

A Transdermal Formulation of Icariin for Use as a Substitute for Sildenafil and a Showing of No Clinical Efficacy

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Abstract: *Icariin, considered to be the active compound in the herb Epimedium (also known as Horny Goat Weed) has been shown to be possibly erectogenic in a variety of animal studies, leading to its popularity as an herbal supplement for ED even though there are essentially no human studies supporting its effectiveness. Icariin is highly insoluble, and many studies have additionally reported that Icariin is not absorbed in the gut due to a very low bioavailability. The purpose of this study was to solubilize Icariin in the formulation of a transdermal cream which would deliver the compound directly to the blood stream bypassing poor intestinal absorption; and by informal testing of the transdermal cream on volunteer subjects, to determine if the erectogenic effects of icariin could be improved. This study demonstrates that icariin, delivered via the transdermal route, failed to demonstrate any efficacy of icariin for ED, conforming to another recent study reporting no effect upon oral administration.*

Keywords: Impotence, erectile dysfunction, Viagra, sildenafil, Epimedium, Icariin, Horny Goat Weed, transdermal herbal extract, transdermal icariin, human study

1. Introduction

We believe this to be the first report of the creation of a transdermal preparation of Icariin to solve the earlier observations of poor solubility and poor bioavailability known for oral administration.

Erectile dysfunction (ED), aka impotence, is the inability to obtain and maintain an erection sufficiently firm enough for sexual activity. Male sexual arousal is a complex process that involves the brain, hormones, emotions, nerves, muscles and blood vessels. Thus, ED may be due to a break in any of these entities, alone or in combination. Likewise, stress and emotional issues may cause or worsen erectile dysfunction. [1]

Sexual stimulation triggers release of neurotransmitters from the cavernous nerve terminals. The nerves to the penis are derived from the pudendal and cavernous nerves. The pudendal nerves derive from the sacral plexus (S2–4) and supplies somatic motor and sensory innervation to the penis. The cavernous nerves are a combination of parasympathetic and visceral afferent fibers and provide the nerve supply to the erectile tissue. [2]

This results in relaxation of these smooth muscles, leading to dilatation of the arterioles and arteries by increasing blood flow in both the diastolic and the systolic phases, trapping of the incoming blood by the expanding sinusoids and compression of the subtunic venular plexuses between the tunica albuginea and the peripheral sinusoids, reducing the venous outflow. Thereafter, stretching of the tunica to its capacity occludes the emissary veins between the inner circular and the outer longitudinal layers and further decreases the venous outflow to a minimum. Also, the understanding of the nitric oxide pathway has aided not only

in the molecular understanding of the tumescence but also aided greatly in the therapy of erectile dysfunction. [3]

A major breakthrough in ED management occurred with the development of Viagra (Sildenafil) and similar agents (Levitra, Cialis) in the same class. They increase blood flow to the penis so an erection can be gotten and kept hard enough for sex. Individual results vary. Effects usually begin within 30–60 minutes. The mechanism of action is the result of nitric oxide (NO) in the corpus cavernosum of the penis binding to guanylate cyclase receptors, which results in increased levels of cGMP, leading to vasodilation. Taking sildenafil with a nitrate type medications can cause a sudden and serious, life threatening decrease in blood pressure. [4]

This untoward side effect and other adverse reactions have motivated a search for a safer, gentler, more natural and equally effective remedy. To that purpose, attention has been drawn to Horny goat weed (Epimedium), which has been reported in the popular press to be a medicinal herb, containing the biochemical icariin, that people have traditionally used to treat many conditions including hay fever, osteoporosis, low libido and erectile dysfunction. [5]

As summarized by Christopher C K Ho and Hui Meng Tan in 2011: “There are about 50 Epimedium (Berberidaceae) species distributed in Asia, Europe, and the Middle and Far East. Among the species, *E. brevicornum*, *E. sagittatum*, *E. pubescens*, *E. koreanum*, and *E. wushanense* were recorded in Chinese Pharmacopoeia, while *E. acuminatum*, *E. myrianthum*, and *E. leptorrhizum* were recorded in Standards for Chinese Traditional Medicines and Ethnic Drugs in Guizhou. Epimedium herbs have been used to treat infertility and impotence for over 2000 years. Icariin, an amorphous yellow powder with a melting point between 231°C and 233°C, has been isolated from the aerial parts of

