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Preface

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The 9th International Symposium for Sustainable Humanosphere (The 9th ISSH 2019)

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Editorial

On behalf of the organizing committee, I would like to present the Proceeding of the 9th International Symposium for Sustainable Humanosphere (ISSH) which was held at Grand Savero Hotel, Bogor Indonesia on 28 – 29 October 2019. The symposium was the 9th annual meeting for ISSH and in conjunction with Humanoshpere Science School (HSS) was also marked as The 409th Symposium on Sustainable Humanosphere for RISH – Kyoto University, Japan. This annual event is an important and prestigious scientific forum organized by RC Biomaterials – LIPI and RISH – Kyoto University dedicated to gather the researchers, academicians, professionals, and general public to sharing knowledge, disseminating research funding, exchanging success stories and expanding both national and international collaboration.

The forum was successfully organized by Research Centre for Biomaterials – Indonesian Institute of Sciences (RC Biomaterials – LIPI), and gratefully (hugely?) supported by many institution and research programs namely: Research Institute for Sustainable Humanosphere (RISH) – Kyoto University, Japan, Japan Science and Technology Agency (JST) through Japan-ASEAN Science, Technology, and Innovation Platform (JASTIP) program; JST – JICA (The Japan International Cooperation Agency) collaborative program through SATREPS (Science and Technology Research Partnership for Sustainable Development); and also National Institute of Aeronautics and Space (LAPAN), Indonesia. The committee.

The insightful and high quality papers have been presented following the symposium theme ”Integrated Smart Technology and Society for Sustainable Humanosphere”, and we are happy to share 56 selected papers that present the best approach to sustainability in humanosphere using an integrated perspective from Applied Science & Technology, Biosciences, Community-based development & socio-economic sciences, Earth & Atmospheric Sciences, and Forest Sciences & Biomaterial Materials.

The organizers appreciate the support and assistance of the co-operating institutions, the participants, the presenters, supporting staffs, and all the sponsors. We are very proud to announce that the present event had generated a great success, as shown by impressive numbers of quality papers with wide-ranging backgrounds and interdisciplinary topics. We hope the proceedings of the 9th ISSH 2019 could provide significant contribution to global research in the field of humanosphere science.

We look forward to see you all again in the future symposium.

Chief Editor

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The Utilization of Oil Palm Leaves (*Elaeis guineensis* Jacq.) Waste as an Antibacterial Solid Bar Soap

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Abstract. Oil palm leaves (*Elaeis guineensis* Jacq), which have been underutilized by the community and become waste that usually stacked around the trees, have a potential to be used as active ingredients for making antibacterial solid bar soap. The chemical content of oil palm leaves are tannins, alkaloids and flavonoids that known had antibacterial activity. This research aims to produce oil palm leaves extracts into an active ingredient of solid bar soap formulation with antibacterial activity against *Escherichia coli* and *Staphylococcus aureus* and also to evaluate the quality of the solid bar soap. The soap was formulated into 3 formulas with varying concentrations of oil palm leaves ethanolic extract that was F1 (1%), F2 (2%), and F3 (4%). Oil palm leaves extract was prepared with maceration using ethanol 70%. Antibacterial activity assay of the solid bar soap was carried out using a disc diffusion method with tetracycline as the positive control (K+) and distilled water as the negative control (K-). The soap quality was evaluated for its organoleptic, foam level and foam stability, pH, hardness, water content and free fatty acid. The results showed that all three solid soap formulas meet the soap quality requirements of SNI No. 3532-2016. The solid bar soap did not have inhibition properties against *Escherichia coli*. However, had inhibition effect against *Staphylococcus aureus* with an average Inhibition Zone Diameter (IZD) on soap base, F1, F2, and F3 was 8.02 mm, 8.53 mm, 10.53 mm, 12.91 mm respectively.

