



ANTICANCER ACTIVITY OF UPAVISHA SNUHI: A COMPREHENSIVE UPDATE

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DOI: 10.7897/2277-4572.096190

Received on: 11/10/20 Revised on: 25/11/20 Accepted on: 29/11/20

ABSTRACT

Cancer is one of the dreaded diseases of 20th century responsible for causing most fatalities and spreading further with increasing incidence in 21st century. Ayurveda an ancient science provides many useful remedies for these types of advanced diseases. Upavishas narrated in Agadtantra contribute for many fruitful therapeutic formulations. Snuhi (*Euphorbia nerifolia* Linn.) is one among these upavishas is knocking out as an effective Anticancer agent. Traditionally it is mainly used in Kushtha, Udara, Shotha, Pandu, Gulma, Dushivisha, Visha chikitsa. Various In Vitro & In Vivo studies has been conducted to evaluate the anticancer activity of Upavisha Snuhi (*Euphorbia nerifolia* Linn.) in the form of its extracts. In this study a special emphasis is on gathering details of Upavisha Snuhi from available classical text and assemble data related to the In Vivo and In Vitro Anticancer activity of Upavisha Snuhi.

Keywords: Agadtantra, Upavisha, Snuhi (*Euphorbia nerifolia* Linn.), Anticancer activity, In Vitro, In Vivo.

INTRODUCTION

Therapeutic use of medicinal plant is as old as humankind. There are about 45,000 medicinal plant species in India. Samhitas, Vedas and holy books has mentioned effective therapeutic use of various medicinal plants. Snuhi is an important medicinal plant included in Upavisha by Rasatarangini.¹ It is botanically identified as *Euphorbia nerifolia* Linn. It grows widely around the dry, rocky, & hilly areas of north, central and south India. Snuhi has been attributed with a number of synonyms depending on its morphology and pharmacological actions. Almost all the parts of Snuhi are of medicinal use. About 462 formulations, having Snuhi as an ingredient are used to combat almost 62 varied diseases.² Acharya Charaka has given its twenty Virechana formulations in Sudha kalpa Adhyaya of Kalpasthana.³ Traditionally it is mainly used in Kushtha, Udara, Shotha, Pandu, Gulma, Dushivisha, Visha chikitsa. Snuhi (*Euphorbia nerifolia* Linn.) extracts and isolates have been reported for its analgesic, anesthetic, anti-anxiety, anti-convulsant, anti-psychotic, anti-arthritis, anti-diabetic, anti-inflammatory, anti-diarrheal, anti-microbial, antioxidant, anti-ulcer, diuretic, hemolytic, immunomodulatory, pesticidal effect, radioprotective, wound healing property, cytotoxic and anticarcinogenic properties using various In Vitro and In Vivo methods.⁴

Cancer in the broader sense refers to more than 277 different types of cancer disease indicating several gene mutations which lead to abnormal cell proliferation.⁵ Cancer is the second leading cause of death globally.⁶ Chemotherapy and surgery have always been the standard methods of treatment but has not been fully effective. Chemotherapeutic agents are responsible for causing severe adverse effects.⁷ As medicinal herbs are known for less side effect; they are moving from border line to mainstream to treat cancer. Nowadays researches are mainly focused to develop new methods for cancer treatment predominantly using different

plant species. Likewise, Snuhi (*Euphorbia nerifolia* Linn.) has been studied for various ethnomedicinal properties including anticancer. But no effort is made to formulate a single-handed document of In Vitro and In Vivo studies done with anticancer activity of Snuhi. Hence present review is mainly concerned with the documentation of In Vivo and In Vitro anticancer activity of Upavisha Snuhi.

LITERATURE REVIEW

Upavisha Snuhi ⁸⁻¹⁵

Snuhi (*Euphorbia nerifolia* Linn.) is commonly known as Indian Spurge tree, it is characterized by presence of latex which exudes when broken and is regarded as toxic part of the plant. It is found throughout the Deccan Peninsula of India.

Botanical Name - *Euphorbia nerifolia* Linn.

Family - Euphorbiaceae

Description – It is a Xerophytic, erect, prickly, succulent, large, much branched shrub, which grows up to 2-6 meters.

