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The Sesame (*Sesamum indicum L.*) Value Chain and Microbiological Quality of Crude Sesame Oil, a Case Study in Western Tigray, Ethiopia

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Abstract

Crude sesame oil (CSO) is the widely used unrefined edible oil. Storage instability, off-flavour, and discoloration are, however, challenges in the industry. The purpose of this study was to map the sesame value chain, assess the suitability of CSO extraction plant, and analyze the microbial quality of CSO and premises environment. A structured Questionnaire and checklist were used to assess the sesame value chain and evaluate the suitability of the CSO extraction plant. Microbiological quality assessment was conducted using standard analytical methods. Stakeholders in the sesame value chain were inclusive of farmers, market mediators, traders, regulatory, extension workers and researchers. Though, illiteracy, inadequate technology, and infrastructure were the drawbacks. The CSO extraction plant was suitable apart from inadequate ingredients and CSO handling and unhygienic practices. Total aerobic bacteria (4.34 - 5.06 $\log_{10}$ CFU/m^2 on swap surfaces, 2.44 $\log_{10}$ CFU/g in CSO), total Coliforms (5.81 $\log_{10}$ CFU/g of animal manure and 1.36 $\log_{10}$ CFU of indoor air after extraction), yeasts and moulds (2.31 $\log_{10}$ CFU/g of sesame seed and CSO and 4.47 $\log_{10}$ CFU/m^2 of swap sample), *Aspergillus* species (1.17 - 1.33 $\log_{10}$ CFU/g of sesame seed/CSO, 3.37 - 3.50 $\log_{10}$ CFU/m^2 of swap samples), and *Staphylococcus aureus* (2.09 $\log_{10}$ CFU/g of CSO, 2.56 - 3.22 $\log_{10}$ CFU/m^2 of surface swaps, 3.26 - 3.77 $\log_{10}$ CFU/protective clothing, 0.74 - 1.82 $\log_{10}$ CFU of the indoor and outdoor air) were detected. *Escherichia coli*, Salmonella and Shigella were not detected. In conclusion, potential microbial pathogens were detected to impose food safety problems and economic loss. To improve the sesame value chain and CSO quality workers training on good handling and hygienic practices and thoughtful regulatory implementation are significant.

Keywords

Crude Sesame Oil, Value Chain, Suitability, Microbial Quality, Pathogens

1. Introduction

Sesame (*Sesamum indicum*) is a flowering annual plant, belonging to the family *Pedaliaceae*. It requires an average of 90 days from the time of plantation till maturity [1]. It is tolerant to drought and flexible to different soil types, harsh conditions, diversified agro-ecological conditions and the pod bursts when it reaches maturity. Sesame is sensitive to a number of environmental and biological factors such as rainfall, wind, hotness and harvesting time [2], soil quality and moistness [3], which impacts productivity and quality. The global sesame production is about 2.211,339 million tons. Sesame production in Ethiopia is about 262,654 tons by 2019 (307 million USD) [4] about 8.96% share in the global sesame market [5]. Sesame productivity in western Tigray is within the range of 2044 kg/ha - 400 kg/ha while highly variable with and without the application of insecticides 651 kg/ha, average [5] variability with sesame breed, plantation location, use of fertilizers and agricultural practices with an average yield obtained 400 kg/ha [4]. Sesame productivity yield is as high as 2044 kg/ha [6] and (1161.5 kg/ha - 756.5 kg/ha) regardless of the variation of cultivation year, varieties and agro-climatic condition [7].

Food-borne illness is a serious problem that is appealing to global attention due to widespread disease and public outcry. Food Safety practices are very important in the food value chain to protect public health as well as food and economic security [8] [9]. It is essential to understand all possible hazards (microbial, chemical, and physical) existing or retrieving within the food value chain for better food safety practices. Microorganisms that exist in the environment, food, soil, water, and other associated animals might be a potential hazard to human, animal, and the environment. Microbial prevalence assessment and their severity to cause complications is critical for safer food distribution, quality assurance and develop operational practices. However, Inadequacy of good practices and written plans remain a challenge due to numerous reasons such as insufficient skill [10], inadequate safety practices, failure to implement and control good processing and handling practices [11], unhygienic conditions [12] [13], lack of transparency [14], inappropriateness of technology, and financial constraints [15].

