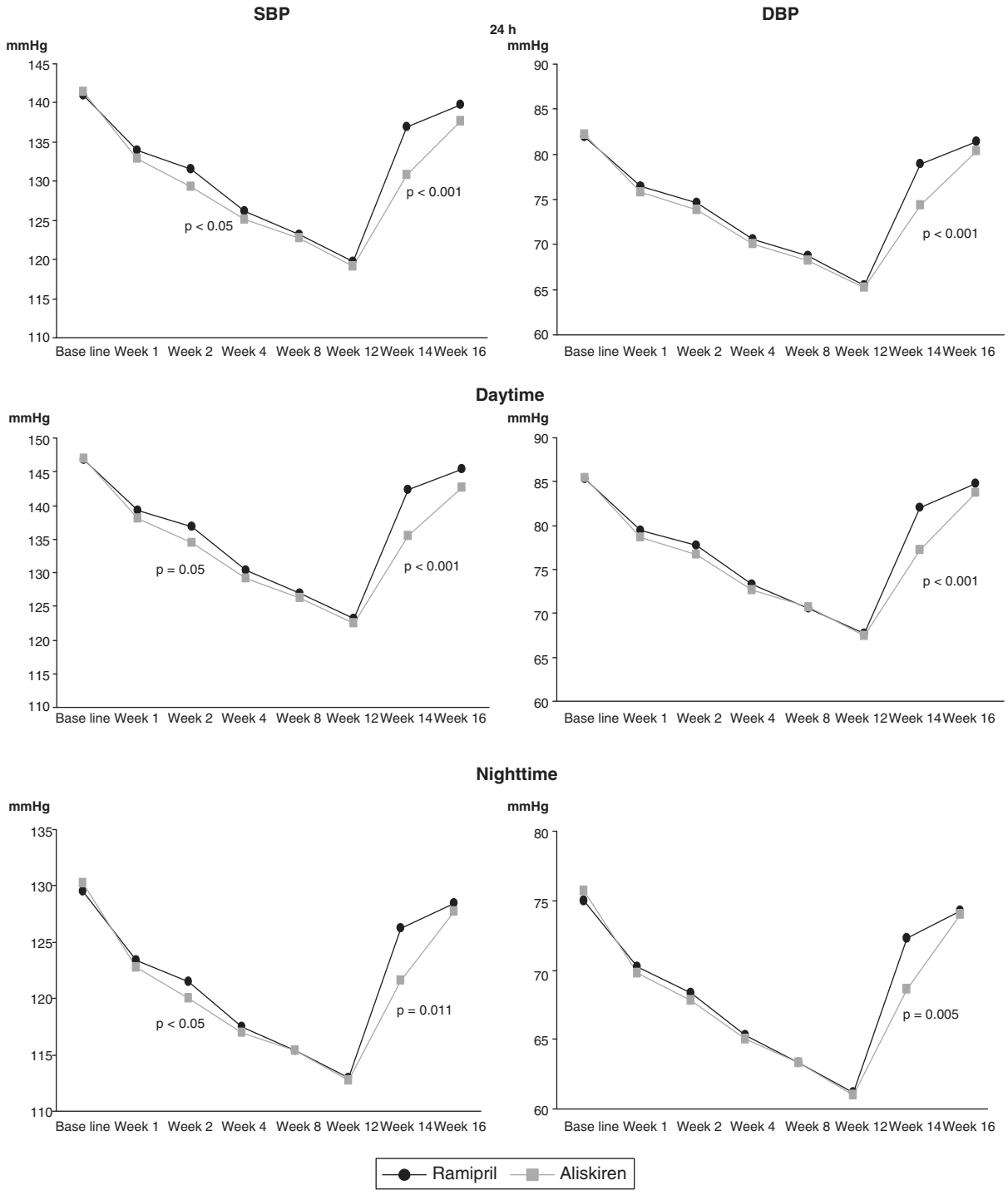

Figure 4. ABPM 24 h, daytime and nighttime.
ABPM: Ambulatory blood pressure monitoring; SBP: Systolic blood pressure; DBP: Diastolic blood pressure.

