

**STUDIES ON SESAME SEEDS AND THEIR CO-
PRODUCT (WHITE/BLACK) FOR ITS FUNCTIONAL
AND NUTRITIONAL APPLICATIONS**

KARNIKA PRAKASH



**CENTRE FOR RURAL DEVELOPMENT AND TECHNOLOGY
INDIAN INSTITUTE OF TECHNOLOGY DELHI**

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PRODUCT (WHITE/BLACK) FOR ITS FUNCTIONAL
AND NUTRITIONAL APPLICATIONS**

by

KARNIKA PRAKASH

Centre for Rural Development and Technology

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This is to certify that the thesis entitled '**Studies on sesame seeds and their co-product (white/black) for its functional and nutritional applications**' being submitted by **Miss. Karnika Prakash** to the Indian Institute of Technology Delhi for the award of the degree of **Doctor of Philosophy** is a record of bonafide research work carried out by him under my guidance and supervision I conformity with the rules and regulations of Indian Institute of Technology Delhi. The research report and results presented in this thesis have not been submitted, in part or in full, to any other university or institute for the award of any degree or diploma.

Dr. S.N. Naik

Centre for Rural Development and Technology
Indian Institute of Technology Delhi
Hauz Khas, New Delhi-110016, India

Dr. Rajender Prasad

Centre for Rural Development and Technology
Indian Institute of Technology Delhi
Hauz Khas, New Delhi-110016, India

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(Karnika Prakash)

Date:

The biggest challenge faced by edible oil industry now a days is the stability of the oils by using non – synthetic antioxidants; which are harmless for consumption over a period of time. The use of natural antioxidants to improve the oxidative stability of food lipids has received special attention because of the worldwide trend to avoid the use of synthetic food additives. The efficacy of natural antioxidants and the oxidative stability of the edible oils are alarming questions in the consumers mind. Along with this we need also focus and draw attention on the food wastage by incomplete utilization of end co-products leading to shortage of food and even environmental pollution. Therefore, looking at the above mentioned concern, the focus of the present investigation is to evaluate the biological activities of sesame lignans on interface with blending and enrichment of linseed and cottonseed oil; value addition of the defatted sesame cake.

Sesame oil soluble lignans i.e. sesamin, sesamol and sesamol were separated, isolated and characterized by TLC, UV- spectra and RP- HPLC. Sesamin (30.09 and 45.86 mg/100g), sesamol (7.12 and 5.70 mg/100g) and sesamol (24.40 and 20.63 mg/100g) respectively were recovered from locally available black/white sesame seeds. Similarly water soluble lignans i.e. sesaminol diglucoside, sesaminol triglucoside and sesamol diglucoside were also isolated, purified and characterized through column chromatography, TLC, UV- spectra and RP- HPLC. Recovery of sesaminol diglucoside (113.34 and 546.02 mg/100g), sesaminol triglucoside (668.74 and 318 mg/100g) and sesamol diglucoside (24.55 and 102.25 mg/100g) respectively from black and white sesame seeds. The antioxidant potential of sesame oil, sesame oil soluble lignans, sesame defatted cake extract and sesame glycosylated lignans were also evaluated through various antioxidant assays i.e. DPPH, ABTS , β carotene- linoleic acid bleaching , TPC and reducing power assay. It was found that black/white sesame extracts at a concentration nearly to 200 ppm

has stabilization proficiency equivalent to the synthetic and commercial antioxidants BHT and BHA, but has lower competence than that of the synthetic antioxidant TBHQ.

Blending of linseed oil (LO) and cottonseed oil (CSO) with white sesame oil (WSO)/ black sesame oil (BSO) in different ratios (20:80, 40:60, 50:50, 60:40 and 80:20). The blends were analyzed for their oxidative stability with Rancimat at 110°C, 120°C and 130°C respectively. It was found that pure BSO has an induction period of 13.62 h at 110°C and WSO has 8.30 h at 110° C on contrary LO and CSO has 3.20 h and 5.23 h respectively. Out of all the blends we tried taking into consideration the nutritional and economical benefits thermal stability of 50:50 ratio were performed. The FFA content results indicated that blending of LO and CSO with BSO lowers the FFA contents followed by WSO. Peroxide value results of all oil samples (pure oil and blends) increased with increase in storage period followed the order BSO < WSO < BSO: CSO (50:50) < WSO: CSO (50:50) < CSO < BSO: LO (50:50) < WSO: LO (50:50) < LO. The pure LO and CSO contains good amount of linoleic acid which is responsible for increase the CDV, but this increment was controlled when blended with BSO and WSO. The FTIR data showed no change in molecular bonds and functional groups after 5 days of frying. It was observed that the absorption peaks are the same for all the oil samples after 5 days of frying. Potato chips used for frying were sensory evaluated by ten panel members and even analyzed for moisture loss and fat uptake and no significant ($p < 0.05$) difference was noticed in moisture content and fat uptake of dried potato chips subjected to periodic frying cycles.

