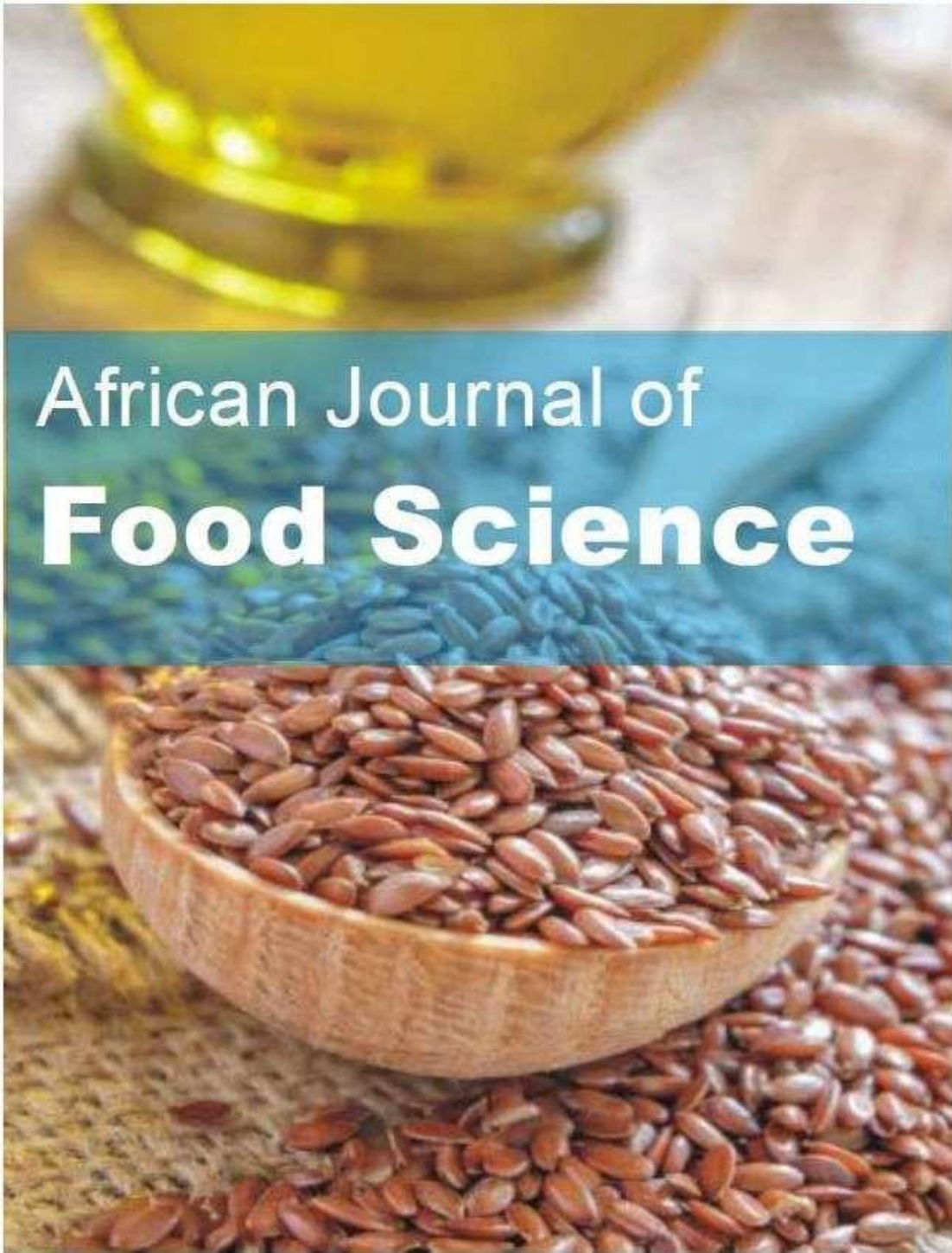


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Full Length Research Paper

Fatty acid and amino acid profiling of chia seeds and physicochemical characterisation of chia seeds oil

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The study analysed the fatty and amino acid compositions of chia seeds and the physicochemical characterisation of chia seeds oil. The quantification of amino and fatty acids was done using high-performance liquid chromatography (HPLC) and gas chromatography equipped with a flame ionisation detector (GC-FID). The amino acids from chia seeds were leucine, valine, phenylalanine, lysine, isoleucine, threonine, histidine, and methionine with 1.63, 1.37, 1.34, 1.14, 0.98, 0.98, 0.88 and 0.81 g/100 g, respectively. Saturated and unsaturated fatty acids were 3.95 and 30.48%, respectively. Meanwhile, the mean ratio of Omega 6/Omega 3 fatty acids was 0.333. The established physicochemical characteristics of CSO were acid value 2.63 mg KOH/g, peroxide value 6.23 meq active oxygen/kg, iodine value 200.721 g I₂/100 g, saponification value 193.345 mg KOH/g, refractive index 1.454 at 40°C relative density 0.8824 g/cm³ at 20°C and specific gravity 0.882 at 40°C. The fatty acid and amino acid profiling of chia seeds revealed the presence of an appreciable amount of polyunsaturated fatty acids (PUFA) and essential amino acids. The physicochemical characteristics of CSO constitute a drying oil category that requires timely usage and proper storage condition to reduce rancidity from air or light exposure.

Key words: Chia seeds, chia seeds oil, amino acids, fatty acids, physicochemical characteristics.

INTRODUCTION

The human body needs protein as one of the macronutrients to help the body to function correctly. Proteins play many crucial roles in the human body, which include building and repairing body tissues, coordination of other body functions, and many others.

The building blocks of proteins are amino acids (AA). There are 22 different AA, with 9 being essential AA. Essential amino acids need ingestion since they cannot be synthesised by the body (Tessari et al., 2016).

Chia seeds have been introduced in East Africa

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recently and have been gaining popularity due to their nutrient composition, mainly the AA and fatty acids (FA). Chia seeds contain fat (30 to 33%) and proteins (15 to 25%) (Ixtaina et al., 2008). More than 60% of its fat contains polyunsaturated fatty acids (PUFA), mainly Omega 3 and 6 fatty acids (Di Marco et al., 2020; Ghafoor et al., 2020; Grancieri et al., 2019; Rajaram, 2014; Segura-Campos et al., 2014; Shen et al., 2018) are vital to human health throughout one's lifespan (Gazem et al., 2017; Wu et al., 2016).

Various researchers have reported various oil content of chia seeds (Suri et al., 2016), Brazilian chia seeds with 31.2 g/100 g, and Argentina, Bolivia, Colombia and Peru with an average of 28.5 to 32.7 g/100 g (Shen et al., 2018). This variation may affect the physicochemical properties of chia seeds oil (Suri et al., 2016; Shen et al., 2018). Also, chia seeds contain 18 amino acids a human body needs and all 9 essential amino acids (Suri et al., 2016; Ullah et al., 2016). Ayerza and Coates (2004), Ixtaina et al. (2011), and Suri et al. (2016) reported that the protein and fat content of chia seeds varies from one place to another in the world due to differences in climatic conditions, soil and agronomic activities. Due to the nutritional potential of chia seeds, the incorporation of chia seeds in food products has gained popularity in dairy (Kibui et al., 2018) and bakery products (Romankiewicz et al., 2017). In all these applications, incorporating chia seeds has contributed to improved texture and nutritional composition in terms of protein content and fatty acids (Segura-Campos et al., 2013) in addition to extending the shelf life (Valdivia-López and Tecante, 2015) of the final product. Protein from chia seeds tends to form plastic films, which have low permeability to oxygen, which prevents food products from undergoing deterioration due to the presence of oxygen and reduces or inhibits the rate of colour change, flavour change and nutrient deterioration (Valdivia-López and Tecante, 2015).

To detect amino acids effectively by high-performance liquid chromatography (HPLC), AAs should be chemically modified into derivatives that absorb or fluoresce ultraviolet light using HPLC (Calull et al., 1991). Reagents commonly used for derivatisation to improve the analytical capability of AA are phenylisothiocyanate (PITC) to obtain PTC-amino acids and ortho-phthalaldehyde (OPA) to derive OPA-amino acids (Calull et al., 1991; Checa-Moreno et al., 2008). However, many other reagents are used for the derivatisation of amino acid analysis but are associated with different shortcomings (Calull et al., 1991; Checa-Moreno et al., 2008; González-Castro et al., 1997). The OPA method has greater sensitivity but is limited to primary amino acids (Checa-Moreno et al., 2008). The PITC is reported to react with all essential amino acids, and the process of derivatisation is simple, rapid and used for precolumn derivatisation (Calull et al., 1991; Checa-Moreno et al., 2008; González-Castro et al., 1997). For fatty acid profiling, gas chromatography equipped with a flame

ionisation detector (GC-FID) has commonly been used by researchers (Nitrayová et al., 2014; Uzunova et al., 2019). This study, therefore, analysed the fatty and amino acids compositions and identified the physicochemical characteristics of chia seeds oil from chia seeds grown in Kagera, Tanzania.

MATERIALS AND METHODS

Chia seeds sample

Chia seeds (*Salvia hispanica* L.) were collected from the Karagwe district in the Kagera region on the western shores of Lake Victoria in Tanzania. Freshly-harvested chia seeds of about 10 kg were cleaned by winnowing to remove debris. Cleaned seeds were wrapped in polyethylene bags and immediately placed in dark storage at a temperature ranging from 25 to 30°C before analysis. Before analysis, raw chia seeds were ground using a laboratory-scale grinding machine (Waring Commercial, made in the USA) and sieved to obtain a uniform particle size of 0.2 mm.

Amino acid (AA) profiling

Free amino acids were determined using phenylisothiocyanate as a derivatising agent using high-performance liquid chromatography with Ultraviolet Detection (Klikarová et al., 2021). The analysis was conducted using laboratory facilities of the Department of Food Science and Technology at the University of Dar es Salaam, the Tanzania Bureau of Standards (TBS) for preliminary analysis, and SGS Tanzania Superintendence Company Limited in Dar es Salaam.

Standard and reagents

Methanol (HPLC grade), phenylisothiocyanate (PITC), trimethylamine (TEA), acetic acid, tetrahydrofuran (HPLC grade), acetonitrile (HPLC grade), sodium hydroxide (anhydrous), sodium acetate and amino acid standards for protein hydrolysate were bought from Sigma Co. (St. Louis, MO, USA). The study used ultra-purified water produced in the laboratory with a pore size of 0.068 µS/cm (Evoqua Water Technologies, made in the USA).

Preparation of standard

A stock solution of AA was prepared from a standard solution containing 0.5 µmol/mL except for L-cystine at 0.25 µmol/mL in 0.2 N sodium citrate. Five-point calibration points of AA were prepared in a range of 0.0238 to 0.2143 µmol/mL, except L-cystine, which was 0.0119 to 0.10715 µmol/mL. The calibration curve was used to calculate AA concentration in chia seeds.

Sample preparation

Ten grams of chia seeds flour were inserted into a 250 mL beaker, followed by 40 mL of extracting solvent (75% methanol in ultra-purified water). The mixture was homogenised for 2 min using a homogeniser machine (Ultra Turrax, IKA T 18, made in Germany). The extract was then transferred to a 100 mL volumetric flask, and the beaker was rinsed four times with 15 mL of extracting solvent.

Table 1. Gradient programme applied for the separation of PTC- Amino Acids.

