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Review Article

Picrorhiza kurroa, Royle ex Benth: Traditional uses, phytopharmacology, and translational potential in therapy of fatty liver disease

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ABSTRACT

Picrorhiza kurroa Royle ex Benth, *Kutki (P.kurroa*) is an important medicinal plant, traditionally recommended and used in *Ayurveda* for millennia, with certain cautions. There has been a significant revival of keen interest in its pharmacology, pharmacognosy, and phytochemistry for the last few decades. The evidence of its hepatoprotective activity, in experimental and clinical studies, accelerated the correlation of the specific phytochemical constituents of *P.kurroa* with precise pharmacological activities. Iridoid glycosides, particularly picrosides, emerged as the active molecules. For effective translation of traditional remedies into modern therapy, value addition by mechanistic understanding of molecular actions, drug targets, the degrees of efficacy and safety as well as convenient dosage forms is needed. Reverse pharmacology approach and phytopharmaceutical drug category facilitate such a translation. The present review illustrates how a potential translation of traditional practices of using *P.kurroa* into a phytochemically standardized, clinically targeted natural product for global unmet medical needs viz. Fatty liver disease can be attained.

1. Introduction

Picrorhiza kurroa Royle ex Benth, *Kutki*- (*P.kurroa*) finds mention for its clinical uses in an ancient and classical treatise of *Ayurveda*-*Charaka-samhita* [1]. Its application for diverse clinical conditions has continued even now since the medieval period [4–7]. Its clinical usefulness, with some precautions, has consensual validity in the contemporary Asian medicine practices [8–10]. Since the second half of the last century until today, *P. Kurroa* has generated a lot of interest in amongst modern biomedical scientists, as evident through the scientific publications [11–14]. The evidence of its hepatoprotective activity, in experimental and clinical studies accelerated the correlation of the specific phytochemical constituents of *P.kurroa* with precise pharmacological activities [15]. Fascinatingly, its potential utility in SARS CoV2 through *in-silico* studies has also been postulated [16] and clinical studies in combination with other herbs has been explored in COVID-19 [17].

Paradoxically, ever-increasing *Ayurvedic* usage demand and interest in the *P.kurroa-Kutki* has led to it's over harvesting, and the plant which has a limited natural habitat mainly at high altitude of sub-Himalayan region is now listed in the category of 'endangered species' [18]. Interestingly, plant tissue culture and genetic engineering techniques are being explored for micropropagation and to enhance the yield of its phytoactives [19–21]. Iridoid glycosides viz. Kutkins, kutkosides and picrosides, cucurbitacin, triterpenes and simple phenols such as apocynin are the most explored phytoactives of *P.kurroa* for their biological activity such as hepatoprotective [22–24], anti-inflammatory [25,26], anti-arthritic [27], anti-diabetic [28], anti-asthmatic [29], collagen synthesis-promoting and collagenase inhibitory [30], hyaluronidase inhibitory [31], anti-fibrotic [32], anti-oxidant [33,34], immunomodulatory [35,36], anti-carcinogenic [37–40], anti-microbial [41,42], anti-leishmanial [43] etc.

Classically, *P.kurroa-Kutki* has been indicated for diverse clinical conditions such as anorexia, burning sensation, fevers, especially

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intermittent fevers, pre-diabetic and diabetic states, asthma, cough, haemorrhoids, dermatoses, helminthiasis etc. Some of these uses have been investigated (*vide supra*). The plant properties, classically described are adipolysis, excretion of wastes, orexigenic, laxative, anti-inflammatory, hepatoprotective, and diuretic activities; it is favourable to the heart action [44]. However, in traditional clinical practice *P.kurroa-Kutki* is predominantly used for hepatobiliary and gastrointestinal disorders.

The present review covers the classical literature and clinical traditions of usage of the plant, its pharmacology with an eye to develop a phytopharmaceutical for the focussed therapeutic indication of non-alcoholic fatty liver disease (NAFLD), by application of Reverse Pharmacology approach to *P.kurroa* for hepatoprotection, based on its specific phytoactives.

2. Classical literature and clinical tradition of *P.kurroa*-Kutki usage

P.kurroa-Kutki, as a medicinal plant, finds mention in all the three major ancient treatises of *Ayurveda* viz. *Charak-samhita, Sushrut-samhita and Ashtang-hrudaya* [1–3]. Moreover, uninterrupted, continued tradition of clinical use of *P.kurroa-Kutki* is evident by its mention in majority of the Nighantus compiled through medieval period like *Bhavprakash Nighantu, Madanpal Nighantu, Kaiyadev Nighantu, Raj Nighantu* and many more [4–7]. Practicing vaidyas of the recent centuries have recorded in their personal notes their clinical experiences using *P.kurroa-Kutki* [45–49]. *Ayurvedic* volumes of periodicals also find articles on different clinical uses of *P.kurroa-Kutki*. Currently, *Ayurvedic* practitioners use *P.kurroa-Kutki* as a single plant formulation or in combination with other herbs or herbo-mineral complex formulations. Picrorhiza plant species has also been used for diverse healthcare problems by many other Asian traditional systems of medicine [8,9].

