Evaluation of the inhibitory activities of acetylcholinesterase, α-glucosidase, pancreatic lipase and tyrosinase by phenolic extracts of honey.

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Abstract

Enzyme inhibition is one of the targeted therapeutic strategies in the treatment of Alzheimer's disease, diabetes, obesity and hyperpigmentation. The objective of this work is to evaluate the inhibitory effect of phenolic extracts of honey on key enzymes that are involved in these diseases (acetylcholinesterase, α-glucosidase, pancreatic lipase and tyrosinase). The color of the honey samples was determined spectrophotometrically and the polyphenols of monofloral and polyfloral honeys were recovered by liquid-solid extraction using amberlite XAD-4 resin. The inhibitory activities of phenolic extracts on the four enzymes were tested with six concentrations and the results are expressed as IC_{50}. The results obtained showed that the color of the tested honeys varied from extra light to dark amber. All the phenolic extracts tested exerted enzymatic inhibition. The phenolic fractions of *Myrtaceae* and polyfloral honeys showed the best anti-acetylcholinesterase, anti-α-glucosidase, anti-lipase and anti-tyrosinase activities, respectively. The inhibitory effect of these honeys was comparable to that obtained with the reference inhibitors (galanthamine, acarbose, orlistat and α-kojic acid). The correlation matrix revealed highly significant correlations between color and inhibitory activity of the honeys. Since acetylcholinesterase, α-glucosidase, pancreatic lipase, and tyrosinase are closely associated with Alzheimer's, hyperglycemia, obesity, and hyperpigmentation, the use of honey polyphenols may have a beneficial effect in treating these diseases.

