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Evaluation of Surfactant Extracted from *Asparagus nelsii Schinz* roots in Detergent Formulation

01 ABSTRACT

The rising demand for eco-friendly personal care products has prompted the exploration of biosurfactants, derived from renewable sources, as sustainable alternatives to synthetic surfactants in detergent formulations. This study investigated the extraction, characterization, and evaluation of surfactant properties from *Asparagus nelsii Schinz*, a plant native to southern Africa and traditionally valued for its medicinal properties and high foaming ability. The extraction process involved isolating saponin-based compounds from the roots of *A. nelsii*, known for their amphiphilic characteristics that facilitate effective surfactant behaviour. Fourier-transform infrared (FTIR) spectroscopy analysis revealed key functional groups, including hydroxyl, carbonyl, and alkyl chains, indicating the biosurfactant's amphiphilic structure essential for surface tension reduction and emulsification. Comparative analysis with synthetic sodium dodecyl sulfate (SDS) highlighted the biosurfactant's significant surface-active properties, showing superior reduction in surface tension at lower concentrations, which is advantageous in formulations aimed at mild yet effective cleansing. Emulsification index testing further demonstrated its capability in stabilizing vegetable oil emulsions, suggesting potential applicability in natural personal care products requiring stable emulsions. Performance assessments of formulated detergent demonstrated effective cleansing ability, with the biosurfactant achieving consistent lather formation and foam stability over time.

02 INTRODUCTION

Driven by growing consumer demand for sustainable personal care products, the detergent industry is increasingly replacing synthetic surfactants with plant-derived biosurfactants like saponins, which naturally excel at reducing surface tension, foaming, and emulsification. This study addresses a key research gap by evaluating the underexplored biosurfactant potential of *Asparagus nelsii Schinz*, an arid-climate southern African plant traditionally used in medicine. By isolating saponins from *A. nelsii* roots, the researchers assess their physicochemical properties—including surface tension reduction, emulsification, and thermal stability—and compare their performance against the commercial synthetic surfactant sodium dodecyl sulfate (SDS). Ultimately, the study seeks to determine if this natural extract can match the cleansing efficacy and foam stability of SDS under varying conditions to serve as a viable, eco-friendly alternative in detergent formulations.

03 EXPERIMENTAL

Sample Collection and Preparation

The roots of a nelsii were collected from Othika village in the Omusati Region of northern Namibia. The identity of A. nelsii was confirmed by the National Herbarium of Namibia (WIND), a division of the Namibian Botanical Research Institute (NBRI), report number 2021/431.
Reagents
All chemicals were of analytical grade. Sodium dodecyl sulfate (SDS), petroleum ether, chloroform, n-butanol, sodium hydroxide, sodium acetate, and sodium phosphate. Deionized water was used for all aqueous solutions and extractions.

Extraction of Biosurfactant

The root samples were washed, air-dried, and crushed into powder. Saponins from *Asparagus nelsii* roots were extracted following the method of Bezerra et al. [9]. Initially, the powder was degreased with petroleum ether and washed with chloroform. It was then boiled in distilled water to extract water-soluble compounds, cooled, and filtered. The filtrate was partitioned with butanol to isolate saponins, and the aqueous layer was discarded. The butanol fraction was treated with 1% NaOH to remove catechins and dihydrochalcones, then the butanol was removed by evaporation. The remaining biosurfactant was re-dissolved in distilled water and lyophilized to yield a saponin-rich extract for analysis.

Characterization and Surface Activity

The functional groups of the lyophilized biosurfactant were identified using the Perkin Elmer UATR spectrum two system through the Attenuated Total Reflectance (ATR) technique, which eliminates the need for KBr pellet preparation. Samples were placed on a diamond crystal, and spectra were recorded to identify functional groups in the transmittance band from 4000 cm-1 to 400 cm-1 Surface tension measurements were taken at 25 ℃ (±0.5℃) using a Sigma 700 Tensiometer across a concentration range of 0 - 0.55 mg/mL for both the biosurfactant and SDS, facilitating a direct comparison of their surfactant properties [10].

