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A PRACTICAL APPROACH TO ASSESS THE OXIDATIVE STABILITY OF COSMETIC BUTTERS: QUALITY CONTROL PERSPECTIVE

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Abstract: Oxidative stability is an important parameter determining the quality of fat products. The oxidation time (OT) of cosmetic butters measured using RapidOxy can be predicted using simple equations based on unsaturated fatty acid / saturated fatty acid content (UFA/SFA), calculated oxidizability (CoxV) and calculated iodine values (CIV). This predictive capability increases the efficiency of the quality control processes for these products. Similar predictive relationships also exist for other fat-based products, such as oils. This simple but innovative approach to assessing the oxidative stability of fats is quick, eliminates chemical reagents and minimizes waste production, making it both cost-effective and environmentally friendly. It is in line with the idea of green chemistry and sustainable development, making it the preferred technique in modern chemical analytics.

Keywords: oxidative stability, RapidOxy, fatty acid, cosmetic butter, quality prediction, green chemistry.

1. INTRODUCTION

Oxidative stability is an important parameter for assessing the quality of fat-based products and their resistance to oxidation during their manufacture, storage and warehousing. The oxidative stability also gives information about the shelf life of raw materials and end products exposed to autoxidation. Lipid oxidation results in, among other things, the oxidation of unsaturated fatty acids, proteins and vitamins, the production of toxic compounds, and changes in the color and odor of the product. The end products of lipid oxidation are highly reactive and may have adverse effects

on human health. For cosmetic products, these may include allergic skin reactions or skin irritation.

Vegetable butter comes from such natural sources as nuts, seeds and grains. They can be applied directly onto the skin or hair, or be an ingredient in such cosmetic preparations as soaps, creams, body lotions, lip balms, etc. They make the skin and hair smooth and soft, helping to soothe rough and irritated skin or the brittleness of hair. Vegetable butters are mainly composed of triglycerides and unsaponifiable matter, which is strongly affected by the method of butter production and origin [Janeš and Kočevar Glavač 2018; Poljšak and Kočevar Glavač 2021]. Butters of the highest quality are obtained through cold pressing and CO_2 extraction; they are not refined and therefore contain no residual solvents. However, some butters used for the cosmetics industry are hydrogenated oils, meaning that they were created by hydrogenating natural vegetable oils. Butters are rich in unsaturated fatty acids, which are crucial active ingredients, responsible for the specific properties of butters, but also making them highly susceptible to oxidation [Thomsen, Chow and Sapijaszko 2021].

There are different methods available to investigate the oxidative stability of fat-containing products [Shahidi and Wanasundara 1996; Barriuso, Astiasarán and Ansorena 2013; Şahin 2019; Abeyrathne, Nam and Ahn 2021]. They include storage tests at room temperature or the accelerated conditions e.g. in the thermostatic Schaal Oven test. Storage tests do not require specific equipment, but they are time-consuming and make it difficult to maintain repeatable storage conditions. Moreover, it is necessary to determine changes in various parameters, such as color, odor, acidic value, *p*-anisidine value, peroxide value, thiobarbituric acid reactive substances, conjugated dienes or triens, which means that they require the use of chemical reagents. The Rancimat [Ciemniewska-Żytkiewicz et al. 2014; Maszewska et al. 2018; Symoniuk et al. 2022a] and RapidOxy [Difonzo et al. 2022; Hassan, El-Sayed Shaltout and Abo-Elyazaid 2022; He et al. 2023; 2025] tests are some other examples of accelerated assays widely used for testing the oxidative stability of fat-containing products. The Rancimat method is based on the conductivity measure of the volatile compounds formed during oxidation. An alternative is the RapidOxy method, utilizing the drop in oxygen pressure caused by the consumption of oxygen by a sample kept at a constant higher temperature. The induction time obtained in both methods is used as an indicator of the oxidative and storage stability of fats, oils and other products. Compared to other methods of oxidative stability and storage testing, the Rancimat and RapidOxy procedures are simple, require only a small sample amount, do not require reagents and are usually faster, thus saving time and money. Fourier transform near-infrared (FT-NIR) [Gliszczyńska-Świgło, Jajor and Kmiecik 2018] and midinfrared (FT-IR) spectroscopies [Guillén and Cabo 2002; Cebi et al. 2017; Mahboubifar et al. 2017; Naz and Saeed 2018; Quantum Analytics 2018; Hu et al. 2019; Kunyaboon et al. 2021], combined with chemometric analysis, such as partial least squares (PLS) regression, have also been proposed as quick, easy

and precise methods to study oils and fat-containing products, but PLS requires the determination of parameter indicating oxidative stability using traditional methods.