Epimedium herbs. There have been no studies yet on humans.” [6]

Sildenafil, commonly sold as Viagra and other brands, works primarily by inhibiting the phosphodiesterase enzymes, particularly Phosphodiesterase 5 (PDE5) thereby leading to strong erections. Icariin has been tested *in vitro* to exhibit PDE5 inhibition leading many to believe that this is its mechanism of action and supporting the idea that icariin may be an herbal substitute for sildenafil. [7]

How a PDE5 inhibitor works to create an erection was nicely summarized by David Rotella: “PDE5 is the primary cGMP-hydrolysing activity in human corpus-cavernosum tissue. Erection is largely a haemodynamic event, which is regulated by vascular tone and blood-flow balance in the penis. Because cGMP levels modulate vascular tone, it is an obvious target for therapeutic intervention in the process. When a man is sexually stimulated, either physically or psychologically, nitric oxide (NO) is released from non-cholinergic, non-adrenergic neurons in the penis, as well as from endothelial cells. NO diffuses into cells, where it activates soluble guanylyl cyclase, the enzyme that converts GTP to cGMP. The cyclic nucleotide then stimulates PKG, which initiates a protein phosphorylation cascade. This results in a decrease in intracellular levels of calcium ions, leading ultimately to dilation of the arteries that bring blood to the penis and compression of the spongy corpus cavernosum tissue. This compression contracts veins, which reduces the outflow of blood and increases intracavernosal pressure, resulting in an erection. A PDE5 inhibitor will retard enzymatic hydrolysis of cGMP in the human corpus cavernosum, leading to the same outcome.” [8]

The Icariin inhibition of PDE5 activity is much weaker than for sildenafil as shown by *in vitro* studies wherein PDE5 inhibition was 1/80th of that found with sildenafil. [7]

Based on widely exaggerated stories and urban myths that have arisen around Icariin within the last few years, Icariin has become a popular herbal supplement, available online at many herbal suppliers. It appears that many re-sellers are buying Icariin powder from Chinese sources and repackaging the powder for sale as is, or after loading into capsules. About 90% of all icariin supplements sold in the US and around the world are manufactured in China, and various studies of testing have shown that a substantial portion of herbal product in the herbal pipeline is fake, contaminated, mislabeled, or in fact adulterated with sildenafil to ensure a positive result. [9] One study found that the most common adulterant found in herbal products for sexual enhancement was sildenafil and that out of 81 samples tested, 28 contained the hidden ingredient. [10]

The fact is that the oral ingestion of icariin powder or capsules has no clinical efficacy whatsoever because of its insolubility. [15, 16] Our own studies have shown that Icariin (in any powdered form up to 98% purity) is not soluble in water, not soluble in any alcohol, not even soluble in the main stomach acid, hydrochloric acid. In addition to insolubility, many studies have confirmed that Icariin is not absorbed in the gut and therefore that oral ingestion yields a very low bioavailability. [13, 14]

So, in reality, with a very poor solubility compounded by almost no uptake in the gut, it is unlikely that the oral ingestion of icariin supplements has any effect whatsoever on improving erections in men, despite the ongoing fanfare and publicity surrounding its proposed use for this purpose.

In this paper, we investigated various wetting agents and report on the preparation of transdermal formulations for Icariin, in which the compound is first dissolved and then combined with cream or gel base for delivery of the compound directly to the blood. Transdermal drug delivery has been shown to bypass the gut and liver metabolism resulting in direct bioavailability in the blood. [15, 16]

Here we report the first formulation of a transdermal Icariin cream. After obtaining a transdermal preparation as described, we tested it on 12 elderly volunteers in an informal trial to determine any efficacy. This paper reports that 98% Icariin combined in a transdermal cream preparation at a concentration of 100 mg/ml, and two week daily dosage of 500 mg had no observed efficacy for erectile dysfunction, nor was the preparation erectogenic, calling into question the usefulness of this supplement as a substitute for sildenafil and related drugs.

Part 1 of the Study: Development of Transdermal Formulation

2. Materials and Methods

Horny Goat Weed Extract (98% Icariin) derived from Epimedium was obtained directly from the manufacturer, Kan Phytochemicals Pvt. Ltd., (1390, HSIIDC Industrial Estate Rai, Rai District, Sonapat 131029, Haryana, India), an herbal extract manufacturer operating in accordance with GMP and norms of WHO and USFDA guidelines, with both ISO 9001 and GMP certifications.