Microorganisms colonize in a dynamic environment and a reason for a wider spectrum of infection [16] in all food categories in the value chain and associated surfaces [17] [18] [19], dairy value chain [20] [21], meat value chain [22] [23], and ready-to-Eat and salad [24] [25] Public health is threatened due to the prevalence of live organisms and their metabolites in food or ingested, contact is made with contaminated surfaces and inhaling of polluted air. Food poisoning/infection occurs after 30 minutes or a few hours of food consumption. The severity of sickness depends on the nature of the microbe and its metabolites apparent for three or more days to manifest [26]. Symptoms of foodborne illness include nausea, vomiting, diarrhoea, abdominal pain, Headache, and chills with/without fever [27], acute gastroenteritis [28], and sepsis infection [29]. Skin and soft tissues inflammation and blemishes atopic dermatitis which triggers expression of cytokine [30]. Microbial poisoning or infection is persistent in a wider group of population [31] causes a high mortality rate among children [32] [33] [34]. Microbial-induced illnesses evade the host immune system and bone impairment [35].

Crude sesame oil (CSO) is the quality oil widely used as cooking oil, salad dressing, blending component in the oil refinery, and ointment. CSO studied in this study is unrefined mechanically extracted sesame oil in a small-scale extraction plant. CSO develops off-flavour, discoloration, and unstable during storage. The causes for the quality and storage instability of CSO might be associated with the prevalence of contaminants due to inappropriate handling and poor hygienic practices. The purpose of this study was to assess the Sesame value chain, CSO extraction plant suitability, hygienic practices, and microbial quality to reduce oil loss, improve stability and promote good practices.

2. Materials and Methods

2.1. Study Design and Setting

Survey data was collected in western Tigray, Ethiopia (latitude 14.032334 and longitude 38.316573), temperature (17.5˚C - 41.7˚C), and altitudes (500 - 1300 meters) above sea level and rainfall (480 - 633 mm) [2] in the period between August 2019 to February 2020. A questionnaire was distributed (320 participants), presented in **Table 1** drawn according to [36] to outline the sesame value chain and stakeholders in the value chain as indicated in **Figure 1**. A checklist was used to assess the suitability of the plant for CSO extraction as summarized in **Table 4**.

2.2. Sample Collection

Sesame seed, CSO, and water were sampled to examine for the prevalence of microbial contaminants. The source of microbial contamination was traced from the environmental air, the surface of the machinery and premises, protective clothes, and supplies. Sesame seed (100 g) was sampled from the CSO extraction store using a grain sampler from the bottom, top, and middle of the package, harvested during 2019/2020 in western Tigray. The seed was cleaned, homogenized, grounded (mortar and pestle grinder), and sieved 1 mm mesh size. About 10 ± 0.05 g of grounded sesame seed was mixed with 90 ml peptone water, diluted, transferred aseptically to the appropriate agar containing petri dish. CSO (100 ml) was sampled in a blue-brown glass bottle and 1 ml of CSO was aseptically transferred to media for microbial prevalence testing. Pipe Water (100 ml)

Table 1. Characteristics of stakeholders involved in the sesame value chain in western Tigray, Ethiopia (n = 320).

Variables	Frequency	Percent
Gender		
Women	119	37.2
Men	201	62.8
Age (years)		
18 - 25	7	2.3
26 - 30	75	23.3
>30	238	74.4
Experience (in years)		
<1	15	4.7
1 - 2	60	18.6
2 - 5	119	37.2
>5	126	39.5
Educational status		
No formal education	82	25.6
8^{th} - 12^{th} grade	90	27.9
Certificate and above	150	46.7
Role of stakeholders		
Market intermediary	21	6.7
Traders	71	22.2
Farmer	86	26.7
Regulatory authorities	29	8.9
Technical expert	87	27.2
Researcher	26	8.2
Communication		
Telephone	160	50
Letter	40	12.5
Telephone, Letter and Internet	120	37.5
Management Support		
No	60	18.9
Yes	260	81.1