Classification -

- i. Ayurveda – Sthavara Upavisha
- ii. Modern Toxicology – Organic irritant poison

Common Names

Sanskrit - Vajri, Snuhi, Sudha, Samantadugdha, Snuk, Sehunda

- i. Hindi - Sehunda, Thohar
- ii. Marathi - Nivdunga
- iii. English - Common Milk Hedge

Gana -

- i. Charaka - Virechana, Shatashodhanavruksha
- ii. Sushruta - Adhobhagahara, Shyamadi

Rasa Panchak -

- a. Guna - Laghu, Tikshna, Guru

- b. Rasa - Katu
- c. Veerya - Ushna
- d. Vipaka - Katu
- e. Doshaghna - Kaphavataghna
- f. Karma - Vishaghna

Bahya - Lekhana, Vedanasthapan
 Abhyantar - Raktashodhak, Shothahara, Twakdosahara,
 Tikshnavirechak, Kaphanissarak

Rogagnata - Gulma, Udararoga, Yakrutplihavruddhi, Shotha,
 Kushtha, Vatarakta, Upadanhsa, Kasa, Shwas, Pratishtay,
 Dushivisha, Visha

Prayojyanga - Moola, Kanda, Patra, Kshira

Actions and Uses – Plant is laxative, carminative, alexipharmic,
 appetizing, useful in abdominal troubles, bronchitis, tumors,
 ulcers.

Juice is useful in – Glandular swellings

Leaves are useful in – Tumors, inflammation, abdominal
 swelling,

Types - 1) Tridhara (*Euphorbia antiquorum* Linn.)

2) Saptadhara (*Euphorbia royleana* Boiss.)

3) Chimiya (*Euphorbia tirucalli* Linn.)

According to Charak Samhita – Alpakantaka, Bahukantaka

Fatal Dose - Uncertain

Fatal Period - Uncertain

Formulations - Citrakadi taila, Abhaya lavana, Avittoladi
 bhasma, Vajraksara.

Table 1: Chemical Constituents^{16,17}

Powdered plant, stem and leaves	Several triterpenoids like Glut-5-en-3 β -ol, Glut 5(10)-en1-one, taraxerol and β -amyron
Latex	Triterpene – nerifoliene, euphol, nerifoliol, nerifolene, euphorbon, resin, gum, caoutchouc, malate of calcium, euphol, monohydroxy triterpene, nerifoliol, taraxerol, beta- amyron, glut-5-(10)-en-1-one, nerifolione, cycloartenol
Leaf	Friedelan-3, D: B- friedolan-5-(10)-en-1-one, taraxerol
Bark	Euphol, Euphorbol, hexacosanoate, n- hexacosanol, 12- deoxy 4- β - hydroxyphorbol-13-dodecanoate-20- acetate, pelargonin – 3, 5- diglucoside, 24-methylene cycloartenol, tulipinin-3, 5- diglucoside
Stem	Euphol, friedelan-3, D: B friedoolan-5(10)-en-1-one, glut-5(10)-en-1-one, taraxerol
Root	Alnus-5(10)-ene-1-one, anthocyanins, euphol, pururate dikinase, terpenes, 24-methylene cycloartenol, tulipinin-3, 5- diglucoside
Ethanollic extract of fresh root	Antiquorin

Anticancer Activity of Upavisha Snuhi (*Euphorbia neriifolia* Linn.) – Various research articles on Snuhi (*Euphorbia neriifolia* Linn.) were studied and data related anticancer activity of Snuhi (*Euphorbia neriifolia* Linn.) was extracted. Both In Vivo and In

Vitro models are included. Data regarding plant part used, phytoconstituent studied and extractive solvent used is also collected.

Table 2: In Vitro Anticancer Activity of Snuhi (*Euphorbia neriifolia* Linn.)

