

Synthetic berberine derivatives as potential new drugs

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The modern process of new drug discovery and development is an exciting, yet a challenging, endeavor. Although it can result in significant financial income and meet the medical needs of patients, it ultimately may result in failure. To achieve a fast and successful new product discovery and development process, natural products which are evolutionarily optimized as drug-like molecules have gained great attention as better potential sources of new chemical entities. Historically, plant species containing berberine are used in various traditional phytotherapy. However, despite the various therapeutic effects it exerts, berberine is not yet developed into a drug product. Addressing the barriers that hinder its successful development and the efforts made to overcome them is thus crucial. The toxicological and pharmacokinetic properties of berberine are the main barriers towards its development into a marketed drug product. It has low aqueous solubility, poor absorption, fast metabolism, and wide tissue distribution which lead to low bioavailability limiting its clinical application. Synthetic berberine derivatives with improved properties are suggested as better alternatives for further development and future therapeutic application. Hence, this paper summarizes the preclinical research studies conducted in the last decade to reveal the therapeutic potential of synthetic berberine derivatives for the treatment of various diseases and hence achieve successful berberine-based drug development in the future. To exploit the value of natural products as a source of leads for the development of effective drugs, collaboration among the different discovery and development scientists is essential.

Keywords: Berberine. Natural product. Synthetic derivatives. Drug discovery. Product development. Collaboration.

INTRODUCTION

Drugs play an important role in maintaining and/or restoring health and hence promote a good quality of life in humans. Although many drugs that treat a wide array of diseases have reached the market, there are still unmet medical needs for which new drugs need to be discovered and developed. Some of the various bases for

a drug discovery process are the isolation of the active constituent in traditional medicine formulations, natural products, *de novo* synthesis, biotechnology, isosteric replacement, and serendipity (Siddiqui AA, *et al.*, 2014). Natural products are chemical compounds produced by living beings (microorganisms, marine organisms, plants, and animals) that usually have therapeutic and/or toxic activity (Siddiqui AA, *et al.*, 2014). Over the years natural products have been utilized as a source of new chemical entities (NCEs) owing to their unique and great chemical diversity, advantageous pharmaceutical properties and potential for synergistic effects in

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traditional formulations containing multiple constituents (Pandey P, Doerksen RJ, 2016). Analysis of the sources of newly approved drugs for the treatment of diseases in human revealed that, of the 1211 small-molecule new chemical entities approved from the year 1981 to 2014, 33% were natural product based (Newman DJ, Cragg GM, 2016).

Medicinal plants which are used to counteract diseases have been playing an important role in the primary health care of humans. They are a potential source of new bio-active substances of wide structural diversity, which can be utilized directly as pharmacologically active compounds or as pro-drugs (Ahmed WJ *et al.*, 2015). In the early 1900's before the start of lead/drug discovery through synthesis 80% of all active pharmaceutical ingredients were obtained from plant sources (Siddiqui AA *et al.*, 2014). However, due to the challenging and laborious nature of natural product-based drug discovery and development process, pharmaceutical companies deemphasized natural product drug discovery since the late 20th century, and advanced technologies namely high

throughput screening and combinatorial chemistry have been introduced to facilitate the discovery of drug leads (Shen B, 2015). However, despite a huge financial incentive that can be obtained from successfully developed and marketed drug products, modern drug discovery and development is also a long and expensive process characterized by low productivity. The high risk of failure in drug discovery and development throughout the pharmaceutical industry statistically shows that, on average, only 1 in 5000 compounds screened in research will successfully reach the market for clinical application (Gibson M, 2009). Furthermore, the emergence of drug-resistance and cost-ineffectiveness of synthetic drugs are increasing the interest in medicinal natural products-based drug discovery (Amritpal S *et al.*, 2010). The utilization of natural products as a safer, better efficacy, and cost-effective therapeutic alternative is thus of paramount importance. Hence it can be concluded that natural products which are evolutionarily optimized as drug-like molecules remain one of the best sources of drugs and drug leads (Cragg GM, Newman DJ, 2013).

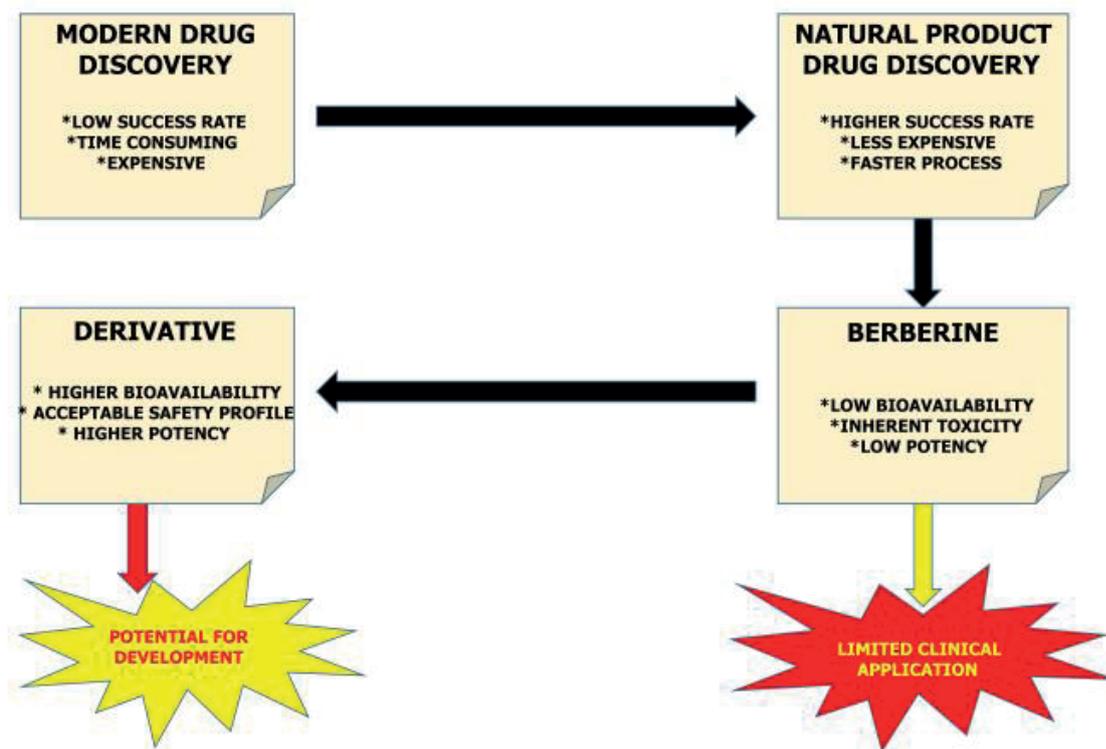


FIGURE 1 - Graphical abstract.

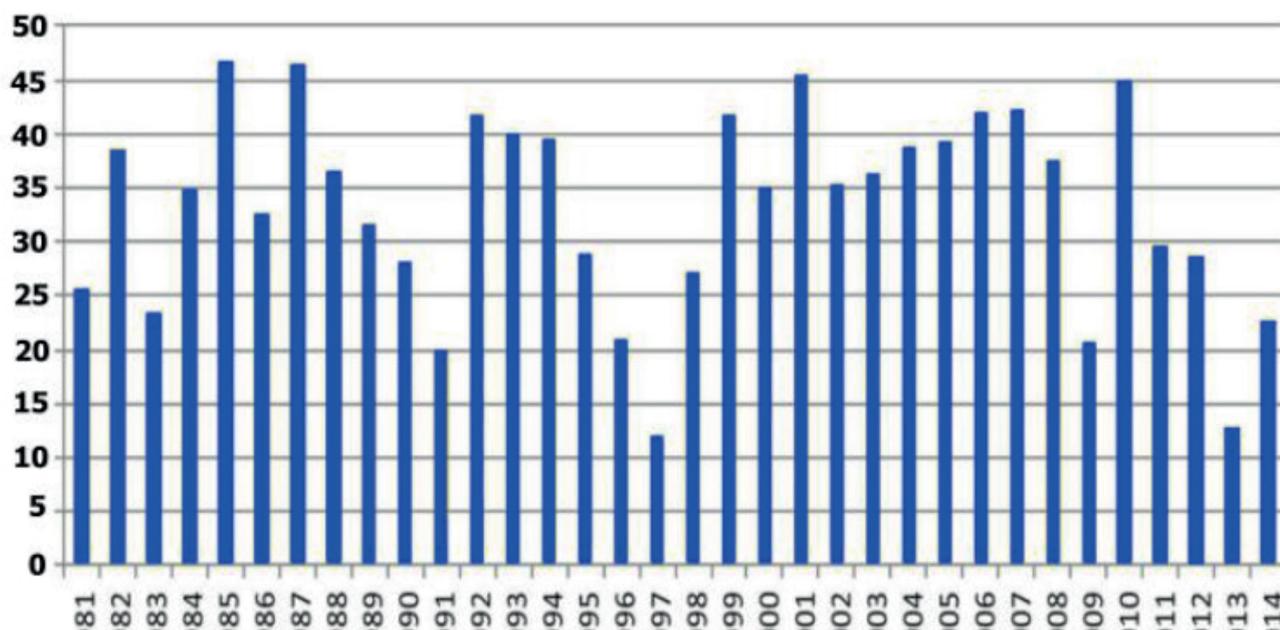


FIGURE 2 - Percentage of natural products (semi- or totally synthetic) approved over the years 1981-2014 [(Newman DJ, Cragg GM. *J. Nat. Prod.* 2016; 79(3)].

