Dioscorea bulbifera-Loaded Phytosomes as a Novel Cardioprotective Agent: A Study on Antioxidant and Pro-Angiogenic Activity

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***Abstract*— The COVID-19 pandemic highlighted the importance of cardiovascular health, emphasizing the need for cardioprotective strategies. Cardiovascular disease (CVD), primarily due to atherosclerosis, remains the leading global cause of mortality. A promising therapeutic approach is revascularization through pro-angiogenesis, which enhances blood vessel formation to restore circulation in ischemic tissues. *Dioscorea bulbifera*, a member of the family Dioscoreaceae, traditionally used for wound healing, contains diosgenin, a bioactive compound known to modulate vascular pathways. However, its pro-angiogenic potential remains unexplored.**

This study involves extraction and yield optimization of *Dioscorea bulbifera* tuber bio actives, followed by phytochemical screening and standardization using UV spectroscopy and Thin Layer Chromatography. Phytosomes were formulated using the solvent injection method and characterized by measuring particle size, zeta potential, and entrapment efficiency. Antioxidant activity was evaluated using the DPPH assay & the pro-angiogenic potential was evaluated through the Chick Chorioallantoic Membrane (CAM) assay, wherein it was found that Dioscorea phytosome formulation induced more pro-angiogenesis as compared to the crude extract. Thus, these findings confirm the antioxidant capacity and suggest a promising pro-angiogenic potential of *D. bulbifera*, that can hence be explored as a possible novel candidate for cardiovascular protection, within phytosomes can provide a strategic approach for enhanced cardio protection.

***Index Terms*— *Dioscorea bulbifera, cardiovascular protection, phytosomes*, pro-*angiogenesis*, and antioxidant**

I. INTRODUCTION

The COVID-19 pandemic served as a significant reminder of the importance of cardiovascular health. Individuals with pre-existing heart conditions were at a heightened risk of severe complications, while the virus itself could trigger serious issues such as myocardial injury, arrhythmias, and acute coronary events[1]. With the global upheaval, consciousness has arisen toward the urgent need of a global cardioprotective agenda to address not just the long-term cardiovascular risks but to also prepare us for similar health emergencies in the future.

Cardiovascular disease (CVD) remains the leading cause of mortality worldwide[2] and significantly contributes to reduced quality of life, with atherosclerosis being the primary underlying factor. Atherosclerosis is the build-up of plaque on arterial walls and is a major risk factor for myocardial infarction (heart attack), a condition resulting from severely reduced or completely obstructed blood flow to the heart, causing damage to cardiac tissue[3].

Current treatments, including lipid-lowering agents, antihypertensives, and antiplatelet drugs, effectively manage symptoms but do not directly address the underlying inflammatory processes driving disease progression. Furthermore, commonly used drugs like statins and aspirin are associated with adverse effects, underscoring the demand for safer and more targeted alternatives[4].

One promising therapeutic approach can be revascularization through therapeutic angiogenesis, which stimulates the formation of new blood vessels to restore perfusion in ischemic tissues. This strategy can enhance blood flow, reduce cardiac damage, and support tissue regeneration. In this context, phytochemicals have garnered increasing interest due to their natural origin, favourable safety profile, and cardioprotective potential.

Among these, *Dioscorea bulbifera*, an edible medicinal plant from the family Dioscoreaceae, commonly known as “air potato” has been traditionally used in African, Chinese, and Indian medicine to treat a wide range of ailments[5].

Botanical Classification of *Dioscorea bulbifera*[6]:
Kingdom: Plantae
Phylum: Tracheophyta
Class: Magnoliopsida
Order: Dioscoreale
Family: Dioscoreaceae
Genus: *Dioscorea*
Species: *bulbifera*

Various phytochemicals present in *Dioscorea bulbifera* tubers includes bioactive agents such as saponins, tannins, flavonoids, phenolic acids, terpenoids, steroids, and cardiac glycosides. Saponins are the most abundant, followed by cardiac glycosides and terpenoids, which contribute to the plant's diverse therapeutic properties such as antioxidant, antimicrobial, and cardioprotective effects.