Time (min)	Flow rate (mL/min)	Mobile phase A (%) (Aqueous buffer)	Mobile phase B (%) (Acetonitrile-water)
0	1.0	90	10
8	1.0	70	30
13	1.0	52	48
15	1.0	0	100
23	1.0	0	100
24	1.0	90	10

Source: González-Castro et al., 1997

Extracting solvents were added to a glass bottle to make a standard volume of 100 mL stored at 4°C for 60 min (Antoine et al., 2001). The content was transferred to centrifuge tubes and centrifuged at 27,000 \times g (4,000 rpm) for 60 min (Rotofix 32A, Germany). The supernatant was filtered using filter paper (Whatman, 90 mm diameter, GE Healthcare UK Limited). Filtered sample extract solutions were stored in a fridge at 4°C. Before HPLC injection, an aliquot of this solution was filtered through 0.45 μ m pore size (HPLC grade, Nagel Germany made) into 1.5 mL HPLC vials. The spiking method helped to determine the accuracy of the method. Meanwhile, for the recovery study, the sample was fortified with the amino acid standard to attain a concentration of 0.1 μ mol/mL for all AA except L-cystine, which was 0.05 μ mol/mL.

Mobile phase preparation

The study used the method of González-Castro et al. (1997) with modifications. Mobile phase A was an aqueous buffer prepared by adding 0.5 mL TEA to 0.14 M of sodium acetate, and the pH of this mixture was adjusted to 6.2 with acetic acid. Mobile phase B comprised acetonitrile and ultra-purified water (60:40). The mobile phases were degassed (3210 BRANSON, Nagtech, UK) for 10 min to remove any air bubbles.

Amino acid derivatisation procedure

Amino acid derivatisation outlined by González-Castro et al. (1997) was followed with modifications. 100 μ L of amino acid standard solution or sample extract were pipetted into 2 mL vials (Agilent Technologies), followed by the addition of 400 μ L of derivatising reagent (methanol: TEA: PITC: water at 7:1:1:1). The mixture of the sample, or amino acid standard solution and derivatising reagent was blended using a vortex machine (Thermo scientific, assembled in China) for 3 s. Finally, the mixture was injected into the HPLC system to detect and quantify amino acids.

Chromatographic procedure

The study used the method of González-Castro et al. (1997) with modifications. HPLC (SHIMADZU Nexera X2, Japan) was used for analysing amino acid profiling. HPLC instrument was equipped with a pump (LC – 30AD), a membrane degasser (DGPU – 20A_{3R}), an autosampler (SIL -30AC), a diode array detector (SPD-M30A) and a column oven (CTO-20AC). Agilent Zorbax Eclipse plus C₁₈ 100A (4.6 mm \times 150 mm \times 5 μ m) was used to separate AA. The analysis was carried out at 30°C, a flow rate of 1.0 mL/min, an injection volume of 25 μ L, and a wavelength of 254 nm. Lab Solution software version 5.86 (200 -2016 Shimadzu, Corporation) was used

for data acquisition, processing and quantification. The gradient programme applied for separating PTC-amino acid is shown in Table 1. After separating the amino acid chromatogram (peaks), integration and quantification were carried out using the standard curve of peak areas obtained from known concentrations of the amino acid standard mixtures.

Fatty acid profiling analysis

Fatty acids profiling was conducted using gas chromatography equipped with a flame ionisation detector (GC-FID) as different researchers (Nitrayová et al., 2014; Uzunova et al., 2019) have commonly applied. The analysis was conducted using the Tanzania Bureau of Standards (TBS) laboratory facilities for preliminary analysis and SGS Tanzania Superintendence Company Limited, based in Dar es Salaam.

Standards and reagents

Pyrogallol acid (AR grade), ethanol (AR grade), hydrochloric acid (AR grade), diethyl ether (AR grade), petroleum ether (30 - 60°C, HPLC grade), toluene, n-hexane (AR grade), triglyceride internal standard solution (C_{11:0}) mixture of 37 fatty acid methyl ester standard (FAMES) and 9 single standard methyl esters of C16:0, C16:1, C17:1, C18:0, C18:1tran, C18:2cis, C18:3(GLA), C18:3(ALA) and C20:0 (Sigma Co., St. Louis - MO, USA). Chloroform (AR grade), boron trifluoride reagent (14% BF₃ in MeOH) and anhydrous sodium sulphate (Na₂SO₄ - AR grade) are manufactured by LOBA Chemie PVT LTD, India. The response factor, the ratio of peak area to the concentration of known standard, was used to calculate the concentration of FA in chia seeds.

Acid hydrolysis

Chia seed flour was hydrolysed with methanolic sulphuric acid (1 M) before profiling the fatty acids present to facilitate the extraction of fats. Sample preparation and acid hydrolysis were conducted following ISO (2017) with some modifications. About 0.5 g of chia seeds flour was weighed using Laboratory analytical balance (Explorer Pro, Switzerland). The weighed sample was placed in a volumetric flask, followed by the addition of 100 mg of pyrogallol acid, 2 mL of 5 g/L triglycerides internal standard solution (C_{11:0}), 2 mL of ethanol, 3 mL of ultra-purified water and 7 mL of hydrochloric acid (37%) and mixed thoroughly. The flasks were kept in a water trough at 75 to 80°C for 40 min and shaken every 10 min. The mixture was cooled at room temperature, and 10 mL of ethanol was added and shaken gently to facilitate fat extraction.

Fat extraction

About 25 mL of diethyl ether was added to the volumetric flask with mixtures from the acid hydrolysis stage and shaken for 5 min. The process was repeated by adding 25 mL of petroleum ether and left to stand until the upper layer was transparent. The upper liquid was then poured into a 250 mL flat bottom flask. The remaining bottom layer re-extraction of fat was repeated for two more times, until there was no more upper liquid separated after adding petroleum ether. All the portion of extract from the upper layer was put in the rotary evaporator to remove petroleum ether.

Methylation to FAMES

Three millilitres of chloroform and 3 mL of diethyl ether were added to the extracted fat residues and placed in a 15 mL reaction tube. The mixture evaporated to dryness at 40°C in a sample concentrator (Techne™ FSC4NCS, Fisher Scientific UK) with a nitrogen purge system. After that, 2 mL of 14% BF₃ in MeOH and 1 mL of toluene were added and closed with a lid and vortex for 1 min. The blend was heated for 45 min at 100°C and shaken every 10 min. The tube was then left to cool down to room temperature, and 5 mL of ultra-purified water, 1 mL of hexane and 1 g of anhydrous sodium sulphate were added. Again, the tube vortex for 1 min until layers separated and the top layer transferred to another vial containing 1 g of anhydrous sodium sulphate. Also, the top layer was transferred to another vial in readiness for analysis in GC-FID. In addition to the oil sample, a sample with a known concentration (BIPEA 20 - 408) of FA was run in parallel with the sample to determine the method's accuracy.

GC-FID setup and quantification

Gas chromatography equipped with flame ionisation detector (GC-FID) Agilent Technologies (Awuchi et al., 2019) equipped with Intuvo 9000 GC system was used to analyse the fatty acid profiling. The GC-FID instrument was equipped with a GC column (diameter 0.25 mm, film thickness 0.2 µm of 100 m length). The analysis of fatty acids was carried out based on ISO (2015). The analysis was carried out at a flow rate of 1.4 mL/min, hydrogen gas flow at 40 mL/min, air flow of 400 mL/min, nitrogen gas at 40 mL/min and oven temperature programmed at 50°C, gradient 5°C/min to 130°C, gradient 25°C/min to 175°C, for 7 min, gradient 2°C/min to 215°C for 1 min, gradient 2°C/min to 230°C hold 10 min. The total run time was 35 min. The sequence of analysis started with the blank (hexane), standard, control sample, sample and standard, respectively.

Chia seed oil characterisation

Extraction of chia seeds oil (CSO)

Chia seeds oil was extracted using automated Soxhlet equipment (Model EV 16, Gerhardt Bonn, Germany) and petroleum ether as extraction solvent (Horwitz, 2010). About 20 g of chia seeds flour was weighed, placed in extraction thimbles, and set in the Soxhlet machine. A ratio of 1:10 of sample and extraction solvent was used. About 200 mL of petroleum ether was added, and the extraction process was left to continue for about 4 h. After the extraction, petroleum ether was recovered using a rotary evaporator, and the flask was placed in an oven at 105°C for 30 min to dry the fat.

Determination of chia seeds oil yield

Chia seeds oil yield was determined by considering the mass of the

extracted oil and the mass of the seeds flour used for oil extraction by the method described by Timilsena et al. (2017), as shown by the equation:

$$\text{Oil yield (\%)} = \frac{\text{Mass of extracted oil (g)}}{\text{Mass of seed flour (g)}} \times 100 \quad (1)$$

Physicochemical characterisation of chia seeds oil

The chia seeds oil physicochemical parameter of acid value (AV) was determined according to ISO (2009). The peroxide value (PV) was determined according to the method described by Animal (2007), the iodine value (IV) (Yuan et al., 2020), the saponification value (SV) (Gu et al., 2017), and the refractive index (RI) following Dumitru (2020) using Abbe refractometer at 40°C (RFM 860, Bellingham UK). The relative density (RD) and specific gravity (SG) were measured by density metre (Mettler Toledo, Switzerland).

Data analysis

All the analyses were carried out in triplicates for amino acids, fatty acids, and physicochemical characterisation of chia seeds oil. Descriptive statistics were done using Microsoft Excel (15.0.5285 of 2013, Microsoft Inc. CA, USA). In addition, analysis of variance was used to determine statistical differences between types of AA or FA analysed using R statistical software (4.0.3 of 2020). Turkey's honest significance test was applied to find the means that are significantly different from each. Subsequently, the results were expressed as mean ± standard error of the mean and p-values of less than 0.05 were significant.

RESULTS AND DISCUSSION

Quality control results for amino acids and fatty acids

The correlation coefficient between peak area and AA concentration ranged from 0.719 to 0.992. The values for leucine, phenylalanine and valine fall within the acceptable range, that is, greater than 0.998 (Bartolomeo and Maisano, 2006), as Table 2 illustrates. The method for extraction and quantification of AA was found accurate for leucine, lysine, threonine, aspartic acid and proline since their recoveries values were 90 to 110% (Reason, 2003). Poor recoveries of other amino acid result from the destruction of some amino acids (tryptophan and cysteine) during acid hydrolysis. In contrast, other amino acids can interfere with each other (threonine and glycine) (Bartolomeo and Maisano, 2006). Table 2 also shows recovery percentage (quality control) results for fatty acids.

Amino acid profiling

The amino acid composition of chia seeds (Table 3) showed the presence of 18 amino acids of different quantities, including essential amino acids such as leucine, valine, phenylalanine, isoleucine and threonine. In this study, chia seeds were found to have significantly

Table 2. Quality control results for amino and fatty acids.