Berberine (BBR), an isoquinoline alkaloid belonging to the class of protoberberine alkaloids, is biosynthesized in plants of the Berberidaceae, Papaveraceae and Ranunculaceae families (Rad SZK, Rameshrad M, Hosseinzadeh H, 2017). It is a constituent of the roots, rhizomes, and stem bark of various plant species including the barberry (*Berberis vulgaris*), the meadow rue (*Thalictrum*), the celandine (*Chelidonium*), the goldenseal (*Hydrastis canadensis* L.), *Phellodendron amurense*, etc. (Nechepurenko I, Salakhutdinov N, Tolstikov G, 2010). It occurs as an intense yellow solid that is odorless with a characteristic bitter taste. Berberine is slightly soluble in ethanol, very slightly soluble in water, and sparingly soluble in methanol (Battu SK *et al.*, 2010). It has a molecular formula of $C_{20}H_{18}NO_4^+$ with the IUPAC name of 9,10-Dimethoxy-5,6-dihydro[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium, and a molecular weight of 336.367g/mole. Typically, berberine is isolated using a method based on extraction by alcohol in neutral or acidic medium followed by removal of side substances and finally its precipitation in the salt form (Nechepurenko IV, Salakhutdinov NF, Tolstikov GA, 2010).

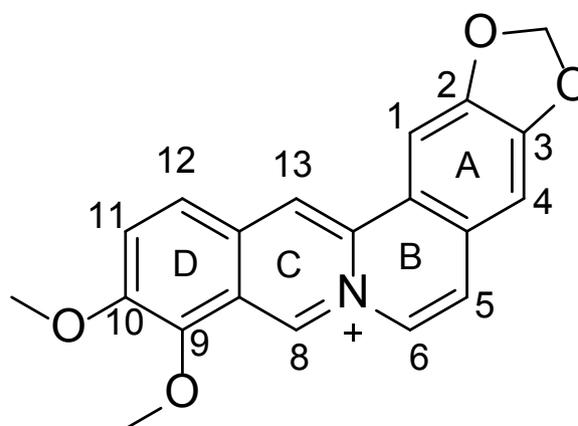


FIGURE 3 - The chemical structure of berberine.

Berberine has been used in traditional and supplementary medicine only in the form of polyherbal formulations and not as a single entity (Singh IP, Mahajan S, 2013). It exerts low therapeutic effects owing to its low permeability and bioavailability (Xiao D *et al.*, 2018). So far, there is no a marketed berberine-based drug product. Therefore, its important to address the barriers toward its development and summarize the efforts made

to facilitate its clinical application. Attempts have been made to improve its intestinal solubility and absorption and hence increase its bioavailability by using novel drug delivery systems such as liposomes and cyclodextrin inclusion complex (Battu SK *et al.*, 2010, Singh IP, Mahajan S, 2013). Interestingly, the chemical structure (isoquinoline skeleton) of berberine offers a scaffold that facilitates the generation of numerous derivatives through rational design and functionalization. Various chemical transformation methods of the berberine molecule have allowed researchers to obtain its derivatives which exhibit valuable pharmacological properties. To facilitate the successful development of berberine-based drug product, summarizing the results of relevant research studies on synthetic berberine derivatives is necessary. This review discusses the research studies conducted on synthetic berberine derivatives and which have proved their potential to be developed as clinically applicable drugs. Besides, the therapeutic applications of berberine in various traditional medicine systems, the scientific researches which have confirmed its pharmacological activities and the properties (pharmacokinetic and toxic) that limit its successful development are summarized.

BERBERINE IN TRADITIONAL MEDICINE SYSTEMS

Berberine-containing plants are widely used in various traditional medicine systems such as Ayurveda (India), Traditional Chinese Medicine (China), Tibet and Japan (Nechepurenko I, Salakhutdinov N, Tolstikov G, 2010). In Traditional Chinese Medicine (TCM) berberine has been used for thousands of years to treat parasitic intestinal infection and bacterial diarrhea (Li D *et al.*, 2017). Moreover, some berberine containing medicinal plants were recorded in the Pharmacopoeia of China (2015) for their therapeutic effects including lowering body temperature, resolving dampness, purging fire and detoxification (Wang K *et al.*, 2017). The use of plants belonging to the genus *Berberis* was reported in the Indian folk medicine for the treatment of leishmaniasis and malaria and was used in the Japanese folk medicine against cholera and bacterial diarrhea (Nechepurenko I, Salakhutdinov N, Tolstikov G, 2010). It is also reported that the plant species *Phellodendron amurense* was used in Japan against dysentery, and *Corydalis* was used as an analgesic and spasmolytic agent against a toothache. Besides, the goldenseal (*Hydrastis Canadensis*) was used against inflammations of gastrointestinal tract and of

upper air passages, as antihypertensive and anticatarral remedy (Nechepurenko IV, Salakhutdinov NF, Tolstikov GA, 2010). Therefore, it can be concluded that berberine represents a valuable lead molecule for obtaining new pharmacologically active drug substances.

EFFORTS TOWARDS CLINICAL APPLICATION OF BERBERINE

Besides to its application in traditional medicine systems a growing number of research studies focused on pharmacological activities of berberine against a wide range of diseases, which have attracted the attention of researchers, clinicians and the public, have been conducted to facilitate its future development and clinical application (Habtemariam S, 2016). The various pharmacological activities of berberine result from its direct interaction with nucleic acids and with several proteins (Tillhon M *et al.*, 2012). A list of therapeutic effects of berberine is given in table I. In this review the preliminary research studies which have confirmed the therapeutic effect of berberine in the treatment of diseases (diarrhea and inflammation) against which berberine is mainly used in traditional medicine systems including the mechanisms of its action are discussed. Moreover, the pharmacological activity of berberine for the treatment of diseases which are the global leading causes of death such as cardiovascular diseases and cancer is included.

TABLE I - Preliminary studies on various therapeutic effects of berberine

Therapeutic effect	References
Anticancer	
DNA damage	Jagetia GC, 2015
Cervical cancer	Lin JP <i>et al.</i> , 2007
Tongue cancer	Ho YT <i>et al.</i> , 2009
Colorectal cancer	Li D <i>et al.</i> , 2017
Breast cancer	Raza A, 2015
Polycystic ovary syndrome (PCOS)	Wang Y <i>et al.</i> , 2016

(continuing)

TABLE I - Preliminary studies on various therapeutic effects of berberine

Therapeutic effect	References
Cardiovascular diseases	
Vasoprotective	Bagade A, Tumbigeremutt V, Pallavi G, 2017
Hyperlipidemia	Li XY <i>et al.</i> , 2015
Diabetes	Li Zheng <i>et al.</i> , 2014, Yin J <i>et al.</i> , 2008, Zhang H <i>et al.</i> , 2010
Heart failure	Marin-Neto J <i>et al.</i> , 1988
antidiarrheal	Habtemariam S, 2016, Birdsall TC, 1997, Marin-Neto J <i>et al.</i> , 1988
Anti-inflammatory	
Suppression of pro-inflammatory cytokine gene expression	Haj-allahyari S, Maghsoudi H, 2018
NF-kB expression	Raza A, 2015
Inhibition of DNA synthesis	Ckless K <i>et al.</i> , 1995
Anti-inflammatory activity against diabetes mellitus	Li Z <i>et al.</i> , 2014
Inflammatory bowel disease (IBD)	Habtemariam S, 2016
Neuroprotective effect	Ahmed T <i>et al.</i> , 2015
Anti-fungal	Freile M <i>et al.</i> , 2006
Anti-parasitic	Birdsall TC, 1997