One of the principal bioactive agents is diosgenin, which is a steroidal sapogenin. It plays a critical role as a precursor in the synthesis of steroid hormones and pharmaceuticals[7]. *Dioscorea bulbifera* also shows the ability to heal wounds, anti-inflammatory, and antioxidant activities[8]. Diosgenin has also shown to protect against hypoxia-reoxygenation (HR)-induced injury in H9c2 cardio myoblast cells, thereby implying its relevance against ischemia-reperfusion damage[9].

The Dioscorea genus is known for a wide range of pharmacological activities, including immunomodulatory, anti-inflammatory, neuroprotective, hypoglycaemic, and hypolipidemic effects[10]. Under hypoxic conditions, diosgenin has been found to upregulate angiogenesis-related genes like *Aggf1* via transcription factors such as HAND2, promoting endothelial cell proliferation and migration[11].

Phytosome technology has emerged as a promising drug delivery strategy. Phytosomes are molecular complexes formed by combining plant extracts with phospholipids, enhancing the solubility, stability, and absorption of bioactive compounds. Phytosomes may improve the bioavailability of herbal constituents by encapsulating hydrophilic plant molecules within the hydrophilic head of phospholipids, leading to better pharmacokinetic profiles and therapeutic effects. Their performance is influenced by properties such as Particle size and shape, Surface charge, Chemical composition, Encapsulation efficiency and Drug release behaviour[12].
Integrating novel delivery systems like phytosomes into herbal medicine may enhance efficacy and reduces side effects. Advances in analytical techniques now support improved standardization, and the use of systems such as nanoparticles, liposomes, and solid dispersions[13].

Angiogenesis refers to the formation of new blood vessels from pre-existing vessels, begins during embryonic development and continues across the life span[14]. To investigate the pro-angiogenic potential of the natural compound, an in vitro model, the Chick Chorioallantoic Membrane (CAM) assay is employed. The CAM assay is a widely accepted, rapid, and cost-effective method for studying angiogenesis, owing to its rich vascular network, ease of manipulation, and lack of a fully developed immune system during early embryonic stages[15].
This assay allows direct visualization and quantification of newly formed blood vessels in response to the applied test substance. The high vascularization of the CAM makes it an ideal environment to assess the angiogenic or anti-angiogenic activity of natural compounds[16]. The observed increase in blood vessel density and branching in the treated CAMs compared to controls suggests that the tested formulation exhibits significant pro-angiogenic activity, potentially contributing to cardio protection through enhanced blood flow in ischemic tissues.

The aim of this study is to evaluate the antioxidant and pro-angiogenic potential of a *Dioscorea bulbifera* tubers delivered via a Phytosome-based nanoscale system, using the Chick Chorioallantoic Membrane (CAM) assay as the primary in vitro model. It is hypothesized that the dioscorea phytosomal formulation may enhance the therapeutic efficacy of the compound, resulting in significantly improved angiogenesis, antioxidant and ultimately cardioprotective potential compared to the unformulated extract.

II. MATERIALS AND METHODS:

1. Procurement, Authentication, and Physicochemical Analysis of Crude Drug:

Dried tubers of Dioscorea bulbifera were procured from A. Amritlal and Co., herbal raw material supplier located in Pydhonie, Mumbai (400003). The plant material was authenticated by Dr. Harshad Pandit, and the specimen was assigned the identification number avm p 25015458**.** The authenticated tubers were cleaned, homogenised into a fine powder, and stored in an airtight container.
Physicochemical parameters such as total ash and acid-insoluble ash values were determined using standard procedures and evaluated against the limits prescribed as per the Ayurvedic Pharmacopoeia of India[17].

2. Extraction and Yield Optimization of Crude Drug

Extraction was carried out using different solvent systems: 80% ethanol, 80% methanol, and water. Initial extraction yields were compared to determine the most suitable solvent. Subsequently, yield optimization was performed using two techniques Soxhlet extraction and ultrasonication[18,19], to identify the method yielding the highest extractive value.

3. Preliminary Phytochemical Screening

The extract was analyzed for preliminary phytochemical screening using standard qualitative tests[20, 21]. The presence of various phytoconstituents such as alkaloids, glycosides, flavonoids, saponins, phenolics, tannins, amino acids, steroids**,** and carbohydrates was assessed in both aqueous and alcoholic extract.