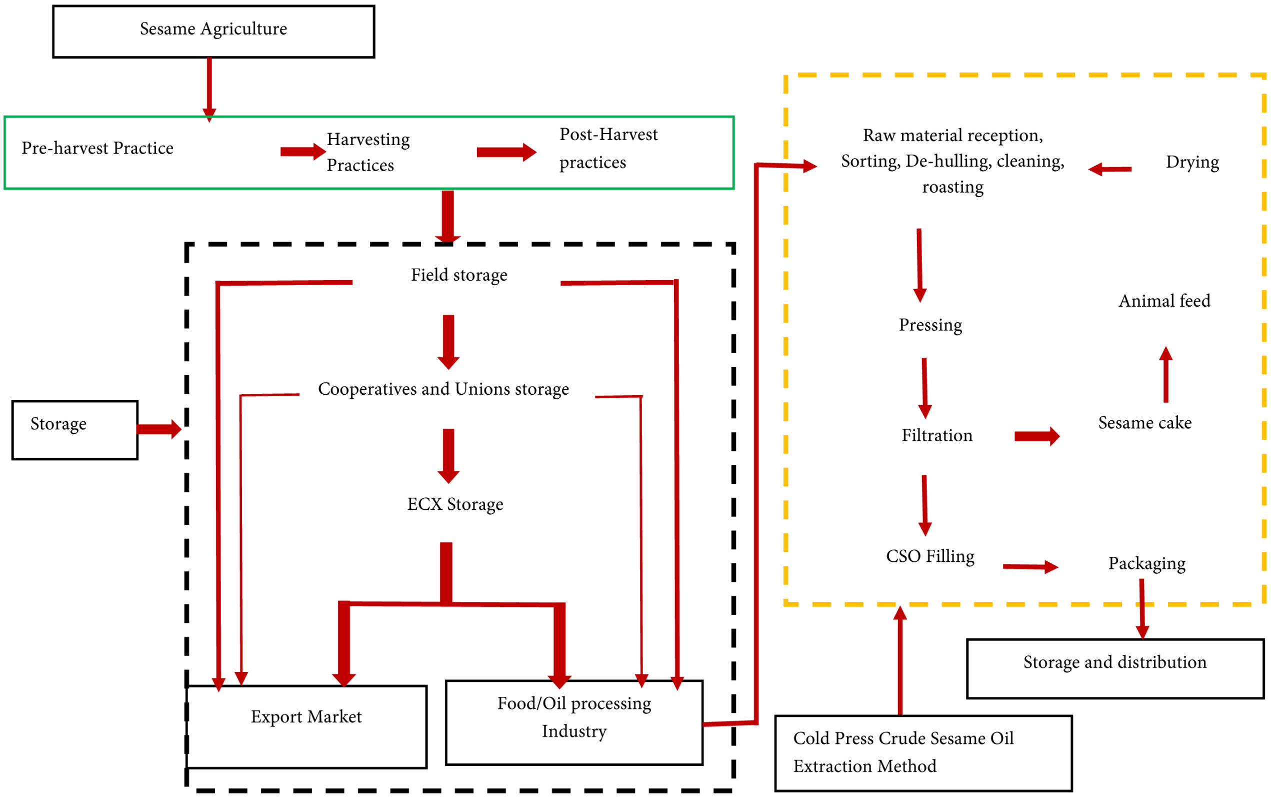

Figure 1. Schematic description of sesame value chain in western Tigray, Ethiopia.

were sampled in a blue-brown glass bottle after 1 - 2 minutes flow and groundwater (100 ml) fetched from the hole in a polyethylene plastic placed in a blue-brown glass (100 ml) in an ice-pack to analyse the microbial quality within 2 hours. The surface of the milling and pressing machine, filtration machine, filling machine, the floor of sesame store, and wall of the extraction room was swabbed. Swabbing was performed in a 10 × 10 cm area, in difficult to clean or reach surfaces scrubbed in two perpendicular directions using a moistened cotton swab in buffered peptone water. Protective gown from inner underarms and the inner side of clove was sampled by peptone water moistened cotton swap scraped, placed, and kept for 5 minutes. The swab sample was placed in a sterilized plastic bag, break-off in a test tube according to the method described by [37]. The indoor and outdoor air sample was collected during/after production using a passive sampling method 1 m above the floor and 1m away from the wall in an agar media containing petri dish (140 mm diameter) left open for 1 hour according to the procedure of ISO 14698 air sampling described by [38]. Incubation methods for microbial cultivation were performed as summarized in Table 2.

2.3. Microbial Enumeration

Microbial colonies were enumerated using an illuminated magnifying colony counter, organized in an excel Microsoft transformed into $\log_{10}$ colony-forming

Table 2. Summery of microbial assessment methods applied to assess CSO microbial quality and CSO plant.