Anticancer effect

As anticancer drugs are characterized by severe side effects development of drug substances from medicinal plants can be considered as a better treatment option. Researches have demonstrated that berberine and/or its derivatives could be promising anticancer agents for clinical development due to the *in vitro* anticancer effects exerted by inhibiting tumour progression and metastasis

and by inducing apoptosis (Tillhon M *et al.*, 2012). Determination of the extent of DNA damage, by alkaline comet assay, in HeLa cells using various concentrations of berberine chloride (BCL) revealed the anti-neoplastic effect of BCL which caused a concentration-dependent rise in the DNA damage (Jagetia GC, 2015). The proapoptotic action of berberine was also confirmed on human cervical cancer cells where *in vitro* treatment with berberine resulted in a decreased percentage of viable cells (Lin JP *et al.*, 2007). Similar results were observed on human tongue cancer cells where it was found to induce apoptosis (Ho YT *et al.*, 2009). *In vivo* study on mice found that berberine effectively inhibited colorectal cancer through its anti-inflammatory effect (Li D *et al.*, 2017). Furthermore, Berberine was found to show a synergistic effect in combined chemotherapy and decreased multidrug resistance in breast cancer therapy (Raza A, 2015).

Cardiovascular disorders

In the recent past, studies have highlighted the therapeutic potential of BBR in metabolic and cardiovascular diseases. It was reported that BBR exerts antiproliferative and vasoprotective actions through the inhibition of intracellular accumulation of reactive oxygen species, cellular apoptosis, and inflammation (Bagade A, Tumbigeremutt V, Pallavi G, 2017). An examination of the effect of BBR on the metabolic pathway of cholesterol showed that BBR lowers blood lipid levels in high-fat-diet-induced hyperlipidemic golden hamsters by promoting the excretion of cholesterol from the liver to the bile (Li XY *et al.*, 2015). It was also demonstrated, in *in vitro* studies, that BBR induces vasodilation by acting on both endothelial and underlying vascular smooth muscle cells (Bagade A, Tumbigeremutt V, Pallavi G, 2017). In a clinical study, BBR lowered fasting blood glucose, hemoglobin A1c, and insulin levels in type 2 diabetic patients at levels similar to metformin and rosiglitazone, indicating its ability to upregulate insulin receptor (InsR). Besides, analysis of the effect of BBR in various human cell lines including CEM, HCT-116, SW1990, HT1080, 293T, and hepatitis B virus-transfected human liver cells, was carried out where it was found that it increased InsR messenger RNA and protein expression (Zhang H *et al.*, 2010). Furthermore, in heart failure patients unresponsive to conventional treatment using digitalis and diuretics, berberine infusion administered at a

known rate of 0.2 mg/kg/min decreased systemic and pulmonary resistance and improved hemodynamics (Marin-Neto J *et al.*,1988).

Diarrhea

Among the reported numerous BBR's therapeutic effects several studies have evaluated the potential application of berberine in the treatment of diarrhea. In a clinical investigation, human patients receiving berberine hydrochloride appeared to benefit from the therapy along with better IBS (irritable bowel syndrome) symptoms and improved quality of life (Marin-Neto J *et al.*,1988). In animals treated with berberine, it was found that the intestinal secretion of water and electrolytes induced by cholera toxin was reduced (Birdsall TC, 1997). Moreover, in mice with established *Clostridium difficile*-induced intestinal injury and colitis, administration of berberine restored the intestinal microbiota (Habtemariam S, 2016).

Inflammation

An inflammatory response which can be induced by allergens, bacterial and viral infections and autoimmune diseases is thought to be threatening especially if the inflammation exists chronically (Zou K *et al.*, 2017). A recent study suggested that an aqueous extract of BBR exerted anti-inflammatory effect probably due to the suppression of pro-inflammatory cytokine gene expression (Haj-allahyari S, Maghsoudi H, 2018). Additionally, BBR suppressed inflammatory agents induced NF-kB expression which leads to down-regulation of genes involved in anti-apoptosis, invasion, and proliferation (Raza A, 2015). Results of an *in vitro* study using stimulated human peripheral lymphocytes proposed that berberine shows an anti-inflammatory effect in part due to inhibition of DNA synthesis (Ckless K *et al.*, 1995). Moreover, in an animal study, it was shown that BBR had antioxidant and anti-inflammatory activities which contribute in part to its efficacy against diabetes mellitus (Li Z *et al.*, 2014). The efficacy of BBR for inflammatory bowel disease (IBD) treatment was also examined. Besides its general anti-inflammatory and antioxidant effects that possibly contribute to its IBD efficacy, IBD-specific mechanisms employing *in vivo* and *in vitro* experimental colitis have been reported. A number of studies suggested that the efficacy of berberine is in part mediated by

reversing the common IBD associated gut epithelial barrier dysfunction (Habtemariam S, 2016). Moreover, the action of berberine to target T-helper cells and/or their products as a potential IBD therapeutic approach has been documented (Habtemariam S, 2016). The plant constituent berberine displays a wide array of pharmacological activities. Other reported therapeutic effects of berberine include: anti-fungal activity (Freile M *et al.*, 2006), growth inhibitory activity against intestinal parasites (Birdsall TC, 1997), neuroprotective effect (Ahmed T *et al.*, 2015) and others.

PROPERTIES THAT LIMIT THE DEVELOPMENT OF BERBERINE

Berberine is a widely used promising biologically active natural product. A review of patents, regarding the therapeutic potential of pure berberine, derivatives of berberine and herbal preparations containing berberine, in the years 2009 to 2012 revealed that berberine or its derivatives possess great potential to be developed as new drug substances for the treatment of cancer, inflammation, metabolic disorders, infectious diseases and others (Singh IP, Mahajan S, 2013). Meanwhile some properties of berberine pose barriers towards its development into a marketed drug product. Understanding these barriers helps in designing a strategy to overcome them and hence achieve a fast and successful development. The toxicological and pharmacokinetic properties are discussed in this review.

Toxicological effects of berberine

Toxicity of candidate compounds is one of the main properties that should be assessed during development. At the doses used in clinical situations (200 mg orally two to four times daily), berberine is not considered to be toxic (Imanshahidi M, Hosseinzadeh H, 2008). Side effects, including dyspnea, lowered blood pressure, and flu-like symptoms can arise from higher dosages. Administration of very high doses for more than 4-6 weeks may cause hepatic side-effects (Singh IP, Mahajan S, 2013). While increasing the oral dose of BBR to obtain optimum efficacy often exerts gastrointestinal side effects, results from animal and *in vitro* studies have indicated that berberine induces immunotoxicity, neurotoxicity, and cardiotoxicity which depends on the route and duration of administration (Yin J, Xing H, Ye J, 2008). In one study, it was reported that UVA

irradiation of HaCaT keratinocytes in the presence of 50 μ M berberine resulted in an 80% decrease in cell viability and in a threefold increase in DNA damage. Hence, it was suggested that exposure to sunlight or artificial light sources emitting UVA should be avoided when using topical preparations containing berberine (Inbaraj JJ *et al.*, 2001). Berberine exerts a toxic effect on both normal and cancer cells in a time and concentration-dependent manner. Besides, as it exhibits uterine stimulatory action, BBR and its derivatives should be used with caution during pregnancy (Kumar A *et al.*, 2015). It also leads to an increase in the free bilirubin concentration, by displacing bilirubin from its serum binding proteins, causing jaundice, kernicterus, and brain damage in infants (Bateman J, Chapman RD, Simpson D, 1998). Along these lines, an assessment of developmental toxicity of berberine in laboratory animals (rats and mice) during gestation found that BBR caused maternal but not fetal adverse effects, indicated by an increase in the lowest-observed-adverse-effect level (LOAEL), in rats. In addition to a significant number of deaths (33%) in the treated mouse, it caused decreased fetal body weight (Jahnke GD *et al.*, 2006). A pilot clinical study performed on herbal medicines which contain berberine from the plants *Rhizoma coptidis* (RC) and *Cortex phellodendri* (CP), for patients with chronic cytopenic haematological conditions reported that appropriate use of BBR did not cause major safety concern although it was suggested that BBR should still be not used in infants and G6PD (glucose-6-phosphate dehydrogenase) deficient individuals of all ages as well as lactating and pregnant women (Linn YC *et al.*, 2012).

