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Berberine: A pioneer remedy for various Ailments

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Abstract
Herbal drug have always been the centre of attraction for the treatment and prevention of various diseases and for the maintenance of healthy lifestyle. Bioactive compounds derived from natural products have been used for the treatment of ailments for thousands of years. Berberine is one such drug which is obtained from various plant sources which has gained massive attention as a therapeutic agent against numerous diseases like hyperlipidemia, diabetes, obesity, fatty liver diseases, and coronary artery disease. But it feels very hard to reach these drugs to the commercial level due to the low solubility and bioavailability problems. Interestingly, herbal drugs offer several advantages over the allopathic system due to the avoidance of severe side effects and toxicity. Recently, advanced techniques have been investigated to enhance the bioavailability of such drugs. In this review, we have summarized the facts about berberine along with the problems associated with its bioavailability and approaches to overcome these problems.

Keywords: Berberine, bioavailability, allopathic, side effects

1. Introduction
Herbal drugs have been used from time immemorial ever since the ancient man searched for an alternative around him to develop remedies which could help to lessen the pain and cure illness [1]. Constituents derived from natural sources especially plants have been the major source of medicine for centuries. [2]. These plants produce a variety of secondary metabolites (SM) which serves to be the defense agents against herbivores and microbes, but also as signal compounds. Generally, SM exhibits a variety of biological and pharmacological properties which have been and are still used to treat infections and health disorders [3]. Majority of herbal medicines lacks data about their ADME and pharmacokinetic properties in humans. Cytochrome P450s (CYPs) and uridine diphosphate glucuronosyl transferases (UGTs) play a major role in Phase I and/or Phase II metabolism of herbal compounds. P-glycoprotein (P-gp/MDR1/ABCB1) is highly expressed in the intestine, liver, brain, and kidney making some herbal ingredients its substrates [4]. Therefore, it is necessary to gain knowledge about the biological fate and disposition pathways of herbal remedies to optimize their usage [5].

1.2 Berberine
Berberine is a natural quaternary benzylisoquinoline plant alkaloid which has been widely used in Ayurvedic and Chinese medicinal systems with proven medicinal properties. Berberine is present as active constituent in stem and bark part of various plants including *Hydrastis canadensis* (goldenseal), *Coptis chinensis* (Coptis or goldenthread), *Berberis aquifolium* (Oregon grape), *Berberis vulgaris* (barberry), and an Indian species *Berberis aristata* (Tree turmeric) [6-7]. Berberine has been used in traditional Chinese, Indian, and middle-eastern folk medicine for more than 400 years [8]. The alkaloid has gained massive attention as a therapeutic agent against numerous diseases like hyperlipidemia, diabetes, metabolic syndrome, polycystic ovary syndrome, obesity, fatty liver diseases, and coronary artery disease. Presence of diverse pharmacological activities in berberine indicates its potential as drug for a wide range of clinical manifestations [9].

1.3 Pharmacokinetics of Berberine
Poor oral bioavailability of berberine suggests that necessary effects may never be achieved by patients taking berberine as medical treatment. Low oral bioavailability of berberine may be the consequences of poor absorption and first pass effect in the intestine and liver. Self-aggregation, poor permeability, p-glycoprotein mediated efflux and hepatobiliary re-excretion further contributes to the poor absorption of berberine [10]. Pharmacokinetic studies in

humans show that the required therapeutic concentration is only achieved after chronic administration which aggravates the conditions of having adverse side effects at high doses. [11].

1.4 Metabolism of Berberine

The plasma level of Berberine is very low in contrast to the significant pharmacological effects [12]. Berberine goes through profound metabolism on oral administration which provides tremendously low plasma coverage. For that reason, it is supposed that the metabolites of berberine too add a lot to its pharmacological effects [13]. A variety of enzymes seems to mediate the metabolism of berberine including CYP1A2, 3A4, 2D6 and UDP-glucuronosyltransferases, which was metabolized with phase I demethylation and phaseII glucuronidation in liver. Metabolites of berberine are berberrubine, demethyleneberberine, jatrorrhizine, thalifendine along with its glucuronidation compounds [14].