Target microorganism	Agar	Incubation condition	Method
Aerobic plate count	Pour plate methods in a Plate count agar (AOAC 966.23)	37˚C/48 h	[39]
Total Coliforms	Pour plate method in a Plate count agar (ISO 9308-1:2014)	37˚C/48 h	[40]
Escherichia coli	Pour plate method in a MacConkey agar	44.5˚C/24 h	[41]
Yeast/mould	Spread plate method in a Rose Bengal Chloramphenicol Agar	22˚C/7 days	[42]
Aspergillus fungi	Sabouraud Dextrose Agar chloramphenicol powder as anti-bacterial growth inhibitor	25˚C/5 - 7 days	[43]
Salmonella and *Shigella*	Xylose lysine desoxycholate agar with Rappaport-Vassiliadis Soy Peptone Broth	37˚C/18 h	[44]
Staphylococcus aureus	Mannitol Salt Agar (Oxoid), gram staining, coagulase test	32˚C/48 h	[45]

units. The microbial colony was expressed as $\log_{10}$ CFU/g for sesame seed, $\log_{10}$ CFU/ml for CSO and water, $\log_{10}$ CFU/m^2 for swab samples, and $\log_{10}$ CFU/hr for air sample.

2.4. Data Analysis

A questionnaire was presented and later translated into local language (Tigrigna) for a comprehensive understanding of the terms and articulated after random questioner distribution. The questionnaire with missing responses was cleaned, inserted into IBM SPSS Statistics version 22 software to generate descriptive data (frequency and percentage) of respondents involved in the sesame value chain. The suitability of the plant for oil extraction and sanitary conditions was tabulated. The APC, total Coliforms, fungi, *Aspergillus species*, *staphylococcus aureus* count colonies data were tabulated and converted $\log_{10}$ colony-forming units in an excel sheet, and analysis was performed using analysis of variance (ANOVA) in a SAS V8 software to separate the means and standard deviations. Tukey was used to test for statistical significance difference at a P-value of 0.05.

3. Results and Discussion

3.1. Stakeholder Characterization

Stakeholders in the sesame value chain were farmers, researchers, regulatory authorities, technical experts, and traders with active and passive participation, as presented in Table 1. Stakeholders were gender-inclusive (37.2% women, 62.8% men) and sesame farmers (26.7%) at different scales of farming. In excess of 74% of the stakeholders were more than 30 years of age increases commitment and effective resource utilization for improved sesame value chain [1]. The composition, education, and experience of stakeholders in a particular value chain improve farming, value addition, and resource utilization [46]. Stakeholders with no formal education (25.6%) were small-scale farmers and low-wage employees. Studies reported that farmers and low-wage employees with inadequate technology and traditional farming practices intensify sesame post-harvest and economic loss [47] [48]. Farmers' farming practices were laborious and time-consuming in particular the small farmers are lack or unaffordability rent of sesame driller/ploughing machine [49]. Besides the economic value, safety might be compromised due to the prevalence of toxigenic microorganisms, see **Table 5**. Stakeholders in the sesame value chain operate their routine practices with inadequate support and lack of training (18.9%) on pesticide use, lack of access to improved seed variety, disease, and pest attack and weed prevention, sesame harvesting and handling, value addition, and market linkage [5].

Stakeholders with technical expertise surveyed (27.2%) were an agricultural extension, soil and water conservation, pest control, post-harvest management, community health, and post-harvest technology. Regulatory (8.9%) were from health extension, agricultural specialist to control the use of pesticides, post-harvest technologist and food scientists regulate agro-processing including sesame based products processing. A multi-disciplined researcher (8.2%) with the aim of improving sesame productivity, introducing sesame new breeds and diseases resistances [50], post-harvest handling, and loss reduction, and value addition [47] [51] were involved. Traders (22.2%) include cooperatives, unions and individuals who supply sesame to the global market guided through the Ethiopian commodity exchange office (EXC) (market intermediary, 6.7%) or to the local market. Obtaining reliable and timely market information, financial constraints, deviation price setting, and identifying marketing channels are the challenges of sesame traders [52] [53]. Sesame traders collect sesame and store in a thin metal sheet-built storage warehouse or well-cemented block warehouses. The metal warehouses were not clean of grasses and bush around. While, sesame storage in a warehouse build of blocks, cemented floor, inlet/exit door, and windows with sufficient space and a recommended side dimensions were clean of grasses and bush. However, extended storage, loading, and unloading, transportation, and packaging incur high economic loss besides the field pre-harvest loss.