slightly, but significantly greater, in the aliskiren than in the ramipril group (-11.6 vs 9.2 mmHg, $p < 0.05$ for 24 h SBP, -12.3 vs 9.9 mmHg, $p < 0.05$ for daytime SBP and -10.3 vs 7.8 mmHg, $p < 0.05$ for nighttime SBP). Similar to what was observed for clinic BP, ambulatory SBP/DBP reductions persisted longer after stopping aliskiren-based therapy than after stopping ramipril-based therapy. Thus, 2 weeks after stopping treatment, the mean values of 24 h, daytime and nighttime SBP were significantly lower in the aliskiren than in the ramipril group (130.8 ± 8.1 vs 136.9 ± 8.1 mmHg, $p < 0.001$ for 24 h SBP, 135.5 ± 8.6 vs 142.3 ± 8.0 mmHg, $p < 0.001$ for daytime and 121.6 ± 8.9 vs 126.2 ± 8.7 mmHg, $p = 0.011$ for nighttime

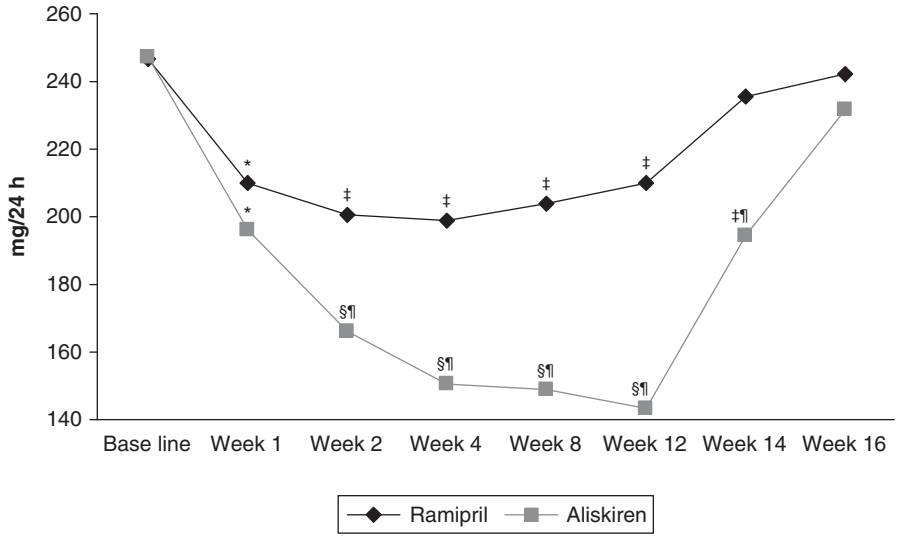

Figure 5. Microalbuminuria levels.
* $p < 0.05$; ‡ $p < 0.01$; § $p < 0.001$ vs baseline.
¶ $p < 0.001$ vs ramipril.

SBP). Similar findings were obtained for ambulatory DBP values, which were significantly lower in the aliskiren than in the ramipril group (74.3 ± 5.5 vs 78.9 ± 3.3 mmHg, $p < 0.001$ for 24 h DBP, 77.2 ± 5.1 vs 82.1 ± 3.8 mmHg, $p < 0.001$ for daytime DBP and 68.6 ± 7.9 vs 72.3 ± 4.3 mmHg, $p < 0.005$ for nighttime DBP). Four weeks after stopping therapy (week 16), ambulatory SBP/DBP values reached pretreatment levels in both treatment groups, with no significant difference between them.

3.2 Urinary albumin excretion rate

Both aliskiren- and ramipril-based therapies were associated with a significant decrease in UAER throughout the treatment period (**Figure 5**). However, the decrease in UAER associated with aliskiren was more pronounced at each time of the study (**Figure 6**). The difference against ramipril was already statistically significant at week 2 (end of monotherapy) and became progressively more evident, being maximal at week 12 (end of active treatment period) (**Figure 6**). At this time point, UAER decreased by a mean of 103.8 mg/day (-42%), $p < 0.001$ vs baseline in the aliskiren-treated patients and by 36.8 mg/day (-15%), $p < 0.01$ vs baseline in the ramipril-treated patients, with a significant difference between the two treatments ($p < 0.001$).

