Distinguishing avocado oil quality based on fatty acid profile using PCA: A review of influencing factors and research gaps

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Abstract

The quality of avocado oil is influenced by multiple factors, including cultivar, growing region, drying method, harvest season, fruit maturity, extraction technique, and storage conditions. This review aims to show how fatty acid profiles (FAP), combined with principal component analysis (PCA), can be used to characterize avocado oil based on various established factors. A total of 23 peer-reviewed articles were included, encompassing 143 data points. PCA was applied as an exploratory tool to reduce dimensionality and visualize patterns in the data. Among the evaluated variables, the fruit part emerged as the most influential determinant, allowing clear categorization of avocado oils based on whether the pulp, peel, or seed was used. Additional separation was achieved based on varietal, geographical origin, harvest month, and extraction method. However, insufficient evidence was found to support consistent differentiation based on ripening stage or drying protocol. These findings also highlight key research gaps and underscore the need to update FAP standards to include oils derived from whole fruits, varied grades, and diverse extraction technologies, advancing sustainability and minimizing food waste.

Keywords: Avocado oil, Chemometrics, Fatty acid, GC-FID, Persea americana Mill.

1. Introductions

The avocado (Persea americana Mill.), which has its origins in Mexico, Central or South America, was believed to be grown in 500 BC [1]. There are over a hundred types of avocadoes registered with the Californian Avocado Society and are documented in their database together with those of three major popular varieties, such as Hass, Fuerte, and Wagner [2–4]. Avocado production was one million tons between 2014 and 2017 and is expected to rise further, with Mexico accounting for one-third of global production [5]. In the 1990s, avocados gained popularity on a global scale.

Plantings of avocado trees have replaced the oakpine forests of central Mexico due to forces from the global market. Over the past 20 years, the demand for avocados has significantly expanded in the United States, Europe, and China, largely because of its characteristics and health advantages [6].

Avocados contain less sugar, monounsaturated fatty acids (MUFA), vitamins C, E, K, B2, B3, B5, B6, B9, magnesium, potassium, omega-3 fatty acids (FA), beta-carotene, and lutein. Additional clinical research comprehends avocados' significance in maintaining cardiovascular health, managing weight, controlling blood glucose levels, and leading a healthy lifestyle [1,7,8]. Many research has

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looked at the health advantages of avocado oil and found that it has a wide range of components, including FA (oleic acid, palmitic acid, etc.) and α -tocopherol. Avocado oils are also known to reduce cholesterol, cardiometabolic risk, hypertension, blood sugar levels, hepatoprotection effect, and antimicrobial effect [9–15].

Whole Foods Market highlighted avocado oil as one of the ten significant culinary trends for 2023. The main reason for the increased usage of avocado fruit oil in many sectors is the high-income development rate in industrialized nations, with the upper-middle class targeting [16]. The market for avocado oil is competitive, but there are presently no criteria to assess whether avocado oil is of the grade stated and real, lacking official or informal criteria for identification or classification. Buyers are susceptible to fraud due to a lack of rules or improper handling of the avocado oil process, which produces rancid or low-quality avocado oils. For example, illicit activities like blending avocado oil with less expensive oils like canola, rapeseed, safflower, or soybean oil may take place [17–19]. Avocado oil's safety and quality are not guaranteed by any set of rules, and it hasn't even been standardized by the CODEX Committee for Fats and Oils yet; instead, it's currently being proposed or standardized under edible oils standard [20,21]. Avocado oil has potential market growth; thus, it is necessary to standardize it to ensure consumer safety and prevent product adulteration [22].

The two primary categories of standards for edible oil are quality and purity. FAP is a popular purity indicator for identifying adulterated oil, and gas chromatography (GC) is primarily utilized for the measurement. GC provided enhanced separation and reduced tailing compared to LC, thanks to the derivatization of FA into FAMEs [23]. In addition, GC is usually combined with highly sensitive detectors, such as a thermal conductivity, electron capture, flame-photometric, nitrogen-phosphorus, photo-ionization, or mass spectrometry [24,25], while conventional HPLC relies on UV detection, which may lack appropriate sensitivity [23]. For FAP profiling in avocado oil, GC coupled with FID is the most often utilized tool. This approach is specifically supported in the proposed modification to the Standard for Named Vegetable Oils (CX-STAN 210-1999), ratified in November 2024 by the Codex Alimentarius Commission. Some factors, namely cultivar, area, drying method, fruit quality, harvest season, maturity, part of avocado used, and extraction method, have all been reported to affect the FAP of avocado oil [5,9,26-30].

Several reviews have underscored the importance of extraction methods and their influence on the FAP of avocado oil. Tapia et al. (1999) explored the chemical characteristics, terminology, and industrial relevance of avocado lipids. Their study noted that avocados from Te Puke, New Zealand, contained higher total FA and dry matter levels but notably less oleic acid compared to those from the Far North, suggesting a temperature-related regional effect that warrants further investigation [31]. Qin and Zhong (2016) reviewed the compositional characteristics and extraction techniques of avocado oil, concluding that oil yield and quality are highly dependent on both conventional and emerging extraction technologies [32]. Similarly, Flores et al. (2019) emphasized that factors such as geographic origin, climate, cultivar, and extraction procedure play pivotal roles in determining avocado oil quality [9]. Satriana et al. (2019) compared various extraction methods and noted that high temperatures during Soxhlet extraction degrade nutritional quality, while cold pressing, though gentler, typically yields lower oil quantities [33]. These studies collectively indicate that extraction techniques can result in oils with distinct characteristics, often necessitating refining to meet compositional standards.

Despite these insights, existing reviews fall short of rigorously analyzing the combined effect of parameters such as cultivar, fruit part, ripeness, harvest timing, extraction method, and solvent type on the FAP of avocado oil. Emerging research also points to additional variables that may be relevant but remain unexplored. To bridge these gaps, this review examines a range of factors that may influence FAP variability, based on data generated through GC-FID. By employing PCA, we aim to reduce data complexity, uncover latent patterns, and support robust decision-making in modeling and interpretation. Through this chemometric approach, the review seeks not only to highlight research gaps but also to provide a methodological framework for future investigations into FAP dynamics under GC-FID.

2. Method

Research articles were extracted from various databases, namely PubMed, Scopus, and DOAJ for PCA. Keywords used in the search: "avocado oil" AND (characteristics OR "fatty acid" OR GC OR chromatography) yielded a total of 223 articles from Scopus. Several exclusion criteria were applied to limit the number of retrieved documents. These

criteria included restricting the timeframe to the year 2023, limiting the document type to original articles, and restricting the language to English. After applying the exclusion criteria, a total of 173 articles remained. No formal bias assessment tool, such as the Cochrane Risk of Bias, was utilized; however, measures were implemented to reduce selection bias through explicitly defined inclusion and exclusion criteria, consensus-driven screening by three independent reviewers, and methodological consistency by restricting the analytical platform to GC-FID. Only research that presented repeatable processes, measurable FA data, and traceable scientific protocols were incorporated into the final analysis.

The abstract and full-text screening process was completed on January 27, 2024, by three researchers working individually. Papers were included if and only if a minimum of two out of the three researchers concurred that the article in question satisfied the predetermined criteria. The article abstract was required to focus on the characterization or analysis of avocado oil and its FA composition. A total of 31 articles were retrieved using this procedure and subsequently underwent full-text screening. Articles that utilized methods apart from GC-FID (such as GC-MS) to analyze avocado oil were also excluded in this process. Limiting the scope to GC-FID ensured methodological coherence across the reviewed studies and facilitated robust cross-comparisons, both among the datasets and against the proposed CODEX benchmark for avocado oil. The FAP was gathered and subjected to PCA using Minitab® 19 in order to extract essential information. This review also included additional publications obtained by a manual search using PubMed and DOAJ, as well as using the snowballing process by examining the 12 accessible articles. Fig. 1 provides a comprehensive breakdown of the quantity of articles that were either excluded or included during each stage of screening or exclusion.

3. Results and discussion

FA data extracted from various journals (Supplementary Data A1 (https://doi.org/10.38212/2224-6614.3561)) is submitted to PCA. The data underwent additional simplification, and the listwise deletion approach was employed to omit the FAP parameters that are not present in all articles from the study [34,35]. Setting the data to 0 will produce bias in the PCA result; hence, it cannot be done (unless the particular FA is not detected). While listwise deletion may reduce sample size and

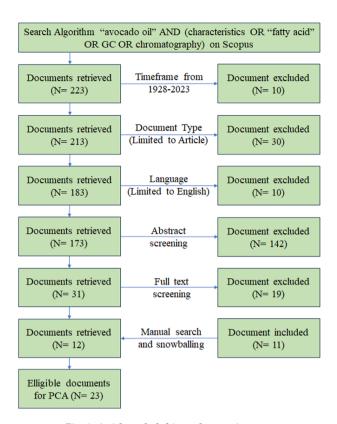


Fig. 1. Article excluded in each screening step.

introduce bias, it was deemed appropriate here given the heterogeneity across studies and to preserve analytical transparency. The raw data and the simplified data are preserved separately (Table 1). The PCA has six independent variables, namely C16:0 (palmitic acid), C18:1 (oleic acid), SFA, USFA, MUFA, and PUFA, with a total of 148 items. The covariance matrix type was selected since the independent variables inhabit the same unit.

