STUDY ON PHYTOCHEMICAL AND ANTIMICROBIAL PROPERTIES OF WHITE INDIAN MULBERRY (Morus alba)

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ABSTRACT

Different plants are rich source of medicines. Since old days, Ayurveda and other disciplines reported the various pharmacological properties of naturally occurring plants against certain specific diseases. The Scientific Name of White Indian Mulberry is Morus alba. Currently, increasing health concern urged the researchers to revitalize the natural products and to alleviate the diseases without harming the body. In spite of medicinal uses of natural products, health supplements from natural products and their use in diet are gaining importance. The objective of this review is to unveil the phytochemistry and antimicrobial activities of Morus alba. The leaves were dried under room temperature and then extracted with cold water, hot water and ethanol (99.7% vol./vol.). The extracts were concentrated using rotary evaporator and kept in desiccators for further analysis. The antibacterial and antifungal activities of aqueous (hot and cold) and ethanol extracts of white mulberry plant (M. alba) were carried out in vitro by agar diffusion-method against some human pathogenic microbes. the Bacitracin and streptomycin were used as the standard reference antibiotics.

KEYWORDS : White mulberry, Morus alba, Phytochemistry, antibacterial, antifungal, ethanolic extract and organisms.

INTRODUCTION

Conventional medicines show reliance on phytochemicals rich plants extracts to cure different maladies because medicines obtained from natural origin are considered to be less toxic and free from undesirable effects as compared to synthetic ones. Genus Morus (Mulberry) is an example that contains more than 150 species, Morus alba L. (white mulberry) is dominant specie among them . *M. alba* is monoecious, deciduous tree and is of medium size with a height of about 30 m and width of about 1.8 m, it is distributed throughout Asia, Africa, Europe and South and North America and found in wide range of tropical areas and in hilly areas of Himalayas at the height of 3300 m. *M. alba* leaves have antioxidant components, which includes rutin, isoquercitrin, astragalin and quercetin-3-(6- malonyl) glucoside among which quercetin - 3-(6-malonyl) glucoside is most abundant in dried mulberry leaf extract. *M.*

alba extracts have 13 known compounds. Koshihara et al. (1984) studied the selective inhibitory effect of caffeic acid on leukotriene biosynthesis and concluded that *M. alba* has high amount of caffeic acid, which selectively inhibits leukotriene biosynthesis, that appreciably play a vital role in various diseases like asthma, allergic reactions and inflammation. The fruits contain anthocyanins, a natural food colourants and modulators of mechanisms for various diseases. Due to increasing demand for natural food colourants, their significance in the food industry is increasing. Anthocyanins are responsible for the attractive colours of fresh plant foods, producing colours such as orange, red, purple, black and blue. They are water-soluble and easily extractable. The aim of this study was to evaluate the phytochemical constituents and antimicrobial properties of Morus alba.

MATERIALS & METHODS:

The two variants of white mulberry leaves (M. alba) were collected. . The leaves were kept away from sun rays and air dried at room temperature. They were milled to powder and stored in air tight containers at room temperature until required for further analysis.

The powdered leaves were subjected to phytochemical screening for the presence of the alkaloids, tannins, saponins, steroids using standard photochemical protocol used by Brain and Turner (1975). *M. alba* leaves have antioxidant components, which includes rutin, isoquercitrin, astragalin and quercetin-3-(6- malonyl) glucoside among which quercetin - 3-(6-malonyl) glucoside is most abundant in dried mulberry leaf extract (Katsube et al., 2006). *M. alba* extracts have 13 known compounds. Koshihara et al. (1984) studied the selective inhibitory effect of caffeic acid on leukotriene biosynthesis and concluded that *M. alba* has high amount of caffeic acid, which selectively inhibits leukotriene biosynthesis, that appreciably play a vital role in various diseases like asthma, allergic reactions and inflammation.

The Paste of plant material was mixed with 5 % Na2CO3 solution and transferred to a 500 ml flask, by adding 50 ml of chloroform. The solution was refluxed for 20 minutes, cooled, filtered and transferred to the agitator for 5 minutes. The upperlayer was removed and made volume up to 5 ml. Further 1% H2SO4 (25 ml) was added and extracted using 20 ml of CHCl3. The aqueous phase was separated, andammonium hydroxide was added to alkaline it and then extracted with 10 ml portions of CHCl3 successively. Then chloroform layers were washed with water followed by reducing volume by distillation. The absolute alcohol was added to the residues and evaporated at 100° C to dryness and solid residue obtained were crude alkaloids. The percentage yield of alkaloids was determined.