LO and CSO were enriched with defatted sesame cake (black/white) extracted with ethanol and methanol. The crude extracts were added in LO and CSO at 50,100,150 and 200 ppm along with synthetic (BHA, BHT and TBHQ). The oil samples were stored at 60°C for 15 days. Rancimat analysis of the extracts found profoundly that the bioactive extract potential was better towards

ethanol and methanol extracts. In relation to the extracts, black sesame extracts (BSME and BSEE) followed by white sesame extracts (WSME and WSEE) shows highest IP value in LO and CSO. The extracts worked better when extracted with ethanol even at lower concentrations. Peroxide value results after 15 days of storage showed that SCE (black/white) inhibited LO and CSO oxidation. Furthermore, the antioxidant effect of SCE (black/white) at 200 ppm was better than BHA and BHT at 200 ppm. p- Anisidine value, Iodine value, Totox value and fatty acid content was observed higher in oils without antioxidants compared to those with antioxidants.

Value addition of defatted sesame cake (black/white) was done by developing biscuits. By analyzing the results obtained, it is concluded that defatted white sesame flour and defatted black sesame flour could be incorporated up to a level of 50% in the formulation of biscuits without affecting their overall quality. Defatted white sesame flour fortification was more acceptable to the consumers in terms of sensorial parameters, the physical and textural attributes of both varieties of biscuits were similar to the control biscuits. But the increased protein, fat, crude fiber, mineral content is an added advantage over the existing wheat flour biscuits. Incorporation of defatted sesame flour increases resistance to microbial contamination thereby improving the shelf life of biscuits. Thus, defatted sesame flour, a co-product from oil processing industry could be utilized in the preparation of biscuits or other economical and value added products with enhanced functional and nutraceutical potential.

सार

खाद्य तेल उद्योग का सबसे बड़ा चुनौती अब एक दिन गैर सिंथेटिक एंटीऑक्सीडेंट का उपयोग करके तेलों की स्थिरता है; जो समय की अवधि में उपभोग के लिए हानिरहित हैं कृत्रिम खाद्य additives के उपयोग से बचने के लिए दुनिया भर में प्रवृत्ति के कारण भोजन लिपिड्स की ऑक्सीडेटिव स्थिरता में सुधार करने के लिए प्राकृतिक एंटीऑक्सीडेंट का उपयोग विशेष ध्यान प्राप्त हुआ है। प्राकृतिक एंटीऑक्सीडेंट्स और खाद्य तेलों की ऑक्सीडेटिव स्थिरता की प्रभावकारिता उपभोक्ताओं के दिमाग में खतरनाक सवाल हैं इसके साथ-साथ हमें अंतः सह-उत्पादों के अपूर्ण उपयोग के कारण खाद्य अपव्यय पर ध्यान केंद्रित करने और ध्यान देने की भी आवश्यकता है जिससे खाद्य की कमी और पर्यावरण प्रदूषण भी हो। इसलिए, उपर्युक्त चिंता को देखते हुए, वर्तमान जांच का फोकस तिल के लिग्नांस की जैविक गतिविधियों का विश्लेषण करना है जो कि अलसी और कपास के तेल के मिश्रण और संवर्धन के साथ अंतरफलक पर है; डिब्बेटेड तिल केक के मूल्य में बढ़ोतरी।