4. Standardization of Extract

a. Thin Layer Chromatography (TLC)

TLC analysis was conducted on silica gel 60 F_{254} pre-coated plates. Diosgenin was used as the reference standard. Several mobile phase combinations of toluene and ethyl acetate were tested to optimize the solvent system and reduce issues like tailing. The developed TLC plate was sprayed with anisaldehyde-sulphuric acid reagent and heated to visualize the spots[22].

b. UV-Visible Spectroscopy

Standard and extract solutions were prepared in ethanol. The solutions were scanned using a Jasco V-730 UV-Visible Spectrophotometer in the range of 200–600 nm to identify and compare the λmax. The UV spectrum of the extract was matched with that of the diosgenin standard to confirm its presence.

Total Diosgenin Content

Total diosgenin content was quantified using the calibration curve method with diosgenin as the standard. A series of standard diosgenin solutions (10-50 µg/mL) were prepared in ethanol. Each standard solution was scanned at λmax of 205 nm using a UV-Visible spectrophotometer (Jasco V-730), and a calibration curve was plotted. The extract sample was treated similarly, and the diosgenin content was calculated based on the standard curve.

5. Quantitative phytochemical analysis:

a. Total Phenolic Content

Total phenolic content was performed using the Folin–Ciocalteu method[23], in which, gallic acid was used as a reference standard & the results were reported as (mg GAE/g) mg gallic acid equivalents per gram of extract.

b. Total Flavonoid Content

Total flavonoid content was estimated using the $AlCl_3$ colorimetric assay[24], in which, quercetin was used as a reference standard. The results were reported as (mg QE/g) mg quercetin equivalents per gram of extract.

6. Formulation of phytosomes & Optimization of phytosomal formulation by QbD.

Phytosomes were formulated using the solvent injection method[25]. In the organic phase, phospholipid and cholesterol were dissolved in ethanol and sonicated. At the same time, in the aqueous phase, the extract was mixed with ethanol and phosphate buffer and also sonicated. The aqueous phase was stirred while the organic phase was added dropwise. After complete addition, the mixture was stirred at an increased speed for a set duration. Finally, the resulting phytosomal dispersion was homogenized to reduce particle size.

The stability of the phytosomal batch was preliminarily evaluated by observing clarity and sedimentation. Further characterization included particle size analysis and entrapment efficiency.

In the 2^3 factorial design, three independent variables were evaluated: concentration of extract (A) ranging from 5 mg (−1) to 10 mg (+1), concentration of lipid (B) from 10 mg (−1) to 20 mg (+1), and stirring time (C) varying between 1 hour (−1) and 3 hours (+1). These variables were studied at two coded levels, low (−1) and high (+1), to assess their effect on the formulation. This design generated eight experimental runs, each representing a unique combination of the coded high and low levels of extract concentration, lipid concentration, and stirring time.

7. Evaluation of Phytosomes:

The optimized phytosomal formulation was assessed by measuring particle size, zeta potential, and entrapment efficiency[26]. To evaluate entrapment efficiency, the phytosomal dispersion was centrifuged, and the supernatant was collected, diluted, and analyzed for unentrapped extract using a UV spectrophotometer[27]. The entrapment efficiency was then calculated by-

$$\text{Entrapment efficiency}(\%) = \frac{\text{Weight of total extract} - \text{Weight of unentrapped extract}}{\text{Weight of total extract}} \times 100$$

8. Drug excipient compatibility studies:

Fourier Transform Infrared Rays (FTIR) was used to analyze the compatibility between extract and other excipients like lipid and cholesterol[28]. Phytosome formation occurs when phospholipids and plant extracts interact, and FTIR can verify this by observing absorption peak shifts. The instrument used was Bruker FTIR spectrometer. IR spectra was obtained in the range of 4000 to 400 cm^{-1}. Comparison of IR spectra of Dioscorea extract, lipid and cholesterol was done with that of dioscorea phytosome.

9. In-vitro evaluation of antioxidant activity of extract:

The antioxidant potential of both the extract and the phytosomal batch was evaluated using the DPPH assay, which involves the reduction of a violet-coloured free radical to yellow in the presence of antioxidants. Ascorbic acid was used as a reference standard. The results reflected the antioxidant activity by comparing the reduction of DPPH. % Scavenging activity was determined using the formula:

$$\%\ \text{Scavenging activity} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

A calibration curve (% scavenging vs concentration) was plotted to determine the IC_{50} value[29,30].