P.kurroa-Kutki is a shrub which grows naturally at high altitude of sub-Himalayan regions. The roots and rhizomes of this plant have major medicinal value, that now we know possess major phytoactives. *Ayurveda* classics give importance to the *Desh* (place of collection), *Kala* (time of collection), *Prayojyanga* (part of the plant) etc as important factors for collecting the plants for medicinal purpose [50]. Now, it has been well documented that the altitude, part of the plant and time and light are the determining factors for the phytoactive contents of *P.kurroa-Kutki* [51–53]. The classical literature enlists *Kutki* by different synonyms such as in *Bhavprakash Nighantu* [54]. These synonyms explain either typical characteristics or biological activity viz. *Katumbhara* (full of bitter principles), *Ashoka* (relieves from grief), *Rohini* (protects the organs by regenerative ability), *Krushnabheda* (potential to cause black stools) etc. To illustrate a few biological effects and clinical indications attributed to *Kutki* and relevant to this review are *Lekhaneeya* (adipolysis), *Bhedaneeya* (piercing through and expelling of waste metabolites), *Aruchihara* (improving appetite), *Deepaniya* (facilitate appetite), *Rechani* (laxative action), *Yakrutdalyodar-nashak* (alleviating liver enlargement), *Shotha-nashak* (anti-inflammatory), *Kamala-nashak* (alleviating jaundice), *Jalodarnashan* (useful in ascites), *Pramehaghna* (useful in pre-diabetic and diabetic states), and *Hrudya* (favourable for the heart).

Conventionally, for most of the clinical indications *P.kurroa-Kutki* rhizome powder is used in a dose range of 300 mg–500 mg, two to three times a day, for an adult [44]. For laxative purpose the dose used is 2 gms to 4 gms as a single dose [44]. Although *P.kurroa-Kutki* is also considered as a detoxicating agent (*Vishapaha*), it has been reported to cause toxicity if not used in an appropriate dose for the correct indication [10]. As per our clinical experience of using *P.kurroa*-Kutki and its traditional products, it is observed to have developed adverse effects such as increased bowel frequency, diarrhoea, abdominal gurgling, abdominal colic etc.

Traditional practitioners more often prefer multi-ingredient formulations over single plant-based prescriptions. *Arogyavardhini, Punarnavashtak kwath*, *Mahatiktaka Ghruta*, *Sarivadyasava, MahayograjGuggulu*, *Sudarshan choorna*, *Katukadyavaleha, Tiktadi kwath*, are some of the commonly prescribed multi-ingredient traditional formulations containing *P.kurroa-Kutki* as one of the ingredients which may be in a major proportion or minor proportion. Although intricacy of pharmacodynamics of these multi-ingredient formulations is beyond the scope of this article the Table 1 takes an account of diverse indications of *P.kurroa-Kutki* as stated in different Nighantus. *Arogyavardhini* in a tablet form is one of the most frequently used complex herbo-mineral multi-ingredient formulation, where *P.kurroa-Kutki* the major ingredient is in an equal proportion of the other 12 ingredients [55]. *Arogyavardhini* is commonly used in traditional practice for liver disorders, hepatosplenomegaly, ascites, dyslipidaemia, obesity, skin disorders, as an anti-inflammatory, for improving appetite, digestion, and laxation [56]. Table 2 summarizes the clinical trials reported with *P.kurroa-Kutki* (alone) and *P.kurroa-Kutki* in combination with other agents in liver related and other disorders.