Proprietary cream and gel bases manufactured by PCCA (9901 South Wilcrest Drive, Houston, TX 77099, USA), namely Lipoderm and Atrevis Hydrogel, as well as all other standard compounding reagents were supplied by Pavilion Compounding Pharmacy LLC., 3200 Downwood Cir NW Suite 210, Atlanta, GA 30327, USA:

Tween-80 (polysorbate 80)
PEG 300 (polyethylene glycol 300)
Saline (Normal saline for irrigation, 0.9% NaCl)
Aloe gel (100%) pharmaceutical grade

Three solutions to be used as solubility reagents were created: (1) Tween 80/PEG 300 50:50, was a solution created by mixing equal parts of Tween-80 and PEG 300; (2) the second solution was created by mixing the following percentages: 10% DMSO, 40% PEG 300, 5% Tween-80, and 45% Saline; (3) The DMSO/Saline solution was created using equal parts of DMSO and normal Saline. For each trial, 2.5 gm of Icariin powder was added first to a mixing jar. Then, 5ml of one of the wetting agent solutions was added slowly and mixed to create a solution, or in some cases only a paste. Finally, a cream or gel base was added for a total of 25 gm to yield a 10% formulation (100mg

Icariin/ml). In the case of a 5% formulation, 1.25 gm of Icariin was used, thereby resulting in a 5% formulation (50mg Icariin/ml). All mixing and handling of reagents occurred at room temperature, 20 °C.

3. Results

We confirm the lack of solubility of Icariin in H₂O, ethanol, methanol, poor solubility in ethanol and polyethylene glycol.

Table 1 presents the results of our study on solubilizing Icariin powder using various wetting agents to achieve a transdermal cream or gel.

Wetting Agent	10% Icariin Concentration	5% Icariin Concentration	Mixed with Lipoderm	Mixed with Hydrogel
Tween-80/PEG 300 50:50:00	Not Soluble. Dark brown suspension containing particles and clumps	Slightly soluble. Dark brown color after 2 hr mixing; particles remain in suspension after 24 hr mixing.	10% concentration yielded a gritty dark brown cream, which after further run through an ointment mill, did yield a smooth and uniform cream. 5% concentration yielded a smooth and uniform cream light brown in color.	The solutions became fully solubilized in the gel, yielding a smooth and uniform gel, brown in color.
10% DMSO/40% PEG/5% Tween	Slightly soluble dark brown solution with particles remaining in suspension after 2 hours of mixing followed by 24hr standing at room temperature	Slightly soluble dark brown solution with particles remaining in suspension after 2 hours of mixing followed by 24hr standing at room temperature	Soluble at both 5% and 10% yielding a smooth uniform cream light brown in color.	Soluble at both 5% and 10% yielding a smooth uniform cream brown in color.
DMSO/Saline 50:50	Instantaneous solubility, best of all wetting agents used, light brown in color	Great solubility, translucent and light brown in color	Soluble at both 5% and 10% yielding an elegant uniform cream, light brown in color	Soluble at both 10% and 5% yielding a smooth and uniform gel brown in color
DMSO/Saline 50:50 mixed with Aloe gel rather than with the PCCA products Lipoderm or Hydrogel.	Smooth and uniform 10% Icariin gel, light brown in color	Smooth and uniform 5% Icariin gel, light brown in color	N/A	N/A
No wetting agent, Icariin powder was mixed directly with either PCCA Lipoderm or Atrevis Hydrogel	N/A	N/A	At both 5% and 10% concentrations of Icariin, a gritty cream suspension, dark brown in color, remained gritty after further run through ointment mill.	At both 5% and 10% concentrations of Icariin, a gritty gel suspension, dark brown in color, remained gritty after further run through ointment mill.

4. Results

The pharmaceutical bases PCCA Lipoderm and Atrevis Hydrogel are standard bases used in the US by compounding pharmacies for the mixture of various drugs and extracts in powdered form to yield transdermal preparations for patients. In many cases, a drug or extract may be mixed directly with the cream or gel base in that the pharmaceutical dissolves directly. In the case of Icariin, which is highly insoluble, direct mixing with either Lipoderm or Hydrogel was ineffective in yielding a good transdermal agent, the result yielding a gritty product with undissolved particles of extract.