Fatty acid composition (FAC) is a key factor for the quality and shelf-life of fat-containing products. Gas chromatography (GC) is the primary technique used for fatty acid analysis. Its accuracy, reproducibility, selectivity and sensitivity reduce the number of samples to be analyzed and thus the consumption of reagents and the amount of chemical waste, decreasing the adverse environmental impact of chemical analytics. As finding fast and sustainable solutions for quality control is of crucial importance in the context of, for example, environmental protection, this study aimed to verify the possibility of predicting the oxidative stability of cosmetic butters based on the parameters calculated from the FAC of these butters, namely the total amount of unsaturated/saturated fatty acids (UFA/SFA), the calculated oxidizability value (CoxV) and the calculated iodine value (CIV). To achieve these goals, FAC was determined using GC while RapidOxy was used as the exemplary method to measure the oxidation time (OT) of cosmetic butters. The relationships between the OT and parameters calculated from the FAC of butters are shown. Moreover, based on literature data for some vegetable oils, we confirmed that similar predictive relationships may also exist for other fat-based products. To the best of our knowledge, this is the first such approach to assess product oxidative stability.

2. MATERIALS AND METHODS

2.1. Materials

Samples of cosmetic butters (100 g) were purchased in an online store: almond butter (INCI *Prunus Amygdalus Dulcis*, *Hydrogenated Vegetable Oil*), vanilla butter (INCI *Helianthus Annuus Seed Oil, Hydrogenated Vegetable Oil, Vanilla Planifolia Oil*), macadamia butter (INCI *macadamia (Macadamia Ternifolia) Hydrogenated Seed Oil*), kokum butter (INCI *Garcinia Indica*), illipe butter (INCI *Shorea Stenoptera*), and cocoa butter 100% natural (INCI *Theobroma Cacao*). They were distributed by Sunniva Med (Wolsztyn, Poland). The samples for each determination were taken in a way that ensured representativeness for the whole study material.

Sodium methoxide and standards of fatty acids were purchased from Sigma-Aldrich (Steinheim, Germany). The standard for α-tocopherol was purchased from Aldrich (Steinheim, Germany), and γ-, δ-tocopherols were from Sigma (St. Louis, USA). Methanol, acetonitrile, n-hexane and 2-propanol were from Chempur (Piekary Śląskie, Poland). All solvents were HPLC grade.

2.2. Determination of the oxidative stability of cosmetic butters

The oxidative stability of cosmetic butters was measured using RapidOxy (Anton Paar, Blankenfelde-Mahlow, Germany). The method is based on accelerated oxidation of the sample at an elevated temperature and oxygen pressure. Samples of each butter (about 2.000 g) were tested in duplicate at 70°C under a pressure of 700 kPa. The results were expressed as the time (in minutes) required to consume 5% of the initial oxygen content present in the test chamber. For this study, this time was called the oxidation time (OT).

2.3. Determination of vitamin E

The content of vitamin E in cosmetic butters, as a sum of α-, β-, γ- and δ-tocopherols, was determined by high-performance liquid chromatography (HPLC) as described by Gliszczyńska-Świgło and Sikorska [2004]. Briefly, three samples of butters were dissolved in 2-propanol (1:20 w/v) at room temperature. HPLC determination of vitamin E was performed using a Waters 600 HPLC equipped with a Symmetry C18 column (150 mm x 3.9 mm, 5 μm) fitted with a μBondapak C18 cartridge guard column (Waters, Milford, MA, USA). A mobile phase consisting of acetonitrile and methanol (1:1) at a flow rate of 1 mL/min was used. Each sample was injected in duplicate. Tocopherols were detected using a Waters 474 scanning fluorescence detector set at 295/325 nm as excitation and emission wavelengths and identified by comparing their retention times with those of the corresponding standards. A Waters 996 photodiode array detector was used to confirm identification of the compounds by their UV spectra. Quantification of α-, (β+γ)- and δ-tocopherols was carried out using the external standard method [Gliszczyńska-Świgło and Sikorska 2004].

