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BACHELOR'S THESIS

Basic Viability of Saponin from Soapwort as a Sustainable Laundry Detergent

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Abstract

Many manufacturers of laundry detergents that contain only biodegradable ingredients from renewable sources frequently use surfactants based on palm oil, although it is widely known for its sustainability problems.

Saponins, in contrast, are a group of natural surfactants that, while seemingly often overlooked as detergents, can be found in various plants including soapwort, *Saponaria officinalis*, which contains a considerably high amount of saponin and is common in Europe.

The basic viability of saponin from soapwort as a sustainable laundry detergent is investigated in this two-part interdisciplinary study. Part I includes an accessible extraction process and a simple laundry experiment. Part II presents a detailed analysis of the actual soil conditions at three locations with large *Saponaria officinalis* plants in a semi-natural habitat.

While the soapwort extracts indicated clear surfactant activity, the results of the laundry experiment using these extracts were inconclusive, showing at most only marginal effects even in the reference treatments. Follow-up studies should improve on the experimental design and employ sophisticated methods for quantification.

The analysis of the soil conditions, however, revealed a gravelly sandy gley with anthropogenic interference and poor nutrient conditions, showcasing that the plant is generally undemanding and seemingly a viable candidate for systematic cultivation.

Keywords: Saponin, Surfactant, Detergent, Soapwort, Saponaria, Saponaria officinalis, Sustainable, Sustainability, Soil

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General introduction

The cultivation of oil palms, *Elaeis guineensis*, as a profitable source of oil for food, industrial, and energy purposes (Pacheco et al. 2017) is a widely known sustainability problem associated with deforestation, soil degradation, and land grabbing (Vijay et al. 2016; Mohd Noor et al. 2017).

While most palm oil is produced for the food sector (Pacheco et al. 2017, 5– 6), laundry detergents often contain a mixture of plant-derived, animal-derived, and petroleum-derived surfactants (see Davis et al. 1992, 37–39), and palm oil is frequently used as the main plant source for oil-based surfactants due to its status as an inexpensive vegetable oil with unique qualities (cf. Hack 2013).

Even though "green" or "eco-friendly" cleaning agents, personal care products, and laundry detergents usually contain only biodegradable ingredients from renewable sources, many of the manufacturers commonly use surfactants based on palm oil and rationalize their choices by implying that palm oil is virtually irreplaceable and shifting the discussion towards the need for improving the sustainability of palm oil production (see Hack 2013). Although there are organic palm oil operations and also initiatives like the Round Table for Sustainable Palm Oil (see Mohd Noor et al. 2017, 2), NGOs point out that these are mostly greenwashing endeavors or at least ill-fated efforts and that an effectively sustainable production of palm oil is hardly feasible, at least on a larger scale (Rettet den Regenwald e. V. 2011; cf. Mohd Noor et al. 2017).

While it is possible to obtain biodegradable surfactants from coconut oil instead of palm oil (Prades, Salum, and Pioch 2016, 2; see Hinrichsen 2016) and – differences in short-term efficiency aside – this seems to be an improvement with regard to land use practices (see Prades, Salum, and Pioch 2016, 1–2), replacing one tropical plant source with another one can hardly be deemed an ideal solution from a wider sustainability perspective. Similar concerns can be applied to importing soapnuts, *Sapindus saponaria*, from India to Europe.

Animal-derived surfactants are not considered at all in this thesis since animal exploitation is not deemed ethically sound, neither from a secondary, strong sustainability (Michelsen and Adomßent 2014) perspective (cf. Steinfeld et al. 2006) nor from a primary perspective of fundamental ethics (see Francione and Charlton 2015).

It should be noted that not only coconut oil, but also other vegetable oils, e.g. olive oil, can be used to synthesize biodegradable surfactants (see Makkar, Cameotra, and Banat 2011; see Santos et al. 2016; see Hanno et al. 2015). However, the utilization of oils and sugars from food plants, although some of these cases might be promising, is out of scope for this thesis.

As for Indian soapnuts, their use as a detergent stems from their high saponin content (Pelegrini et al. 2008, 922). Saponins, however, can be found in many different plants to varying degrees (Sparg, Light, and van Staden 2004; cf. Schwarzbach 2004), and one plant with a considerably high saponin content is soapwort, *Saponaria officinalis* (see Fig. 1), which is a common plant species in Europe (Khela 2012).

Apparently, the extraction of saponin from soapwort used to be local ecological knowledge (Martín-López and Montes 2015, 3) in certain parts of Europe (Svanberg and Łuczaj 2014, 168 & 194; see Truttwin 1920, 35), and it has been reported that soapwort extract has been used to clean the fabric of museum pieces (Motz and Kinder 2012). The current use of saponin as a detergent in general and of soapwort extract in particular, however, seems to be a niche phenomenon limited to hobbyist circles (see R. Blume and Meiners 2017; see Motz and Kinder 2012) and a small number of commercial distributors of "green" or "eco" cleaning products (see Weber 2010; cf. Triaz GmbH 2018; cf. memo AG 2018; cf. Hess 1956).

This thesis aims to incorporate the examination of saponin from soapwort as a detergent into a broader sustainability framework from a perspective of empirical science. It is a two-part interdisciplinary study investigating the basic viability of saponin from soapwort as a sustainable laundry detergent: To what extent can saponin be extracted from soapwort with a view to its potential use in laundry detergents, and how do soapwort extracts from different sources compare to other plant-based surfactants in this context? How do abiotic and habitat conditions relate to *Saponaria officinalis* being described as undemanding, and what does this imply for sustainable cultivation and large-scale production?

Part I, the experimental chemistry part, includes an accessible extraction process and a simple laundry experiment. Part II, the soil ecology part, presents a detailed analysis of the actual soil conditions at three locations with large *Saponaria officinalis* plants in a semi-natural habitat. The results and the implications that might be inferred from them as empirical examples are embedded into an overarching discussion.

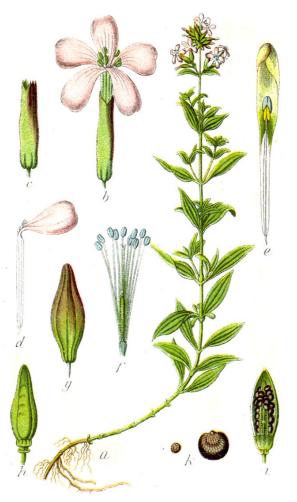


Fig. 1: Soapwort (SAPONARIA OFFICINALIS). Illustration by Jacob Sturm (Sturm 1796)

I. Saponin from soapwort as a detergent

1. Introduction

Saponaria officinalis is a herbaceous perennial plant from the pink family, Caryophyllaceae, (Hilty 2017) with a creeping rhizome (Meyer 1911, 135).

The whole plant is high in saponins (Jia, Koike, and Nikaido 1998; Koike, Jia, and Nikaido 1999), with a saponin content of approximately 3–5 % in the rhizomes in particular (R. Blume and Meiners 2017).

Saponin is an umbrella term for many structurally different glycosides, although they can usually be categorized into either steroidal or triterpenoid saponins (Sparg, Light, and van Staden 2004, 219–20).

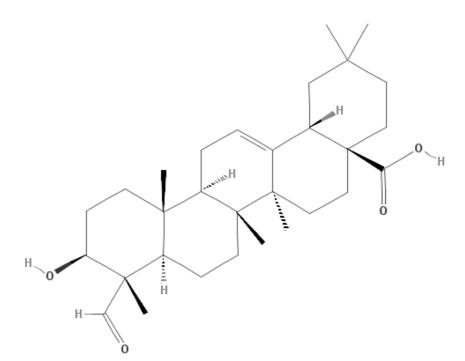


Fig. 2: 2D molecular structure of gypsogenin, $C_{30}H_{46}O_4$ (NCBI 2018a), one of the main triterpenoid saponins found in soapwort (SAPONARIA OFFICINALIS)

Traditionally, saponins are classified as such due to their shared physical and biochemical properties, e.g. surfactant characteristics, hemolytic action, and toxicity in fish (Tschesche and Wulff 1973, 462).

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The saponin group in *Saponaria officinalis* consists mostly of several different triterpenoid saponins (Sparg, Light, and van Staden 2004, 230; Jia, Koike, and Nikaido 1998; Koike, Jia, and Nikaido 1999), see for example Fig. 2.

A basic approach for the extraction from different soapwort samples is described below, followed by a simple laundry experiment that aims to compare the extracts with other plant-based surfactants.

In addition, the pH values of the extracts and the reference surfactants were measured and are presented for further context.

2. Material and methods

2.1. Extraction

Samples were taken from different soapwort sources. Above-ground (ag) and below-ground (bg) biomass was collected from cultivated plants (C), field plants that were not in blossom (F), and field plants that were in blossom (Fbl), resulting in six different kinds of extracts (C ag, C bg, F ag, F bg, Fbl bg). This was done to allow for the examination of possible differences in detergent behavior that might indicate varying levels of saponin between different parts and growth conditions of the plants.