AA	R ²	Recovery (%)	FA	Recovery (%)
Histidine	0.976	72.47	Eicosenoic acid	50
Isoleucine	0.959	-	Lauric acid	80
Leucine	0.998	85.87	Linolenic acid	100
Lysine	0.789	116.70	Myristic acid	95
Methionine	0.980	-	Linoleic acid	100
Phenylalanine	0.988	75.64	Palmitic acid	100
Threonine	0.965	98.27	Margaric acid	50
Valine	0.992	34.94	Stearic acid	97
Arginine	0.815	70.11	Arachidic acid	70
Cystine	0.719	55.73	Behenic acid	20
Tyrosine	0.978	66.66	Oleic acid	101
Alanine	0.866	-	Palmitoleic acid	110
Aspartic acid	0.968	95.49		
Glutamic acid	0.937	74.15		
Glutamine	0.714	50.70		
Glycine	0.885	-		
Proline	0.933	96.18		
Serine	0.973	59.05		

Source: Authors

high levels of glutamic acid (4.41 ± 0.023) and arginine (3.73 ± 0.009) in g/100 g. Similarly, other researchers reported comparable high levels of glutamic acid and arginine. For example, Kulczyński et al. (2019) reported values of 3.5 and 2.14, Dautant et al. (2007) reported values of 7.08 and 4.23, and Valdivia-López and Tecante (2015) reported values of 12.3 and 8.06 in g/100 g for glutamic acid and arginine, respectively.

All the values are expressed in mean \pm standard error, $n=3$. Different small-scale letters within amino acids indicate a statistical difference at a 5% significant level according to Tukey's HSD multiple ranks test, whereas different capital letters within average AA indicate a statistical difference according to the student t-test at a 5% significant level.

The essential amino acid, leucine 1.63 g/ 100 g, had the highest amount, followed by valine (1.37) and phenylalanine (1.34) in g/100 g. For non-essential amino acids, glutamic acid had the highest quantity (4.41), followed by arginine (3.73) and aspartic acid (2.05), all in g/100 g. The chromatograms of amino acid profiling are presented in Figure 1, indicating the peak area of each amino acid detected.

In general, chia seeds were found to have a high amount of non-essential amino acids; however, 43% (that is 1.14 g/100 g) of the total AA found in chia seeds was essential AA, as Table 3 illustrates. Chia seeds from Kenya (Kibui et al., 2018) reported lower values of essential amino acids: leucine 1.02, valine 0.62, phenylalanine 0.75, lysine 0.73, isoleucine 0.54, threonine 0.4, histidine 0.33, and methionine 0.52 in g/100 g than the one reported in the present study. Nitrayová et al.

(2014) reported chia seeds from Poland with 1.42, valine 0.79, phenylalanine 1.16, lysine 0.93, isoleucine 0.74, threonine 0.54, histidine 0.61 and methionine 0.67 in g/100 g, which are lower than the one reported in the present study but higher than what Kibui et al. (2018) reported. These variations might be attributable to different geographical conditions, agronomic practices occurring during the cultivation of chia seeds and seed maturation (Ayerza, 2019), and the application of different analytical techniques. However, all the results conclusively show that chia seeds contain various amino acids in varying quantities (Table 3). As Table 3 illustrates, these results indicate that the chia seeds contain 18 AAs out of 22 amino acids the human body needs as building blocks for proteins for different functions in the body (Kulczyński et al., 2019; Nitrayová et al., 2014). These amino acids play different roles and functions in the body, which include blood sugar regulations, body tissue growth, muscle growth, energy production, immunity functioning of the body and brain functioning (Kulczyński et al., 2019; Nitrayová et al., 2014).

Fatty acid profiling

Analysis of fatty acids profiling of chia seeds revealed a significantly high amount of unsaturated fatty acids, 30.48% attributable to linolenic and linoleic acid, as compared to saturated fatty acids of 3.95% (Table 4). The monounsaturated fat acids (MUFA) were palmitoleic and oleic, whereas polyunsaturated fatty acids (PUFA)

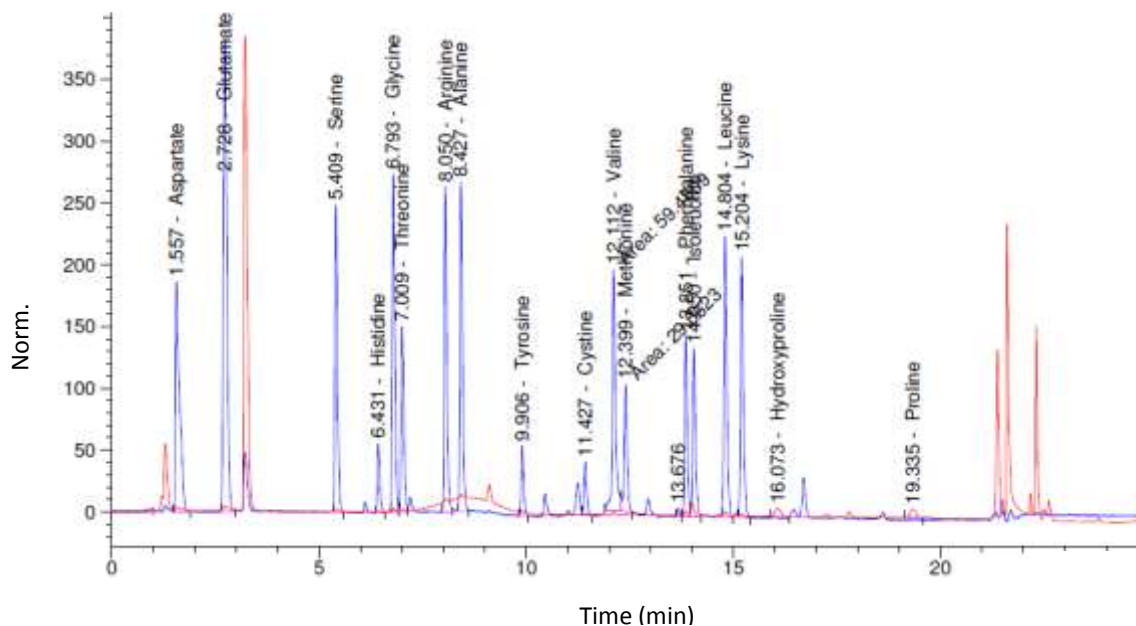


Figure 1. Chromatogram of amino acid profiling of chia seeds. The blue peaks are the amino acids identified from the chia seeds.

Source: Authors

Table 3. Amino acids' profile of chia seeds.

Amino acid	Abbreviation	Quantity (g/100 g) DM
Essential amino acid		
Histidine	His (H)	0.88±0.002 ^j
Isoleucine	Ile (I)	0.98 ±0.004 ⁱ
Leucine	Leu (L)	1.63±0.007 ^d
Lysine	Lys (K)	1.14±0.006 ^g
Methionine	Met (M)	0.81±0.001 ^k
Phenylalanine	Phe (F)	1.34±0.004 ^{ef}
Threonine	Thr (T)	0.98±0.005 ⁱ
Valine	Val (V)	1.37±0.003 ^e
Average		1.14±0.055^B
Non-essential amino acids		
Arginine	Arg (R)	3.73±0.009 ^b
Cystine	Cys (C)	0.43±0.004 ^m
Tyrosine	Tyr (Y)	0.59±0.009 ^l
Alanine	Ala (A)	0.94±0.003 ⁱ
Aspartic acid	Asp (D)	2.05±0.014 ^c
Glutamic acid	Glu (E)	4.41±0.023 ^a
Glutamine	Gln (Q)	0.00±0.001 ^m
Glycine	Gly (G)	1.08±0.009 ^h
Proline	Pro (P)	1.31±0.008 ^f
Serine	Ser (S)	1.35±0.009 ^{ef}
Hydroxyproline	Hyp (O)	0.94±0.006 ⁱ
Average		1.53±0.231^A

All the values are expressed in mean ± standard error n=3. Different small-scale letters within amino acids indicate a statistical difference at a 5% significant level according to Tukey's HSD multiple ranks test, whereas different capital letters within average AA indicate a statistical difference according to the student t-test at a 5% significant level.

Source: Authors

Table 4. Fatty acid profile of chia seeds.

Component	Chemical formula	Quantity (%) w/w
Saturated fats (SFA)		
Caprylic acid	C8:0	Less than 0.01
Capric acid	C10:0	Less than 0.01
Undecanoic acid	C11:0	Less than 0.01
Lauric acid	C12:0	Less than 0.01
Tridecanoic acid	C13:0	Less than 0.01
Myristic acid	C14:0	Less than 0.01
Myristoleic acid	C14:1	Less than 0.01
Pentadecanoic acid	C15:0	Less than 0.01
Pentadecanoic acid	C15:1	Less than 0.01
Palmitic acid	C16:0	2.41±0.012 ^d
Margaric acid	C17:0	0.05±0.001 ^g
Heptadecenoic acid	C17:1	0.05±0.001 ^g
Stearic acid	C18:0	1.31±0.011 ^e
Arachadic acid	C20:0	0.19±0.006 ^f
Arachidic acid	C20:4	Less than 0.01
Heneicosanoic acid	C21:0	Less than 0.01
Behenic acid	C22:0	0.05±0.001 ^g
Monounsaturated fats (MUFA)		
Palmitoleic acid	C16:1	0.09±0.001 ^{fg}
Oleic acid	C18:1	2.87±0.006 ^c
Gadoleic acid	C20:1	0.05±0.001 ^g
Polyunsaturated fats (PUFA)		
Linoleic acid	C18:2	6.82±0.021 ^b
Linolenic acid	C18:3	20.62±0.079 ^a
Eicosadienoic acid	C20:2	Less than 0.01
Eicosatrienoic acid	C20:3 n6	Less than 0.01
Eicosapentaenoic acid	C20:5 EPA n3	Less than 0.01
Summary of fat acids		
Omega 3 fatty acid		20.62±0.079
Omega 6 fatty acid		6.86±0.021
Saturated fatty acid		3.95±0.029
Unsaturated fatty acid		30.48±0.099
Ration n-6/n-3		0.333

All values are expressed in terms of mean ± standard error n=3. According to Tukey's HSD multiple ranks test, different small-scale letters within fat acids indicate a statistically significant difference at a 5% level.

Source: Authors

were linoleic and linolenic acids, as Table 4 illustrates. The fatty acid profile composition of chia seeds is presented in Table 4. The detection limit for the fatty acids was 0.01%. The saturated fatty acids recorded (Table 4) were palmitic, margaric, heptadecenoic, stearic, arachidic, and behenic. Mono-unsaturated acids were palmitoleic, oleic, and gadoleic acids. On the other hand, polyunsaturated fatty acids were linoleic and linolenic acids. The saturated fatty acids, palmitic acid (2.41%) and stearic acid (1.31%) were predominantly present. The ratio of Omega

6/Omega 3 fatty acid was 0.333, comparable with other research studies of chia seeds from Argentina/Guatemala, Mexico and Australia, respectively (Ixtaina et al., 2011; Segura-Campos et al., 2014; Timilsena et al., 2017). The nutritious richness of chia seeds is popular due to the high amount of polyunsaturated fatty acids, particularly the Omega 3 and 6 fatty acids. In recent years, people have suffered from non-commutable diseases such as diabetes, cardiovascular diseases, and various cancers, which are highly associated with eating

Table 5. Physicochemical properties of chia seeds oil (CSO).