3. Reverse pharmacology approach to Kutki (*P. kurroa*) for hepatoprotection

Continued tradition of clinical use of *P.kurroa-Kutki* and *Kutki*-based multi-ingredient formulations, attracted attention of contemporary bio-medical investigators to investigate this plant. Major clinical and experimental studies of *P.kurroa-Kutki* were undertaken at BHU by Pandey VN, as a post-graduate thesis work in mid 1960s [65] and later submitted Ph.D. thesis on “The effect of indigenous drugs in the management of certain liver disorder” in 1979 [66]. Meanwhile Dhar, ML [67], Das, PK [68] and Kanitkar, SV [69] investigated this drug for its biological activity and hepatoprotective activity in early 70's. Tiwari, NS and Jain, PC investigated *Arogyavardhini* (*Kutki*-50%) clinically for its hypocholesterolaemic activity with special reference to obesity [70]. Vohora, SB and colleagues investigated *P.kurroa-Kutki* with special reference to its choleretic and antimicrobial properties [12]. The plant was also taken up by CDRI, Lucknow for extensive investigations and came out with a hepatoprotective product ‘Picroliv’ in 1990s. Ramesh Chander et al. [13], Dhawan BN and group [71] extensively worked on ‘Picroliv’ to investigate its hepato–protective activity whereas Shukla B et al. [72] investigated the choleretic effect of ‘Picroliv’. Saraswat B, investigated protective effect of Picroliv, active constituent of *P.kurroa*, against oxytetracycline induced hepatic damage [73]. Sporadically clinical trials of *P.kurroa-Kutki* (alone) and *P.kurroa-Kutki* in combination with other agents in liver related and other disorders has been investigated with variable outcomes [59–63].

Our research team has been consistently pursuing for the purpose of translational medicine on the path of reverse pharmacology [74,75]. *Arogyavardhini* formulation (50% *P.kurroa-Kutki*) was studied in acute viral hepatitis (HAV) on the basis of ‘clinical-usage-experience’. Since *Arogyavardhini* was often used along-with *Punarnavadi-Kwath* in traditional practices, the same was adhered-to, and studied in an exploratory open-labelled clinical trial in acute viral hepatitis. This study ensured activity and safety [57]. Taking cue from this exploratory study, a double-blind, placebo controlled, clinical study of *Arogyavardhini* was conducted. This study demonstrated early recovery of symptoms, faster reduction of liver transaminases, quicker clearance of bilirubin and a total reduction in disease duration [58]. Subsequent to this a placebo-controlled study was undertaken with the standardized

Table 1
Diverse clinical indications of *P.kurroa-Kutki* noted from Ayurveda Nighantus' (Compendia of Ayurvedic medicinal plants).

Sr.No	Roghaghnata (Indications)	B.N [1].	K.N [2].	M.N [3].	R.N [4].	D.N [5].	A.N [6].	V.N [7].	P.N [8].	V.G [9].	DG.H [10].	V.V [11].
1	*Malabhedan/Rehan* (Piercing through and expelling of waste metabolites)	+	+		+				+		+	
2	*Krumighna* (Anti-helminthic)	+	+	+					+		+	+
3	*Dahanashaka* (Useful in burning)	+	+	+		+	+			+		+
4	*Jwraghna* (Antipyretic)	+	+	+	+	+	+		+	+	+	+
5	*Kamala nashak* (Alleviating jaundice)						+		+	+	+	+
6	*Raktavikar nashak* (Useful in blood disorders)	+	+			+			+	+	+	+
7	*Sheetapittaghna* (Anti-urticarial)				+		+			+		
8	*Kaphaghna* (Pacifying vitiated Kapha)		+	+	+	+	+		+	+	+	+
9	*Pittaghna* (Pacifying vitiated Pitta)		+	+		+	+		+		+	+
10	*Aruchinashaka* (Improving distaste)				+	+	+			+		
11	*Kushthaghna* (useful in skin diseases)	+	+						+	+		+
12	*Vishaghna* (Detoxicant)									+		
13	*Rajayakshmanivarini* (Alleviating Tuberculosis)				+					+		
14	*Hridrogaharana/Hrudya* (Useful in Heart diseases)	+			+					+	+	
15	*Pramehanashaka* (Anti-diabetic)	+	+							+	+	+
16	*Kasahara/Shwasahara* (Anti-tussive/Anti-asthmatic)	+	+		+						+	+
17	*Chardinashana* (Anti-emetic)											+
18	*Hikkanashana* (Useful in Hiccups)											+
19	*Pandunashaka* (Helpful in Anemia)											+
20	*Kantharoga* (Useful in throat disorder)											+
21	*Raktashodhaka* (Blood purifier)	+	+		+				+		+	
22	*Agnimandya nashaka/Deepani* (Anti-anorexic/Appetizer)	+									+	
23	*Anaha nivarana* (Relieving painful Blotting)										+	
24	*Yakrutdalyudara pleehavruddhi har* (Useful in Ascites & Hepato- splenomegaly)								+		+	
25	*Shothanashaka* (Anti-inflammatory)										+	

(B.N. – Bhavaprakasha Nighantu, K.N.-Kaiyadeva Nighantu, M.N.-Madanapala Nighantu, R.N.-Raja Nighantu, D.N.- Dhanvantari Nighantu, A.N.-Adarsha Nighantu, V.N.-Vanaushadhi Nighantu, P.N.-Priya Nighantu, V.G.-Vanaushadhi Gunadarsha, DG.H.-Dravyaguna Hastamalaka, V.V.-Vanaspati Vijnana).