1. Introduction

Oil palm (*Elaeis guineensis* Jacq.) is an important commodity for the Indonesian economy. Oil palm plays a role in generating more foreign exchange than oil and gas [1]. One of the uses of oil palm is as a producer of vegetable cooking oil. Oil produced from oil palm able to produce oil seven times higher than rapeseeds (*Brassica napus*) and eleven times higher than soybean per hectare. Since 2004, oil palm has slowly become a worldwide vegetable oil with a total production of 30 million tons and an average growth rate of 8% per year [2]. Unfortunately, this industry leaves waste, such as kernel shells, mesocarp fibers, oil palm trunks (stems) and oil palm fronds (leaves) [2,3]. Hambali & Rivai (2017) reported that oil palm leaf waste in 2015 reached 124,032,861 tons. This value is likely to continue to increase with time [2].

Oil palm leaves, normally underutilized by the community. They usually left alone to rot between the oil palm trunks. The abundance aims to maintain the sustainability of the soil and nutrient cycle in the plantation [4]. The oil palm leaves contain major compounds such as alkaloids, tannins, and flavonoids. These compounds are known to act as antimicrobials because they can damage cell walls, disrupt cell permeability, and inhibit enzyme or protein [5,6,7,8].

Several studies have proven that oil palm leaf extract has antimicrobial activity. Previous studies reported that oil palm leaf extracts were able to inhibit the growth of *Escherichia coli* and *Staphylococcus aureus* [8,9,10]. Other research stated that in addition to its potential as an antibacterial, oil palm leaf extract also has the potential to be a skin protective agent from UV radiation. Based on this, Yusof et al. (2006) suggest that oil palm leaf extracts can be made into a topical application for skin protection [4].

One form of topical application that can be used to maintain health is soap. Soap is a product that is produced from the reaction between fatty acids with strong bases that function to wash and clean fat or dirt. There are two types of soap, namely solid soap and liquid soap. Most people come down using solid soap (bar soap) to clean the body because bar soap is cheaper, easier to use, and efficient in cleaning the skin [11]. This study aims to test the formulation of soap from oil palm leaves extracts for its activity as antibacterial soap.

2. Material and Methods

2.1. Materials

The oil palm leaves were obtained from Desa Bogatama, Lampung, Indonesia. Chemicals being used 70% ethanol, aquadest, H_2SO_4, HCl, acetic acid, Nutrient Agar (NA) [Oxoid], olive oil, coconut oil, palm oil, sodium lactate, dyes, fragrance, Sodium hydroxide (NaOH) [Merck], NaOH 0.1 N [Merck], plastic wrap. The microorganisms used were *Staphylococcus aureus* and *Escherichia coli* bacteria collection from the Microbiology Laboratory of ISTN.

2.2. Extraction of Oil Palm Leaves

The oil palm leaves were cut into small pieces and washed thoroughly with water and then dried using the oven. The dried leaves were mashed using a blender into a powder. The leaf powder then sieved using a 60 mesh sieve to obtain uniform particle sizes. The powder then macerated using 70% ethanol with a ratio of 1:10 for 1 x 24 hours with stirring every 6 hours. The filtrate obtained was filtered and the dregs were re-macerated 2 times. Furthermore, the filtrate obtained from maceration was concentrated using a vacuum rotary evaporator, then evaporated on a water bath until a thick extract was produced from oil palm leaf powder [12].

2.3. Extracts Examination

2.3.1. Organoleptic. The organoleptic examination based on Monograph Ekstrak Tumbuhan Indonesia [13].

2.3.2. Phytochemical Screening. Phytochemical screening was carried out based on Materia Medika Indonesia (Depkes RI, 1989) and Pandey & Tripathi (2014). Screening includes testing for alkaloids, flavonoids, tannins, saponins, steroids and triterpenoids [14,15].

2.4. Soap Formulation

The formulation refers to Hambali *et al.* (2006) with modification [16]. The composition of the formula in 100 ml aquadest add presented in Table 1.

Solid soap formulations were carried out by dissolving NaOH in distilled water. NaOH solution is mixed with sodium lactate. The mixture of solutions was called the first mixture. Meanwhile, at different container, mixed the coconut oil, olive oil and palm oil, known as the second mixture. The first mixture was poured dropwise into the second mixture. The mixture was stirring until homogeneous using a hand blender and a *trace* occurs (a condition in which the soap has formed with a sign of the thickening soap mass). The ethanol extract of oil palm leaves was added at the time of the *trace*. After that, the mixture was stirred again until homogeneous, then added fragrance and colouring. The liquid soapy mass is poured into a mold and incubated for 24 hours until it hardens.