Two weeks after stopping therapy, UAER remained significantly below baseline values in the aliskiren (194.5 ± 49.3 vs 247.4 ± 26.8 mg/day, $p < 0.001$) but not in the ramipril group (235.6 ± 29.5 vs 246.8 ± 26.4 mg/day, not significant), the difference between the two treatments being statistically significant ($p < 0.001$) (**Figure 6**). In both treatment groups, UAER returned to pretreatment values 4 weeks after stopping therapy.

A significant correlation was found between the change from baseline in UAER and in clinic SBP in both treatment groups (**Figure 7**), although such a correlation was more marked with aliskiren ($r = 0.98$, $p < 0.01$) than with ramipril ($r = 0.76$, $p = 0.027$).

3.3 RAAS biomarkers

Results about RAAS biomarkers are shown in **Figure 8**.

Mean PRA at baseline was 1.32 ± 0.36 and 1.32 ± 0.38 ng/mL/h in the aliskiren and ramipril treatment groups, respectively. During the active treatment period, aliskiren-based therapy reduced PRA by 67.4% from baseline to week 12 endpoint, whereas ramipril-based therapy increased PRA by 120.4% at this time point, the difference between the two treatments being statistically significant ($p < 0.001$). After stopping therapy, PRA levels gradually increased and remained below pretreatment levels 4 weeks after stopping aliskiren-based therapy, whereas PRA rapidly decreased in patients who stopped ramipril-based therapy, returning to baseline levels 2 weeks after treatment withdrawal (**Figure 8**).

Mean PRC at baseline was 21.1 ± 9.0 and 21.2 ± 8.7 ng/L in the aliskiren and ramipril treatment groups, respectively. During the active treatment period, PRC increased significantly from baseline to week 12 endpoint with both regimens, but such an increase was more marked in the aliskiren-treated patients (+389%, $p < 0.001$ vs baseline) than in the ramipril-treated patients (+158%, $p < 0.01$ vs baseline), with a significant difference between the two groups ($p < 0.001$). After stopping therapy, PRC values decreased rapidly in both treatment groups, returning to baseline levels 2 weeks after treatment withdrawal.

Baseline plasma aldosterone mean values were 76.9 ± 52.3 and 76.6 ± 51.3 ng/L in the aliskiren and ramipril treatment groups, respectively. During active treatment period, in the aliskiren-treated patients, plasma aldosterone levels progressively decreased from baseline to week 12 endpoint (-37%,

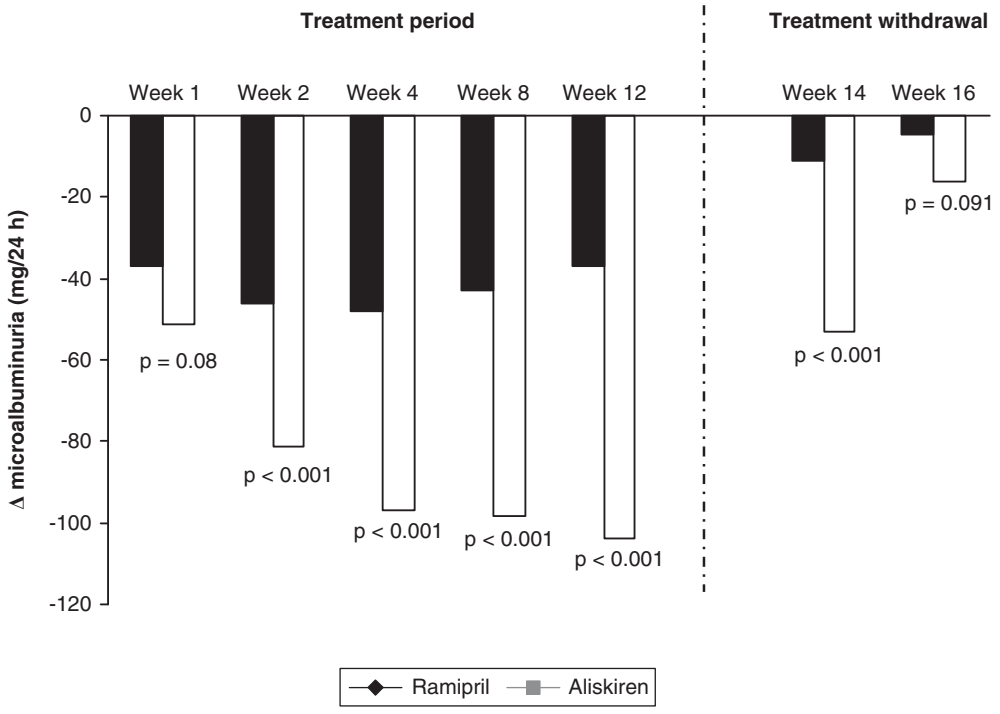

Figure 6. Changes in microalbuminuria.