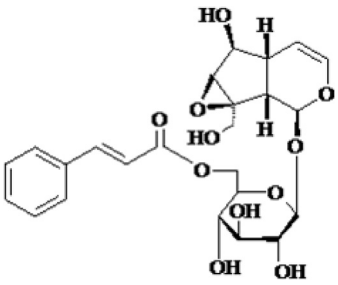
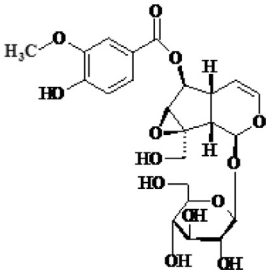
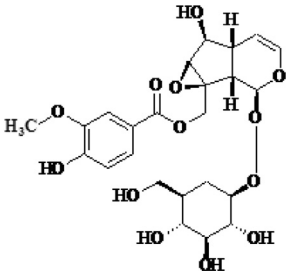
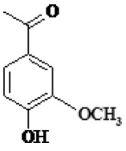
extract of *P.kurroa-Kutki* the major ingredient of *Arogyavardhini*. The efficacy and safety of this extract (standardized for Picroside-I and II) in viral hepatitis was also established [15].

Later researchers at KHS-MRC pursued the activity of *P.kurroa-Kutki* in NAFLD (Non-Alcoholic Fatty Liver Diseases), the emerging major healthcare concern [76]. In the bedside to bench mode, a rat model for NAFLD was established which showed by histological tissue studies of the liver that oral administration of *P.kurroa-Kutki* extract protected the rats from accumulation of fat and from the tissue damage induced by NAFLD. *P.kurroa-Kutki* was better when compared with control group and also compared with silymarin. The serum alkaline phosphatase (ALP) levels decreased faster in the *P.kurroa-Kutki* group than in the silymarin group [77]. Further to comprehend the molecular mechanism in a targeted condition of NAFLD, an *in-vitro* study was undertaken. This experimental work in *in-vitro* NAFLD model has shown that Picroside-II, the active ingredient in *P.kurroa-Kutki*, reduces fatty acid accumulation in HepG2 cells via modulation of fatty acid uptake and synthesis [78]. Subsequent recent *in-vitro* study has further shown that Picroside-II reduced not only lipid accumulation, but also oxidative stress and

Table 2
Summary of clinical trials of *Picrorhiza kurruoa* (alone) and *Picrorhiza kurroa* in combination with other agents in liver related and other disorders.

Formulation and Study Design	Dose	Duration and No of subjects	Adversity	Activity	Reference
Arogyavardhini &Punarnavadi (Open label)	500 mg x 3 & 30 ml x 2	3 weeks N = 24	No ADR	Acute Viral Hepatitis	[57]
Arogyavardhini (Double Blind)	750 mg x 3	2 weeks N = 20	No ADR	Acute Viral Hepatitis	[58]
Kutaki (Double Blind)	375 mg x 3	2 weeks N = 15	No ADR	Acute Viral Hepatitis	[15]
Kutaki + Methoxasalen (Oral & Topical)	200 mg x 2 20 mg once	3months N = 30	No ADR	Vitiligo	[59]
Kutaki with Sita (Two arms)	One gm x 2	3 weeks N = 30 (15 in each group)	No ADR	Amlapitta	[60]
Arogyavardhini &Triphala Guggulu and Pathya (Two arms)	250 mg x 2	3months N = 21	No ADR	NAFLD	[61]
Phalatrikadi Kwath (Open Label)	40 ml x 2	6months N = 59	No ADR	HbsAg + ve	[62]
Kutaki processed in Guduchi with Atorvastatin (Open Label)	Atovarstatin: 20 mg twice daily+2 gm Katukai (P. Kurroa) processed in Guduchi twice daily	3months N = 32	No ADR	Hepatoprotective	[63]
Elastographic liver evaluation of Katukyadi churna in the management of Non-Alcoholic Steatohepatitis (NASH) – A single arm clinical trial	6 gm (Sachets) twice a day with water. Katukyadichurna comprises 1 part each of Katuki + Nimba + Amrita + Bhringaraj + Bhumyamalki	180 days	11 patients participated in the study. Two patients suffered from loose stools 2–3 times/day for the first 8 days.	NASH	[64]

Table 3
Phytoconstituents of *Picrorhiza kurroa* and their hepatoprotective activity.