10. In-vitro evaluation of Proangiogenic activity of the formulated phytosomes by CAM assay:

The in vitro pro-angiogenic activity of the extract and dioscorea phytosomes was evaluated using the Chick Chorioallantoic Membrane (CAM) assay, a widely accepted model for assessing new blood vessel formation. The CAM assay was employed to examine the effect of test samples on angiogenesis, using the protocol established by Ribatti and co-workers (1997)[31]. 12 days old Fertilized Chicken eggs were collected from a local hatchery & were cleaned with ethanol and incubated at 37°C with constant humidity. A small volume of albumin was withdrawn to detach the CAM, followed by the creation of a window on the eggshell. Sterile gel foam pieces impregnated with phosphate-buffered saline (PBS) as a negative control, VEGF-A as a positive control, and test samples were placed on the CAM surface. After 24 hours of incubation, the eggs were opened, and CAM images were captured using a stereo microscope. The number of vessel branch points in a defined region was counted to determine the angiogenesis index. This index, representing the average number of new vessel branches, was used to compare the pro-angiogenic potential of the extract and phytosomal formulation.

11. Formulation and evaluation of Phytosomal tablets:

The optimized batch of lyophilized *Dioscorea bulbifera* phytosomes was formulated into tablets by direct compression. The formulation included dried dioscorea phytosomes as the active drug, Avicel PH 102 as binder and diluent, Anhydrous Lactose as filler, Talc as glidant, Aerosil 200 as flow enhancer, and Magnesium stearate as lubricant. All ingredients were weighed, sieved, mixed, and compressed using an 8 mm punch to obtain tablets of 250 mg. The tablets were evaluated for weight variation, hardness, disintegration time, and friability as per pharmacopoeial standards to ensure quality and performance[32].

III. RESULT & DISCUSSION:

1. Physicochemical Analysis of crude drug:

The total ash value of the crude drug was determined to be 4.5%w/w, which lies in pharmacopoeial limit of NMT 6% and the Acid insoluble ash value of the crude drug was determined to be 0.86%w/w, which lies in pharmacopoeial limit of NMT 1%.

2. Extraction and yield optimization of crude drug:

The extraction yield was significantly influenced by both the solvent and extraction technique. Among the solvents, 80% ethanol gave the highest yield at 3.862% w/w, followed by 80% methanol with 3.26% w/w and water with 2.35% w/w. In terms of technique, ultrasonication yielded 3.862% w/w, proving more efficient than Soxhlet extraction, which gave 2.296% w/w, likely due to better solubility and enhanced cell disruption.

3. Preliminary Phytochemical Screening & evaluation of extract:

Preliminary phytochemical screening results showed that flavonoids, tannins, and phenolic acids were present in both aqueous and alcoholic extracts, whereas glycosides, steroids, saponins, alkaloids, and carbohydrates were present selectively, depending on the polarity of the solvent used.

4. Standardization of Extract

TLC analysis was carried out using different mobile phase ratios of toluene and ethyl acetate to optimize the separation of constituents. At a 7:3 ratio, the Rf value was 0.79 for standard and 0.813 for the sample. With 8:3, the standard showed 0.63 and the sample 0.67. For 6:4, the standard was 0.72 and the sample 0.74. At a 5:5 ratio, the standard and sample showed 0.77 and 0.79 respectively. The best separation was observed at a 5:6 ratio, where the standard has a Rf of 0.81 and the sample has 0.802, confirming the presence of diosgenin in the extract.

Std	Extract
Rf1: 0.81	Rf2: 0.802

Table no.1: TLC profile of standard diosgenin and *Dioscorea bulbifera* extract using toluene: ethyl acetate (5:6).

The extract of *Dioscorea bulbifera* tubers was standardized using UV-Visible spectroscopy and Thin Layer Chromatography (TLC). The UV spectrum of the extract displayed a prominent absorption peak at 207.2 nm, closely matching the peak observed for standard diosgenin at 206.6 nm, confirming the presence of diosgenin in the extract (figure 1).