तिल का तेल घुलनशील लिग्नांस, जैसे कि सेसमैन, सेसमोलिन और सेसमोल अलग-अलग, टीएलसी, यूवी-स्पेक्ट्रा और आरपी-एचपीएलसी द्वारा अलग-थलग किए गए थे। सेस्मिन (30.0 9 और 45.86 मिलीग्राम / 100 ग्राम), सेसमोल (7.12 और 5.70 मिलीग्राम / 100 ग्रा) और सेसमोलिन (24.40 और 20.63 मिलीग्राम / 100 ग्राम) स्थानीय रूप से उपलब्ध काले / सफेद तिल के बीज से वसूल किए गए थे। इसी तरह पानी में घुलनशील लिग्नांस जैसे एसएसमाइनोल डिग्लुकोसाइड, सेसमिनोल ट्राइल्कोसाइड और सेसमोलिनोल डिग्लूकोसाइड को स्तंभ क्रोमैटोग्राफी, टीएलसी, यूवी-स्पेक्ट्रा और आरपी-एचपीएलसी के माध्यम से पृथक, शुद्ध और लक्षणीत किया गया था। काले और सफेद तिल के बीज से क्रमशः सेमिनोल डिग्लुकोसाइड (113.34 और 546.02 मिलीग्राम / 100 ग्रा), सेसमिनोल ट्राइल्लुसाइड (668.74 और 318 मिलीग्राम / 100 ग्रा) और सेसमोलिनोल डिग्लूकोसाइड (24.55 और 102.25 मिलीग्राम / 100 ग्रा) की वसूली। तिल के तेल, तिल का तेल घुलनशील लिग्नांस की एंटीऑक्सीडेंट क्षमता, तिल डिब्बेटेड केक निकालने और तिल ग्लाइकोसिलेटेड लिग्नांस का भी विभिन्न एंटीऑक्सीडेंट अभिकरणों के माध्यम से मूल्यांकन किया गया था। डीपीपीएच, एबीटीएस, बीओ कैरोटीन- लिनोलिक एसिड विरंजन, टीपीसी और बिजली परख को कम करना। यह पाया गया कि काले / सफेद तिल का अर्क लगभग 200 पीपीएम तक होता है।

सिंथेटिक और वाणिज्यिक एंटीऑक्सीडेंट बीएचटी और बीएचए के बराबर स्थिरीकरण प्रवीणता है, लेकिन सिंथेटिक एंटीऑक्सीडेंट टीबीएचक्यू की तुलना में कम क्षमता है।

सफेद तिल का तेल (डब्ल्यूएसओ) / काले तिल का तेल (बीएसओ) के साथ अलग अनुपात (20:80, 40:60, 50:50, 60:40 और 80:20) के साथ अलसी तेल (लो) और कपास का तेल (सीएसओ) का मिश्रण।)। क्रमशः 110 डिग्री सेल्सियस, 120 डिग्री सेल्सियस और 130 डिग्री सेल्सियस पर रैंकिमेट के साथ उनके ऑक्सीडेटिव स्थिरता के मिश्रण का विश्लेषण किया गया। यह पाया गया कि शुद्ध

बीएसओ में 110 डिग्री सेल्सियस पर 13.62 घंटे की एक प्रेरण अवधि है और डब्ल्यूएसओ में क्रमशः LO पर 110 डिग्री सेल्सियस पर 8.30 घंटे और सीएसओ 3.20 एच और 5.23 एच है। सभी मिश्रणों में से हमने पोषण और आर्थिक लाभों को ध्यान में रखते हुए 50:50 अनुपात की थर्मल स्थिरता का प्रदर्शन किया। एफएएस सामग्री परिणामों से संकेत मिलता है कि बीएसओ के साथ लो और सीएसओ का सम्मिश्रण एफएसए सामग्री को डब्ल्यूएसओ द्वारा अनुपालन करता है। सभी तेल के नमूनों (शुद्ध तेल और मिश्रणों) के पेरोक्साइड मूल्य के परिणाम भंडारण अवधि में वृद्धि के साथ क्रम में बीएसओ > डब्ल्यूएसओ > बीएसओ: सीएसओ (50:50) < डब्ल्यूएसओ: सीएसओ (50:50) < सीएसओ < बीएसओ: लो 50:50 > डब्ल्यूएसओ: लो (50:50) > LO शुद्ध LO और सीएसओ में लिनोलिक एसिड की अच्छी मात्रा होती है जो सीडीवी को बढ़ाने के लिए जिम्मेदार है, लेकिन बीएसओ और डब्ल्यूएसओ के साथ मिश्रित होने पर यह वेतन वृद्धि नियंत्रित किया गया था। एफटीआईआर आंकड़ों ने 5 दिनों के फ्राइंग के बाद आणविक बांडों और कार्यात्मक समूहों में कोई परिवर्तन नहीं दिखाया। यह देखा गया कि फ्राइंग के 5 दिनों के बाद सभी तेल के नमूनों के लिए अवशोषण चोटियां एक समान हैं। फ्राइंग के लिए इस्तेमाल किए जाने वाले आलू के चिप्स दस पैनल के सदस्यों द्वारा संवेदी मूल्यांकन किए गए थे और यहां तक कि नमी हानि और वसा की तेजता के लिए विश्लेषण किया गया था और नमी सामग्री में कोई महत्वपूर्ण (पी < 0.05) अंतर नहीं देखा गया था और आवर्ती फ्राइंग चक्रों के अधीन सूखे आलू के चिप्स की वसा तेज थी।