The five potted Saponaria officinalis plants that were used for the cultivated plant samples, see Fig. 3, were acquired from an organic gardening operation (Brunkhorst and Brunkhorst 2018). The plants were 16 months old and had been grown in polypropylene pots with a volume of approximately 0.5 L each. The potting soil had the following properties (according to the supplier): a medium texture (0–25 mm, 20 kg/m³ ground clay), a water capacity of 73–78 % by volume, a pH value of 5.5 (in CaCl₂ 1:2.5 v/v), 480–620 mg/L N, 250-450 mg/L P₂O₅, 350–700 mg/L K₂O, and 100–200 mg/L Mg.

For the field samples, parts of three large plants from a semi-natural habitat, see II. 2.1, were collected.



Fig. 3: Cultivated soapwort (SAPONARIA OFFICINALIS) plants

The above-ground biomass and the below-ground biomass of each kind of sample were chopped up separately in a food processor (Moulinex *La Moulinette*, DP700, 1000 W). 25 g of each of the six resulting samples were weighed (balance: G&G pocket scale, model No. LS2000, capacity: 2000 g, d = 0.1 g) into a 500 ml amber bottle each, and 50 g of 70.55 % ethanol (w/w) (see Tschesche and Wulff 1973, 465–66) were added to each bottle. The bottles were closed and left to sit for 10 days with intermittent shaking in a circular motion for approximately three seconds twice a day (cf. Cheok, Salman, and Sulaiman 2014, 21).

Subsequently, each solution was filtered through a paper filter (Profissimo coffee filter, size 4) into a beaker (cf. R. Blume and Meiners 2017), and the filtrate

was transferred to semitransparent polyethylene bottles.

Unfortunately, the saponin content of the extracts could not be quantified, as a high-performance liquid chromatography (HPLC) or similar method of quantification was not available due to resource and time constraints. Therefore, the saponin content was only estimated, based on a general percentage of 3-5 % (see R. Blume and Meiners 2017) \approx 4 % and conservatively assuming an effective extraction of only 50 % to be on the safe side.

2.2. Laundry experiment

The experiment included nine different treatments: a blank treatment of 70.55 % ethanol (w/w), extracts 1–6 (C bg, C ag, F bg, F ag, Fbl bg, Fbl ag), and two reference surfactants.

The substances chosen as reference surfactants were coco glucoside (55 % active substance) and cocamydopropyl betaine (30 % active substance), since both are plant-based biosurfactants (see NCBI 2018b, 2018c; cf. Prades, Salum, and Pioch 2016, 2). These were diluted with 70.55 % ethanol (w/w) to adjust their respective percentages to the estimated saponin contents (see 3.1) of the soapwort extracts, with the estimates being used in lieu of an actual quantification (see 2.1).

For each treatment, a batch of three times three different replicable stains of balsamic vinegar (Rapunzel Aceto Balsamico di Modena I.G.P., 6 % acid), tomato juice (Alnatura Tomaten Saft Direktsaft, 99.6% tomato juice, sea salt), and rapeseed oil (Alnatura Raps Öl nativ, virgin rapeseed oil) were applied to a sheet of cotton satin (undyed satin, GOTS, 100 % cotton, 157 cm width, 135 g per lineal m) with five drops from a pipette per stain, see Fig. 4.

The first run of the experiment consisted of letting the stains settle for ten days before applying five drops of the respective treatment to each stain per batch, waiting for 10 minutes, and then processing the batch in a washing machine (Bauknecht WATE 9585) at 30° C in a delicate cycle with 400 rpm for 35 minutes.

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The second run consisted of letting the stains settle for only five hours before pouring 10 g of the respective treatment into the washing machine's detergent chamber instead of applying it directly to the stains and then processing the batch at 30° C in a delicate cycle with 400 rpm for 35 minutes.

Each batch was treated and washed separately in both runs. The pieces of cotton satin were let air-dry on a laundry rack overnight and then scanned with an image scanner (Epson GT9300 using xsane 0.999) at full color range, 300 dpi resolution, gamma: 0.85, brightness: -100.00, and contrast: 100.00.



Fig. 4: Replicable stains on undyed cotton satin. From top to bottom: Balsamic vinegar, tomato juice, and rapeseed oil

The scanned images were inspected visually for any possible cleansing effects compared to the blank treatment of mere ethanol (70.55 % w/w) as a baseline, and the observations were rated accordingly on an ordinal scale with five steps from no observable effect • to highly increased stain removal +++.

2.3. pH values

The pH values of the soapwort extracts, the diluted reference surfactants, and 70.55 % ethanol (w/w) were measured with a pH meter (inoLab WTW series pH/ION 735, pH-Electrode SenTix 41 pH $0..14 / 0..80^{\circ}$ C, stored in 3 mol/L KCl) that had been pre-calibrated with buffer solutions (pH 4.01, HI 7004, H906.1; pH 7.01, HI 7007, H908.1).

Each pH value was measured three times, and the electrode was rinsed with demineralized water and dried off with a paper towel between measurements. The respective pH values of each substance were averaged.

3. Results

3.1. Extraction

The colors of the extracts ranged from amber to dark drab tones, see Fig. 5. Every extract showed clear surfactant activity, i.e. pouring a stream of water onto a drop of the extract resulted in visible foaming.



Fig. 5: Soapwort extracts

On average, the extraction yielded a mass of 23.5 g per filtered extract, see Tab. 1. Estimating the total extractable saponin from each sample at $3-5 \% \approx 4 \%$ and conservatively assuming an effective extraction of only 50 % leaves the estimated extracted saponin at 1 % of the sample mass and thus at 0.23 g on average.

					Tab.	1: Extr	action	results
Extra	act:	1	2	3	4	5	6	Average
Sam	ple:	C bg	C ag	F bg	F ag	Fbl bg	Fbl ag	
Mass sample [g]		25.0	25.0	25.0	25.0	25.0	25.0	25.0
Estimated extractable sapon in in sample, 3–5 % \thickapprox 4 % \rightarrow^{*} 2 % [g]		0.5	0.5	0.5	0.5	0.5	0.5	0.5
Mass solvent (ethanol 70.55 % w/w) [g]		50.0	50.1	50.1	50.1	50.1	50.0	50.1
Estimated extracted saponin in solution [%]	≲	1	1	1	1	1	1	1
Mass extract [g]		26.2	28.4	22.0	22.6	21.5	20.5	23.5
Estimated extracted saponin [g]	≲	0.26	0.28	0.22	0.22	0.21	0.20	0.23

 * conservatively assuming an effective extraction of only 50 %

3.2. Laundry experiment

The visually observable results showed only marginal differences at most between the blank treatment and any of the extracts or reference surfactants. This applies to both runs (see 2.2) of the experiment, see Tab. 2–3. There was no observable effect for the rapeseed oil stains at all, as these were already hardly discernible in the blank treatments.

Evidently, the extracts, although colored themselves (see 3.1), did not introduce any additional discolorations to the fabric.

Tab. 2: Results of laundry experiment I. Cleansing effect compared to mere ethanol treatment.•: no observable effect, (+): marginally increased stain removal,

+: slightly increased stain removal	, $++:$ considerably	increased stain removal,
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						+++:	highly in	creased	stain n	remova	
	Stain:	Г	Tomato juice			Balsamic vinegar			Rapeseed oil		
Treatment	Repeat:	Ι	II	III	Ι	II	III	Ι	II	III	
Extract 1 (C bg), saponin 1 $\%$ w/w		(+)	(+)	•	(+)	(+)	(+)	•	•	•	
Extract 2 (C ag), saponin 1 $\%$ w/w		•	•	(+)	•	(+)	(+)	•	•	•	
Extract 3 (F bg), saponin 1 $\%$ w/w		•	•	(+)	•	(+)	(+)	•	•	•	
Extract 4 (F ag), saponin 1 $\%$ w/w		•	(+)	•	(+)	•	(+)	•	•	•	
Extract 5 (Fbl bg), saponin 1 % [*] w/w	7	(+)	•	•	(+)	•	(+)	•	•	•	
Extract 6 (Fbl ag), saponin 1 $\%$ w/w	v	•	(+)	•	•	(+)	(+)	•	•	•	
Cocamidopropyl betaine 1 % w/w		(+)	(+)	(+)	(+)	(+)	(+)	•	•	•	
Coco glucoside 1 % w/w		(+)	•	(+)	(+)	(+)	•	•	•	•	
						* es	timated p	percenta	qe, see	Tab.	