Stability and Surface Activity of the Biosurfactant

The stability and surface activity of the biosurfactant were evaluated using a modified method from Kiran et al. (2017)[12]. pH stability involved incubating a 0.2% (w/v) solution at 37°C in buffer solutions at pH 4.0–9.0. Salt stability was tested by dissolving the biosurfactant in distilled water with 0-12% NaCl and incubating at 37°C. Foam stability was measured at room temperature over 180 minutes. Temperature stability was assessed with a 1% (w/v) solution across 25°C to 65°C, with foam stability measured at each temperature.

Emulsification Activity with Different Hydrophobic Substrates

The biosurfactant's emulsifying activity with various hydrophobic substrates was assessed using modified established methods[11]. Each test involved mixing 2 mL of the biosurfactant with an equal volume of 0.1 M sodium acetate buffer and 1 mL of a hydrophobic liquid (e.g., vegetable oil, paraffin, crude oil, or engine oil). SDS served as a positive control. The mixtures were vortexed for 2 minutes and left to stand for 24 hours. Emulsifying activity was measured using the emulsification index (E24), following the method outlined by Lawrance et al. (2014)[11].
as follows:

$$E24\% = \frac{H_e}{H_t} \times 100$$

where He is the height of the emulsion layer formed and Ht total height of the mixture in the tub, both after 24 hours.

Hand-Washing Detergent Formulation and Detergency Evaluation

The detergent formulation utilized a biosurfactant as the main surfactant and foam booster, with added fragrance and preservatives for stability. *Moringa oleifera* seed oil, extracted following methods adapted from Oladipo and Betiku [13, served as a moisturizing agent. Foamability and stability were assessed through specific tests adapted from Lunkenheimer and Malysa (2003) [14], including mixing with distilled water and evaluating foam height over time. Both SDS-based and extract-based detergents were analyzed for their cleansing capabilities using engine oil-stained materials.

04 RESULTS AND DISCUSSION

FTIR Analysis

Saponins are amphiphilic glycosides characterized by a hydrophilic sugar and a hydrophobic aglycone. FTIR analysis identifies functional groups such as O–H, C–H, C=O, and C–O, confirming their surfactant properties. Hydroxyl groups improve solubility and interfacial orientation, reducing surface tension through hydrophobic interactions. The C–H stretching at 2924 cm^{-1} indicates alkyl chains that enhance nonpolar interactions and emulsification [15]. The C=O stretching at 1728 cm^{-1} suggests ester or carboxylic acid groups, optimizing the hydrophilic-hydrophobic balance for oil-water applications [9]. Additional bands at 1025 cm^{-1} and 1237 cm^{-1}, linked to ether groups, enhance molecular flexibility and stability, making saponins effective for industrial use.

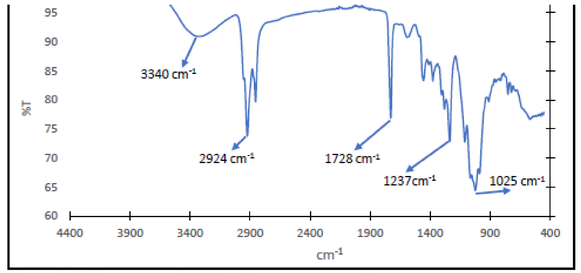

Figure 1: FTIR spectrum of the biosurfactant extract sample

Surface Tension and Surface Surfactant Adsorption

Figure 2 shows the concentration dependence of surface tension for SDS and the *A. nelsii* extract. Both surfactants reduce surface tension with increasing concentrations; however, the biosurfactant extract displays a more notable decrease, especially between 0 - 0.15 mg/mL, highlighting its superior surfactive properties at lower concentrations. This concentration is below SDS's critical micelle concentration (CMC) of roughly 8.2 mM (about 2.35 mg/mL). Statistical analysis revealed significant differences in surface tension reduction between the two surfactants at 0.025 to 0.2 mg/mL ($p < 0.05$). At concentrations above 0.225 mg/mL, no significant differences were noted ($p > 0.05$), indicating similar effects at higher levels. The surface tension reduces from 72 mNm-1 for pure water to approximately 40-45 mNm-1 at higher concentrations for both surfactants, suggesting their effectiveness converges at those levels.