Country	AV (mg KOH/g)	PV (meq active oxygen/kg)	IV (g I ₂ /100 g)	SV (mg KOH/g)	RI at 40°C	RD at 20°C (g/cm ³)	SG at 20°C	Source
Tanzania	2.63±0.044	6.23±0.205	200.72±1.192	193.35±1.463	1.454±0.0008	0.8824±0.0001	0.882±0.0001	This study
Australian	2.54±0.02	4.33±0.03	204 ±4	197 ±4	1.48±0.00	-	0.93±0.03	Timilsena et al. (2017)
Peru	1.68	1.95	208.3	197.9	1.473	0.9288	-	Uzunova et al. (2019)
Mexico	2.053	17.5	222.66	193.45	1.4684	0.9241	-	Segura-Campos et al. (2014)
Argentina	2.05	-	210.5	193.09	1.4710	-	-	Ixtaina et al. (2011)
Guatemala	1.64	-	215.0	193.01	1.4705	-	-	Ixtaina et al. (2011)

AV, acid value; PV, peroxide value; IV, iodine value; SV, saponification value; RI, refractive index; RD, relative density; SG, specific gravity.
Source: Authors

processed foods that are mostly less nutritious and have high carbohydrate and fat content. These foods mostly contain saturated fats and less polyunsaturated fatty acids, which have biologically active functions for human health (Swanson et al., 2012). Omega 3 and 6 are linked with human health benefits, which during pregnancy are associated with proper foetal development and reduction of cardiovascular diseases (Swanson et al., 2012). Emerging knowledge of chia seeds, especially their composition, including fatty acids, has opened the possibility of improving health using vegetarian sources through which chia seeds are cultivated. Doing so would reduce the burden of over-dependence on sea foods for sourcing Omega 3 and 6, particularly because sea foods are increasingly becoming scarce. Still, they are also associated with risks of environmental degradation in the sea (Kaale and Eikevik, 2014). In developing countries such as Tanzania and most sub-Saharan African nations, malnutrition is still one of the causative factors of child stunting. This causation might be associated with low income resulting in poor health. As chia seeds are a cultivated crop rich in polyunsaturated fatty acids, the crop could be introduced to farmers and used

at different levels in the household to combat this problem. Omega 3 and 6 fatty acids reportedly contribute positively to one's lifespan in terms of brain functioning and cognition (Rahmawaty and Meyer, 2020).

Chia seeds oil physicochemical characterisation

In this study, chia seeds oil yield was 27.4±0.128%. Researchers reported chia seeds oil (CSO) yield varies between 26 and 36% (Segura-Campos et al., 2014; Timilsena et al., 2017). The variation in oil yield may be attributable to agronomic and geographical conditions (Ayerza, 2019) and the method and extraction solvent applied (Ixtaina et al., 2011). Table 5 shows the physicochemical characteristics of chia seeds oil obtained using solvent extraction from different geographical locations, including results from this study.

The abbreviation of acid value (AV) is expressed in mg KOH/g, peroxide value (PV) is expressed in meq active oxygen/kg, iodine value (IV) is expressed in g of I₂/100 g, saponification value (SV) is expressed in mg KOH/g, refractive index

(RI) measured at 40°C, relative density (RD) measured at 20°C in g/cm³, and specific gravity measured at 20°C.

In this study, the oil was characterised based on its physicochemical properties. Different parameters were established as follows: the acid value of 2.63 mg KOH/g, peroxide value of 6.23 meq active oxygen/kg, iodine value of 200.72 g I₂/100 g, the refractive index of 1.454 at 40°C, the relative density of 0.8824 g/cm³ at 20°C and specific gravity of 0.882 at 20°C, as detailed in Table 5.

The chia seeds used in this analysis were dark with some slight whitish seeds and CSO characterised by golden colour. The colour of the seeds mostly depends on the level of maturity of the seeds, from dark seeds to black/black spotted. In all chia seeds, a few white seeds were mixed with dark/black spotted ones, sometimes not fully matured (white seeds). The oil extracted from grounded chia seeds flour was golden yellow (Figure 1).

The physicochemical characteristics commonly identify the oil type are saponification value (SV), iodine value, refractive index, and relative density. The saponification value refers to potassium hydroxide (KOH), which should be in milligrams to

convert 1 g of fat into soap (Ibeto et al., 2012). The SV of chia seeds oil of 193.345 mg KOH/g showed a high number of fatty acids. Other oils, such as shea-nut oil and jatropha oil, were reported to have SV of 195 and 193.55, respectively (Ibeto et al., 2012). Meanwhile, the SV for chia seeds ranged from 193 to 198 mg KOH/g. The high value of SV in CSO shows an increased number of fatty acids but also signals the possibility of using the oil for cosmetic purposes in producing soap-like products.

The AV of CSO measured was 2.63 ± 0.044 mg KOH/g. Any oil's AV represents the rancidity level as it usually occurs during the decomposition of triglycerides. The lower the AV, the better the oil quality. The acid value and PV commonly serve as the parameter to describe oil quality. The PV of CSO was 6.23 meq active oxygen/kg, which is within the acceptable limit of ≤ 10 meq active oxygen/kg for most edible oils and fats (Balley, 1982). Table 5 also shows the values of AV and PV of CSO that ranged from 1.6 to 2.6 mg KOH/g and 4.3 to 17.5 meq active oxygen/kg, indicating the CSO's easiness of undergoing rancidity. Implicitly, this suggests the possibility of CSO undergoing proper refining or protection from oxidation after extraction. Moreover, immediate usage should be recommended due to the instability of oil, which can be associated with shorter shelf life than other types of oils. Furthermore, the oil should be stored at a low temperature and dark to avoid oxidation and rancidity.

CSO's IV was 200.721 $I_2/100$ g. The IV of oil explains the oil's identity but also classifies the degree of unsaturation (Knothe, 2002). The IV of oils with a value < 125 is described as non-drying (e.g. olive), IV of 125 to 150 as semi-drying (e.g. sunflower oil), whereas IV of >150 are described as drying oils (Knothe, 2002). The CSO values (Table 5) ranged from 200 to 222 $I_2/100$ g, which signals that all the CSO from different locations is drying. On the other hand, non-drying oils are good oils which can be used in sauces and dressing as they will not harden when exposed to air (e.g., olive oil). On the other hand, drying oils have a high level of unsaturation which reveals the instability of oil.

This study established a refractive index of 1.454 at 40°C for CSO. The values of CSO established from different studies (Table 5) ranged from 1.45 to 1.48. The RI of oil is also used to show the rancidity levels of oil as it measures the passage of light rays traversing through the material. The RI is measured at a specific temperature. At every degree of temperature, the change of RI is at a rate of 0.000385 (Shahidi, 2005). The RI of 1.454 for CSO is still comparable for most common oils as the values range from 1.447 to 1.482 (Shahidi, 2005). This value of RI, PV, AV and IV of CSO has shown a high level of unsaturation of the oil. Due to the unsaturation level of fatty acids present in chia seeds oil, the same trend was noted for the RD value. The relative density of oil serves as an identification criterion of the oil type, but the higher RD value also shows an unsaturation level of fatty acids (Segura-Campos et al., 2014). This study's

relative density values and specific gravity of chia seeds oil were lower than previously reported values (Table 5). The oil's specific gravity (SG) is an essential factor when improving the oil's process and quality control (Timilsena et al., 2017). Both SG and RD also facilitate the identification of the type of oil.

Conclusion

Chia seeds' fatty acid and amino acid profiling revealed the presence of appreciable amounts of PUFA and essential amino acids, which offer significant health benefits to the human diet. The physicochemical characteristics of CSO showed it is a drying oil category necessary for timely usage and proper storage condition to reduce rancidity from exposure to either air or light. Chia seeds grown in Kagera, Tanzania, can thus be used to improve human health through different means of consumption. Therefore, the study recommends the scaled-up usage of chia seeds due to their amino and fatty acids richness.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Nutrient content of complementary foods for children in Kilimanjaro, Tanzania

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Complementary feeding is an effective strategy in reducing the levels of malnutrition among children aged 6-23 months. Little is known about the preparation and nutrient content of locally made complementary foods in Tanzania. This study was carried out with the aim of analysing nutrient content of the frequently used complementary foods for children of age 6 to 23 months. A cross-sectional study was conducted in three randomly selected villages in Rombo district, Kilimanjaro region, Tanzania. Information on the types of complementary foods was collected using semi-structured and 24-h dietary-recall questionnaires. Seven samples of frequently consumed complementary foods (banana, rice and maize-based porridges) were collected and analysed for proximate, vitamins and minerals composition. The results were as follows: Energy 317.98-379.23 kcal, vitamin A 81.32-971.05 µg RE, vitamin C 3.48-9.56 mg, iron 2.48-22.86 mg, zinc 0.92-9.57 mg, calcium 73.13-400.58 mg and iodine 10.18-200.93 µg/100 g dry-weights. Conclusively, the amount of vitamin C, iron, zinc, calcium and iodine of the frequently used complementary foods in the study area was low as compared to what is recommended by World Health Organization. It is important to develop recipes that may either fill or narrow this gap by using low-cost, locally available and culturally acceptable ingredients.

Key words: Complementary foods, nutrient content, energy, micronutrients, porridge.

INTRODUCTION

The first 1000 days of life, from conception until the child's second birthday, are considered the critical window of opportunity for preventing under nutrition and its long-term consequences (Hemalatha et al., 2018). Poor breastfeeding patterns, low nutrient density and poor quality of complementary foods account for nutrient deficiency, illness and infections in children, leading to

malnutrition at an early age (Singh et al., 2019). This in turn prevents children from reaching their full physical and mental potential later in life. This is due to delayed physical growth and motor development, low intellectual quotient (IQ), greater behavioural problems, deficient social skills as well as their increased susceptibility to contracting diseases (Kamath et al., 2017). The common

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nutritional problems among children aged 6-23 months in many countries include protein-energy malnutrition (PEM), vitamin A deficiency (VAD) as well as iodine deficiency disorders (IDD) and iron deficiency anaemia (IDA) (IFPRI, 2016).

Globally, an estimated 156 million (23.8%) children under-five years of age are stunted, 95 million (14%) are underweight, 50 million (7.5%) are wasted and 16 million (2.4%) are severely wasted (IFPRI, 2016; UNICEF/WB/WHO, 2016). In addition, over 160 million children worldwide are vitamin A deficient with a prevalence of about 30% in all developing countries and over 293 million (47.4%) of pre-school age children are anaemic (UNSCN, 2016; Yisak et al., 2020).

In Africa, 58.5 (37%), 13.9 (28%) and 10.3 (25%) million children under the age of five years are stunted, wasted and overweight, respectively (IFPRI, 2016). TDHS-MIS (2015-2016) and IFPRI (2016) reported that Tanzania ranks 105 out of 132 countries surveyed with prevalence of stunting (34.7%), wasting (3.8%), underweight (14%), severely underweight (3%) and anaemia prevalence of about 40%.

Complementary feeding refers to the introduction of other foods and liquids to a breastfed child when breast milk alone is no longer sufficient to meet the nutritional requirements of the infant (Fewtrell et al., 2017). It is an effective strategy in reducing the levels of malnutrition among children aged 6-23 months (Kassa et al., 2016; Mitchodigni et al., 2017). Breast milk alone can be used to properly feed infants for the first six months of life, but as infants grow and become more active, breast milk alone falls short of providing the full nutritional requirements and the gap keeps expanding with increasing age of the infants and young children; hence complementary feeding plays critical role in bridging these gaps (Abeshu, 2016). Major problems at this stage include poor timing of introducing complementary foods (too early or too late), poor food preparation and feeding practices, the use of complementary foods with low energy and nutrient density, low nutrients' bioavailability as well as poor processing methods and all of these are exacerbated by poverty and food insecurity (Arikpo et al., 2018; Oladiran and Emmambux, 2020).