Since berberine is a high-affinity substrate for P-glycoprotein (P-gp), organic cation transporters and some cytochrome P450 isoenzymes, which all have important roles in the transport and metabolism of drugs, co-administration of berberine was reported to increase the blood concentration of other drugs transferred by these pathways enhancing their therapeutic/toxic effect (Kwon M *et al.*, 2015). Furthermore, the inhibitory effect of berberine on cytochrome-P (CYP) enzymes could result in toxicity when co-administered with drugs that are mainly metabolized by these enzymes (Rad SZK, Rameshrad M, Hosseinzadeh H, 2017). It was reported that CYP2D6 and other CYPs (such as CYP1A and CYP3A) are involved in berberine metabolism, and thus when berberine or berberine-containing products are used, potential drug interactions and genetic polymorphisms of CYP2D6 should be considered

(Guo Y *et al.*, 2011). Therefore, for a rational use of berberine, continuous examination of toxicity against its therapeutic potential based on research evidences is important (Fung FY, Linn YC, 2015).

Bioavailability and pharmacokinetic properties of berberine

Pharmacokinetic data is necessary to design a rational dosage regimen. Berberine's low aqueous solubility, poor absorption together with extensive metabolism and wide tissue distribution results in low plasma concentration hindering its oral administration (Wang K *et al.*, 2017). Gastrointestinal absorption of berberine has been demonstrated in rabbits, rats and humans (Wang K *et al.*, 2017, Jahnke GD *et al.*, 2006). However, its absolute bioavailability is very low (Cheng M *et al.*, 2016). In a study designed to assess the role of small intestine and liver in the extensive first-pass elimination of berberine in the body, it was observed that, approximately half of the drug was absorbed by the gastrointestinal tract and another half was distributed to the liver after intragastric administration to rats, leading to very low plasma concentration (Liu Y *et al.*, 2010). Similarly, in a pilot pharmacokinetic study which involved twenty healthy male volunteers, the maximum mean plasma concentration was found to be only 0.4 ng/mL, $t_{1/2}$ was 28 h and t_{max} was 9.8 h after a single oral dose of 400 mg (Hua *et al.*, 2007), suggesting that berberine has a long half-life and low systemic bioavailability in humans. Investigation of tissue distribution in rats found that, four hours after oral administration the concentrations of berberine and its active metabolites were higher in most of the studied tissues (liver, kidneys, muscle, lungs, brain, heart, pancreas and fat) than in blood (Tan XS *et al.*, 2013).

The fast elimination rate of berberine is also one of the factors that leads to its low blood concentration. Results of excretion study of berberine in rats revealed that most of BBR and its metabolites were found in feces after oral administration (Ma JY *et al.*, 2013). In an *in vivo* study, sixteen phase I and phase II metabolites of berberine were detected in the urine, feces and bile of rats after oral administration (Ma JY *et al.*, 2013). Thalifendine was the metabolite with the highest amount excreted in all the samples including bile, urine, and feces. Moreover, *in vitro* incubation of berberine with 14 intestinal bacterial strains showed that berberine is converted to dihydroberberine by nitroreductases of gut microbiota

(Feng R *et al.*, 2015). The major circulating metabolite of BBR in rats, after intravenous administration, were demethyleneberberine, berberrubine and their corresponding glucuronide conjugates (Liu Y *et al.*, 2009). Moreover, the major metabolites after intragastric dosing in rats were found to be glucuronides of phase I metabolites and the unconjugated metabolites (berberrubine, demethyleneberberine and jatrorrhizine) were the major forms present in tissues, including liver,

heart, and kidney (Liu Y *et al.*, 2010, Tan XS *et al.*, 2013). Although BBR is characterized by very low absolute oral bioavailability, it exerts marked therapeutic activities *in vivo*, and the organ and blood concentrations of its major metabolites is relatively high (Wang K *et al.*, 2017), and hence it was proposed that its metabolites also contribute to the various pharmacological effects (Wang K *et al.*, 2017; Zuo F *et al.*, 2006).

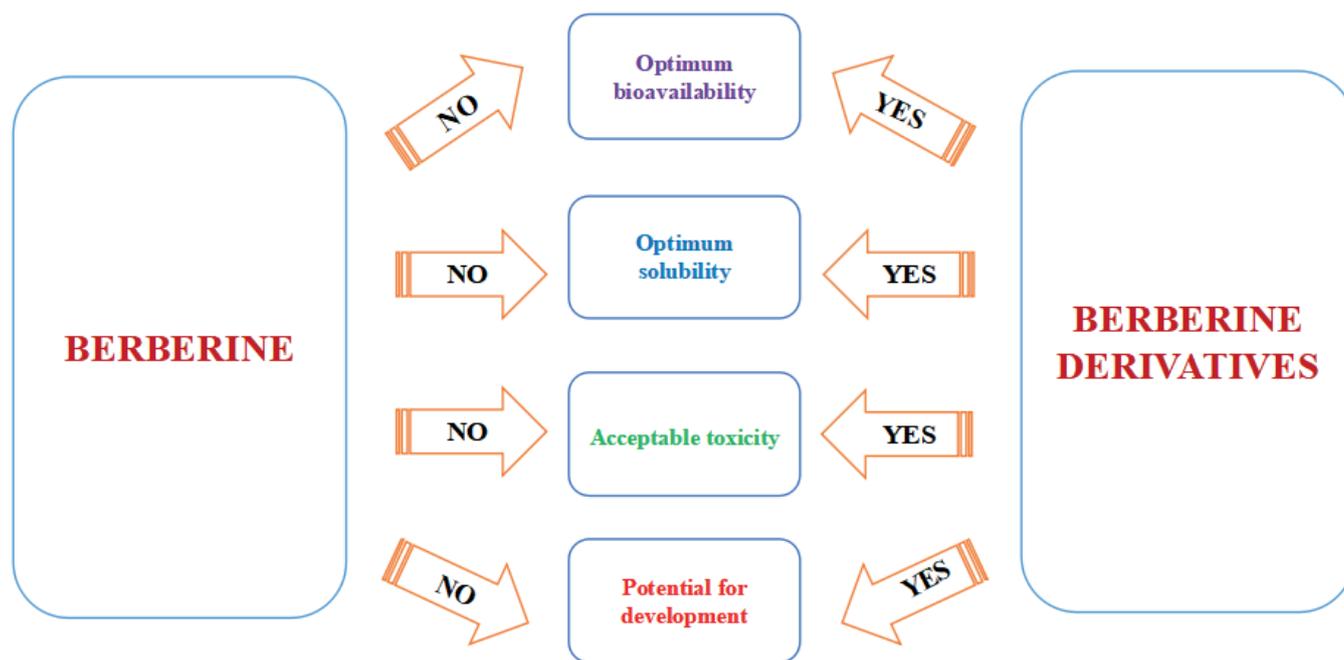


FIGURE 4 - A schematic presentation of BBR and its synthetic derivatives' potential for development.

ENHANCING THE THERAPEUTIC EFFECTS OF BERBERINE VIA STRUCTURAL MODIFICATION

For successful development of berberine-based drug product, enhancing the bioavailability of BBR and decreasing its toxicity are thus essential. Structural modification, which has been the focus of several recent studies, as a strategy to overcome the barriers toward a successful development of berberine is discussed in this review. Development of berberine analogues or derivatives is expected to increase its bioavailability and hence its therapeutic effect (Wang Y *et al.*, 2016, Han Y, Kim MJ, Lee KW, 2018). Hence, to improve the pharmacokinetic properties and obtain better

pharmacological effects of berberine for treating systemic disorders researchers have been designing and synthesizing its derivatives, by modifying its structure, in the recent past (Yang Z *et al.*, 2018, Mistry B *et al.*, 2017, Chen J *et al.*, 2014, Wang YX *et al.*, 2017, Sun N *et al.*, 2014).