% yield of alkaloids= Weight of alkaloids obtained × 100/ Total weight of Sample

The plant material was placed in soxhlet extractor with solvent (200 ml) i-e ether for six hours at temperature 60°C to be defatted. The residue was kept in open air for overnight to evaporate the solvent. This plant material was then placed in the soxhlet extractor again along with methanol solvent (200 ml) till the colour less extraction. The solvent was evaporated and calculated percent yield of saponins.

% yield of saponins = Weight of saponins obtained × 100 / Total weight of Sample.

The grounded plant material was boiled with ethyl alcohol and filtered. The filtrate was mixed with lead sub acetate solution (30 ml) to remove chlorophyll and other pigments. The filtrate was treated with distilled water (45 ml saturated with H2S) to remove lead sub acetate. The pure filtrate was then dried on an electric water bath and the percentage (%) yield of crude glycosides was calculated. % Yield of Glycosides = Weight of glycosides obtained × 100 / Total weight of Sample

Methanolic extract of *M. alba* leaves showed brown precipitates when Mayers & Wagner's reagents were added. These results showed alkaloids were present in this plant. Quantitatively alkaloids isolated from *M. alba* leaves were 40%. Methanolic extract of *M. alba* showed light brown precipitates with Fehling solution as well as with Benedict solution .These observations suggested the presence of glycosides in *M. alba* leaves. Quantitatively glycosides isolated from *M. alba* leaves were 20.05%. When small quantity of ground plant materiel of *M. alba* was shaken with distill water, considerable froth was produced which lasts for several hours. Saponins are used for hypercholesterolemia, antioxidant, anti inflammatory, anti-cancer & gentle blood cleanser .Quantitatively 11.5% saponins were present in *M. alba* leaves.

The use of antibiotics in excess is harmful for human body and also resistance occurred against harmful pathogens. So the demand of exploring natural compounds having activity against harmful pathogens is increasing day by day. Antimicrobial activity of the aqueous and organic extracts of the plant samples were evaluated. For determination of antibacterial activity, bacterial cultures were adjusted to 0.5 McFarland turbidity standards and inoculated onto 15 cm diameter nutrient agar (petri-) plates. For the determination of antimycotic activity, all the fungal isolates were first adjusted to the concentration of 10 cfu/ml. All the 6 cultures were inoculated onto Potato Dextrose Agar plates. Sterile filter paper discs diameter(6 mm) impregnated with 100 µl of reconstituted extract (10 mg/ml) to give final concentration of 1mg/disc were placed on the culture plates previously seeded with the 0.5 McFarland and

10 cfu/ml cultures of bacteria and 6 fungi respectively. Bacterial cultures were then incubated at 37°C for 24 h, while the fungal cultures were incubated at room temperature (30-35°C) for 48 h. Paper discs impregnated with 20 µl of a solution of 10 mg/ml of bacitracin (for bacteria) and streptomycin (for fungi) as standard antibiotics were used for comparison. Antimicrobial activity was determined by measurement of inhibition zone around each paper disc. For each extract three replicate trials were conducted against each organism. evaluated the effect of compounds isolated from *M. alba* leaves against oral pathogens, commonly Streptococcus mutan. Compounds were purified by using silica gel chromatography and analyzed with different analytical techniques and by micro dilution method MICs were evaluated. The purified *M. alba* compound (1- deoxynojirimycin) showed 8-fold reduction of MIC against biofilm development of S. mutans than crude extract and it was revealed that 1-deoxynojirimycin inhibits the proliferation and formation of biofilm by S. mutans and can be used as therapeutic agent.

RESULTS AND DISCUSSION:

Due to the global trend towards improved quality of life there is significant demand for medicinal plant-based supplements from natural sources that have no contamination from synthetic fertilizers or chemicals and have lesser side effects. *M. alba* now a days has been investigated in various scientific instigations in order to explore its active constituents, which may have medicinal worth. It is a rich source of flavonoids and other compounds which showed antimicrobial potential and free radical scavenging activity . The phytochemical analysis of the two variants of *M. alba* showed the presence of saponin having high percentage in both the two variants of *M. alba* screened. These phytochemicals exhibit diverse pharmacological and biochemical actions when ingested by animals Saponin reduces the uptake of certain nutrients including glucose and cholesterol at the gut through intra luminal physicochemical interactions. Alkaloids are often toxic to man and may have dramatic physiological activities hence their wide use in medicine. The antimicrobial activity of the ethanolic extracts of the *M. alba* variants showed some inhibitory power against the microbes used for this research work with S14 extract showing the highest activity (diameter of inhibition zone 11 mm) against *P. aureginosa* and S34 (diameter of inhibition zone 11 mm). The lowest activity was shown by S34 (diameter of inhibition zone 03 mm) against P. oxalicum. Leaves of *M. alba* are rich in protein and widely used in food formulations.

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