Article | Received 19 November 2024; Accepted 17 February 2025; Published 21 February 2025
https://doi.org/10.55092/exrna20250002

MicroRNA profiling as novel biomarkers for detecting gutter oil using machine learning

Jiaxin Li[1,2], Lin Cong[1,2], Yuyu Liu[1,2], Limin Li[3,*] and Yujing Zhang[1,2,*]

[1] Nanjing Drum Tower Hospital Center of Molecular Diagnostic and Therapy, State Key Laboratory of Pharmaceutical Biotechnology, Jiangsu Engineering Research Center for MicroRNA Biology and Biotechnology, School of Life Sciences, NJU Advanced Institute of Life Sciences (NAILS), Institute of Artificial Intelligence Biomedicine, Nanjing University, Nanjing 210023, China

[2] Chinese Academy of Medical Sciences, Research Unit of Extracellular RNA, Nanjing 210023, China

[3] State Key Laboratory of Natural Medicines, Jiangsu Key Laboratory of Druggability of Biopharmaceuticals, School of Life Science and Technology, China Pharmaceutical University, Nanjing 211198, China

* Correspondence authors; E-mails: liminli@cpu.edu.cn (L.L.); yjzhang@nju.edu.cn (Y.Z.).

Table S1. Reviews of technologies for detecting gutter oil.

Category	Method	References	Comments
Physical Chemistry	Planar toroidal dipole metamaterial sensor	[15]	These methods typically require specialized detection equipment and are still in developmental stages, resulting in relatively high testing costs. Additionally, conductivity detection is influenced by the refining process of gutter oil, leading to significant fluctuations under deep refining conditions.
	Highly sensitive optical fluid detection with hybrid-waveguide couplers	[16]	
	Raman spectroscopy for identifying polycyclic aromatic hydrocarbons (PAHs)	[17]	
	Conductivity measurement	[56]	
Analytical Chemistry	Mass spectrometry and chromatography techniques (e.g., MALDI-MS, UPLC-MS/MS) to detect a variety of substances in gutter oil, including aldehydes, ketones, capsaicin, cholesterol, surfactants, and species-specific fatty acids	[18–23]	a) The refining process can effectively remove a significant portion of aldehydes and ketones, making direct detection challenging;
			b) Capsaicin levels are highly affected by cooking practices, as certain foods may be cooked without spices, thus making capsaicin detection less reliable;
			c) Cholesterol detection suffers from low sensitivity, and detecting surfactant residues can be influenced by the refining process;
			d) Mass spectrometry and chromatography require large-scale equipment, leading to high costs for testing.
Biochemical	Antigen-antibody interactions (detect capsaicin)	[24–26]	Capsaicin detection is still influenced by cooking practices, and antigen-antibody interactions are not reliable enough.
	Metabolomics	[27,28]	Metabolomics is a highly complex and expensive technique.

Table S2. Sequences of primers and probes used for reverse transcription and TaqMan qPCR.

miRNA (Accession number)	Label	Sequence
MIR162a (MI0000194)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACctggat
	F	TCGCTtcgataaacctctgc
MIR168a (MIMAT0001045)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACgtcccg
	F	TCGCTtcgcttggtgcagat
MIR166 (MIMAT0000635)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACggggaa
	F	TCGCTtcggaccaggcttca
MIR156a (MIMAT0000618)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACgtgctc
	F	TCGCTtgacagaagagagt
let-7a (MIMAT0000062)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACaactat
	F	TCGCTtgaggtagtaggttgt
miR-223 (MIMAT0000280)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACtggggt
	F	TCGCTtgtcagtttgtcaaat
miR-16 (MIMAT0000069)	RT	GTCGTATCCAGTGCAGGGTCCGAGGTATTCGCACTGGATACGACcgccaa
	F	TCGCTtagcagcacgtaaata
Universal Primer	R	GTGCAGGGTCCGAGGT

Reference

[15] Jianwei X, Shoujian O, Shouxin D, Liner Z, Xiaohua D, *et al*. Terahertz planar toroidal dipole metamaterial sensor for detecting gutter oil. *Chin. J. Quantum Electron.* 2023, 40(3):333.

[16] Lin C, Liao C, Zhang Y, Xu L, Wang Y, *et al.* Optofluidic gutter oil discrimination based on a hybrid-waveguide coupler in fibre. *Lab Chip* 2018, 18(4):595–600.

[17] Su M, Jiang Q, Guo J, Zhu Y, Cheng S, *et al.* Quality alert from direct discrimination of polycyclic aromatic hydrocarbons in edible oil by liquid-interfacial surface-enhanced Raman spectroscopy. *Lwt* 2021, 143:111143.

[18] HE W, Fang R, LI Y, Zheng S, Lin J. Determination of cholesterol content in vegetable oil for rapid screening waste oil. *Chin. J. Anal. Chem.* 2015:394–398.

[19] Ng TT, So PK, Zheng B, Yao ZP. Rapid screening of mixed edible oils and gutter oils by matrix-assisted laser desorption/ionization mass spectrometry. *Anal. Chim. Acta* 2015, 884:70–76.

[20] Teng C, Wu S, Sun Y, Gong G. Determination of parent and oxygenated polycyclic aromatic hydrocarbons (PAHs) in waste cooking oil and oil deodorizer distillate by GC–QQQ–MS. *J. AOAC Int.* 2019, 102(6):1884–1891.

[21] Lu Q, Guo H, Li D, Zhao Q. Determination of capsaicinoids by magnetic solid phase extraction coupled with UPLC-MS/MS for screening of gutter oil. *J. Chromatogr. B* 2020, 1158:122344.

[22] Zhang Y, Han Y, Hu W, Pan Q, Liu Z, *et al.* Diacylglycerols ions as novel marker indicators for the classification of edible oils using ultrahigh resolution mass spectrometry. *Food Res. Int.* 2021, 145:110422.

[23] Zhou Q, Yao C, Wang H, Zhu KZ, Peng J, *et al.* Application of chromatographic technology in the identification of swill-cooked dirty oil. *China Oils Fats* 2021, 46(5):93–96.

[24] Wu Q, Yao L, Qin P, Xu J, Sun X, *et al.* Time-resolved fluorescent lateral flow strip for easy and rapid quality control of edible oil. *Food Chem.* 2021, 357:129739.
[25] Zhang J, Zhang M, Yang Q, Wei L, Yuan B, *et al.* A simple and rapid homogeneous fluorescence polarization immunoassay for rapid identification of gutter cooking oil by detecting capsaicinoids. *Anal. Bioanal. Chem.* 2022, 414(20):6127–6137.
[26] Nie K, Zhang J, Xu H, Ren K, Yu C, *et al.* Reverse design of haptens based on antigen spatial conformation to prepare anti-capsaicinoids&gingerols antibodies for monitoring of gutter cooking oil. *Food Chem. X* 2024, 22:101273.
[27] Cavanna D, Righetti L, Elliott C, Suman M. The scientific challenges in moving from targeted to non-targeted mass spectrometric methods for food fraud analysis: A proposed validation workflow to bring about a harmonized approach. *Trends Food Sci. Technol.* 2018, 80:223–241.
[28] Liu SJ, Wu YN, Chan L. Application of metabonomics approach in food safety research-a review. *Food Rev. Int.* 2020, 36(6):547–558.
[56] Jie Y, Haishen H, Yangjun L. A high precious gutter oil detector based on complementary metal oxide semiconductor. *Food Mach.* 2016, 32(9):44–48,153.