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Quick Dissociation and Selective Affinity of Actives from Akshun Results in Rapid Pain Relief

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Abstract: ***In the present study we have reverse interpreted the scope of zein protein hydrolysis and collagen swell test. Akshun is quick remedy for various types of pain such as cervical spondylitis, low back pain and pain in the joints and further the product is presented in the form of bathing lotion. The surfactant base is expected to cause small corrosion in the stratum corneum thereby the actives may enter the system. We believed the above possibility because the relief from Akshun is instantaneous however its contact time is incredibly short. Akshun has not hydrolyzed Zein protein and collagen therefore it can be presumed that the base of Akshun may not corrode stratum corneum cells. The selective bindings may be due to special affinity is believed to be the reason for faster therapeutic benefit of Akshun with least contact time. Details are presented in the paper.***
Keywords: Akshun, bathing lotion, rapid pain relief, permeation

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Introduction

A complete, sustained and instant solution for pain of multi-various origin is still far from reach for the sufferer. Whatever be the underlying causes, the pain whether we describe it as a problem or as a symptom, poses great agony to the sufferer.

Chronic pain due to auto-immune causes like arthritis, cancer etc., requires lifelong treatment and palliative care. Most of the analgesic and anti-inflammatory preparations are bound to gift serious side effects in long term use. Therefore palliative/supportive products that might offer moderate relief from pain without any side effect is necessary. [1, 2, 3]

Several topical analgesic and anti-inflammatory preparations are available however most of such products involve cumbersome usage procedure (massage oil/ointments) or are strongly pungent like diclofenac and many other Ayush products. [4, 5, 6 7, 8]

Akshun is a product of great innovation and unconventional science where science and tradition has been blended in equal proportion to offer instantaneous pain relief to the sufferer and at the same time the procedure involved in the usage of Akshun is quite simple.

Akshun is formulated with the following herbs such as *Clerodendron phlomoides, Cardiospermum helicacabum, Cinnamamonum camphora* and *Mentha piperata* in bathing lotion base.

The clinical trial findings have shown that Akshun is quite effective and quick to relieve pain in those who suffer from cervical spondylitis, low back pain and pain in the joints. [9]

In the present paper we have explained the possible reason for such an effect is due to competitive absorption of the actives from the medicinal herbs used in the formulation and such competitive absorption is not due to micro- abrasion of the skin or dermal corrosion as a result of the base ingredients (detergents) used in the formulation base of Akshun. The above findings really make the Akshun a unique and an extraordinary formulation for quick pain relief. Details are presented in the paper.

Materials and Methods

Principle

Akshun formulation was subjected to the following tests such as zein protein hydrolysis and collagen swell test in order to understand whether the formulation in toto or any of the actives hydrolyze zein protein and collagen.

Zein protein is one of the softest fine proteins used widely for testing the primary irritant property of several personal care and toiletry preparations. The extent of hydrolysis of zein protein is directly proportional to the irritant or harshness of the product.

Similarly collagen is the major skin protein and is highly permeable to damage due to harsh chemicals and detergents. When the damage implicated on collagen increases the rate of absorption of water do increase and as a result the collagen would swell.

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Zein protein hydrolysis by measuring the residue through filtration

In brief the sample (Akshun bathing lotion for pain relief) was diluted with water to achieve different concentrations viz., 1%, 5 %, 10 % and 15 %. Similarly a positive control containing 0.5 % SLS in water was prepared. Then the Whatman filter paper No.1 was weighed and initial weight was noted. One gram of Zein protein was weighed accurately and was added separately to all the above solutions including positive control. Then the above mixture was allowed to remain for 24 hours. After 24 hour incubation the above mixture were filtered separately through the pre-weighed filter paper. After complete filtration, the filter papers were collected, marked and then dried overnight at 45°C. The rate of Zein protein hydrolysis was calculated using the formulae

Total Zein (g) added to the solution - Total Zein after drying x 100

Total Zein (g) added to the solution

The rate of hydrolysis was compared between test and positive control

Zein protein hydrolysis by measuring the residue by centrifugation

In brief the sample was diluted with water to achieve different concentrations viz., 1%, 5 %, 10 % and 15 %. Similarly a positive control containing 0.5 % SLS in water was prepared. Then the Whatman filter paper No.1 was weighed and initial weight was noted. One gram of Zein protein was weighed accurately and was added separately to all the above solutions including positive control. Then the above mixture was allowed to remain for 24 hours. After 24 hour incubation the above mixture was centrifuged using pre weighed centrifuged tube at 1500 rpm for 15 minutes.[10] Then the weight of unhydrolyzed zein protein hydrolysis was calculated using the formulae

Total Zein (g) added to the solution - Total Zein after centrifugation x 100

Total Zein (g) added to the solution

The rate of hydrolysis was compared between test and positive control

Zein protein hydrolysis by determination of OD value by spectrophotometer

In brief the sample was diluted with water to achieve different concentrations viz., 1%, 5 %, 10 % and 15 %. Similarly a positive control containing 0.5 % SLS in water was prepared. Then the Whatman filter paper No.1 was weighed and initial weight was noted. One gram of Zein protein was weighed accurately and was added separately to all the above solutions including positive control. Then the above mixture was allowed to remain for 24 hours. After 24 hour incubation the above mixture was centrifuged at 1500 rpm for 15 minutes. Then the supernatant was collected and read using a spectrophotometer at 244 nm. The OD Value of the test was compared with positive control.