The data was fitted to the linear Gibbs adsorption equation: Equation 2 is the linearised form of the Gibbs adsorption isotherm, derived from the thermodynamic relationship between surface tension and surface excess concentration.

$$\ln \gamma = -RT\Gamma_B \ln C + \text{Constant} \quad (2)$$

where is the solution surface tension, TB is the surface excess concentration,As is the area occupied per surfactant molecule at the interface ($Å^2$/molecule), C is the concentration of the biosurfactant extract or SDS, R equal to 8.314 $JK^{-1}mol^{-1}$ is the universal gas constant, and T is thermodynamic room temperature taken to be 298 K. The results of the in a linear relationship between γ and lnC, shown in Figure 3 and Supplementary Material Figure S2, further support the finding that the biosurfactant extract is more effective at reducing surface tension than SDS in the low concentration range studied. The steeper slope for SDS indicates a more rapid decrease in surface tension with increasing concentration, whereas the biosurfactant extract exhibits a less steep slope. There is, however, a crossover point (about 0.40 mg/mL) such that the surface tension of SDS becomes lower than that of the biosurfactant extract.

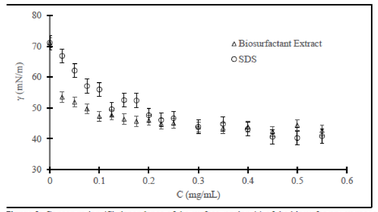

Figure 2: Concentration (C) dependence of the surface tension (γ) of the biosurfactant extract and SDS

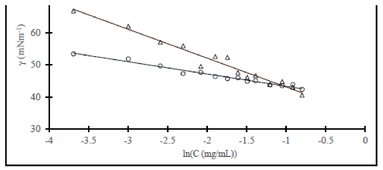

Figure 3: Surface tension data fitted according to the linear form of the Gibbs adsorption equation.

Table 1: Surface excess concentration and area per molecule of biosurfactant extract and SDS at 298 K

Parameter	Biosurfactant Extract	SDS
Surface Excess Concentration, Γ_B (molm^{-2})	3.60 x 10^{-6}	1.53 x 10^{-6}
Area per Molecule, A_s, Å^2/molecule	46.2	108.4

B

The estimated values of surface excess concentration, Γ_B, and the corresponding area per molecule, A , are calculated as $A_s = \frac{1}{\Gamma_B N}$ where N is Avogadro's number, for both the biosurfactant system and SDS are given in Table1 and Supplementary Material Table S2. The biosurfactant extract exhibited higher Γ and lower in the concentration range studied. This suggests a greater number of extract molecules accumulating at the airwater interface, consistent with its lower surface tension values and superior surface activity in the studied concentration range. This observation aligns with established principles of surfactant adsorption, where increased molecular accumulation at the air-water interface directly correlates with reduced surface tension [19,20].

Foamability and Stability of the Surfactant

The study investigated how added salt, pH, temperature, and time affect the foamability and stability of a biosurfactant extract. Increased NaCl concentration significantly reduced foam height from 26 mm (100% foam retention) at 0% NaCl to 7 mm (26.92% foam retention) at 10% NaCl, indicating that higher salt levels destabilize foam by screening electrostatic repulsions among surfactant molecules. While low NaCl concentrations minimize interactions among surfactants and maintain foam stability, elevated salt levels weaken these interactions leading to thinner foam films. This aligns with findings by Qiao et al. (2021) and Yekeen et al. (2017) [21,22], which noted reduced stability and accelerated bubble coalescence, respectively, with increased salt.

There exists a critical concentration range where ionic interactions can destabilize foam structure; excessive salt in detergent formulations can hinder foam performance and cleansing effectiveness [23]. Conversely, low salt concentrations can initially enhance foam stability, but adding too much salt results in rapid foam collapse beyond a specific threshold. Interestingly, at NaCl concentrations between 8-12%, foam height stabilizes, suggesting a limiting threshold where additional salt does not further destabilize the foam.