लो और सीएसओ को तिल के केक (काला / सफेद) से युक्त किया गया जो एथेनॉल और मेथनॉल से निकाला गया था। कच्चे निष्कर्षों को LO और सीएसओ में 50,100,150 और 200 पीपीएम में सिंथेटिक (बीएचए, बीएचटी और टीबीएचक्यू) के साथ जोड़ दिया गया था। तेल के नमूनों को 15 दिनों के लिए 60 डिग्री सेल्सियस पर संग्रहीत किया गया था। निष्कर्षों के Rancimat विश्लेषण गहराई से पाया कि जैव सक्रिय निकालने की क्षमता के लिए बेहतर था

इथेनॉल और मेथनॉल अर्क निष्कर्षों के संबंध में, काले तिल का अर्क (बीएसएमई और बीएसईई) सफेद तिल के अर्क (डब्ल्यूएसएमई और डब्ल्यूएसईई) द्वारा पीछा किए जाने के बाद LO और सीएसओ में सर्वोच्च आईपी वैल्यू दिखाता है। कम सांद्रता पर भी इथेनॉल निकाले जाने पर अर्क बेहतर काम करता है। 15 दिनों के भंडारण के बाद पेरोक्साइड मान के परिणाम से पता चलता है कि एससीई (काला / सफेद) ने लो और सीएसओ ऑक्सीकरण को रोक दिया। इसके अलावा, 200 पीपीएम पर एससीई (काले / सफेद) का एंटीऑक्सीडेंट प्रभाव बीएचए और बीएचटी 200 पीपीएम से बेहतर था। पी-एनिसिडिन वैल्यू, आयोडिन वैल्यू, टोटोक्स वैल्यू और फैटी एसिड सामग्री एंटीऑक्सिडेंट वाले लोगों की तुलना में एंटीऑक्सिडेंट बिना तेल में ज्यादा देखी गई थी।

डिबेटेट तिल केक (काला / सफेद) के मूल्य में वृद्धि बिस्कुट के विकास के द्वारा किया गया था। प्राप्त परिणामों का विश्लेषण करके, यह निष्कर्ष निकाला है कि डिब्बाबंद सफेद तिल का आटा और

बदली काले तिल का आटा बिस्कुट तैयार करने में उनके समग्र गुणवत्ता को प्रभावित किए बिना 50% के स्तर तक शामिल किया जा सकता है। डिस्पैटेड सफेद तिल के आलू कचरे को संवेदी मापदंडों के संदर्भ में उपभोक्ताओं को स्वीकार्य था, बिस्कुट की दोनों किस्मों के भौतिक और वस्त्र संबंधी गुण नियंत्रण बिस्कुट के समान थे। लेकिन बढ़ी हुई प्रोटीन, वसा, कच्ची फाइबर, खनिज सामग्री मौजूदा गेहूं आटा बिस्कुट पर एक अतिरिक्त लाभ है। डिस्पैटेड तिल आटे का निगमन माइक्रोबियल प्रदूषण के प्रतिरोध को बढ़ाता है जिससे बिस्कुट के शेल्फ लाइफ में सुधार होता है। इस प्रकार, तिल के आटे को ढक दिया गया, तेल प्रसंस्करण उद्योग से सह-उत्पाद का इस्तेमाल बिस्कुट या अन्य किफायती और मूल्यवर्धित उत्पादों की तैयारी में उपयोग किया जा सकता है, ताकि बढ़ाया कार्यात्मक और पौष्टिक चिकित्सीय क्षमता हो।

Sl.No.	Title	Page No.
	Certificate	i
	Acknowledgements	ii-iv
	Abstract	v-vii
	Contents	viii-xii
	List of Figures	xiii-xvii
	List of Tables	xviii-xix
	Symbols	xx-xxi
	Abbreviations	xxii-xxv
Chapter I	Introduction	1-14
1.1	Background	1-4
1.2	Sesame seed	5
1.3	Cottonseed oil	5-6
1.4	Linseed or flaxseed oil	6
1.5	Sesame lignans	6-7
1.6	Antioxidant activity of sesame oil	8
1.7	Oil blending and fortification of vegetable oil	8-10
1.8	Utilization of sesame seed as food	10
1.9	Research gaps and the aim of the study	10-11
1.10	Objectives of the study	11
1.11	Organization of the thesis	11-14
Chapter II	Review of Literature	15-58
2.1	Sesame seeds- Cultivation and Production	16-18
2.2	Utilization of sesame seed	18-
2.2.1	Human food	19-22
2.2.1.1	Seeds and kernels	19-21

