

ABSTRACT

Asthma is a chronic inflammatory disease and many people are living with it worldwide. Although conventional drugs demonstrate beneficial effects, their adverse effects limit their long term use. Reliance on the use of medicinal plants has also increased. It is therefore pertinent to investigate the chemical constituents of some plants used in the management of asthma in Alternative Medicine. Such plants include: the leaves of *Adansonia digitata* (AD), *Calotropis procera* (CP), *Coix lacrymajobi* (CL), *Datura metel* (DM), *Deinbollia pinnata* (DP), aerial parts of *Euphorbia hirta* (EH) and roots of *Pterocarpus osun* (PO). This study was carried out to obtain and characterize the essential oils from the listed plants and also to extract, fractionate, analyze and investigate the *in vitro* antioxidant, anti-inflammatory and antibacterial activities of these medicinal plants, since oxidative stress, inflammation and bacterial infections are major contributing factors to the development of asthma. Essential oils were extracted from AD, CP, CL, DM, DP, EH and PO by modified hydrodistillation and Gas Chromatography–Mass Spectrometry (GC-MS) analyses of the essential oils were carried out with appropriate temperature programs on HP5 column with 5% phenylmethylsiloxane as the stationary phase. Collection of the essential oils were in two modes; a single continuous collection over four hours and hourly collections over the same period. The 3 plant parts with highest contents of known anti-inflammatory compounds in the essential oils as indicated by GC-MS were selected for further investigations. These compounds include phytol, n-hexadecanoic acid, oleic acid, polyunsaturated fatty acids (PUFA), 2-Pentadecanone, 6,10, 14-trimethyl and squalene hence solvent extractions, analyses using High Performance Liquid Chromatography coupled to Photodiode Array Detector (HPLC-PDA) and *in vitro* anti-inflammatory, antioxidant and antibacterial assays were carried out on AD, CP and EH. 50% aqueous methanol (aq.MeOH) and soxhlet extraction methods were used to fractionate the components of the selected plants into non-polar, medium polar and polar fractions. Solid Phase Extraction (SPE) was used for the cleanup of the methanol extracts and five different SPE fractions were obtained for each of the plants. The antioxidant assay was carried out based on the scavenging activities of the plant extracts on 2, 2-diphenyl-1-picrylhydrazyl (DPPH) free radicals. Anti-inflammatory assay investigated the effects of the plants on the production of nitric oxide (which is a compound involved in inflammation) induced by Lipopolysaccharide (LPS) in RAW 264.7 cells. The antibacterial potentials of the plants were evaluated against 5 bacterial strains: *Acinetobacter baumannii*, *Bacillus cereus*, *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* and minimum inhibition concentrations (MIC) of the extracts were determined by resazurin based microtiter dilution assay. Antioxidant activities, determined by percentage inhibitions (%I) were obtained for all the extracts and fractions investigated and 50% Inhibition Concentrations were calculated for those with up to 50% minimum inhibitions. The extracts and fractions with the highest antioxidant activities were: ethyl acetate fraction of AD {AD(EtOAc)} from 50% aq.MeOH [%I: $89.895 \pm 0.001\%$, IC_{50} : 3.48×10^{-2} mg/mL]; methanol fraction of AD {AD(MeOH)} from soxhlet [%I: $89.633 \pm 0.226\%$, IC_{50} : 4.02×10^{-2} mg/mL] and AD(MeOH) from SPE [%I: $94.962 \pm 0.004\%$, IC_{50} : 2.93×10^{-2} mg/mL]. Antioxidants are known to be very effective in the management of asthma hence isolation of an antioxidant from the plants were carried out using reversed phase HPLC-PDA and quercetin was isolated from the ethyl acetate fraction of *Adansonia digitata* using bioactivity-guided isolation method. The anti-inflammatory activities were concentration–dependent and the highest values were obtained at 100 µg/mL for all extracts. The activities were in the order: AD > EH > CP at all concentrations. Antibacterial results revealed that seven extracts had activities against *Staphylococcus aureus* which had been implicated in asthma while six of them had activities against *Bacillus cereus*. The findings established the presence of some bioactive compounds in the plants which are useful in the management of asthma. It also suggests that the plants are strong antioxidants, suppress inflammation and contain antibacterial compounds hence could be highly effective in the management of asthma in Alternative Medicine.

Keywords: Asthma, essential oil, inflammation, medicinal plants

