Chemical composition and anti-hemolytic activity of Algerian honey samples

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INTRODUCTION

Aluminum (Al) is the most abundant metal in nature. Although, it has no known biological function, Al compound has become integral part of our modern life and is widely used in processed foods, water and as a component of many cosmetic and pharmaceutical preparations.
Aluminium was only recently recognized as a possible source of human intoxication. Different clinical symptoms related to aluminium toxicity have been reported and toxicological reports revealed that Al may present a major threat for humans by causing many diseases such as dialysis dementia, Alzheimer's disease, Parkinson's disease, osteomalacia and microcytic anemia.

Management of aluminium intoxication was at the basis of many studies. Metal-targeted strategies have been proposed and different families of chelating agents have been tested. Recently, aluminium interactions with natural substances were also tested. The main goal of this study was to evaluate the protective role of honey on red blood cells (RBCs) osmotic fragility and hemoglobin (Hb) degradation under Al-induced oxidative damage in human erythrocytes.

MATERIAL AND METHODS

Honeys samples

Honey samples were collected from beekeepers in the city of Bordj Bou Arreridj, Algeria. Honey samples were given numbers prior experiment (Honey 1 for sample collected form Bir Snab and Honey 2 for sample collected from Colla). 1 g of each honey was diluted with PBS (100 mg/mL).

Phenolic profiling

Total phenolic compounds were estimated using the Folin-Ciocalteu method. Total flavonoid content in each honey sample was measured by colorimetric assay.

Isolation of human erythrocytes

After the prior informed consent, peripheral blood was obtained from healthy adult volunteers. For each analysis, 4 ml of blood were centrifuged at 2500g for 10 min at 4 °C. Platelets, leukocytes and plasma were removed by aspiration. Red Blood Cells (RBCs) were washed three times and re-suspended in phosphate buffered saline (PBS).

Studied samples

The samples tested were: negative control (50% RBC suspension: 50% PBS), positive control (50% RBC suspension: 50% PBS), ascorbic acid (50% RBC suspension: 50% ascorbic acid [50 mmol/l]), honey 1 (50% RBC suspension: 50% honey 1 [100 mg/ml]) and honey 2 (50% RBC suspension: 50% honey 2 [100 mg/ml]). RBC suspensions (positive control and samples) were incubated with 800 µL AlCl3 for 30 min. Negative control was treated with a PBS solution.

Evaluation of the antioxidant activity

All samples were evaluated simultaneous for hemoglobin concentration and cellular turbidity. In addition, kinetics of hemoglobin stability were investigated.

RESULTS AND DISCUSSION

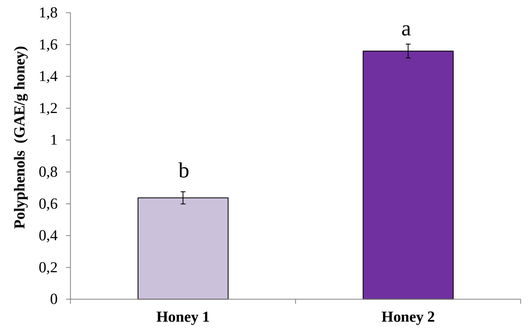

Fig. 1. Total phenolic contents of honey samples

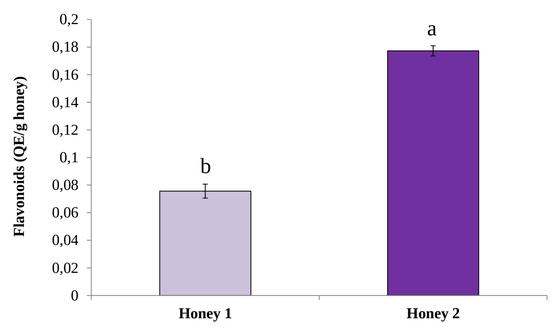

Fig. 2. Total flavonoid contents of honey samples

Tab. 1. Cellular turbidity of human RBC and Hb levels when exposed to the hypotonic conditions and oxidation by $AlCl_3$.

	Negative control	Positive control	Ascorbic acid	Honey 1	Honey 2
RBC	0.097 ± 0.038^{a}	0.039 ± 0.010^{c}	0.061 ± 0.002^{b}	$0.063 \pm 0.013^{a,b}$	$0.069 \pm 0.004^{a,b}$
Hb	1.52 ± 0.34^{a}	1.36 ± 0.13^{b}	0.87 ± 0.24^{c}	1.58 ± 0.42^{a}	1.93 ± 0.12^{a}

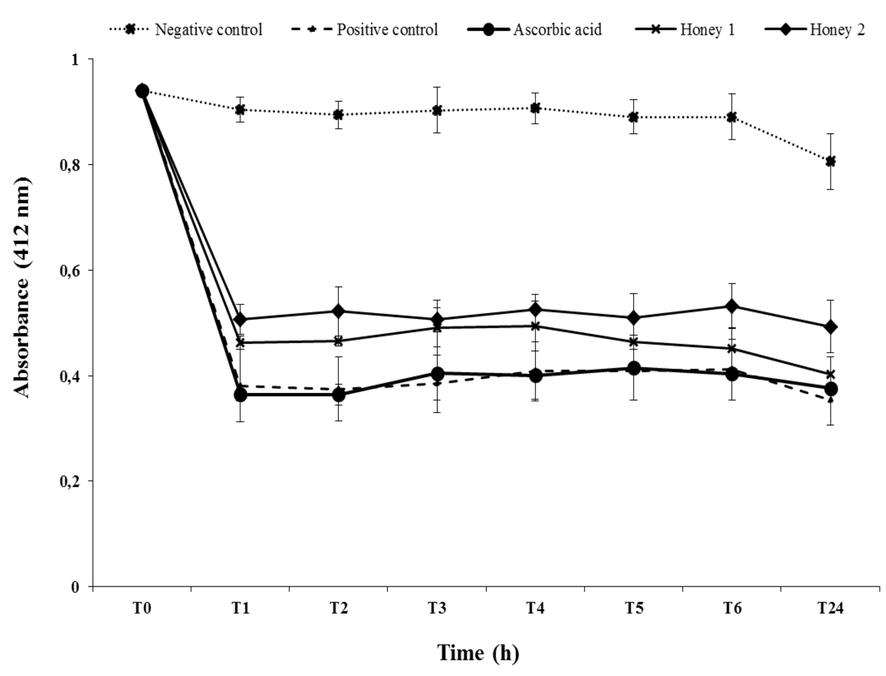

Fig. 3. Kinetics of hemoglobin breakdown exposed to $AlCl_3$.

Results of the colorimetric analysis of phenolic contents revealed that honey 2 contained the highest phenolic content (1.55 ± 0.04 mg GAE/g) compared to honey 1 (0.63 ± 0.03 mg GAE/g). As well, honey 2 showed the highest levels of flavonoid content (0.17 ± 0.003 mg QE/g) when compared to honey 1 (0.075 ± 0.005 QE/g).

The tested honeys were effective in protecting RBCs against hypotonic stress with respect to cell and Hb concentrations (Tab. 1). These findings were supported by the measurement of kinetics stability of Hb exposed to $AlCl_3$. We showed that honey, unlike ascorbic acid, significantly improved hemoglobin stability (Fig. 3).

CONCLUSION

This study showed that honeys contained important quantity of phenolic and flavonoid compounds. The tested honeys were effective in protecting erythrocytes against hypotonic stress and Al-induced adverse effects with respect to cell absorbance and hemoglobin concentration.