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SHORT COMMUNICATION



Nigerian propolis: chemical composition, antioxidant activity and α -amylase and α -glucosidase inhibition

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ABSTRACT

Propolis is an attractive natural ingredient to design health products due to its pharmacological effects. Our chemical investigation of a polar extract of Nigerian propolis (NP) led the isolation and identification of five isoflavonoids (**1-4**, **6**), one diarylpropane (**5**) and one prenylated flavanone (**7**) by the combination of chromatographic and spectroscopic techniques. Compounds **1**, **4** and **7** were found to be the main markers in NP (8.0, 5.0 and 4.0 mg/g of dry extract, respectively). Moreover, NP and its phenolic constituents exhibited *in vitro* free radical scavenging activity together with a promising antidiabetic effect against α -amylase and α -glucosidase enzymes. Finally, NP showed also a moderate inhibition of *Helicobacter pylori* growth. These results suggested that NP could be a good candidate in nutraceuticals and food products.

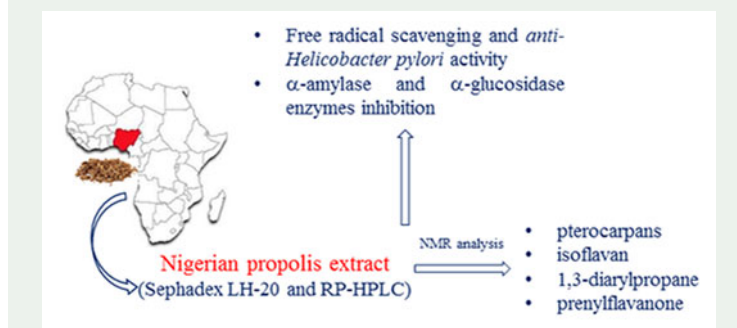
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Nigerian propolis; polyphenols; free-radical scavenging activity; α -amylase and α -glucosidase enzymes; *Helicobacter pylori*



1. Introduction

Nowadays the market of nutraceuticals, functional foods, and foods supplement are rising fast worldwide both their nutritional value and ability to improve human health.

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Their effects are linked to the presence of bioactive compounds (BACs) including phytochemicals, antioxidants and probiotics that may play a crucial role in the prevention and treatment of age-related neurodegeneration and chronic diseases such as stroke, hypertension, arthritis, heart diseases, diabetes, obesity, cancer, Alzheimer's and Parkinson's disease (Sansone et al. 2018). Propolis, a resinous material collected by honeybees from different plants and which they use as defense substance for bee's hives (Piccinelli et al. 2011), is a rich source of BACs. Resin bee wax, aromatic and essential oils, pollen, and organic compounds (polyphenols, terpenes, and steroids) together vitamins B, C, E, and several minerals (magnesium, iron, calcium, potassium, sodium, zinc, and manganese) are the mixture substances present in propolis (Pasupuleti et al. 2017). Nevertheless, the chemical composition of propolis, with provides a wide range of functional properties including antioxidant, antibacterial, anti-inflammatory, and anticancer, is highly variable depending upon the season, the species of bee and the flora around the hive (Alday et al. 2015). Furthermore, some studies have highlighted a correlation between chemical composition and specific biological activity of propolis (Rufatto et al. 2018). Thus, the chemical standardization of propolis is a need for its use as a functional ingredient. Currently, to obtain new active ingredients to developing health products, there is a renewed interest in the research of composition and biological activities of unknown propolis. In Nigeria, the Fulani Nomads use propolis to manage diabetes and as an emollient in the treatment of measles, ringworm, and chickenpox disease. The marketed Nigerian propolis exhibited hypoglycemic, hypolipidemic, hepatoprotective, and pancreatoprotective properties on alloxantin-induced diabetic rats (Babatunde et al. 2015). Moreover, xanthenes, triterpenes, and isoflavonoids against *Trypanosoma brucei* have been isolated from various Nigerian propolis samples. The main botanical source of this resin has been hypothesized to belong to *Dalbergia* and *Macaranga spp.* (Omar et al. 2016, 2017). This paper aimed to investigate the chemical composition and the biological activity of a polar extract from Nigerian propolis (NP) to support its potential use as a functional ingredient in the pharmaceutical and food industry.

2. Results and discussion

The chemical investigation of NP led to the isolation of seven phenols and their structures (Figure 1) were elucidated by NMR and MS data in comparison to those reported in the literature. They include astrapterocarpan (**1**) (Spencer et al. 1991), 3,8-dihydroxy-9-methoxy-pterocarpan (**2**), vesticarpan (**3**), medicarpan (**4**), vestitol (**6**) (Piccinelli et al. 2011), broussonin B (**5**) (de Almeida et al. 1999), and 8-prenylnaringenin (**7**) (Omar et al. 2016). Some of them (**2-4** and **6-7**) are reported in Brazilian, Cuban and Nigerian red propolis (Piccinelli et al. 2011; Omar et al. 2016) while **1** and **5** are new in propolis. The compounds **1**, **4** and **7** were found to be the main markers of NP (8.0, 5.0 and 4.0 mg/g of dry extract, respectively). The Total Phenolic Content, expressed as gallic acid equivalent, was 69.9 mg/g in the extract (Table S1).