2.4. Determination of fatty acids

The fatty acids profile was determined in two samples of each cosmetic butter by gas chromatography-flame ionization detection (GC-FID). Fatty acid methyl esters (FAME) were prepared using the transesterification method [PN-ISO 12966-2011] and analyzed by applying the Agilent 7820A GC equipped with a capillary column BPX-70 (60 m x 0.25 mm x 0.25 μm). The temperature gradient used was from 140ºC to 240ºC with a temperature rate of 6˚C. The carrier gas was helium at the flow rate of 0.8 mL/min. The injection port was set to 250ºC, split mode 50:1, and the FID was set to 270ºC. Each sample was injected in duplicate. Commercially available standards of pure compounds (Supelco 37 component FAME mix) were used for FAME identification. The quantitation was based on a relative percentage basis.

2.5. Calculated oxidizability (CoxV) and iodine (CIV) values

The calculated oxidizability value (CoxV) of cosmetic butters was computed from the formula proposed by Fatemi and Hammond [1980] as described by Hassanien et al. [2014]: CoxV = (C18:1 + 10.3*C18:2 + 21.6*C18:3)/100, where: C18:1 (oleic acid), C18:2 (linoleic acid), and C18:3 (linolenic acid) in %.

The calculated iodine value (CIV) of cosmetic butters was obtained from the FAC according to AOCS Official Method Cd 1c-85 [2009]: CIV = (0.95*C16:1) + (0.86*18:1) + (1.732*18:2) + (2.616*18:3) + (0.785*20:1) + (0.723*22:1), where: C16:1 (palmitoleic acid), C18:1 (oleic acid), C18:2 (linoleic acid), C18:3 (linolenic acid), C20:1 (eicosenoic acid), and C22:1 (erucic acid) in %.

2.6. Statistics

The data are presented as mean ± SD. The linear relationship between two sets of data (Pearson correlation coefficient, *r*) was determined using Statistica 13.3 (2017) (Stat-Soft, Inc., Tulsa, OK, USA). The significance of the Pearson correlation coefficient was determined at $\alpha = 0.05$. Principal component analysis (PCA) was performed for standardized data for the main fatty acids and parameters calculated from the FAC of cosmetic butter. Leave-one-out cross-validation was used. The analysis was performed by Unscrambler 7.0 software (CAMO, Oslo, Norway). The Pearson correlation coefficient distribution table was used to determine the significance of the correlations obtained in the PCA.

3. RESULTS AND DISCUSSION

Cosmetic butters are source of unsaturated fatty acids, which makes them highly prone to oxidation. The composition of fatty acids, the content of vitamin E, OT, CoxV and CIV of the cosmetic butters tested in this study are presented in Table 1. The FAC of the tested butters was generally consistent with the data published in the literature for butters or their oil counterparts. The main fatty acids in cocoa butter (CB) are stearic acid (C18:0) (34–40%), followed by oleic acid (C18:1) (29–38%) and palmitic acid (C16:0) (24–34%) [Kalse, Sawant and Swami 2021; Diomandé 2022]. These fatty acids content in CB of the present study was 37.0%, 33.3%, and 26.0% for C18:0, C18:1 and C16:0 fatty acids, respectively. The kokum butter (KB) mainly contains C18:0 fatty acid (49–62%) and C18:1 fatty acid (30–42%) [Matulka et al. 2016; Kalse, Sawant and Swami 2021], which corresponds to the content of these fatty acids in KB tested (59.3% and 37.6%, respectively). The main fatty acid in macadamia butter (MB) of the present study is C18:1 fatty acid (50.1%). It also dominates in macadamia oil (41-58%) [Dubois et al. 2007; Aquino-Bolaños et al. 2017; Gliszczyńska-Świgło, Jajor and Kmiecik 2018].

Table 1. Fatty acid composition, CoxV, CIV, OT and vitamin E content of cosmetic butters