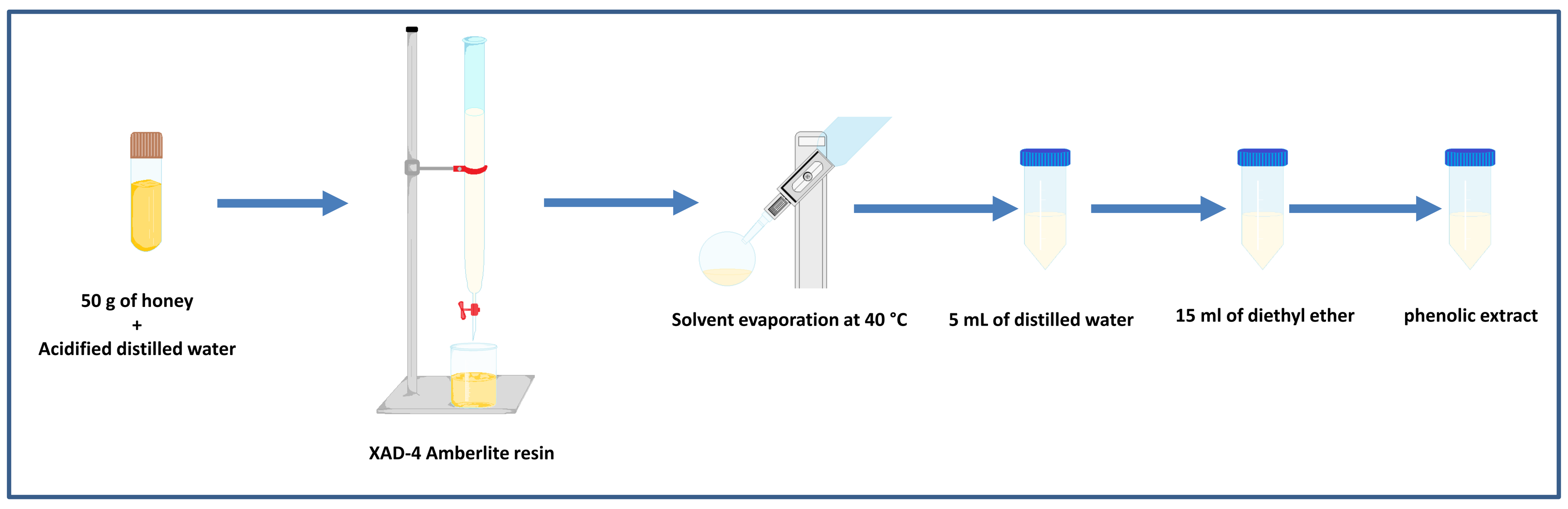
Introduction

Today, neurodegenerative diseases, such as Alzheimer's disease, are a huge health problem as they affect the majority of the elderly and are incurable. Acetylcholinesterase (AChE) inhibition is one of the targets for the treatment of Alzheimer's disease. The inhibitory effect of honey on AChE is attributed mainly to phenolic compounds that allow restoration of deficient cholinergic neurotransmission **(Ahrari-Roodi et al., 2018; Muhammad et al., 2015)**. Diabetes is a disease that is of great concern. The α-glucosidase is the main enzyme responsible for hydrolyzing carbohydrates and increasing blood glucose levels. Phenolic compounds in honey exert inhibition on these digestive enzymes and consequently control hyperglycemia **(Lakshmana-Senthil et al., 2019; Zaidi et al., 2019)**. Pancreatic lipase plays an important role in the hydrolysis of triglycerides into glycerol and fatty acids. Several studies have reported that natural polyphenols can inhibit pancreatic lipase and subsequently influence fat digestion and decrease energy intake **(McDougall et al., 2009)**. Melanin formation in human skin occurs in the presence of a catalyst (tyrosinase) and UV light, which can lead to hyperpigmentation. Therefore, there is a need to find bioactive and harmless compounds from natural sources with tyrosinase inhibitory activity **(Arrowitz et al., 2019)**. In search of natural and sustainable treatments for the mentioned diseases, the inhibitory activities of BSA, acetylcholinesterase, α-glucosidase, pancreatic lipase and tyrosinase of phenolic extracts from honey are investigated.

Samples

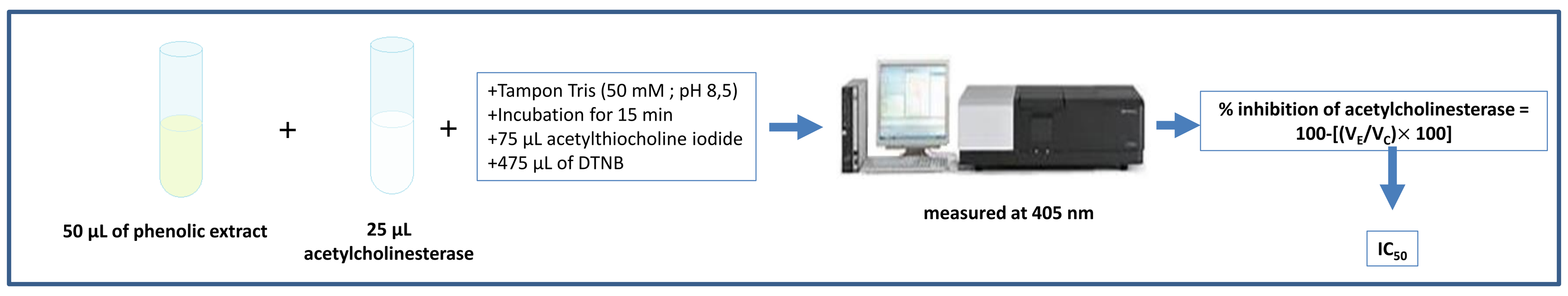
A total of 26 honeys are provided by beekeepers from 16 wilayas of Algeria from East to West (Skikda, Batna, Jijel, Sétif, Bejaia, Bordj Bouarrerid, Tizi-Ouzou, Boumerdès, Djelfa, Médéa, Aïn Defla, Chlef, El Bayadh, Mostaganem, Naâma and Tlemcen). The samples were stored at room temperature and protected from light and humidity. the color intensity of the studied honeys is determined with the method Pfund.