Kilimanjaro region, just like other regions of Tanzania has malnourished children of which 20, 9.6 and 1.5% of children less than five years of age are stunted, underweight, and wasted, respectively (TFNC, 2019). About 48.9% of children under this age group are also anaemic while 34.2% are vitamin A deficient (TDHS-MIS, 2015-2016). One of the possible explanations for malnutrition could be inadequate nutrient intake from the commonly consumed foods.

In Rombo district, studies on the nutrient content of the frequently used complementary foods for children aged 6-23 months are very limited. The few available previous studies were on mycotoxins level in complementary foods

as well as prevalence and predictors of exclusive breastfeeding among breastfeeding women (Shirima et al., 2000; Mgongo et al., 2013). The specific objectives of the present study were to identify the frequently used complementary foods as well as their nutrient contents. The results will help other food and nutrition stakeholders in planning diet modification studies to improve nutrient content of commonly used complimentary foods.

MATERIALS AND METHODS

Study area

This study was conducted in Kilimanjaro region, Rombo district, Tanzania Mainland. Rombo district is one of the six districts in Kilimanjaro region. The district is bordered to the North and East by United Republic of Kenya, to the West by the Hai district and to the South by Moshi Rural district (URT, 2017). The district receives annual rainfall ranging from 500 to 1000 mm per annum and the mean monthly temperature is 22 to 26°C with maximum temperatures of 35°C. The main economic activity practiced in Rombo district is agriculture. This carries about 90% of the total activities while 7% of the residents are doing small businesses and 3% are the employed workers (URT, 2017). Food crops include banana, maize, sorghum, sweet potatoes, cassava and legumes, fruits and vegetables; whereas the main cash crop is coffee.

Study design and sample size

A cross sectional study was conducted in three randomly selected villages of Urauri, Kibaoni and Kikelelwa in November to December when the residents were planting crops. The study population comprised mothers/caregivers and their children who were 6 to 23 months old during data collection period. Children who were under special nutritional therapies and those with medical disorders or chronic health conditions were excluded from the study. The sample size was obtained from the general stunting percentage (18.3%) for children less than 5 years in Kilimanjaro region (TNNS, 2018). The formula used was adopted from SMART (2012) and a total of 230 respondents were involved in this study.

Data collection

Identification of the frequently used complementary foods was done by using semi-structured questionnaires using 24 h dietary recall. The information collected included social and demographic characteristics of the mother/caregiver and the child, types of complementary foods and how they were prepared.

Before administration of the questionnaires, five enumerators were enlightened on the main and specific objectives of the research and familiarized with data collection instruments. Pre-testing of the questionnaire was done at Mazimbu in Morogoro region before the beginning of data collection in a randomly selected sample of 10 individuals who were not included in the study but had similar characteristics to the study sample. After pre-testing the questionnaires, corrections were made to avoid misleading information, ambiguous sentences and repeated questions.

Before the beginning of the interview, the enumerators introduced themselves, explained the purpose of the study as well as the potential benefits and risks and then the respondents were

asked to voluntarily sign the consent form. For those respondents who were unable to read and write, they were helped by a closer relative, neighbour or the enumerator and give their oral consent. The questionnaire was administered through face to face interview. The children who were not living with their mothers were represented by their caregivers and most of them were their grandmothers. This is because some of the young mothers tend to leave their children with their grandmothers and go to big towns to work.

Collection of food samples and laboratory analysis

Seven samples of frequently used complementary foods which were collected using 24 h dietary recall questionnaire (Table 2) were taken to the laboratory for analysis of proximate composition and vitamin A, vitamin C, iron, zinc, calcium and iodine contents.

Before collection of cooked samples, there was a focus group discussion with ten women who came to clinic (RCH unit) at Tarakea Health Centre on preparation methods of the selected complementary foods. When the common procedures, ingredients, amounts, preparation and cooking methods were agreed, the ingredients such as meat, fish, milk, onions, tomatoes, rice, pumpkins and bananas were purchased at Tarakea market (it is the market that was used by most of the respondents). Complementary food samples were prepared by seven randomly selected mothers/caregivers (one for each recipe) at different households in the three villages. Mothers/Caregivers from Urauri prepared banana porridge with meat and with fish while those from Kibaoni village prepared banana porridge with pumpkin and with milk. Mothers/Caregivers from Kikelelwa village prepared maize, rice and composite flour porridges. This was done without the interference from the researcher. The task of the researcher was only to provide ingredients and record the procedures.

During preparation of complementary foods, the ingredients (name and amount used) were recorded first before cooking and then the mother/caregiver prepared and cooked the food while the researcher was observing and recording the procedures. When the food was ready, it was left to cool and then served in clean tight plastic food containers, weighed again and then stored in a cool box with iced water tightened in plastic bag then transported for analysis at Sokoine University of Agriculture laboratory on the next day. The remaining foods and ingredients were given to the mothers/caregivers who prepared the foods. CAMRY kitchen weighing scale (Model: EK3651, Max. 5000 g/11 lb, Japan) was used to weigh the ingredients and cooked samples.

Sample preparation

The cooked food samples were stored in the freezer for five days waiting for analysis at Sokoine University of Agriculture, Morogoro. During analysis, the food samples were thawed in running water and then mixed thoroughly (homogenization) while maintaining its representativeness (without changing its original composition and form). Nutrient composition analysis was done in duplicate for all seven samples and the results were presented in grams (g), milligrams (mg) or micrograms (μg) per hundred grams.

Laboratory analysis

The proximate composition of each of the frequently used complementary food samples were determined by using the standard methods of AOAC (2000) and the results were presented as an average of the duplicate determinations. Crude protein was

determined by Kjeldahl method (AOAC, 2000, official method 925.09), total fat by using Soxhlet system (HT model 1043-extraction unit AB, Sweden) following the procedures shown in AOAC (2000; official method 4.5.01.) while Ancom fibre analyser (Model ANCOM 220, USA) was used to determine crude fibre content as outlined by AOAC (2000) in official method 962.09. Moisture and ash contents were determined using oven drying (AOAC, 2005; method 925.09) and (AOAC, 2000; method 923.03), respectively for 5 h. The ash content of the samples was calculated as the difference between weight of the sample before and after incineration. Energy values of all the complementary food samples were determined by calculation using Atwater's conversion factors (FAO, 2003).

Vitamin A (Beta carotene) content was determined using UV-Visible Spectrophotometer following the procedures described by Rodriguez-Amaya and Kimura (2004). A conversion factor of 6 μg of β -carotene equivalent to 1 μg of Retinol Equivalent (RE) was used. Retinol was determined following the procedures described by Lietz et al. (2000), Rutkowski and Grzegorzczak (2007) and Kandar et al. (2012). Vitamin C determination was done following AOAC (2000) procedures using method No. 985.33 by titration. Iron, zinc, calcium and iodine contents were determined using the AOAC (2000) procedures, method No. 985.35 by using atomic absorption spectrophotometer.

Data analysis

Data was cleaned to adjust for inconsistent, conflicting and implausible responses and carefully subjected to the descriptive analyses using the computer Microsoft Office Excel 2007, Statistical Products and Service Solution software (SPSS) version 20.0. Means were calculated for continuous variables and for categorical variables frequencies and percentages were used. For laboratory results of the nutrient content of the frequently used complementary foods, each determination was carried out in duplicate and results were reported as an average value (mean \pm standard deviation (SD)). Turkey's Honest Significant Difference test was used for multiple mean comparison tests. Statistical significance was set at $p < 0.05$.

Ethical clearance

The study protocol was approved by the National Institute for Medical Research (NIMR/HQ/R.8a/Vol. IX/2362). Also, Sokoine University of Agriculture and Rombo District Executive Director provided us with the written permit to conduct research in the area. Written informed consent was obtained from all mothers/caregivers who took part in this study as well as the village leaders who issued a letter of acceptance for the research. All the participants were ensured of confidentiality and autonomy and that the information obtained will not be misused.

RESULTS

Social and demographic characteristics of the study participants

Table 1 shows the socio-demographic characteristics of the 230 mothers/caregivers from three villages, namely Kikelelwa (30.4%), Kibaoni (38.7%) and Urauri (30.9%) who participated in the survey. Majority of the children

Table 1. Socio-demographic characteristics of the study participants

Variable	Number	%
Age of children (months)		
6-8	60	26.1
9-11	52	22.5
12-23	118	51.4
Age of mothers (years)		
<20	31	13.5
20-35	143	62.2
> 36	56	24.3
Marital status		
Single	63	27.4
Married	167	72.6
Education level		
Informal	10	4.3
Primary school	147	63.9
Secondary school	67	29.2
Post-secondary school	6	2.6
Occupation		
Housewife	33	14.3
Agriculture	117	50.9
Employed formal	11	4.8
Employed informal	6	2.6
Self employed	63	27.4
Number of under five children per household		
1-2	216	93.9
3 or more	14	6.1

(51.4%) were aged between 12 and 23 months at the time of data collection. Most of the mothers/caregivers (95.65%) were able to at least read and write their names. The mean age was 27 years, 63.9% had completed primary school education and 50.9% were involved in agriculture. The average number of people per household was 5 (53.5%). Predominantly produced food crops were cereals, legumes and banana. About 39% of the respondents reported to keep poultry in their households.

Frequently used complementary foods

Table 2 shows the frequently used complementary foods. These included banana porridge with beef (53.5%, n=123), banana porridge with fish (30.9%, n=71), maize porridge (36.2%, n=84), banana porridge with milk (37.0%, n=85), composite flour porridge (57.8%, n=133),

banana porridge with pumpkins (2.61%, n=6) and rice porridge (13.5%, n=31). Composite flour porridge was the highly consumed complementary food (57.8%, n=133) while porridge with pumpkins was the least consumed food (2.61%, n=6).

Nutrient content of the frequently used complementary foods

Proximate composition and energy content

Proximate composition of the seven frequently used complementary foods (banana porridge with beef, fish, milk or pumpkins, composite flour porridge, maize porridge and rice porridge with milk) for children 6-23 months of age in Rombo district on wet basis are shown in Table 3.

Moisture content of the samples ranged from 65.51 to

Table 2. Frequently used complementary foods selected for laboratory analysis.

Name of the food sample	Swahili name (Local name)	N	%
Banana porridge with beef	<i>Mtori wa nyama</i>	123	53.5
Banana porridge with fish	<i>Mtori wa samaki</i>	71	30.9
Maize porridge (with sugar and cooking oil)	<i>Uji wa mahindi wenye sukari na mafuta (Uji)</i>	84	36.5
Banana porridge with milk	<i>Mtori wa maziwa (kitawa/ kena)</i>	85	37.0
Composite flour porridge	<i>Uji wa unga mchanganyiko / lishe</i>	133	57.8
Banana porridge with pumpkins	<i>Mtori wa maboga (mtori wa masidi)</i>	6	2.6
Rice porridge (with milk)	<i>Uji wa mchele/wali madida (Mshele)</i>	31	13.5

N=Number of children who consumed the food, %=percentage of children who consumed the food.

Table 3. Proximate composition of frequently used complementary foods (g/100 g dry weight).