1.5 Chemistry of Berberine:

The term berberine was invented by Buchner and Herberger for a yellow extract obtained from Berberis vulgaris in 1830 [15]. Berberine is a plant quaternary ammonium salt from the group of isoquinoline alkaloid (2, 3-methylenedioxy-9,10-dimethoxyprotoberberine chloride; $C_{20}H_{18}NO_4^+$), with a molar mass of 336.36122g/mol which can be isolated from a variety of plants, such as *Coptis chinensis* (Coptis or Goldthread), *Hydrastis Canadensis* (goldenseal), *Berberis aquifolium* (Oregon grape), *Berberis aristata* (Tree Turmeric) and *Berberis vulgaris* (Barberry), and *Acrangelisia flava* [16].

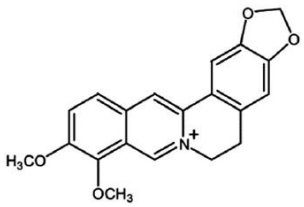

Fig 1: Chemical Structure of Berberine [17]

The isoquinoline skeleton of berberine opens new ways to create various derivatives using rational drug design. A structure-cytotoxicity relationship of berberine derivatives reveals importance of changes to functional group which creates a great impact on the biological activity. Further changes in functional groups, modification of the size and units of the chemical structure is primarily done to enhance the biological activity, lessen the toxicity, and attain the required physicochemical properties. But there is still an enormous scope of bringing in specific moieties for different therapeutic areas at various positions in the berberine skeleton and also at other positions of the isoquinoline moiety [18].

1.6 Pharmacology of Berberine

Berberine is a vital therapeutic herb that has been successfully employed as folk medicine for a long time in china and other countries for treating dysentery, diarrhea, stomatitis, throat infections, and hepatitis [19]. Barberry fruit has been used by ancient Egyptians with fennel seeds to fight off pestilent fevers. In Indian Ayurvedic system barberry was used for treating dysentery and as sedative in traditional Iranian medicine. Barberry was used to treat gall bladder and liver problems in northern Europe, whereas it was utilized in the management of abnormal uterine bleeds and rheumatism in Russia and Bulgaria [20]. Berberine has verified a broad variety of pharmacological activities including; antihypertensive, anti-inflammatory, antioxidant, antidepressant, anticancer, anti-diarrhoeal, cholagouge, hepatoprotective and above all, antimicrobial. Latest learning's about berberine have thrown light on antidiabetic and hypolipidemic activities of the alkaloid [21].

1.6.1 Berberine as antidiabetic and hypolipidaemic agent:

According to some latest clinical studies berberine has been found to show its antidiabetic effect by controlling the efficacy of various effectors, stimulation of glucose uptake in various places (muscles, liver and adipose), inhibiting gluconeogenesis by inhibition of enzymes involved in it. [22].

Table 1: Research work on anti-diabetic activity of berberine

S. No.	Author (year)	Antidiabetic activity	Hypolipidemic activity	Ref.
1.	Liu C *et al.* (2015)	Glucose tolerance tests documented that berberine-treated mice were more glucose tolerant.	Histological analyses revealed that the treatment of berberine inhibited hepatic fat accumulation.	[23]
2.	Gu Y *et al.*(2010)	Berberine is shown to aid in the treatment of type 2 diabetes via down regulating the high levels of free fatty acids in patient serum, thus mediating glucose and lipid metabolism.	Fatty acids are pharmaceutical targets for the treatment of diabetes.	[24]
3.	Yin J *et al.* (2008)	Berberine exerted similar hypoglycemic effect similar to metformin by lowering fasting blood glucose and postprandial blood glucose.	Total cholesterol and low density lipoprotein cholesterol (LDL-C) were considerably decreased.	[25]
4.	Zhang Y *et al*. (2008)	In berberine reduced fasting and post load plasma glucose.	Changes in serum lipid concentrations were observed.	[26]
5.	Yin J *et al.*(2002)	Berberine exerted a glucose lowering effect in hepatocytes which is insulin independent and similar to that of metformin.	Not studied	[27]

1.6.2 Anticancer activity of berberine

Lately, *in vitro* studies using cancer cell lines have shown that berberine inhibits cancer cell proliferation and migration, and stimulates apoptosis in a range of cancer cell lines, encouraging supplementary expansion of derivatives for drug-base cancer avoidance and management. Numerous results proposed that the molecular structure of berberine is capable to bind DNA, other nuclear and cytoplasmic targets [28].