Extraction of phenolic compounds

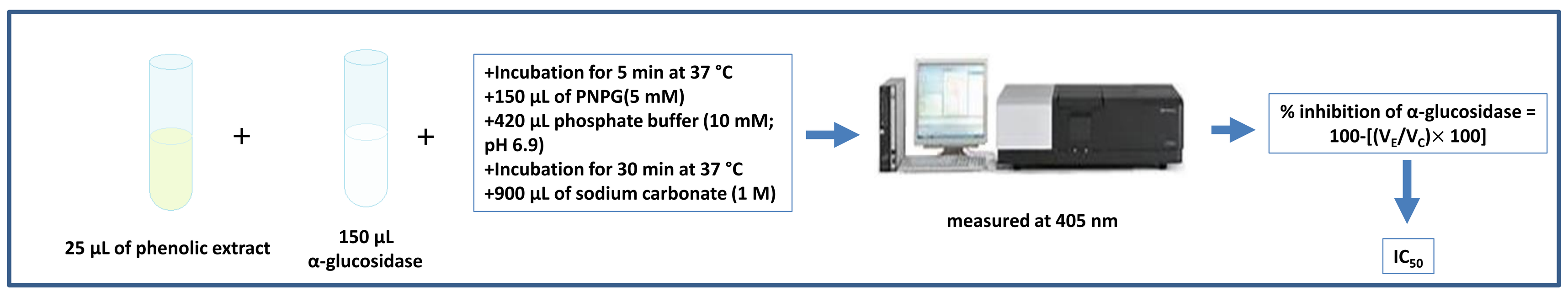


Enzymatic inhibition

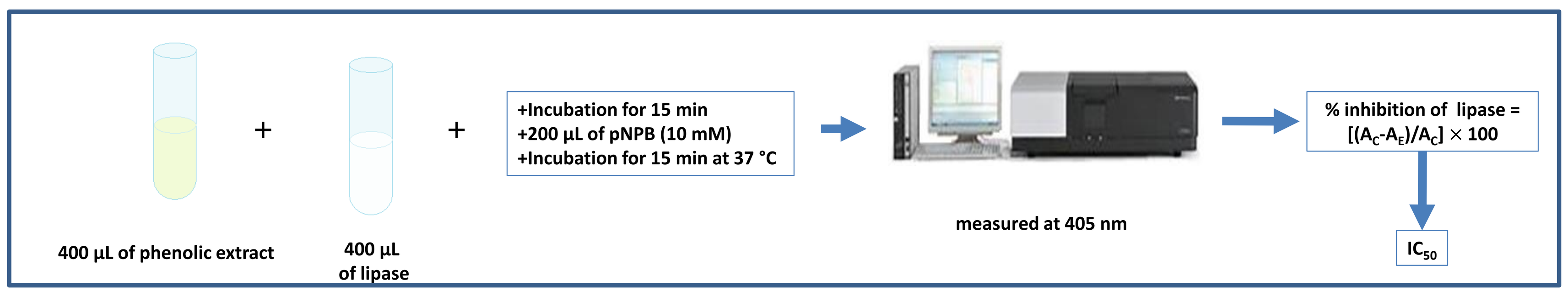
Acetylcholinesterase



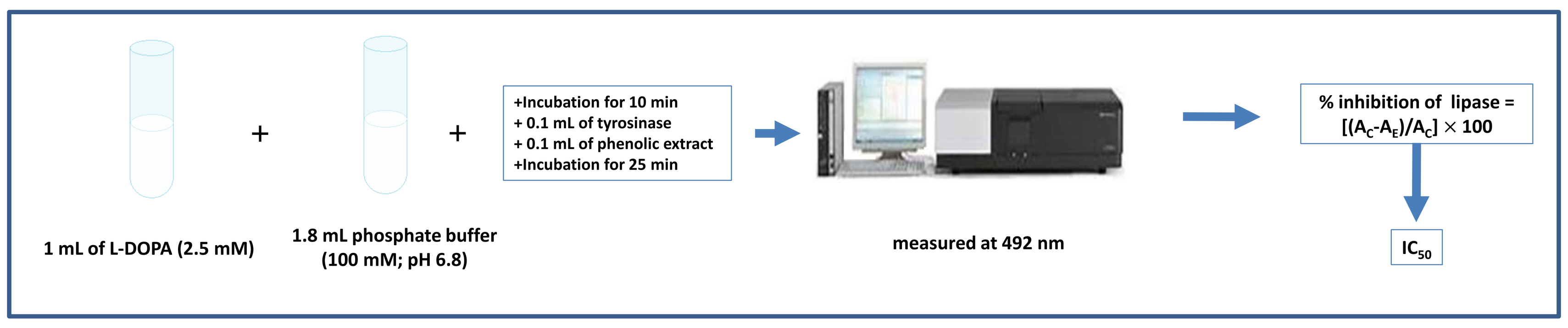
α-glucosidase



Pancreatic lipase



Tyrosinase



Statistical analysis

Student's t-test, ANOVA (analysis of variance with a single classification criterion) and correlation matrix are applied to the data presented and are performed using STATISTICA 7.1 software.

IC50 of phenolic extracts from honey samples to inhibit enzymes (AChE, α-glycosidase, lipase and tyrosinase) are calculated by regression analysis using Excel 2013.

Results and discussion

Table 1 : Intensities and color names (Pfund scale) of analyzed honeys

Sample		Color intensity mm Pfund	Name of the color	Sample		Color intensity mm Pfund	Name of the color
Monofloral honey	H2	150±3 **f**	Dark amber	Polyfloral honey	H1	191±0 **f**	Dark amber
	H4	412±4 **b**	Dark amber		H3	210±1 **e**	Dark amber
	H5	402±6 **c**	Dark amber		H7	115±0 **h**	Dark amber
	H6	112±3 **i**	Amber		H8	342±1 **c**	Dark amber
	H12	255±3 **d**	Dark amber		H9	61±4 **k**	Light amber
	H13	195±7 **e**	Dark amber		H10	520±9 **b**	Dark amber
	H14	53±3 **j**	Light amber		H11	619±6 **a**	Dark amber
	H15	133±4 **g**	Dark amber		H16	73±2 **j**	Light amber
	H18	46±1 **k**	Extra light amber		H17	174±2 **g**	Dark amber
	H20	123±1 **h**	Dark amber		H19	331±1 **d**	Dark amber
	H22	462±0 **a**	Dark amber		H21	62±1 **k**	Light amber
	H23	130±0 **g**	Dark amber		H24	108±3 **hi**	Amber
	H25	33±1 **l**	White		H26	107±2 **i**	Amber

