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ORIGINAL ARTICLE

In vitro Antiproliferative Effects of a Bulb Lectin from *Scadoxus multiflorus* (SmL) on Human Prostate Cancer Cells

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ABSTRACT

Background: Prostate cancer is a major health burden, and new therapeutic agents from natural sources are urgently needed. Plant lectins, known for their selective binding to cancer cell surface glycans, show promising antiproliferative potential. This study investigates the *in vitro* effects of a bulb lectin from *Scadoxus multiflorus* on human prostate cancer cells. **Methods:** A purified lectin from *Scadoxus multiflorus* bulbs was evaluated against human prostate cancer cell lines. It's cytotoxic and antiproliferative effects were measured using the MTT assay, while the scratch assay assessed its influence on cell migration. Data were analysed to establish its dose-dependent antiproliferative potential. **Results:** The MTT assay demonstrated that SmL significantly inhibited prostate cancer cell proliferation in a concentration-dependent manner after 48 h of treatment, with an IC_{50} value of 51.36 µg. In contrast, SmL exhibited no appreciable cytotoxicity toward normal Human Embryonic Kidney (HEK293) cells, even at higher concentrations, indicating its selectivity for tumour cells. Furthermore, the scratch assay revealed that SmL effectively suppressed cancer cell migration. **Conclusion:** These findings suggest that the lectin from *Scadoxus multiflorus* bulbs (SmL) possesses selective antiproliferative activity against human prostate cancer cells by inhibiting proliferation and migration while sparing normal cells. SmL thus represents a promising natural candidate for further investigation as a potential therapeutic agent for prostate cancer.

Keywords: *Scadoxus multiflorus*, Bulbs, Lectin, Antiproliferative activity, Wound healing assay

INTRODUCTION

Prostate cancer is one of the most frequently diagnosed malignancies among men worldwide and remains a major cause of cancer-related mortality. Despite advances in early detection and therapeutic strategies, the progression to advanced and treatment-resistant forms of prostate cancer poses a significant clinical challenge. This has prompted growing interest in the search for novel therapeutic agents, particularly those derived from natural sources with selective cytotoxicity and fewer side effects [1]. Lectins, a diverse group of carbohydrate-binding proteins, are increasingly recognized for their biomedical potential, including antimicrobial, immunomodulatory, and anticancer activities. They can bind with specific carbohydrate moieties reversibly, but they are neither antibodies nor enzymes in nature. Besides, they do not modify the bound carbohydrates biochemically. Many plant lectins exhibit the ability to bind selectively to altered glycan structures present on cancer cell surfaces, thereby interfering with cell proliferation, inducing apoptosis, or modulating immune responses. Such properties make lectins attractive candidates for anticancer drug development [2].

Scadoxus multiflorus (family: Amaryllidaceae), commonly known as the blood lily, is an ornamental and medicinal plant traditionally used in African and Asian ethnomedicine for treating various ailments. The bulbs of *S. multiflorus* are rich in bioactive compounds, including alkaloids, flavonoids, and lectins, which have been reported to exhibit antimicrobial, anti-inflammatory, and cytotoxic activities [3]. However, systematic evaluation of its lectin fraction against human cancers, particularly prostate cancer, remains limited. Given the pressing need for novel and effective therapeutics against prostate cancer, investigating the antiproliferative potential of lectins derived from *S. multiflorus* bulbs offers a promising approach. This study aims to evaluate the *in vitro* antiproliferative effects of a bulb lectin from *Scadoxus multiflorus* on human prostate cancer cell lines, with a focus on its antiproliferative properties.

MATERIALS AND METHODS

For all *in vitro* assays previously purified lectin from *Scadoxus multiflorus* bulbs (SmL) was used. Stock solution of SmL was prepared by dissolving in sterile PBS and filtered (0.22 µm). Working concentrations of SmL were prepared fresh in culture medium.