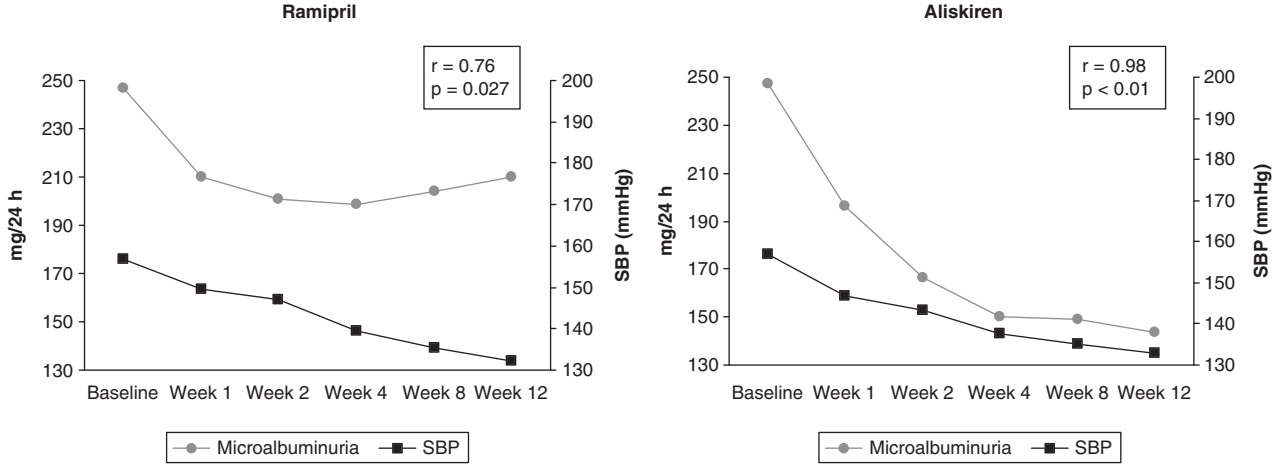

Figure 7. Correlation between microalbuminuria and clinic SBP during the 12 weeks of active treatment.
SBP: Systolic blood pressure.

p < 0.001), whereas in the ramipril-treated patients, we observed the aldosterone escape phenomenon, since plasma aldosterone significantly decreased from baseline to week 4 (-26%, p < 0.01) and thereafter increased toward baseline values at week 12 (71.5 ± 47.7 ng/L, -6%, not significant vs baseline and p = 0.009 vs aliskiren) (**Figure 9**). After stopping therapy, plasma aldosterone values increased in both treatment groups returning to baseline levels at week 16.

3.4 Safety and tolerability

Both aliskiren- and ramipril-based therapies were well tolerated. In the overall patient population, most AEs were mild or moderate in intensity. The rate of AEs was 10% in the aliskiren-treated patients and 23% in the ramipril-treated patients (**Table 2**), whereas the discontinuation rate due to AEs was 4 and 7%, respectively. Cough occurred in four patients in the ramipril group (8%) and in no patient in the