Sr.	Phytoconstituent	Chemical structure	Biological Activity	References
1	Picroside I		Hepato- protective, Antioxidant, Anti-inflammatory	[26,39,92]
2	Picroside II		Hepato- protective, Antioxidant	[39,65,92]
3	Kutkoside		Anti- inflammatory Hepato-protective	[26,93]
4	Apocynin		Neuro-protective, Anti- inflammatory	[94]

mitochondrial dysfunction in *in-vitro* NAFLD-Hep G_2cell model [79]. *P.kurroa-Kutki* product development through reverse pharmacology provide wide scope for developing standardized traditional formulation, traditional or non-traditional plant extract, bioactive fraction, single active principle, its analogues and derivatives. However, for a plant like *P. kurroa-Kutki*, pharmaceutical excellence, formal regulation, and ethical prescription is desired [Fig. 1].

4. Phytoactives of *P.kurroa-Kutki* for the indication of fatty liver disease

Picrorhiza species has been explored since 1940s for its chemical composition [11]. Since then, several phytocompounds have been isolated from different parts of this plant species which have shown the presence of glycosides, aromatic esters, bis-iridoid, phenyl propenoids, alcoholic compounds and fatty acids [80]. Iridoid glycosides viz. kutkins, kutkosides and picrosides, cucurbitacins, triterpenes and simple phenols such as apocynin are the most explored phytoactives of *P.kurroa* for their biological activity and potential clinical applicability. The active principles of *P.kurroa* are the iridoid glycosides picrosides I, II and III. Kutkin is a mixture of picrosides I and II. *P.kurroa* also contains Picroside IV and V, verminoside, catapol, veronicoside, specioside 6- feruloylcatapol, pikuroside, aucibin and many more. *P.kurroa* also contains other active constituents such as apocynin, drosin, nine cucurbitacin glycosides, D-mannitol, phenolic acids and phenylethanoids. As has been reported by Sah & Varsheya, 132 chemical constituents belonging to different class of compounds from roots, rhizomes, seeds, stem and leaves of two Picrorhiza species (*P. kurroa* Royle ex Benth and *Picrorhiza scrophulariiflora* Pennel) are listed between 1949 and 2013 [81]. In the last decade or so, 53 more phytoconstituents have been identified [80]. This reflects the ever-increasing enthusiasm and interest of biomedical scientists in *Picrorhiza species.*

However, Picroside-II is one of the iridoid glycosides from *P.kurroa* which has been extensively studied for its pharmacological effects in both *in vitro* and *in vivo* models. In a recent review, S Ma et al., 2020, have discussed in details the therapeutic potential of picroside-II for organic ischemia and reperfusion injury (to organs and tissues such as cerebrum, myocardium, kidney, testes, skeletal muscles and erythrocytes), liver damage, inflammation, cancer metastasis and osteoclastogenesis. Although the effects of picroside-II are multiple and complex, with the intricate