Table 1. Soap Bar Formulation.

Materials	Concentration (%)			
	Base (BF)	F1	F2	F3
Oil Palm Leaves Extract	0	1	2	4
NaOH	9.5	9.5	9.5	9.5
Coconut Oil	20	20	20	20
Palm Oil	35	35	35	35
Olive Oil	10	10	10	10
Sodium lactate	1.95	1.95	1.95	1.95
Distilled water	22.1	22.1	22.1	22.1
Colouring	0.5	0.5	0.5	0.5
Fragrance	0.5	0.5	0.5	0.5

2.5. Soap Evaluation

2.5.1 Organoleptic Evaluation. Organoleptic evaluation by observing the colour, texture, and aroma of the soap [20].

2.5.2. pH Test. An amount of 1 g soap sample was dissolved into 10 mL distilled water and stirred until homogenous. The solution was measured using pH meter. According to ASTM (2002) the pH of a relatively safe soap is 9-11 [17].

2.5.3. Foam Level and Stability. About 1 gram of soap was put into a test tube containing 10 ml of distilled water and then shaken with vortex for 30 minutes. The height of foam formed is measured using a ruler (initial foam height). The height of the foam is measured again after a few minutes (high foam end).

$$\text{Foam Stability} = 100\% - \%\ \text{Foam Loss} \quad (1)$$

$$\%\ \text{Foam loss} = (\text{High initial foam} - \text{High final foam})/(\text{High initial foam}) \times 100\% \quad (2)$$

2.5.4. Soap Hardness. The hardness of the soap was done using a penetrometer. The needle on the penetrometer was inserted into the sample and allowed to penetrate the material for 5 seconds at a constant temperature (27°C). The depth of needle penetration into the material is expressed in 1/10 mm of the number indicated on the penetrometer scale.

2.5.5. Moisture Content. Measurement of moisture content was using Moisture Content Balance [Ohaus].

$$\text{Free Fatty Acid} = 0.282 \times V \times N / w\ 100\% \quad (3)$$

Information:
Free fatty acids in units of% mass fraction
V = volume of KOH used (mL)
N = normality of KOH used
B = sample weight (g)
282 = equivalent weight of oleic acid ($C_{18}H_{34}O_2$)

2.5.6. Hedonic Test. The hedonic test was performed to compare BF, F1, F2, and F3. This test aims to determine which formula was preferred by consumers. The criteria of the test were appearance, colour, aroma, skin irritation test, and skin moisture test. The preference test was carried out using 20 panellists consisting of men and women at random, with an age range of 18-35 years. The score for the panellist preference level used was around 1-8.

Criteria:
1 = very, very dislike
2 = really don't like it
3 = don't like it
4 = rather not like
5 = normal / neutral
6 = rather like it
7 = like
8 = really like it

2.6 Antibacterial Test

2.6.1. Bacterial Suspensions. About 1-2 ose of bacterial cells were suspended in 5 ml of a 0.9% NaCl solution. Turbidity of bacterial suspension was equivalent to Mc. Farland 3 (3×10^8). The suspension was then diluted to obtain 3×10^7 CFU / ml.

2.6.2. Diffusion Test. The antibacterial activity test was using the disk diffusion method (Kirby-Bauer Method). A total of 20 µl of a solid soap solution is dripped onto disk paper and awaited until it dries. Meanwhile, petri dishes containing NA media were inoculated with 0.1 ml of bacterial suspension and spread evenly over the plates. The disk that has been dripped by the sample was then placed on the surface of the media. The same method was also done on tetracycline as a positive control and aquadest as a negative control. Samples were incubated at 37ºC for 24 hours and then observed inhibitory zones were formed [18].

3. Results and Discussion

3.1. Extracts Examination

Extraction was done by maceration method using 70% alcohol. The extract obtained as much as 178 g thick extract from 830 g of oil palm leaves powder. From that results, the yield was 21.4%. The results can be seen in Table 2.

Table 2. The Yield of Oil Palm Leaves Extracted Using 70% alcohol.

Leaves Powder (g)	Thick Extract (g)	Yield (%)
830	178	21.4

3.1.1 Organoleptic Results. The organoleptic results showed that the extract of oil palm leaves in the form of thick and concentrated liquid, blackish-brown, and has a typical aroma of oil palm leaves. The results were presented in Table 3.