Cell culture and maintenance

PC-3 (Human prostate cancer cell line) and HEK-293 (Human embryonic kidney normal cell line) were purchased from the National Center for Cell Sciences (NCCS), Pune (India). The cells were maintained aseptically at 37˚C in Dulbecco's Modified Eagle's Medium (DMEM) supplemented with 10% FBS, two mM L-glutamine, 100 IU/mL penicillin, and 100 µg/mL streptomycin in a humidified atmosphere, containing 5% CO_2. Cells were seeded and allowed to adhere overnight, then treated with different doses of SmL for 48 h. Culture medium was used as a control. All chemicals and reagents used in the study were of analytical grade and were procured from SRL (Sisco Research Laboratories, India), India, Hi-Media, India and standards were from Sigma, USA.

Determination of Antiproliferative Activity by MTT assay

In vitro inhibition effects of SmL on PC-3 (human prostate cancer cell line) and HEK-293 (Human embryonic kidney normal cell line) were demonstrated using MTT assay as previously described by Mosmann [4]. Briefly, cells were seeded in 96-well plates at a density of 10,000 cells/well and incubated for 24hr. After 24hr, cells were replenished with fresh media. Then 100µl of different concentrations of the lectin (SmL) (6µg, 12µg, 24µg, 36µg, 48µg and 60µg / 500µl) were added to wells in triplicate and incubated for 24 hr. After incubation, the culture medium was removed and adhered cells were washed with PBS. Subsequently, 100µl of MTT reagent diluted in culture medium to the final concentration of 0.5 mg/mL was added and incubated for 4 h. MTT reagent was then removed and 200µL of Dimethyl sulfoxide (DMSO) was added to dissolve the formazan crystals. The absorbance of the solution was recorded at 570 nm and cell viability was determined for each assay including control wells that did not contain lectin. All measurements were performed in triplicates. The results were expressed as a percentage of the control/vehicle group (considered as 100% viable). The IC_{50} value was determined to be 50% of the SmL ability to suppress cell growth. The percentage (%) of cell viability is calculated using the following formula:

Cell viability % = [As/Ac] x 100

Where: As-absorbance of treated cells; Ac-absorbance of control cells

Evaluation of Cell Migration Suppression through Wound Healing Assay

In vitro scratch assay was performed as described previously [5]. Briefly, 6×10^5 PC-3 cells were seeded onto six-well plate, after reaching confluency, a scratch was made using 200 µL pipette tip. The open space was tracked using live cell imaging microscope (Motic AE2000 series camera), photographed at 0, 6, 12, and 24 h interval, and the percentage of wound closure was quantified using Image J (IJ 1.46r). The migration was expressed as % wound closure relative to 0 h.

Statistical analysis

All experiments were performed in triplicate ($n \geq 3$). Data are presented as mean ± SD and were analysed using two-way ANOVA followed by a non-parametric t-test, with $p < 0.05$ considered statistically significant.

RESULTS AND DISCUSSION

Source of Lectin (SmL)

SmL (Scadoxus multiflorus lectin) from *Scadoxus multiflorus* bulbs was purified and characterized earlier using conventional analytical techniques was used for its potential cell growth inhibitory and anti migratory effect .

Antiproliferative Effect of SmL on PC-3 and HEK-293

In this study, we investigated the antiproliferative activity of SmL on PC-3 (human prostate cancer) and HEK-293 (Human embryonic kidney normal) cell lines by MTT assay. Fig. 1A & Fig. 1B represents the percentage of cell viability shown by PC-3 and HEK-293 on lectin treatment in a concentration-dependent manner after 48 h of exposure respectively. SmL showed no significant cytotoxicity towards HEK-293 even at higher concentration (50µg/500µl), demonstrating selectivity for

tumour cells. After 48 h, IC_{50} of SmL on the viability of PC-3 was 51.36 µg/500µl. The discrepancy in lectin cytotoxicity may be caused by the divergence of glycoprotein expression on different cell lines.

In recent decades, several antiproliferative and apoptosis-inducing lectins have been reported from plant bulbs, including those from *Sauromatum venosum* and *Arisaema jacquemontii* [6, 7]. These lectins typically exert their anticancer activity by recognizing specific carbohydrates on the surface of cancer cells, thereby interfering with cell survival mechanisms.