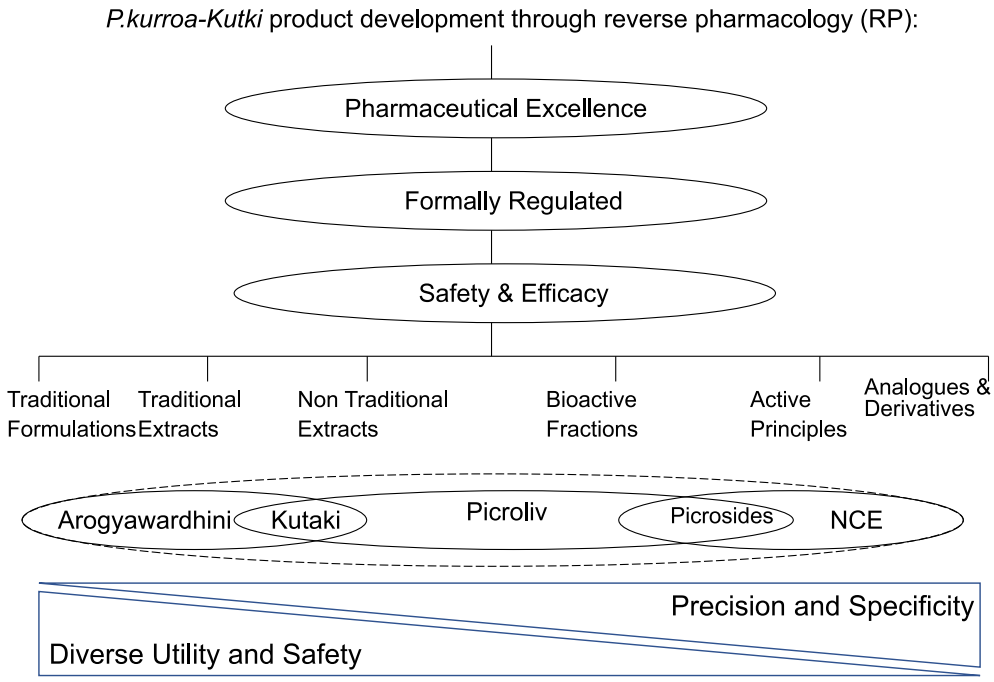


Fig. 1. Prerequisites being pharmaceutical excellence, formal regulation, and ethical prescription. RP provide wide scope for developing standardized traditional formulation, traditional or non-traditional plant extract, bioactive fraction, single active principle, its analogues and derivatives. However, the traditional formulations have diverse utility and better safety profile but the precision and specificity is relatively less. Whereas, actives and derivatives have more precision and specificity in the drug action but the safety and diversity are potentially compromised. Hence, the phytopharmaceutical product such as Picroliv is expected to strike this balance.

involvement of a number of pathways, the mechanisms of picroside-II appear to be mainly acting through anti-oxidant, anti-inflammatory and anti-apoptotic mechanisms [82].

Recently our group has reported results of *in-vitro* studies that Picroside-II effectively attenuated fatty acid accumulation in Hep G2cells pre-treated with Picroside-II. In this study Picroside-II pre-treatment inhibited FFAs-induced lipid accumulation by attenuating the expression of fatty acid transport protein 5, sterol regulatory element binding protein 1 and stearoyl CoA desaturase. It was also observed that pre-treatment with Picroside-II decreased the expression of forkhead box protein O1 and phosphoenolpyruvate carboxykinase. These findings suggest that Picroside-II effectively attenuated fatty acid accumulation by decreasing FFAs uptake and lipogenesis. Picroside-II also decreased the expression of gluconeogenic genes [79,80]. The molecular mechanisms of Picroside-II in fatty liver are depicted in Fig. 2 [83]. Interestingly Hua Han et al., recently have reported many synthesized derivatives from picroside-II and demonstrated the hepatoprotective activity of the derivatives evaluated on SMMC-7721 cells [84].

Bioavailability is an important pharmacokinetic parameter that is essential to be correlated with the clinical effect of most drugs. Some studies have been published of late regarding bioavailability of picrosides and its metabolites [85–89]. However, bioavailability of picrosides is variable or low, most probably due to primary metabolism through gut microbial flora and the hydrophilic nature of these iridoid glycosides [85]. However, information gained from Zahiruddin S et al. postulates that after oral administration of iridoid rich fraction the basic pharmacokinetic profiling of picroside I, II and apocynin as well as fate of other metabolites, demonstrates scientific basis of *P.kurroa-Kutki's* traditional use in *Ayurveda* [86]. Gao et al., have proposed four metabolic pathways for picroside-II viz. I) Picroside-II is de-glycosylated to generate the aglycone, which is isomerized to a dialdehyde-type intermediate. A series of metabolic reactions, including glucuronidation, subsequently occurs. II) Picroside-II is subjected to ester bond hydrolysis to form vanillic acid, which is further subjected to sulfate conjugation, glycine conjugation, glucuronidation, and demethylation. III) Picroside-II is directly conjugated with glucuronic acid to yield a predominant metabolite in plasma. IV) Picroside-II is directly conjugated with sulphate [87]. Interestingly, Zhu J et al. have demonstrated higher concentration of picroside-II in liver tissues after I.V. administration in Sprague Dawley rats [88]. This correlates well with the traditional clinical usage of *P.kurroa-Kutki* in hepatic disorders. Moreover, wide spectrum hepatoprotective activities of isolated chemical constituents and plant extracts of *P.kurroa-Kutki* such as anti-lipogenic [79,80], anti-inflammatory [25,26], anti-fibrotic [32], anti-oxidant [33,34], anti-hepatocellular carcinogenic [34], make this product applicable to the entire spectrum of alcoholic/non-alcoholic fatty liver disease [90].

5. Scope and challenges of *P.kurroa-Kutki* natural products/ phytopharmaceutical

Founded on insights from the *Ayurveda* knowledge-base and traditional clinical usage-experiences a novel natural product of *P.kurroa-Kutki* is possible while integrating current scientific and experimental database. Such a natural product is expected to have targeted efficacy, wide safety profile and desired reproducibility. The new natural product category of phytopharmaceutical has formally been approved in India [91].