Additionally, the foam ratio data revealed a pH-dependence in foamability, with buffered pH values (4-9) maintaining a relatively constant foam ratio (45.06% to 49.49%). However, an unbuffered pH of 6.7 caused a drop to 38.24%, indicating that buffering significantly enhances the foam stability of the biosurfactant, with unbuffered conditions leading to a notable decrease in foamability [24].

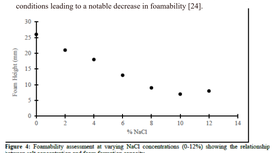

Figure 4: Foamability assessment at varying NaCl concentrations (0-12%) showing the relationship between salt concentration and foam formation capacity

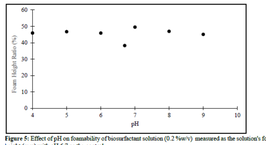

Figure 5: Effect of pH on foamability of biosurfactant solution (0.2 %w/v) measured as the solution's foam height (mm) with pH 6.7 as the control

The ANOVA results indicate a significant difference in foamability between buffered and unbuffered conditions ($p = 0.0345$). This suggests that buffering affects foam stability in biosurfactants, with a decline in foamability at pH 6.7 due to unbuffered conditions. Buffers may stabilize the biosurfactant's structure, preventing destabilization from pH fluctuations. Thus, maintaining buffered conditions is essential for optimizing biosurfactant performance in applications requiring consistent foamability.

The data presented in Figure 6 reveal that foam stability diminishes as temperature rises. At lower temperatures (25–35°C), foam height remains stable, indicating good foam integrity of the biosurfactant. However, as temperatures climb, foam height decreases, peaking at a minimum at 65°C due to reduced liquid thickness and accelerated drainage processes leading to foam collapse. This observation aligns with findings by Wang et al. (2017) [25], who noted that while increasing temperatures can enhance foam production, they generally compromise foam stability by facilitating faster drainage and shorter foam lifespans.

Additionally, the foam loss over time, tracked as foam ratio in Figure 7, serves as an indicator of stability and structural retention. Initially (0 minutes), foam loss is at 0%, reflecting complete retention. After 30 minutes, there is a 7.14% foam loss, marking the beginning of liquid drainage while showcasing relatively stable structural integrity during this early phase. At 60 minutes, foam loss rises to 17.86%, suggesting a moderate decline in stability likely due to drainage and coalescence where liquid exits the foam structure and larger bubbles collapse [26]. From 90 to 150 minutes, foam loss levels off at 25%, indicating a stabilization phase attributed to the biosurfactant's strong structural retention, despite ongoing degradation processes.

Finally, after 180 minutes, foam loss slightly increases to 32.14%, showcasing the biosurfactant's capability to maintain a significant portion of its structural integrity over time, indicative of its effectiveness in sustaining foam stability in various applications. This pattern, characterized by an initial moderate decline followed by a stable period and a gradual eventual decrease, highlights the biosurfactant's potential for consistent foam performance, crucial for systems where prolonged foam retention is necessary [27,28].

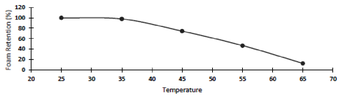

Figure 6: Effect of temperature on stability of foam resulting from a biosurfactant extract solution (1 %w/v)

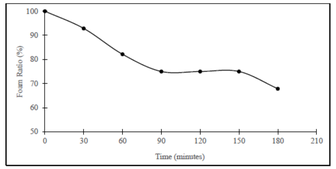

Figure 7: Effect of time on the stability of the foam of the biosurfactant extract (1 %w/v) solution

Emulsification Activity of the Surfactant

The emulsification index is crucial in detergency, as it measures how well emulsifying agents work in cleaning products by stabilizing emulsions, which aids in the removal of oils and greases. A higher index signifies better detergent performance in keeping oil droplets dispersed in water, thus enhancing cleaning efficacy.

Comparative analysis of emulsification indices reveals that *A. nelsii* biosurfactant extract and SDS perform comparably for engine oil, crude oil, and paraffin; however, the biosurfactant extract shows significantly higher efficacy for vegetable oil ($p = 0.003$). ANOVA results indicate no significant differences among the surfactants across various agents ($p > 0.05$), except for vegetable oil, reinforcing the extract's specialization for this application. The findings suggest that while both surfactants perform similarly for certain hydrophobic agents, the *A. nelsii* extract is particularly effective for vegetable oil emulsions, backed by significant statistical results from independent t-tests and various supplementary data tables.