Parameter	Banana porridge with beef	Banana porridge with fish	Maize porridge	Banana porridge with milk	Composite flour porridge	Banana porridge with pumpkin	Rice porridge with milk
Energy	317.98±16.49 ^b	348.14±10.55 ^{ab}	334.51±12.88 ^{ab}	373.44±22.34 ^a	379.23±7.51 ^a	333.29±5.31 ^{ab}	345.79±10.10 ^{ab}
Protein	9.74±2.48 ^c	8.34±2.57 ^c	13.47±1.07 ^c	8.33±1.48 ^c	13.84±1.58 ^{bc}	23.65±5.02 ^{ab}	25.12±1.27 ^a
Fat	3.59±1.08 ^c	2.71±0.25 ^c	6.55±1.93 ^{bc}	10.66±2.77 ^b	20.72±0.97 ^a	1.05±0.37 ^c	2.36±0.02 ^c
CHO (available)	61.67±0.76 ^{ab}	72.61±5.76 ^a	55.41±2.20 ^b	61.05±0.82 ^{ab}	34.34±2.48 ^c	57.31±4.51 ^b	56.01±3.79 ^b
Moisture	72.36±1.34 ^b	67.72±0.84 ^{bc}	81.66±1.43 ^a	65.51±1.32 ^c	79.72±0.58 ^a	71.44±2.78 ^b	68.67±1.14 ^{bc}
Ash	2.71±0.25 ^b	2.10±0.01 ^c	1.05±0.02 ^e	1.68±0.07 ^{cd}	3.54±0.19 ^a	1.47±0.05 ^{de}	1.89±0.003 ^{cd}
Fibre	22.28±3.04 ^{ab}	14.25±2.93 ^c	23.52±0.83 ^{ab}	18.28±2.18 ^{ab}	27.55±2.90 ^a	16.52±0.92 ^c	14.61±2.53 ^c
Dry matter	27.64±1.34 ^b	32.28±0.84 ^{ab}	18.34±1.43 ^c	34.49±1.32 ^a	20.28±0.58 ^c	28.56±2.78 ^b	31.33±1.14 ^{ab}

Values are means ± SD of duplicate determinations. Values with different superscripts in a row differ significantly (p<0.05).

81.66%. Banana porridge with milk had significantly lower moisture content than the rest of the formulations. The lower moisture content could be attributed to the addition of milk instead of plain water during stirring. Maize porridge had higher moisture content but it was not significantly (p>0.05) different from composite flour porridge. Banana porridge with beef, banana porridge with fish, banana porridge with pumpkins and rice porridge were not significantly different (p>0.05) in terms of moisture content.

The value of ash content for all the formulations ranged from 1.05 to 3.54%. Composite flour had significantly higher ash content (3.54) (p < 0.05). Milk based samples (banana porridge with milk and rice porridge with milk) had similar levels of ash content. Maize porridge had the lowest ash content followed closely by banana porridge with pumpkins and they were not significantly different from one another.

Banana porridge with fish, rice porridge with milk and banana porridge with pumpkins had lower total fibre content (14.25, 14.61 and 16.52%, respectively). The values of fibre content ranged from 14.25 to 27.55%. The highest fibre content was found in composite flour porridge followed by banana porridge with beef.

Protein content ranged from 8.33 to 25.12%. There was no significant difference in protein content among the samples. Porridge made from composite flour as well as banana porridge with milk had significantly higher fat content than other formulations (p<0.05). Fat content for all the samples ranged from 1.05 to 20.72 g/100 g (dry weight). Banana porridge with pumpkins had the lowest fat score followed by rice porridge with milk and banana porridge with fish.

Available carbohydrate ranged from 34.34 to 72.61%. Banana porridge with fish had relatively higher carbohydrate content compared to the rest of the samples. The lowest carbohydrate content was reported in composite flour porridge.

Energy content of the frequently used complementary foods ranged from 317.98 to 379.23 kcal per 100 g (dry weight). Composite flour and banana with milk porridges were characterised by the highest levels of energy as compared to the rest of the analysed complementary food samples. Banana porridge with beef had the lowest energy value (317.98 kcal). Energy content of banana porridge with fish, banana porridge with pumpkins and maize porridge were not significantly different (p > 0.05) from one another.

Table 4. Vitamins A and C composition of the frequently used complementary foods in Rombo district (g/100 g dry weight basis).

Complementary foods	B-carotene ($\mu\text{g}/100\text{ g RE}$)	Retinol ($\mu\text{g}/100\text{ g}$)	Total vitamin A ($\mu\text{g}/100\text{ g}$)	Vitamin C ($\text{mg}/100\text{ g}$)
Banana porridge with beef	170.13 \pm 11.45 ^{de}	582.62 \pm 49.22 ^a	752.75 \pm 60.67 ^{bc}	3.48 \pm 0.12 ^c
Banana porridge with fish	143.73 \pm 0.22 ^{de}	401.02 \pm 9.21 ^b	544.76 \pm 8.98 ^d	7.02 \pm 0.31 ^a
Maize porridge	195.83 \pm 16.77 ^{cd}	0.00 ^c	195.83 \pm 16.77 ^f	5.23 \pm 0.52 ^b
Banana porridge with milk	105.30 \pm 3.93 ^e	76.01 \pm 37.84 ^a	81.32 \pm 33.91 ^b	5.62 \pm 0.18 ^b
Composite flour porridge	971.05 \pm 10.75 ^a	0.00 ^c	971.05 \pm 10.75 ^a	9.56 \pm 0.23 ^b
Banana porridge with pumpkin	401.49 \pm 39.05 ^b	0.00 ^c	401.489 \pm 39.05 ^e	6.46 \pm 0.58 ^b
Rice porridge with milk	262.66 \pm 9.54 ^c	387.51 \pm 10.34 ^b	650.17 \pm 0.80 ^{cd}	6.05 \pm 0.36 ^a

Values are means \pm SD of duplicate determinations. Values with different superscripts in a column differ significantly ($p < 0.05$). The sample with 0.00 were from plant sources and therefore retinol was not analysed.

Table 5. Mineral composition of frequently used complementary foods for children aged 6-23 months at Rombo district (g/100 g dry weight).

Complementary foods	Iron (mg/100 g)	Zinc (mg/100 g)	Calcium (mg/100 g)	Iodine ($\mu\text{g}/100\text{ g}$)
Banana porridge with beef soup	22.86 \pm 1.09 ^a	9.57 \pm 0.85 ^a	400.58 \pm 40.22 ^a	10.18 \pm 4.23 ^d
Banana porridge with fish soup	5.99 \pm 0.17 ^d	1.17 \pm 0.02 ^{bc}	82.73 \pm 1.47 ^d	40.24 \pm 4.54 ^{cd}
Maize porridge with sugar and oil	9.12 \pm 1.24 ^b	2.53 \pm 0.15 ^b	149.75 \pm 5.11 ^{bc}	42.04 \pm 8.66 ^{cd}
Banana porridge with milk	4.88 \pm 0.39 ^{cd}	1.38 \pm 0.12 ^{bc}	194.56 \pm 6.19 ^b	56.86 \pm 9.46 ^{bc}
Composite flour porridge	9.21 \pm 0.53 ^b	1.05 \pm 0.12 ^c	111.55 \pm 6.84 ^{cd}	200.93 \pm 15.34 ^a
Banana porridge with pumpkin	2.58 \pm 0.40 ^d	0.92 \pm 0.05 ^c	135.15 \pm 11.37 ^{bcd}	31.73 \pm 10.52 ^{cd}
Rice porridge with milk	2.48 \pm 0.24 ^d	1.36 \pm 0.35 ^{bc}	73.13 \pm 3.77 ^d	85.12 \pm 6.52 ^b

Values are means \pm SD of duplicate determinations. Values with different superscripts in a column differ significantly ($p < 0.05$).

Vitamins A and C composition of the complementary foods

Table 4 shows values of vitamins A and C of the seven frequently used complementary food samples in Rombo district. Vitamin A content ranged from 81.32 to 971.05 $\mu\text{g}/100\text{ g}$ (dry weight). The lowest vitamin A content was observed in banana porridge with milk. Food samples with animal products such as meat, fish and milk had relatively higher vitamin A contents.

Vitamin C content ranged from 3.48 to 9.56 $\text{mg}/100\text{ g}$ dry weight, with the highest contents found in composite flour porridge and banana porridge with fish; and the lowest in banana porridge with beef. Milk based complementary food samples (banana porridge with milk and rice porridge with milk) as well as maize porridge and banana porridge with pumpkins had relatively similar levels of vitamin C.

Iron, zinc, calcium and iodine content of the complementary foods in Rombo

Iron content ranged from 2.48 to 22.86 $\text{g}/100\text{ g}$ of the dry

sample shown in Table 5. There was no significant difference in iron content between banana porridge with fish and banana porridge with milk. Banana porridge with beef had significantly ($p < 0.05$) higher iron content as compared to the rest of the sample. Banana porridge with pumpkins had lowest iron content.

With regard to zinc content, samples had zinc content below the minimum recommended levels for complementary foods with the exception of banana porridge with beef. It ranged from 0.92 to 9.57 $\text{mg}/100\text{ g}$ (dry weight). Banana porridge with beef had the highest zinc content (9.57) as compared to banana porridge with pumpkin which had less than 1 $\text{mg}/100\text{ g}$ (Table 5). Banana porridge with pumpkin had the lowest zinc content but it was not significantly different from composite flour porridge, banana porridge with fish and banana porridge with milk.

Calcium levels ranged from 73.13 to 400.58 mg shown in Table 5. Banana porridge with beef had significantly ($p < 0.05$) higher calcium content than the rest of the samples. The lowest calcium content was reported in rice porridge with milk (73.13 $\text{mg}/100\text{ g}$ dry weight).

Composite flour porridge had significantly ($p < 0.05$) higher iodine content (200.93 $\mu\text{g}/100\text{ g}$ dry weight) while

banana porridge with beef had the lowest score (10.18 µg/100 g dry weight). The rest of the sample had almost similar iodine levels.

DISCUSSION

This study aimed at assessing the nutrient content of frequently used complementary foods in Rombo district. Maize porridge had significantly higher moisture content when compared with the rest of the recipes. This means that a child needs to eat a lot of food in order to meet the nutrient needs because the food is more energy and micronutrient-dilute. The moisture content of the porridges was similar to the findings of Kulwa et al. (2015) but lower than that of Mandha et al. (2021). High moisture content in food products have also been associated with increased growth of microorganisms, which in turn causes spoilage and low nutritional qualities of the food products (Tapia et al., 2020).

The ash content reported in the present study was lower than what was reported by Tiencheu et al. (2016) but similar to that of Ezeokeke and Onuoha (2016). The lower values of ash content in the samples used in this study may probably indicate lower mineral contents. Composite flour porridge had higher ash content relative to other foods due to the presence of variety of ingredients such as maize, rice, soya beans, groundnuts and finger millet that may have more minerals (particularly iron, zinc and calcium) relative to other samples.

The findings from this study revealed that most of the complementary foods in Rombo district meet the recommended amount of protein needed from complementary foods. Even the foods that could not meet WHO recommendations (banana porridge with fish and banana porridge with milk); they were still able to provide at least 50% of the amount of protein needed from complementary foods (Abeshu et al., 2016). The higher protein content in rice porridge may be contributed by the addition of milk which is a good source of protein.

In order to meet the amount of energy, essential fatty acids and uptake of fat soluble vitamins by lipids, fat from complementary foods should provide approximately 30 to 45% of the total energy required by infants and young children based on their age and development stage (Khor and Lee, 2021). Fat content of the complementary food samples ranged from 1.05 to 20.72 g/100 g (dry weight). Composite flour porridge had significantly higher fat content relative to the rest of the complementary food samples ($p < 0.05$). The high fat content in composite flour may be due to the inclusion of oily seeds such as groundnuts and soya beans and whole grains as well as addition of vegetable oil during cooking. Also addition of whole milk in banana porridge may have contributed to the high fat content of the sample.