Systematic structural modification of berberine, mainly on its C-8, C-9, C-13 positions as shown in figure 5, has been proven to be an effective method to change its biological activities and improve its efficacy for the treatment of different diseases (Xiao D *et al.*, 2018). Berberine can be readily converted to berberrubine, by pyrolysis, to generate a series of 9-O-substituted derivatives with specific pharmacological properties.

The polar C=N⁺ bond in berberine nucleus is sensitive to nucleophilic attack leading to the formation of derivatives with substituent at C-8. Modification of the berberine skeleton by functionalization at C-13 through enamine-7,8-acetyl intermediate is another method employed to obtain a series of derivatives (Tillhon M *et al.*, 2012, Singh IP, Mahajan S, 2013). Preliminary pharmacological studies of these derivatives are demonstrating their promising therapeutic effects for further development and clinical application in the future.

Preliminary pharmacological studies on synthetic berberine derivatives

Several studies have revealed that derivatives of berberine can exert an optimum therapeutic effect for the treatment of various diseases. This review summarizes the research studies conducted in the last decade to explore the applicability of synthetic berberine derivatives as potential new active pharmaceutical ingredients for the treatment of a variety of diseases including their pharmacological profile and the underlying mechanisms of action. The various pharmacological effects of synthetic berberine derivatives are given in table II.

TABLE II - Preliminary studies on pharmacological activity of synthetic berberine derivatives

Therapeutic effect	References
Antimicrobial	
Anti-mycobacterial	Wang Y <i>et al.</i> , 2012, Mahapatra A <i>et al.</i> , 2014
Antibacterial	Gao WW <i>et al.</i> , 2018, Sun N <i>et al.</i> , 2014, Jeyakkumar P <i>et al.</i> , 2016
Antifungal	Ling Y <i>et al.</i> , 2018
Antiprotozoal	Bahar M <i>et al.</i> , 2011
Antioxidant	
	Mistry B <i>et al.</i> , 2017, Pongkittiphon V, Chavasiri W, Supabphol R, 2015, Roselli M <i>et al.</i> , 2016

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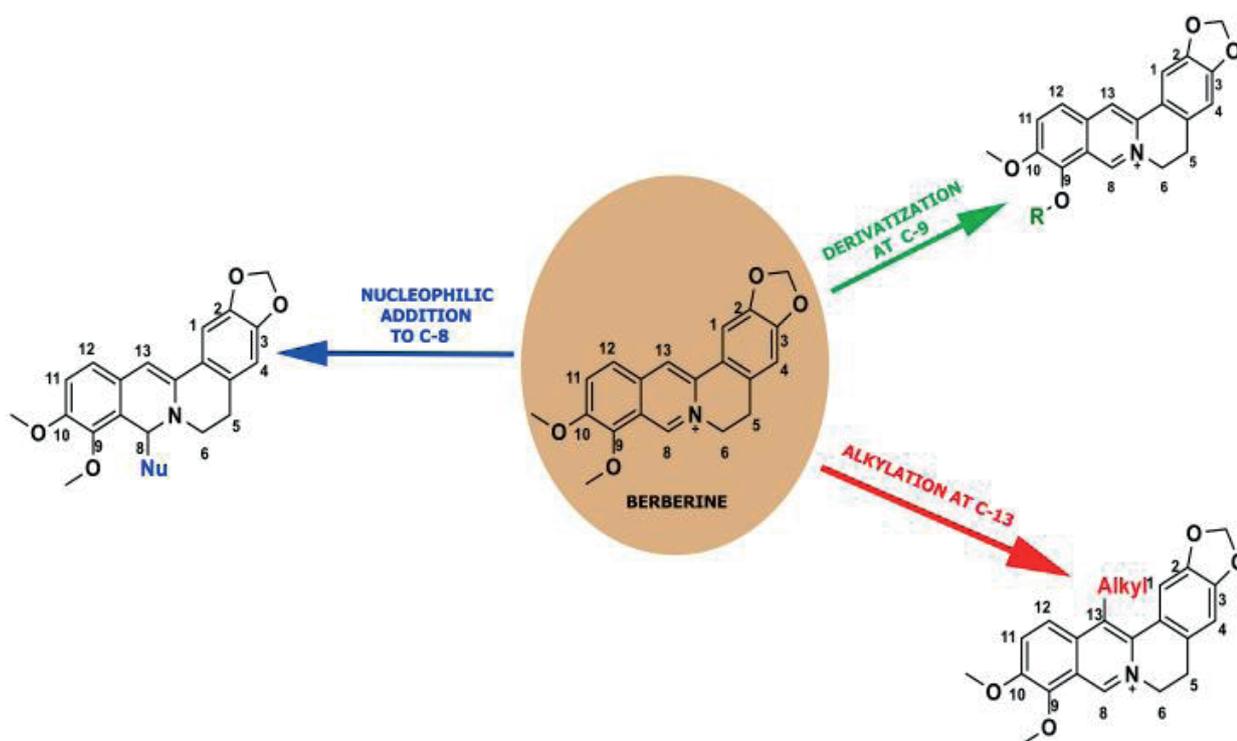


FIGURE 5 - Common methods for functionalization the chemical structure of berberine.

TABLE II - Preliminary studies on pharmacological activity of synthetic berberine derivatives

Therapeutic effect	References
Lipid-lowering effect/ atherosclerosis	Chen J <i>et al.</i> , 2014
Anticancer	
Liver, breast, pancreatic cancer	Jin X <i>et al.</i> , 2014
Colon cancer	Albring KF <i>et al.</i> , 2013, Guaman Ortiz LM <i>et al.</i> , 2014, Guamán Ortiz LM <i>et al.</i> , 2015,
Histone acetyltransferase (HAT) dysfunction treatment	Yang Z <i>et al.</i> , 2018
G-quadruplex telomeric DNA stabilization	Ma Y <i>et al.</i> , 2009
Breast cancer	Pierpaoli E <i>et al.</i> , 2013
inhibitor of MAPK (mitogen- activated protein kinase)	Kim N <i>et al.</i> , 2014
Skin cancer	(Kaboli PJ, Ismail P, Ling K-H, 2018)
Cancer immunotherapy	Wang YX <i>et al.</i> , 2018
Antidiabetic	Ren G <i>et al.</i> , 2017, Turner N <i>et al.</i> , 2008
Anti-inflammatory	Wang YX <i>et al.</i> , 2017, Huang MY <i>et al.</i> , 2016, Chen YY <i>et al.</i> , 2017, Deng X <i>et al.</i> , 2012
Hepatic effects	Wang Y <i>et al.</i> , 2016, Zhang P <i>et al.</i> , 2015, Qiang X <i>et al.</i> , 2015, Al LX, 2016,
Neurodegenerative diseases	Huang L <i>et al.</i> , 2010, Ribaud G <i>et al.</i> , 2018, Huang L <i>et al.</i> , 2010
Osteoporosis	Han Y, Kim MJ, Lee KY, 2018

Antimicrobial action

Natural products and/or their novel structures have been utilized to discover drugs/leads against microbial infections. Many scientific researches have investigated the antimicrobial activity of synthetic BBR derivatives. To overcome the problem of, multidrug resistance which hinders the effective treatment of tuberculosis, the anti-mycobacterial activities of eighteen newly synthesized 8-substituted berberine derivatives were investigated against *Mycobacterium tuberculosis* (*M. tuberculosis*) strain H₃₇Rv (Wang Y *et al.*, 2012). Results of SAR analysis indicated that for anti-mycobacterial activity a large group at the 8-position might be required. Interestingly, one of the tested compounds (Figure 6, A) which exerted a potent effect against clinically isolated rifampicin- and isoniazid-resistant *M. tuberculosis* strains showed its potential for further anti-TB drug development. Moreover, the anti-tubercular of 13-benzyl, 13-allyl, 8-(2-oxopropyl) and 9-hydroxy BBR derivatives for tuberculosis was studied against *Mycobacterium tuberculosis* strain H₃₇Rv using microdilution assay and compared with rifampicin as a standard drug (Mahapatra A *et al.*, 2014). Some of the tested compounds with 13-benzyl and allyl substitution displayed 2–4 fold higher activity than berberine demonstrating their therapeutic potential.

