

Comparative Evaluation of the Physicochemical, Thermal, and Antioxidant Properties of Seed Cake Oils Extracted via Soxhlet and Microwave-Assisted Methods

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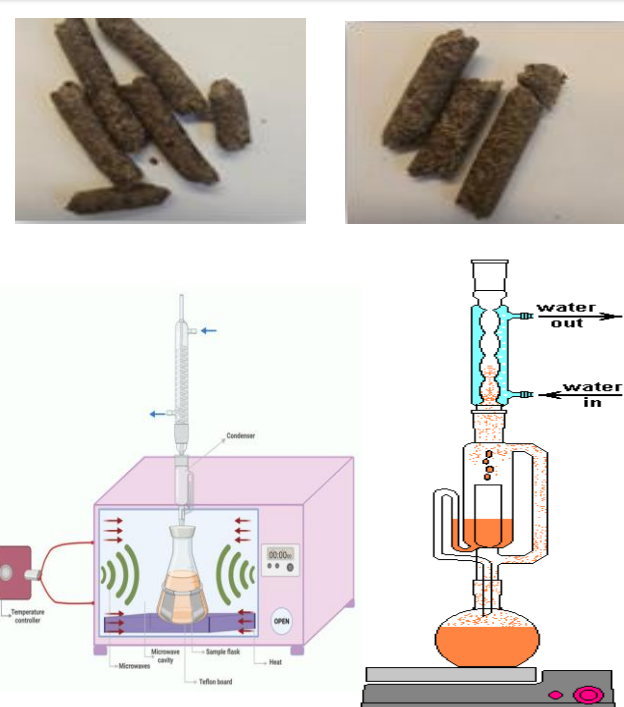
INTRODUCTION & AIM

- Milk thistle (*Silybum marianum*) seed cake, a by-product of oil extraction, represents an underutilized yet valuable source of bioactive lipids and antioxidants with significant nutraceutical potential. The growing emphasis on sustainable food processing has intensified efforts to recover high-value compounds from agro-industrial by-products, including seed cakes.
- Milk thistle seed cake is particularly rich in unsaturated fatty acids, proteins, dietary fiber, vitamin E, and essential minerals such as calcium, potassium, and copper.
- Objective:** This study aims to compare Soxhlet extraction (Sox) and Microwave-Assisted Extraction (MAE) methods for oil recovery, focusing on extraction efficiency, antioxidant retention, and thermal stability.



METHODS

- Samples: Milk thistle seed cakes obtained from two Polish producers (MTC1 and MTC2).
- Solvent used: n-Hexane.
- Extraction methods:
 - Soxhlet Extraction (Sox):** Conventional extraction technique involving continuous solvent reflux for 6 hours.
 - Microwave-Assisted Extraction (MAE):** Innovative method utilizing microwave energy (300 W, 3 min) to enhance solvent penetration and accelerate the release of bioactive compounds.