Parameter	Unit	AB	VB	CB	KB	MB	IB
C12:0	(%)[1]						0.21 ±0.01
C14:0	(%)						0.34 ±0.02
C16:0	(%)	8.97 ±0.13	8.28 ±0.05	25.98 ±0.05	1.70 ±0.06	11.22 ±0.04	12.79 ±0.00
C16:1	(%)			tr		7.50 ±0.04	
C18:0	(%)	23.03 ±0.42	22.80 ±1.73	37.02 ±0.10	59.31 ±0.08	22.77 ±0.20	47.77 ±0.05
C18:1	(%)	50.28 ±0.18	36.75 ±0.99	33.34 ±0.05	37.62 ±0.05	50.10 ±0.23	36.33 ±0.05
C18:2	(%)	17.76 ±0.37	32.17 ±0.78	2.57 ±0.02	0.71 ±0.07	5.12 ±0.01	1.97 ±0.00
C20:0	(%)			1.08 ±0.01	0.50 ±0.03	1.36 ±0.03	0.44 ±0.02
C20:1	(%)				0.17 ±0.02	1.11 ±0.02	0.15 ±0.01
C22:0	(%)					0.83 ±0.05	
Total UFA	(%)	68.04	68.93	35.91	38.49	63.82	38.46
CoxV		2.33	3.68	0.60	0.45	1.03	0.57
CIV		74.00	87.33	33.13	33.71	52.82	34.78
OT	min	103.7 ±0.3	77.2 ±0.4	635.3 ±42.6	620.7 ±44.2	232.8 ±5.6	435.2 ±7.3
Vitamin E	mg/100 g	115.3 ±1.3	107.0 ±3.3	29.9 ±1.2	1.34 ±0.29	118.4 ±6.1	1.24 ±0.13

AB – almond butter, VB – vanilla butter, CB – cocoa butter, KB – kokum butter, MB – macadamia butter, IB – illipe butter, tr – trace amount (<0.01%), UFA – unsaturated fatty acids, CoxV – calculated oxidizability value, CIV – calculated iodine value, OT – oxidation time.

Source: own study.

Vanilla (VB), almond (AB) and MB butters contained the highest amounts of UFA (63.8–68.9%), thus their CoxVs (1.0–3.7) were considerably higher than those calculated for other butters (0.5–0.6). Their CIV, an index of unsaturation, ranged from 52.8 to 87.3. These butters occurred to be the most susceptible to oxidation. Their stability expressed as OT was considerably lower (77–229 min) than measured for illipe butter (IB), KB and MB (430-605 min) for which the CIVs were 33.1–34.8. Based on the CoxV, which is generally used to evaluate the tendency of fat-containing products to undergo autoxidation, the oxidative stability of six tested cosmetic butters was in the following order KB>IB≈CB>MB>AB>VB (a lower CoxV means better oxidative stability). This order generally corresponds to the results of RapidOxy. The oxidative stability of cosmetic butters decreases as the

CoxV increases. Among the butters analyzed, VB was the most susceptible to oxidation due to the highest content of C18:2 fatty acid, which, together with C18:3 fatty acid (not observed in our samples), has the highest relative oxidation rate [Fatemi and Hammond 1980].