Tab. 3: Results of laundry experiment II. Cleansing effect compared to mere ethanol treatment. •: no observable effect, (+): marginally increased stain removal, +: slightly increased stain removal, ++: considerably increased stain removal, +++: highly increased stain removal

	Stain:	Т	Tomato juice			samic vin	egar	Rapeseed oil			
Treatment	Repeat:	Ι	II	III	Ι	II	III	Ι	II	III	
Extract 1 (C bg), saponin 1 $\%$ w/w		(+)	•	•	(+)	(+)	(+)	•	•	•	
Extract 2 (C ag), saponin 1 $\%$ w/w		•	•	•	•	(+)	(+)	•	•	•	
Extract 3 (F bg), sapon in 1 $\%^*{\rm w/w}$		•	(+)	•	(+)	(+)	(+)	•	•	•	
Extract 4 (F ag), saponin 1 $\%$ w/w		•	(+)	•	•	(+)	(+)	•	•	•	
Extract 5 (Fbl bg), saponin 1 $\%^*\mathrm{w/w}$		•	•	•	(+)	•	(+)	•	•	•	
Extract 6 (Fbl ag), saponin 1 $\%$ w/w		•	•	(+)	(+)	(+)	(+)	•	•	•	
Cocamidopropyl betaine 1 % w/w		•	•	(+)	(+)	•	(+)	•	•	•	
Coco glucoside 1 % w/w		•	•	•	(+)	(+)	(+)	•	•	•	
						$^{*} es$	timated	percenta	ge, see	Tab. 1	

3.3. pH values

The pH values of the soapwort extracts in 70.55 % ethanol (w/w) range from 5.3 to 5.7 with an average of 5.6. This is slightly more acidic than the cocamidopropyl betaine and considerably milder than the coco glucoside, which appears to be highly alkaline, see Tab. 4.

Tab. 4: pH values of extracts, ref	erence solutions, and solvent
	pH value
Solution	(in ethanol 70.55 $\%~{\rm w/w})$
Ethanol 70.55 $\%$ w/w	6.7
Cocamido propyl betaine 1 $\%~{\rm w/w}$	6.5
Coco glucoside 1 % w/w	12.1
Extract 1 (C bg), sapon in 1 $\%^*{\rm w/w}$	5.6
Extract 2 (C ag), sapon in 1 $\%^*{\rm w/w}$	5.7
Extract 3 (F bg), sapon in 1 $\%^*{\rm w/w}$	5.5
Extract 4 (F ag), saponin 1 $\%$ w/w	5.3
Extract 5 (Fbl bg), sapon in 1 $\%^*{\rm w/w}$	5.7
Extract 6 (Fbl ag), saponin 1 $\%^* \mathrm{w/w}$	5.6
Average (extracts 1–6)	5.6
* esti	mated percentage, see Tab. 1

Tab.	4: pH	values	of	extracts,	reference	solutions,	and solvent
						pH valı	ue

4. Discussion

With only marginal detectable differences that might not even be due to a cleansing effect, but for example due to uncertainties with respect to the applications of stains and/or treatments, the results of the laundry experiment are inconclusive.

The extraction itself severely lacks an adequate method of quantification. While the estimated saponin percentage is based on known general information, it resembles an educated guess more than a valid estimate. Any potential misjudgment is further amplified by the fact that the dilution of the reference surfactants is in turn based on this educated guess, rendering a meaningful comparison moot.

Since the design and setup of the laundry experiment as presented here seem to be inadequate for any meaningful findings with regard to cleansing effects, an improved experimental design utilizing sophisticated methods is deemed necessary for any follow-up studies.

A future laundry experiment should employ larger concentrations and/or amounts of extracted saponin and reference surfactants. Only a fraction of the available biomass was used in this study. For a considerable yield of highly concentrated extracts, a less expensive method than industrial-grade pressure applications or the use of microwaves (see Schmitt et al. 2014), for example, could be the basic heating-induced evaporation of the employed ethanol in combination with its recapture and subsequent reuse (cf. Cheok, Salman, and Sulaiman 2014, 21; cf. Koike, Jia, and Nikaido 1999), ideally for a plethora of extraction cycles. Since boiling temperatures are used occasionally for different saponin extractions (see Cheok, Salman, and Sulaiman 2014, 21; see R. Blume and Meiners 2017), there are apparently no issues with thermal stability (see Tschesche and Wulff 1973, 466) in this context.

The potential highly concentrated extracts should be quantified using a

sophisticated method such as a high-performance liquid chromatography (HPLC) (see Koike, Jia, and Nikaido 1999) before the final setup of the improved laundry experiment and its reference surfactants.

Although the treatments in this study did not cause any additional discoloration and the usual amber color reportedly turns clear when emulsified (cf. R. Blume and Meiners 2017), it should also be investigated whether highly concentrated extracts, especially those with a darker drab color, introduce a potential risk of discoloring fabrics and if so how to mitigate this, i.e. how to easily separate any possibly interfering dyes from the extracts.

II. Soil conditions in a semi-natural habitat

1. Introduction

Saponaria officinalis is native to or has become naturalized in many parts of the world including central, southeastern, and northern Europe (Khela 2012). It can be found in loamy, gravelly, or sandy locations (Hilty 2017), especially gravelly riverbanks (Meyer 1911, 135).

With regard to cultivation, the plant is described as "easy to grow" (Hilty 2017) and "not fussy about soil characteristics" (Hilty 2017), i.e. generally undemanding. The following detailed analysis of the actual soil conditions in a semi-natural habitat is supposed to aid in clarifying what this means from a perspective of empirical science.

While a habitat in the rural district of Lüneburg was chosen chiefly for convenience, as a part of Lower Saxony the area also serves well as a general example for an agricultural region with a moderate European climate (see 2.1). Moreover, Lower Saxony is home to two pioneering vegan organic growing (Hall and Tolhurst 2015) operations: *Gärtnerhof Bienenbüttel* (Verbeck 2018), approximately 12 km from Lüneburg, and the Community Supported Agriculture *Gemeinschaftsgärtnerei Wildwuchs* (SoLaWi Gemeinschaftsgärtnerei Wildwuchs e.V. 2018) near Hannover. This recent history of sustainable (cf. Steinfeld et al. 2006) agricultural endeavors further suggests the wider region as a suitable area.

Although Saponaria officinalis can be found in almost all parts of Lower Saxony (Schacherer 2007, 400), see Appendix A, App. Fig. 1, the habitat investigated is the only habitat of soapwort in the rural district of Lüneburg officially documented on a mesoscopic level (see Appendix A, App. Tab. 1). The main reason for this is that Saponaria officinalis is not an endangered species (see Khela 2012).

2. Material and methods

2.1. Area

The habitat, reference number 7157, is located between Klein Sommerbeck and Eimstorf, see Fig. 6, subdistrict of Dahlenburg, in the rural district of Lüneburg, Lower Saxony, Germany.

According to the Köppen-Geiger classification (Kottek et al. 2006), the region has an oceanic climate (Cfb) (AM Online Projects 2015a, 2015d) with borderline characteristics towards a warm humid continental climate (Dfb) (see AM Online Projects 2015b). The approximate average annual temperature is 8.5° C with an approximate average annual rainfall of 638 mm (AM Online Projects 2015a, 2015c, 2015d).

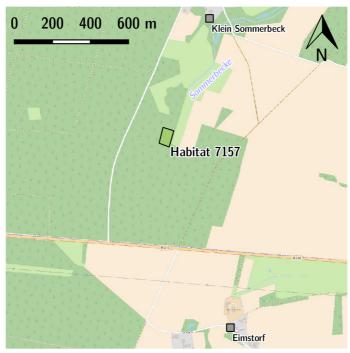


Fig. 6: Map of the area. Location of the habitat between Klein Sommerbeck and Eimstorf. Background map: OpenStreetMap © OpenStreetMap contributors (OSMF 2012)

Habitat 7157 is a grassland area of 3398 m^2 managed for conservation (see Appendix A, App. Tab. 1), partially surrounded by woods and featuring a small

artificial pond. A branch of a creek named Sommerbecke is approximately 200 m away, see Fig. 7.

According to the generalized soil map, the expected soil type in the area would be a secondary podzol (brown podzolic soil), a gley, and/or a related soil type, see Fig. 8.

Saponaria officinalis can be found at a low to medium frequency (see Appendix A, App. Tab. 1).

Three large plants, each with a height of more than 100 cm and several offshoots, were chosen for the locations of the trial holes (see Fig. 7–10). Plant No. 3 was in blossom, see Fig. 9.

The phytocoenosis included Urtica dioica, grasses, and other herbaceous plants (see Fig. 9 & Tab. 5–7). For a detailed list of the general vegetation, see Appendix A, App. Tab. 1. According to the indicator values of Saponaria officinalis and Urtica dioica (Ellenberg and Leuschner 2010, 55 & 63), a medium to high amount of N in the soil could be expected.

The forest official at *Junkernhof* (see Landkreis Lüneburg 2018, 154) stated that the soil might have a considerable amount of anthropogenic alterations due to management measures, e.g. the digging of the artificial pond. It was also suggested that the soapwort in the habitat was not autochthonous (cf. Appendix A, App. Tab. 1).