Table 2: Research work on anticancer activity of berberine

S.No.	Author (year)	Design of experiment	Anticancer activity	Ref.
1.	Balakrishna A *et al*. (2015)	Evaluation of synergistic anticancer activity of berberine and curcumin on specified human cell lines (A549, HepG2, MCF-7, Jurkat, and K562).	Results proved the synergetic anticancer activity of Berberine with Curcumin inducing cell death greater percentage of >77% when compared to pure curcumin with <54% and pure berberine with <45% on average on all cell line models.	[29]
2.	Xi S *et al*. (2014)	Effect of berberine on activity and mRNA expression of N-acetyl transferase in human lung cancer cell line A549.	The N-acetyltransferase content in human lung cancer A549 cells decreased with the increasing of berberine concentration, significantly lower than that in the control group.	[30]
3.	Mittal A *et al*. (2013)	To study the anticancer effect of berberine in combination with doxorubicin on murine melanoma B16F10 cells *in vitro* and *in vivo*.	This drug combination strongly inhibited cell growth strongly inhibited cell growth and induced cell death, and caused G2/M arrest.	[31]
4.	Lin CC *et al*. (2007)	Examination of the effects of berberine on cell growth, apoptosis and cell cycle regulation in oral squamous carcinoma HSC-3.	Berberine induced dose and time dependent irreversible inhibition of cell growth and DNA synthesis.	[32]
5.	Letasiova S *et al*. (2006)	Studies on antiproliferative activity *in vitro* and induction of apoptosis/necrosis of the U937 (human tumor cell line) and B16 (murine melanoma) cells.	Berberine induces apoptosis of the U937 and B16 cells. Cell lysis/necrosis of the berberine treated B16 cells was observed as a result of the integrity damage of the cytoplasmic membrane.	[33]

1.6.3 Antibacterial activity of berberine

Extract of berberine have been known to be efficient against various organisms, including bacteria, viruses, fungi, protozoans, helminthes and Chlamydia. Research work envisaged about antibacterial activity of berberine confirmed its inhibitory effect on the augmentation of a number of bacterial pathogens such as *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *E. coli,* and *Bacillus subtilis* [34].

Table 3: Research work on antibacterial activity of berberine

S.No.	Author (year)	Design of experiment	Antibacterial activity	Ref.
1.	Y Xiang D *et al*. (2017)	To study the effect of berberine on *Clostridium perfringens* induced necrotic enteritis in broiler chickens.	Berberine controlled necrotic enteritis caused by *Clostridium perfringens.*	[35]
2.	Bandyopadhyay S *et al*. (2013)	The study was aimed to investigate the antimicrobial activity of berberine.	The results showed that berberine can be used as a good antibacterial agent against multi drug resistant *E.coli*.	[36]
3.	Anubhuti P *et al*. (2011)	The research work was aimed to monitor the antimicrobial activity of berberine against five bacterial strains by agar diffusion method.	Berberine exhibited antimicrobial activity against tested strains.	[37]

1.7 Toxicology studies of Berberine:

On the basis of animal and *in vitro* studies the efficacy of berberine to provoke gestrointestinal upset and ulceration, immune-toxicity, photo-toxicity, neuro-toxicity and cardio-toxicity in a dose dependent approach. Berberine inhibits adenine nucleotide translocase while promoting reactive oxygen species formation which induces its toxicity on cancer as well as normal cells in a time and dose dependent behavior. In addition, berberine is known to be quite controversial during pregnancy, so it is suggested to be cautious while using berberine in pregnancy and neonatal. Lastly its inhibitory effects on CYP enzymes should be taken under consideration because it can lead to indirect toxicity. This interaction is significant while administering narrow therapeutic window drugs which may enhance the drug plasma concentration and toxicity [38].

1.8 Problems associated with the delivery of berberine:

Berberine has lately achieved great interest because of vast therapeutic efficacy, low toxicity, and low cost properties. Nevertheless, poor intestinal absorption limits the wide application of berberine [39-40].