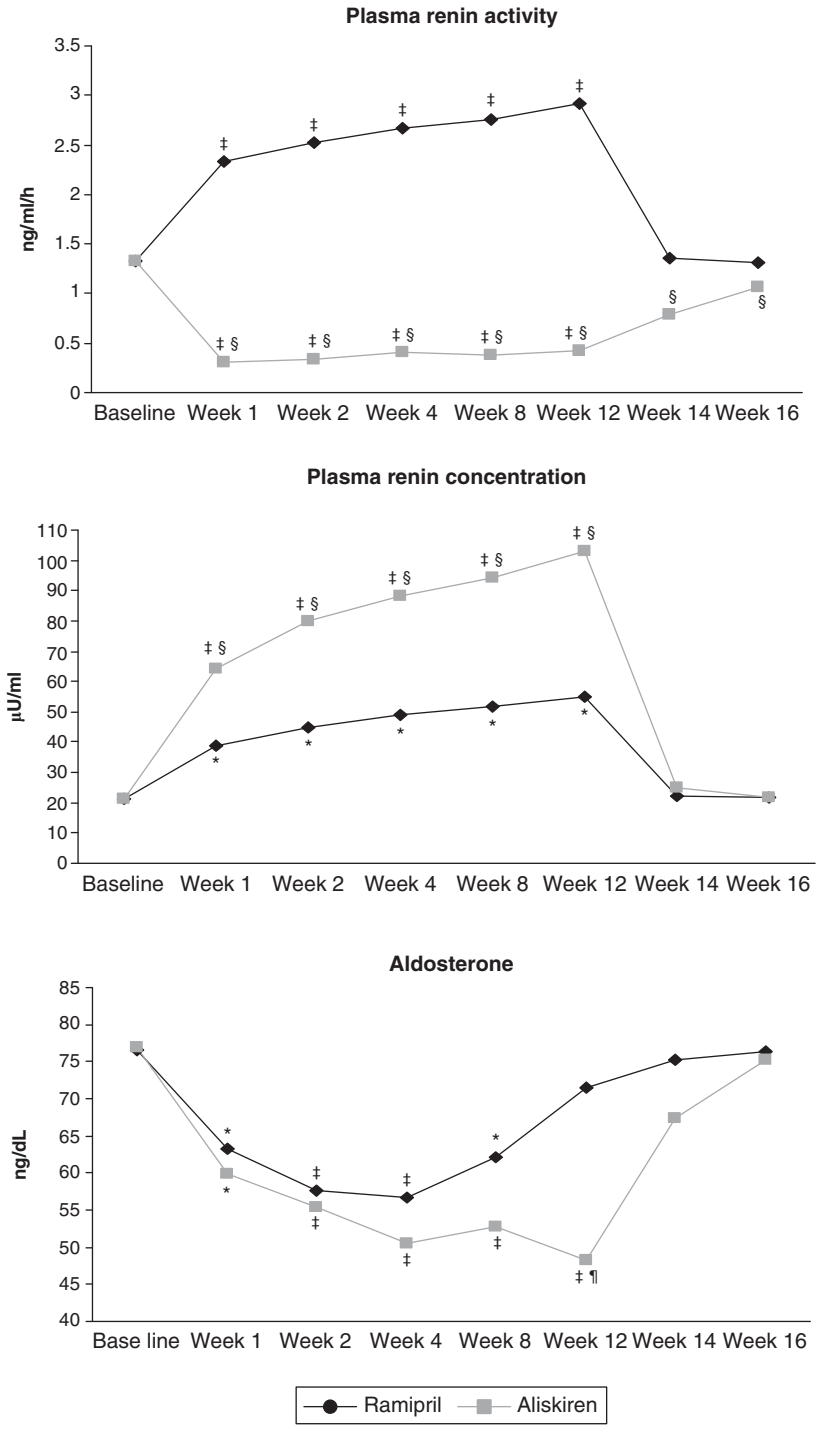

Figure 8. RAS biomarkers.

* $p < 0.01$; ‡ $p < 0.001$ vs baseline.

¶ $p = 0.009$; § $p < 0.001$ vs ramipril.