3.1. Part of fruit used

The PCA result indicates that part of the fruit used (pulp, seed, and peel) played an important role in the differentiation of FAP under GC-FID (Fig. 2A). Among all factors assessed, the fruit part produced the clearest clustering in PCA (Supplementary Data B1 (https://doi.org/10.38212/2224-6614.3561)). The eigenvectors and loading plot demonstrated the significant contribution of oleic acid (PC1, with an eigenvector value as high as 0.598) and PUFA (PC2 with an eigenvector value of up to 0.674) to sample clustering as x and y, respectively. As much as 94.1% of the variance can be explained by the two PCs combined. The dependent variables MUFA and oleic acid have similar vectors (Fig. 2B) indicating that the groups

Table 1. Effect of various variables on fatty acid profile output.

Ref	Variables								Fatty A	Fatty Acid Composition ^a (%)					
	Variety	Source	Continent	Maturity	Part	Drying	Extraction Solvent	Extraction Method	C16:0	C18:1	SFA	USFA	MUFA	PUFA	
[36]	NS	Harcourt, Nigeria	Africa	Ripe	Pulp	Oven	Water	Floatation	12.6	43.23	35.31	64.50	44.60	19.90	
	NS	Harcourt, Nigeria	Africa	Ripe	Seed	Oven	Water	Floatation	55.0	20.67	69.00	30.95	23.25	7.70	
[37]	Bacon	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	28.20	43.29	33.64	65.93	52.66	13.27	
	Bernecker-43	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	37.08	29.84	41.84	58.14	43.22	14.92	
	Dade-3	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	32.21	33.46	36.58	63.41	45.40	18.01	
	Day	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	27.00	41.75	31.48	68.50	51.30	17.20	
	FL Hass	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	31.56	28.08	38.07	61.91	44.59	17.32	
	Lula	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	30.12	34.63	36.58	63.40	47.31	16.09	
	Miguel	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	28.20	39.23	33.10	66.87	48.26	18.61	
	Monroe	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	30.24	42.48	35.63	64.36	51.34	13.02	
	PA-6206	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	22.84	44.42	28.95	71.02	55.23	15.79	
	Pflume	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	28.71	40.40	34.29	65.69	50.84	14.85	
	Simmonds	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	31.66	33.47	34.47	65.51	44.60	20.91	
	Zutano	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	27.00	35.67	34.84	65.13	49.69	15.44	
	35,706	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	32.37	36.89	34.98	65.00	46.45	18.55	
	35,707	Florida, USA	NA	Ripe	Pulp	Oven	IH	Pisani	22.94	45.14	28.52	71.45	53.82	17.63	
[38]	Hass	Antalya, Turkey	Asia	Unripe	Peel	Fresh	PE	Soxhlet	20.00	50.12	21.88	71.43	50.12	21.31	
[JO]	Hass	Antalya, Turkey	Asia	Unripe	Peel	Air	PE	Soxhlet	17.82	54.85	20.07	74.68	54.85	19.83	
	Hass	Antalya, Turkey	Asia	Unripe	Peel	Microwave	PE	Soxhlet	19.48	49.98	21.13	72.96	49.98	22.98	
	Hass	Antalya, Turkey	Asia	Unripe	Peel	Oven	PE	Soxhlet	19.14	53.93	20.05	73.56	53.93	19.63	
	Hass	Antalya, Turkey Antalya, Turkey	Asia	Ripe	Peel	Fresh	PE	Soxhlet	18.59	54.57	20.03	72.75	54.57	18.18	
	Hass	Antalya, Turkey Antalya, Turkey	Asia	Ripe	Peel	Air	PE	Soxhlet	16.51	48.15	21.35	73.11	48.15	24.96	
					Peel	Microwave		Soxhlet	19.08	47.71	20.25	73.66	47.71	25.95	
	Hass	Antalya, Turkey	Asia	Ripe	Peel	Oven	PE	Soxhlet		51.21	20.23		51.21		
	Hass	Antalya, Turkey	Asia	Ripe					19.51			72.94		21.73	
	Hass	Antalya, Turkey	Asia	Unripe	Pulp	Fresh	PE	Soxhlet	19.77	57.37	20.38	72.73	57.37	15.36	
	Hass	Antalya, Turkey	Asia	Unripe	Pulp	Air	PE	Soxhlet	19.85	56.19	20.54	73.04	56.19	16.85	
	Hass	Antalya, Turkey	Asia	Unripe	Pulp	Microwave		Soxhlet	21.57	58.50	22.40	70.96	58.50	12.46	
	Hass	Antalya, Turkey	Asia	Unripe	Pulp	Oven	PE	Soxhlet	23.03	59.43	23.69	69.44	59.43	10.01	
	Hass	Antalya, Turkey	Asia	Ripe	Pulp		PE	Soxhlet	20.40	51.96	21.18	71.91	51.96	19.95	
	Hass	Antalya, Turkey	Asia	Ripe	Pulp		PE	Soxhlet	19.16	54.88	19.87	73.64	54.88	18.76	
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Microwave		Soxhlet	20.95	57.21	21.89	71.47	57.21	14.26	
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	20.89	57.41	21.86	71.54	57.41	14.13	
	Hass	Antalya, Turkey	Asia	Unripe		Fresh	PE	Soxhlet	23.56	37.14	28.21	68.30	37.67	30.63	
	Hass	Antalya, Turkey	Asia	Unripe	Seed		PE	Soxhlet	17.12	42.14	21.12	76.19	42.72	33.47	
	Hass	Antalya, Turkey	Asia	Unripe		Microwave	PE	Soxhlet	17.90	29.17	19.60	75.64	29.68	45.96	
	Hass	Antalya, Turkey	Asia	Unripe		Oven	PE	Soxhlet	26.05	28.42	28.76	67.79	31.26	36.53	
	Hass	Antalya, Turkey	Asia	Ripe		Fresh	PE	Soxhlet	18.78	36.53	24.18	72.72	38.12	34.60	
	Hass	Antalya, Turkey	Asia	Ripe	Seed		PE	Soxhlet	19.12	27.56	26.70	69.85	28.51	41.34	
	Hass	Antalya, Turkey	Asia	Ripe	Seed	Microwave	PE	Soxhlet	20.78	27.10	24.56	72.12	29.47	42.65	
	Hass	Antalya, Turkey	Asia	Ripe	Seed	Oven	PE	Soxhlet	21.75	42.61	26.10	71.47	44.14	27.33	
[39]	Fuerte	Paraíba, Brazil	SA	Ripe	Pulp	Oven	n-hexane	Soxhlet	21.312	64.436	22.799	77.047	67.433	9.614	
	Fuerte	Paraíba, Brazil	SA	Ripe	Seed	Oven	n-hexane	Soxhlet	20.874	17.41	31.569	67.438	20.712	46.726	

(continued on next page)

Table 1. (continued)