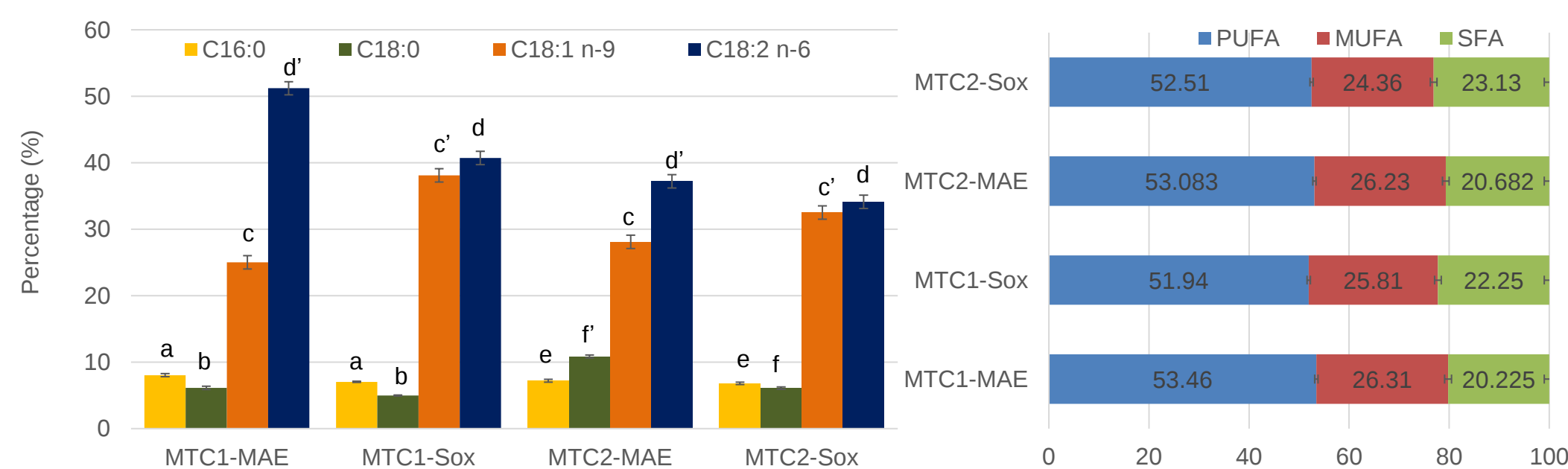


RESULTS & DISCUSSION

Composition of Fatty Acids - characterized by gas chromatography:

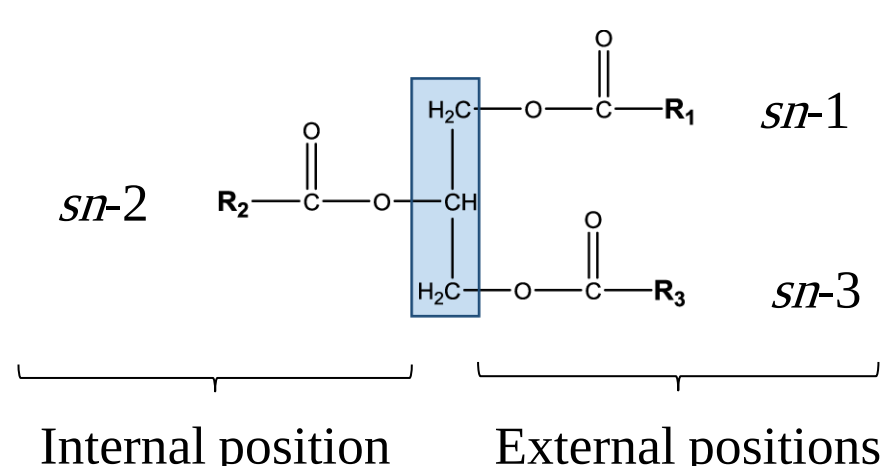
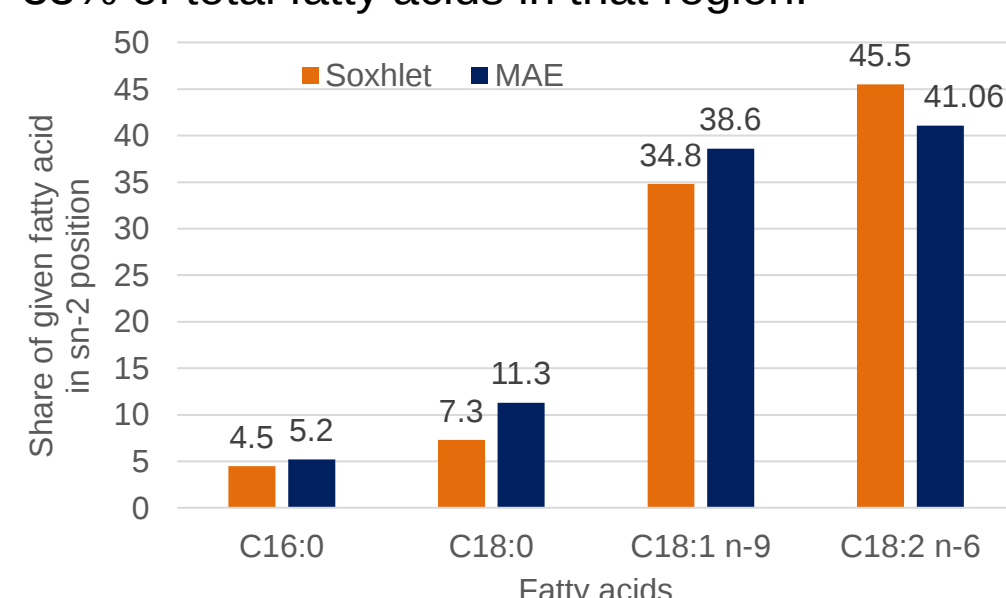
For **MTC1**, the MAE method yielded the highest **linoleic acid (C18:2)** content, while **oleic acid (C18:1)** was more abundant in the Soxhlet extract. Linoleic acid was predominant overall, with the highest level observed in MTC1-MAE.