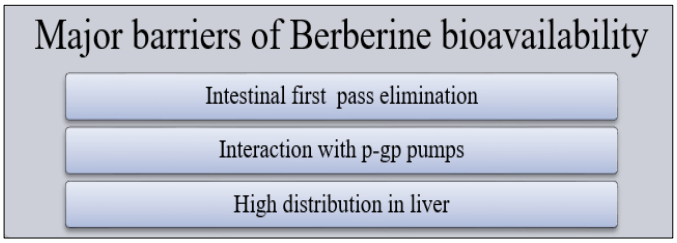

Fig 2: Major barrier in berberine bioavailability

1.8.1 Interaction with p-gp pumps

P-glycoprotein (P-gp) is a membrane-bound protein with 170 kDa molecular weight which is an energy-dependent efflux transporter. The main physiology of p-gp is to limit the damaging contact of toxins, drugs, and xenobiotics to the body by throwing them out of the cells. It plays a significant role in modulating the pharmacokinetic characteristics of several clinically essential therapeutic agents which leads to its screening into the drug discovery process [41]. It has been revealed that berberine is a substrate compound of p-glycoprotein which plays an important role in the absorption of berberine -**.** As expected, when berberine was administered

With P-gp inhibitor (cyclosporine or verapamil), there was a significant improvement in the absorption of berberine which indicates the involvement of intestinal efflux transporters in the excretion of berberine into intestinal lumen causing poor absorption and thus low bioavailability [42].

1.8.2 Intestinal first pass elimination

A study in rats concluded that the first pass elimination of berberine takes place primarily in the small intestine rather than in the liver and stomach among which liver dominates over the other organs in its tissue distribution. Oxidative demethylation of berberine and consequent glucuronidation are the most important means for the intestinal metabolism of berberine after oral intake [42].

1.8.3 High distribution in liver:

A research work by Tan XS *et al.* in 2013 indicated that berberine has been mainly distributed in the tissues, and the concentration of berberine in tissues was found to be much higher than that in the plasma. Berberine was detected in liver, kidneys, muscle, lungs, brain, heart, pancreas and fat in a decreasing manner of distributed amounts within 48h. The pharmacokinetic studies recommended that berberine get to the tissues within 0.25 h after dosing and its level in most of the examined tissues was higher (or much higher) than that in the plasma at 4 h after administration [43].

1.9 Patents related to Berberine:

A lot of work related to berberine has been patented by different researchers which makes it a promising candidate for future work. Some of the work related to berberine patents is mentioned in the table.

Table 4: Patents related to berberine

S.No.	Patent No.	Patent Title	Inventor/Assignee	Year	Ref.
1.	WO2016210230 A1	Therapeutic uses of berberine formulations	Carl Oscar BROWN, III, Po-Yuan TSENG, IYin Lin, Chen-En Tsai, Chih-Kuang Chen	2016	[44]
2.	US20150320738 A1	Berberine-containing pharmaceutical composition for inhibiting cancer stem cell growth or carcinoma metastasis and application thereof.	Hsiu-Mei Hsieh, Chen-Yu Lee, Chih-Chien Shen, Tien-Chun Wang/Chen-Yu Lee, National Taiwan Normal University	2015	[45]
3.	US20150258078 A1	Compositions containing berberine and pantethine and methods for treatment of lipid metabolism disorders	Alexander R. Shikhman/Alexander R. Shikhman	2015	[46]
4.	WO2013063271 A1	Artemisinin and berberine compositions and methods of making	Bob Rosen	2013	[47]
5.	WO2012108892 A1	Combinations of berberine, artemisinin, loperamide and their derivatives to treat malaria, diarrhea, travelers' diarrhea, dysentery, dengue fever, parasites, cholera and viruses	Kirk Seubert, James Spencer, John Colman	2012	[48]
6.	WO2011119649 A3	Pharmaceutical compositions containing berberine for treatment or prevention of weight gain and obesity associated with antipsychotic drugs	Gareth Davies, Yueshan Hu	2012	[49]
7.	US20110086872 A1	Berberine as a selective lung cancer agent and other compositions and methods	Maung Tin-Wa/Maung Tin-Wa	2011	[50]
8.	US006280768B1	Berberine alkaloids as a treatment for chronic – protozoally induced diarrhea	Joseph T‘ McDevltt	2001	[51]

