RASĀYAN *J. Chem.*
Vol. 19 | Special Issue | 215-222 | 2026
ISSN: 0974-1496 | e-ISSN: 0976-0083 | CODEN: RJCABP
http://www.rasayanjournal.com
http://www.rasayanjournal.co.in

COMPARATIVE EFFECTS OF SUNLIGHT AND OVEN HEATING ON THE FATTY ACID PROFILE AND CHEMICAL CHARACTERISTICS OF BLACK ROUGH SHARK (*Carrhigaelus barbifer*) LIVER OIL

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ABSTRACT

Shark liver oil is a valuable source of bioactive lipids, particularly monounsaturated fatty acids (MUFAs) (MUFAs) and polyunsaturated fatty acids (PUFAs), which are highly susceptible to oxidative degradation during processing. This study aimed to evaluate the effects of sunlight and oven-heating extraction methods on the chemical characteristics and fatty acid composition of black rough shark (*Carrhigaelus barbifer*) liver oil. Liver samples were extracted using sunlight exposure and oven heating at 60 and 70 °C. The extracted oils were analyzed for yield and moisture content, while their chemical profiles were characterized using Fourier Transform Infrared Spectroscopy (FTIR) and Gas Chromatography–Mass Spectrometry (GC–MS). FTIR analysis showed identical characteristic absorption bands at 2922, 1744, 1446, 1379, and 834 cm^{-1} in all samples, indicating the preservation of triglyceride structures and unsaturated fatty acids regardless of extraction method. GC–MS results revealed that the oils were predominantly composed of unsaturated fatty acids. Sunlight-heated oil contained higher levels of methyl nervonate (61.58%) and total MUFAs (61.71%), whereas oven-heated oil exhibited higher docosahexaenoic acid (DHA) content (45.81%) and total PUFAs (47.61%). These findings demonstrate that both extraction methods effectively maintain the chemical integrity and bioactive lipid composition of shark liver oil. Sunlight heating represents a simple, low-cost, and sustainable alternative to conventional oven extraction without compromising the fundamental chemical properties of the oil. Further studies involving oxidative stability parameters are recommended to comprehensively assess oil quality during processing and storage.

Keywords: *Carrhigaelus Barbifer*, Fatty Acids, FTIR, GC–MS, DHA, Extraction Method, Nervonic Acid, Shark Liver Oil.

RASĀYAN *J. Chem.*, Thematic Issue, 2026

INTRODUCTION

Fish oil, including shark liver oil, is an important source of long-chain polyunsaturated fatty acids (LC-PUFAs), particularly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), which have considerable nutritional and functional value. However, the high degree of unsaturation of these fatty acids makes fish oil highly susceptible to oxidation during processing and storage. Oxidative degradation can reduce the nutritional and functional value of the oil and may lead to the formation of undesirable compounds. Therefore, processing conditions, particularly temperature, heating duration, and extraction method, are important factors in determining the yield and quality of the resulting oil.[1-2] Thermal extraction is one of the commonly applied methods for recovering oil from fish tissues and can be performed using direct solar exposure or controlled heating systems. The temperature applied during extraction plays an important role in determining the balance between lipid recovery and oxidative stability. At relatively low temperatures, oxidation reactions tend to be limited, which may help preserve