It offers an opportunity to develop *P.kurroa-Kutki* phytopharmaceutical, standardized for phytoactives, in a targeted clinical condition of NAFLD. This phytopharmaceutical should identify picroside-II as a main phytoactive for the indication of the fatty liver disease besides minimum three other phytomarkers [92–94]. Table 3 summarizes the structure and biological activity of picroside-II and other three relevant phytomarkers suitable for NFALD. Although pre-clinical studies have demonstrated ample evidence; clinical studies with the *P.kurroa-Kutki*

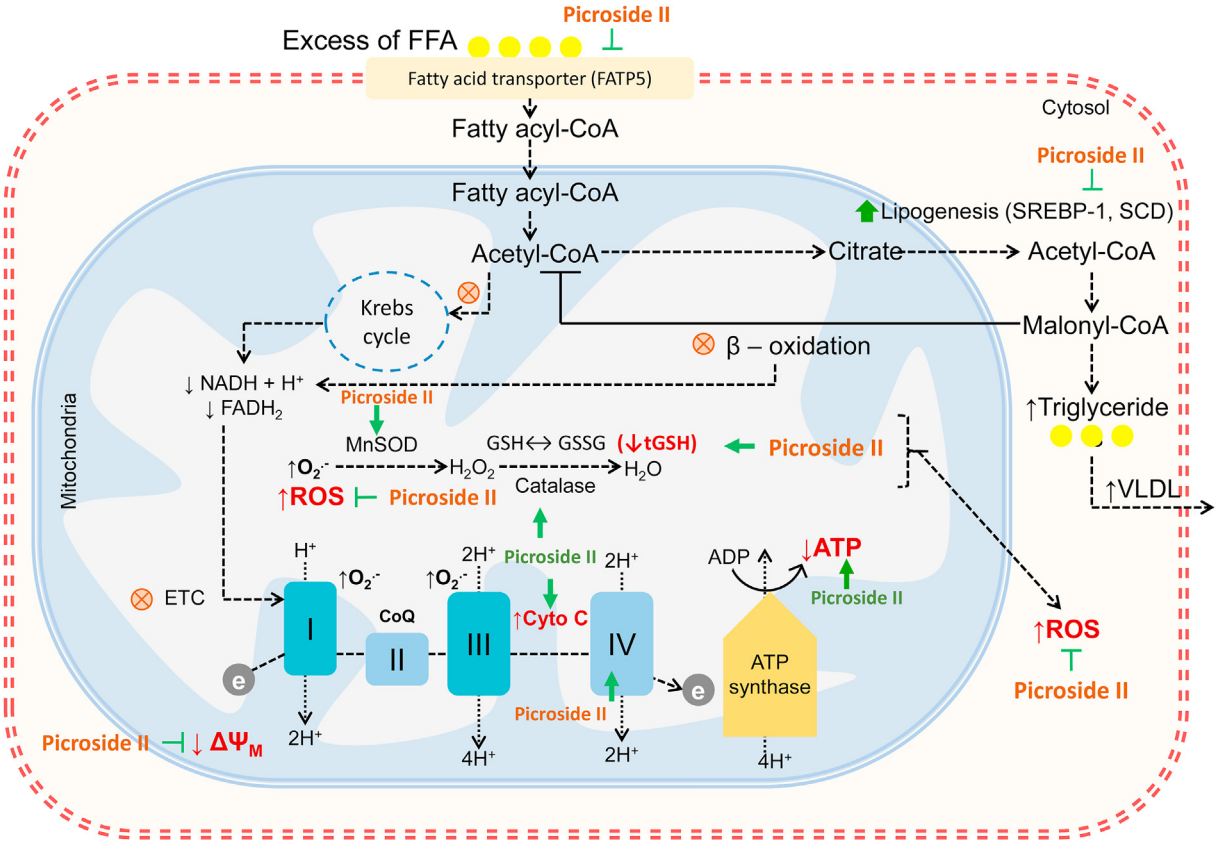


Fig. 2. Molecular mechanisms of Picroside II in fatty liver. Proposed mechanism of action of Picroside II on FFA accumulation, oxidative stress and mitochondrial dysfunction: Picroside II attenuated FFA accumulation in HepG2 cells by downregulation of FATP5, SREBP-1 and SCD, hence decreasing fatty acid uptake and lipid synthesis. Picroside II decreases the production of ROS, increases the levels of antioxidants (glutathione, Manganese superoxide dismutase and Catalase). Picroside II improves mitochondrial functions by increasing ATP production, decreasing ΔΨM loss and increasing cytochrome *c* expression. (The figure is adopted and modified further from; Dhami-Shah H, Vaidya R, Talwadekar M, Shaw E, Udipi S, Kolthur-Seetharam U, Vaidya ADB. Intervention by Picroside II on FFAs induced lipid accumulation and lipotoxicity in HepG2 cells. J Ayurveda Integr Med. 2021 Jul–Sep; 12(3):465–473′). FFA, Free Fatty Acid; FATP5, Fatty Acid Transporter Protein 5; SREBP-1, Sterol Regulatory Element-Binding Protein-1; SCD, Stearoyl CoA Desaturase; ROS, Reactive Oxygen Species; ATP, Adenosine Triphosphate; ΔΨM, Mitochondrial Membrane Potential.

phytopharmaceutical may face challenges of the bioavailability and biocompatibility of phytoactives.