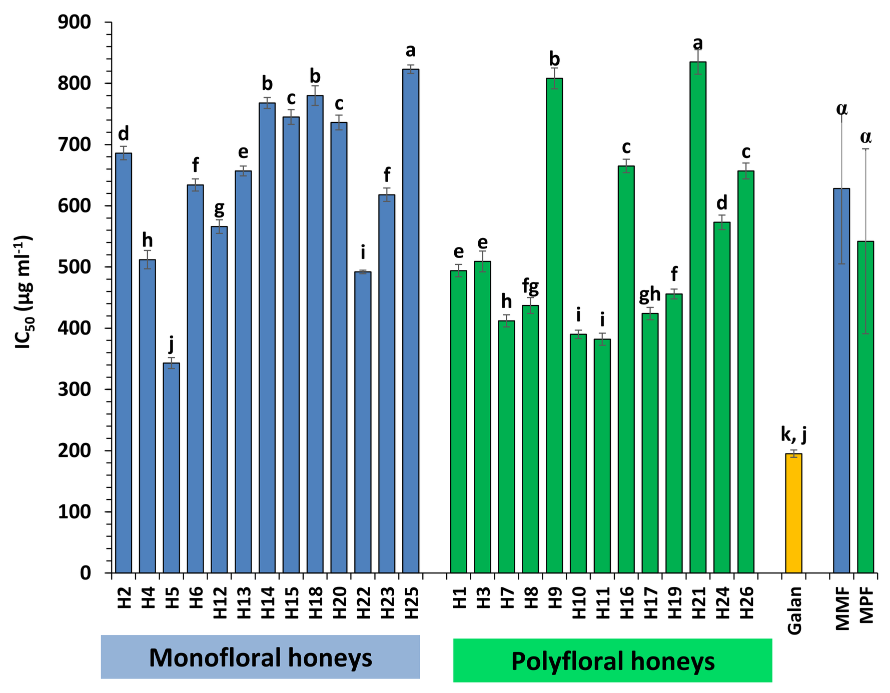


Figure 1 : IC_{50} values of phenolic extracts of honeys analyzed for acetylcholinesterase inhibition.

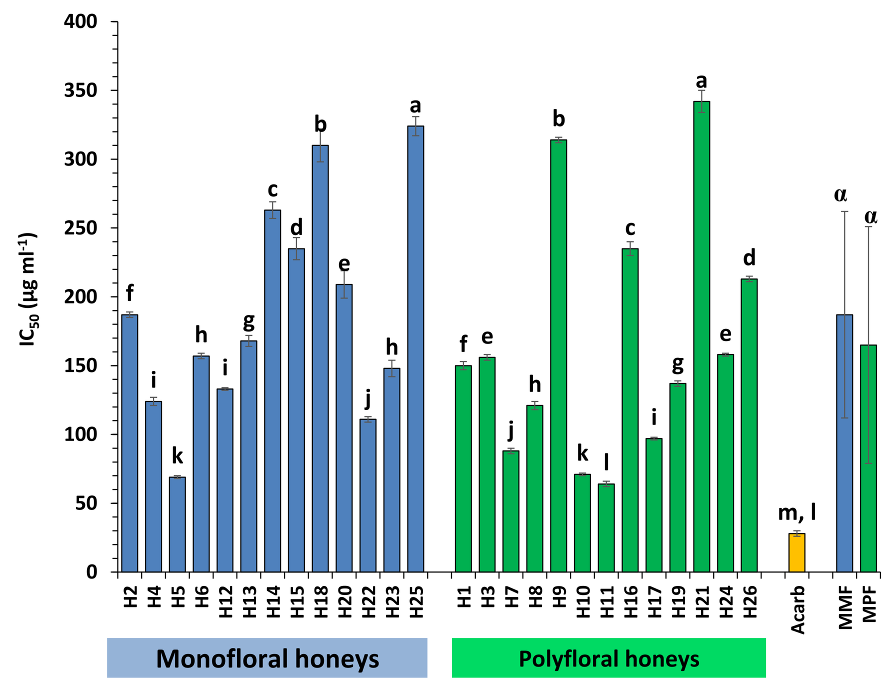


Figure 2 : IC_{50} values of phenolic extracts of honeys analyzed for α-glucosidase inhibition.