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Collagen swell test

A piece of pre- weighed collagen sheet (1 cm^2) was incubated in various conc. of test solutios (1, 5, 10 and 15 %) vis-à-vis positive control and then incubated for 24 hours, at 50˚C. after 24 hour incubation the collagen sheet was collected separately from each test sample and was weighed carefully.[10] The results are expressed as follows:

CST = (weight after incubation – initial weight)/initial weight

Results

Akshun bathing lotion for pain relief up to 15% does not hydrolyze zein protein significantly when compared to sodium lauryl sulfate. Table- 1 , 2 & 3

a) Zein protein hydrolysis by measuring the residue through filtration

S.No	Concentration	% un-hydrolysed zein	% zein hydrolysis
1	1 % Akshun	95 %	5 %
2	5 % Akshun	85 %	15 %
3	10 % Akshun	80 %	20 %
4	15 % Akshun	65 %	35 %
5	Control (SLS)	8%	92%

Zein protein hydrolysis by measuring the residue by centrifugation

S.NO	Concentration	% un-hydrolysed zein	% zein hydrolysis
1	1 % Akshun	98 %	2
2	5 % Akshun	90 %	10
3	10 % Akshun	84 %	16
4	15 % Akshun	79 %	21
5	Control (SLS)	5%	95

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Zein protein hydrolysis by Determination of OD value by spectrophotometer

S.NO	Concentration	OD Value
1	1 % Akshun + Zein	0.01
2	5 % Akshun + Zein	0.05
3	10 % Akshun + Zein	0.12
4	15 % Akshun + Zein	0.20
5	Control (SLS)	0.35

Up to 15% Akshun did not induce significant damage to collagen as the absorption rate of water by collagen was quite abysmal when compared to SLS. table- 4

COLLAGEN SWELL TEST

S.NO	Concentration	% Collagen Swelling
1	1 % Akshun	10 %
2	5 % Akshun	25 %
3	10 % Akshun	30 %
4	15 % Akshun	32%
5	Control (SLS)	95 %

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Discussion

Our present study has revealed a new possibility of dermal absorption of various herbal actives. When we formulated Akshun with various select herbal extracts such as *Clerodendron phlomoides*, *Cardiospermum helicacabum*, *Cinnamamonum camphora* and *Mentha piperata* in bathing bar lotion base constituted by several surfactants, we initially doubted the stability of the above herbal extracts in the finished formulation. Further we also presume that the mild damage that may be caused by the surfactants might aid the absorption of the actives into the skin and thereby offers instantaneous therapeutic benefit.

Although such an effect may be appreciable only from the point of view of pain relief but the possible derml damage due to prolonged usage cannot be ignored while achieving relief from pain. In order to exclude such possibility we have undertaken the present study. We have used to two key candidate proteins such as Zein and Collagen for the study. Both these proteins represent great value to skin as the product that does not hydrolyze either of these proteins has to be safe to skin during prolonged usage.

When we subjected Akshun to zein protein hydrolysis and collagen swell test, we found that Akshun did not hydrolyze either of the above proteins significantly when compared to SLS. The above findings suggest that none of the surfactants used in Akshun are likely to cause any dermal damage.

The herbal actives due to their micronized state and special affinity to skin may be permeating into the skin and thereby brings down the pain quickly. The safe delivery of actives into skin must be seen also from the perspective of efficacy and safety in equal measure.

The competitive absorption of herbal actives through the skin may be the reason for the faster therapeutic effect. Akshun is formulated as bathing lotion and hence its residency time over skin is very short. Under such circumstances competitive dermal absorption of the actives into the skin is must when the formulation is intended to offer pain relief. Up to 15% Akshun did not hydrolyze zein and collagen protein. This supports the safe and non-irritant nature of the product. Perhaps this may be the first product for pain relief formulated in the form of bathing lotion where the contact time of the product to the skin is very short, however the competitive absorption of the actives enables the rapid permeation in to the skin and thereby offers quick relief.

Our present study has not only establishes the safety of Akshun through zein and collagen hydrolysis assay but also the possible mechanism of action responsible for therapeutic benefit that is competitive permeation.

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