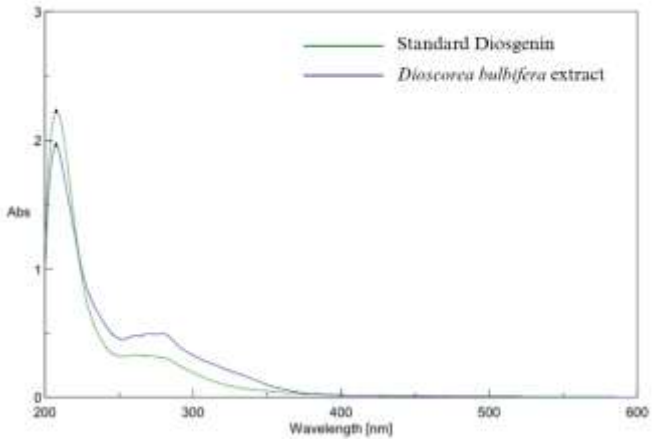


Figure no. 1: UV-Visible absorption spectra of standard diosgenin and *Dioscorea bulbifera* extract

The extract of *Dioscorea bulbifera* tubers was found to be rich in bioactive constituents. The total diosgenin content, determined through a calibration curve with a linear equation of $y = 0.0127x + 0.0487$ and an R^2 value of 0.9985, was found to be **9.73% w/w.**

5. Quantitative phytochemical analysis:

- Total phenolic content, calculated using the equation $y = 0.0582x + 0.0493$ with an $R^2 = 0.9906$, was found to be 32.03 mg GAE/gm of extract.
- Total flavonoid content was assessed using the equation $y = 0.0176x + 0.02$ with an R^2 of 0.9949, which equated to 28.64 mg QE/gm**.**

6. Formulation of phytosomes & Optimization of phytosomal formulation by QbD.

Runs	Factor A: Concentration of Extract	Factor B: Concentration of Lipid	Factor C: Stirring Time	Response 1: Entrapment efficiency (%)	Response 2: Particle size (nm)
1	5	10	1	87	497
2	10	20	1	89	483
3	10	20	3	93	469
4	5	20	3	92	480
5	5	10	3	90	490
6	10	10	3	91	475
7	10	10	1	89	482
8	5	20	1	88	489

Table no. 2: Experimental Design Matrix with Observed Responses

Following Table 2, a 2^3 full factorial design was employed with extract concentration (Factor A), lipid concentration (Factor B), and stirring time (Factor C) as independent variables, while entrapment efficiency (EE) and particle size (PS) as the responses. The experimental data were analyzed using Design-Expert software (version 13.0.15). The significance of each model term was evaluated at a 95% confidence level ($p < 0.05$).

Effects of Independent variables on Entrapment efficiency:

The polynomial equation for entrapment efficiency (EE) in terms of coded factors is: EE (%) = +89.88 + 0.6250*A+ 0.6250*B+ 1.62*C. The model was statistically significant (F-value 24.33, p = 0.0055), with all three variables showing significant positive effects ($p < 0.05$) on EE. Factor C had the greatest impact, indicating that increasing the stirring time increases entrapment by improving dispersion and vesicle formation. Increased concentration of extract and lipid also contributed by promoting better interaction with the lipid bilayer. The model demonstrated strong reliability with $R^2 = 0.9481$, Adjusted $R^2 = 0.9091$, and Predicted $R^2 = 0.7922$, along with an Adequate Precision of 13.279.

Effects of Independent variables on Particle size:

The effect of formulation variables on particle size was analyzed using a polynomial model in Design Expert: Particle size (nm) = +482.63 – 5.38*A – 3.38*B – 4.13*C. The model was statistically significant (F-value 10.27, p = 0.0238), with all three factors showing significant negative effects ($p < 0.05$) on particle size. Factor A had the greatest impact, indicating that increased extract concentration reduces the particle size. Increased lipid concentration and stirring time also reduced particle size by enhancing homogenization. The model showed $R^2 = 0.8851$, Adjusted $R^2 = 0.7989$, Predicted $R^2 = 0.5404$) and Adequate Precision of 9.4420. These findings highlight the importance of optimizing formulation parameters to achieve smaller particle sizes, enhancing stability in phytosomal systems.