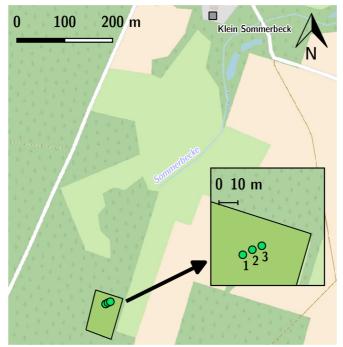


Fig. 7: Map detail. Locations of the trial holes at three large SAPONARIA OFFICINALIS plants. Background map: OpenStreetMap © OpenStreetMap contributors (OSMF 2012)

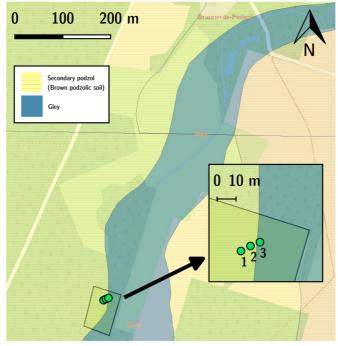


Fig. 8: Map detail. Generalized soil map of the area according to geo data (LBEG 2015). Background map: OpenStreetMap © OpenStreetMap contributors (OSMF 2012)



Fig. 9: Three large (height > 100 cm) soapwort (SAPONARIA OFFICINALIS) plants in the habitat. From left to right: Plants No. 1-3

2.2. Soil survey

A trial hole (AG Bodenkunde 2005, 38) with a width of ~ 80 cm, a length of 120 cm, and a depth of 70–90 cm to uncover all the relevant horizons of the rhizosphere, see Fig. 10, was dug and troweled at each of the three locations (see Fig. 7–9) using a spade, a shovel, a trowel, and a knife.

The horizons and soil types were determined according to the German soil classification standard *Bodenkundliche Kartieranleitung* (AG Bodenkunde 2005).



Fig. 10: Soil profiles. From left to right: Trial hole 1, width ~80 cm, depth ~90 cm. Trial hole 2, width ~80 cm, depth ~80 cm. Trial hole 3, width ~80 cm, depth ~70 cm.

Random samples from each horizon were taken and mixed with a small amount of water to determine their color and texture using the Munsell soil color chart (AG Bodenkunde 2005, 108–9; Munsell Color 2012) and a finger test (AG Bodenkunde 2005, 144–47), respectively, and the percentage of organic matter was estimated according to the color (see H.-P. Blume, Stahr, and Leinweber 2011, 29).

2.3. Soil sampling

Three volume samples per horizon were taken from the two uppermost horizons with a depth of more than 10 cm each. For each volume sample a standardized metal cylinder of 100 ml with one lid was put into a core cutter (see AG Bodenkunde 2005, 40–41). The core cutter was hammered into the horizon and then removed with a trowel securing the sample. Any material exceeding the cylinder's volume was carefully cut away with a knife, and then the second lid was put on the cylinder.

Composite samples of approximately 500 g along the full depth of each horizon (AG Bodenkunde 2005, 40) were taken from all the horizons that were fully uncovered. In addition, small random samples of approximately 50 g were taken from the horizons that were only partially uncovered.

2.4. Soil analysis

2.4.1. Preparation of volume samples and analysis of pore volume

Each volume sample was placed on a glass plate with a piece of fabric in between and then put in a basin that was filled with water up to the edges of the cylinders, see Fig. 11. The samples were left to soak up water until they were fully saturated. After five days, the saturated samples were weighed (balance: sartorius AX4202, max. 4200 g, d = 0.01 g) and then transferred into a drying cabinet, where they were left to dry at 105° C for two days. The dried samples were put into an exsiccator to let them cool down without reabsorbing moisture from the air. They were then weighed again. The total pore volume (AG Bodenkunde 2005, 349) and the soil density (AG Bodenkunde 2005, 124–26) were calculated from the mass differences between the water-saturated and the dried condition of each sample, facilitated by the cylinder volume being 100 ml and the density of water being 1 g/ml.



Fig. 11: Volume samples in water basins with added cylinder numbers (see 3.1 & Appendix B, App. Tab. 7)

2.4.2. Preparation of composite samples and random samples

The composite samples (S1,1-3, S2,1-4, S3,3) and the random samples (S1,4, S2,5, S3,4) were spread on paper plates and left to air-dry for five days. Each sample was sieved in a vibratory sieve shaker (Retsch AS 200 basic) using a sieve with a mesh size of 2 mm for at least 5 minutes at an amplitude of 70 % (100 % \triangleq 3 mm) to remove the skeleton fraction (AG Bodenkunde 2005, 155) and homogenize the sample (see Fig. 12). The percentage of the skeleton fraction was determined by weighing each sample before and after sieving. A random small amount of each composite sample was checked for carbonates by applying 10 % hydrochloric acid (HCl 10 % v/v) and looking for any fizzing reaction (H.-P.

Blume, Stahr, and Leinweber 2011, 25).

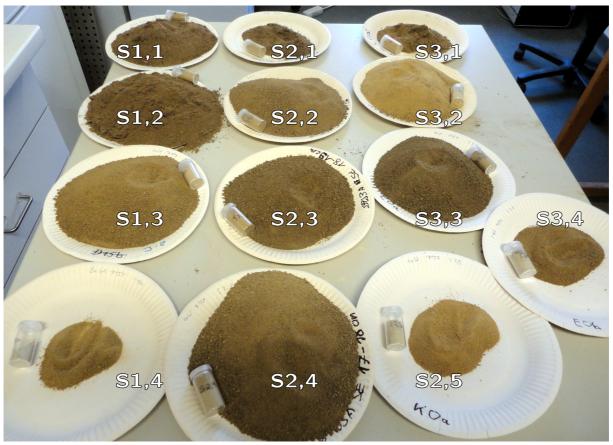


Fig. 12: Sieved (paper plates) and ground (small containers) soil samples with added identifiers (see 3.1)

2.4.3. CN analysis

Approximately 10 ml of each composite sample and each random sample were ground in a ball mill (Retsch Mixer Mill MM400) at 30 Hz for three minutes and then stored in small containers (see Fig. 12). Approximately 8 g of these finely ground samples were transferred into tin capsules (PerkinElmer Tin capsules for solids, 5 x 8 mm, precleaned, N241-1255) using a small metal spatula.

Each of the samples in the tin capsules was weighed twice with an analytical scale (sartorius analytical scale), discarding the first result. The capsules were double-folded carefully, but tightly with two pairs of tweezers to prevent any leakage and then stored in separate slots of a cartridge with unique identifiers (H1–12, F1).

These samples were analyzed with a sophisticated CN analyzer based on combustion and reduction (PerkinElmer precisely Series II CHNS/O Analyzer 2400, equipped for CN).

2.4.4. pH values

10 g of each composite sample and 2 g of each random sample were suspended in 25 ml and 5 ml $CaCl_2$ (0.01 mol/L), respectively. This was repeated for a second batch. Each suspension was stirred with a glass rod and left to settle for one hour. The glass rod was rinsed with demineralized water and dried off with a paper towel each time.

A pH meter (inoLab IDS Multi 9310 WTW) was calibrated with technical buffer solutions (NIST/PTB pH 4.01, TEP 4, 108 700; pH 7.01, TEP 7, 108 702; pH 10.01, TEP 10 Trace, 108 703). The pH value of each suspension was measured using the pH meter. The electrode was rinsed with demineralized water and dried off with a paper towel between measurements. For each sample the respective pH values of both batches were averaged.

2.4.5. Electrolytic conductivity

10 g of each composite sample and 2 g of each random sample were suspended in 100 ml and 20 ml demineralized water, respectively. The suspensions were shaken in an automatic shaker (uni jogger) at 200 rpm for two hours.

An electrolytic conductivity measuring device (Hanna Instruments HI 8733) was calibrated with a reference solution (12880 μ s/cm, HI 7030). The electrolytic conductivity (H.-P. Blume, Stahr, and Leinweber 2011, 26–27) of each suspension was measured using the device. The electrode was rinsed with demineralized water and dried off with a paper towel between measurements.

2.4.6. Sieve analysis

Each composite sample and each random sample was sieved in a vibratory sieve shaker (Retsch AS 200 basic) using a stack of sieves with mesh sizes of 630 µm, 200 µm, and 63 µm (see AG Bodenkunde 2005, 141) for 5 minutes at an amplitude of 70 % (100 % \triangleq 3 mm). The contents of the three sieves and of the bottom container were weighed separately in order to calculate the percentages of the different sand fractions (gS: coarse sand / mS: medium sand / fS: fine sand) and the silt/clay fraction (U: silt / T: clay). The silt/clay fraction itself was not differentiated.

3. Results

3.1. Soil survey

All the trial holes revealed the soil type as a gley, i.e. a type of soil affected by groundwater, with almost all of the horizons clearly showing a considerable amount of rust spots and/or mottle. For the most part, the textures appeared to be slightly or medium silty sand (Su2, Su3), while horizon No. 4 (Go3/rBv/rBv) of all three trial holes, horizon No. 5 (rGo) of trial hole No. 2, and horizon No. 2 (M) of trial hole No. 3 were found to consist of pure sand (Ss).