2.2.1.2 Oil	21
2.2.1.3 Cake and meal	22
2.2.2 Medicinal uses	22-23
2.3 Nutritional Profile of Sesame seed	23-27
2.4 Health Benefits of Sesame Seeds	27-30
2.5 Sesame Lignans	30-41
2.5.1 Chemistry and Biosynthesis of sesame seed lignans	32-38
2.5.1.1 Oil Soluble (or oil dispersed) Lignans	32-33
2.5.1.2 Glycosylated Lignans	33-38
2.5.2 Bioactivity of Sesame Seed Lignans	38-41
2.6 Antioxidants	41-58
2.6.1 Synthetic antioxidants	43-44
2.6.2 Natural antioxidants	44-45
2.6.3 Regulatory status and safety issues of synthetic and natural antioxidants	45-46
2.6.4 Legal implications on the usage of antioxidants	46-49
2.6.5 Potential health effects of dietary antioxidants	49-50
2.6.6 Antioxidant Potential of sesame oil	51-52
2.7 The Oxidative Stability of Sesame Seed Oil	52-53
2.8 Blending of vegetable oils	53-54
2.9 Frying quality and stability of vegetable oils	54-56
2.10 Modifications of oils and fats	57-58
2.10.1 Nutritional Reasons for Modifying a Fat	57
2.10.2 Physical Reasons for Modifying a Fat	57-58
2.11.3 Economic Reasons for Selecting a Fat	58
Chapter III Material and Methods	59-85
3.1 Materials	59
3.1.1 Collection of raw material	60
3.1.2 Chemicals and Reagents	60-61

3.2	Characterization of sesame seeds and defatted sesame cake	61-67
3.2.1	Solvent extraction method	61
3.2.2	Extraction, separation, isolation and characterization of sesame oil soluble	61-62
3.2.3	Extraction, separation, isolation and characterization of sesame glycosylated lignans	63-65
3.2.4	To evaluate the antioxidant potential of sesame oil, sesame oil soluble lignans, sesame defatted cake extract and sesame glycosylated lignans	65-67
3.3	Blending and enrichment of vegetable oil with sesame oil and sesame extract respectively	67-78
3.3.1	Blending of linseed oil (LO) and cottonseed oil (CSO) with white sesame oil (WSO)/ black sesame oil (BSO) in different ratios	67-76
3.3.2	Enrichment of linseed oil (LO) and cottonseed oil (CSO) with defatted sesame cake (DSC) extract of white/black sesame	76-78
3.4	Value addition of defatted sesame cake (black/white) for healthy bakery products	79-85
3.4.1	Preparation of Biscuits	79
3.4.2	Physical analysis of biscuit characteristics	79-80
3.4.3	Nutritional and mineral analysis	80
3.4.4	Textural properties of dough and biscuits	81-83
3.4.5	Color analysis	83
3.4.6	Microbial contamination analysis of the biscuits	83-84
3.4.7	Cytotoxicity analysis	84
3.4.8	Sensory analysis	84-85
3.5	Statistical analysis	85

	Results and Discussions	86-181
Chapter IV	Characterization of black and white sesame seeds	88-106
4.1	Isolation, TLC, spectroscopic analysis and HPLC of oil – soluble lignans from sesame oil	88-90
4.2	Isolation, TLC, spectroscopic analysis and HPLC of water soluble lignans from defatted sesame cake	91-94
4.3	Antioxidant potential of sesame oil, sesame oil soluble lignans, defatted sesame cake and sesame glycosylated lignans	95-106
Chapter V	Blending and fortification of vegetable oil by with sesame oil and extract	107-158
5.1	Blending of Linseed oil and cottonseed oil	107-136
5.1.1	Oxidative stability of blended oils at different temperature range with sesame oil (black/white)	107-111
5.1.2	Fatty acid composition of pure and blended oils at initial stage	112-116
5.1.3	Chemical Properties of Pure and Blended Oil	116-136
5.2	Enrichment of linseed oil and cottonseed oil with sesame extract (methanol / ethanol)	136-157
5.2.1	Effect on oxidative stability of cottonseed oil and linseed oil with DSC extract	137-143
5.2.2	Effect of the extract on the oxidation of linseed oil and cottonseed oil	144-158
Chapter VI	Value addition of defatted sesame cake (white/black) for healthy bakery product	159-182
6.1	Standardization and Development of Biscuits Using Defatted sesame Flour (Black/White)	159-160
6.2	Nutritional and mineral composition	160-136
6.3	Physical attributes of defatted sesame flour biscuits	163-165