1.10 Approaches to improve the bioavailability of Berberine

Research work that has been endeavored regarding berberine showed that it displays very low plasma level after oral administration in humans due to low oral bioavailability and poor absorption through the gut wall along with extensive metabolism. Several techniques can be used to improve its oral bioavailability which can be divided into different categories as pharmaceutical (development of suitable formulations) or pharmacokinetic (using bioenhancer) [52]. Some studies reveal that berberine alkaloids belong to a large group of cationic toxins that bind with P-glycoprotein which is an important aspect in limiting the absorption of berberine [53]. Therefore it is necessary to solve the bioavailability related problems of the drug to enhance its therapeutic efficiency. This can be done by one of the following methods [54]:

- Preparation of novel non-P-gp substrates
- Combination of P-gp inhibitors with drug
- Developing formulations which allow the drug to bypass efflux pump transport

For that reason, it is needed to develop a novel formulation of berberine which can improve its oral bioavailability and therapeutic effect [55]. Luckily, a range of formulation ideas have been studied to enhance the solubility and bioavailability of such drugs. Numerous approaches such as absorption enhancers, chemical modification and dosage forms have been discovered to facilitate peroral delivery of drugs [56].

1.11 Research Envisaged in enhancing the bioavailability of berberine

A lot of research work has been done for increasing the bioavailability of berberine. Some of it is mentioned in the table below;

Table 5: Research work on bioavailability enhancement of berberine

S.No.	Formulation	Author (year)	Findings	Reference
1.	Berberine nanoparticles	Khayam MU *et al.* (2018)	Berberine nanoparticles showed marked improvement in solubility and dissolution rate by converting crystalline structure into semi-crystalline form.	[57]
2.	Berberine Cremochylomicrons	Elsheikh MA *et al.* (2018)	There was considerable improvement (about 2 fold) in bioavailability of cremochylomicrons as compared to plain berberine.	[58]
3.	Berberine solid dispersion with Eudragit S100	Guo S *et al.* (2017)	Solid dispersion of berberine with Eudragit S100 showed decrease in IC_{50} values as compared to pure berberine.	[7]
4.	Berberine bioadhesive microspheres	Zhang Y *et al.*	Prepared formulation enhanced the bioavailability of berberine by 1.5	[59]

		(2016)	fold as compared to commercial tablets and also exhibited sustained release for 48 h.	
5.	Berberine phytosomes	Yu F et sl. (2016)	This novel formulation was effective in increasing the bioavailability of berberine by 3 fold.	[60]
6.	Berberine novel hydrogel loaded with gold nanoparticles	Souza CR *et al*. (2015)	Novel hydrogel loaded berberine gold nanoparticles enhanced the release of berberine.	[61]
7.	Amorphous solid dispersion of berberine with hydrogenated hosphatidylcholine	Shi C *et al*. (2015)	Results indicated marked improvement in absorption and bioavailability of solid dispersion as compared to pure berberine.	[62]
8.	Berberine loaded self nanoemulsifying drug delivery system (SNEDDS)	Ke Z *et al*. (2015)	Optimized batch of berberine SNEDDS released 90% of drug in just 20 min whereas it took to 2h for the commercial products to release 90% of drug.	[63]
9.	Solid dispersion of berberine with absorption enhancer (Sodium Caprate)	Zhaojie M *et al*. (2014)	Pharmacokinetic evaluation showed 3-fold improvement in *in vitro* membrane permeation and a 5-fold improvement *in vivo* bioavailability of optimized batch in comparison to berberine or berberine tablets.	[64]
10.	Berberine loaded solid lipid nanoparticles	Xue M *et al*. (2013)	Bioavailability of berberine was considerably improved as compared to pure berberine along with enhancement in antidiabetic efficacy.	[65]

Conclusion and future directions

In conclusion, we can say that berberine is a highly efficacious drug for various disorders. However, this drug faces significant challenges to improve its oral bioavailability which include interaction with P-gp pump, intestinal first pass elimination and high distribution rate in liver. To overcome these problems various approaches can be applied like preparation of novel non-P-gp substrates, combination of P-gp inhibitors with drug, developing formulations which allow the drug to bypass efflux pump transport along with the preparation on novel drug delivery systems such as nanoparticles, liposomes, solid dispersion etc. A lot of research work has been done in improving the bioavailability profile of berberine but there are still some barriers which hinder the commercialization of this phytoconstituent. Clinical trials need to be conducted to confirm its efficacy and potential which will provide a basis for its commercialization and will help in reaching to the market.

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