http://doi.org/10.31788/RJC.2026.19510023
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oil quality. However, insufficient heating may restrict lipid release from the tissue matrix, resulting in relatively low extraction efficiency. Previous studies have reported that low-temperature extraction can produce oil with favorable oxidative and organoleptic characteristics but may provide limited oil recovery.[3] This condition is also relevant to shark liver oil, in which efficient disruption of the tissue matrix is required to facilitate lipid release. Increasing the extraction temperature to a moderate range can improve lipid release by promoting tissue denaturation and facilitating the diffusion of oil from the tissue matrix. Temperatures of approximately 50–65°C have been reported to provide a favorable balance between extraction efficiency and oxidative stability in fish oil processing. At these temperatures, increased lipid recovery may occur without substantial acceleration of lipid oxidation.[4] Thus, moderate-temperature extraction may represent a potential approach for improving the efficiency of shark liver oil production while maintaining acceptable oil quality. In contrast, excessive heating may adversely affect the quality of fish oil. At temperatures of approximately 70°C or higher, lipid autoxidation can be accelerated through free-radical formation and subsequent propagation reactions. These reactions promote the formation of primary oxidation products, particularly hydroperoxides, which can subsequently decompose into secondary oxidation products such as aldehydes and ketones.[5] The accumulation of these compounds may contribute to undesirable changes in odor, color, and other quality characteristics of the oil. In addition, excessive heating may promote tissue degradation and affect the physical characteristics and purity of the extracted oil. Consequently, a higher extraction yield does not necessarily indicate superior oil quality. The quality of fish oil can be evaluated using several physicochemical indicators, among which peroxide value (PV) and acid value (AV) are commonly used. PV provides an indication of primary lipid oxidation, whereas AV reflects the amount of free fatty acids generated mainly through lipid hydrolysis. An increase in these parameters generally indicates deterioration of oil quality and reduced oxidative stability.[6-8] Therefore, evaluation of both extraction yield and quality parameters is necessary to determine whether an extraction condition provides an appropriate balance between efficient lipid recovery and preservation of oil quality. International standards also emphasize the importance of controlling the oxidative and hydrolytic quality of fish oils. The Codex Alimentarius Standard for Fish Oils (CXS 329-2017) specifies maximum values for peroxide value and acid value, while the International Fish Oil Standards (IFOS) provide stricter criteria for high-quality fish oil products. These standards demonstrate that the quality of fish oil cannot be assessed solely on the basis of extraction yield but must also consider its chemical stability and suitability for further utilization.

Although previous studies have established the relationship between extraction temperature, oil yield, and oxidative stability, an important research gap remains regarding the identification of suitable heating conditions specifically for shark liver oil. Much of the existing literature has examined fish oil extraction more broadly or has considered extraction efficiency and oxidative quality as separate aspects. Consequently, the temperature condition that provides an appropriate balance between oil recovery and quality characteristics of shark liver oil remains insufficiently characterized. In particular, comparative information on solar heating and controlled heating at moderate and elevated temperatures, together with simultaneous evaluation of organoleptic characteristics, extraction yield, PV, and AV, is still limited. Addressing this gap is important because optimization based solely on yield may result in excessive thermal exposure and deterioration of the functional quality of the extracted oil.[7,9] Therefore, this study aimed to evaluate the effect of different heating conditions on the extraction yield and quality of shark liver oil. Solar heating and controlled heating at different temperatures were compared based on organoleptic characteristics, extraction yield, peroxide value, and acid value. The study specifically sought to identify the heating condition that provides the most favourable balance between extraction efficiency and oxidative stability.[10] The findings are expected to contribute to the optimization of shark liver oil extraction by providing practical information on temperature control as an important factor for maintaining oil quality while achieving efficient lipid recovery.

EXPERIMENTAL

Sample preparation

The prepared liver samples were placed in heat-resistant containers and subjected to dry rendering using an oven at different temperatures (60 and 70 °C) for 1–3 h to facilitate protein denaturation and oil release.

The extracted oil was periodically collected during heating. The crude oil was filtered using filter paper to remove solid residues and subsequently centrifuged at 5000 rpm for 2 h to obtain purified oil. The final oil samples were stored in dark glass bottles at 4°C until further analysis. For sunlight heating extraction, liver pieces were spread evenly on aluminium trays and covered with sterile gauze to prevent contamination. The samples were exposed to direct sunlight for several hours per day over 1–3 days, depending on solar intensity. The oil released during the process was collected immediately using a pipette or funnel to minimise oxidation. The crude oil was filtered using filter paper, allowed to settle, and then centrifuged at approximately 4000 rpm for 2 h. The purified oil was stored in dark glass bottles at refrigerated conditions (4°C) prior to analysis.