Ref	Variables								Fatty A	Acid Co	mpositio	on ^a (%)		
	Variety	Source	Continent	Maturity	Part	Drying	Extraction Solvent	Extraction Method	C16:0	C18:1	SFA	USFA	MUFA	PUFA
[40]	Hass	Antioquia, Colombia	SA	Ripe	Pulp	Oven	PE	Soxhlet	18.14	55.38	19.85	81.64	66.68	14.96
	Hass	Antioquia, Colombia	SA	Ripe	Pulp	Oven	PE	Soxhlet	18.21	59.19	19.50	80.50	67.91	12.59
	Hass	Antioquia, Colombia	SA	Ripe	Pulp	Oven	PE	Soxhlet	19.63	50.63	21.28	78.73	64.37	14.36
	Hass	Antioquia, Colombia	SA	Ripe	Pulp	Oven	PE	Soxhlet	21.45	42.14	23.70	76.35	59.68	16.67
	Hass	Antioquia, Colombia	SA	Ripe	Pulp	Oven	PE	Soxhlet	20.27	46.76	22.29	77.71	63.24	14.47
	Hass	Antioquia, Colombia	SA	Ripe	Pulp	Oven	PE	Soxhlet	20.54	43.23	22.51	77.49	59.29	18.20
[27]	Bacon	Málaga-Spain	Europe	Ripe	Pulp	_	Water	Malaxation	12.16	71.55	12.65	87.35	78.61	8.74
	Fuerte	Málaga-Spain	Europe	Ripe	Pulp	_	Water	Malaxation	12.37	73.57	13.00	86.99	78.06	8.93
	Hass	Málaga-Spain	Europe	Ripe	Pulp	_	Water	Malaxation	18.17	61.56	18.65	81.35	69.64	11.71
	Pinkerton	Málaga-Spain	Europe	Ripe	Pulp	_	Water	Malaxation	16.93	65.92	17.53	82.46	73.65	8.81
	Hass	Brazil	SA	Ripe	Pulp	_	Water	Malaxation	21.05	71.62	21.77	89.79	76.86	12.93
[41]	Fortuna	Itambé, Brazil	SA	Ripe	Peel	Oven	n-hexane	Soxhlet	28.93	39.85	31.93	67.97	48.52	19.45
	Collinson	Itambé, Brazil	SA	Ripe	Peel	Oven	n-hexane	Soxhlet	19.79	43.01	23.27	77.42	51.04	26.38
	Barker	Itambé, Brazil	SA	Ripe	Peel	Oven	n-hexane	Soxhlet	24.25	42.87	28.42	71.62	50.40	21.22
	Fortuna	Itambé, Brazil	SA	Ripe	Pulp	Oven	n-hexane	Soxhlet	20.51	51.40	22.30	77.73	60.79	16.94
	Collinson	Itambé, Brazil	SA	Ripe	Pulp	Oven	n-hexane	Soxhlet	27.47	51.26	28.99	70.59	56.91	13.68
	Barker	Itambé, Brazil	SA	Ripe	Pulp	Oven	n-hexane	Soxhlet	36.39	32.66	41.27	58.76	37.49	21.27
	Fortuna	Itambé, Brazil	SA	Ripe	Seed	Oven	n-hexane	Soxhlet	22.41	10.88	39.73	56.12	16.81	39.31
	Collinson	Itambé, Brazil	SA	Ripe	Seed	Oven	n-hexane	Soxhlet	12.64	17.59	29.01	69.27	27.05	42.22
	Barker	Itambé, Brazil	SA	Ripe	Seed	Oven	n-hexane	Soxhlet	17.87	16.09	35.36	63.21	24.94	38.27
[42]	NS	Bantul, Indonesia	Asia	Ripe	Pulp	Sun	n-hexane	Percolation	30.91	34.79	33.22	55.74	46.09	9.65
	NS	Purwokerto, Indonesia	Asia	Ripe	Pulp	Sun	n-hexane	Percolation	28.73	42.77	30.49	62.83	51.11	11.72
	NS	Garut, Indonesia	Asia	Ripe	Pulp	Sun	n-hexane	Percolation	25.28	47.99	27.01	68.94	55.86	13.08
[26]	Margarida	Brazil	SA	Ripe	Pulp	_	_	Centrifugated	23.28	57.33	23.28	76.07	59.98	16.09
	Hass	Brazil	SA	Ripe	Pulp	_	_	Centrifugated	19.43	54.72	19.43	80.12	66.07	14.05
[43]	Breda	São Sebastião do Paraíso, Brazil	SA	Ripe	Pulp	Oven	_	Cold-pressed	19.9	59.3	22.3	77.7	65.8	11.9
	Breda	São Sebastião do Paraíso, Brazil	SA	Ripe	Pulp	Oven	PE	Soxhlet	21.0	57.1	23.7	76.5	64.1	12.4
	Breda	São Sebastião do Paraíso, Brazil	SA	Ripe	Pulp	Oven	_	Cold-pressed	21.2	58.6	23.0	77.0	65.1	11.9
	Breda	São Sebastião do Paraíso, Brazil	SA	Ripe	Pulp	Oven	PE	Soxhlet	21.3	57.7	23.9	76.1	64.2	11.9
	Breda	São Sebastião do Paraíso, Brazil	SA	Ripe	Pulp	Vacuum	_	Cold-pressed	20.7	64.5	21.6	78.4	67.2	11.2
	Breda	São Sebastião do Paraíso, Brazil	SA	Ripe	Pulp	Vacuum	PE	Soxhlet	21.3	57.7	23.8	76.2	64.3	11.9
[44]	Hass	Chile, USA	NA	Ripe	Pulp	_	Water	Soxhlet	13.33	71.93	14.10	85.72	77.21	8.51
	Hass	Chile, USA	NA	Ripe	Pulp	_	n-hexane	Soxhlet	12.99	71.20	13.73	84.64	75.32	9.32
	Hass	Chile, USA	NA	Ripe	Pulp	_	_	Cold-pressed	12.75	73.63	13.64	84.69	78.14	6.55
[45]	NS	Mexico	NA	Ripe	Pulp	_	_	Cold-pressed	21.10	52.33	21.60	63.50	61.54	1.96
	NS	Mexico	NA	Ripe	Pulp	_	n-hexane	Soxhlet	18.10	56.54	18.64	66.02	63.91	2.11
	NS	Mexico	NA	Ripe	Pulp	_	n-hexane	Soxhlet	15.71	60.58	16.43	69.80	68.21	1.59
	NS	Mexico	NA	Ripe	Pulp	_	Acetone	Distillation	14.96	60.47	15.44	69.28	67.13	2.15
[46]	Ettinger	Rabat-Salé-Kenitra, Morocco	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	15.23	60.79	15.69	84.08	69.67	14.41
	Fuerte	Rabat-Salé-Kenitra, Morocco	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	15.63	57.50	16.46	81.25	59.84	21.41
	Hass	Rabat-Salé-Kenitra, Morocco	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	20.91	54.53	21.40	78.35	64.52	13.83
	Reed	Rabat-Salé-Kenitra, Morocco	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	18.43	61.18	18.92	80.39	68.96	11.43

[29]	Fuerte	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	22.4	59.3	23.05	75.94	65.47	10.47
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	23.3	47.2	24.66	74.94	58.40	16.54
	Fuerte	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	17.7	63.4	18.35	81.42	69.89	11.53
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	21.2	52.1	22.10	77.79	62.70	15.09
	Fuerte	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	12.0	73.0	12.24	87.74	77.22	10.52
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	16.8	59.5	17.21	82.84	68.94	13.90
	Fuerte	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	17.4	65.7	17.82	82.04	71.30	10.74
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	21.3	52.8	22.22	77.72	63.00	14.72
	Fuerte	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	18.7	65.1	19.20	80.67	71.10	9.57
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	20.3	53.1	21.19	78.46	63.40	15.06
	Fuerte	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	16.1	65.0	16.72	82.57	70.28	12.29
	Hass	Antalya, Turkey	Asia	Ripe	Pulp	Oven	PE	Soxhlet	19.8	53.0	20.65	79.36	63.70	15.66
[47]	Hass	Chile, USA	NA	Ripe	Pulp	Freeze	n-hexane	Malaxation	15.7	61.1	15.7	84.0	65.4	18.6
	Hass	Chile, USA	NA	Ripe	Pulp	Freeze	n-hexane	Malaxation	12.9	65.9	12.9	87.1	69.0	18.1
	Hass	Chile, USA	NA	Ripe	Pulp	Freeze	n-hexane	Malaxation	13.1	67.7	13.1	86.8	70.4	16.4
	Hass	Chile, USA	NA	Ripe	Pulp	Freeze	n-hexane	Malaxation	13.7	67.4	13.7	86.3	70.8	15.5
[48]	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS	_	Malaxation	18.5	50.6	19.0	80.9	58.7	22.2
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	19.0	51.4	19.6	80.5	60.4	20.1
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	18.8	51.4	19.2	81.2	61.0	20.2
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	18.4	51.6	19.0	81.5	60.1	21.4
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	19.6	53.1	20.1	83.8	62.3	21.5
	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS	_	Malaxation	21.3	52.6	21.8	78.4	62.6	15.8
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	19.5	52.4	20.1	80.0	61.4	18.6
	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS	_	Malaxation	18.5	50.6	19.0	80.9	58.7	22.2
	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS	_	Malaxation	19.5	50.7	20.1	79.8	60.5	19.3
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	19.4	51.2	19.9	80.2	60.9	19.3
	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS	_	Malaxation	19.8	51.3	20.4	80.1	60.9	19.2
	Hass	Jalisco, Mexico	NA	Ripe	Pulp		_	Malaxation	18.9	51.2	19.4	80.8	60.5	20.3
	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS	_	Malaxation	21.3	51.0	21.8	78.2	60.0	18.2
	Hass	Jalisco, Mexico	NA	Ripe	Pulp	NS		Malaxation	20.6	52.2	21.2	78.7	61.1	17.6
[49]	Hass	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	24.33	48.57	24.67	61.58	61.58	0
	Fuerte	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	18.03	59.00	18.03	65.23	65.23	0
	Hass	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	UAE	23.91	48.57	23.91	61.99	61.99	0
	Fuerte	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	UAE	17.64	58.19	17.64	64.93	64.93	0
	Hass	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	24.22	47.46	24.77	60.63	60.63	0
	Fuerte	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	17.77	59.45	17.77	65.68	65.68	0
	Hass	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	25.29	46.28	25.29	64.15	64.15	0
	Fuerte	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	n-hexane	Soxhlet	18.03	60.14	18.03	68.16	68.16	0
	Hass	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	CO ₂	SCO ₂	21.70	41.57	36.08	54.65	54.65	0
reor	Fuerte	KwaZulu-Natal, South Africa	Africa	Ripe	Pulp	Oven	CO_2	SCO ₂	15.60	50.35	15.60	55.26	55.26	0
[50]	Hass	Morelia, Mexico	NA	NS	Pulp	Freeze	n-hexane	NS	15.23	65.66	15.79	83.59	71.5	12.09