The carbohydrate (excluding fibre) contents of the

complementary food in this study were in the range of 34.34 to 72.61%. The carbohydrate values of maize and banana-based complementary foods are similar to what was reported by Ezeokeke and Onuoha (2016) as well as Okoye and Egbujie (2018). This could probably be due to the addition of other ingredients such as milk or preparation methods of the ingredients that may have reduced losses during peeling or de-hulling.

The recommended energy intake from complementary foods varies according to the age of child, amount of breast milk consumed, fat content in breast milk and the frequency of feeding (Feyisa et al., 2020). For children aged 6-8, 9-11 and 12-23 months and are breastfed, it is recommended that they take 202, 307 and 548 kcal per day, respectively (Abeshu et al., 2016). Studies conducted in Tanzania revealed that, most of the complementary foods are bulky but with lower energy and micronutrient concentrations (Ogbo et al., 2018). The findings from this study support this review because the energy content of all the samples of the frequently used complementary food were below the recommended amount of energy need from complementary foods. The energy content of the samples ranged from 317.6 to 379.23 kcal/100 g (dry weight) and banana porridge with beef had the lowest score. This means that, mothers should be advised to reduce water content (dilution) in complementary foods and increase frequency of feeding in order to meet the recommended dietary allowance.

In this study, total vitamin A content of all the samples was higher than the range reported by Isingoma et al. (2015), with the exception of maize porridge. Samples that contained animal products such as meat, milk and fish had higher vitamin A which is supported by Eggersdorfer and Wyss (2018). Maize porridge had the lowest vitamin A content which is in agreement with what was reported by Jemberu et al. (2016). The observed lower than recommended vitamin A content in the maize porridge which is one of the frequently used complementary food in Tanzania, encourages the formulation complementary foods using more nutritious ingredients. Examples of ingredients include orange-fleshed sweet potatoes, carrots, legumes and seeds as well as using improved traditional processing methods such as fermentation, soaking, germination/malting and de-hulling (Jemberu et al., 2016).

Studies have shown that most homemade complementary foods in Ethiopia have low vitamin C content (Abeshu et al., 2016). This is not different from what has been reported by this study whereby vitamin C content of the frequently used complementary foods in Rombo district ranged from 3.48 to 9.56 mg/100 g of dry sample. Composite flour porridge and banana porridge with fish had the highest vitamin C content relative to the rest of the complementary food samples. This may be due to the addition of fish at the end of cooking which reduced the cooking time of fish since ascorbic acid is

heat labile. Carletti et al. (2017) and Idris et al. (2019) recommend addition of vitamin C rich ingredients such as citrus fruits, tomatoes, green, yellow and red peppers as well as green leafy vegetables to home-made complementary foods.

Iron, zinc, and calcium have always been reported as limiting nutrients in unfortified plant-based complementary foods commonly used in developing countries (Gibson et al., 2010; Abeshu et al., 2016). Similarly, most of locally used complementary foods in Tanzania were poor in iron, zinc and calcium because they are mainly plant-based with little or no addition of animal products. The findings from this study have also shown lower than recommended amount of iron, zinc and calcium in the frequently used complementary foods in the study area.

Banana porridge with beef was the only sample that was able to provide more than the amount of iron needed from complementary foods assuming moderate bioavailability. With the exception of banana porridge with milk, banana porridge with pumpkin and rice porridge with milk, all the other samples were able to provide at least half of the recommended iron intake from complementary foods according to WHO recommendations. Several studies have suggested addition of animal products, the use of commercial infant formulas (Idris et al., 2019) as well as micronutrient powders (Solomon et al., 2017) to improve iron status of infants and young children in developing countries. In order to increase iron content and reduce anti-nutrients such as phytates, some studies suggested soaking and germination of cereals and legumes prior to processing (Mihafu et al., 2017).

According to (FAO/WHO, 2017) complementary foods should provide 86 to 100% of zinc based on the age and breastfeeding status of the child. With the exception of banana porridge with beef, all the other samples of frequently used complementary foods in Rombo district had less than 3 mg/100 g of zinc. This value is lower than what is recommended by World Health Organization (WHO, 2011). Even though they are lower than the recommended amount, they are still higher than the findings of Yanmife et al. (2020) in Pakistan and Kulwa et al. (2015) in Tanzania. The higher zinc content in banana porridge with meat could be due to the presence of meat, which was a good source of zinc.

The recommended amount of calcium (196-353 mg/day) needed from complementary foods for children aged 6-23 months could not be met by plant-based complementary foods in Rombo district. Only banana porridge with beef had the calcium value above the recommended range. Even the milk containing foods such as rice porridge and banana porridge with milk had lower calcium values relative to beef-containing foods. This could be due to the fact that only a small amount of milk is added to complementary foods and sometimes the milk over-diluted. Another study conducted by Laryea et

al. (2018) have also reported lower than recommended amount of calcium in complementary foods.

The lowest iodine concentration was found in banana porridge with beef (10.18 µg/100 g dry weight) and the highest was found in composite flour porridge (200.93 µg/100 g dry weight). With the exception of banana porridge with beef, iodine content of all other samples were above the recommended intake of 19, 30 and 51 µg/day from complementary foods for infants and young children aged 6-8, 9-11 and 12-23 months, respectively (Laryea et al., 2018). The reason for low iodine concentration in banana porridge with beef might be the use of poorly stored and expired salt.

Conclusion

Most of the frequently used complementary foods were found to contain lower than recommended amount of energy, protein, vitamin C, iron, zinc, calcium and iodine. Therefore, this study provides a benchmark for educating mothers on the importance of including nutrient-dense ingredients and proper preparation methods for complementary foods.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Full Length Research Paper

Drying fish (*Rastrineobola argentea*) on the bank of Lake Victoria in a prototype solar-heated enclosure: Using renewable energy to reduce drying-time and improve product quality and value

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Silver cyprinids (*Rastrineobola argentea*) are small pelagic fish endemic to Lake Victoria. High-quality dried fish are an important protein-rich human food. This study was carried out to determine if it would be practical to use renewable energy to dry silver cyprinid. Drying is presently achieved by laying fish out in the sun on the ground or on racks. In the wet seasons, however, drying is compromised and much of the catch becomes fit only for animal consumption, or spoils. Lake Victoria's surface waters are c. 25°C year-round. Its enormous volume of tropical water offers a source of thermal energy that could be used to dry fish. As a proof of concept, we used solar-generated electricity to drive a heat pump to harvest heat energy from water in a 10,000-L rainwater tank beside Lake Victoria, used the energy to heat air, and blew the air over fish in a tent-like enclosure. Fish in the enclosure dried in about 4 h versus about 7 h outside, were free from insects, and not at risk of theft by or defecation upon by birds. The drying processes inside and outside the enclosure were modelled. The model correctly reproduced observed drying times, and enabled exploration of options to improve drying performance. Up-scaling the prototype could provide year-round sustainable fish-drying capability, reduce waste, boost food security, and add value to the catch.

Key words: *Rastrineobola argentea*, dagaa, mukene, omena, drying, renewable energy, food security.

INTRODUCTION

Fish is an important source of protein, omega-3 poly-unsaturated fatty acids and micronutrients for humans

(Golden et al., 2021) and is also valuable as an animal feed. Lake Victoria, East Africa, yields c. 1 million tonnes

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of fish per annum (LVFO, 2016a). Nile perch, an introduced species achieves a high price (up to UGX23,000 per kg wet mass in 2022), and most of the annual catch of c. 250 thousand metric tonnes (approximate median annual value for the period 1994 to 2014 (LVFO, 2015)) is exported in frozen fillets as a luxury food for the developed world, generating foreign income (\$260 Million in 2012, which was c. 2% of the riparian states' trade balance (LVFO, 2105)). The small (maximum length c. 9 cm) and endemic Silver cyprinid (*Rastrineobola argentea* - known locally as dagaa, mukene or omena) is less expensive (maximum price around UGX1,300 per kg wet mass in 2022), and is sold locally for human consumption or animal feed. Approximately 650 thousand tonnes of Silver cyprinid are landed each year (LVFO, 2016a). These fish are caught at night from open canoes using purse-seine-like nets after being attracted towards the surface by lamp light: catches are higher on dark nights around the new moon when lamp light stands out strongly against the background dark sky.

Fish are landed at dawn and spread on the ground or on elevated racks to dry in the sun in the open air. This open-air drying can be effective in the dry seasons (June to September, and January to February) and is sometimes achieved in a single day (it can take two days or more though). The target water content for safe, long-term preservation is around 15% by mass (Abdulmajid, 2015), but 'dryness' is judged by fish-dryers on appearance and feel rather than by formal measurement. In any case, since dried fish are sold by weight, and because reducing water content reduces weight, dryers can find it preferable to sell fish as soon as possible. Fish laid out to dry in the open are exposed to birds (that eat them and defecate on them) and insects, and the fish can become contaminated with foreign material. Sun drying also results in vitamin loss, discoloration, and if fish are not well spread, uneven residual moisture distribution can occur and leads to spoil (Sreekumar et al., 2008). In the wet seasons (March to May and October to December) when rainfall is high, open-air drying can be impossible and the low water content required for safe long-term storage is often unachievable (Oduor-Odote et al., 2010). Product-quality in the wet season is generally low, and much of it is fit only for animal feed: fish that fail to dry are dumped and losses can exceed 50% (Owaga et al., 2011). High post-harvest loss, including rotting or spoilage due to insufficient sun-heat to dry the catch, particularly during the wet seasons, has been identified as a key feature of Silver cyprinid trade in the Lake Victoria region (LVFO, 2016b). Paradoxically, the wet season is the time of year when catches are highest (fish migrate inshore to spawn and become more accessible to fishers), but because of product-quality issues, catch can fetch as little as UGX200 per kg wet mass. In 2014, Silver cyprinid contributed 55.4% to the total annual catch

from Lake Victoria but contributed only 16.1% to the total catch value: development of a reliable and sustainable method to dry these fish in the wet season would bring substantial benefit in terms of food-availability, nutrition and value-chain improvement (LVFO, 2016b).

Several methods have been explored to improve fish drying. Wood-fired kilns are used to dry larger, more valuable species such as *Tilapia* and catfish, but kiln-drying is not viable for the large volumes of smaller and lower-value Silver cyprinid. In any case there are environmental concerns around unsustainable harvest of firewood, and wood smoke is damaging to human health. Passive indirect solar dryers have been able to dry fish to acceptable levels, and have sometimes achieved drying times quicker than in the open air (Abdulmajid, 2015). These passive dryers rely on absorption of solar radiation by black materials that then heat air indirectly, but the process is inefficient, especially during times of cloud cover. Active dryers have not been adopted because of the cost of energy, and the fact that some landing sites (fish are often dried close to where they are landed) are not on the electricity grid. A reliable low-cost and sustainable heat source is required.

