

ORIGINAL PAPER

# BENEFICIAL EFFECTS OF TRIPLE ANTIHYPERTENSIVE THERAPY WITH AMLODIPINE, VALSARTAN AND HYDROCHLOROTHIAZIDE ON COPPER STATUS IN A TRIAL OF GERIATRIC PATIENTS IN SOUTHWESTERN ROMANIA

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## ABSTRACT

**Background.** Inadequate mineral status involving the biometals copper and zinc is linked to cardiovascular homeostasis, anemia and arterial hypertension. In our previous studies, we revealed that long-time therapy with angiotensin-converting enzyme (ACE) inhibitors causes excessive zinc deficiency in comparison with the triple fixed-dose combination antihypertensive therapy, which is safer for the elderly hypertensive patient.

**Objectives.** The aim of our study was to evaluate the pharmaco-therapeutic effects of a fixed-dose combination of Amlodipine + Valsartan + Hydrochlorothiazide (Aml + Val + HCTZ) 10/320/25 mg on the copper status, in a clinical setting of geriatric patients from Southwestern Romania.

## RÉSUMÉ

Les effets bénéfiques du triple traitement antihypertenseur avec amlodipine, valsartan et hydrochlorothiazide sur l'état du cuivre dans une cohorte de patients gériatriques dans le Sud-Ouest de la Roumanie

**Introduction.** L'état minéral inadéquat impliquant les oligo-éléments cuivre et zinc est lié à l'homéostasie cardiovasculaire, à l'anémie et à l'hypertension artérielle. Dans nos études précédentes, nous avons révélé que la thérapie de longue date avec inhibiteurs de l'enzyme de conversion de l'angiotensine cause une carence en zinc excessive par rapport au traitement antihypertenseur à triple combinaison à dose fixe, qui est plus sûr pour le patient âgé.

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**Methods.** We performed a randomized longitudinal, retrospective and comparative survey on 115 hypertensive elderly patients (65 to 80 years, 40 males and 75 females), during April 2013-May 2016. The patients received fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg, as a single-tablet, for a study period of three years. Plasma copper concentrations and Copper/Zinc ratio (CZr) were measured comparatively, by atomic absorption spectrophotometry (AAS), at baseline and at the end of the study.

**Results.** The copper serum levels and plasma CZr ratio of the study group were significantly higher at the end of the study in comparison with baseline values ( $P < 0.01$ ).

**Conclusions.** Our study reveals beneficial pharmacological and therapeutic activities of triple fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg on copper mineral status in the elderly hypertensive patient with comorbidities (coronary heart disease, anemia and systemic inflammatory diseases).

**Key words:** copper serum levels, Copper/Zinc ratio, fixed-dose combination Amlodipine + Valsartan + Hydrochlorothiazide 10/320/25 mg, geriatric patient.

#### Abbreviations list

Aml + Val + HCTZ = amlodipine + valsartan + hydrochlorothiazide

ACE = angiotensin converting enzyme

AAS = atomic absorption spectrometry

CZr = Cu/Zn ratio

## INTRODUCTION

Inadequate mineral status involving the biometals copper and zinc is linked to cardiovascular homeostasis, anemia and hypertension. In previous studies, we revealed that long-time therapy with angiotensin converting enzyme (ACE) inhibitors cause excessive zinc deficiency in comparison with the triple fixed-dose combination of amlodipine + valsartan + hydrochlorothiazide (Aml + Val + HCTZ) 10/320/25 mg antihypertensive therapy, which is safer in elderly hypertensive patients<sup>1</sup>.

Numerous observational studies have revealed that zinc (Zn), copper (Cu), magnesium (Mg), and

**Objectifs.** Le but de notre étude était d'évaluer les effets pharmaco-thérapeutiques d'une combinaison à dose fixe d'amlodipine + valsartan + hydrochlorothiazide (Aml + Val + HCTZ) 10/320/25 mg sur le statut du cuivre dans une cohorte de patients gériatriques du Sud-Ouest de la Roumanie.

**Méthodes.** Nous avons mené une étude randomisée, expérimentale, longitudinale, rétrospective et comparative sur 115 hypertendus (65 à 80 ans, 40 hommes et 75 femmes), au cours d'Avril 2013-Mai 2016. Les patients ont reçu une combinaison à dose fixe de l'Aml + Val + HCTZ 10/320/25 mg en comprimé unique pour une période d'étude de trois ans. Les concentrations plasmatiques du cuivre et le rapport plasma Cu/Zn ont été mesurés comparativement par spectrométrie d'absorption atomique (AAS), au départ et à la fin de l'étude.