Trial hole No. 1 revealed a fully intact gley with a grayish topsoil (Ah) horizon and three Go horizons. Go2 was determined as a transitional horizon towards Go3. While the survey of trial holes No. 2 and 3 still showed a functional gley, in both cases a relatively young Ah horizon of only 2 cm and 3 cm, respectively, was found above a gravelly mineral (M) horizon interspersed with cobble stones. This M horizon was assumed to be an anthropogenic fill on top of a relict topsoil (rAh) horizon of the original gley. Tab. 5–7 show the results of the soil survey in detail.

Tab. 5: Soil survey. Trial hole at plant No. 1

Soil type		Gley	GK_R: GK_H:		5896735					Date:	3 July 2018
Land use	: (Conservation	Vegetatio	on:	Saponaria officinalis (not in	i blossom), <i>Urtica dio</i>	<i>ica</i> , grasses, and other	herbaceous plants			
							Organic				Volume
Horizon	Depth						matter ^{**}		Composite	Random	samples
No.	[cm]	Horizon	$Texture^*$	Color		Mottle	[%]	Comment	sample	sample	(Cyl. No.)
1	0-18	Ah	Su2	10 YR 3,2	very dark grayish brown	slight rust spots	1.5–3		\$1,1		18, 38, 94
2	18–75	Go1	Su3	10 YR 2.5,2	very dark (grayish) brown	rust spots	3–6		S1,2		41, 43, 81
3	75-85	Go2	Su2	10 YR 3,3	dark brown	rust spots	1.5-3	transitional horizon to Go3	S1,3		
4	85	Go3	Ss	10 YR 3,6	dark yellowish brown	severe rust spots	0.9–1.5			S1,4	

4411701

m · 11 1

* via finger test (AG Bodenkunde 2005, 144-47)

** according to color (see H.-P. Blume, Stahr, and Leinweber 2011, 29)

Tab. 6: Soil survey. Trial hole at plant No. 2

Trial hol		2	GK_R:		4411726					Date:	3 July 2018
Soil type	:	Gley	GK_H:		5896738						
Land use	:	Conservation	Vegetati	on:	Saponaria officinalis (not in	i blossom), <i>Urtica dioica</i> , gra	sses, and other	herbaceous plants			
							Organic				Volume
Horizon	Depth						matter ^{**}		Composite	Random	samples
No.	[cm]	Horizon	Texture [*]	Color		Mottle	[%]	Comment	sample	sample	(Cyl. No.)
1	0-2	Ah	Su2	10 YR 2.5,2	very dark (grayish) brown		3-6		S2,1		
2	2–40	М	Su2	10 YR 3,3	dark brown	rust spots	1.5-3	probably anthropogenic fill	S2,2		1, 9, 54
3	40-57	rAh	Su3	10 YR 3,2	very dark grayish brown	rust spots	1.5-3		S2,3		37, 45, 66
4	57-74	rBv	Ss	10 YR 3,3	dark brown	severe rust spots, slight	1.5-3		S2,4		
						mottle, dark spots					
5	74	rGo	Ss	10 YR 3,4	dark yellowish brown	yellow-reddish mottle	0.9-1.5			S2,5	
				,.	, , , , , , , <u>, , , , , , , , , , , , </u>	,					
-											

* via finger test (AG Bodenkunde 2005, 144-47)

** according to color (see H.-P. Blume, Stahr, and Leinweber 2011, 29)

Tab. 7: Soil survey. Trial hole at plant No. 3

Trial hole Soil type:		iley	GK_R: GK_H:		4411731 5896740					Date:	3 July 2018
Land use:	C	onservation	Vegetatio	on:	Saponaria officinalis (in blo	ssom), <i>Urtica dioica</i> , grasses	, and other her	baceous plants			
Horizon No.	Depth [cm]	Horizon	Texture [*]	Colon		Mottle	Organic matter ^{**} [%]	Comment	Composite sample	Random sample	Volume samples (Cyl. No.)
1	0–3	Ah	Su2	10 YR 3,2	very dark grayish brown	slight rust spots	1.5-3	Comment	S3,1	sample	(091. 100.)
2	3–30	М	Ss	10 YR 3,6	dark yellowish brown	rust spots	0.9–1.5	probably anthropogenic fill	\$3,2		59, 80, 84
3	30-63	rAh	Su2	10 YR 2.5,2	very dark (grayish) brown	severe rust spots, slight mottle	3–6		\$3,3		3, 4, 64
4	63	rBv	Ss	10 YR 3,3	dark brown		1.5–3			S3,4	

* via finger test (AG Bodenkunde 2005, 144-47)

** according to color (H.-P. Blume, Stahr, and Leinweber 2011, 29)

3.2. Soil analysis

With the exception of the Ah horizon of trial hole No. 1, all the textures were revealed by the sieve analysis to be pure sand (Ss), i.e. coarse-sandy medium sand (mSgs) in the two uppermost horizons and fine-sandy medium sand (mSfs) in the other ones. The texture of the Ah horizon of trial hole No. 1 was the only one determined as slightly silty sand (Su2). The Ah horizons were shown to have a skeleton fraction of 5.7-6.8 %, the Go1 horizon one of 4.6 %, and the M horizons a considerably higher one of 10.5-12.1 %

The analysis of the volume samples resulted in total pore volumes of 42-54 %. The M horizons were shown to have higher densities than the rAh horizons below them.

Within the relevant horizons of the rhizosphere the average pH value in $CaCl_2$ was 4.8. The most acidic value was a pH of 4.3 in the Ah horizon of the fully intact typical gley at trial hole No. 1.

The electrolytic conductivity was found to be considerably low at only 17 –45 $\mu\mathrm{S/cm}.$

No carbonates were detected. The C content was overall low with a percentage of 2.1-2.4 % in the Ah horizons, 1.4-1.9 % in the rAh and Go1 horizons, and 0.7 % in the rBv and Go2 horizons. The N content was overall low with an average percentage of 0.2 %.

The M horizons were shown to have lower C and N contents than the horizons both directly above and below them, the M horizon of trial hole No. 3 considerably so.

For a detailed summary of the full laboratory analysis, see Tab. 8. The results from the sieve analysis and the C analysis are compared with the corresponding results from the soil survey in Tab. 9.

			0	U	0	(/ 1	1	/ 11	1 /
					Skeleton	Total pore	Soil				Electrolytic
Trial hole	Horizon	Depth		Texture	fraction	volume	density	C content	N content	pH value	conductivity
No.	No.	[cm]	Horizon	(sieve analysis)	[%]	[%]	$[g / cm^3]$	[%]	[%]	$(\mathrm{in}\;\mathrm{CaCl}_{_{2}})$	$[\mu S / cm]$
1	1	0–18	Ah	Su2	6.78	54.14	1.10	2.11	0.22	4.3	27.3
	2	18-75	Go1	Ss: mSgs	4.64	46.05	1.38	1.51	0.20	4.7	26.7
	3	75–85	Go2	Ss: mSfs	3.28	n/a	n/a	0.71	0.14	5.0	17.4
	4	85-	Go3	(Ss: mSfs)	(22.37)	n/a	n/a	(0.33)	(0.12)	(5.1)	(21.8)
2	1	0–2	Ah	Ss: mSgs	5.72	n/a	n/a	2.39	0.27	4.7	40.3
	2	2–40	М	Ss: mSgs	10.45	42.86	1.44	1.60	0.20	4.6	21.4
	3	40-57	rAh	Ss: mSfs	1.26	50.61	1.32	1.88	0.23	4.9	28.4
	4	57–74	rBv	Ss: mSfs	0.26	n/a	n/a	0.72	0.14	5.1	21.8
	5	74–	rGo	(Ss: mSfs)	(0.12)	n/a	n/a	(0.37)	(0.11)	(5.2)	(21.0)
3	1	0–3	Ah	Ss: mSgs	6.41	n/a	n/a	2.12	0.27	4.9	45.1
	2	3–30	М	Ss: mSgs	12.08	45.83	1.30	0.51	0.11	4.9	19.5
	3	30-63	rAh	Ss: mSfs	1.99	51.23	1.27	1.38	0.19	4.9	33.3
	4	63–	rBv	(Ss: mSfs)	(0.58)	n/a	n/a	(0.66)	(0.10)	(5.4)	(26.8)

Tab. 8: Results of the laboratory analysis. (For raw data, see Appendix B, App. Tab. 4-11)

Tab. 9: Comparison of texture and C content from soil survey and laboratory analysis.