FTIR Analysis

Prior to analysis, oil samples were clarified by filtration or mild centrifugation to remove residual particles. A drop of oil was directly placed onto the ATR crystal (diamond/ZnSe) without dilution. Spectral measurements were performed in the range of 4000–650 cm^{-1} with a resolution of 4 cm^{-1}. The spectra were processed using baseline correction and normalisation to improve spectral accuracy and comparability.[11-12]

Unsaturated fatty acids were identified based on the presence of =C–H stretching around 3010 cm^{-1}, while bands at 2925–2850 cm^{-1} corresponded to aliphatic C–H stretching and the 1740 cm^{-1} band indicated ester carbonyl groups. These characteristic peaks are widely used to identify lipid functional groups and fatty acid composition in oils. The relative intensity of the unsaturation band was used as an indicator of MUFA and PUFA content, as commonly applied in FTIR-based lipid analysis.[9] After each measurement, the ATR crystal was cleaned using isopropanol and lint-free tissue to prevent cross-contamination.

GC-MS Analysis

Shark liver oil samples were filtered and converted into fatty acid methyl esters (FAMEs) through derivatisation using methanolic NaOH followed by BF_3-methanol. The resulting FAMEs were analysed using GC–MS equipped with an HP-5MS capillary column, with helium as the carrier gas and electron ionisation at 70 eV. Individual compounds were identified by comparison with the NIST mass spectral library, and fatty acid composition was expressed as the relative percentage of total chromatographic peak area for classification of lipid fractions and profiles.

RESULTS AND DISCUSSION

FTIR Characterisation of Shark Liver Oil

The FTIR spectra of the positive control, solar-heated extract, and oven-heated extracts showed similar major absorption bands at 2922, 1744, 1446, 1379, and 834 cm^{-1} (Figs.-3 to 5). The similarity of these spectral profiles indicates that the different extraction conditions did not cause substantial changes in the major functional groups of shark liver oil. This finding suggests that the primary lipid structure, which is predominantly associated with triglycerides and long-chain fatty acids, was retained following both solar and oven heating. Similar FTIR profiles have been reported for fish oils, in which characteristic absorption bands associated with aliphatic hydrocarbons, ester groups, and unsaturated fatty acids are commonly observed.[13-15] The absorption band at 2922 cm^{-1} is attributed to the asymmetric stretching vibration of aliphatic C–H bonds, particularly those associated with $–CH_2$ groups in long hydrocarbon chains. The presence of this band in all samples indicates the preservation of the aliphatic lipid structure after extraction. The strong absorption at 1744 cm^{-1} corresponds to the stretching vibration of the ester carbonyl (C=O) group, which is characteristic of triglycerides. The persistence of this band across all treatments indicates that the fundamental ester structure of the major lipid components remained detectable following thermal extraction. [14] The absorption bands at 1446 and 1379 cm^{-1} are associated with CH_2 bending and symmetric CH_3 bending vibrations, respectively. The presence of these bands with similar spectral characteristics among the samples further supports the preservation of the hydrocarbon chains during extraction. These observations are consistent with previous FTIR investigations of fish oils, which have reported comparable absorption regions for aliphatic C–H and methyl/methylene groups.[15] The band observed at approximately 834 cm^{-1} can be tentatively associated with out-of-plane =C–H bending, which is related to unsaturated hydrocarbon structures. This is consistent with the presence of