In an attempt to develop a new class of antibacterial agents against antibiotic-resistant bacteria the inhibitory effect of 9-phenoxyalkyl berberine derivatives on the bacterial cytokinesis protein (FtsZ) was tested (Sun N *et al.*, 2014). The compounds exhibited potent antimicrobial activity against Gram-positive bacterial strains such as *Staphylococcus aureus* and a broader spectrum of action than the parent compound and inhibited the FtsZ polymerization dose-dependently. Along these lines, to overcome the problem of resistance of pathogens against common drugs and develop effective small-molecule antimicrobial drugs, berberine was used as a lead compound to synthesize a series of derivatives by introducing substituted benzyl groups at its C-9 position. *In vitro* screening of the compounds against some pathogenic bacterial and fungal strains resulted in a good antimicrobial activity compared to reference drugs (ciprofloxacin and fluconazole). The interaction of the synthesized compounds with DNA gyrase (a target for effective treatment of microbial infection) was confirmed using docking studies. Based on SAR studies it was proposed that the substituents (such as

electron withdrawing and electron donating groups) in the benzyl moiety were essential for the antimicrobial activity (Ling Y *et al.*, 2018). Moreover, to exploit the antimicrobial effect of benzimidazole moiety in the development of novel antimicrobial drugs against drug-resistant microbial strains, a series of berberine-benzimidazole derivatives were synthesized. Most of the compounds were found to be effective against the tested bacterial strains (Jeyakkumar P *et al.*, 2016).

In an effort to find a better alternative antiprotozoal drug, the synthesis of 5,6-didehydro-8,8-diethyl-13-oxodihydro berberine chloride was carried out and its effect against protozoan parasites of leishmaniasis, African trypanosomiasis and malaria were evaluated using *in vitro* models. The compound was showed a highly potent effect against *in vitro* models of leishmaniasis (*Leishmania amazonensis*), malaria (*Plasmodium falciparum*) and trypanosomiasis (*Trypanosoma brucei brucei*) as well as activity in an *in vivo* visceral leishmaniasis model (mouse peritoneal macrophages infected with *L. amazonensis parasites*). Results of the comparative studies indicated that it had higher selectivity towards parasites compared to mammalian cells (green monkey epithelial kidney) and a multi-fold increase in inhibitory potency than berberine (Bahar M *et al.*, 2011). In another study, a series of aminothiazolyl berberine derivatives were designed as a new class of antimicrobial agents some of which displayed better antibacterial activity than natural berberine (Gao WW *et al.*, 2018). The biological activity determined based on *in vitro* evaluation of minimum inhibitory concentration for a panel of microbes indicated that some prepared compounds showed moderate to good antibacterial and antifungal activities including the clinically drug-resistant *acinetobacter baumannii*. The aminothiazole and Schiff base moieties were predicted to be beneficial for aqueous solubility and antibacterial activity.

Antioxidant effect

Oxidative stress in cells can damage cytoplasmic biomolecules causing several diseases including diabetes, Alzheimer's disease, rheumatoid arthritis, cancer, and inflammation, etc. (Pongkittiphon V, Chavasiri W, Supabphol W, 2015). Recently, direct and indirect antioxidant activities of berberine derivatives have been demonstrated in numerous researches. The *in vitro* biological activity of 9-demethylated (mannich base) synthetic berberine derivatives was tested on cervical cancer cell lines HeLa and CaSki and normal cell lines (Madin-

Darby canine kidney (MDCK) cell lines) to determine their anticancer and antioxidant effects (Mistry B *et al.*, 2016). All the tested compounds showed better antioxidant potential in DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS•+ (2,2-diphenyl-1-picrylhydrazyl) bioassays than parent berberine. While compounds bearing a furoyl piperazine substituent, a pyridyl piperazine ring system and a carbazole moiety displayed the highest anticancer activity against the CaSki cell line those with a pyridyl piperazine substituent, and having a carbazole substituent, were the most potent and active analogues against the HeLa cell and were almost twice as active as parent berberine.

In a study aimed to enhance the antioxidant effect of BBR, investigation of both direct and indirect antioxidant activities of phenolic derivatives was carried out (Pongkittiphon V, Chavasiri W, Supabphol R, 2015). DPPH assay results indicated that the derivatives had improved direct-antioxidant activity one of which showed a better DPPH scavenging activity than a standard antioxidant (BHT). The cytotoxicity against human fibrosarcoma cells (HT1080) was tested using 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) reagent. Although BBR exerted a better cytotoxic activity, its phenolic derivatives (Figure 6, E) showed improved up-regulation of superoxide dismutase (SOD) gene expression, indicating cytotoxicity might not be their main effect. Furthermore, two derivatives namely, demethyleneberberine bromide and tetrahydroxyberberine bromide which contain catechol rings showed good antiacetylcholinesterase and more potent anti-oxidant activities (Roselli M *et al.*, 2016). Similarly, *in vitro* examination of antioxidant and anticancer potential of halogenated analogs resulted in a better efficacy compared to berberine (Mistry B *et al.*, 2017).

Lipid-lowering effect

Berberine can decrease blood cholesterol and triglyceride (TG) levels in both human and animal studies (Bagade A, Tumbigeremutt V, Pallavi G, 2017). To enhance the effect of berberine in the treatment of atherosclerosis the therapeutic potential, on extracellular matrix metalloproteinase inducer (EMMPRIN) suppression and on plaque size and vulnerability, of its derivatives was examined in macrophages and mice (Chen J *et al.*, 2014). Compared with BBR treatment of macrophages with dihydroberberine (dhBBR) and 8,8-dimethyldihydroberberine (Di-MeBBR) resulted in a significant reduction of EMMPRIN expression.

Additionally, the derivatives reduced the aortic atherosclerotic plaque size and improved plaque stability and decreased EMMPRIN expression in apoE^{-/-} mice fed with a Western diet.

Anticancer activity

Of the 175 anti-cancer drugs approved during the period from around 1940s to 2014, a large percentage (49%) were either natural products or their direct derivatives (Newman DJ, Cragg GM, 2016). Berberine and its derivatives too have been the focus of many researches in the area of cancer. In an effort to improve the anticancer effect of berberine, a series of phenyl-substituted berberine triazolyl derivatives were designed and synthesized via copper-catalyzed azide-alkyne cycloaddition (CuAAC) click chemistry, and their impact was examined against three human cancer cell lines including MCF-7 (breast), SW-1990 (pancreatic), and SMMC-7721 (liver). From the results obtained it was evident that the anticancer activity of most of the compounds, against all the tested cancer cell lines, was better than berberine. Moreover, the compounds showed lower cytotoxicity to the noncancerous HUVEC (human umbilical vein endothelial cell) cells as compared to berberine (Jin X *et al.*, 2014). Berberine and its 13-arylalkyl derivatives were identified as natural inhibitors of Wnt/ β -catenin signaling pathway which is reported to be abnormally activated in many tumors (Albring KF *et al.*, 2013). Comparative study on the effects of berberine and the derivatives on the viability, cytotoxicity, and Wnt/ β -catenin signaling on HCT116 colon carcinoma cells revealed that the screened derivatives exhibited superior activity in Wnt suppression without affecting viability or showing increased cytotoxicity.

In a study aimed to develop potential new drugs for the treatment of diseases associated with histone acetyltransferase (HAT) dysfunction, B-homo palmatine and berberine derivatives were synthesized from 3,4-dimethoxybenzaldehyde and benzo[d][1,3]dioxole-5-carbaldehyde respectively (Yang Z *et al.*, 2018). An isotope labeling method used to measure their inhibitory activities (IC₅₀) against p300 HAT revealed that the derivatives exhibited potent inhibition of p300 HAT, one of which was suggested for further structural optimization as it was found to be more potent than the known inhibitor C646 against HCG27, HT1080, and Z-138 cancer cell lines (Figure 6, B). The anti-tumor activity of 9-O-substituted berberine derivatives

containing aza-aromatic terminal group was measured by evaluating their ability to stabilize human G-quadruplex telomeric DNA (Ma Y *et al.*, 2009). Results of molecular docking studies indicated that the terminal aza-aromatic group N⁺ fitted well in the grooves of G-quadruplex with much higher binding affinity compared to berberine. It was proposed that for G-quadruplex binding activity and selectivity central N⁺-containing chromophore ring and terminal positively charged aza-aromatic substituent might be beneficial. A similar study aimed to find a compound which might show a higher affinity and specificity to G-quadruplex structures (GqS), seven derivatives of palmatine (an isoquinoline alkaloid) with peptide side chains were synthesized. It was confirmed, from molecular docking assays results of the peptide side chains on four different parallel G-quadruplex structures, that the derivatives possessed a higher affinity to GqS than palmatine alone (Franceschin M *et al.*, 2018).