RAS: Renin-Angiotensin System.

aliskiren group. Serum potassium levels increased from 4.5 ± 0.3 at baseline to 4.7 ± 0.3 mEq/L at 12 week endpoint with aliskiren-based therapy, whereas ramipril-based therapy did not significantly change them (from 4.4 ± 0.33 to 4.5 ± 0.2 mEq/L), the difference between treatments being statistically significant ($p = 0.013$). However, no event of hyperkalemia (serum potassium > 5.5 mEq/L) was recorded during the study. Serum creatinine levels were unaffected by both

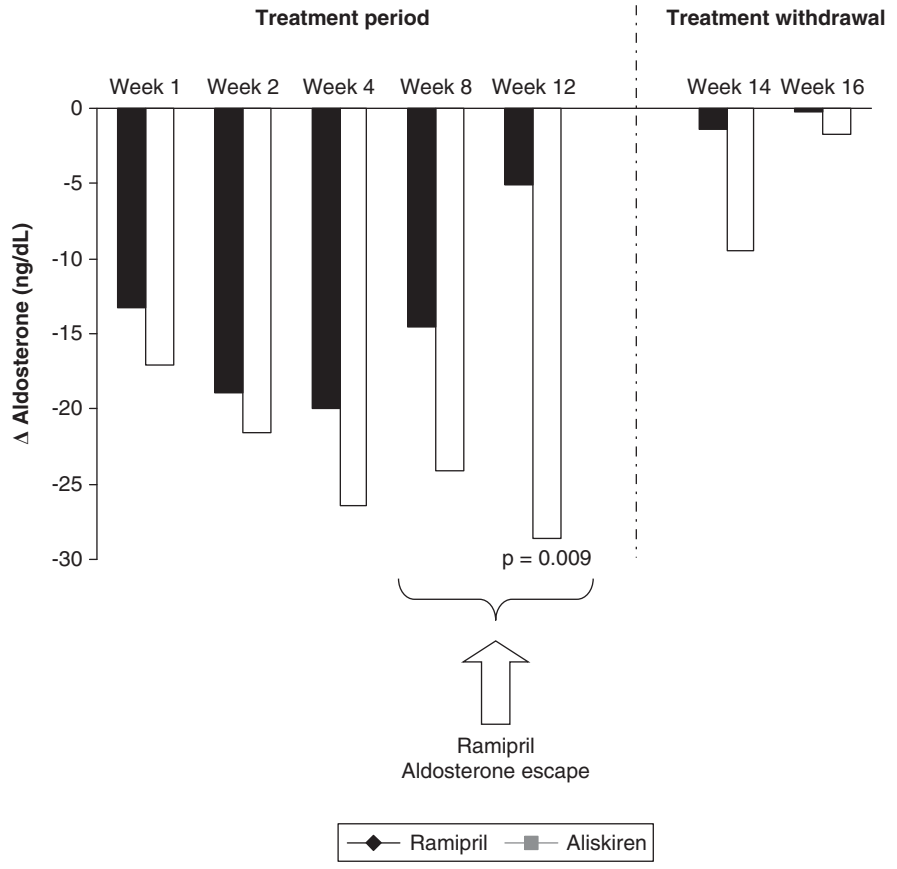

Figure 9. Changes in aldosterone.

treatment regimens throughout the study. No cardiovascular events or deaths occurred throughout the study.

4. Discussion

The results of this study demonstrated that in hypertensive patients with type 2 diabetes and microalbuminuria, both aliskiren- and ramipril-based therapies were effective in reducing clinic as well as ambulatory BP values at week 12 endpoint, without significant difference between the two treatments. However, there was a less need for add-on therapy with amlodipine in the aliskiren-treated group to reach the BP goal requested by the study protocol and more patients on aliskiren were receiving monotherapy than patients on ramipril at the end of the active treatment period (26 vs 17%). In clinical practice this could be beneficial from therapeutic adherence perspective, thereby contributing to better BP control. Besides, at week 2, at the end of monotherapy with each drug, the aliskiren-induced reduction in both clinic and ambulatory SBP was significantly greater than that observed with ramipril. This resembled the results of some previous comparative studies between aliskiren and ramipril monotherapy in both diabetic and non-diabetic hypertensive patients [23,24]. The greater antihypertensive efficacy of aliskiren alone observed in the present study would probably be more evident if monotherapy would have been prolonged for longer time. The ability of aliskiren to achieve more prompt BP reduction than ramipril might have a relevant impact on cardiovascular disease, since large clinical trials have demonstrated that the risk of cardiovascular events is significantly influenced by the time it takes to achieve BP target [25,26]. At this regard, results from the VALUE trial showed a greater reduction in cardiovascular risk in patients who reached target BP after the first month of treatment compared with those who achieved BP goal at a later time [25]. It is also believed that prompt reduction of BP as early as the first week of treatment may improve patient therapeutic compliance [27]. Interestingly, both clinic and ambulatory SBP/DBP reductions persisted for longer after stopping aliskiren-based therapy than after stopping ramipril-based therapy, which confirmed previous observations [28]. Potential explanations for these findings include: i) a more complete and effective 'upstream' inhibition of the RAAS by aliskiren as compared with ramipril, with consequent prevention of the reactive rise in PRA and the compensatory increase in Ang I, that accompanies treatment with ACE-Is and partially restores Ang II production by ACE-independent pathways; this explanation is supported by our observation that aldosterone values, which were progressively reduced by aliskiren, under