3.2. Sesame Value Chain

Sesame (*Sesamum indicum L.*) locally named as "Selit" is economically important. The sesame value chain presented in **Figure 1**, involves organic sesame agriculture, sesame post-harvest practices, distribution and marketing, and processing. The economic value of sesame was reported within the range of USD 307, 512 - 619,033 million annually in the year of 2011-2017 with an average yield of 756.5 - 906.3 kg/ha considering agro-climatic condition, season, rainfall, and other determinant parameters variability [7]. Sesame plantation and production include land preparation, accessibility of improved seed and fertilizer, cultivation, weed prevention and use of pesticides, labour, and technology. Farmers interviewed revealed rainfall variability, weed prevention, climatic factors such as unexpected rainfall and wind at the stage of maturity, inadequate labour, or absences of technology for harvesting are critical challenges. Capsule colour or aging of sesame leaves are the indicators for harvesting. The cuts of sesame bundle locally named as "Hilla" placed in the field are susceptible to insects such as termite, sesame webworm, sesame seed bug, gall midge, and others causes sesame post-harvest loss and quality defect. To reduce the loss and quality defect of sesame studies suggested the bundles placed in a plastic mat of clean area or application of pesticides around the bundle to prevent insect attack. After 15 days of drying with its parent plant seed is thresh separated in a plastic mat. However, During harvesting and early post-harvest practices about 17% of sesame was lost due to poor/inappropriate farming, delaying harvesting time, shattering, and field drying, threshing, loading, and unloading during bundle transportation excluding storage loss [51].

Sesame stored in cooperative warehouse build-up of stainless steel experiences high loss. The stainless steel warehouse increases inside environment hotness which is conducive for microbial growth. Furthermore, the external environment of the warehouse was not clear of grass, shrub, and trees to hide out macro-organisms such as mice and rats, birds, and termites to attack sesame during storage. The unions' warehouses were, however comparably clean, built of cinder concrete block smoothly cemented floor and wall. The arrangement of sesame packs in a sack of 100 kg in the cemented warehouses with adequate space and recommended opening for ventilation. Unprocessed sesame seed value addition includes cleaning, colour sorting, and de-hulling process satisfying the sanitary and phytosanitary requirement. Regulatory authorities regulate sesame agriculture and trains farmers to avoid the use of unregistered or unpermitted pesticides in the farm, harvesting, and storage. To overcome the challenges in the sesame value chain studies conducted in Myanmar suggested that integrated sesame agriculture and value chain is critical to improve sesame productivity and reduce sesame loss [54].

Sesame is one of the main export crops in foreign currency earnings to the country and income generation. EXC certifies sesame via examining the quality and grading based on impurity level, contrasting colour, and identifying the appropriate market channel, as presented in **Table 3**. Sesame marketing continues to grow conversely quality defects and inadequate value addition, illegal broker interference, lack of storage and transportation, inadequate infrastructure, the disparity between stakeholders, limited access to market, and low price are the major challenges. Besides export earnings from the unprocessed seed, sesame is locally used for food and non-food application. It is a primary ingredient for refined and unrefined edible oil extraction. Sesame cake is a by-product of edible oil extraction used for animal feed and soap production. Sesame straw is also used for animal feed and biomass energy sources in the cement industry.

Table 3. Grading parameters of western Tigray sesame seed based on the Ethiopian commodity exchange.

Parameters	Grade 1	Grade 2	Grade 3	Grade 4	UG
Whitish Humera and reddish sesame seed					
Total Impurity (Foreign Matter and DSW) Max % by weight	1	3	5	7	15
Contrasting Colour, max % by Weight	1	2	4	6	7
Mixed Humera and Mixed Reddish Sesame Seed					
Total Impurity (Foreign Matter and DSW) Max % by weight	1	3	5	7	15
Contrasting Colour, max % by Weight			>7		

DSW: Damaged, Shriveled, Weevil bored.

3.3. CSO Extraction Establishment Suitability and Hygienic Condition

The suitability of the extraction plant for oil extraction was determined, preconditions and criteria to fulfil in the CSO extraction premises summarized in Table 4. Oil extraction plant located in a government-built lodging designed for small and medium manufacturing enterprise in proximity to the sesame farm, infrastructure, and market free from nearby chemical manufacturing plant or sewage accumulation/treatment. The plant was built with cinder concrete blocks, the wall and floor were smoothly cemented and a stainless-steel cover of the roof with appropriate drainage system and sanitation friendly. Though the exterior environment was clear of grass, shrub, and trees and yet dusty due to the dry and windy environment and randomly pole-installed light for security purposes. Studies suggested that lightening ensures quality preservation, sanitary purpose, security, and avoid insect attraction and contamination [55]. The opening was sealed to avoid insects and rodent entrances, however, the only door for raw material and end product dispatch was old and folded somehow. The extraction equipment (milling, pressing, and filtration machine) was suitable. Nevertheless, overall sanitation and handling practices were unfortunate. The challenges identified were limited access to sanitary inputs such as enough water, detergents, and other cleaning inputs, poor sanitation infrastructure/structure including operation and maintenance, and poor hygiene habits. Lack of written sanitation program and presentation in the appropriate location, sporadic cleaning of the machines and premises, inadequate supply of hygienic materials, and movement of workers liable for dirt accumulation and cross-contamination. Sesame seed was procured either from farmers, cooperatives and other retailers without quality specification contribute to affect quality and safety aspects. Sesame store in the extraction plant lacks ventilation and temperature control creates a conducive condition for the revival of contaminants due to the warm climate condition and unclean premises.