To exploit the anticancer effect of berberine, three 13-arylalkyl derivatives were synthesized, and their effects were tested against two human colon cancer cell lines, HCT116, and SW613-B3. The derivatives induced cell cycle arrest and cell death through apoptosis and were more potent than the lead compound (BBR) (Guaman Ortiz LM *et al.*, 2014). Similar results were observed using other five derivatives which were found to be more cytotoxic for both p53wt (HCT116) and p53mutated (SW613-B3) colon cancer cell lines (Guamán Ortiz LM *et al.*, 2015). It was confirmed through fluorescence analysis that they were localized within the cytoplasm of the cell and triggered autophagy. Additionally, an investigation of the efficacy of BBR and some of its derivatives, which have aromatic moieties bonded to the 13-position of berberine, in the management of metastatic breast cancer was carried out in HER-2/neu overexpressing SK-BR-3 breast cancer cells (Pierpaoli E *et al.*, 2013). The synthetic derivatives and BBR exerted a dose- and time-dependent inhibition of cell viability and two derivatives showed greater effectiveness than berberine.

In a search for a selective inhibitor of the mitogen-activated protein kinase (MAPK) which regulates proliferation, differentiation and cell death in eukaryotes, a protoberberine derivative library was screened against MAPK (Kim N *et al.*, 2014). One compound (HWY336) which selectively inhibited the kinase activity of mitogen-activated protein kinases 4 and 7 (MKK4 and MKK7) by interfering with access of a protein substrate to their binding site was identified and suggested for further structural modification to improve its selectivity.

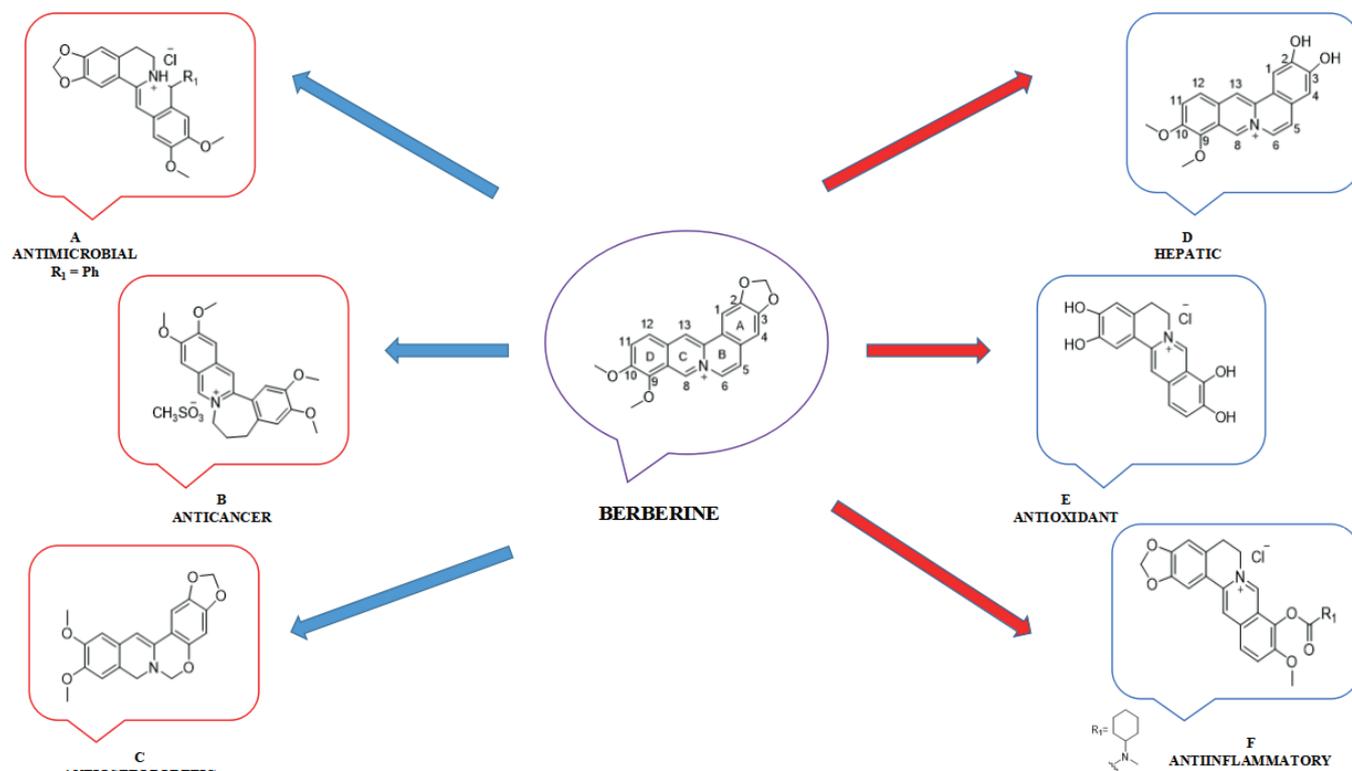


FIGURE 6 - The chemical structures and therapeutic effects of promising synthetic berberine derivatives.

Besides, twenty five new berberine (BBR) derivatives with substituents on position 3 or 9 of BBR targeting indoleamine 2,3-dioxygenase 1 (IDO1) were synthesized as cancer immunotherapy agents (Wang YX *et al.*, 2018). It was proposed that for potency enhancement large group at the 9-position might be beneficial. Two compounds which inhibited IFN-gamma-induced IDO1 expression through the activation of AMPK and subsequent inhibition of STAT1 phosphorylation were suggested as promising IDO1 modulators for cancer immunotherapy for further investigation.

To overcome the limitation (development of drug-resistance) of BRAF (an enzyme in the mitogen-activated protein kinase signaling pathway) inhibitors in the treatment of cancer, compounds with similar structure to berberine were sorted (from PubChem database) and their binding efficiency was studied by molecular docking. Of the 1544 sorted compounds, ten chemical structures were ranked as suitable berberine derivatives based on thermodynamics parameters. Based on the highest inhibition constants scores (greater than 6), BBR and most of its derivatives were found to be optimal BRAF inhibitors. Interestingly, three novel

BBR derivatives, which were not reported previously in the PubChem database, were generated (BBR-9, BBR-7, and BBR-10). Addition of trifluoromethyl benzyl group to BBR at three positions (9, 10, and 13 for BBR-9, BBR-7, and BBR-10, respectively) increased the inhibition constants of these compounds. BBR-10 which was found to bind to the active sites of BRAF better than BBR was identified as a BBR-derived lead candidate for BRAF inhibition (Kaboli PJ, Ismail P, Ling K-H, 2018).

Antidiabetic activity

Modification of the structure of BBR is expected to enhance its glucose-lowering effect for the treatment of diabetes. In a study designed to discover new synthetic anti-diabetic BBR derivatives, thirty five compounds were screened for their activities on glucose consumption in HL-7702 cells. One of the candidates with an amide bond and amidogen at 9-position showed stronger potency than BBR (Ren G *et al.*, 2017). Results indicated that amide bond at 9-position and the introduction of electron-donating groups at 9-position are essential to maintain BBR's glucose-lowering effect and to enhance

its potency respectively. Moreover, the *in vivo* efficacy of berberine and its derivative (dihydroberberine) in the improvement of insulin sensitivity was compared in rodent models of insulin resistance. By inhibiting mitochondrial complex I, dihydroberberine was able to counteract tissue triglyceride accumulation, adiposity, and insulin resistance which was likely due to enhanced oral bioavailability (Turner N *et al.*, 2008).