Sr. No.	Plant Part Used	Extractive Solvent	Phytoconstituent Studied	Study Design	Assessment	Results
1.	Dried Powder of Leaf ¹⁸	Ethanol	Sapogenin	Murine F ₁ B16 Melanoma cell line	Cytotoxicity Assay	cell viability – At 10 μ g/ml-76.6% At 500 μ g/ml – 13.6%
2.	Whole Plant ¹⁹	Methanol	-	B16F10 Melanoma	SRB Assay MTT Assay	IC ₅₀ by – SRB Assay – 198.26 MTT Assay – 212.78
3.	Leaves and Bark - Dried Powder ²⁰	Methanol	-	HepG2 cell line	MTT Assay	Cytotoxicity for HepG2 cell line- 89.25%
4.	Whole Plant ²¹	-	MacKay-03 (3,12-o-diacetyl-7-o-angeloyl-8-methoxyingol)	K562 HEL cells	MTT Assay	Inhibited growth of Human leukemic cells
5.	Dried Powder of Leaves ²²	MeOH	Neriifolin 1 Neriifolin 2 Neriifolin 3	HCT116 cell line MCF7 cell line MDA-MB-231 cell line	MTT Assay	Compound 1-3 showed cytotoxicity to MCF 7 cell line
6.	Arial parts ²³	Ethyl Acetate	Four Triterpenoids a) 3 beta -friedelinol b) 3 alpha- taraxerol c) 3 beta -taraxerol d) 3 alpha-friedelinol. Four lingols – a) 3,7,12 -o-triacetyl-8-o- tigloylingol. b) 3,7,12 -o-triacetyl-8-o- benzoylingol. c) 3,12-o-diacetyl-7-o-angeloyl-8- methoxyingol. d) 3,7,12-o-diacetyl-8- methoxy-7-o- benzoylingol.	K562 cell line Panc-1 cell line 81T cell line BE3 cell line	MTT Assay	3 beta -friedelinol 3 beta -taraxerol and 3 alpha- friedelinol cytotoxic on -Panc-1 cell line 81T cell line BE3 cell line Inhibition 60% at conc. 10 μ m. and 3 beta -friedelinol, 3,7,12-o-triacetyl-8-o- tigloylingol and 3,12-o- diacetyl-7-o-angeloyl-8- methoxyingol, cytotoxic on - K562 cell line with 45%, 42%, & 53% inhibition at conc. 10 μ m
7.	Latex ²⁴	Acetone	Terpenoids	EAC cell line DLA cell line	Trypan blue exclusion method	DLA cell line – IC ₅₀ Conc. 51 μ g/ml EAC cell line – IC ₅₀ Conc. 82 μ g/ml

In Vivo Anticancer Activity of Snuhi (*Euphorbia neriifolia* Linn.) -

1. N-nitrosodiethylamine (DNA) Induced Renal Carcinogenesis²⁵ –

Plant Part Used – Dried Leaves

Extractive Solvent – Pet-ether, benzene, chloroform, ethyl acetate & ethanol.

Phytoconstituents Studied –

- i. Isolated Flavonoids – ENF [2-(3,4-dihydroxy-5-methoxy-phenyl)-3,5-dihydroxy-6,7-dimethoxychromen-4-one]
- ii. Hydro Ethanolic Extract of *Euphorbia Neriifolia* (EN)

Experimental Animals – Healthy male Swiss albino mice

Groups - Twelve groups of 6 mice each.

Parameters Studied –

- i. Levels of renal markers - urea and creatinine,
- ii. Xenobiotic metabolic enzymes - Cyt P450 and Cyt b5
- iii. Lipid peroxidation - LPO
- iv. Antioxidants - SOD, CAT, GST and GSH
- v. Biochemical Parameters - AST, ALT, ALP, total protein (TP), total cholesterol (TC)

Experimental Procedure – Experimental mice were pretreated with 150 and 400 mg/kg body weight of EN, 0.5% and 1% mg/kg body weight of butylated hydroxyanisole (BHA) as a standard antioxidant and 50 mg/kg body weight of ENF for 21 days prior to the administration of a single dose of 50 mg/kg body weight of DNA. DNA administration significantly ($p < 0.001$) decreased the body weight and increased the tissue weight. It significantly ($p < 0.001$) enhanced the levels of Cyt P450, Cyt b5 and LPO and decreased the levels of SOD, CAT, GST and GSH content. The activities of AST, ALT and ALP and the TP content and renal markers were also significantly decreased ($p < 0.001$), while TC level was markedly increased after DNA administration, as compared with the normal control group ($p < 0.001$). Pretreatment with EN and ENF counteracted DNA-induced oxidative stress (LPO) and exerted its protective effects by restoring the levels of antioxidants (SOD, CAT, GST and GSH), biochemical parameters (AST, ALT, ALP, TP and TC), renal markers (urea and creatinine) and xenobiotic enzymes (Cyt P450 and Cyt b5) in renal tissue.