6.4	Textural profile analysis of dough and biscuits	165-169
6.5	Color analysis	169-171
6.6	Microbial contamination analysis	171-174
6.7	Cytotoxicity analysis	174-176
6.8	Sensory attributes of defatted sesame flour biscuits	176-182
Chapter VII	Conclusion and future scope	183-192
	References	193-227
	Appendix	228
	Bio Data	229-232

Figure No.	Title	Page No.
1.1	World scenario of sesame seed production (2013-2014)	1
1.2	States producing different varieties of sesame seeds	3
2.1	Sesame farming	17
2.2	A flowering sesame plant	17
2.3	Sesame seeds	17
2.4	Oil soluble or oil dispersed sesame lignans	34
2.5	Lignan glycosides isolated from defatted sesame flour	35
2.6	Possible interactions of primary and secondary antioxidants with lipid oxidation pathway in foods.	42
4.1	Sesame oil soluble lignans obtained from sesame oil (black and white) after extarction	89
4.2	Spots of sesamin/sesamolin/sesamol on a TLC plate	89
4.3	HPLC chromatograph of sesamol, sesamin and sesamolin	90
4.4	Experimental set up for column chromatography for sesamolinol diglucoside (SD-1)	91
4.5	PLC plates of black sesame (BS) and white sesame (WS) for separation of sesaminol diglucoside (SD-2)	92
4.6	Experimental set up for column chromatography for sesaminol triglucoside (ST)	92
4.7	TLC plates of black sesame (BS) and white sesame (WS) with anisaldehyde, sulphuric acid and iodine vapor	93

4.8	HPLC chromatograph of sesamolinal diglucoside, sesaminol diglucoside and sesaminol triglucoside	94
4.9a	Antioxidant potential of sesame oil and oil soluble lignans with reducing power assay	96
4.9b	Antioxidant potential of sesame defatted cake and glycosylated lignans with reducing power assay	97
4.10a	Antioxidant potential of sesame oil and oil soluble lignans with DPPH assay	98
4.10b	Antioxidant potential of sesame defatted cake and glycosylated lignans with DPPH assay	99
4.11a	Antioxidant potential of sesame oil and oil soluble lignans with ABTS assay	101
4.11b	Antioxidant potential of sesame defatted cake and glycosylated lignans with ABTS assay	101
4.12a	Antioxidant potential of sesame oil and oil soluble lignans with β carotene bleaching assay	103
4.12b	Antioxidant potential of sesame defatted cake and glycosylated lignans with β - carotene bleaching assay	103
5.1	Free fatty acid formation in oils and oil blends during five frying cycles	117
5.2	Peroxide value formation in oils and oil blends during five frying cycles	119
5.3	Conjugated diene value (CDV) in oils and oil blends during five frying cycles	121
5.4	Color value in oils and oil blends during five frying cycles	122

5.5	Photographs of the fresh oils and fried oils obtained from during five frying days	123
5.6	FTIR spectra of fried oil and oil blends (LO) obtained before frying and after completion of frying	128
5.7	FTIR spectra of fried oil and oil blends (CSO) obtained before frying and after completion of frying	129
5.8	Effect of frying cycles on moisture content during frying of potato chips	131
5.9	Effect of frying cycles on fat uptake during frying of potato chips	132
5.10a	Quantitative response of potato chips odor, color, crispiness and taste after day 1 of frying	133
5.10b	Quantitative response of potato chips odor, color, crispiness and taste after day 3 of frying	133
5.10c	Quantitative response of potato chips odor, color, crispiness and taste after day 5 of frying	134
5.11	Photographs of the fresh potato chips and fried potato chips obtained from during five frying days	135
5.12	Photographs of the cottonseed oil and linseed oil sample added with methanol / ethanol defatted sesame cake extract (black/white) before keeping in oven at 60 °C	137
5.13	Effect of time and concentration of added defatted sesame extract on stability of cottonseed oil	139
5.14	Effect of time and concentration of added defatted sesame extract on stability of linseed oil	140