Table 2. AEs complained by the patients who completed the study.

	Ramipril (n = 51)	Aliskiren (n = 50)
Edema	4	2
Cough	4	0
Rush	2	1
Diarrhea	0	1
Headache	1	1
Nausea	1	0

ramipril-based treatment decreased during the first 4 weeks. but thereafter increased toward baseline levels; this aldosterone escape might have limited the antihypertensive efficacy of ramipril; ii)long-term elimination half-life of aliskiren, which approximates 40 h [29] and contrasts with the shorter half-life of ramipril, the active metabolite of ramipril, which approximates 13 – 17 h [30].The longer maintenance of the antihypertensive effect following aliskiren treatment interruption may be of clinical relevance in consideration of the high proportion of patients who occasionally miss a dose of treatment [31].

Both aliskiren- and ramipril-based therapies produced a significant reduction of UAER throughout the study period. However, the decrease in UAER associated with aliskiren was significantly greater than that associated with ramipril at each time of the study, the difference between the two regimens being maximal after 12 weeks of active treatment. Moreover, the UAE lowering effect persisted longer after stopping aliskiren-based therapy than after stopping ramipril-based therapy. Mechanisms for such different antiproteinuric effects are unclear. RAAS blockade by ACE-Is and ARB is believed to reduce proteinuria through several mechanisms including i) reduction of intraglomerular hydraulic pressure by preferential vasodilation of the efferent arterioles; ii) improvement of perm selective properties of the glomerular membrane; iii) reduction of the deficiency in glomerular nephrin expression, a protein located at the slit-diaphragm of the glomerular podocyte, which is suggested to play a central role in the function of the glomerular filtration; iv) reduction of renal levels of pro-atherosclerotic cytokines, such as transforming growth factor-beta (TGFβ) and connective tissue growth factor [32-35]. Aliskiren probably shares the same mechanisms for reducing proteinuria with ACE-Is and ARBs, but a peculiar effect on renin/prorenin receptor and intrarenal RAAS has been suggested. Renin/prorenin receptors have been demonstrated to stimulate the mitogen-activated protein kinase pathway and to increase TGFβ, plasminogen activator inhibitor-1, fibronectin and type 1 collagen in renal mesangial cells [36]. Both *in vitro* conditions with high glucose [37] and in animals with diabetes [38,39], aliskiren reduced the number of (pro)renin receptors in the kidney, attenuated profibrotic activity in the kidney and nearly abolished the apoptotic effects on cultured podocytes. In transgenic (MRen-2)27 rats with diabetes, aliskiren showed a greater renoprotective effect than ACE-Is by inducing a greater reduction in tubular interstitial fibrosis as compared with perindopril [40]. The mechanisms underlying such a different effect of the two drugs are unclear, but the aliskiren-mediated reduced expression of prorenin/renin receptor, which is predominantly expressed in the renal tubular epithelium, could have played a pivotal role [41].

Aliskiren treatment has also been demonstrated to exert an Ang II-dependent vasodilatory effect on the efferent arteriole in the glomerulus, which produces a rise in renal plasma flow and a reduction in filtration fraction that far exceeds the effect previously seen with ACE-Is and ARBs. In salt-depleted healthy humans, the vasodilator response induced by aliskiren was twice than reported with ACE-Is and exceeded the effects of ARBs by about 40%. Besides, renal vasodilatation persisted for 48 h after a single oral dose [42]. Similar favorable effects of aliskiren on renal hemodynamics have been also demonstrated in type 1 diabetic subjects [43]. These findings suggest that aliskiren may provide more effective blockade of the RAAS in the kidney. Since the RAAS is activated in patients with diabetes as compared with controls [44], a more pronounced difference in renal vasodilation may be expected during aliskiren therapy [20].