For **MTC2**, both oleic and linoleic acids were predominant in the MAE extract, whereas the Soxhlet method resulted in higher oleic acid levels. The palmitic acid (C16:0) content showed no significant differences between the two extraction techniques.



Fatty Acid Distribution in Triacylglycerols (TAG) - performed by GC after prior enzymatic hydrolysis:

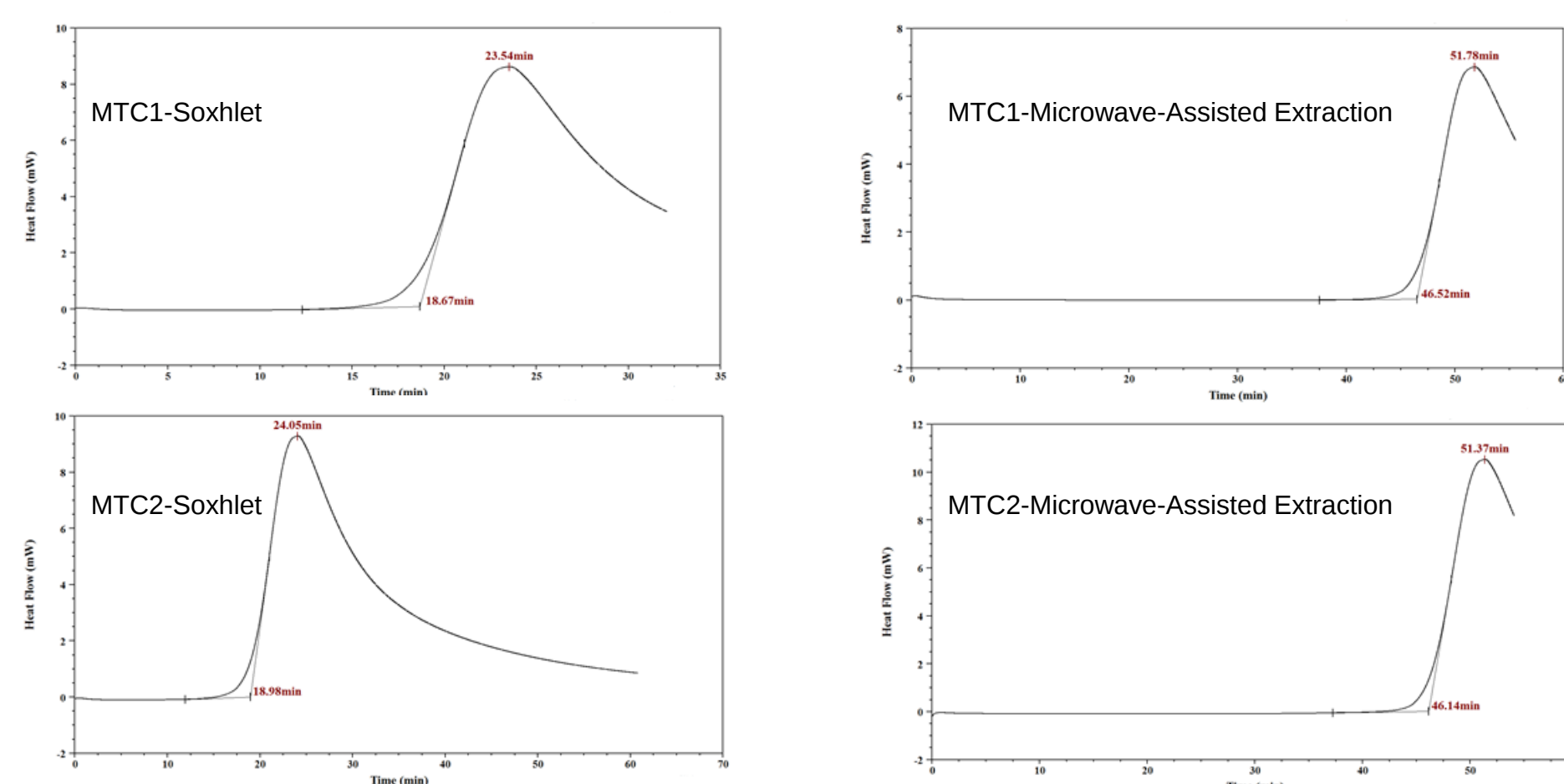
In the oils extracted using both methods, saturated fatty acids - **palmitic (C16:0)** and **stearic acid (C18:0)** - were mainly located at the **outer (sn-1,3) positions** of triacylglycerols (TAG), with only a minor share at the sn-2 position. In contrast, unsaturated fatty acids - **oleic (C18:1)** and **linoleic acid (C18:2)** - were predominantly concentrated at the **internal sn-2 position**, accounting for over 33% of total fatty acids in that region.