Table 4. Assessment of the CSO extraction establishment suitability and hygienic condition.

Premises Assessment	Yes	No	Status
Is the extraction establishment located proximity to raw material, infrastructure and market?	√		Suitable
Is the extraction establishment located in a weed, grass, brush, and dust free environment, so that pest and macro-organisms cannot hide?		√	Unsuitable
Is the internal premises smooth and hygiene friendly?		√	Unsuitable
Does the establishment have a separated storage of raw material and end product?	√		Suitable
Is the storage and production area doors and windows have fine mesh screens or tightly sealed to keep out insects?	√		Suitable
Is there any leaks/cracks in the floor, roof, window to allow sky light, and pest entry or water leak?		√	Suitable
Is the establishment designed with a proper drainage and waste disposal system?	√		Suitable
Suitability for CSO extraction			
Is the establishment primarily designed for oil extraction?		√	Suitable
Is the equipment primarily designed or suitable for oil extraction?	√		Suitable
Does the establishment have equipment calibration, maintenance and monitoring plan with appropriate layout?	√		Suitable
Is the oil extraction establishment have quality Specification, control and monitory?		√	Unsuitable
Is the establishment equipped with temperature, ventilation, lightening, pest, and humidity control?		√	Unsuitable
Does the establishment have transportation?	√		Suitable
Sanitary condition			
Is the water used in the establishment treated?		√	Unsuitable
Is the cleaning and washing facilities furnished with soap, detergents, and cleaning equipment for personal and establishment sanitary with enough water supply?	√		Suitable
Is there any presence of domestic animals around the establishment?	√		Unsuitable
Is there enough protective clothing such as gown, air cover and glove to oil handlers and operators?	√		Suitable
Are the workers provided a separated area for eating and drinking?		√	Unsuitable
Is the waste quickly removed to appropriate bins and kept covered?	√		Suitable
Does the establishment have pest control system?		√	Unsuitable

3.4. Microbial Quality

Microbial contaminants of aerobic bacteria, total Coliforms, fungi, *Aspergillus species*, and *staphylococcus aureus* were predominantly detected and quantified, presented in Table 5. *Staphylococcus aureus* and *Aspergillus species* were the potential pathogens identified. However, Escherichia coli, salmonella, and Shigella were not detected. The source of microbial contaminants was found to be the extraction establishment after examining the surface of equipment and premises, protective clothing, indoor, and outdoor air and water. Aerobic plate count bacteria (APC) are microbial hazards grow and survive in an aerobic and mesophilic environment. APC are important microbial indicators to assess product quality such as organoleptic quality and shelf life, manufacturing and hygienic practices, and cleanness of food establishment and product safety to a lesser extent [56] [57] and process efficiency for safer produce [58]. The APC for

Table 5. The Microbial count ($\log_{10}$ colony forming unit) of supply, CSO, surface, protective cloth, indoor and outdoor environmental air during and after production in the crude sesame oil extraction facilities.