The challenge of bioavailability of *P.kurroa-Kutki* phytoactives has been explored through the application of nanoencapsulation technique. Alcoholic extract of *P.kurroa-Kutki* was successfully encapsulated into pluronic-F-68-PLA nanoparticles by nano-precipitation method. Encapsulation efficiency for picrosides I and II was determined 60.17 ± 2.8% and 67.2 ± 7.4% respectively [95]. The authors stated that the release dynamics profile suits intestinal absorption and uptake in humans and could be a promising approach for enhancing intestinal absorption, biocompatibility as well as bioavailability, making it a suitable form of administering the alcoholic extract. However, limited studies on oral bioavailability indicate the need for further work along with exploring the potential for encapsulation and controlled release of the preparations, followed by appropriate studies for safety and toxicity of the nano-encapsulates.

Another important challenge is of drug interactions with the *P.kurroa-Kutki* phytopharmaceutical when co-administered with conventional chemical drugs. Under experimental conditions hydro alcoholic extract of *P. kurroa* rhizome has displayed β-cell regeneration with enhanced insulin production and antihyperglycemic effects in streptozotocin induced rat model and in insulin producing Rin5f cells [96]. Also, Husain et al., have reported that standardized *P.kurroa-Kutki* extract increased the insulin-mediated translocation of GLUT-4 from cytosol to plasma membrane which resulted in better glucose uptake by skeletal muscles and improved glycaemic control in diabetic rats [28]. This certainly warrants an attention for potential interaction of *P.kurroa-Kutki* products with anti-diabetic drugs. Hence, concomitant administration of other herbal products and chemical drugs demands attention from the drug interaction perspective. Although no known major reports of drug interactions with *P.kurroa-Kutki* specifically are reported in literature [97–99]. It is possible to record such a potential drug interaction in an organized clinical study. Not all interactions are harmful, and some can be beneficial, and some insignificant. However, it is worthwhile to have data from post marketing studies and pharmaco-epidemiological studies for such a phytopharmaceuticals as has been recommended recently for *Ayurvedic* products [100].

The wisdom of using traditional formulations containing *P.kurroa-Kutki* in combination with other herbs and herbo-minerals also should be explored for understanding the synergistic activity. The unique processes involved in these formulations' development needs to be comprehended to provide further insights through investigations. Although, *P.kurroa-Kutki* and its traditional products are sometimes known to lead to adverse effects such as increased bowel frequency, diarrhoea, abdominal gurgling, abdominal colic etc, vigilance about such side effects is desired during clinical trials of *P.kurroa-Kutki* products. Observations regarding compatibility and potentiation of *P.kurroa-Kutki*-products with certain food substances is also relevant. This should facilitate scheduling the time of oral administration of the *P.kurroa-Kutki* natural product.

6. Conclusion and prospects

Starting from bedside with the hint from observational therapeutics of classical *Ayurveda* treatment for hepatitis and further, well organized clinical studies, has given convincing clinical lead for the hepatoprotective potential of *P.kurroa-Kutki* standardized product. However, extensive phytochemicals identification, and isolation of phytoactives has given further boost to the experimental *in-vitro* and *in-vivo* studies of *P.kurroa-Kutki* plant molecules and products. Well organized, multicentric controlled-clinical trial, with a standardized phytopharmaceutical for phytoactives in fatty liver disease are likely to be undertaken soon. However, in the last couple of decades scientists have unravelled many more phytoactives from *P.kurroa-Kutki* with biological activities at different organ systems and molecular targets. These experimental bench-side findings encourage clinical scientists to go back to bedside and traditional clinical practices to get further hints and leads to undertake the clinical studies with *P.kurroa-Kutki* phytopharmaceuticals exploring clinical targets beyond hepatology.

Authors contribution

Ashwinikumar Raut: Conceptualization, writing original draft, writing – review & editing, visualization, supervision; Hiteshi Dhami-Shah: Writing – review & editing, visualization; Aashish Phadke: Writing – review & editing, visualization; Anand Shindikar: Writing – review & editing, visualization; Shobha Udipi: Writing – review & editing, supervision; Jayashree Joshi: Writing – review & editing, supervision; Rama Vaidya: Conceptualization, writing – review & editing, supervision; Ashok DB Vaidya: Conceptualization, writing – review & editing, supervision.

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Conflict of interest

The authors do not have conflict of interest. Although AAR is a member of the J-AIM Editorial Board, he was not involved in peer review process and editorial decisions related to this paper.

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