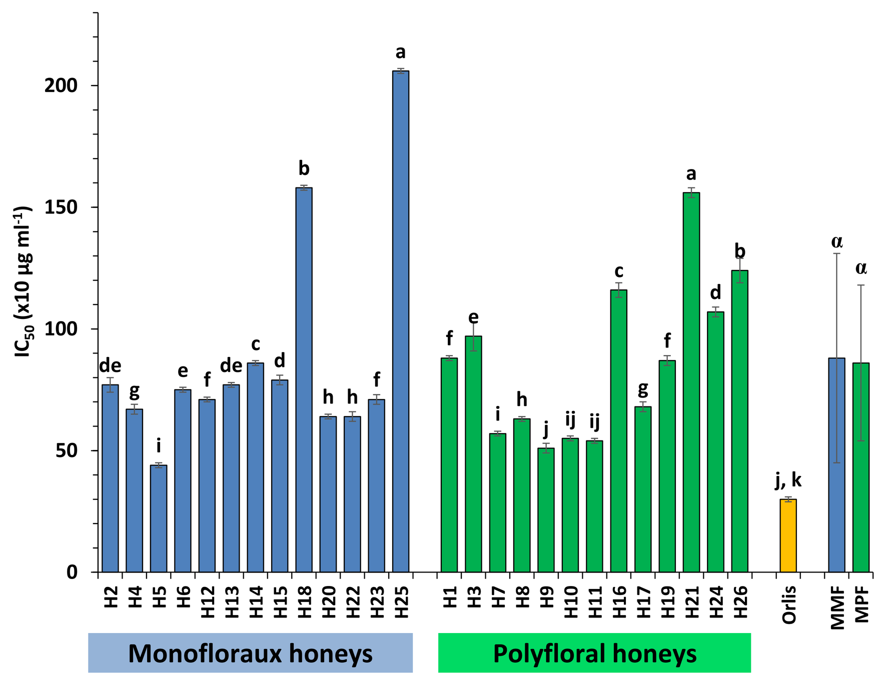


Figure 3 : IC_{50} values of phenolic extracts of honeys analyzed for lipase inhibition.

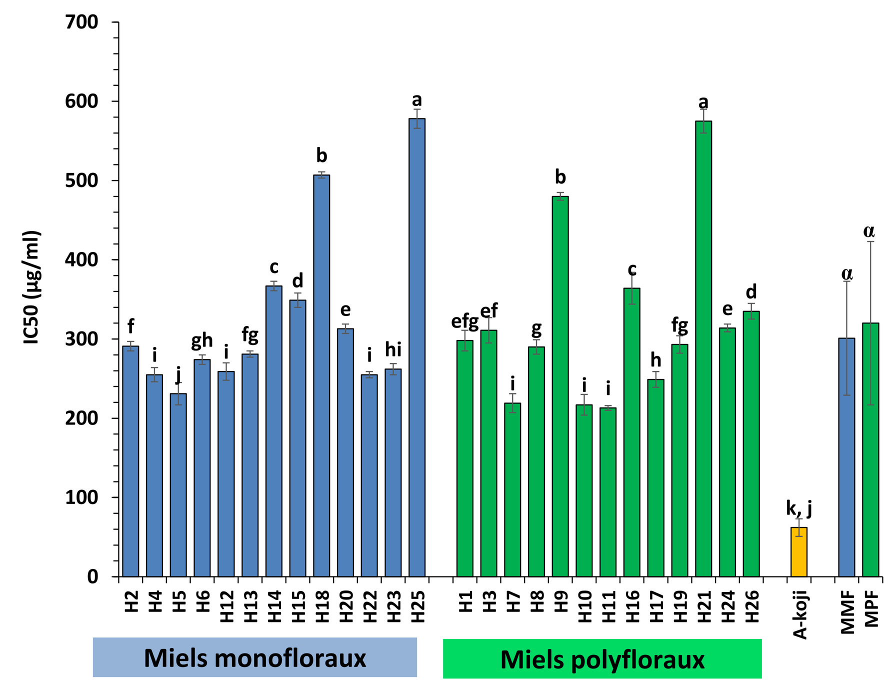


Figure 4 : IC_{50} values of phenolic extracts of honeys analyzed for tyrosinase inhibition.

Conclusion

The phenolic extracts of all the honeys analyzed show acetylcholinesterase and α-glucosidase inhibitory activity with IC_{50} values ranging from 343 to 835 µg mL^{-1} and 64 to 343 µg mL^{-1}, respectively. The extracts also reveal an inhibitory capacity of pancreatic lipase and tyrosinase with IC_{50} values of 440 to 2060 µg mL^{-1} and 213 to 578 µg mL^{-1}, respectively.

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