PREFACE

India is renowned for its rich heritage of herbal medicinal knowledge. In India, approximately 80% of the rural population uses medicinal herbs or indigenous medical systems. Among the numerous medicinal plants that originate from India, *Ichnocarpus frutescens* and *Hemidesmus indicus* are notable for their various traditional uses in treating a range of ailments. This research focuses on the extraction, purification, and characterization of bioactive compounds from root of *Hemidesmus indicus* and *Ichnocarpus frutescens*. The antidiabetic potential of these compounds was evaluated using *invitro* and *insilico* studies, targeting α-Amylase (α-A) and α-Glucosidase (α-G) inhibition. Additionally, *invitro* glucose uptake and wound healing assays were conducted to assess their therapeutic efficacy. The antioxidant potential of the plant extracts and isolated compounds was also investigated. The anticancer properties of the phytoconstituents were explored through biological and computational studies. Additionally, zinc oxide nanoparticles were also synthesized from the methanolic root extract of *Hemidesmus indicus* and *Ichnocarpus frutescens*, and their biological evaluation, including antioxidant, antidiabetic, and anticancer potentials, was conducted. Bioactive compounds silychristin was isolated from the roots of *Hemidesmus indicus* and hyperoside was isolated from *Ichnocarpus frutescens* using Column chromatography and were characterized using TLC, HPLC, 1H and 13C NMR, and HR-MS techniques. Hyperoside and silychristin exhibited significantly inhibitory effects against α-A and α-G than the standard drug, acarbose. The NBDG glucose uptake assay indicated that both compounds significantly enhanced glucose uptake compared to the control, with silychristin showing a more pronounced effect. DPPH assay, revealed that hyperoside exhibited the strongest antioxidant activity, while Silychristin and ascorbic acid showed similar antioxidant activities.

Biological evaluation and computational studies were conducted to assess the anticancer potential of phytoconstituents in the A549 lung cancer cell line. The studies included apoptosis, cell cycle analysis, and gene expression analysis. Silychristin and hyperoside exhibited 50% cell growth inhibition at concentrations of 96.46 µg/mL and 74.61µg/mL, respectively. The Annexin V apoptosis assay revealed that Hyperoside marked increase in late apoptotic and necrotic cell populations. Silychristin, while less potent, still induced apoptosis. Cell cycle analysis revealed distinct mechanisms of action for the two compounds. Hyperoside caused G2-M phase arrest, indicating interference with mitotic progression, while silychristin induced a strong G0-G1 phase arrest, effectively blocking DNA synthesis. Gene expression analysis indicated that the Bcl-2 gene was downregulated, whereas p21, p53, and Caspase-3 genes were upregulated compared to the housekeeping gene. Hyperoside treatment increased p21 expression 9-fold and silychristin 7-fold. For p53 expression, hyperoside caused a 2.4-fold increase, while silychristin led to a 3.7-fold increase. Caspase-3 expression was enhanced 4-fold by hyperoside and 6.7-fold by silychristin.

The zinc oxide nanoparticles using root extracts of *Hemidesmus indicus* (HI ZnOxNP) and *Ichnocarpus frutescens* (IF ZnOxNP) were also explored for their therapeutic applications, including antioxidant antidiabetic and anticancer potential. The synthesized nanoparticles were characterized through UV-VIS Spectroscopy, Particle Size Distribution and Zeta Potential Analysis, FT-IR Spectroscopy, Scanning Electron Microscopy (SEM), and Transmission Electron Microscopy (TEM). HI ZnOxNP exhibited superior free radical scavenging and ferric ion-reducing power compared to IF ZnOxNP. HI ZnOxNP demonstrated stronger inhibition of α-A, while IF ZnOxNP exhibited more potent inhibition of α-G.The anticancer studies on the A549 lung cancer cell line revealed that both HI ZnOxNP and IF ZnOxNP exhibit significantly higher

anticancer potential compared to their parent root extracts. Flow cytometry analysis demonstrated that HI ZnOxNP induced apoptosis more effectively than IF ZnOxNP, with a predominant late apoptotic response and minimal necrosis, highlighting its targeted cytotoxic mechanism. Both ZnOxNP also caused cell cycle arrest, with HI ZnOxNP inducing G0/G1 phase arrest and IF ZnOxNP exhibiting dual-phase inhibition, disrupting both DNA synthesis (S phase) and mitosis (G2/M phase). The combination of ZnOxNP with natural phytochemicals offers a novel strategy to improve the bioavailability, cellular uptake, and therapeutic efficacy of plant-derived compounds. These findings contribute to a deeper understanding of the therapeutic applications of *Hemidesmus indicus* and *Ichnocarpus frutescens*, suggesting their potential in treating type 2 diabetes, oxidative stress-related conditions, and cancer.

Keywords: *Hemidesmus indicus, Ichnocarpus frutescens, Bioactive Compounds, Cancer, Type 2 diabetes.*