						$\operatorname{Estimated}^*$	$\operatorname{Estimated}^{**}$	
Trial hole	Horizon	Depth		$Texture^*$	Texture	organic matter	C content	C content
No.	No.	[cm]	Horizon	(finger test)	(sieve analysis)	[%]	[%]	[%]
1	1	0–18	Ah	Su2	Su2	1.5–3	0.9–1.7	2.11
	2	18–75	Go1	Su3	Ss: mSgs	3–6	1.7–3.5	1.51
	3	75–85	Go2	Su2	Ss: mSfs	1.5–3	0.9–1.7	0.71
	4	85–	Go3	Ss	(Ss: mSfs)	0.9–1.5	0.5–0.9	(0.33)
2	1	0–2	Ah	Su2	Ss: mSgs	3–6	1.7–3.5	2.39
	2	2–40	Μ	Su2	Ss: mSgs	1.5–3	0.9–1.7	1.60
	3	40–57	rAh	Su3	Ss: mSfs	1.5–3	0.9–1.7	1.88
	4	57–74	rBv	Ss	Ss: mSfs	1.5–3	0.9–1.7	0.72
	5	74–	rGo	Ss	(Ss: mSfs)	0.9–1.5	0.5–0.9	(0.37)
3	1	0–3	Ah	Su2	Ss: mSgs	1.5–3	0.9–1.7	2.12
	2	3–30	М	Ss	Ss: mSgs	0.9–1.5	0.5–0.9	0.51
	3	30–63	rAh	Su2	Ss: mSfs	3–6	1.7–3.5	1.38
	4	63–	rBv	Ss	(Ss: mSfs)	1.5–3	0.9–1.7	(0.66)
								* see 3.1
			** C c	ontent = org	ganic matter \div .	1.72 (cf. AG Be	odenkunde 2	005, 111)

4. Discussion

While the soil types at two of the three trial holes should have been found to be secondary podzols instead of gleys if the geo data (see Fig. 8) were assumed to be highly accurate, considerable uncertainties are expected from a generalized soil map as a matter of course. Thus, finding a gley at all three trial holes can be considered well in line with the data represented in Fig. 8.

That the M horizons have resulted from an anthropogenic fill is not only

probable due to their morphology (see 3.1) and the information from the forest official (see 2.1), but also supported by their C and N contents being lower than those of the horizons both above and below them and their higher densities compared to the rAh horizons.

The overestimation of silt in the soil survey (see Tab. 9) was probably due to a misinterpretation of iron oxides as silty material in the finger test, considering the high amount of visible rust spots and mottle (see 3.1).

With its relatively low C content (cf. AG Bodenkunde 2005, 111; cf. H.-P. Blume, Stahr, and Leinweber 2011, 29), its unexpectedly (see 2.1) low N content (cf. H.-P. Blume, Stahr, and Leinweber 2011, 211–13) and its marginal electrolytic conductivity (cf. H.-P. Blume, Stahr, and Leinweber 2011, 26–27), see Tab. 8, the soil's nutrient conditions are overall rather poor.

Generally favorable conditions that should be noted are the high pore volume and the proximity of groundwater. This probably results in high water availability due to capillary water rising from the groundwater, as indicated by the rust spots found in even upper horizons. The pH values are also only moderately acidic (see AG Bodenkunde 2005, 367).

Overarching discussion

1. Results in context

While the results of the laundry experiment were inconclusive, the obvious foaming activity of the soapwort extracts showed that saponin extraction might be an accessible source of biosurfactants without the need for a synthesis step (cf. Makkar, Cameotra, and Banat 2011; cf. Santos et al. 2016). The relatively low pH value looks promising for applications such as shampoos and other personal care products (cf. Lambers et al. 2006), as well as detergents for delicate fabrics.

Although the examination of the soil conditions was only a small-n study and a comprehensive investigation of several different locations would be needed for representative results, the survey and analysis presented here nevertheless work well to showcase what it means when *Saponaria officinalis* is described as undemanding (see II. 1). From a perspective of sustainable agriculture, this supports the notion of cultivating soapwort as a saponin source that is easily grown.

2. Limitations and caveats

2.1. Silty and clayey soils

The overall sandy texture and the amount of gravel and cobble stones found in the examination of the soil conditions are well in line with the typical growth conditions described in the literature (Hilty 2017; Meyer 1911, 135). Whether more silty and/or clayey soils pose actual problems for *Saponaria officinalis* in an agricultural context should be further investigated.

2.2. Water availability

The water availability of the highly porous gley in the presented habitat is reminiscent of soapwort being typically found in moist to slightly dry (Hilty 2017) locations at riverbanks (Meyer 1911, 135). Cultivation of *Saponaria officinalis* on soils that are not directly affected by groundwater might require an additional amount of irrigation in dryer regions.

2.3. Creeping rhizomes

Although the aggressive growth of soapwort's creeping rhizomes (Meyer 1911, 135) might be desirable with respect to the yield in harvestable belowground biomass, it might also have the undesirable effect of outcompeting other crops (Hilty 2017) when integrating *Saponaria officinalis* into polycultural setups. The use of physical barriers could be necessary in these cases.

2.4. Toxicity in fish

One of the defining characteristics of saponins is their toxicity in fish (Tschesche and Wulff 1973, 462). This, however, does not have to present a problem for the use of saponin from soapwort as a laundry detergent, since the high biodegradability of saponins as naturally occurring surfactants mitigates the issue when a wastewater treatment facility is in place (see Knieper, Kalewski, and Carlson 2011).

It still poses two potential sustainability issues that need to be addressed. First, people's reasonable concerns about the toxicity in fish might cause them to avoid or boycott saponin-based detergents or even campaign for their ban.

Secondly, campers, hippies, and/or inhabitants of rural areas might use

saponin-based detergents in open bodies of water, unwittingly jeopardizing fish in the process.

It is thus of utmost importance to communicate well that saponin is safe for use in households of industrialized areas, but unsafe for use in open bodies of water. A well-designed, noticeable warning label on commercial products might help with this. In addition, advice for sensible practices in situations outdoors, e.g. using Aleppo or Castile soap and vinegar for washing and rinsing clothes and hair, could be included.

3. Beyond region

Growing soapwort in Europe instead of importing palm oil or soapnuts from tropical regions might be desirable in itself. Potentially establishing large monocultures of *Saponaria officinalis*, however, would be less than ideal from a sustainability perspective. When considering the systematic cultivation of soapwort as a saponin source, the integration into existing agricultural operations with sustainable practices should be key.

As Saponaria officinalis is a flowering plant that attracts wild pollinators, e.g. butterflies (Hilty 2017) and bumblebees (Wolff et al. 2006), it could be grown in dedicated wildflower strips and patches.

Additionaly, since saponin can be found in many different plants (Sparg, Light, and van Staden 2004, 219; cf. Schwarzbach 2004) it might be advisable to diversify saponin sources in general, e.g. integrate soapwort into vegan organic growing (Hall and Tolhurst 2015) operations as described, cultivate horse chestnuts (see Truttwin 1920, 35) in agroforestry (see Nair and Garrity 2012) systems, collect waste saponin from processing sugar beets (cf. Knieper, Kalewski, and Carlson 2011), and so forth.

Transdisciplinary (Dubielzig and Schaltegger 2004, 9–11) research projects monitoring these efforts could be established to both further the availability of sustainable detergents and improve the scientific understanding of feasibility and efficiency issues.

4. Conclusion

The examination of the soil conditions in a semi-natural habitat showcases the undemanding nature of *Saponaria officinalis* and thus indicates the plant as a viable candidate for systematic cultivation as a source of saponin. The integration into existing sustainable agriculture and a diversification of saponin sources seem advisable.

Actual risks of toxicity in fish and their mitigation via biodegradation in wastewater treatment facilities need to be addressed carefully and properly.

The general efficacy of saponin from soapwort as a laundry detergent as well as efficiency issues should be further investigated using sophisticated quantitative methods.

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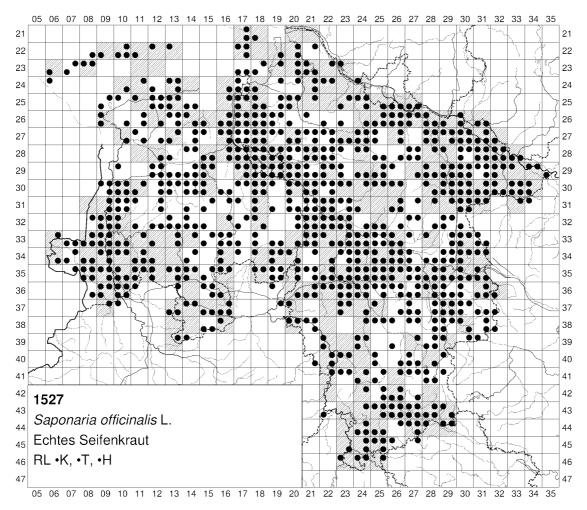
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Appendix

A: Supplementary data. Vegetation



App. Fig. 1: Distribution of SAPONARIA OFFICINALIS in Lower Saxony (Schacherer 2007, 400).

