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Evaluación del poder de detergencia de un extracto procedente del fruto de la Parapara (*Sapindus saponaria)*.

Evaluation of the detergency power of an extract from of the fruit of Parapara (*Sapindus saponaria)*

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Abstract

In plants there are secondary metabolites that present many properties that favor their industrial use since they possess interfacial activity, among these are the saponins that are characterized by the production of foams in aqueous solutions. In this research an extract of parapara (Sapindus saponaria) was obtained by total reflux, where the FTIR spectrum showed carboxylic and hydroxyl groups, in addition to cyclic and aliphatic chains, agreeing with the groups of some metabolites such as flavonoids and saponins among others. The Salkowski test showed the presence of saponin and the Liebermann-Burchard test determined the presence of triterpenoid structures. The extract in aqueous solution decreases the surface tension to 31 dynes/cm, has a CMC 0.17 % (m/v), and an area per molecule of 0.67 nm2 at the interface. Its HLB of 9.4 indicates that it has emulsifying and foaming properties in the aqueous phase. Detergency tests on cotton showed a low removal in static (< 50%) but with agitation the removal is higher than 50%. In the case of cotton-polyester, the percentages of removal were higher.

Keywords: Natural surfactant; detergent; Sapindus saponaria extract

Resumen

En las plantas existen metabolitos secundarios que presentan muchas propiedades que favorecen su uso industrial ya que poseen actividad interfacial, entre estos se encuentran las saponinas que se caracterizan por la producción de espumas en soluciones acuosas. En esta investigación se obtuvo un extracto de la parapara (Sapindus saponaria) por reflujo total, donde el espectro FTIR mostró grupos carboxílico e hidroxilo, además de cadenas cíclicas y alifáticas, concordando con los grupos de algunos metabolitos como flavonoides y saponinas entre otros. Se realizó la prueba de Salkowski que mostro presencia de saponina y la prueba de Liebermann-Burchard determinó la presencia estructuras triterpenoide El extracto en solución acuosa disminuye la tensión superficial a 31 dina/cm, posee una CMC 0,17 % (m/v), y un área por molécula de 0,67 nm^2 en la interfase. Su HLB de 9,4; indica que tiene propiedades emulsionante y espumante en fase acuosa. Las pruebas de detergencia sobre algodón mostró una baja remoción en estático (< 50%) pero con agitación la remoción es superior al 50%. En el caso de algodón-polyester, los porcentajes de remoción fueron superiores.

Palabras clave: Surfactante natural; detergente; Extracto Sapindus saponaria.

1 Introduction

Detergents are a key group of products of industrial importance and are intended for general use, for the removal of dirt, whether for the maintenance of environments, textiles or skin. These are generally manufactured synthetically, including surfactants, emulsifiers, stabilizers, and other additives which could affect health and cause environmental pollution. (Do *et al*., 2019). Currently, the trend is the production of detergents of natural origin, replacing some substances with compounds present in nature that allow the removal of dirt, but without generating pollution in bodies of water.

Secondary metabolites are found in plants, which are organic compounds that perform non-essential functions in them. Within these metabolites there are families of polyphenols, saponin, among others, which can have an amphiphilic nature and are responsible for the properties of adsorption, emulsion, foaming, among others (Sochacki & Vogt 2022). The *Sapindus saponaria* tree is a species that contains many of these compounds, with saponin being found in a greater proportion, which is a compound that has the property of forming foam when placed in aqueous solution (Thomas *et al*., 2010). The presence of saponins offers excellent physicochemical and biological properties that make them a promising source of natural surfactants, both for research and commercial purposes. (Lozsan *et al*., 2017).

Saponins are secondary metabolites produced by plants belonging to more than 500 species (Rai *et al*., 2021). In terms of chemical structure, saponins result from their amphiphilic structure, which consists of a hydrophobic backbone known as aglycone (or genin) and hydrophilic sugar groups (glycone). The two glycoside-forming parts are the basis for the structural deviation of saponins in nature. The glycone part consists of one or more sugar chains, which are then linked to the aglycone through a glycosidic bond. The O-glycosidic bond separates the two structural parts of saponins. (Sochacki &Vogt, 2022). Chemically, they are amphiphilic glycosides with hydrophilic glucones composed of sugar units linked to hydrophobic aglycones (steroids or terpenoids) (Rai *et al*., 2023). Due to their amphiphilic nature, they tend to form micelles and reduce the surface tension in water and manifest their foam properties (Lorent *et al*., 2014).