**Résultats.** Les niveaux du cuivre sérique et le Cu/Zn ratio (CZr) du groupe d'étude ont été significativement plus élevés à la fin de l'étude par rapport aux valeurs initiales ( $P < 0,01$ ).

**Conclusions.** Notre étude révèle des activités pharmacologiques et thérapeutiques bénéfiques de la triple combinaison à dose fixe d'Aml + Val + HCTZ 10/320/25 mg sur l'état minéral du cuivre chez les personnes âgées hypertendues atteintes de comorbidités (maladie coronarienne, anémie et maladies inflammatoires systémiques).

**Mots-clés:** niveaux de cuivre sérique, Cu/Zn ratio, triple combinaison à dose fixe d'amlodipine + valsartan + hydrochlorothiazide 10/320/25 mg, patient gériatrique.

#### Abréviations

Aml + Val + HCTZ = amlodipine + valsartan + hydrochlorothiazide

ECA = enzyme de conversion de l'angiotensine

AAS = spectrométrie d'absorption atomique

CZr = Cu/Zn ratio

manganese (Mn) in the serum have a key role in maintaining the human hypertension, especially in the geriatric patient, marked by deficiencies of trace elements<sup>2,3</sup>. There is evidence that these biometals act on the intracellular oxidative balance and the renin-angiotensin system<sup>4</sup>.

Copper is a complex trace element with pleiotropic effects, but studies of its elemental alteration in human fluids in patients are still conflicting. While some researchers found that serum copper levels in hypertensive patients were significantly increased<sup>5</sup>, other revealed decreased values<sup>6</sup>.

Due to the fact that serum copper concentrations modulate the effects of numerous reactions involved

in blood pressure regulation, maintaining an adequate copper status and a favorable Cu/Zn ratio is an important objective to achieve in clinical practice.

**THE AIM OF OUR STUDY** was to evaluate the pharmacotherapeutic effects of a fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg on the copper status in a clinical setting of geriatric patients.

## MATERIAL AND METHODS

### Study design and protocol research

We performed a three year randomized experimental, longitudinal, retrospective and comparative survey on 115 hypertensive geriatric patients (65 to 80 years, mean age  $68.4 \pm 2.6$  years). The study was conducted during April 2013-May 2016, at the Timis County Center of the Ministry of the Internal Affairs, Timisoara, allowing the enrolment of patients from several counties located in Southwestern Romania. The patients were diagnosed with arterial hypertension based on current European Society of Hypertension guidelines. The patients with medical or surgical conditions that may affect the absorption, distribution, metabolism or excretion of drugs have been excluded from the study.

The patients received fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg, as a single-tablet once-daily during the whole study period.

Plasma copper concentrations and plasma Cu/Zn ratio were measured comparatively by atomic absorption spectrophotometry (AAS), at baseline and at the end of the study.

In order to participate in this survey, all patients (40 males and 75 females) signed written consent, according to the criteria set out in the Helsinki Declaration<sup>7</sup>. Special attention was paid to early detection of micronutrient deficiencies and evaluation of dietary copper intake, by performing a carefully food survey in every case.

#### *Inclusion criteria:*

In the study were enrolled geriatric patients with diagnosis of arterial hypertension, according to current European Society of Hypertension guidelines, and with comorbidities of moderate severity.

#### *Exclusion criteria:*

Patients with use of nutritional supplements with copper content, chronic use of medications or polypharmacy were excluded from the study.

Comparative plasma copper concentrations were measured by atomic absorption spectrophotometry<sup>7</sup>. The serum Cu to Zn ratio (CZr) was determined

by using an air/acetylene flame atomic absorption spectrometer (AAS)<sup>8</sup>. All laboratory determinations were performed at the Toxicology Laboratory of the Timisoara County Emergency Clinical Hospital, Romania.

Statistical analyses were performed using the SPSS software package (version 21.0 for Windows, SPSS Inc, Chicago, IL.). Results were expressed as mean and standard deviation. The difference was considered significant when the p-value  $< 0.01$ .