Table 3. Organoleptic Results of Oil Palm Leaves Extract.

Criteria	Organoleptic Results
Shape	thick and concentrated liquid
Aroma	typical aroma of oil palm leaves
Colour	Blackish-brown

3.1.2. Phytochemical Screening. The results of the phytochemical screening showed that the ethanolic extract of oil palm leaves contain an alkaloid, saponin, tannin, flavonoid and triterpenoid (Table 4). These results were suitable with research conducted by Febrina *et al.* (2018) who used 96% ethanol as a solvent [10].

Table 4. Phytochemical Screening of Oil Palm Leaves Extract.

Plant Constituents	Tests	Results
Alkaloid	Dragendorff	(+)
	Bouchardart	(+)
	Mayer	(+)
Saponin	Water and HCl	(+)
Tanin	$FeCL_3$	(+)
Flavonoid	Na Nitrit 5%, HCl 1%, NaOH 1 N	(+)
Triterpenoid	Cloroform, H_2SO_4 P	(+)

(+): present.

3.2. Solid Soap Bar Formulation

Solid soap bar was formulated into three concentrations, namely 1%, 2%, and 4%. The process of making soap was done by the cold process method. The cold process is recommended by small-scale enterprise soap makers in low-resource contexts, particularly in tropical regions due to quick solidification of local oils [19]. The results of the formulation showed that the base formula looked brighter, whereas in soap with concentrations of 1%, 2%, and 4% are slightly darker. The higher the concentration of the extract, the darker the soap preparations will be. [Figure 1]

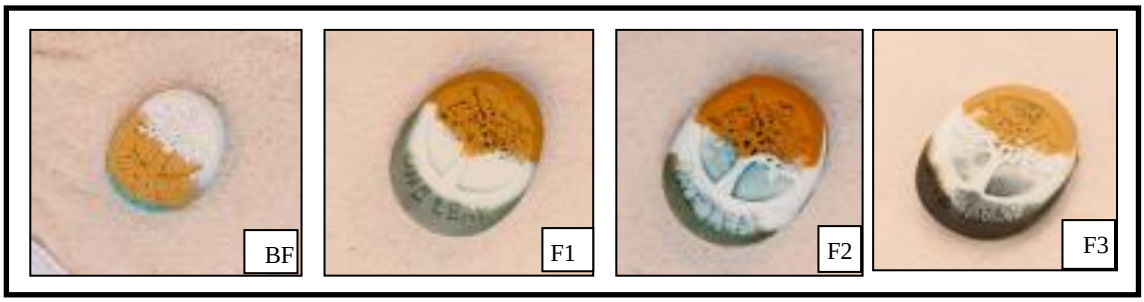

Figure 1. Solid bar soap formulation. BF: basis formula; F1: formula with 1% oil palm leaves extract; F2: formula with 2% oil palm leaves extract; F3: formula with 4% oil palm leaves extract.

3.3. Soap Evaluation

The soap evaluation was carried out aiming to see whether the solid soap meets the requirements to SNI (Indonesian National Standard) 2016 on bath soap which includes organoleptic tests, hardness test, moisture content, foam level test, free alkali content, and pH test [20].

3.3.1. Organoleptic Tests. The organoleptic test was done by visually observing the solid bar soap including form, colour, and aroma.

Table 5. Organoleptic Tests of Solid Bar Soap.

Formula	Basic colour of the soap	Aroma	Form
BF	White	Typical of oil palm leaves	Solid
F1	Brownish-yellow	Typical of oil palm leaves	Solid
F2	Brown	Typical of oil palm leaves	Solid
F3	Blackish-brown	Typical of oil palm leaves	Solid

BF: basis formula; F1: formula with 1% oil palm leaves extract; F2: formula with 2% oil palm leaves extract; F3: formula with 4% oil palm leaves extract.

Organoleptic tests showed that whether BF, F1, F2, and F3 has solid form and has a distinctive aroma of oil palm leaves, which is not commercial to promote. Whereas based on the colour produced, the basis formula soap showed white colour, F1 has a brownish-yellow colour, F2 has a brown colour, and F3 has a blackish-brown colour.