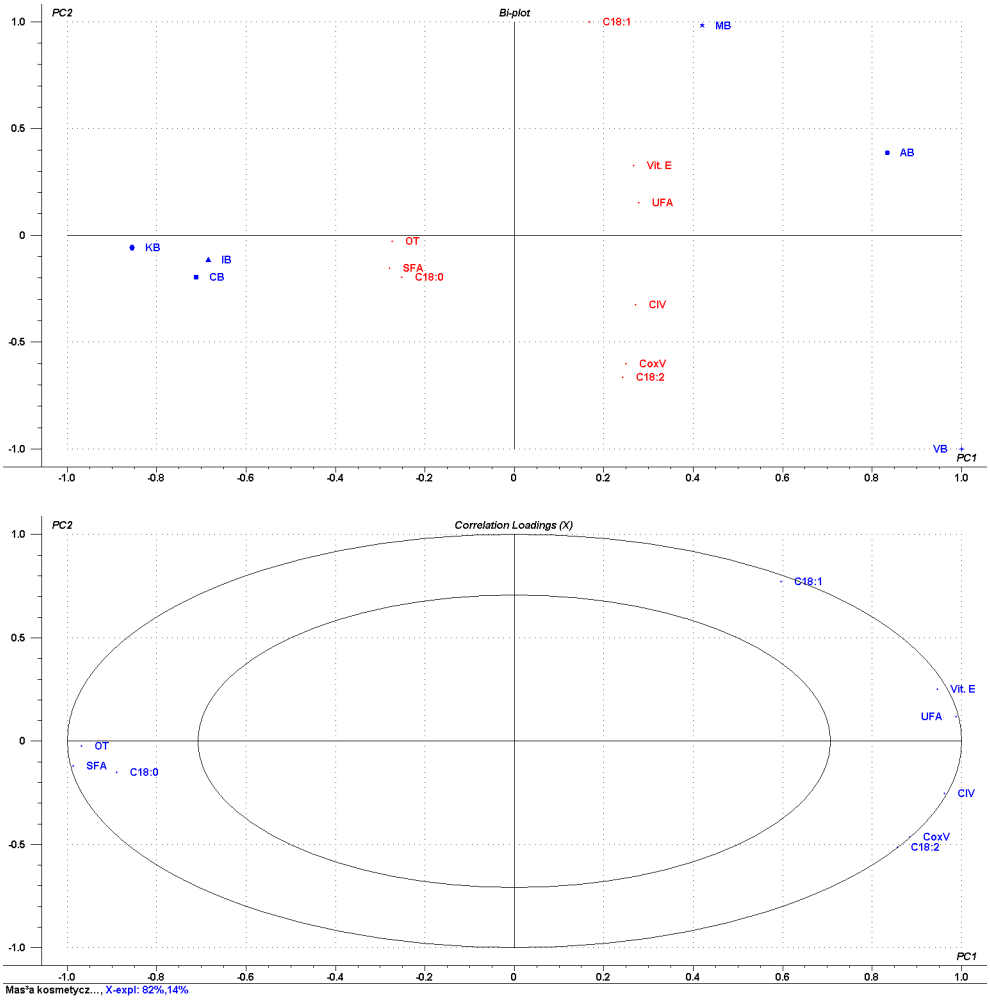

Fig. 1. Biplot from PCA of C18:0, C18:1, C18:2 fatty acids, SFA, UFA, CoxV, CIV, vitamin E content and OT of cosmetic butters

CB – cocoa butter, IB – illipe butter, KB – kokum butter, MB – macadamia butter, AB – almond butter, VB – vanilla butter, C18:0 – oleic acid, C18:1 – linoleic acid, C18:2 – linolenic acid, UFA – unsaturated fatty acids, SFA – saturated fatty acids, CoxV – calculated oxidizability value, CIV – calculated iodine value, Vit. E – vitamin E, OT – oxidation time.

Source: own study.

The above findings were confirmed by PCA performed for the parameters tested, namely: the contribution of C16:0, C18:0, C18:1 C18:2 fatty acids in total fatty acid contents, UFA, SFA, CoxV, CIV, vitamin E content and OT. The first and second principal components (PC1 and PC2) explained 74% and 13% of the total data variance, respectively. However, the values of correlation loadings for C16:0 fatty acid were lower than 0.63 (-0.279 for PC1, 0.270 for PC2) indicating the insignificant impact of this parameter on the differentiation of cosmetic butters, so it was excluded from the subsequent PCA. The value of 0.63 comes from the Pearson correlation coefficient distribution table for a degree of freedom of 8 and a significance level of 0.05. Figure 1 shows the PCA biplot (scores and loadings) and correlation loadings. PC1 and PC2 explained 82% and 14% of the total data variance, respectively. The cosmetic butter samples were spread along the PC1 and PC2 axes, from positive to negative values. The PC1 was positively and strongly correlated with UFA (0.987), CIV (0.962), vitamin E (0.946), CoxV (0.884), C18:2 fatty acid (0.857), and negatively with SFA (-0.987), OT (-0.968) and C18:0 fatty acid (-0.890). The C18:1 fatty acid content (0.773) contributed significantly to PC2. Considering PC1, KB, CB and IB (butters with higher oxidative stability) were separated from MB, AB and VB (less oxidatively stable butters, as determined by RapidOxy). KB, CB and IB had a higher percentage of SFA (61.5–64.5%), including C18:0 fatty acid (37.0–59.3%), than MB, AB and VB (31.1–32.0% of SFA and 22.8–23.0% of C18:0 fatty acid). Considering PC2 and less oxidatively stable cosmetic butters, VB (the least oxidatively stable butter) was separated from MB and AB due to its highest amount of C18:2 fatty acid, the highest CoxV and CIV. Both AB and MB had a similar high percentage of C18:1 fatty acid (50.3 and 50.1%, respectively Tab. 1).

A statistically significant positive linear correlation was observed between OT and C18:0 fatty acid content ($r = 0.827$) and a negative linear correlation between OT and C18:2 fatty acid content ($r = -0.815$). The linear correlation coefficients for C16:0 and C18:1 fatty acids (other fatty acids common to all the butters tested) were $r = 0.311$ and $r = -0.612$, respectively, but they were statistically insignificant.