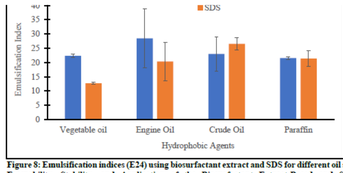

Figure 8: Emulsification indices (E24) using biosurfactant extract and SDS for different oil substrates

Foamability, Stability, and Application of the Biosurfactant Extract-Based and SDS-Based Detergents

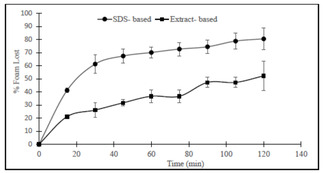

Figure 9: Foamability and Stability of the Formulated Detergent Over Time.

The comparison of biosurfactant-based and SDS-based detergent formulations highlights distinct differences in foamability and stability, as illustrated in the provided Figure 9. The SDS-based detergent initially produces a larger foam volume but suffers from a rapid reduction in foam stability, losing 41.19% of its foam by 15 minutes and 80.49% by 120 minutes. This decline is attributed to bubble collapse due to drainage and coalescence. Conversely, the biosurfactant extract-based detergent shows a steadier reduction in foam height, with only 21.11% loss at 15 minutes and 52.22% at 120 minutes, implying better structural retention. This slower foam degradation indicates that the biosurfactant enhances foam stability by minimizing bubble coalescence and liquid drainage, resulting in a more durable foam.

In terms of cleaning efficacy, both detergent formulations were tested on black engine oil-stained materials, with distilled water serving as a control. The biosurfactant-based detergent demonstrated cleansing ability (Figure 10) comparable to that of the SDS detergent, effectively reducing stains on materials, as shown in the results. Overall, while the SDS formulation offers superior initial foamability, the extract-based detergent provides a more sustained foam presence and effective cleaning performance over time.

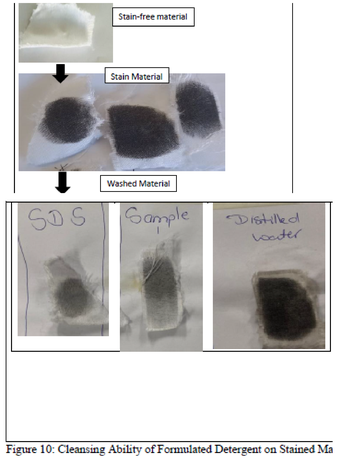

Figure 10: Cleansing Ability of Formulated Detergent on Stained Ma

05 CONCLUSION

Taken together, these findings form a coherent performance narrative for the *A. nelsii* biosurfactant. The amphiphilic functional group profile confirmed by FTIR — combining hydrophilic O-H and C-O groups with hydrophobic C-H alkyl chains — directly underpins the superior surface tension reduction at low concentrations, reflected in the high surface excess concentration (Table 1) and efficient interfacial packing. This surface activity translates into effective foam generation and a robust stability plateau, sustained by viscoelastic interfacial film formation. Emulsification performance is broadly comparable to SDS, with a substrate-specific advantage for vegetable oil attributed to functional group polarity matching. The formulated detergent successfully integrates these properties, demonstrating that *A. nelsii* biosurfactant is a functionally viable and structurally coherent alternative to synthetic surfactants..

Author Contributions: Conceptualization - SA and HMK; Data Collection – TNAN and SA; Writing original draft-TNAN and SA; Review and editing – HMK and SA; Final Compilation of Manuscript – TNAN, SA, and HMK
Conflict of Interest: The authors declare that no conflict of interest exists.
Data Availability: The authors declare that the relevant data supporting the findings of this study are available in this paper.
AI Use Disclosure: AI tools were utilized to assist with data analysis, wording, phrasing, and grammar review during manuscript preparation. The author accepts full responsibility for all interpretations, conclusions, and final content

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