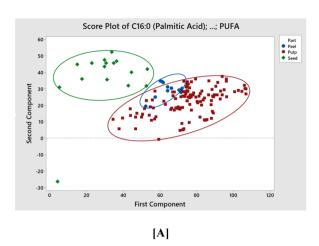
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Table 1. (continued)

Ref	Variables								Fatty Acid Composition ^a (%)					
	Variety	Source	Continent	Maturity	Part	Drying	Extraction Solvent	Extraction Method	C16:0	C18:1	SFA	USFA	MUFA	PUFA
[51]	Hass	Carandaí, Brazil	SA	Ripe	Pulp	Microwave	_	Cold-pressed	25.9	48.0	26.35	72.97	61.4	11.57
	Hass	Carandaí, Brazil	SA	Ripe	Pulp	Oven	_	Cold-pressed	24.7	49.0	25.12	73.28	61.8	11.48
	Hass	Carandaí, Brazil	SA	Ripe	Pulp	Oven	_	Cold-pressed	25.1	47.9	25.56	72.78	60.9	11.88
	Hass	Carandaí, Brazil	SA	Ripe	Pulp	Oven	PE	Soxhlet	26.1	47.4	26.51	72.79	60.6	12.19
	Hass	Carandaí, Brazil	SA	Ripe	Pulp	Oven	PE	Soxhlet	26.0	47.3	26.36	72.91	60.6	12.31
	Hass	Carandaí, Brazil	SA	Ripe	Pulp	Oven	Ethanol	Soxhlet	25.1	46.2	25.55	71.94	59.7	12.24
[52]	Fuerte	Numazu, Japan	Asia	Ripe	Pulp	_	CM	Folch	19.9	59.7	20.8	78.2	65.8	12.4
	Bacon	Numazu, Japan	Asia	Ripe	Pulp	_	CM	Folch	18.6	63.1	19.5	80.0	68.5	11.5
	Hass	Tokyo, Japan	Asia	Ripe	Pulp	_	CM	Folch	20.0	55.6	20.5	78.8	65.2	13.6
	Fuerte	Numazu, Japan	Asia	Ripe	Seed	_	CM	Folch	17.8	27.6	22.1	74.8	34.2	40.6
	Bacon	Numazu, Japan	Asia	Ripe	Seed		CM	Folch	17.7	26.9	23.6	73.5	33.3	40.2
	Hass	Tokyo, Japan	Asia	Ripe	Seed	_	CM	Folch	19.0	25.1	23.8	72.9	29.3	43.6
[28]	Hass	Australia	Australia	Ripe	Pulp	Oven	PE	Soxhlet	25.63	42.59	26.08	73.94	49.88	24.06
	Hass	Mexico	NA	Ripe	Pulp	Oven	PE	Soxhlet	22.59	49.19	22.83	77.17	60.82	16.35
	Hass	New Zealand	New Zealand	Ripe	Pulp	Oven	PE	Soxhlet	20.61	50.97	20.91	79.10	61.28	17.82
	Hass	California, USA	NA	Ripe	Pulp	Oven	PE	Soxhlet	22.24	47.69	23.17	76.84	60.83	16.01
[53]	NS	University Putra Malaysia Campus, Malaysia	Asia	Ripe	Pulp	Oven	n-hexane	Soxhlet	34.48	40.73	35.55	64.44	47.37	17.07
	NS	University Putra Malaysia Campus, Malaysia	Asia	Ripe	Pulp	Oven	CO ₂	SCO ₂	30.88	42.72	31.11	68.79	49.52	19.27
	NS	University Putra Malaysia Campus, Malaysia	Asia	Ripe	Pulp	Oven	Water	UAE	28.12	41.74	28.75	71.26	50.24	21.02
[54]	NS	West Malaysia	Asia	Ripe	Pulp	Oven	PE	Soxhlet	27.63	51.22	29.19	70.82	55.62	15.20
	NS	West Malaysia	Asia	Ripe	Pulp	Oven	PE	Soxhlet	30.37	43.65	31.67	68.35	48.87	19.48
	NS	East Malaysia	Asia	Ripe	Pulp	Oven	PE	Soxhlet	26.41	51.18	27.44	72.57	58.62	13.95

NS = Not specified, SA = South America, NA = North America, IH = isopropanol/n-hexane 4:6, PE = petroleum ether, CM = Chloroform/methanol, - = Not done/no usage.

a After listwise deletion, Pisani extraction method = maceration.



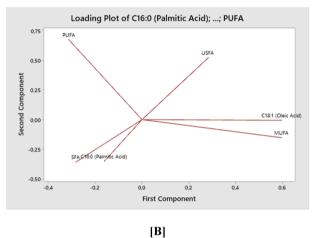


Fig. 2. PCA score plot [A] and loading plot [B] of fatty acids according to part of fruit used.

that follow the vector's direction have an abundance of the two FA parameters. This indicates that whereas MUFA and oleic acid concentrations are higher according to pulp > peel > seed. In contrast, PUFA concentrations are higher according to the order of seed, peel, and pulp.

The results are also in line with those of Green et al. (2022), who noted that the mesocarp had more oleic acid than the fruit as a whole [55,56]. This implies that the component of the fruit that was utilized is significant (in determining the FAP) and ought to be mentioned in any study featuring avocado oil. Nevertheless, the conclusion reflects only one aspect of the complex relationships among the fatty acid variables assessed by PCA. Given that only five publications conducted comparisons of different parts of fruits (Table 1), these journals were chosen to investigate the relationship between the fruit portion and the other FAP. All avocado samples used as FAP comparisons are ripe, and the validity of the finding increases with the number of agreeing references. Table 2 displays the findings

on the influence of a subset of the fruits utilized on the FAP under GC-FID.

The MUFA is found to be highest in the mesocarp/pulp, followed by the peel and seed, in accordance with the PCA result. The peel and mesocarp have the lowest levels of PUFA, whereas the seed has the highest levels. However, we can only conclude that pulp contains higher oleic acid than seed because neither of the five journals compares the oleic acid of pulp to peel as well as seed to peel. In contrast to avocado pulp and seed, the majority of research indicates that avocado peels contain a large quantity of myristic, palmitic, and stearic acid. On the other hand, the greatest source of total lauric, linoleic, and arachidic acid appears to be avocado seeds. This result, however, is limited to the outcome of the agreeing journals and should not be extended generally. It could also vary if additional journals were included.

3.2. Variety, cultivar, and genotypes

Within the kingdom of plants, a cultivar refers to a plant variety that has been created in cultivation by selective breeding, whereas a variant of plant is one that grows and reproduces naturally and deviates in some manner from its regular species due to natural evolution. The PCA result of Table 1 does not identify anything valuable, due to the mass of variations in the samples (Supplementary Data B1 (https://doi.org/10.38212/2224-6614.3561)). For instance, the utilization of various parts of fruits as a variable in PCA, which is observed to be divided into distinct clusters, has the ability to modify the outcome when the labels are converted into the

Table 2. Comparative fatty acid contents according to fruit part used.

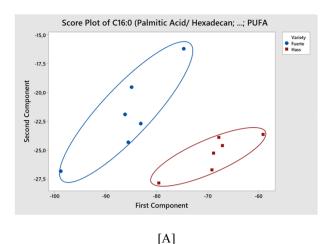
Fatty Acid	Part of	f Fruit U	References		
	Pulp	Seed	Peel		
C12:0 (Lauric Acid)	*	***	**	[39,41,52]	
C14:0 (Myristic Acid)	*	**	***	[39,41]	
C16:0 (Palmitic Acid)	**	*	***	[36,39,41,52]	
C17:0 (Margaric Acid)	*	**	NA	[39,52]	
C18:0 (Stearic Acid)	*	**	***	[39,41,52]	
C18:1n-9 (Oleic Acid)	**	*	NA	[36,39,52]	
C18:2n-6 (Linoleic Acid)	*	***	**	[38,39,52]	
C20:0 (Arachidic Acid)	**	***	*	[36,38,52]	
C20:1 (Eicosenoic Acid)	*	**	NA	[36,39,52]	
C22:0 (Behenic Acid)	*	**	NA	[39,52]	
C24:0 (Lignoceric Acid)	*	**	NA	[39,52]	
SFA	*	**	NA	[39,52]	
MUFA	***	*	**	[39,41,52]	
PUFA	*	***	**	[39,41,52]	

Note: ***>**>, NA = Comparison not available, SFA = Saturated Fatty Acid, MUFA = Monounsaturated Fatty Acid, PUFA = Polyunsaturated Fatty Acid.

independent variable of variety. Despite the setback, data from other researchers indicated a connection between FAP and avocado cultivars/varieties.