Table 3: Animal Groups Involved

Groups	Drug Received
Group I (normal control, NC)	-
Group II (carcinogen control, CC)	distilled water for 21 days prior to a single dose of DNA, 50 mg/kg body wt, p. o
Groups III (E. neriifolia low dose, ENL)	ENL 150 mg/kg body wt/day
Group IV (E. neriifolia high dose, ENH)	ENH 400 mg/kg body wt/day, p.o for 21 days
Groups V (BHA low dose, BHAL)	BHAL 0.5%
Group VI (BHA high dose, BHAH) (standard treated group)	BHAH 1% mg/kg body wt/day, p.o for 21 days, dissolved in 0.5% acetone
Group VII (ENF)	ENF 50 mg/kg body wt/day; p.o, for 21 days, dissolved in distilled H ₂ O
groups VIII-XII	Pre-treated with EN, BHA (low and high dose) and ENF for 21 days and on day 22 DNA was administered and left for 10 days.

ALP - alkaline phosphatase; ALT - alanine aminotransferase; AST - aspartate aminotransferase; BHA - butylated hydroxyl anisole; BHAL - BHA lower dose; BHAH - BHA higher dose; CAT - catalase; CC - carcinogen control; CR - creatinine; Cyt - cytochrome; DNA - N- Nitrosodiethylamine; EN - *Euphorbia neriifolia*; ENF - EN flavonoid; ENH - EN higher dose; ENL - EN lower dose; GSH- reduced glutathione; GST – glutathione S-transferase; LPO - lipid peroxidation; NC - normal control; SOD- superoxide dismutase; TC - total cholesterol; TP - total protein.

2. N-nitrosodiethylamine induced hepatocarcinoma in mice²⁶ –

Plant Part Used – Leaves

Phytoconstituents Studies – ethanolic extract, isolated flavonoid.

Parameters Studied –

- i. Levels of liver markers - AST, ALT & ALP
- ii. Xenobiotic metabolic enzymes - Cyt P450 and Cyt b5
- iii. Lipid peroxidation (LPO)
- iv. Antioxidants - SOD, CAT, GST and GSH
- v. Biochemical parameters – TP and TC

Experimental Procedure – Experimental mice were pretreated with 150 and 400 mg/kg body wt of EN, 0.5% and 1% mg/kg body wt of butylated hydroxyanisole as a standard antioxidant and 50 mg/kg body wt of ENF for 21 days prior to the administration of a single dose of 50 mg/kg body wt of DNA. DNA administration significantly ($p < 0.001$) decreased the body weight and increased the tissue weight. Activities of liver markers, antioxidants and TP content were significantly decreased ($p < 0.001$), while Cyt P450, Cyt b5, LPO and TC levels were significantly ($p < 0.001$) increased after DNA administration as compared with the normal control group ($p < 0.001$). Pretreatment with EN and ENF counteracted DNA-induced oxidative stress (LPO) and exerted its preventive effects by restoring the levels of liver markers (AST, ALT and ALP), antioxidants (SOD, CAT, GST and GSH) and other biochemical parameters (TP and TC) and xenobiotic enzymes (Cyt P450 and Cyt b5) in liver tissue.

DISCUSSION

The data obtained in this study reveals anticancer activity of Snuhi (*Euphorbia neriifolia* Linn.). Most studies reveal use of dried leaf as a plant part for extraction of phytoconstituents and methanol is a common extractive solvent used. Phytoconstituents studied like Isolated flavonoids, sapogenin, triterpenoids, lingols are proven to be anticancerous on variety of cancer cell lines. Leukemia and melanoma cell lines are frequently used for anticancer study of *Euphorbia neriifolia* Linn. MTT assay was the most commonly found method of detection of cell cytotoxicity.

In-Vitro testing of total sapogenin against the murine F1 B16 Melanoma cell line showed 76.6 % cell viability at 10 µg/ml compared to 13.6 % at 500 µg/ml of total sapogenin. The assay data show that the IC₅₀ (over a period of 72 h) concentration of total sapogenin that inhibited growth of mouse melanoma cells by 50 % was 173.78 µg/ml compared to 120 ng/ml for vincristin. Study reports the potential anticancer property of *Euphorbia neriifolia* Linn. (Table 2, Sr. No. 1)