RESULTS & DISCUSSION

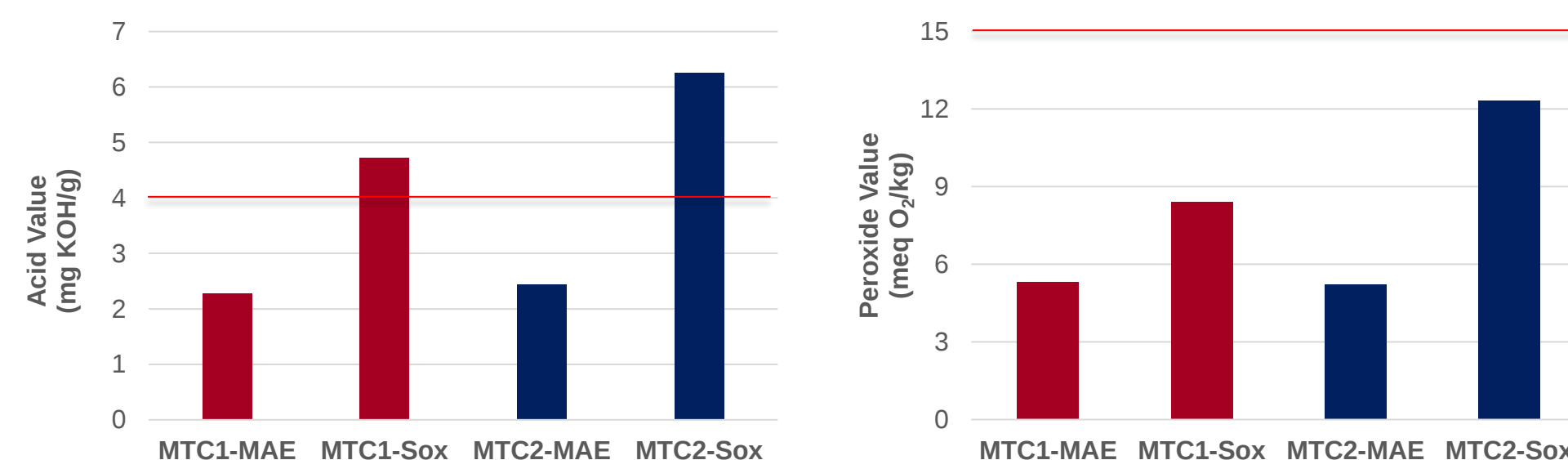
Oxidative Stability - determined at 140°C using PDSC:

The Soxhlet-extracted oil exhibited a significantly shorter induction time ($t_{ind} = 18.67$ min) compared to the MAE - extracted oil (46.52 min). Similarly, the time to maximum oxidation (t_{max}) was longer for MAE oil (51.78 min) than for Soxhlet oil (23.54 min). The greater oxidative stability of the MAE-extracted oil is attributed to its higher content of antioxidant compounds, which effectively delay oxidation despite the oil's higher proportion of polyunsaturated fatty acids (PUFA).



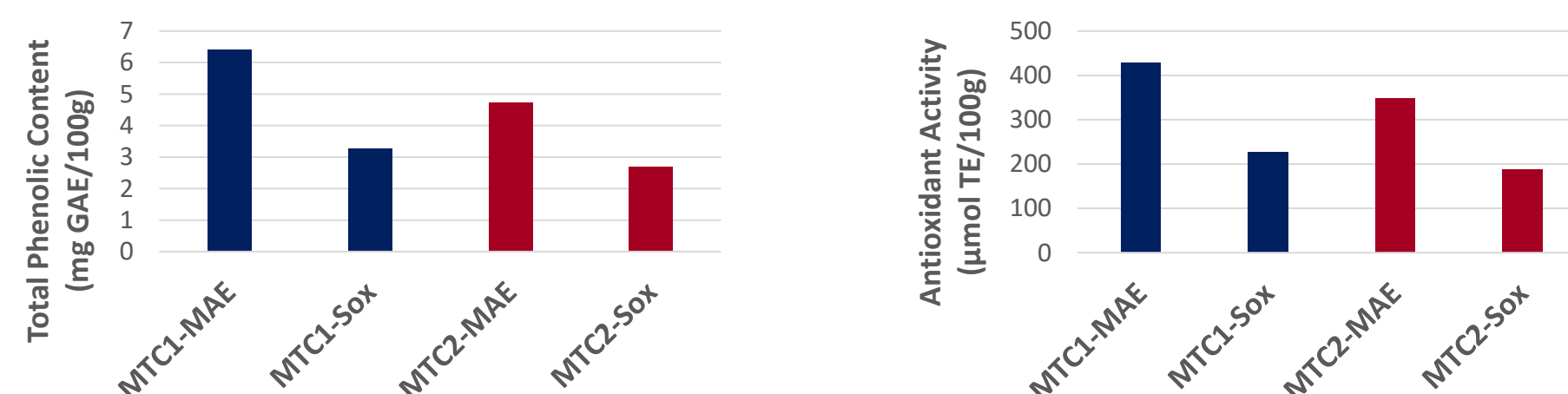
Acid Value (AV) and Peroxide Value (PV) - measured by titrimetric methods:

According to the Codex Alimentarius, the permissible limits for acid value (AV) and peroxide value (PV) are 4 mg KOH/g and 15 meq O₂/kg, respectively. The Soxhlet-extracted oil exceeded the acceptable acid value, indicating that **hydrolytic rancidity** likely occurred during the extraction.



Total Phenolic Content (TPC) and Antioxidant Activity (AA) - assessed by spectrophotometric methods:

For both seed cake samples, **MAE** significantly enhanced **bioactive compound recovery**. **MTC1** showed a **97% increase in TPC** and a **90% increase in AA**, while **MTC2** showed a **76% increase in TPC** and an **86% increase in AA** over the Soxhlet method.



CONCLUSION

- Microwave-assisted extraction (MAE)** is recommended for the valorization of milk thistle seed cake, as it produced oil with superior oxidative stability, enhanced antioxidant activity, and acid and peroxide values within permissible limits.
- The **higher antioxidant content** in MAE-extracted oil effectively **prevented oxidative degradation of polyunsaturated fatty acids (PUFA)**.
- Overall, MAE demonstrates a more sustainable and efficient approach for valorizing milk thistle seed cake, resulting in improved oil quality and enhanced functional properties.

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