unsaturated fatty acids commonly found in marine-derived oils.[16] Importantly, the similarity of the FTIR spectra among the different treatments indicates that the effects of solar and oven heating were not sufficient to produce major qualitative changes in the functional groups detected by FTIR. Thus, the differences observed between extraction conditions are more likely to be reflected in quantitative characteristics, such as extraction yield and the relative composition of individual fatty acids, as well as in oxidative quality parameters. This interpretation is important because FTIR provides information about functional groups but does not, by itself, provide a comprehensive measurement of lipid oxidation. Oxidative deterioration should therefore be evaluated using complementary parameters such as peroxide value, acid value, p-anisidine value, or other validated oxidation indices.[17] From a processing perspective, the preservation of the major FTIR absorption bands under both solar and controlled oven heating suggests that thermal extraction under the conditions investigated did not substantially disrupt the fundamental chemical identity of the extracted oil. This finding supports the potential use of solar heating as a low-cost extraction approach, particularly where access to conventional heating equipment is limited. However, the suitability of solar heating cannot be determined from FTIR spectra alone and should be considered together with extraction yield, physicochemical quality parameters, and fatty acid composition. Therefore, the FTIR results provide complementary evidence that the extraction treatments primarily preserved the major lipid functional groups, while other analytical parameters are required to distinguish differences in oil quality and oxidative stability.[18]

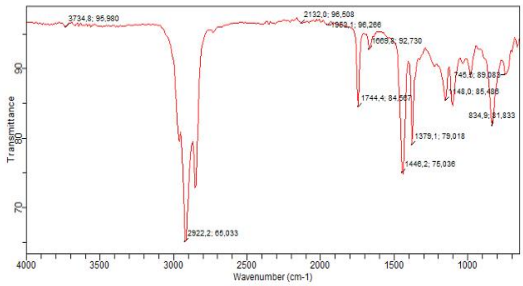


Fig.-1: FTIR Spectrum of Shark Liver Oil Extracted using the Sunlight Heating Method for 3 hours

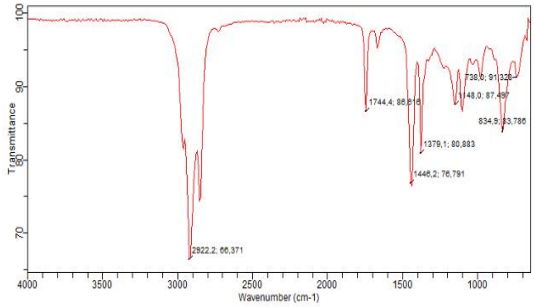


Fig.-2: FTIR Spectrum of Shark Liver Oil Extracted using the Oven-Heating Method at 60°C for 3 Hours

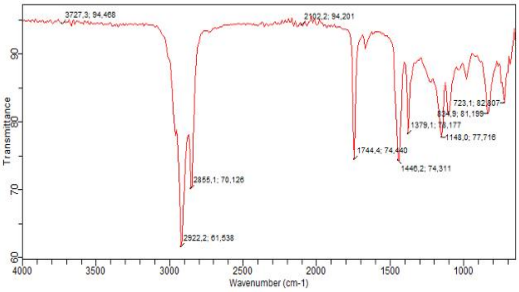


Fig.-3: FTIR Spectrum of Shark Liver Oil Extracted using the Oven-Heating Method at 70°C for 3 Hours

GC–MS Characterisation of Shark Liver Oil

Solar Heating

GC–MS analysis of shark liver oil extracted using solar heating showed that the oil was predominantly composed of unsaturated lipid components. The major compounds detected were methyl nervonate (61.58%) and docosahexaenoic acid (DHA) (37.14%) (Fig.-4). The predominance of these compounds indicates that the extracted oil contains a substantial proportion of unsaturated fatty acid-derived components. Shark liver oil and other marine oils have previously been reported to contain considerable amounts of unsaturated fatty acids and other bioactive lipid components, supporting the potential nutritional and functional significance of the present findings.[19] The relatively high proportion of methyl nervonate is notable because nervonic acid (C24:1) is an important very-long-chain monounsaturated fatty acid associated with sphingolipids and myelin structure in neural tissues. Nevertheless, its detection by GC–MS should be interpreted primarily as evidence of the lipid composition of the extracted oil, rather than direct evidence of health benefits, which require further nutritional and biological studies.[20] The detected DHA content of 37.14% is also significant, as DHA is a major long-chain omega-3 PUFA commonly found in marine oils. Its presence indicates that solar extraction successfully recovered DHA-containing lipids from shark liver. However, GC–MS detection alone cannot confirm protection against oxidation during processing or storage. Because DHA contains multiple double bonds, it is highly susceptible to oxidative degradation; therefore, its preservation should be evaluated alongside peroxide value, acid value, and other oxidative stability parameters.[21] The reported MUFA and PUFA proportions of 61.71% and 37.81%, respectively, indicate an oil dominated by unsaturated lipids. This agrees with FTIR results showing characteristic lipid functional groups. Thus, FTIR and GC–MS provide complementary evidence: FTIR confirms major lipid structures, whereas GC–MS identifies specific lipid components. Nevertheless, the high unsaturated fatty-acid content may increase oxidation susceptibility, so oil quality should be assessed together with physicochemical oxidation indicators. [22]