Nasri et al., 2021 stated that Ettinger's FAP is more Reed-like, however, not with the Hass and Fuerte variety. The Reed variety has a significantly higher oleic acid percentage than the others, at 61.18% [46]. Reddy et al., 2012 concluded that the Fuerte type was healthier than the Hass variety since it had the highest MUFA:SFA ratio [49]. Jorge et al., 2015 examined the FAP of avocado oil that is already commercially available on the market with two avocado cultivars, Margarida and Hass. The FAP produced by the two cultivars varied, with Margarida exhibiting larger levels of n-6 and n-3 PUFAs, linoleic acid (14.84 g/100 g) and α-linolenic acid (1.25 g/100 g), respectively [26]. Ali et al., 2020 displayed varying genotypes produce varying contents of FA. Hass has a higher percentage of oleic acid than genotypes like Bacon, Monroe, or Zutano. According to this study, however, additional research is needed to prove that FA concentration varies with location and climate [37]. Amado et al., 2019 also found that four avocado cultivars (Hass, Ouintal, Fortuna, and Margarida) have different FAP. The predominant FA in the pulps of the Quintal, Fortuna, and Margarida cultivars was oleic acid; in contrast, the Hass cultivar had a larger proportion of palmitic acid [57]. When the oils of the three locally grown cultivars in west and east Malaysia were compared to the oil from the imported Hass from Australia, Hass was found to have a higher degree of unsaturation in its FA and triacylglycerol compositions [54]. In order to obtain a more definitive outcome about the impact of avocado variety on the distribution of FAP, we do a PCA analysis of the data from Ozdemir et al. (2004) as part of an exploratory study [29].

A dataset of 12 items and 11 dependent variables (palmitic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, arachidic acid, SFA, USFA, MUFA, and PUFA) were used in this PCA (Supplementary Data A2 (https://doi.org/10. 38212/2224-6614.3561)). The Fuerte and Hass avocado varieties are successfully distinguished in a different group (Fig. 3A). PC1 and PC2 are oleic acid (eigenvector score as high as -0.673) and USFA (eigenvector score as high as -0.458). Remarkably, the combined explanation of the variation by the two PCs is up to 99.6%. The Fuerte variety has low levels of palmitoleic acid, linoleic acid, palmitic acid, and PUFA, while the Hass variety has high levels of oleic acid and USFA (Fig. 3B). This finding provides evidence that the FAP assessed by GC-FID



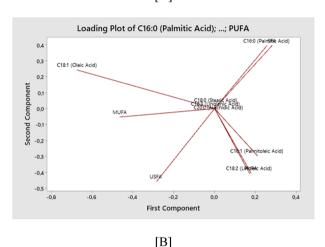


Fig. 3. PCA score plot [A] and loading plot [B] of fatty acids according to variety of avocado [29].

is affected by different avocado cultivars. All six papers that compare avocado varieties (Table 1) were utilized to substantiate this claim and to further examine the FAP that is not addressed by the PCA analysis.

Table 3 presents a further comparison of the FAP of a few commonly consumed avocado types from the six selected journals. The avocado pulp/ mesocarp used in all the chosen publications is ripe. In comparison to Fuerte and Bacon, the majority of the chosen studies concur that Hass has the highest levels of palmitic, palmitoleic, linoleic, and linolenic acid. In comparison to Hass, the majority of the study concurs that Bacon and Fuerte have the highest amount of oleic acid. However, the studies contradict each other with regard to stearic and arachidic acid, suggesting the avocado variety is rarely a decisive influence on these two FA. This fact is demonstrated by the quantity of references that contradict each other. This result is in line with Fig. 3B, which shows that stearic and arachidic acids

Table 3. Comparative fatty acid contents based on different avocado varieties.

Fatty Acid	Varie	ty		References		
	Hass	Fuerte	Bacon			
C16:0 (Palmitic Acid)	***	**	*	[27,29,37,46,49,52]		
C16:1n-7	***	*	**	[27,29,37,46,49]		
(Palmitoleic Acid)						
C18:0 (Stearic Acid)	*	**	*	[27,37,46]		
C18:1n-9 (Oleic Acid)	*	**	**	[27,29,37,49,52]		
C18:2n-6	***	**	*	[27,29,37,52]		
(Linoleic Acid)						
C18:3n-3	**	*	*	[27,29,52]		
(Linolenic Acid)						
C20:0 (Arachidic Acid)	**	*	*	[29,37]		

Note: ***>**>*.

have little effect on the PC used to distinguish between different varieties of avocados. Additionally, linoleic acid has no effect on the PC that is utilized in the grouping. Nonetheless, the Hass type has higher linoleic acid, followed by Fuerte and Bacon, according to the majority of the chosen papers. This finding indicates that while linoleic acid varies throughout the three types, it is not statistically significant when compared to the other PCA-utilized variables. The variation in FA production across different avocado varieties is primarily determined by distinct genetic features that influence the composition and quantity of FA synthesized [58]. Studies have also shown that the upregulation of important genes and regulators involved in FA metabolism, such as PaWRI1, PaACP4-2, and PapPK-β-1, is in line with the overall rise in FA and changes in FA composition during the growth of avocado fruit [59].

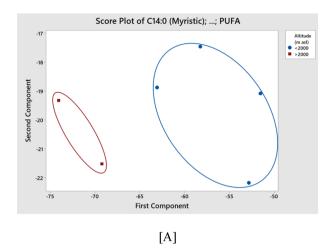
3.3. Different farms and origin

Various farms and growth practices lead to variations in soil composition, elevation, shading, temperature, relative humidity, light intensity, ecological relationships, and many other factors. Many investigations have been conducted to demonstrate that these criteria have an impact on the FAP. For instance, Tan et al., 2017, has compared Hass avocado oil from many countries (Mexico, Australia, New Zealand, and the United States). New Zealand's FA analysis showed 50.97% oleic acid and 61.28% MUFA, which is the highest compared to the others [28]. Five years later, Arpi et al., 2023, investigated the FAP of Hass avocado from an agricultural resource in Takengon, Central Aceh, Indonesia, resulting in a palmitoleic acid rate of 52.9%. However, it is relatively low compared to Tan's [60]. Indrivani et al., 2016, also observed that compared to the other examined avocado oils of Bantul and Purwokerto (Indonesia), avocado oil from Garut, Indonesia, has the largest amount of USFA (68.94%) and oleic acid (47.99%) [42]. However, these studies do not specify which climatic or edaphic (soil condition, aeration, etc.) variables contributed to the FAP shift findings.

An experiment conducted by other researchers looked at a number of the variables influencing various planting locations. Pedreschi et al., 2016 [47], for example, designed experimental conditions using different solar radiation, temperature, humidity, and evapotranspiration. In an attempt to provide the membranes more flexibility, colder growth temperatures have been linked to higher concentrations of MUFA and PUFA [61]. Consistent with the theory, avocados cultivated in regions with the lowest average temperature (14.3 °C) in the study exhibit reduced levels of oleic acid while displaying elevated levels of palmitic and palmitoleic acids [47]. A study on the relationship between FAP and growth altitude was also released in 2015 by Carvalho et al. [40]. The highest levels of linoleic/ palmitoleic index were found in Entrerrios and Rionegro (over 2000 m ASL), while the lowest levels were found in Jerico, Venecia, and Tamesis (2000 m ASL). The data released by Carvalho et al., 2015, were further analyzed using PCA (Supplementary Data A3 (https://doi.org/10.38212/2224-6614.3561)).

There are six total items in the dataset, which includes 13 dependent variables (myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid, linoleic acid, arachidic acid, and gadoleic acid). The mentioned findings are consistent with our PCA results (Fig. 4A), which indicate pattern recognition between avocado oils grown above and below 2000 m ASL.

Oleic acid (with an eigenvector value as high as -0.754) and PUFA (having an eigenvector value as high as -0.600) are the two factors that affect PC1 and PC2, respectively. The samples can be explained by both PCs up to a cumulative 99.0%. Fig. 4B shows that the concentration of oleic acid is higher in avocado oils grown above 2000 m ASL, whereas the concentrations of palmitic acid, palmitoleic acid, and SFA are higher in oils grown below 2000 m ASL. However, this component might affect the FAP because only the latter study discusses the effects of the various ascending elevations. Furthermore, the shift is inconsistent that the author was forced to divide the samples into two groups rather than performing a regression analysis using the concentration versus altitude level data. This hypothesis needs to be further investigated for consistency. It must be verified, for example, with



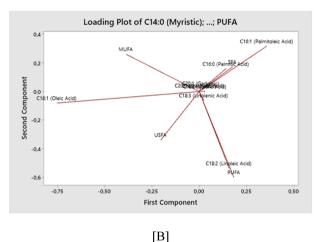


Fig. 4. PCA Score Plot [A] and Loading Plot [B] of Avocado Oil Fatty Acids based on Growing Altitudes [40].