RL: Red List (as at 1.1.2004). K: Region coast, T: Region lowland, H: Region lowland. •: not endangered in this region

		el district of Lüneburg. Mappin			
Designati		Mesophile grassland south of Klein S	ommerbeck	Date:	13 July 2016
Conservat	tion category:	§22 GLB			
Descriptio	on:	Humid grassland area managed for co	onservation at a small boo	ly of water,	
		in part with presumably planted non-	autochthonous species		
Rating:		4 (high)			
Area:		3398 m ²			
Land pare	cels				
No.		District Ca	dastral section	Parcel	Size $[m^2]$
1		Eimstorf 5		41/3	84821
Habitat t	ypes				
No.	Code	Name			Percentage
1	GMF	Mesophile grassland of moderately h	umid habitats		100
Plant spe	cies				
No.		Latin name		Frequency	Red List
1		Agrimonia eupatoria L. ssp. eupatoria	1	1 (rare)	3
2		Anthyllis vulneraria L.		1 (rare)	3
3		Cirsium palustre (L.) Scop.		1 (rare)	*
4		Heracleum sphondylium L. ssp. spho	ndylium	1 (rare)	*
5		Phalaris arundinacea L.		2 (medium)	*
6		Veronica chamaedrys L. ssp. chamae	drys	2 (medium)	*
7		Dianthus deltoides L.		2 (medium)	3
8		Filipendula ulmaria (L.) Maxim.		2 (medium)	*
9		Galeopsis speciosa Mill.		2 (medium)	V
10		Galium aparine L.		2 (medium)	*
11		Galum verum L.		2 (medium)	V
12		Geranium palustre L.		2 (medium)	2
13		Glechoma hederacea L.		2 (medium)	*
14		Vicia cracca L.		2 (medium)	*
15		Holcus mollis L.		2 (medium)	*
16		Lotus pedunculatus Cav.		2 (medium)	*
17		Lysimachia nummularia L.		2 (medium)	*
18		Athyrium filix-femina (L.) Roth		2 (medium)	*
19		Origanum vulgare L. ssp. vulgare		2 (medium)	*
20		Plantago lanceolata L.		2 (medium)	*
21		Ranunculus acris L.		2 (medium)	*
22		Rubus fruticosus agg.		2 (medium)	*
23		Rumex acetosa L.		2 (medium)	*
24		Saponaria officinals L.		2 (medium)	*
25		Alopecurus pratensis L. ssp. pratensis	1	2 (medium)	*
26		Urtica dioica L. ssp. dioica		2 (medium)	*
27		Arrhenatherum elatius (L.) P. Beauv	. ex J. Presl & C. Presl	2 (medium)	*
				. ,	

App. Tab. 1: Mapping of habitat 7157. Data according to the local environmental authorities of the rural district of Lüneburg. Mapping done by PLANUNGSGEMEINSCHAFT MARIENAU

B: Raw data. Results

				App.	Tab.	2: Extre	action	results
	Extract:	1	2	3	4	5	6	Average
	Sample:	C bg	C ag	F bg	F ag	Fbl bg	Fbl ag	
Mass sample [g]		25.0	25.0	25.0	25.0	25.0	25.0	25.0
Mass solvent (ethanol 70.55 % w/w) [g]		50.0	50.1	50.1	50.1	50.1	50.0	50.1
Mass extract [g]		26.2	28.4	22.0	22.6	21.5	20.5	23.5
Mass loss (solvent–extract) [g]		23.8	21.7	28.1	27.5	28.6	29.5	26.5

		pH value							
		(i							
Solution	Measurement:	Ι	II	III	Average				
Ethanol 70.55 % w/w		6.695	6.680	6.800	6.725				
Extract 1 (C bg), saponin 1 $\%^* \mathrm{w/w}$		5.627	5.594	5.585	5.602				
Extract 2 (C ag), sapon in 1 $\%^*{\rm w/w}$		5.745	5.743	5.728	5.739				
Extract 3 (F bg), sapon in 1 $\%^*{\rm w/w}$		5.545	5.542	5.528	5.538				
Extract 4 (F ag), sapon in 1 $\%^*{\rm w/w}$		5.335	5.333	5.334	5.334				
Extract 5 (Fbl bg), saponin 1 $\%^*\mathrm{w/w}$		5.765	5.765	5.717	5.749				
Extract 6 (Fbl ag), saponin 1 $\%^*{\rm w/w}$		5.566	5.555	5.563	5.561				
Cocamido propyl betaine 1 % w/w		6.471	6.473	6.470	6.471				
Coco glucoside 1 % w/w		12.152	12.129	12.080	12.120				
			* estimated	percentage, s	ee Tab. 1				

App. Tab. 3: pH values of extracts, reference solutions, and solvent

					11pp: 100: 2	. Steele analysts.	Shereron jraction
						Mass	
		Total mass	Mass	Mass	Mass	skeleton fraction	Skeleton fraction
Composite	Random	(incl. plate)	plate	roots	soil $\leq 2~\mathrm{mm}$	(> 2 mm)	(> 2 mm)
sample	sample	$[\mathbf{g}]$	[g]	[g]	[g]	[g]	[%]
S1,1		766.02	12.89	24.79	678.98	49.36	6.78
S1,2		943.87	13.73	0.00	886.98	43.16	4.64
S1,3		766.81	12.44	0.00	729.64	24.73	3.28
	S1,4	46.57	14.39	0.00	24.98	7.20	22.37
S2,1		204.37	14.80	10.33	168.98	10.26	5.72
S2,2		837.34	14.92	0.38	736.12	85.92	10.45
S2,3		527.64	15.37	1.68	504.15	6.44	1.26
S2,4		618.08	17.77	0.00	598.73	1.58	0.26
	S2,5	62.18	11.88	0.00	50.24	0.06	0.12
S3,1		184.55	14.51	0.59	158.58	10.87	6.41
S3,2		991.33	14.59	0.00	858.76	117.98	12.08
S3,3		553.07	12.52	0.00	529.77	10.78	1.99
	S3,4	70.49	11.87	0.00	58.28	0.34	0.58

App. Tab. 5: Sieve analysis. Texture

									11pp.	100.0	. Dicti	s unu	<i>you</i> .	1 cature
Sample	source													
		Weighed	Mass	Mass	Mass	Mass	Mass	Mass	gS	mS	$_{\rm fS}$	\mathbf{S}	U/T	
Composite	Random	mass	loss	sum	$2000630~\mu\mathrm{m}$	$630200~\mu\mathrm{m}$	200–63 μm	63– μm	fraction	fraction	fraction	fraction	fraction	
sample	sample	g	[g]	[g]	[g]	[g]	[g]	[g]	[%]	[%]	[%]	[%]	[%]	Texture [*]
S1,1		64.73	0.49	64.24	6.16	23.48	27.52	7.08	9.59	36.55	42.84	88.98	11.02	Su2
S1,2		52.82	0.21	52.61	14.94	25.38	11.21	1.08	28.40	48.24	21.31	97.95	2.05	mSgs
S1,3		64.35	0.24	64.11	5.81	33.37	21.18	3.75	9.06	52.05	33.04	94.15	5.85	mSfs
	S1,4	14.69	0.23	14.46	1.47	7.09	4.66	1.24	10.17	49.03	32.23	91.42	8.58	mSfs
S2,1		50.55	0.35	50.20	12.20	20.92	14.76	2.32	24.30	41.67	29.40	95.38	4.62	mSgs
S2,2		62.39	0.27	62.12	14.23	23.25	20.04	4.60	22.91	37.43	32.26	92.59	7.41	mSgs
S2,3		52.30	0.30	52.00	7.63	24.92	17.22	2.23	14.67	47.92	33.12	95.71	4.29	mSfs
S2,4		54.41	0.22	54.19	3.96	25.54	21.64	3.05	7.31	47.13	39.93	94.37	5.63	mSfs
	S2,5	37.18	0.37	36.81	2.35	19.51	12.65	2.30	6.38	53.00	34.37	93.75	6.25	mSfs
S3,1		51.70	0.33	51.37	13.76	19.74	14.38	3.49	26.79	38.43	27.99	93.21	6.79	mSgs
S3,2		61.59	0.41	61.18	12.32	24.79	20.10	3.97	20.14	40.52	32.85	93.51	6.49	mSgs
S3,3		57.81	0.13	57.68	6.28	25.84	22.88	2.68	10.89	44.80	39.67	95.35	4.65	mSfs
	S3,4	44.64	0.35	44.29	2.75	22.86	15.23	3.45	6.21	51.61	34.39	92.21	7.79	mSfs
	* (see AG Bodenkunde 2005, 144-48)													