Based on the colour and aroma we added additional dye and bubble gum fragrance to cover the basic colour and aroma of the soap. That additional were expected can make the soap colour more attractive and cover the distinctive aroma of the leaves that are less pleasant (Figure 1).

3.3.2. Foam Level and Stability. The results of foam level and stability presented at Table 6.

Table 6. Foam Level and Stability of Solid Bar Soap from Oil Palm Leaves Extract.

Formula	High Initial Foam (cm)	High Final Foam (cm)	Foam Stability (%)
BF	9.5	6.7	99.70
F1	10.3	7.7	99.74
F2	9.7	7.5	99.77
F3	9.5	7.5	99.79

The results of the foam level and foam stability tests showed that BF has a stability of 99.70%, F1 of 99.74%, F2 of 99.77%, and at F3 of 99.79%. From these results, it can be concluded that F3 has the best stability among other formulations.

3.3.3. pH Test

The pH measurement was done at week 3 because making soap using the cold process method takes 2-4 weeks for a stable pH. Soap will experience a curing time where the soap will undergo a maturation process. Curing time is the time needed to evaporate water in natural soap so that the soap will be safe to use, harder, better foam, stable pH, softer if used, and more durable. The results of the pH test showed that BF, F1, F2 and F3 had a pH that was still within the limits allowed for the preparation of soap that is 9-11 [17].

Table 7. pH Test Result of Solid Bar Soap from Oil Palm Leaves Extract.

Formula	pH
BF	9.27 ± 0.06
F1	9.58 ± 0.06
F2	9.82 ± 0.06
F3	9.79 ± 0.06

3.3.4. Soap Hardness Test. Requirements for the value of soap hardness are not yet available so there are no requirements that indicate hardness in soap. the result of the test can be seen in Table 8.

Table 8. Hardness Test of Solid Bar Soap.

Formula	**Hardness (mm/g/s)**
BF	16.00×10^{-1}
F1	16.05×10^{-1}
F2	17.00×10^{-1}
F3	18.00×10^{-1}

From the results (Table 8), we can see that soap which is 3 weeks old shows hardness on a basis 16.00×10^{-1}, whereas in Formulation 1% the extract shows hardness 16.05×10^{-1}, Formulation 2% extract shows hardness 17.00×10^{-1}, The 4% extract formulation showed a hardness of 18.00×10^{-1}. These results indicate that the ethanol extract of oil palm leaves can affect the hardness of the soap. The higher the concentration of the extract given to the soap, the softer the soap.

3.3.5. Moisture Content. Moisture content testing on soap aims to measure the per cent of water content contained in solid soap after drying at a temperature of 105 ºC for 60 minutes using a Moisture Content Balance Analyzer (Table 9). The maximum permissible moisture content in soap is 15% [20].

Table 9. Moisture Content on Solid Bar Soap from Oil Palm Leaves Extract.

Formula	Moisture Content (%)
BF	10.60
F1	12.97
F2	10.90
F3	8.28

The testing was done after the soap has been stored for 3 weeks. The soap was made using the cold process method which will be stable within 2-4 weeks. So that the soap undergoes a perfect saponification process. Besides that, the duration of soap storage affects the hardness of the soap due to the water content in the soap had evaporated. The results of the test showed that all formulas meet the requirements of SNI 3532:2016 (less than 15%).

3.3.6. Free Fatty Acid Test. Free fatty acids are fatty acids in soap that are not bound as sodium compounds or triglyceride compounds (neutral fat). The high free fatty acids in soap will reduce the power to clean the soap because free fatty acids are undesirable components in the cleaning process. The presence of free fatty acids can be checked if there is no red colour on the phenolphthalein indicator after boiling in neutral alcohol. Free fatty acids which dissolve in neutral alcohol are then titrated with KOH [20].

The results of testing free fatty acid levels in the BF was 1.128%, in the F1 was 0.846%, in F2 was 0.958%, and in F3 was 0.789%. (Table 10). Based on the data, it is known that the amount of free fatty acids produced meets the quality requirements of bath soap according to SNI, which is a maximum of 2.5%. This means that the solid bar soap from the ethanol extract of the oil palm leaves has a low amount of free fatty acids so that the soap has good clean power and also has a good ability to clean oil from oily material.