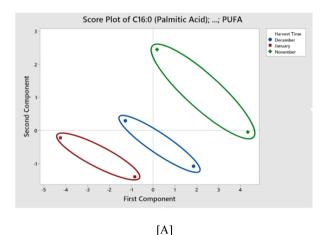
other avocado kinds, different avocado components, or with different variable subsets.

3.4. Harvesting period

The period of harvest correlates with various factors, such as overall temperature, humidity, etc. Ozdemir *et al.*, 2004 [29], have detected changes in the FA composition of avocado oil at various harvesting months (November, January, and December). PCA was used to further examine the data (Supplementary Data A4 (https://doi.org/10.38212/2224-6614.3561)) in relation to the harvesting period. The data consisted of 11 dependent FAP variables. Fig. 5A shows how samples can be clustered based on their harvest month using multivariate clustering, specifically PCA with FAP as parameters. PC1 (oleic acid) with an eigenvector value as high as -0.339 and PC2 (stearic acid) with

an eigenvector value of 0.469 are the variables that play a major role in distinguishing the samples. This suggests that changes in harvest month have the greatest impact on oleic acid and stearic acid, albeit more data is needed in this area. Fig. 5B shows that the order of harvesting avocado oil with the highest palmitic and SFA concentration was November > December > January. The avocado oil with the highest SFA concentration, on the other hand, was harvested following the order of January > December > November. Nevertheless, the study did not include any data regarding crucial factors such as climate, temperature, or humidity, nor did it establish a connection between these factors and the change of FAP.

Another research by Vekiari et al., 2004, found that the Hass variety produced higher oleic acid in January (612 \pm 3.2 g/kg), March (653 \pm 19.9 g/kg), and November (651 \pm 8.5 g/kg); the Fuerte variety showed high oleic acid production in May



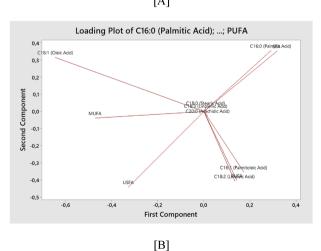


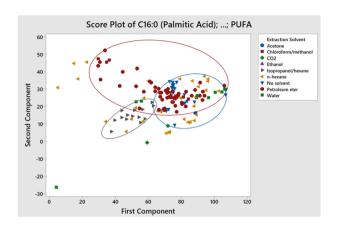
Fig. 5. PCA score plot [A] and loading plot [B] of fatty acids based on different harvesting months [29].

 $(695.3 \pm 1.2 \text{ g/kg})$ and September $(756.8 \pm 10.8 \text{ g/kg})$. These results suggest that the variation in FAP of avocado oils is correlated with the month of harvest [62]. Similar to the previous study, this study also lacks data on important aspects such as climate, temperature, and humidity. Additionally, given that the results differed between the two avocado varieties, the harvest month variable can also be associated with the variety-independent factor. Therefore, more investigation is required to confirm this relationship.

3.5. Extraction of avocado oil

There are many optimizable parameters when it comes to the process of oil extraction, for example, method of extraction, time, temperature, duration, and type of solvent used. It has been reported that the extraction yield and FAP of avocado oil are affected by these processes. SFAs (such as palmitic acid, lauric acid, and stearic acid) typically exhibit lower solubility in organic solvents compared to their unsaturated equivalents. Linoleic acid has greater solubility in n-hexane compared to oleic acid at the same temperature, attributable to the presence of a double bond in linoleic acid. The presence of double bonds diminishes the strength of van der Waals interactions among FA molecules, facilitating the penetration and dissolution by nonpolar liquids [63,64]. Generally, adding isopropanol to n-hexane can help dissolve FA more effectively because it is a polar solvent, and it can interact with the polar parts of the FA molecules.

The examination of the data in Table 1 indicates a pattern in the PCA, distinguishing samples extracted with isopropanol:hexane from those extracted with petroleum ether or those without solvent extraction (Fig. 6A). According to the eigenvector in Fig. 6B, SFAs and palmitic acid are more effectively extracted using an isopropanol:hexane (4:6) solvent, but USFAs are more prevalent with petroleum ether or non-solvent extraction methods. This is consistent with the concept, as the isopropanol: hexane (4:6) mixture exhibits greater polarity than petroleum ether, allowing SFA (including palmitic acid) to dissolve more readily in a polar environment due to the lack of double bonds, in contrast to USFA. The isopropanol:hexane (4:6) system, however, cannot be distinguished from the n-hexane extraction, despite the theoretical premise that SFA solubility is greater in isopropanol:hexane (4:6) than in pure n-hexane. This could be attributable to the large variability of experimental designs used by different authors regarded in this study. Although SFA and palmitic acid are extracted more efficiently



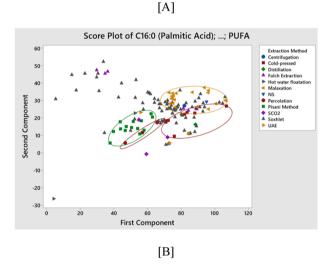


Fig. 6. PCA score plot of fatty acids based on different solvents [A] and extraction methods [B] used (Pisani method = maceration with isopropanol: Hexane = 4:6).

utilizing the isopropanol:hexane (4:6) system compared to petroleum ether, this does not imply that the former is preferable to petroleum ether extraction. In summary, USFAs are recognized as superior to SFAs, especially in reducing bad cholesterol and mitigating cardiovascular disease. Prospective studies and randomized controlled trials have provided robust evidence that substituting dietary SFA for USFA, including MUFA and PUFA, enhances cardiovascular health [65].

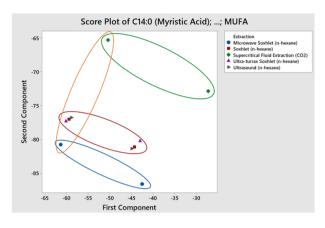
Regarding the extraction method, we noted that several patterns of extraction methods can be differentiated from one another. The Pisani extraction method (maceration) utilizes different solvent systems compared to percolation, which employs n-hexane; yet, both procedures are closely clustered (Fig. 6B). Malaxation approaches are categorized distinctly from the maceration-percolation group, despite there being three malaxation procedures utilizing three different solvent systems (water,

n-hexane, and a solvent-free method available in the dataset). Formulating further conclusions based on this data, however, is challenging due to the variety of the extraction conditions and methodologies applied.

Tan et al., 2018, have detected that different extraction methods can produce significantly different avocado oil FA compositions. SFA results obtained using the Soxhlet (n-hexane) extraction method (35.55%) were significantly higher than subcritical carbon dioxide (SCO₂) and ultrasoundassisted aqueous extraction (UAAE) extracted oil (31.11% and 29.21%, respectively) [53]. This outcome is not consistent with research conducted in 2021 by Pérez-Saucedo et al., which compared the characteristics of avocado oil extracted by centrifugation, ultrasonic-aided technique, and Soxhlet. According to this study, SFA is superior when employing the ultrasound-aided approach as opposed to the Soxhlet method [66]. There are differences between the two approaches; Tan's method provided water to the UAAE system, but Pérez-Saucedo's method did not. Thus, the decrease in SFA in avocado oil may be related to the addition of water. This is in line with research by Li et al., 2019 [44], which found that when compared to the n-hexane extraction and cold pressing procedures, the water generation method had the highest amount of palmitic and palmitoleic acids as well as the largest proportion of USFA (85.72%).

When comparing the extraction methods, Reddy et al., 2012 [49] used Duncan's multiple range test to find that supercritical fluid extraction produced the highest percentage of palmitoleic acid and the lowest percentage of palmitic and oleic acids (among the five compared extraction methods). PCA was performed using the Reddy et al., 2012 dataset, which included 10 items and 8 dependent FAPs (myristic acid, palmitic acid, palmitoleic acid, stearic acid, oleic acid, SFA, USFA, and MUFA), to further explore the relationship between the extraction technique and the FAP shift (Supplementary Data A5 (https://doi.org/10.38212/2224-6614.3561)).

The PCA result (Fig. 7A) shows that the samples are generally grouped into 3 groups. Oleic acid (PC1; eigenvector -0.592) and USFA + MUFA (PC2; eigenvector -0.486) were the parameters that influenced the grouping. On the other hand, oleic acid contributes to the majority of the x-axis (Fig. 7B), which is used to distinguish the samples according to their variety. Two varieties of avocado were used in this research, namely the Fuerte and Hass varieties. The Fuerte variety was categorized according to the oleic acid vector, as indicated by



[A]

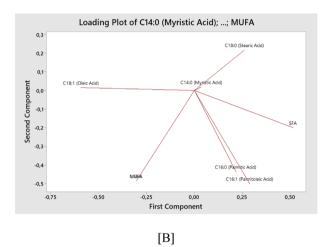


Fig. 7. PCA score plot [A] and loading plot [B] of fatty acids based on different extraction methods [49].

the orange grouping. The Fuerte variety has higher oleic acid content than the Hass variety, which is consistent with Table 3.