In terms of non-food applications, saponin extracts can be used in shampoo and liquid detergents (Do et al., 2019), being natural compounds that can be used to replace synthetic detergents (Güçlü-Üstündağ & Mazza, 2007; Rai *et al*., 2021). The objective of this research was to evaluate the interfacial properties and the detergency power of the crude extract obtained from the parapara fruit (*Sapindus saponaria)* to be incorporated into detergent formulations, as a substitute for highly branched alkyl chains, which makes synthetic detergents poorly biodegradable.

2 Experimental Procedure

2.1 Extract of the fruit of the parapara

Sapindus saponaria fruits were harvested when ripe, the pericarp was manually separated from the seed, and dried in the sun for 7 days. The dried material was ground to a particle size of 180 µm. Proximal analysis of the fruit was performed using near infrared spectroscopy equipment (NIRS-2500F). The extraction was carried out at total reflux in a solid-liquid ratio of 1:10 at 70°C/3h, using 70% (v/v) ethanol as solvent. After extraction, it was filtered through Whatman No. 5 paper, and then concentrated using an evaporator (Büchi B-480 Labortechnik AG, Switzerland) at 78 °C. The extract was subjected to the Salkowski test for saponin and the Liebermann-Burchard test to identify the type of saponin

2.2 Physicochemical characterization of the extract

Using a PARAGON 1000PC model spectrometer, Fourier Transform Infrared Spectrophotometry (FTIR) was performed, and pH was also determined in 1% solution (Accumet AB-30 pH meter) and its solubility in alcohols of different polarities (ethanol to heptanol). The solubility test consisted of placing 2.5 mL of alcohol in a test tube and then adding 0.5 mL of crude extract, shaking gently and verifying whether or not it dissolved

2.3 Interfacial characterization of the parapara extract.

The surface tension was determined at room temperature using the Du Noüy ring method, using a Cole Palmer model 21 tensiometer, the platinum-iridium ring with a circumference of 6 cm. All the solutions are prepared and allowed to stand for 24 h. The result is plotted as surface tension versus the logarithm of the solute concentration. The changes of the slope allow obtaining the critical micellar concentration (CMC). The Gibbs adsorption equation (Eq. 01) was used to determine the interfacial parameters (area per molecule, surface excess, tension at the CMC), where ϒ surface tension, C is the solute concentration, Γ surface excess, R constant gases, T absolute temperature (Dabestani *et al*., 2021).

$$\Gamma = -\frac{d\gamma}{RTLnC} \quad \text{(Eq. 01)}$$

For the determination of HLB, the methodology proposed by Salager & Anton (1983) was followed. Known HLB surfactants such as Tween 80 and 85, Span 20 and 80 were used as reference, these were mixed in different proportions (w/w), NaCl (1%) and a mixture of secbutanol/pentanol alcohol were added. A different ratio of the 4 surfactants is placed in each tube to obtain different

HLB values ranging from 4.3 to 15.0. Once the reference HLB is obtained, a 20% proportion of the surfactant mixture is removed and replaced with the test sample. They are mixed gently and allowed to stand at room temperature. Once the diffusion process was completed, the optimum system was selected, represented by a three-phase system where the volumes of water and oil are equal. The presence of surfactants in each phase of the formulated tubes was verified by means of the Tyndall effect. The HLB was calculated according to the rule of mixtures (Eq.0 2).

$$HLB_{muestra} = \frac{HLB_{ref} - \frac{HBL^*}{0,8}}{0,2} \quad \text{(Eq.02)}$$

A foamability test was also performed at concentrations of 0.1 %, 0.3 %, 0.5 %, 1 % and 3 %, 5 mL aliquots were taken, which were added in 30 mL graduated tubes, shaking each tube vigorously for 30 s, the height reached and the collapse time of 1 mL of the foam were taken. The test was performed in duplicate.