5.15	Effect of changing antioxidants and oils on antioxidant index (AI)	141
5.16	Effect of changing concentration of antioxidants on Antioxidant Index after 15 days of storage (Cottonseed oil)	143
5.17	Effect of changing concentration of antioxidants on Antioxidant Index after 15 days of storage (Linseed oil)	143
5.18	Effect of time and concentration on peroxide value of added defatted sesame extract on stability of linseed oil	145
5.19	Effect of time and concentration on peroxide value of added defatted sesame extract on stability of cottonseed oil	146
5.20	Effect of time and concentration on p- anisidine value of added defatted sesame extract on stability of linseed oil	147
5.21	Effect of time and concentration on p- anisidine value of added defatted sesame extract on stability of cottonseed oil	148
5.22	Effect of time and concentration on Totox value of added defatted sesame extract on stability of linseed oil	149
5.23	Effect of time and concentration on Totox value of added defatted sesame extract on stability of cottonseed oil	150
5.24	Effect of time and concentration on Iodine value of added defatted sesame extract on stability of linseed oil	151
5.25	Effect of time and concentration on Iodine value of added defatted sesame extract on stability of cottonseed oil	152
5.26	Effect of time and concentration on free fatty acid of added defatted sesame extract on stability of linseed oil	154

5.27	Effect of time and concentration on free fatty acid of added defatted sesame extract on stability of cottonseed oil	155
6.1a	Comparison of dough stickiness among control, white sesame biscuits and black sesame biscuits	166
6.1b	Comparison of dough strength among control, white sesame biscuit and Black sesame biscuits	167
6.2	Microbial count of Defatted sesame flour biscuit during 90 days of storage	171
6.3	Photographs of microbial analysis for biscuit samples during 90 days of storage	173
6.4a	Viability percentage of water extract of defatted sesame flour biscuits	175
6.4b	Viability percentage of methanol extract of defatted sesame flour biscuits	175
6.5	Photographs of cell growth observed after 90 days of storage under microscope	176
6.6	Sensory attributes of defatted sesame flour biscuits	178
6.7	Consumer preference for experimental biscuits	179
6.8	Photographs of control and experimental biscuits	180

TABLE NO.	TITLE	PAGE NO.
1.1	Growing areas of sesame seeds in India	2
2.1	Ancient recipes of sesame seeds across the world	19
2.2	Nutritional profile of sesame seed	25-27
2.3	Major sesame seed lignans and lignan glucosides range, molecular formula, UV absorption and extinction coefficient	30-31
2.4	Bioactivities of major sesame lignans	36-38
2.5	Permissible limit of additive as antioxidants source allowed by Codex Alimentarius Commission for vegetable oils and fats	48-49
3.1	Details of the preparation of blends with sesame oil (Black/ White)	68-69
3.2	Operational parameters of Rancimat apparatus	70
3.3	Different oil blends for frying stability test	71
4.1	Yields of oil soluble lignans from sesame oil (black/white)	88
4.2	Quantification of sesame oil soluble lignans	90
4.3	Yields of water soluble lignans from defatted sesame cake (black/white)	94
4.4	Total Phenol content (TPC) of sesame oil, sesame lignans and defatted sesame cake	95
5.1	Induction period of BSO, WSO, LO, BSO: LO and WSO: LO during different temperature range	109
5.2	Induction period of BSO, WSO, CSO, BSO: CSO and WSO: CSO during different temperature range	110

5.3	Preliminary assessment of effect of blending BSO and WSO with LO in different ratios at different temperatures	111
5.4	Preliminary assessment of effect of blending BSO and WSO with CSO in different ratios at different temperatures	111
5.5	Fatty acid composition of BSO, WSO, LO and their blends (in different ratios) at initial stage	114
5.6	Fatty acid composition of BSO, WSO, CSO and their blends (in different ratios) at initial stage	115
5.7	Fatty acid composition of BSO, WSO, LO, CSO, BSO: LO, WSO: LO, BSO: CSO and WSO: CSO before frying and after frying	125
5.8	Functional groups of BSO, WSO, LO, BSO: LO and WSO: LO before frying and after frying	126-127
6.1	Incorporation levels of defatted sesame white/black flour in biscuit preparation	159
6.2	Nutritional composition of wheat flour, defatted sesame flour and defatted sesame flour biscuits	161
6.3	Mineral composition of wheat flour, defatted sesame flour and defatted sesame flour biscuits	162
6.4	Physical attributes of defatted sesame flour biscuits	164
6.5	Textural profile analysis of defatted sesame flour dough and biscuits	168
6.6	Color analysis of defatted sesame cake biscuits	170