Methanolic extract of *Euphorbia neriifolia* was tested for its inhibitory action on B16F10 melanoma cell line under concentration range 10 µl to 100 µl. The percent viability was tested using Trypan blue dye exclusion method and cytotoxicity by SRB assay and MTT assay. IC₅₀ and R2 value by SRB assay was 198.26 and 0.710 respectively. Whereas IC₅₀ and R2 value by MTT assay was 212.78 and 0.762 respectively. Results have

shown significant activity that means it can be used as anticancer agent. (Table 2, Sr. No 2)

In Vitro Antitumor study of methanolic extract of *Euphorbia neriifolia* Linn. extract was counted as active possessing (scoring) >50% in HepG2 cytotoxicity. (Table 2, Sr. No.3)

MacKay-03 compound from *Euphorbia neriifolia* is able to inhibit growth, induce megakaryocytic differentiation, and to a lesser extent cause apoptosis in K562 and HEL human leukemia cells. (Table 2, Sr. No. 4)

The isolated compounds Neriifolin 1, 2, and 3, were evaluated for their cytotoxicity against three cancer cell lines HCT-116 (colon) and MCF-7 and MDA-MB-231(Breast) by MTT assay. Doxorubicin was used as a positive control with IC50 values ranging from 1.59 to 2.13 μ M. All three compounds (Neriifolin1,2,3) displayed more cytotoxicity on breast cancer MCF-7 cells than other two cell lines. The IC50 values of Neriifolin 1,2, and 3 on MCF-7 cells were 13.14, 7.12 and 9.50 μ M, respectively. Compound showed more cytotoxicity on MCF 7 cell line which suggests that present compound is active on estrogen receptor positive cancer cell line but not on triple negative MDA- MB - 231. (Table 2, Sr. No. 5)

The triterpenoids, 3 beta -friedelinol, 3 beta -taraxerol and 3 alpha-friedelinol cytotoxic on Panc-1 cell line,81T and BE3 cell line having inhibition 60% at conc. 10 μ m. Whereas 3 beta -friedelinol, 3,7,12-o-triacetyl-8-o-tigloylingol and 3,12-o-diacetyl-7-o-angeloyl-8-methoxyingol, cytotoxic on - K562 cell line with 45%, 42%, & 53% inhibition at conc. 10 μ m. The triterpenoids were found cytotoxic on Panc-1 cell line,81T and BE3 cell line. Whereas lingols were found cytotoxic on only K562 cell line. (Table 2, Sr. No. 6)

The acetone extract was found to contain terpenoids as the major component which was screened for its cytotoxic activity against the DLA and EAC cancer cells. The IC50 value of the Acetone extract of the latex of the EN on DLA cells was 51 μ g/ml. On EAC cells the Acetone extract of latex of EN had an IC50 value of 82 μ g/ml. Study concluded that acetone extract of *Euphorbia neriifolia* has significant anticancer activity. (Table 2, Sr. No. 7)

Whereas animal models reveal the anticancer activity of Snuhi (*Euphorbia neriifolia* Linn.) against DENA-induced renal carcinogenicity using hydro-ethanolic extract *E. neriifolia* and ENF.

Hepatocarcinoma animal study showed significant anti-carcinogenic potential of the hydro-ethanolic extract of *E. neriifolia* and ENF against DENA induced hepatic carcinogenicity. Study also reveals that Snuhi (*Euphorbia neriifolia* Linn.) releases the oxidative stress and exerts its preventive effect which restores level of liver markers, antioxidants, biochemical parameters, and xenobiotics in liver tissue.

CONCLUSION

Present review accomplishes anticancer activity of Upavisha Snuhi (*Euphorbia neriifolia* Linn.). However, the studies conducted include only extracted phytoconstituents as an active anticancer agent. There are number of formulations which constitutes Snuhi (*Euphorbia neriifolia* Linn.) as an ingredient containing similar phytoconstituents. Hence the need is to conduct further studies on formulations of Upavisha Snuhi (*Euphorbia neriifolia* Linn.) as an anticancer agent. More

thorough experimental studies and clinical data can suffice the validation of Snuhi as an anticancer agent.

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How to cite this article:

Asolkar Geeta Govindrao *et al.* Anticancer activity of upavisha snuhi: A comprehensive update. *J Pharm Sci Innov.* 2020;9(6):162-166.

<http://dx.doi.org/10.7897/2277-4572.096190>

Source of support: Nil, Conflict of interest: None Declared

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