2.4 Detergency power of parapara extract

The detergency test was performed using the International Association for Soaps, Detergents and Maintenance Products (A.I.S.E) as a reference. The percentage of stain or soiling removal was considered, placed on two cotton-polyester (TAP) and cotton (TA) textiles, which are capable of retaining dirt in their fabric. These are soiled with two common stains, mud and coffee. The percentage of cleanliness or soiling removal was determined as proposed by Pradhan & Bhattacharyya (2017) who determined the percentage removal according to (Eq. 03).

$$\%Removal = \frac{W_2 - W_3}{W_2 - W_1} * 100 \quad \text{(Eq.03)}$$

w_1: initial weight cloth; w_2: weight of the soiled cloth; w_3: weight after washing

Tests were carried out in duplicate under static (soaking) and agitation conditions. Once the washing process was completed, the percentage of removal was recorded. The substrate (cloth) is checked using a portable optical microscope. A commercial surfactant used to formulate surface and textile detergents with an HLB of 10.8 was used as a reference**.**

3 Discussion and Results

3.1 *Characterization of parapara fruit extract*

Proximal analysis of the parapara fruit showed 12.38% moisture, 2.07% ash, 2.41% crude fat, 17.80% starch. The crude extract obtained from this fruit (ECP) presented an acid pH (4.89), with brownish color, viscous appearance and sweet aroma, the latter is attributed to the degradation of the sugar present in the same, which represent approximately 50% of the weight of the molecule. The *Salkowki test* confirmed the presence of saponin in the extract when it turned from reddish to orange, while the Liebermann-Burchard test established that the type of aglycone corresponds to a triterpenoid. Schreiner *et al*. (2022) mention that aglycones (or sapogenins) compose the hydrophobic parts of saponins and may include steroidal or triterpenoid main chains, predominantly glycosides with one or more sugar residues linked to the aglycone by glycoside bonds, among the most common monosaccharides are D-glucuronic acid, L-rhamnose, D-glucose, Dfucose, L-arabinose, D-xylose, D-apiose and D-galactose.

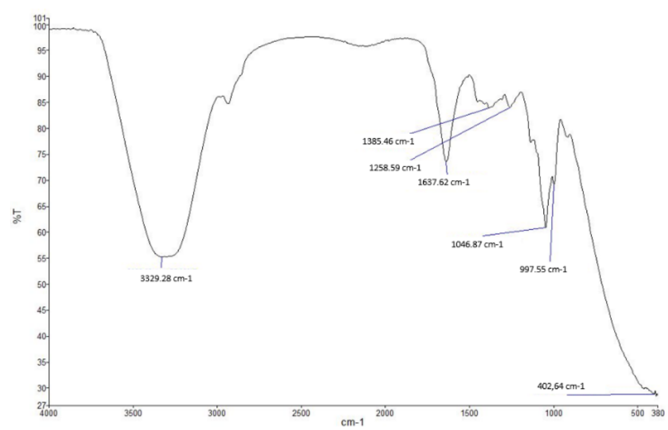

Fig. 1. FTIR spectrum of the crude extract of parapara

Figure 1 shows the FITR spectrum of the ECP, which presents an absorption band at 3329 cm-1, associated with the stretching of O-H groups of carboxylic acids and hydroxyl groups. The band between 2922 cm-1 and 2854 cm-1, are asymmetric and symmetric stretching of the C3sp, this indicates the presence of saturated carbons, the band 1637 cm-1, corresponds to a stretching of the C=O of the carbonyl group, the signal between 1385 cm-1 and 1258 cm-1, correspond to the out-of-plane bending of the CH2 and CH3 indicating the presence of alkyl chains. C-O-C band at 1046 cm-1, which corresponds to oligosaccharide binding to sapogenins, and at 997 cm-1, corresponds to what is known as rocking, and indicates the presence of at least 3CH2 consecutive to give this signal, typical of geminal methyl groups in cyclic chains.

Table 1. Solubility test of the parapara extract**.**

Solvents	Water	Etanol	Propanol	Butanol	Pentanol	Hexanol	Heptanol
Solubility	+++	+++	+	---	---	---	---

(+++) Soluble (+) Partially soluble (---) Insoluble

Jarzebski *et a*l. (2020) comment that for this type of extract the signals observed may also correspond to compounds such as flavonoids, phenolic derivatives, saccharides, anthocyanins, steroidals and triterpenes.