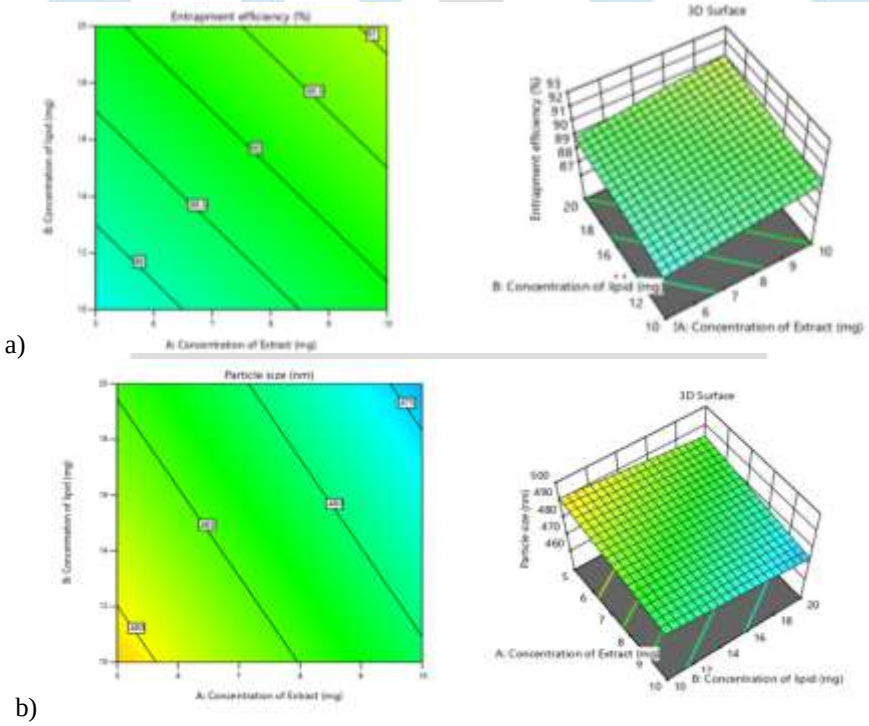
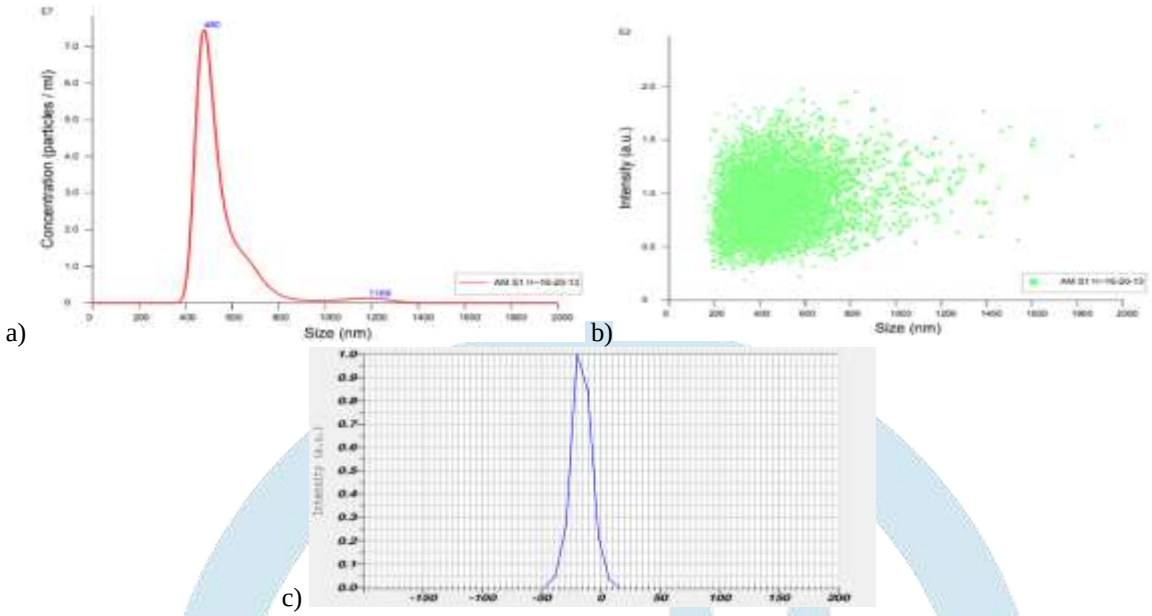


Fig.2: a) Contour plot and 3D surface plot showing the effects of Factor A, B, C on EE. b) Contour plot and 3D surface plot showing the effects of Factor A, B, C on PS.