In the present study, the bulb lectin from *Scadoxus multiflorus* (SmL) demonstrated significant inhibition of cell growth, as evidenced by the MTT assay. At a concentration of 51.36 µg/500 µl (IC_{50}), SmL effectively reduced cell viability, suggesting a cytotoxic effect. The observed reduction in cell growth may be attributed to the lectin's binding to surface glycoconjugates on prostate cancer cells, consistent with the glycan-mediated mechanism reported for other plant lectins. For instance, mannose-binding lectin from *Remusatia vivipara* has been shown to induce strong glycan-dependent cytotoxicity and suppress the proliferation and motility of human breast cancer cells [8].

To the best of our knowledge, this is the first report describing the antiproliferative activity of a bulb lectin from *Scadoxus multiflorus*. While lectins from other botanical sources have been extensively documented for their antiproliferative potential [9, 10], our findings highlight SmL as a novel plant-derived candidate with promising cytotoxic effects on prostate cancer cells.

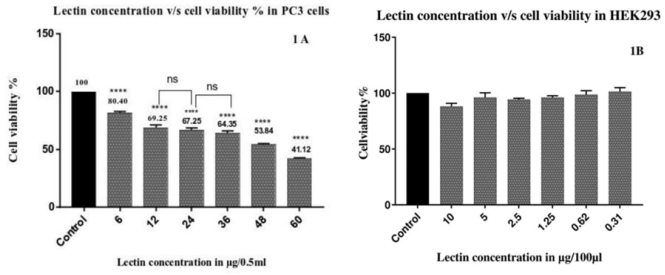

Fig. 1: Effect of SmL on cell viability

Growth inhibition assessed by MTT assay in (A) PC-3 prostate cancer cells treated with increasing concentrations of SmL (6, 12, 24, 36, 48, and 60 µg/500 µl) and (B) HEK-293 noncancerous cells treated with serial concentrations of SmL (0.31, 0.62, 1.25, 5, and 10 µg/100 µl) for 48 h. Cell viability of untreated controls was considered 100%. Data are expressed as mean ± SE (PC-3) or mean ± SD (HEK-293) from two independent experiments performed in triplicate.

SmL Decrease Capacity of Cell Migration in PC-3 Cells

Cellular migration and motility pathways play a crucial role in the invasive behaviour of prostate cancer cells. In the scratch assay, treatment with SmL at its IC_{50} concentration (51.36 µg/500 µl) significantly inhibited cell migration. At 0, 6, 12, and 24 hours, the percentage of wound area remaining in SmL-treated cells was approximately 100%, 75%, 60%, and 25%, respectively, compared with ~100%, 75%, 25%, and 3% in control cells. While the control group achieved near-complete wound closure (~3%) within 24 hours, SmL treatment markedly delayed closure, indicating strong anti-migratory activity Fig. 2.

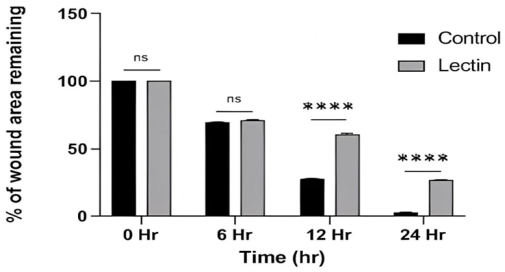

Fig. 2: Effect of SmL on migration of PC-3 cells

Inhibition of PC-3 cell migration following treatment with SmL at its IC_{50} concentration (51.36 µg/500 µl) across different time points. Data are presented as mean ± SE from two independent experiments performed in triplicate.

CONCLUSION

The present study demonstrates that the bulb lectin from *Scadoxus multiflorus* (SmL) exerts significant *in vitro* antiproliferative effects against human prostate cancer cells. SmL inhibited cell proliferation, suppressed migratory potential, and induced cell death in a dose-dependent manner, highlighting its potential as a novel bioactive protein with therapeutic relevance. The observed selective cytotoxicity toward cancer cells underscores its promise as a safer alternative to conventional chemotherapeutics. Further studies focusing on mechanistic pathways, molecular targets, and *in vivo* validation are warranted to establish SmL as a potential candidate for prostate cancer therapy.

DISCLOSURE

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Competing Interest: None

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