For two of the wetting agent solutions, namely Tween-80/PEG 300, and the DMSO/PEG/Tween/Saline solutions, although the icariin was not fully solubilized, it became so for the 5% Icariin concentration when Lipoderm or Hydrogel finished off the mixture, demonstrating that both Lipoderm and Hydrogel have certain solubility effects on their own.

The best solubility mixture, yielding instantaneous results was a 50:50 mixture of DMSO/Saline. Topping off with either Lipoderm or Hydrogel yielded very excellent preparations. And topping off even with just aloe vera gel

yielded a very nice transdermal preparation. Although our focus was to end with a 10% transdermal cream (100mg Icariin/ml), we believe that even higher concentrations can be achieved using the 50:50 DMSO/Saline.

As lipoderm and hydrogel may not be readily available, we conclude that dissolving the Icariin in the DMSO/Saline wetting agent followed by the addition of any good quality penetrating cream or aloe vera gel make the most convenient transdermal formulation. Mixing in DMSO/Saline alone, being watery, makes for a difficult solution to handle.

Part 2 of the Study: Informal Trial of Transdermal Icariin

Study Population

N = 12 adult males (Ages: 55, 62, 62, 63, 63, 65, 65, 66, 67, 68, 68, 73) were recruited at an apartment complex in Wilmington, NC during July 2019 and completed the 14-day study. Although 17 in toto were recruited, only 12 completed the informal trial. A Interview Questionnaire was used to select participants who (1) were in generally good health; (2) who experienced erection difficulties from mild to severe and/or stated they "could use some help"; (3) had tried sildenafil or similar drugs previously and had successful outcomes; (4) who had discontinued sildenafil

due to cost and/or perceived dangerous side effects. The purpose of requesting information about past sildenafil was to ensure that the biochemical pathway for phosphodiesterase inhibition was available. The participants were not known previously to the researcher nor to each other but were located around the recreational and pool areas of the apartment complex.

Preparation of Icariin Transdermal Lotion

Following our observation that 50:50 DMSO/Saline was the best wetting agent resulting in an instantaneous solution, we selected at random a readily available penetrating cream manufactured by Clinique Laboratories (New York, NY), namely "Clinique Moisture Surge 72-hour replenishing hydration." Any good quality penetrating cream will serve to create an easily handled preparation for application to the skin. Commonly available plastic syringes were used both for the transdermal preparation as well as for dosing. A batch of transdermal cream was made every 4 days as follows: To 25 gm of Icariin (98%, Kan Phytochemicals Pvt. Ltd) was added 50:50 DMSO/Saline to 50 ml total. After solubilization, an additional DMSO/Saline mixture was added to 100 ml total, creating a 25 gm/100 ml preparation. To this was added to a sufficient quantity of Clinique cream to make 250 total ml preparation, thereby yielding 25 gm Icariin/250 ml cream preparation, or 100 mg Icariin/ml cream. 5 ml of preparation (500 mg Icariin) was applied daily to clean skin under forearm, using both forearms as necessary. The transdermal cream preparation was absorbed readily, and after 5 minutes, there was virtually no telltale sign on the skin that the cream was applied.

Treatment Procedure

In order to ensure compliance with daily application, volunteers would meet individually at a private area of the recreation area and 500 mg icariin formulated as described was administered trans dermally and rubbed into the skin until fully absorbed within 5 minutes. Participants were advised not to shower for 2 hours and to return the next day. The study continued for 14 days, July 14-July 28, 2019. After that time, the participants were interviewed as to any results obtained.

Results

No participant observed any positive result from the transdermal icariin at 500 mg per day. No participant observed any change in or benefit in erections, erection strength, erection duration, or in size of tumescence commonly observed with sildenafil or its analogs. While it was not necessarily expected that the effects of transdermal icariin would be as dramatic as that found with sildenafil, it was disappointing to demonstrate that icariin had no observable effect whatsoever in this study.

5. Discussion

To our knowledge, this is the first report of attempts to create a transdermal preparation for Icariin which will deliver the herbal extract directly to the blood stream, bypassing both the gut and liver metabolism. We had postulated that the poor solubility as well as poor bioavailability of icariin due to poor absorption in the gut were the reasons why, though highly touted as a sildenafil

substitute, the herbal extract has never achieved any consensus as to its efficacy.