Figure 2 shows the relationships between OT and UFA, SFA, CIV or CoxV. A statistically significant linear relationship between OT and UFA or SFA content ($r = -0.960$ and $r = 0.960$, respectively) (Fig. 2A), as well as the decimal logarithm of OT and CIV ($r = -0.987$) or CoxV ($r = -0.939$) was found, although a better fit of the relationship curve was obtained for OT and CIV or CoxV when a non-linear equation was used (Fig. 2B and 2C). Using the linear models shown in Figures 2B and 2C and assuming that oxidatively stable butters are those for which the oxidation time in the conditions applied in the RapidOxy test is at least 8–10 hours (480–600 min), it was calculated that butters with CoxV of 0.18–0.52 and CIV of 31.0–36.8 can be considered products with good oxidative resistance. However, it should be noted that in assessing the oxidative stability of fat-containing products, in addition to the fatty acid composition, which determines the CoxV and CIV, other

compounds such as antioxidants, e.g. tocopherols, may also be important. The content of vitamin E in the tested butters ranged from about 1.2–1.3 mg/100 g in IB and KB through about 30 mg/100 g in CB up to about 107–118 mg/100 g in VB, AB and MB. No significant relationship was found between OT and vitamin E content in the tested butters.

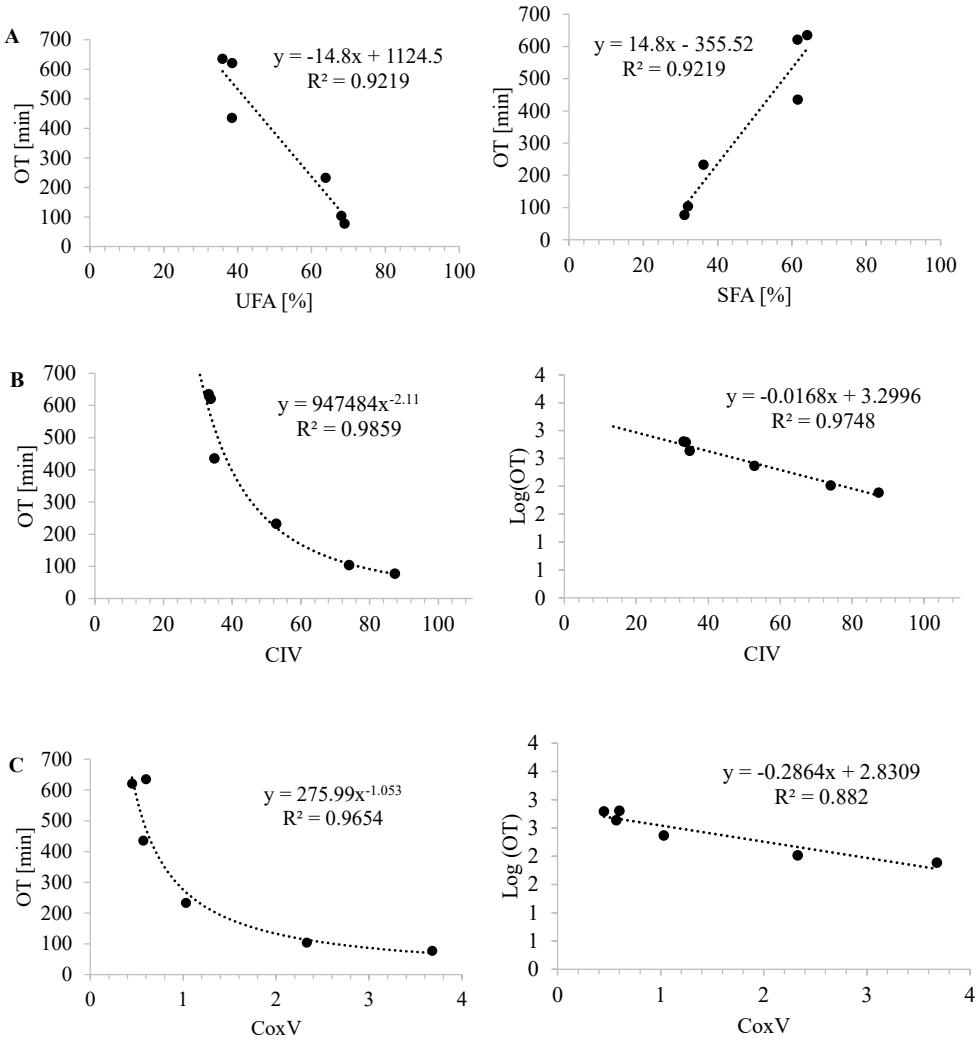

Fig. 2. Relationship between (A) OT and the content of UFA and SFA, (B) OT or logOT and CIV (C) OT or logOT and CoxV

Source: own study.