7. Evaluation of optimized phytosomal batch.



a) b) c)

Fig.3: a) Particle Size Distribution Curve of the Optimized Phytosomal Batch; b) PDI Scatter Plot of the Optimized dioscorea phytosome; c) Zeta Potential Distribution of the Optimized dioscorea phytosome

The optimized *Dioscorea bulbifera* phytosomal batch showed a particle size of 480 nm and a zeta potential of –16.3 mV, indicating good stability. The entrapment efficiency was found to be 92.2%, suggesting effective incorporation of the extract into the phytosomal formulation.

8. Drug excipient compatibility studies:

FTIR analysis was conducted to assess compatibility between *Dioscorea bulbifera* extract and excipients. The extract showed O–H stretching at 3333 cm^{-1}, C–H stretching at 2979–2931 cm^{-1}, and C=O/aromatic vibrations between 1762–1396 cm^{-1}. Lipid peaks appeared at 3313 cm^{-1} (O–H) and 2956–2849 cm^{-1} (C–H), while cholesterol showed peaks at 3408 cm^{-1}, 2929–2849 cm^{-1}, and 1730 cm^{-1}. The phytosomal formulation retained key peaks, including broad O–H at 3333 cm^{-1} and C–H around 2980 cm^{-1}, with minor shifts at 1639 and 1084–904 cm^{-1}. Absence of new or missing peaks confirmed compatibility and no degradation.

9. *In-vitro* evaluation of antioxidant activity of extract:

The antioxidant activity of the extract, its phytosomal formulation, and ascorbic acid (standard) was assessed using the DPPH free radical scavenging assay. The percentage scavenging activity (%SCV) increased in a concentration-dependent manner across all samples. At 100 µg/mL, the %SCV of the dioscorea extract, dioscorea phytosome, and ascorbic acid was 75.62%, 77.23%, and 76.74%, respectively. Notably, the phytosomal formulation demonstrated slightly higher antioxidant activity compared to the extract alone, indicating enhanced efficacy. The IC_{50} values were calculated as 67.78 µg/mL for ascorbic acid, 52.16 µg/mL for the extract, and 51.82 µg/mL for the phytosomes, suggesting improved free radical scavenging potential upon phytosomal encapsulation.

10. *In-vitro* evaluation of Proangiogenic activity of the formulated phytosomes by CAM assay.

In this study, given samples *Dioscorea bulbifera* (DT) extract and its phytosomal formulation were evaluated to measure the therapeutic angiogenic activity on the Fertile Chick eggs. The No of Branching Points resulted in each culture condition were mentioned as follows:

Sr. no.	Condition	No. of branching vessels ± SD
1	Control	41 ± 9.54
2	Std control (VEGF-10 ng/ml)	121.33 ±21.73
3	Dioscorea tubers extract- 1000ppm	104.33 ±7.7
4	Dioscorea Phytosome- 1000ppm	151.33 ±13.01

Table 3: Details of concentrations of test compounds along with controls. Presented values were the average of 3 independent individual experiments.

The CAM assay (Figure 4) demonstrated enhanced angiogenesis in fertilized eggs treated with the Dioscorea phytosomal formulation, as evidenced by increased vascular branching and density. This observation is quantitatively supported by the data in Table 6, and visually reinforced by the bar graph in Figure 5, where the Dioscorea phytosome group showed a greater number of branching points compared to the Dioscorea extract and the control. These findings highlight the potential of *D. bulbifera* tubers phytosomes in promoting neovascularization, supporting their role in cardiovascular regenerative therapy.