Table 10. Free Fatty Acid Test

Formula	Free Fatty Acid (%)
BF	1.128
F1	0.846
F2	0.958
F3	0.789

3.3.7. Hedonic Test. The results of the hedonic tests presented at Table 10.

Table 11. Average of Hedonic Test

Criteria	BF	F1	F2	F3
Appearance	7.65	7.15	6.90	6.40
Colour	7.60	7.25	6.50	5.75
Aroma	7.80	6.05	4.45	4.30
Moisture	7.00	7.05	7.35	7.45
Irritation	6.15	6.15	6.20	6.25

Based on the table 11, it shows that in terms of appearance, colour and aroma, the BF was more dominant than other formulas, with average values of 7.65, 7.6 and 7.8, respectively (Table 11). The appearance of BF is more eye-catching and fresh. The aroma of the soap was also quite fresh. While, the formula containing ethanol extract of oil palm leaves has a distinctive aroma of oil palm, making it less attractive to panellists. The higher concentration of the extract, the fewer enthusiasm of the panellists, because the extract has a less pleasant aroma, even though a fragrance has been added but it still cannot cover the original aroma of the extract itself.

On the other hand, the hedonic test results in terms of humidity and irritation, showed that F3 has the highest average value are F3 with an average point of 7.45 and 6.25, respectively. Soap added with ethanol extract of oil palm leaves has a pretty good humidity when compared to BF. Hedonic observations or preferences in terms of irritation test have a little difference, of the 20 panellists there is one panellist who is irritated to the skin that is a change in the colour of the skin becomes reddish and itchy. It happens possibly because has a different skin type, and maybe the panellist have sensitive skin types that have a negative impact on the skin.

3.4. Antibacterial Activity

The data from **Table 12** showed that the solid bar soap from the oil palm leaves extract with a concentration of 1%, 2%, and 4% have inhibitory action against *S. aureus* with a diameter of the inhibitory zone about 8.53 mm, 10.53 mm, 12.91 mm, respectively. However, it did not inhibit the growth of *E. coli*.

Table 12. The Diameter of Inhibitory Zone of Oil Palm Leaves Extract Against *Staphylococcus aureus* and *Escherichia coli*.

Samples	Diameter of Inhibitory Zone (mm)	
	Escherichia coli	*Staphylococcus aureus*
BF	-	8.02
F1	-	8.53

F2	-	10.53
F3	-	12.91
K+	26.96	13.70
K-	-	-

K + : Postive Control; K-: negative control; - : no inhibitory zone

The diameter of the inhibition formed, presumably due to the presence of secondary metabolites such as alkaloid, tannins, saponins, flavonoids, and terpenoid contained in ethanol extracts of oil palm leaves which known as a potential antibacterial agent. These metabolites action provide the diameter of the inhibition as an inhibitory activity against the growth of *S. aureus*. Alkaloid compounds work by disrupting the peptidoglycan component of bacterial cells so that the cell wall layer is not formed intact and causes the death cell. Tannins are a group of polyphenol compounds which have antibacterial activity, the mechanism of action of tannins as an antibacterial is thought to be able to shrink the cell walls so that they interfere with the permeability of bacterial cells, due to disruption of permeability, bacterial cells cannot carry out living activities so that growth is inhibited or even dies. Flavonoids have antibacterial activity caused by the ability of flavonoids to form complexes with extracellular proteins and are dissolved so that the bacterial cell membranes will be damaged and lose their function to detergents, as a result, saponins will reduce the surface tension of bacterial cell walls and damage membrane permeability. Damage to the cell membrane is very disturbing survival of bacteria [5, 21].

4. Conclusion

All formulas of solid bar soap from ethanol extracts of oil palm leaves showed good physical characteristics, pH 9.27 – 9.82 (ASTM requirement 9-11), moisture content 8.28 – 12.97 % (SNI requirement less than 15%), free fatty acid levels 0.78 - 1.128% (SNI requirement maximum 2.5%), so it cat be concluded that all formulas meet the requirements established by SNI 3532: 2016 and ASTM 2002. The soap has antibacterial activity against *S. aureus*, but does not inhibit the growth of *E.coli*.

5. References

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