## RESULTS

115 elderly patients have been enrolled in the study, 40 women (34.78%) and 75 men (65.22%). Of these, 92 completed the three year survey period, respectively 35 women (38.05%) and 57 men (61.95%). Regarding the blood pressure values, 20% of the patients had grade 1 hypertension, 70% grade 2 and 10% grade 3. All the patients have been diagnosed with arterial hypertension before their enrolment in the study. In order to obtain high accuracy data, any other pre-existing antihypertensive medication was discontinued 6 months before enrolment, as well as OTC drugs, nutritional supplements or vitamins. The use of baby aspirin for cardiovascular protection purposes was the only one allowed. The data collection was represented by three tools used to evaluate geriatric patients with hypertension:

- *Data collection sheet.*
- *Participant information sheet about the study.*
- *Informed consent form.*

Patient adherence to treatment was evaluated by electronic monitoring, using the ICeMed computer program system (Syonic services software solutions), used by most healthcare providers from Southwestern Romania. Adherence to triple antihypertensive therapy was similar for both sexes ( $p = 0.52$ ). Treatments were well tolerated, with similar overall incidence of adverse events (men: 22.8%, women: 22.2%). Among comorbidities, coronary heart disease was diagnosed in 38% of the cases, iron deficiency anemia in 42%, and systemic inflammatory diseases (lupus erythematosus and rheumatoid arthritis) in 23% of the patients, with no significant differences concerning the severity of the disease ( $p = 0.32$ ).

Reference copper serum levels ranged between 11 and 22  $\mu\text{mol/L}$ <sup>9</sup>. The copper serum levels in the study group were significantly higher at the end of the survey, in comparison with baseline values ( $p < 0.01$ ) (Table 1).

Normal serum Cu/Zn ratio ranged between 0.9 and 1.13<sup>10</sup>. Data dynamics for CZr is revealed in Table 2.

**Table 1.** Serum copper levels at baseline and after 3 year Aml + Val + HCTZ 10/320/25 mg therapy

Study Group Aml + Val + HCTZ 10/320/25 mg/day	No. Subjects	Copper level ( $\mu\text{mol/L}$ )	Statistical significance
Baseline	115	$8.2 \pm 12.1$	
After 3 years	92	$12 \pm 4.0$	$p < 0.01$

**Table 2.** Serum Cu/Zn ratio at baseline and after 3 year Aml + Val + HCTZ 10/320/25 mg therapy

Study Group Aml + Val + HCTZ 10/320/25 mg/day	No. Subjects	Serum Cu/Zn ratio CZr	Statistical significance
Baseline	115	$0.8 \pm 6.2$	
After 3 years	92	$0.9 \pm 1.4$	$p < 0.01$

## DISCUSSION

The elderly enrolled at baseline in our research were copper deficient, as an expression of nutritional microelement deficiency specific to this patient population, aspects outlined in table 1: baseline copper serum level in the study group observed in this study was  $8.2 \pm 12.1 \mu\text{mol/L}$ , with no significant difference between sexes ( $p = 0.682$ ). After 3 years of follow-up therapy with fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg, a significant rise of the values was revealed, respectively at  $12.3 \pm 4.0 \mu\text{mol/L}$ . Thus, because the values were at the lower limit of the normal range, a balanced diet with copper supplementation is highly required in geriatric patients with arterial hypertension.

Fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg is very useful in reducing blood pressure and monitoring it in case of long-term administration, due to optimal pharmacokinetic profile of the three component drugs<sup>11</sup>.

In accordance with recent researches undertaken in our country, we tried to reveal and understand sex differences in the comorbidities of patients with arterial hypertension<sup>12</sup>. In this context, we followed an important link between baseline copper deficiency and anemia in elderly female patients but further research is needed.

We consider that the knowledge of Cu/Zn ratio serum values is important for health care providers who follow-up geriatric hypertensive patients. CZr higher values and clinical importance were revealed in patients with comorbidities represented by systemic inflammatory diseases (lupus erythematosus and rheumatoid arthritis). Further research is designed to study the relationship between CZr and C-reactive protein values.

## CONCLUSIONS

Our study reveals beneficial pharmacological and therapeutic properties of triple fixed-dose combination of Aml + Val + HCTZ 10/320/25 mg on

copper mineral status in the elderly hypertensive patient with co-morbidities (coronary heart disease, anemia and systemic inflammatory diseases).

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