To the best of our knowledge, there are no examples in the literature concerning the oxidative stability of cosmetic butters as determined using RapidOxy-type apparatus. Therefore, a comparison of the results of the present study with the studies concerning cosmetic butters and the relationship between their oxidative stability and UFA/SFA, CoxV and CIV is not possible.

However, there are studies on the oxidative stability of oils measured using the Rancimat apparatus, in which the results of oxidative stability were correlated with those derived from the FAC of the oils tested. In a study by Symoniuk et al. [2022b], no significant linear correlation was found between the content of C18:2, SFA, UFA and the oxidative stability of commercial cold-pressed oils (8 derived from various oilseeds and 3 mixtures) determined by the Rancimat method at 100°C. In another study [Symoniuk et al. 2022a], a significant correlation (r = –0.87) was found for CoxV and induction time (IT) at 120°C determined using the Rancimat apparatus for oils from unconventional raw materials (n = 7).

Using the data published by Symoniuk et al. [2022b], we calculated CoxV and CIV for the tested oils and correlated them with the induction time (IT) obtained by the Rancimat method at 100°C, 105°C and 110°C. Two oils with very high peroxide values (much higher than the maximum specified in the Codex Alimentarius) were excluded from the calculations. The same was done for data published by Redondo-Cuevas et al. [2018] for seventeen oils and their oxidative stability at 120°C determined by the Rancimat method.

The values of correlation coefficients are shown in Table 2. Significant linear correlation coefficients were found between IT and CoxV or CIV. Slightly higher r values were obtained using the logarithm of IT. The results indicate that CoxV, as a parameter showing the tendency of a fat product to autoxidize, or CIV, an index of fat unsaturation, can be used to build simple models for predicting the oxidation stability of various oils.

Table 2. Correlation coefficients (*r*) between IT and CoxV or CIV of oils calculated based on the results of [Redondo-Cuevas et al. 2018; Symoniuk et al. 2022b]

r	CoxV	CIV	Ref.
IT (105°C)	–0.917	–0.919	Symoniuk et al. 2022b
LogIT (105°C)	–0.960	–0.961	
IT (110°C)	–0.924	–0.927	
LogIT (110°C)	–0.964	–0.966	
IT (120°C)	–0.936	–0.941	
LogIT (120°C)	–0.971	–0.975	
IT (120°C)	–0.799	–0.803	Redondo-Cuevas et al. 2018
LogIT	–0.917	–0.916	

Source: own study.

4. CONCLUSIONS

The results of this study suggest that the oxidative stability of cosmetic butters, expressed as OT measured with RapidOxy, can be predicted from the content of UFA/SFA, CIV and CoxV using a linear or non-linear equation. This approach is cost-effective and environmentally friendly, and appears to be a rapid screening tool for assessing the oxidative stability of cosmetic butters. The preparation of simple linear or non-linear models can help the manufacturers of fat products, not only cosmetic butters, to assess the oxidation resistance. If such simple relationships exist between parameters calculated from the FAC and the oxidative stability of fat products, measured using a RapidOxy-type apparatus or similar, they can become an important part of the quality control of fat products, as demonstrated for the cosmetic butters analyzed in this study and for some oils using data available in the literature.

4.1. Future perspectives

Our practical approach applied to assess the oxidative stability of cosmetic butters suggests that it is possible to create universal and precise predictive models, not only for different types of cosmetic butters, but also for other fat products, considering any differences in product composition and the variable conditions of oxidation stability measurements. Both the FAC (along with the parameters calculated from them) and the oxidation time create a multidimensional dataset; if the historical dataset is large enough, then machine learning (ML) can be incorporated to create appropriate predictive models. The ML-based approach can help automate the process of predicting oxidation times based on known parameters, such as fatty acid composition, thereby facilitating fast and reagent-free assessment of the oxidative stability of fat products and increasing the efficiency of quality control in the cosmetics and food industries.

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