We found Icariin extract 98% easily soluble only in 50:50 DMSO/Saline, requiring this mixture to serve as a wetting agent prior to admixing a penetrating cream for ease of use. Furthermore, almost any good quality cosmetic cream can be used, whether these be formulated by a cosmetic cream manufacturer or off the shelf cosmetic cream. The purpose of adding any cream to the DMSO/saline solution containing icariin is to make the compound easier to handle, use, and administer to the skin by adding bulk to the final preparation.

We further found that Icariin 98%, administered transdermally at a dosage of 500 mg, had no effect whatsoever on erectile problems in our study. Not even one of 12 volunteer subjects noted any erectogenic effect, although all had tried sildenafil in the past with positive results. We find these results disappointing, as it was postulated that the transdermal route of administration, by delivering the compound directly to the blood, would bypass the solubility and low bioavailability issues, causing a beneficial erectogenic effect. This was not the case.

The most cited paper on the subject of icariin serving as a possible herbal substitute is the paper by Dell'Agli et al. "Potent Inhibition of Human Phosphodiesterase-5 by Icariin Derivatives." [7] The report states: "Compound 1 [icariin] was a good PDE5 inhibitor (IC₅₀ of 5.9 μ M) but required improvement in order to have equivalent potency to sildenafil, which gave an IC₅₀ of 75 nM."

While the paper can be commended for the head to head comparison of icariin analogs to sildenafil in an enzyme inhibition assay, the bottom line is that the data is based solely on inhibition rates on a phosphodiesterase-5 (PDE-5) enzyme inhibition assay, *in vitro*, far away from actual *in vivo* activity arising from oral ingestion of an herbal extract. Moreover, what the Dell'Agli paper in fact showed was that the activity of icariin was 1/80th that of sildenafil in the test-tube enzyme assay. ONLY a synthesized analog of icariin, known as 3, 7-bis(2-hydroxyethyl) icaritin was comparable in activity to sildenafil in PDE-5 enzyme inhibition assay.

Our study herein showing no efficacy of transdermal icariin, is supported by a recent study looking at the pharmacokinetics of orally administered Icariin.[16] In the 2019 Brown et al study, the authors noted: "Human data on icariin are limited," and proposed a study "to determine the pharmacokinetic profile of oral icariin in humans using serial blood draws over a 24 hours following a single dose of the drug." Five daily dose levels were tested: 100, 200, 400, 840, 1680 mg icariin. Plasma testing resulted in no finding of any icariin. The authors stated: "...the analysis of plasma samples revealed a low bioavailability of icariin, such that the pharmacokinetic profile of the drug could not be determined."

Even over a 5-day course of oral supplementation, no change in erectogenic activity was noted: "There was no statistically significant differences between placebo and icariin groups on the Arizona Sexual Experience Scale"

which asks about penis erection, arousal, sexual desire and orgasm. This study adds further support to our conclusion that icariin has no efficacy against ED in humans.

Finally, despite all the hype involved in promoting icariin as a substitute for sildenafil, a top research scientist in the Chinese herbal extract industry, Dr. Annette Wang, reported to us in a terse statement concerning our inquiry about solubility, that "Icariin has no clinical efficacy" (Annette Wang, PhD, Sinoway Industrial Co. Ltd, personal communication, July 18, 2018). Based on all the above, we question previous news articles and anecdotal reports drawing conclusions from convoluted animal studies, mostly *in vitro*, purporting to claim any erectogenic effect of icariin in humans.

We have come to question previous notions that icariin may be a useful substitute for Sildenafil in treating Erectile Dysfunction. We believe that while the herb, Epimedium, may well have mild erectogenic effects, icariin is not the active ingredient. We postulate that icariin, as the most abundant compound extracted from Epimedium, was presumed to be the active ingredient, but is not in fact so. We conclude that there is no basis whatsoever to believe that icariin taken orally, even in mega doses, has any beneficial effect on erectile dysfunction. Moreover, our own small study delivering icariin via the transdermal route also failed to demonstrate any efficacy of icariin for ED.

This paper is important in defining a procedure for composing a transdermal formulation for icariin. Additionally, this preliminary clinical study, delivering icariin via the transdermal route, and demonstrating no erectogenic effect, confirms a recent study in humans that megadoses of the compound taken orally lack any erectogenic effect as well.

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