%	Percent
°C	Degree celsius
µg	Microgram
mg	Miligram
g	Gram
nm	Nanometre
µm	Micrometre
mm	Millimetre
cm	Centimetre
µL	microlitre
mL	Mililitre
L	Litre
rpm	Revolutions per minute
h	Hour
min	Minute
s	Second
mM	Milimolar
ppm	Parts per million
>	Greater than
Na ₂ CO ₃	Anhydrous sodium carbonate
Na ₂ SO ₄	Anhydrous sodium sulphate

CH_3COONa	Sodium acetate
$\text{K}_3\text{Fe}(\text{CN})_6$	Potassium ferricyanide
FeCl_3	Ferric chloride
R_f	Rate of flow
v/v	Volume / Volume
λ_{max}	Maximum wavelength
ϵ_{max}	Absorption coefficient
pH	Potential of hydrogen
M	Molar
mg GAE/g	milligrams gallic acid equivalents per gram
Abs^0	Absorbance at zero minute
Abs^{120}	Absorbance at 120 minute
~	Approximately
CO_2	Carbon dioxide
β	Beta

FAO	Food and agriculture organization
IOPEPC	Indian Oilseeds and Produce Export Promotion Council
ICMR	Indian Council of Medical Research
ICAR	Indian Council of Agricultural Research
IUPAC	International Union of Pure and Applied Chemistry
ROS	Reactive Oxygen Species
DNA	Deoxyribonucleic acid
PUFA	Polyunsaturated fatty acids
ALA	α -linolenic acid
FA	Fatty acid
HPLC	High-performance liquid chromatography
LCMS	Liquid chromatography–mass spectrometry
BHA	Butylated hydroxyanisole
BHT	Butylated hydroxytoluene
TBHQ	Tertiary-butyl hydroquinone
TLC	Thin layer chromatography
PLC	Preparative liquid chromatography
DPPH	1, 1- diphenyl-2-picrylhydrazyl radical
ABTS	2, 2-azinobis-(3-ethyl-benzothiazoline-6-sulphonic acid)
CFTRI	Central Food Technology Research Institute
RP	Reverse phase
ODS	Octadecyl-silica

SD 1	Sesamolinol diglucoside
SD 2	Sesaminol diglucoside
ST	Sesaminol triglucoside
DSF	Defatted sesame flour
UV	Ultra Violet
TEAC	Trolox equivalent antioxidant capacity
LO	Linseed oil
CSO	Cottonseed oil
BSO	Black sesame oil
WSO	White sesame oil
OSI	Oxidative stability index
FAME	Fatty acid methyl esters
FTIR	Fourier Transform Infra-Red
FFA	Free fatty acid
NaOH	Sodium chloride
PV	Peroxide value
CDV	Conjugated Diene value
IV	Iodine value
AV	p- Anisidine value
L*	Lightness
a*	Redness
b*	Yellowness
MSA	Mannitol salt agar

NA	Nutrient agar
TSA	Tryptose soy agar
EMB	Eosin methylene blue
PDA	Potato dextrose agar
MTT	(3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide)
DMSO	dimethyl sulfoxide
DMEM	Dulbecco's Modified Eagle Medium
PBS	phosphate-buffered saline
BS	Black sesame
WS	White sesame
TPC	Total phenol content
BSOL	Black sesame oil lignan
WSOL	White sesame oil lignan
BSO	Black sesame oil
WSO	White sesame oil
WSDCL	White sesame defatted cake lignan
BSDC	Black sesame defatted cake
WSDC	White sesame defatted cake
BSDCL	Black sesame defatted cake lignan
IP	Induction period
PF	Protection factor
SFA	Saturated fatty acid
MUFA	Mono unsaturated fatty acid

SMP	SFA: MUFA: PUFA
WHO	World health organization
BSME	Black sesame methanol extract
WSME	White sesame methanol extract
BSEE	Black sesame ethanol extract
WSEE	White sesame ethanol extract
AI	Antioxidant index
WF	Wheat flour
WSB	White sesame biscuit
BSB	Black sesame biscuit
DWSF	Defatted white sesame flour
DBSF	Defatted black sesame flour
Control H ₂ O	Control biscuit extract in water
BSB H ₂ O	Black sesame biscuit extract in water
WSB H ₂ O	White sesame biscuit extract in water
Control MEOH	Control biscuit extract in methanol
BSB MEOH	Black sesame biscuit extract in methanol
WSB MEOH	White sesame biscuit extract in methanol
DBSB	Defatted black sesame biscuit
DWSB	Defatted white sesame biscuit