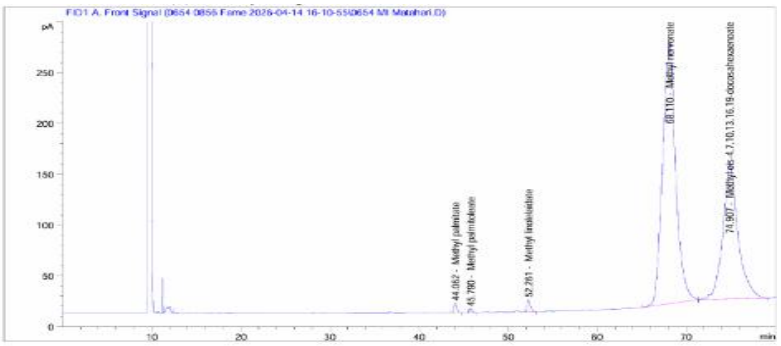


Fig.-4: GC–MS Spectrum of Fish Oil Obtained Through Sunlight Heating Extraction

Oven Heating

The GC–MS/FID analysis of shark liver oil extracted by oven heating showed a similar predominance of unsaturated lipid components. Methyl nervonate was the major detected MUFA-related component at 50.26%, while DHA was the dominant PUFA component at 45.81% (Fig.-5). The presence of these components demonstrates that controlled oven heating was also effective in recovering unsaturated lipid components from shark liver tissue.[23] Compared with the solar-heated extract, the oven-heated extract showed a lower relative proportion of methyl nervonate (50.26%) but a higher relative proportion of DHA (45.81%). The MUFA and PUFA proportions were reported as 51.12% and 47.61%, respectively. These differences indicate that the relative composition of the compounds detected by GC–MS varied between extraction conditions. However, differences in relative peak percentages should not automatically be interpreted as evidence of thermal degradation or formation of individual fatty acids. Changes in relative composition may also reflect differences in extraction efficiency, lipid recovery, sample preparation, or the relative abundance of individual components in the analyzed fractions. Therefore, conclusions regarding thermal degradation should be supported by additional oxidation markers and quantitative fatty acid analyses.[24] The relatively high DHA proportion in the oven-heated extract is particularly relevant because DHA is an important long-chain omega-3 PUFA in marine oils. Its detection after controlled

heating indicates that the extraction treatment did not eliminate this important lipid component. At the same time, the high proportion of PUFA emphasizes the need for careful control of processing and storage conditions because highly unsaturated fatty acids are susceptible to oxidation.[25] Thus, the GC–MS findings should be interpreted together with the peroxide value and acid value results to provide a more complete assessment of oil quality. The similarities between the solar- and oven-heated samples are also consistent with the FTIR results. Both extraction methods retained the characteristic functional groups associated with triglycerides and unsaturated lipid structures. This suggests that neither extraction approach caused a major qualitative alteration in the principal lipid components detectable by the analytical methods used. Nevertheless, the differences in relative fatty acid composition demonstrate that the extraction method may influence the quantitative profile of the recovered oil.