App. Tab. 6: CN analysis

							11 P P .	1001 01 011	anargere
Sample so	ource		Analyzed					Spare	
		Finely	Weighed	Weighed			Finely	Weighed	Weighed
Composite F	Random	ground	mass I	mass II	C content	N content	ground	mass I	mass II
sample s	ample	sample	[g]	[g]	[%]	[%]	sample	[g]	[g]
S1,1		H1	8.615	8.616	2.11	0.22	G1	7.724	7.725
S1,2		H2	7.560	7.559	1.51	0.20	G2	6.448	6.447
S1,3		H3	6.174	6.174	0.71	0.14	G3	6.050	6.050
S	51,4	H4	7.329	7.330	0.33	0.12	G4	7.453	7.454
S2,1		H5	7.209	7.208	2.39	0.27	G5	9.048	9.048
S2,2		H6	8.340	8.340	1.60	0.20	G6	7.740	7.739
S2,3		H7	9.862	9.860	1.88	0.23	G7	8.514	8.512
S2,4		H8	8.029	8.029	0.72	0.14	G8	8.996	8.994
S	52,5	H9	8.186	8.185	0.37	0.11	G9	8.974	8.973
S3,1		H10	6.164	6.162	2.12	0.27	G10	8.011	8.012
S3,2		H11	9.636	9.636	0.51	0.11	G11	7.344	7.344
S3,3		H12	7.506	7.507	1.38	0.19	G12	8.016	8.016
S	53,4	F1	9.995	9.995	0.66	0.10	E1	7.558	7.559

TT + 11 1 NT	App			lume and	soil densit			lant No. 1
Trial hole No.	1	1	1	1	$\frac{1}{2}$	1 2	$\frac{1}{2}$	1
Horizon No. Horizon	1 Ah	1 Ah	1 Ah	1 Ah		Go1	Go1	2 Go1
Cylinder No.	All 18	38	94	All	41	43	81	GOI
Depth [cm]	10-15	0-5	5-10	_	25-30	35-40	47–53	
Direction		Horizontal			Horizontal		Horizontal	
	TIGHEORICA	TIGHEORE	TIGHZONU		Tionzontar	Ventical	TIGHEORICA	
Weight cylinder [g]	95.99	95.85	97.35		95.11	99.26	97.32	
Weight Cymider [g]	55.55	93.03	51.55	-	95.11	55.20	51.52	_
	107.10	100.04	100.00		100.05	100.00	100.00	
Weight plate [g]	127.16	128.24	129.38	_	128.25	129.09	128.06	
Weight water-saturated sample								
(incl. cylinder & plate) [g]	387.24	386.00	393.81		425.72	402.13	400.07	
Weight dry sample								
(incl. cylinder & plate) [g]	332.45	329.95	342.22		388.40	353.99	347.38	
(men eyimder & prace) [8]	002.10	020100	•	Average			0.11.00	Average
Water-saturated mass [g]	164.09	161.91	167.08	164.36		173.78	174.69	183.61
Dry mass [g]	109.30	105.86	115.49	110.22		125.64	122.00	137.56
Water content $[g \triangleq m]$	54.79	56.05	51.59	54.14	37.32	48.14	52.69	46.05
Total pore volume [%]	54.79	56.05	51.59	54.14	37.32	48.14	52.69	46.05
Soil density $[g / cm^3]$	1.09	1.06	1.15	1.10	1.65	1.26	1.22	1.38
	An	n Tab 8	$R \cdot Pore vo$	lume and	soil densit	u Trial	hole at ni	lant No 2
Trial hole No.	2	2	2	2		2	$\frac{none}{2}$	2
Horizon No.	2	2	2	2		3	3	3
Horizon	М	М	М	М		rAh	rAh	rAh
Cylinder No.	1	9	54		37	45	66	
Depth [cm]	25-30	33–38	35–40		40–45	40–45	50–55	
Direction	Horizontal	Vertical	Horizontal		Horizontal	Vertical	Vertical	
Weight cylinder [g]	99.32	96.21	95.08		96.12	97.80	98.59	
Weight plate [g]	130.84	127.98	128.44		127.22	126.64	128.22	
Weight water-saturated sample								
(incl. cylinder & plate) [g]	412.17	403.11	424.04		410.18	403.23	408.41	
Weight dry sample								
(incl. cylinder & plate) [g]	369.16	360.14	381.45		359.05	350.63	360.30	
TT <i>t</i> t t t t t t t t t t	102.01	170.00	200 52	Average		170 70	101.00	Average
Water-saturated mass [g] Dry mass [g]	182.01 139.00	178.92 135.95	200.52 157.93	187.15 144.29		178.79 126.19	181.60 133.49	182.41 131.80
Water content $[g \triangleq m]$	43.01	42.97	42.59	42.86		52.60	48.11	50.61
Total pore volume [%]	43.01	42.97	42.59	42.86		52.60	48.11	50.61
Soil density $[g / cm^3]$	1.39	1.36	1.58	1.44		1.26	1.33	1.32
Son density [g / em]	1.00	1.50	1.00	1	1.00	1.20	1.00	1.02
	Apr	p. Tab. g): Pore vo	lume and	soil densit	y. Trial	hole at pl	lant No. 3
Trial hole No.	3	3	3	3	3	3	3	3
Horizon No.	2	2	2	2	3	3	3	3
Horizon	Μ	Μ	Μ	Μ	rAh	rAh	rAh	rAh
Cylinder No.	59	80	84		3	4	64	
Depth [cm]	10–15	13–18	18-23		35–40	35–40	43–48	
Direction	Horizontal		Vertical		Horizontal		Vertical	
Weight cylinder [g]	96.99	97.38	97.39		96.49	97.85	95.43	
Weight plate [g]	131.14	112.39	127.91		127.95	128.02	127.23	
Weight water-saturated sample								
(incl. cylinder & plate) [g]	394.29	385.43	411.86		397.17	407.82	403.95	
Weight dry sample								
	242.00	242 55	267.22		240.66	257 12	257.45	
(incl. cylinder & plate) [g]	343.22	343.55	367.33	Average	340.66	357.13	357.45	Average
Water-saturated mass [g]	166.16	175.66	186.56	Average 176.13		181.95	181.29	Average 178.66
Dry mass [g]	115.09	133.78	142.03	130.30		131.26	134.79	127.42
Water content $[g \triangleq m]$	51.07	41.88	44.53	45.83		50.69	46.50	51.23
Total pore volume [%]	51.07	41.88	44.53	45.83		50.69	46.50	51.23
Soil density $[g / cm^3]$	1.15	1.34	1.42	1.30		1.31	1.35	1.27
- [0 / · ·]								

T. Oldridge: Basic Viability of Saponin from Soapwort as a Sustainable Laundry Detergent 40

				Sample source		Suspen		
						Demineralized	Weighed	Electrolytic
Trial Hole	Horizon	Depth		Composite	Random	water	sample mass	$\operatorname{conductivity}$
No.	No.	[cm]	Horizon	sample	sample	[ml]	[g]	$[\mu S / cm]$
1	1	0–18	Ah	\$1,1		100	10.02	27.3
	2	18–75	Go1	S1,2		100	10.00	26.7
	3	75–85	Go2	S1,3		100	10.00	17.4
	4	85–	Go3		S1,4	20	2.01	21.8
2	1	0–2	Ah	\$2,1		100	10.01	40.3
	2	2–40	М	S2,2		100	10.00	21.4
	3	40–57	rAh	S2,3		100	10.01	28.4
	4	57–74	rBv	S2,4		100	10.00	21.8
	5	74–	rGo		S2,5	20	2.00	21.0
3	1	0–3	Ah	S3,1		100	10.00	45.1
	2	3–30	М	S3,2		100	10.01	19.5
	3	30–63	rAh	S3,3		100	10.00	33.3
	4	63–	rBv		S3,4	20	2.01	26.8

$App. \ Tab. \ 10: \ Electrolytic \ conductivity$

App. Tab. 11: Soil pH values

						11771 1001	11. 2000 1			
Sample	source	S	uspension I		Suspension II					
		CaCl_{2}	Weighed		CaCl_2	Weighed				
Composite	Random	(0.01 mol/L)	sample mass		(0.01 mol/L)	sample mass		average		
sample	sample	[ml]	[g]	pH value	[ml]	[g]	pH value	pH value		
S1,1		25	10.02	4.449	25	10.03	4.245	4.347		
S1,2		25	10.00	4.751	25	10.02	4.670	4.711		
S1,3		25	10.03	5.093	25	10.01	4.973	5.033		
	S1,4	5	2.00	4.986	5	2.01	5.139	5.063		
S2,1		25	10.00	4.784	25	10.00	4.598	4.691		
S2,2		25	10.01	4.650	25	10.01	4.533	4.592		
S2,3		25	10.02	4.916	25	10.00	4.877	4.897		
S2,4		25	10.00	5.132	25	10.01	5.093	5.113		
	S2,5	5	2.01	5.003	5	2.01	5.348	5.176		
S3,1		25	10.07	4.933	25	10.01	4.820	4.877		
S3,2		25	10.01	4.904	25	10.02	4.809	4.857		
S3,3		25	10.00	4.875	25	10.00	4.952	4.914		
	S3,4	5	2.00	5.266	5	2.00	5.471	5.369		