Anti-inflammatory effect

To improve the anti-inflammatory effect of berberine, discovery of novel derivatives is essential. A preliminary study on twenty three new berberine analogues, which revealed their anti-inflammatory action, suggested that BBR derivatives could be developed into a new class of anti-inflammatory drugs (Wang YX *et al.*, 2017). Based on Structure-activity relationship (SAR) analysis it was indicated that their anti-inflammatory potency was enhanced probably due to the suitable tertiary/quaternary carbon substitutions at the 9-position or rigid fragment at position 10. All of the compounds exerted their effect by inhibiting TNF- α -induced NF- κ B activation. Along these lines, among a series of 9-O-substituted-berberine synthetic derivatives, two promising compounds (Figure 6, F) which displayed their effect in part through suppression of the of the NF- κ B signal pathway were identified to be better antiinflammatory agents than berberine (Huang MY *et al.*, 2016). Their activity was confirmed by both a decrease in the *in vitro* levels of NO, TNF- α and IL-6 in lipopolysaccharide (LPS)-induced RAW264.7 cells, and by the *in vivo* inhibition of migration of neutrophils and primitive macrophage in injured transgenic zebrafish larvae. It was suggested that the introduction of a hydrophobic group to the C- 9-O position of BBR might be beneficial for improved anti-inflammatory effect.

Furthermore, the therapeutic potential of demethyleneberberine (DMB) in inflammatory intestinal disorders was studied on dextran sulfate sodium (DSS)-induced mice colitis where DMB markedly inhibited *in vitro* production of ROS (reactive oxygen species) and pro-inflammatory cytokines in RAW264.7 cell line (Chen YY *et al.*, 2017). Moreover, *in vivo* results indicated that it significantly diminished myeloperoxidase (MPO) activity, reduced the production of pro-inflammatory cytokines [interleukin (IL)-6 and tumor necrosis factor-a (TNF-a)], alleviated the weight loss and inhibited the activation of NF- κ B signaling

pathway. DMB also decreased interferon (IFN)-c, increased IL-4 concentration in the mice splenocytes and the ratio of IgG1/IgG2a in the serum.

Irritable bowel syndrome and osteoporosis treatment

In an attempt to discover better 5-hydroxytryptamine (5-HT) receptor modulators with fewer side effects for irritable bowel syndrome (IBS) treatment, biological evaluation tests were done on twelve BBR derivatives on a rat model of IBS (Deng X *et al.*, 2012). All the tested compounds showed potential for IBS therapy which was measured based on the feces water ratio and rectal distension-induced nociception of IBS rats. To improve the activity of berberine in the promotion of osteogenic differentiation, a new compound was synthesized by substituting its quaternary amine group with uncharged tertiary amine and changing the position of the methoxy group. Evaluation of the osteogenic effects of the compound (Figure 6, C) in C2C12 pre-osteoblastic myoblast cells revealed that it enhanced alkaline phosphatase (major osteogenic marker) activity and osteoblast differentiation through the regulation of PPARgGSK3b/b-catenin axis (Han Y, Kim MJ, Lee KY, 2018).

Hepato-therapeutic and -protective effects

The therapeutic potential of berberine and its synthetic derivatives have also been tested for the treatment and protection of some liver diseases. A recent study has demonstrated the hepatoprotective and anti-fibrotic effects of DMB, *in vivo*, using thioacetamide (TAA)-induced hepatic fibrosis model in mice (Wang Y *et al.*, 2016). By modulating the nuclear factor- κ B (NF- κ B) signaling pathway, DMB suppressed the activation of hepatic stellate cells (HSCs) and induce cell apoptosis. Moreover, DMB reduced the expression of TGF- β 1 (transforming growth factor β 1) and TIMPs (tissue inhibitors of MMPs) which consequently decreased ECM (extracellular matrix) synthesis and promotes collagen degradation. Results of the study suggested that it has a better safety profile as compared to the parent compound (BBR). In a study aimed to evaluate the protective effect of DMB on alcoholic liver disease (ALD), DMB was employed as a natural mitochondria-targeted antioxidant in *in vivo* as well as in *in vitro* studies (Zhang P *et al.*, 2015). DMB was detected in significant amount in both mitochondria and cytoplasm

isolated from HepG2 cells. It significantly ameliorated oxidative stress and mitochondrial dysfunction in acute ethanol-treated mice model. Additionally, DMB Suppressed CYP2E1, HIF-1 α (hypoxia-inducible factor α , and iNOS (inducible nitric oxide synthase), which contribute to ethanol-dependent oxidative stress. In the chronic ethanol-fed mice, DMB treatment suppressed lipid peroxidation and macrosteatosis in the liver.

Examination of the therapeutic effect of DMB on non-alcoholic fatty liver disease (NAFLD) was conducted using both *in vivo* (choline-deficient (MCD) high-fat diet mice) and *in vitro* (Palmitic acid (PA)-treated HepG2 cells) models (Qiang X *et al.*, 2015). DMB treatment resulted in significant reduction of PA- and MCD-induced hepatic oxidative stress and inflammation as well as lipid accumulation in both *in vitro* and *in vivo* NAFLD models which was proposed to probably be through the activation of the AMPK (AMP-activated protein kinase) signaling pathway. Furthermore, the effect of DMB (Figure 6, D) on isoniazid (INH) induced liver injury was studied in 8-week-old male C57 mice model in which it was reflected that DMB reversed the INH-induced liver injury in a dose-dependent manner (Xu L, Qiang X, Zhang Y, 2016). The protective effect of DMB was proposed to be associated with its regulation of lipid metabolism, i.e. reductions in liver TG (triglyceride) and TC (total cholesterol), reduction of CYP2E1 enzyme expression and inhibition of ER (endoplasmic reticulum) stress. Moreover, photographic and microscopic observation of livers by hematoxylin and eosin (HE) staining showed much fewer lipid droplets deposited inside the parenchyma cells.

Effect on neurodegenerative diseases

There is a need for the treatment of the neurodegenerative disease which are incurable disorders with an increasing prevalence (Duraes F, Pinto M, Sousa E, 2018). Natural products and/or their derivatives are potential sources of new therapies for these disorders too. Berberine and its derivatives have been tested for the treatment of these diseases. In a study determination of the potential of a series of 9-O substituted berberine derivatives as dual *in vitro* inhibitors of acetylcholinesterase/butyrylcholinesterase (AChE/BuChE) was done. It was proposed that these compounds might show better clinical efficacy for the treatment of Alzheimer's disease (AD). Results of the study revealed that most of the compounds showed

potent activity to BuChE and that the AChE inhibitory activities were affected dramatically by the bulk of the scaffold of the compounds at the end of the chain (Huang L *et al.*, 2010). A quaternary derivative (berberine linked with 3-methylpyridinium by a 2-carbon spacer) was found to be the best potent inhibitor bound at both the ligand binding sites of AChE and another compound, berberine linked with 2-thionaphthol by a 4-carbon spacer, was the most potent inhibitor for BuChE. To find a better treatment alternative for Parkinson's disease (PD), *in silico* tools were employed to synthesize a set of semi-synthetic berberine derivatives and were tested against proteins involved in the onset or progression of the disease (Ribaud G *et al.*, 2018). The predicted pharmacokinetic properties, based on *in silico* physicochemical parameters evaluation, were confirmed by *in vitro* assay on MAO-B (monoamine oxidase). One of the tested derivatives appeared to be a potential anti-PD agent targeting α -synuclein, phosphodiesterase (PDE4), and MAO-B (Huang L *et al.*, 2010).

CONCLUSION AND FUTURE PERSPECTIVES

Historically natural products, especially those from plants, have been used as a source of new drug molecules owing to their favorable biological properties, acceptable safety profile and synergistic effect of formulations containing multiple constituents. Berberine is a promising natural product with a variety of therapeutic activity. However, it is not yet developed into a marketed drug product. To achieve a successful development of berberine, addressing the barriers and summarizing the efforts made to facilitate its development is important. Structural modification of berberine to obtain derivatives with improved physicochemical properties, therapeutic effects and safety profile is perceived to be an effective approach which can facilitate the development of berberine-based drug products. Preclinical scientific researches carried out in the recent past have confirmed the therapeutic potential of a significant number of synthetic berberine derivatives against various diseases. This review discusses the pharmacological activities of berberine, the barriers that limit its development and the strategy (structural modification) used to enhance its therapeutic effects and hence its clinical applicability. For successful future development, the synthetic pathways should be designed in such a way that allows their easy and cost-effective large-scale manufacture. An important consideration is the assessment of

physicochemical, biopharmaceutical and toxicological properties of promising synthetic derivatives in the early development stage. This assessment enables the development of products with suitable pharmaceutical properties and consequently secure a quick progress of stable, effective, and safe berberine-based drug products to the market. Obviously, this requires multidisciplinary collaboration.

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