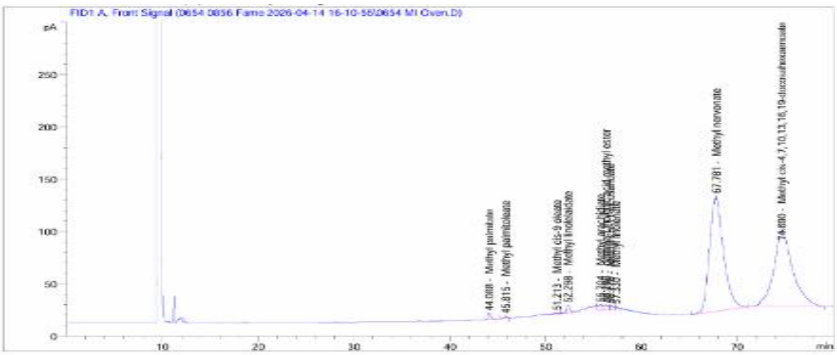


Fig.-5: GC–MS Spectrum of Shark Liver Oil Extracted using the Oven-Heating Method at 60°C for 3 hours

Significance of the FTIR and GC–MS Findings

The FTIR and GC–MS results provide complementary information on the effects of extraction conditions on shark liver oil. FTIR analysis showed that the major lipid functional groups remained detectable across treatments, while GC–MS identified substantial proportions of unsaturated fatty acid-derived components, particularly methyl nervonate and DHA. These findings suggest that both solar and controlled oven heating can recover shark liver oil containing desirable lipid components.[26] However, chemical identity, relative composition, and oxidative stability must be distinguished when evaluating extraction methods. Similar FTIR spectra indicate the presence of comparable functional groups, whereas differences in GC–MS profiles reflect variations in the relative proportions of detected components. Neither technique alone establishes oxidative stability. Therefore, these results should be interpreted alongside peroxide value, acid value, extraction yield, and organoleptic characteristics. Overall, an appropriate extraction method should balance efficient oil recovery with acceptable physicochemical quality and oxidative stability rather than relying solely on extraction yield or the relative abundance of desirable fatty acid-derived components.

CONCLUSION

This study demonstrated that both solar and controlled oven heating successfully extracted shark liver oil while preserving the major lipid functional groups identified by FTIR. The extracts showed similar characteristic bands associated with aliphatic C–H, ester carbonyl, and unsaturated lipid structures, indicating no substantial qualitative changes. GC–MS analysis revealed that the oils were predominantly composed of unsaturated lipid components, particularly methyl nervonate and docosahexaenoic acid (DHA). The solar-heated extract contained 61.58% methyl nervonate and 37.14% DHA, while the oven-heated extract contained 50.26% and 45.81%, respectively. These differences may reflect extraction conditions, efficiency, or sample preparation rather than thermal degradation alone. Overall, solar heating shows potential as a simple and low-cost extraction approach. However, further evaluation using extraction yield, peroxide value, acid value, p-anisidine value, and other oxidative stability parameters is recommended. Future studies should optimise temperature, extraction time, yield, fatty acid composition, and oxidative stability to ensure efficient oil recovery and maintain its chemical and physicochemical quality. The potential use of solar heating also supports SDG-12 (Responsible Consumption and

Production) by promoting resource-efficient processing and the sustainable utilisation of available biological resources.

ACKNOWLEDGEMENTS

The authors would like to express their sincere gratitude to the Head of the Biochemistry Laboratory, Universitas Sumatera Utara, and the Research Laboratory of Institut Kesehatan Helvetia for granting permission and providing facilities to conduct this study.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHOR CONTRIBUTIONS

The present work is related to *SDG-9: Industry, Innovation, and Infrastructure*. All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

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Cite this article as: D. A. Tanjung *et al.*, *Rasayan J. Chem.*, Vol.19, Special Issue, 215-222(2026), https://doi.org/10.31788/RJC.2026.19510023

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