Table 1 shows the solubility of ECP in different solvents. It is soluble in water and ethanol, and insoluble in alcohols with a higher number of carbon atoms. The solubility in these solvents can be attributed to the OH groups in its structure, which allow the formation of hydrogen bridges with the solvent. Yoong *et al*. (2014) mentioned that saponins are soluble in polar solvents**.**

3.2. Interfacial characterization

Surface tension is related to the amount of adsorbed amphiphile and the nature of the interfacial layer. This tension is a parameter that controls the stability of foams, emulsions, etc. Figure 2 shows that with increasing concentration there is a reduction of the tension until the critical micellar concentration (CMC) is reached, where the interface is saturated and the tension tends to be constant. The strain at the CMC of the ECP was 48 dynes/cm, which evidences its action as a detergent, due to the mixture of compounds present in the extract. Pradhna & Bhattacharyya (2017) and Schmitt *et al*. (2014) mention that the saponins exhibit a surface tension reduction of up to 40 mN/m**.**

The formation of micelles in aqueous solution and the reduction of surface tension are the characteristic features of any surfactant, responsible for its versatile industrial applications as detergent, foaming agents, emulsifiers, solubilizers, etc. (Sochacki &Vogt, 2022). On the other hand, interfacial parameters were determined finding a CMC of 0.17 % (m/v) and an area per molecule 0.67 nm^2, these values may vary depending on the Sapindus family. Pagureva *et al.* (2016) reported for a triterpenoid saponin an area per molecule between 0.4 and 0.6 nm^2.

The HLB of the ECP was 9.4, this value on the Griffin scale indicates that it has properties to form oil/water (o/w) emulsions and good foaming in the aqueous phase. However, there is little information on HLB values for this species. One of the relevant properties of the presence of saponins in an extract is its ability to foam in aqueous solutions. When studying the foaming capacity of ECP, it was found that the height depends on the concentration, and the higher the concentration, the greater the foam column, which ranged between (10.6 - 18.2) mL. The reduction of the surface tension of the water causes foam to form. The presence of saponins in the extract allows the decrease of surface tension and increase the foamability of aqueous solutions, even at low concentrations (Chen *et al*., 2010).

The foam generated with the dissolution of the extract is not very stable. The highest stability was found at a concentration of 0.5% (m/v) with a collapse time of 58 sec. Although foam generation has little relation to detergency capacity, it is an important criterion for evaluating a detergent (Wisetkomolmat *et al*.,2020), since it is associated with the reduction of air-liquid interfacial tension (Amankeldi *et al*., 2018). Stability also depends on the foam generation method, morphology and, consequently, decay time (Santinia *et al*., 2019).

A comparison was made of the ECP with a commercial surfactant (SC), which has a CMC of 0.015 % (m/v), and a surface tension of 31 dynes/cm, this value coincides with that shown in its data sheet (30 dyne/cm). The area per molecule occupied by this molecule is 0.755 nm2. When the interfacial parameters were compared, it was found that SC decreases the surface tension as much as ECP, however, its CMC is almost ten times lower than ECP. It produces foam of higher stability than those obtained with ECP.

3.3 Detergency power of parapara extract.

The removal of a soil depends on the type of surface, the soil to be removed and the cleaning condition. The cleaning action of a surfactant is associated with its ability to reduce surface tension and form micelles in aqueous media (Rakowska, *et al*.,2017). The results obtained for removing common soils showed that DBS solutions is able to remove the soil in the conditions of static washing and agitation, in the selected ones, the removal percentages are shown in Table 2. The removal for mud and coffee for the ECP in static is below 50%, while a better performance in static of the SC with respect to the ECP is evidenced.

On the other hand, when washing was performed with constant agitation, it was found that it favored cleanliness being higher than 50% removal. This increase in removal is attributed to the turbulence generated during agitation, which favors mass transfer, suspension of the dirt and dragging of the same (George & Raymon, 2016). On the other hand, Yakimchuk *et al*., (2004) mention that, above the CMC, micellar aggregates remain suspended in water with the hydrophilic head portions forming the outer layer and the hydrophobic tails forming the core of the micelles. Dirt is trapped within the core by these aggregates and solubilized and then easily removed with water.

A microscopic analysis of the textiles was carried out to verify the removal of the mud, these are compared with the reference before washing as shown in Figure 2, where it is observed how the dirt is removed and the fiber of the textile does not show any damage. This shows the detergent power of ECP and its efficiency, so it could be considered for inclusion in the formulation of cleaning and hygiene products, with the advantage of being a biodegradable product since it is composed mostly of sugars.

Table 2. Percentage of impurity removal on textiles.