When compared to the other four n-hexane extraction methods, the SCO₂ approach stands out as the most unique. The technique produces the least amount of MUFA, USFA, palmitic acid, and palmitoleic acid while preserving the most stearic acid compared to the other solvent extraction methods (Fig. 7B). In contrast to the SCO₂ method, microwave-assisted Soxhlet extraction maintains the highest concentration of MUFA, USFA, palmitic acid, and palmitoleic while preserving the least amount of stearic acid. Based on its FAP, it is not possible to differentiate between the Soxhlet extraction, ultrawave-assisted extraction, and Ultra-Turrax assisted Soxhlet extraction. The outcome implies that certain extraction techniques are similar to one another with respect to FAP when subjected to GC-FID analysis. The research by Santana *et al.*, 2015, also supports this theory. Under

regulated circumstances, avocado oils obtained through mechanical pressing and petroleum ether maceration display comparable FAP [51].

The use of microwaves to collect a higher yield of FA is the subject of additional investigation. Moreno *et al.*, 2003, found notable changes of FAP in four extraction processes (microwave + squeezing, microwave + hexane soxhlation, hexane soxhlation, and acetone maceration). The oils that underwent microwave processing, regardless of whether they were extracted using hexane or squeezing, have a comparable profile [45]. This may be because microwaves quickly loosen and rupture cells, which enhances component extraction by altering the microstructure of biomass [67,68].

Another factor that determines the output of FAP is the temperature of extraction. Diverse FAs may demonstrate unique optimal solubilization characteristics, which can be enhanced through varying temperatures. At reduced temperatures, USFAs such as linoleic acid and oleic acid have greater solubility in organic solvents than SFAs like palmitic acid and stearic acid [69]. Ramirez et al., 2018 designed an experiment to determine the FA compound by using different temperatures and malaxation times. This experiment resulted in the highest FA concentration at 40 °C (120 min) and 50 °C (180 min) [48]. Nevertheless, additional evidence is required to substantiate this assertion, as the aforementioned study exclusively examines the malaxation extraction procedure. Rigorous experimental protocols must be implemented to validate this assertion, utilizing various extraction techniques and conditions. The variation in temperature and extraction duration also correlates with the efficiency and yield of oil extraction [44]. However, as this parameter is not the primary focus of this review, it is not further assessed.

3.6. Other factors (drying process, maturity, and post-ripening period)

Before the oil is produced from avocados, the fruit is dried to remove any remaining water. The drying process has a number of adjustable factors, including temperature, duration, drying instrument, etc. Some studies indicate that the drying procedure does not affect the content of FAP, which is consistent with our PCA findings. For example, Wang et al., 2023 [70], stated that the FAPs observed are similar between 3 pre-dry treatments (ovendried, vacuum-dried, and freeze-dried). Peroxide value, anisidine value, and acid value, however, differ between the three pre-drying treatments, which highly suggests that the drying process

influences the oxidation of avocado oil. In another study, Santana *et al.*, 2015, discovered that although several drying techniques are used, the FAP profiles are identical. The microwave-dried sample, however, exhibits a higher degree of oxidative stability compared to the oven-dried samples in terms of peroxide value [51]. Since GC-FID cannot be used to determine the peroxide, acid, or anisidine values, these variables are not further examined in this review.

Alkatham et al., 2021 [38], reported that oleic acid, linoleic acid, and palmitic acid vary depending on the drying systems (air-dried, oven-dried, microwave-dried, and fresh), which is in opposition to the findings of the previous two research studies. Using the data published by Alkatham et al., 2021, no groups were detected when PCA was utilized. This could be because of the other variables included in the study, like part of the fruit used (seed, peel, and pulp), maturity (ripe and unripe), or the fact that the drying process itself has little to no effect on the FAP. Krumreich et al. (2018) assert that, for the majority of FAs, the highest values were achieved through drying at 40 °C and solvent extraction, with the exception of oleic acid, which reached its peak values when dried at 60 °C under vacuum and extracted via cold pressing, and palmitic acid, for which no differences were noted when drying the pulp at 60 °C [43]. The author ascribes this phenomenon to the increased activity of lipase enzymes [71], which remain active at 40 °C but are nearly inactivated at 60 °C. This enzyme activity leads to oil with elevated acidity, as indicated by the acid value of samples under the author's specified experimental method. A more focused investigation is essential to ascertain if the drying process affected the FAP under GC-FID, considering that acid value is not the primary focus of this review.

Avocado maturity and the post-ripening period are still subjects of controversy as potential factors that may influence fatty acid composition. While still attached to the tree, avocado fruit does not mature (soften into an edible shape), and it ripens approximately 6-10 days after being harvested. The cultivar, maturity level, and other environmental factors, including storage time, temperature, and ethylene exposure, all have an impact on the timing and variability of ripening [72]. According to Pedreschi et al., 2016 [47], the FAP is deemed not affected by postharvest ripening. This finding is also in agreement with the results obtained from Ozdemir et al., 2004 [29]. The assertion aligns with the non-significant PCA (Supplementary Data B2 (https://doi.org/10.38212/2224-6614.3561)) outcome observed in both investigations.

The unripe and ripe avocado oil group did not exhibit distinct clustering, according to the PCA result as well (Supplementary Data B1 (https://doi. org/10.38212/2224-6614.3561)), suggesting that the two oils cannot be distinguished solely on this category. These findings might not accurately represent the value because of the various factors that affect the PCA plot. Contradicting the research and prior PCA findings, Villa-Rodríguez *et al.*, 2011 [73] observed a significant increase in monounsaturated and saturated FA during avocado ripening while PUFA content decreased (p < 0.05). This observation shows that other factors might influence the PCA result, implying that more research on the subject is required.

3.7. Study limitations and evaluation of avocado oil

This review aims to assess whether GC-FID can differentiate avocado oils based on several parameters, in conjunction with PCA. The coupling of GC-FID with PCA enables reliable detection of FA variations with different fruit sections (Fig. 2). This outcome concerning the fruit component, however, achieved the best clustering when all 148 data points were analyzed in the PCA, suggesting that the other variables in the data have minimal or are not as significant as the fruit component variable. Furthermore, several aspects related to the extraction process, such as temperature settings, solvent systems, and extraction methods, are noted to affect the variation of FAPs identified by GC-FID (Fig. 6). Regarding avocado varietals, growth origins, maturity, and drying process, our PCA indicates no significant results (Supplementary Data B1 (https:// doi.org/10.38212/2224-6614.3561)), which can be attributed to the variability in experimental designs utilized among the studies. It is also important to note that PCA serves as an exploratory tool for pattern recognition and dimensionality reduction; while it can reveal clustering and trends, it does not establish causal relationships or independently authenticate samples.

Upon evaluating the PCA results of multiple experiments separately, there is compelling evidence that the variations in varietals, growing area origins, and harvesting months can be differentiated using GC-FID. However, significant aspects, including differences in soil composition, elevation, shade, temperature, relative humidity, light intensity, ecological interactions, and several other elements, are often neglected or unreported in the primary study materials. This is unfortunate, as these supplementary elements considerably affect the FAP of avocado oil. The identical circumstance occurs

during the avocado harvesting period. Crucial elements, such as climate, temperature, and humidity, are frequently not reported despite their significance to the subject matter. A more profound explanation cannot be derived without the data.

Concerning avocado extraction, we encountered significant heterogeneity in data, including extraction method, time, temperature, duration, and type of solvent employed. Each author employs distinct experimental designs; hence, even when comparing the extraction method and solvents used, deriving a conclusion is challenging. A more controlled experimental design is necessary to validate, or a more concentrated review must be conducted to ascertain the impact of these variables. While this review method is robust to conclude which factors matter more to FA composition (in this case, part of avocado and extraction procedures), a significant variety of the data can result in inconsistencies in the outcomes. The result may be different, however, if more data points, references, or databases are incorporated into the study. Furthermore, we must eliminate the majority of the identified FAs (Supplementary Data A1 (https://doi.org/10.38212/2224-6614.3561)) due to the inconsistency in FA datasets among the papers. Inclusion of additional FA variables may alter the identification of the most distinguishing FA between samples.

GC-FID is considered a principal tool for analyzing the FA composition of oil, alongside other quality assessments (such as acid value and peroxide value). PCA has been employed to identify adulterants in various culinary oils, as different plant species yield distinct FA compositions [74]. However, researchers must note that many internal characteristics inside avocado species might significantly distort the results of GC-FID combined with PCA when evaluating the approach for identifying or detecting adulterants in avocado oil. These sources of natural variability may resemble compositional shifts caused by adulteration, potentially leading to misclassification or false positives. Consequently, these potentially distorting parameters must be thoroughly evaluated, documented, or at a minimum acknowledged in studies pertaining to the identification of avocado oil utilizing this method.