Sample	Aerobic bacteria	Total coliforms	Yeasts and moulds	Aspergillus species	Staphylococcus aureus
Sesame seed ($\log_{10}$ CFU/g)	-	-	$2.31^{cd} \pm 0.13$	$1.17^{b} \pm 0.13$	-
Crude sesame oil ($\log_{10}$ CFU/mg/ml)	$2.44^{cd} \pm 0.07$	$2.39^{d} \pm 0.14$	$2.31^{cd} \pm 0.08$	$1.33^{b} \pm 0.01$	$2.09^{dc} \pm 0.12$
Municipal water ($\log_{10}$ CFU/mg/ml)	$1.58^{f} \pm 0.44$	$1.46^{f} \pm 0.28$	-	-	$0.67^{g} \pm 0.49$
Ground Water ($\log_{10}$ CFU/mg/ml)	$2.76^{c} \pm 0.23$	$3.41^{c} \pm 0.14$	-	-	$0.83^{gf} \pm 0.31$
Animal manure ($\log_{10}$ CFU/mg/ml)	-	$5.81^{a} \pm 0.48$	-	-	$1.24^{gef} \pm 0.28$
Surface ((log CFU/m^2)					
Milling and pressing machine	$4.55^{ab} \pm 0.12$	$4.54^{b} \pm 0.12$		-	$2.87^{b} \pm 0.19$
Filtration machine	$5.06^{a} \pm 0.09$	$4.52^{b} \pm 0.13$		-	$2.56^{bc} \pm 0.31$
Filling machine	$4.34^{b} \pm 0.15$	$3.99^{bc} \pm 0.05$	$3.73^{b} \pm 0.09$	-	-
Floor of sesame store	$4.83^{ab} \pm 0.13$	$4.47^{b} \pm 0.20$	$4.47^{a} \pm 0.07$	$3.50^{a} \pm 0.09$	$3.22^{ab} \pm 0.05$
Wall of the extraction plant	$4.62^{ab} \pm 0.18$	$4.28^{b} \pm 0.11$	$4.27^{ab} \pm 0.20$	$3.37^{a} \pm 0.16$	-
Protective clothing					
Clove (log CFU/glove)	$2.11^{def} \pm 0.20$	$1.78^{def} \pm 0.22$	$1.18^{e} \pm 0.29$	-	$3.77^{a} \pm 0.36$
Gown (log CFU/gown)	$2.05^{def} \pm 0.10$	$2.23^{ed} \pm 0.20$	$3.01^{c} \pm 0.33$	-	$3.26^{ab} \pm 0.14$
Air (log CFU/hr)					
Indoor air during production	$2.55^{cd} \pm 0.19$	$1.63^{ef} \pm 0.28$	$2.06^{d} \pm 0.11$	-	$1.82^{de} \pm 0.04$
Indoor air after production	$2.17^{de} \pm 0.18$	$1.36^{f} \pm 0.39$	$1.73^{de} \pm 0.23$	-	$1.54^{def} \pm 0.09$
Outdoor air during production	$1.18^{ef} \pm 0.10$	$1.70^{ef} \pm 0.16$	$1.70^{de} \pm 0.57$	-	$1.35^{def} \pm 0.18$
Outdoor air after production	$1.79^{ef} \pm 0.08$	$1.73^{def} \pm 0.12$	$1.79^{de} \pm 0.08$	-	$0.74^{g} \pm 0.20$

All values indicates $\log_{10}$ colony forming unit ± standard deviation. Values with different superscript letters in each column are significantly different (P-value < 0.05).

CSO was 2.44 log_{10} CFU/ml oil lower than the acceptance limit of <6 log_{10} CFU/ml for foods without cooking [59]. The sources of APC contamination were the surface of equipment and premises (4.68 log_{10} CFU/m^2), water (2.16 log_{10} CFU/ml), Indoor and outdoor air (2.08 log_{10} CFU/hr), and personal protective clothes (2.08 log_{10} CFU/clothes) with significant variability. Groundwater was immensely contaminated than pipe water and indoor air during production significantly with higher counts. The prevalence of APC explains unhygienic handling, inefficient and sporadic cleaning as well as environmental factors such as warm climatic conditions, dry and dusty environments.

Coliforms are non-spore-forming and Enterobacteriaceae group of gram-negative road-shaped microbial flora capable of lactose fermentation and survive in soil, water, human and animal intestine during aerobic and facultative anaerobic conditions at nearly body temperature. Coliforms are indicator pathogenic microorganisms originating from the gastrointestinal tract with a potential to cause food and waterborne illness as a result of poor environmental sanitation of food establishment, food handlers, and improper food processing and handling conditions [60] [61]. Total Coliforms identified in CSO were as large as 2.39 log_{10} CFU/ml oil lower than the acceptance limit of <4 log_{10} CFU/ml applied to processed foods. However, coliforms detected in animal manure (5.81 log_{10} CFU/ml), the surface of equipment, and establishment premise (4.36 log_{10} CFU/m^2) were above the limit explains the inefficiency of hygienic practices in the premises and equipment. Coliforms in water (2.43 log_{10} CFU/ml), protective clothing (2.0 log_{10} CFU/m^2) and environmental air (1.6 log CFU/m^3) below the limit. The prevalence of coliforms in the product, protective clothing, surface, and environment confirms that poor hygienic practices compromise CSO safety and quality. Nevertheless, *E. coli* was not detected in CSO.