		Mud		Coffe	
	Textile*	**Static**	**Agitation**	**Static**	**Agitation**
ECP	CT	20,89	54,76	28,30	68,07
	CPT	47,00	63,83	31,65	71,52
SC	CT	48,06	56,00	38,91	76,77
	SPC	49,23	67,11	45,50	76,21

*CT: Cotton textile CPT: Cotton-polyester textile

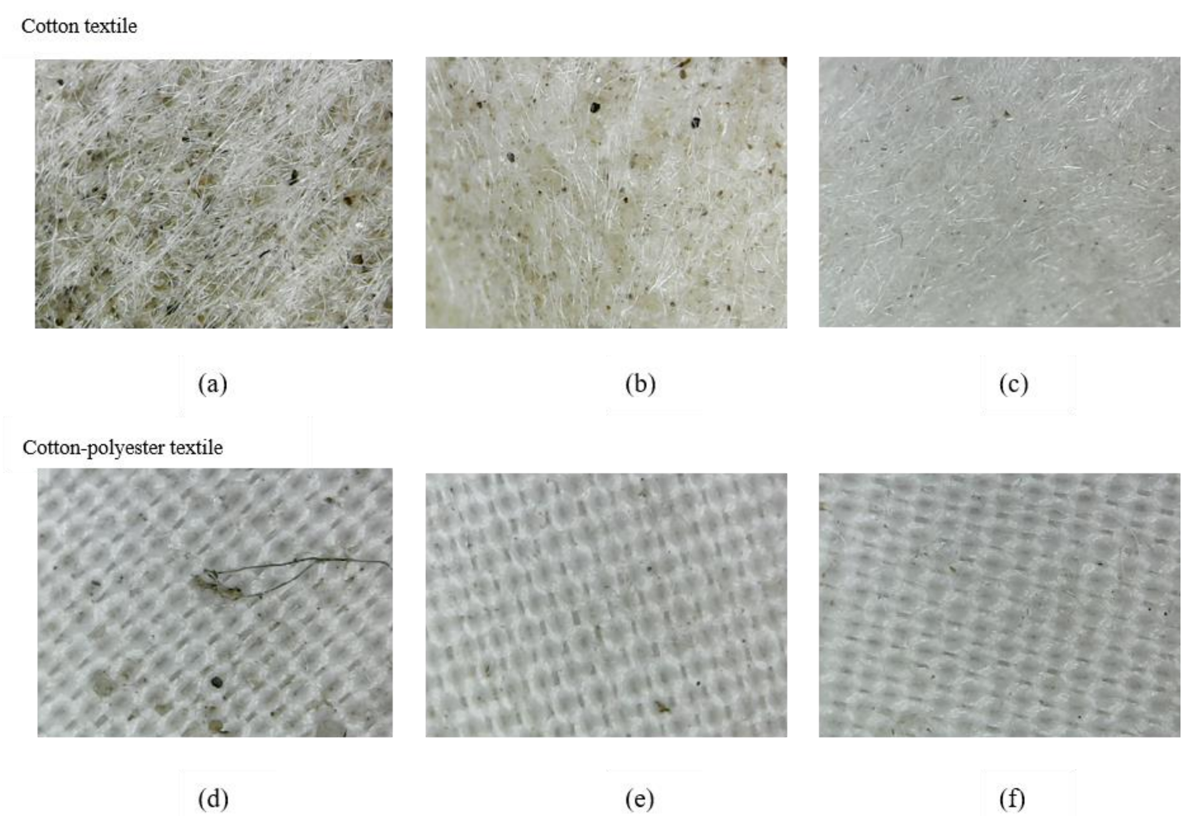

Fig. 2. Mud removal using ECP (a,d) Dirty mud reference, (b,e) static washing, (c,f) agitated washing**.**

4 Conclusion

The extract extracted from parapara contains a mixture of secondary metabolites including flavonoids and saponins, the latter being evidenced by the formation of foams in the aqueous phase. The FTIR showed the presence of carboxylic and hydroxyl groups, in addition to aliphatic cyclic chains, corresponding mostly to sugars, these groups confer interfacial properties and make it soluble in water. The ECP achieves a significant decrease in surface tension and foam formation in aqueous solutions, in addition to a good detergent power to remove soils such as mud and coffee when used in washing with and without agitation, its action is comparable to a commercial surfactant.

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