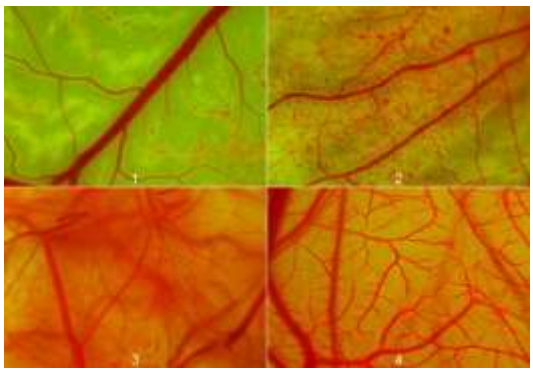


Fig.4: CAM assay. (1) Control, (2) Std control (VEGF-10ng/ml), (3) Dioscorea tuber extract with 1000ppm (4) Dioscorea Phytosome with 1000ppm treated on CAM

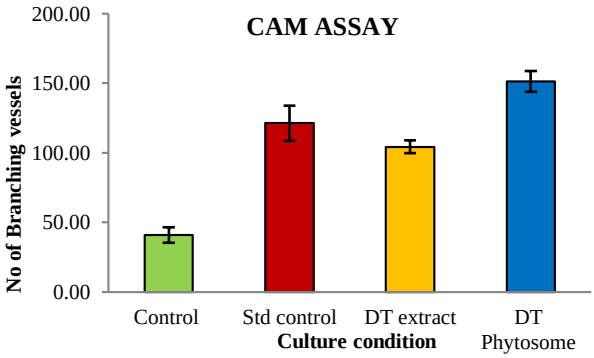


Fig.5: Bar graph showed the counted branching points in different conditions of CAM.

11. Formulation and evaluation of Phytosomal tablets.

The optimized phytosomal formulation of *Dioscorea bulbifera* was successfully compressed into tablets and evaluated for key pharmaceutical quality parameters. Weight variation was assessed across 20 tablets, with all individual weights falling within the acceptable limit of ±7.5%, as per pharmacopoeial guidelines. The average tablet weight was found to be approximately 243.3 mg, indicating good uniformity. Friability was measured using a Roche Friabilator. The percentage friability ranged from 0.20% to 0.98%**,** remaining well below the acceptable limit of 1%, thus confirming adequate mechanical strength. The hardness of the tablets was recorded at 4.5 kg/cm², indicating sufficient compressional force to maintain integrity during handling. Disintegration time was found to be 337 seconds, or approximately 5 minutes and 37 seconds, which complies with the general requirement for uncoated tablets. These findings confirm that the phytosomal tablets possess acceptable physical characteristics suitable for oral administration.

IV. CONCLUSION:

The COVID-19 pandemic brought global attention to the critical need for effective cardioprotective strategies, as individuals with cardiovascular comorbidities faced higher risks of severe complications. In light of this, the present study focused on developing a novel Phytosome-based formulation of *Dioscorea bulbifera* tuber extract, a plant known for its traditional use and rich content of bioactive compounds like diosgenin for the management of cardiovascular disease.

Through methods of extraction, enrichment, and standardization, a diosgenin-rich extract was obtained and subsequently formulated into phytosomes using the solvent injection method. The formulation was optimized using a Quality by Design (QbD) approach, to improve entrapment efficiency and reduce particle size. The dioscorea phytosomal formulation showed enhanced antioxidant activity via DPPH assay due to the contribution of flavonoids and phenolic acids present. The dioscorea phytosome formulation showed significant enhanced therapeutic angiogenesis in the CAM assay compared to the crude extract, indicating its potential to support vascular repair and reduce oxidative stress.

Furthermore, the dioscorea phytosomes were successfully developed into tablets through direct compression, which had acceptable pharmacopoeial properties, which enhances their practical applicability for oral administration.

Given the study results, this study not only confirms the cardioprotective potential of *Dioscorea bulbifera*-based phytosomes but also demonstrates the value of integrating herbal medicine with advanced drug delivery systems. The findings suggest that such phytosomal formulations can serve as promising, plant-based interventions for cardiovascular protection, especially relevant in pandemic scenarios where vascular complications are frequent. This work lays the foundation for future preclinical and clinical research aimed at establishing phytosomes as viable alternatives in phytopharmaceutical and pandemic-responsive healthcare.

V. CONFLICT OF INTEREST:

The authors declare no conflicts of interest concerning this investigation**.**

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