The avocado oil quality is generally divided into 4 categories, which are extra virgin, virgin, pure, and blends [72]. The categorization is inherently subjective (Table 4), especially regarding fruit quality, as the percentage of rot is not specified. Although the degree of rot correlates with fruit maturity, which our PCA results deem insignificant (Supplementary Data B1 (https://doi.org/10.38212/2224-

6614.3561)), further investigation is necessary to determine whether this variable could distort the findings on FAP. Furthermore, this necessity arises because most studies referenced in this review focus on ripe and unripe avocados rather than those that are rotten. One study by Green and Wang, 2023, has designed a study to evaluate the effect of rotten avocados to FAP [22]. The PCA results, like the prior review, indicated that the samples cannot be categorized according to the fruit's maturity. This outcome is, nonetheless, subjective, as it depends on the categorization of decaying fruit; the author did not disclose the criteria employed to determine if the fruit meets the Grade 4 categorization, nor did the studies referenced in this review establish a criterion for distinguishing ripe from unripe avocados.

The parameters, including FFA content (reported as % oleic acid) and the FAP standard of extra virgin avocado oil, can be significantly influenced by internal factors such as the fruit variety or the specific section of the fruit utilized. Consequently, these factors must be articulated and evaluated. Certain avocado cultivars, like Bacon and Fuerte (Table 3), may possess higher levels of oleic acid compared to the Hass variety. It is of the utmost importance to emphasize that oleic acid is regarded as a standard quality parameter and should not be strictly restricted to a narrow range. This concern must also be applied to the other FAP.

The variety of avocado oil also poses a considerable challenge to its standardization. Palmitic acid is more abundant in the peel than in the pulp and seed of the avocado, and it is also more prominent in the Hass variety compared to the Fuerte and Bacon varieties. Proposed CODEX standards, for instance, restrict the palmitic acid content to 11.00–26.0% [21]. Research conducted by Green and Wang in 2023 has devised an experiment utilizing different harvest times, avocado grades,

planting origins, and specific parts, particularly pulp only or whole fruit [22]. Out of the 68 sample variances, 1 sample fails to meet the palmitic acid restriction, 2 samples fail to meet the palmitoleic acid limitation, and 4 samples fail to meet the restrictions for 10-Heptadecanoic acid and linoleic acid (Table 5). The author also concluded that standards for FAP need to be modified to account for oils derived from the entire fruit as opposed to solely the mesocarp, or for grade 1 versus grade 4 fruits. This may mitigate food waste and the byproducts of avocado oil production by utilizing lower-grade whole avocados for oil extraction instead of discarding them entirely or disposing of the seed and exocarp.

The data in this review also shows that myristic acid (C14:0) did not meet standards in 18 out of 70 samples, suggesting that differences were caused by how the samples were extracted and the types of avocados used. Similarly, palmitic acid (C16:0) showed inconsistencies in 26 of 148 samples, pointing out the difficulties in achieving uniformity. Palmitoleic acid (C16:1) did not meet the standards in 10 out of 124 samples, while 10-heptadecanoic acid (C17:1) was not compliant in 11 out of 21 samples, and linoleic acid (C18:2) exceeded the limits in 34 out of 138 samples. Substantial noncompliance was identified for linolenic acid (C18:3), with 54 out of 130 samples failing to meet standards. These findings highlight the necessity to amend fatty acid standards to include oils derived from whole fruits, different grades, and various extraction methods, thereby promoting sustainability and minimizing food waste.

While the proposed CODEX standards aim to establish compositional benchmarks for avocado oil, the observed non-compliance in authentic samples suggests that natural variability may confound authentication assessments. PCA based on FAP is effective for differentiating oils by

Table 4. Standards of avocado oil related to discussed factors [72].

	Extra Virgin	Virgin	Pure	Blends
Quality of Fruit	Minimal level of rot	Some rots are tolerated	Rots are not important	Specification depends on claim and labeling
Extraction	Mechanical extraction (no solvent extraction)	Mechanical extraction (no solvent extraction)	Not specified	Not specified
Free fatty acid (% as oleic acid)	≤0.5%	0.8-1.0%	≤0.1%	As specified/claimed
Fatty acid composition (% relative values)	Palmitic acid (10–25%) Palmitoleic acid (2–8%) Stearic acid (0.1–0.4%) Oleic acid (60–80%) Linoleic acid (7–20%) Linolenic acid (0.2–1%)	_	_	_

Table 5. Quantity of samples that fail to comply with the proposed CODEX standards [21].

Fatty Acid	Notation	Proposed CODEX Standard (relative %)	Samples in this review that do not meet the criteria (Supplementary Data A1 (https://doi.org/10.38212/2224-6614.3561))
Myristic Acid	C14:0	ND-0.3	18/70
Palmitic Acid	C16:0	11.0-26.0	26/148
Palmitoleic Acid	C16:1	4.0 - 17.1	10/124
Margaric Acid	C17:0	ND-0.3	7/27
10-Heptadecanoic acid	C17:1	ND-0.1	11/21
Stearic Acid	C18:0	0.1-1.3	39/124
Oleic Acid	C18:1	42.0-75.0	30/148
Linoleic Acid	C18:2	7.8-19.0	34/138
Linolenic Acid	C18:3	0.5-2.1	54/130
Arachidic Acid	C20:0	ND-0.7	25/88
Gadoleic Acid	C20:1	ND-0.3	25/58
Behenic Acid	C22:0	ND-0.5	2/18
Tetracosanoic Acid	C24:0	ND-0.2	4/18
Nervonic Acid	C24:1	ND-0.2	_

ND = Not detected.

cultivar, origin, or processing method, but the specificity required to detect adulteration against other oils requires further study, particularly in borderline cases. PCA using FAP data alone may misclassify oils due to overlapping natural variation, fail to identify minor blends or targeted fraud (e.g., deodorized oils), and does not rely on unique biomarkers exclusive to avocado oil. Complementary techniques, such as Fourier Transform Infrared Spectroscopy (FTIR), Nuclear Magnetic Resonance (NMR), and isotope ratio mass spectrometry, could be explored to aid in the precise detection of adulteration and compositional anomalies beyond the resolution of conventional FAP-based screening.

These findings also underscore the importance of aligning avocado oil specifications with established standards for other edible oils, such as those defined for olive oil. Notably, frameworks like those from the International Olive Council (IOC) and CODEX STAN 33-1981 incorporate thresholds for FA composition and oxidative stability to ensure product authenticity and shelf-life consistency. Harmonizing avocado oil standards with such benchmarks would enhance regulatory coherence strengthen consumer protection across sectors. Nevertheless, this pursuit presents a regulatory trade-off. Expanding FAP thresholds to include oils derived from whole fruits, lower-grade avocados, and varied extraction techniques may advance sustainability and reduce agricultural waste. However, broader compositional allowances risk weakening the analytical precision of FAP-based authentication and complicating the detection of adulteration. To address this concern, any standard revision should be accompanied by robust traceability mechanisms

and complementary analytical techniques, such as triacylglycerol profiling, sterol quantification, or stable isotope analysis.

To facilitate future harmonization and improve transparency, we recommend establishing a comprehensive reference database of authentic avocado oil fatty acid profiles (FAPs). This database would encompass rigorously validated samples produced from both mesocarp-only and whole-fruit inputs, reflecting diverse cultivars, fruit grades, and extraction methods. Such a resource would serve as a scientific benchmark for assessing compositional variability and refining authenticity criteria within regulatory and industrial settings.

4. Conclusion

The FAP of avocado oil is primarily determined by which part of the fruit is used. Avocado peels contain high levels of myristic, palmitic, and stearic acid, while the seeds are the best source of lauric, linoleic, and arachidic acid. The avocado variety also impacts FAP, though to a lesser extent for some acids. Most research agrees that Hass avocados contain higher quantities of linoleic, palmitic, and linolenic acid compared to Fuerte and Bacon. The majority of studies also show that Bacon contains the largest amount of oleic acid. However, findings on stearic and arachidic acid appear to contradict one another, suggesting that variety has little significant impact on these two FAs.

Additional variables, such as planting origin, harvest timing, ripeness level, and drying method, have been noted in relation to FAP variation. Observational patterns imply that planting location and harvesting month may correspond to

compositional shifts, although conclusive trends remain limited. Conversely, the influence of ripening duration and drying technique is not wellsupported across the included studies, indicating the need for further investigation. While the proposed CODEX standards aim to establish compositional benchmarks for avocado oil, instances of non-compliance among authentic samples imply that natural variability may confound purity assessments and should be accounted for in regulatory evaluations. These findings also highlight the importance of revising FA standards to accommodate avocado oils produced from whole fruits, varied grades, and diverse extraction practices, an effort that may promote sustainability and reduce food system waste.

Ethical clearance

In the process of writing this review, no animals were hurt.

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