MOLECULAR PROFILING OF BIOACTIVE CONSTITUENTS FROM *MONOTHECA BUXIFOLIA* (FALC.) FRUIT

Abstract

This Ph.D. thesis describes the phytochemical and pharmacological investigation of *Monotheca buxifolia* fruit, used as laxative, digestive, purgative, vermicidal, antipyretic, and for the treatment of gastritis and urinary tract infection without scientific evidence(s).

In the present research work, *Monotheca buxifolia* hydroethanolic extract (MBHE), its sub-fractions and isolated compounds were screened for various biological activities, to provide a valid scientific rationale to its ethno-medicinal uses.

The MBHE and its fractions showed moderate antibacterial potential with maximum activity exhibited by *n*-hexane (69.56 and 60%) and ethyl acetate fractions (69.56 and 60%) against *P. aeruginosa* and *E. coli* respectively while other fractions showed mild antibacterial activity. Similarly the MBHE and its fractions showed a mild antifungal and no leishmanicidal potential.

Significant cytotoxicity against brine shrimp was observed for ethyl acetate and chloroform fractions with LD_{50} values of 14.74 and 31.22 µg/mL respectively. The MBHE, aqueous and *n*-hexane fractions exhibited moderate cytotoxicity with LD_{50} values of 147.99, 117.06 and 279.95 µg/mL respectively. The phytotoxic potential observed against *Lemna minor*, was significant at dose of 1000 µg/mL and in order of chloroform > aqueous > *n*-butanol > ethyl acetate fractions. A mild insecticidal activity was showed by MBHE, chloroform, ethyl acetate and *n*-hexane fractions. The MBHE and its fractions were found inactive in protein antiglycation assay.

In urease inhibition assay, ethyl acetate fraction showed a moderate inhibition (IC_{50} value of 151.3 µg/mL) while other fractions exhibited mild inhibition. Among the isolated compounds, compound **9** showed significant inhibition (IC_{50} value of 51.6 µg/mL), while mild activity was observed for the rest of the compounds. In acetylcholinesterase and α-chymotrypsin inhibitory assays all the tested samples were found insignificant.

A significant anticancer activity was observed for compound **9** (55.55%) against PC3 cell lines, while compound **1** exhibited the least (5.34%) cytotoxic potential against NIH 3T3 cell lines at 100 µM concentration. A moderate immune modulatory activity was also observed for compound **7**.

The MBHE showed promising *in-vivo* anti-pyretic, analgesic, and anti-inflammatory activities at test dose of 150 mg/kg. MBHE also exhibited laxative activity in both charcoal meal transit and laxative models. The laxative effect was partially antagonized by atropine. Furthermore, MBHE exhibited significant hepatoprotective activity against isoniazid and rifampicin induced hepatotoxicity in rats.

In phytochemical investigation, ethyl acetate fraction was subjected to various chromatographic techniques. Structures of the isolated compounds were elucidated using advanced spectroscopic and spectrometric techniques i.e. ^{1}H-NMR, ^{13}C-NMR, COSY, HMBC, NOESY, HSQC, IR, UV, EI-MS, HR-EI-MS, FAB-MS, HR-FAB-MS and $[\alpha]^{26}_{D}$. Among the isolated compounds, 4 were new, 1 was new natural product and 4 were previously reported but first time isolated from this plant. The first time isolated from natural sources was buxifoline-A (**1**), new were buxifoline-B (**2**), buxitriol (**3**), buxilide (**4**) and buxiglucoside (**5**) while the other 4 compounds were oleanolic acid (**6**), glucosidic β-sitosterol (**7**), 2-hydroxy- epikatonic acid (**8**) and iso-quercetin (**9**).