Animal studies indicated that aliskiren partitioned to the kidney and exhibited a prolonged renal residence, which was not seen with ACE-Is or ARBs [45]. As documented by autoradiography, aliskiren accumulated in renal tissue after 2 weeks of treatment and localized in the glomeruli and in the arterial walls of the small cortical blood vessels. Besides, aliskiren inhibited intrarenal RAAS, even several days after cessation of treatment [45].

In patients with advanced diabetic nephropathy, the aliskiren-induced suppression of albuminuria was suggested to correlate to its action of suppressing oxidative stress and tubulointerstitial damage [46]. Thus, it has been suggested that the increase in oxidative stress and inflammations caused by increased PRA might be related to the mechanism by which proteinuria increases in patients already on treatment with ACE-Is or ARBs but who still see their nephropathy worsening [47].

In the present study, the reductions in albuminuria were significantly correlated with the change from baseline in clinic SBP in both treatment groups, which confirmed the importance of BP reduction in decreasing UAE. However, since the degree of BP lowering of aliskiren-based therapy was similar to that of ramipril-based therapy after 12 weeks of treatment, the greater antiproteinuric effect of aliskiren was probably related to factors other than mere BP decrease.

In addition to the above-described hypothetic mechanisms for aliskiren antiproteinuric effect, the aldosterone escape that we observed with ramipril, but not with aliskiren, could also have contributed to their different effects on UAER. Indeed, an association has been demonstrated in animal models between elevated aldosterone levels and proteinuria [48]. Besides, there is experimental and clinical evidence that

aldosterone can contribute to the development of nephrosclerosis and renal fibrosis in models of diabetes and hypertension and that aldosterone-induced vasculopathy may underlie progressive renal disease [49,50]. Failure of ACE-Is to suppress aldosterone production during long-term treatment (so-called aldosterone escape) may result in no amelioration of the direct renal effect of aldosterone, thus, limiting the antiproteinuric and renal protective effect of ACE inhibition over time [51]. By converse, the ability of aliskiren to reduce steadily aldosterone levels in the long-term may be relevant from the standpoint of proteinuria reduction and renal protection.

As expected, at the end of active treatment, PRC levels were increased with both aliskiren-based and ramipril-based treatments, due to loss of Ang-II mediated feedback inhibition of renal renin release. Despite the increase in PRC, aliskiren-based therapy lowered PRA, whereas ramipril-based therapy significantly increased PRA. Such a reactive PRA increase might have contributed to the less marked antiproteinuric effect of ramipril.

Whereas the AVOID trial [20] showed that aliskiren had additional renoprotective effects when added in hypertensive type 2 diabetic patients with nephropathy who were receiving the recommended maximal renoprotective treatment with an ARB, the more recent results of the ALTITUDE trial [52] indicated that the addition of aliskiren to standard therapy with an ACE-I or an ARB did not reduce renal or cardiovascular outcomes and resulted in an increased number of AEs. Given these findings, combination therapy with aliskiren and another RAS blocker is not recommended in diabetic patients with nephropathy. In the present study, aliskiren was not added, but compared to the ACE-I ramipril and the greater antiproteinuric effect that we observed with aliskiren suggests that this drug may be a valid alternative to this ACE-I in the treatment of diabetic hypertensives with microalbuminuria. Further studies, however, are needed to confirm beneficial outcome data with aliskiren-based regimen in these patients.

5. Conclusions

In conclusion, this study suggested that in hypertensive microalbuminuric type 2 diabetic patients, aliskiren-based therapy produced a greater and more sustained decrease in UAER than ramipril-based therapy. This could be due to a higher degree of intrarenal RAAS blockade, which might also explain the more prolonged antihypertensive effect of the drug and the lack of aldosterone escape as compared to ramipril.

Declaration of interest

The authors have no relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript. This includes employment, consultancies, honoraria, stock ownership or options, expert testimony, grants or patents received or pending or royalties. No writing assistance was utilized in the production of this manuscript.

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