The antioxidant effect of NP and its constituents was evaluated by TEAC (Trolox Equivalent Antioxidant Capacity) assay (Kerbab et al. 2019), one of the methods used

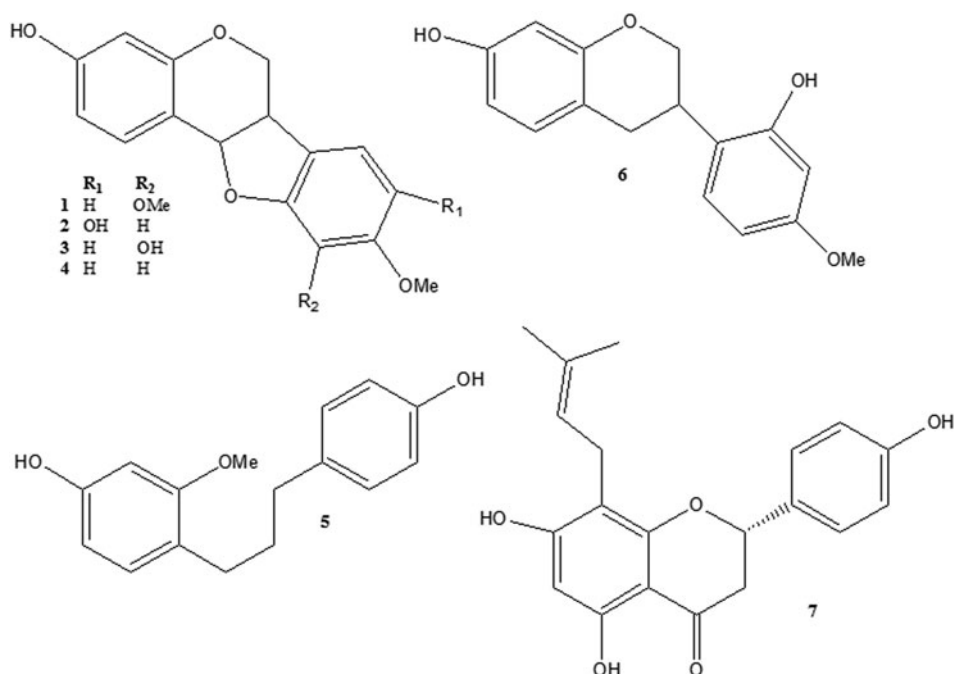


Figure 1. Chemical structures of compounds 1–7 isolated from NP.

in the measurement of AOC (antioxidant capacity) of food supplements (Prior et al. 2005). NP was more potent to scavenging ABTS^{*+} radical in comparison with BHT, a known antioxidant used as a positive control (Table S1). In the same test, all tested compounds (1–7) exhibited ABTS^{*+} scavenging activity lower than that of parental extract (Table S1), suggesting a potential synergy in the action of all antioxidant molecules.

Nowadays, the involvement of oxidative stress in the pathogenesis of diabetes mellitus, in particular, type 2, was extensively investigated (Asmat et al. 2016). So, the antioxidants may play a focal role in the treatment and prevention of type 2 diabetes and its complications. Therefore, here we have evaluated *in vitro* the hypoglycemic effect of NP and its isolated compounds by inhibition of α -amylase and α -glucosidase enzymes. Both enzymes were involved in the carbohydrate digestion and their suppression would delay the absorption of glucose with a decreasing of postprandial hyperglycemia (Kerbab et al. 2019). The NP inhibited both enzymes with a potency about six times lower than that positive control, acarbose ($IC_{50} = 368.0 \pm 55.0$ and $55.0 \pm 2.2 \mu\text{g/mL}$ for α -amylase, and 51.1 ± 3.5 and $7.5 \pm 1.6 \mu\text{g/mL}$ for α -glucosidase, respectively). All compounds exhibited a higher inhibitory effect against α -glucosidase (Figure S1(a)) respect to α -amylase (Figure S1(b)). In particular, 2 and 7 were strong inhibitors of α -glucosidase with IC_{50} values lower than that of acarbose ($IC_{50} = 5.0, 5.2, \text{ and } 7.5 \mu\text{g/mL}$, respectively).

Recent studies have shown that some polyphenols possess anti-*H. pylori* activity, a human pathogen associated with several gastric disorders (Liu et al. 2018). Moreover, the *H. pylori* infection in diabetic patients may be linked with inflammations, insulin resistance, and diabetic complications (Hosseininasab-Nodoushan and Nabavi 2019).

Accordingly, NP was tested *in vitro* against clinical and reference *H. pylori* strains by agar dilution method. NP exhibited a similar inhibitory activity on both strains with a MIC value about eightfold higher than that of positive control, amoxicillin (Table S2). Furthermore, the total growth inhibition of *H. pylori* observed for the incubation time assayed, suggest a bactericidal effect, expressed as MBC (Table S2).

3. Conclusion

Several phenols (1–7) have isolated from a polar extract from Nigerian propolis. The extract exhibited high free-radical scavenging activity together with a promising anti-diabetic effect, suppressing α -amylase and α -glucosidase enzymes, that may be correlated to its chemical constituents. Furthermore, NP showed also an interesting antimicrobial effect against *H. pylori*. These preliminary data suggested that a polar extract from Nigerian propolis might be a good candidate as an active ingredient for developing natural health products.

Disclosure statement

No potential conflict of interest was reported by the authors.

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