Yeasts/moulds a microbial flora has grown and survived in a wide range of environmental conditions such as humidity, water activity, temperature, time, acidity, and air [62] [63] and in wider agricultural produce, food, food processing and storage establishments [64]. Fungi are categorized as beneficial and hazardous microbial flora with diverse physical features [65]. However, hazardous yeasts/moulds to humans and animals are gaining attention due to their diverse pathogenic character and toxic metabolites [66] [67], resistances during handling, processing, and cooking [68]. Fungi were detected in sesame seed, sesame oil, and associated surfaces, environment and protective clothing. Surfaces of extraction equipment and premises were found convenient environments for yeasts/moulds growth and survival (4.15 log_{10} CFU/m^2) whereas the filling machine was detected a lower colony count. The average fungi identified in the indoor and outdoor environmental air during and after production was also as high as 1.7 - 2.06 log_{10} CFU/hr with significant variability. Yeasts and moulds detected were variable in size, shape, texture, and colour explains diversified yeasts and moulds species were prevalent due to convenient environmental temperature (22˚C - 27˚C), humid environment, and poor hygienic practices. Yeasts and mould colonies with powdery and fluffy white, fluffy black, deep brown, green-black, mountain-like rough yellow colonies of different sizes and structures were detected. In the meantime, yeasts/moulds pathogenicity was confirmed as the *Aspergillus* species were detected in a Sabouraud Dextrose Agar with chloramphenicol powder added as a bacterial growth inhibitor at 25˚C during 5 - 7 days of incubation. The prevalence of *Aspergillus* species was confirmed (3.45 log_{10} CFU/m^2) on the selected surface of the storage and extraction facilities and 1.25 log_{10} CFU/ml in sesame seed and CSO with insignificant variability.

Staphylococcus aureus is a small (0.5 - 1.5 μm) spherical gram-positive facultative anaerobic multi-resistant bacteria of Micrococcaceae family staphylococcus genera. *Staphylococcus aureus* is catalase-positive, and thermostable [69] and a potential animal and human pathogen associated with poor hygienic human, animal, and environmental conditions. *Staphylococcal* poisoning is known worldwide as it produces *Staphylococcal* enterotoxin when ingested the strain/its toxins in food or in contact with surfaces contaminated with *Staphylococcus aureus*. *Staphylococcus aureus*, a potential infectious pathogenic bacteria were detected in CSO as large numbers as 2.09 log_{10} CFU/ml few hours after production which is below the acceptable limit (<4 log_{10} CFU/ml). However, *Staphylococcus aureus* growth might be continued during CSO storage. It was also detected at the surface of the equipment and different sites in the CSO establishment, protective clothing. This was due to unhygienic and inappropriate handling practices. Studies have reported *Staphylococcus aureus* to survive in a wide range of surfaces including plastics and fabrics [70], household and marketing equipment, glass, pipes, and metallic containers [71] [72], bottled and unblotted water of different sources [73], animal and animal by-products [74] and food [75], bio-aerosol [76], public areas and hospitals [9]. It survives in a wide range of conditions such as relative humidity, high gradient temperature, salinity, diverse environment, time and light exposure [70] [73] due to unhygienic practices and inappropriate food-handling [77].

4. Conclusion

Sesame value chain involves sesame agriculture, processors and consumers. However, value addition and local processing skills ineffectiveness experiencing post-harvest and quality loss. The sesame oil extraction establishment was suitable. However, the design of premises and hygienic practices were insufficient. And yet, quality degradation predominantly discoloration, off-flavour, and storage instability of sesame oil is a challenge. Both spoilage and pathogenic microorganisms were detected in CSO, sesame seed, water, and related surfaces in the establishment. Aerobic bacteria, total Coliforms, and yeasts and moulds were among the spoilage microbes, and *Aspergillus species* and *staphylococcus aureus* were potential pathogens detected in CSO, supplies, and establishment environment while *E. coli*, salmonella and Shigella were not detected. The causes for the prevalence of spoilage and pathogenic microorganisms were primarily poor hygienic practices and the inappropriate handling such as high moisture content of the seed and CSO. In the meantime, the use of raw CSO should be avoided unless cooked optimally. Further investigation is critical to minimize health burden, promote appropriate handling and hygienic practices, and use organic anti-microbial preservatives.

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Conflicts of Interest

The authors declare there is no conflict of interest.

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