Lake Victoria straddles the equator. It is the world's largest tropical lake (c. 60,000 km² area, and 2,400 km³ volume). The lake has an annual average surface temperature of around 25°C and acts as a storage reservoir of radiational heat (Yin and Nicholson, 1998). In this study, we investigated whether this heat-energy could be harvested using a heat pump powered by solar-generated electricity and used to dry Silver cyprinid. Heat pumps transfer energy in the form of heat from one location to another. Domestic refrigerators, for example, cool down inside when a pump moves heat from inside to the cooling vanes outside (which get hot), from where the heat dissipates as 'waste' to the air. We report here some preliminary experiments to use heat-energy extracted with a heat pump from a 10,000-L rainwater tank adjacent to Lake Victoria using renewable off-grid electricity (solar power) to dry Silver cyprinids in a tent-like enclosure that served also to shield fish from birds and insects. In addition, we develop a numerical model of the drying process under ambient conditions and within the enclosure, and use the model to identify the key optimisation parameters for the system under constrained energy budgets. The data reported may be useful in up-scaling fish-drying capacity in the wet season (the tent enclosure provides shelter from rain), and for extending drying beyond periods of daylight since solar-generated electricity can be stored in batteries.

MATERIALS AND METHODS

Location of the study and physical setup

A 3 m long × 2 m wide × 2 m tall plastic tent-like enclosure (Figure



Figure 1. The drying enclosure with the heat pump in position in the eastern end (gaps yet to be closed), the 10,000-L rainwater tank, and the pipe along which water was pumped from the tank to the heat pump. Source: Authors

1) was set up to dry Silver cyprinid at the National Fisheries Resources Research Institute (NaFiRRI) in Jinja, Uganda (0.416°N, 33.206°E) in February 2022. The enclosure was erected in the NaFiRRI grounds less than 1 km from Lake Victoria and only 10 m or so above lake level. The location chosen was flat, unshaded and adjacent to a 10,000-L rainwater tank that was used as a 'mini Lake Victoria' heat source. Water in the tank was c. 26°C. A nylon net rack (190 cm × 45 cm; mesh size = 1 mm) was suspended at waist height inside the enclosure to spread fish out onto dry (Figure 2A). The rack enabled air to circulate beneath the fish as well as over the top, facilitating evaporation from most of the fish surface.

Heat source

A Proteam Europe Model P8 swimming-pool heat pump (230 Volt AC, 1100 Watt) was positioned in the eastern end of the plastic enclosure and fed with water pumped along a 25 mm internal-diameter flexible pipe from the rainwater tank using a 24 Volt DC in-water pump (Figure 1). Water was returned to the tank after passing through the heat pump. The water pump and the heat pump were driven by electricity generated from four 845 × 670 × 30 mm/100 Watt solar panels. Electricity from the panels (42 Volt DC) was directed to the heat pump via an inverter (230 Volt; Iconica 3,000 Watt, 24 V hybrid pure sine wave inverter with 40 Amp MPPT Solar controller). This inverter also charged two 75-Amp Hour lead acid car batteries wired in series. The battery pack was included to bridge periods of cloud cover and to provide efficient power to the 24 Volt DC water pump. It also provided electricity for lighting that could enable start up before sunrise. The heat pump had the capacity to cool water to a minimum of 8°C, and it was set at that to give the maximum output of heated air. The heat pump

specifications indicate that this configuration would produce a Coefficient of Performance (COP) of ca. 5.4 and therefore a heat output of ca. 6 kW. The heat-pump fan had an air displacement of 2,200 m³ h⁻¹. For the tent cross section of 4 m², this corresponds to a minimum air velocity of 0.15 m s⁻¹. As air left the pump in a directed flow though, and because the fan outlet was quite close to the fish (c. 3 m range), we estimate that the air velocity over the fish was approximately an order of magnitude larger, at about 1 m s⁻¹.

Fish

Fish were obtained fresh from the landing site at Kikondo each morning. The site, on the opposite side of Napoleon Gulf to NaFiRRI, was about 2 km away by boat. The boat journey took around 10 minutes, and fish were at our experimental site within an hour of being landed. Two experiments were carried out to compare the fish-drying trajectory inside the enclosure with outside. The first, on February 24, 2022 involved the solar-powered heat pump described earlier blowing heated air into the enclosure. In the second, on February 25, the heat pump was replaced with a simple electric fan (Figure 3) that blew ambient air into the enclosure at the same rate as blown by the heat pump. Evaporation is a function of air speed, temperature and relative humidity (Hisatake et al., 1995), and we were interested to determine if gains in drying time in the enclosure compared with those in the open could be achieved with blown air alone since a simple fan has a lower energy requirement (80 Watts) and purchase cost (£30) than a heat pump (1100 Watts and £1000). As a control for both experiments, we suspended a rack of the same nylon mesh as used in the enclosure at waist height outside the enclosure to spread fish out on to dry (Figure 2B).

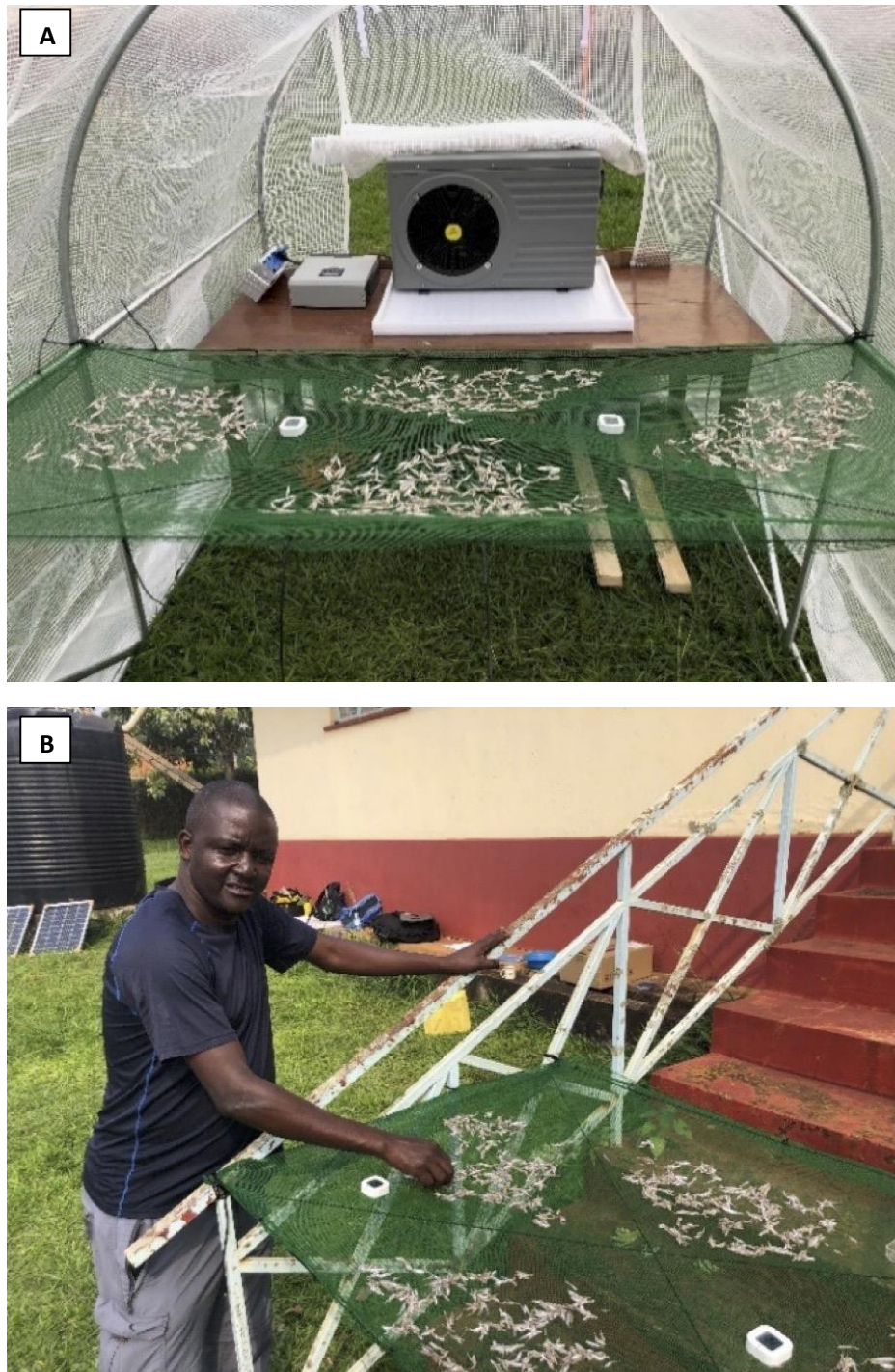


Figure 2. (A) Drying racks inside the enclosure and (B) outside, showing four 100-g samples of fish spread out to dry. The white cubes on the drying racks are SensorBlue temperature/humidity loggers.
Source: Authors

Experimental approach

For both experiments (heat-pump versus ambient, and fan versus

ambient), we weighed 8 samples of 100.00 g of freshly caught fish and spread 4 samples on the rack in the enclosure and 4 samples on the control rack outside. Every hour for the next 7 h (experiment



Figure 3. The drying enclosure as used in Experiment 2 on February 25, with a simple fan in place of the heat pump used the day before in Experiment 1. The fan delivered air at the same rate that the heat pump had, but the fan diameter was smaller so fan speed was higher.

Source: Authors

1) or 6 h (experiment 2), we gathered each of the 8 samples, weighed them to the nearest 0.01 g on a portable electronic balance, and returned them to their respective racks. Temperature and relative humidity were recorded inside the enclosure and outside throughout each experiment using SensorBlue WS08 Smart Hygrometers. At the end of each experiment, we placed the 8 samples in a drying oven at 100°C and left them overnight to desiccate completely before weighing again. Finally, for experiment 1 samples, we weighed the fish again after they had been left for 4.5 h open to the air after desiccation to determine mass of water reabsorption.

Modelling

A physics-based model of Silver cyprinid drying was developed to enable us (a) to consider our empirical observations in the context of the drying processes that should, in theory, be in action, and (b) to explore options for optimizing the drying process energetically (e.g. to explore whether additional air-flow or heat or both would be beneficial). The drying characteristics of Silver cyprinid have been studied in detail (Oduor-Odote et al., 2010). Drying occurs in two stages: an initial constant rate, followed by a decreasing rate. In the initial constant-rate period, evaporation occurs from the fish surface and can be reasonably well approximated as the rate of evaporation from a free water area (Oduor-Odote et al., 2010; Hisatake et al., 1995). In the second phase, the limiting factor is internal diffusion of water from the inside of the fish to the outer surface: the rate of this reduces over time and so the drying rate also decreases over time.

Our model is framed around the moisture content per dry weight M of fish (sometimes referred to as dry basis). M has units of kg/kg_{dw} corresponding to kg water per kg dry weight (dw), and is defined as:

$$M = \frac{W_w - W_d}{W_d} \quad (1)$$

With W_w being the wet weight and W_d the dry weight of the fish. Initially, fish have a moisture content of $M_0 = 0.8$ kg/kg_{dw} and this decreases toward 0 kg/kg_{dw} at full desiccation. During the constant-rate drying phase, the drying rate dM/dt is a function of the relative humidity h_r of the surrounding air, temperature T (we assume for simplicity that the fish are at the same temperature as the air within the tented enclosure in experiment 1 and as ambient air outside), and air velocity v (which is wind velocity outside, and blown air velocity in the enclosure in the respective experimental conditions). Under the turbulent flow conditions in our experiments dM/dt for a single fish is given by the following semi-empirical formula (Hisatake et al., 1995):

$$\frac{dM}{dt} = -c_1 n_s (1 - h_r) \left(\frac{v}{v_1}\right)^{0.9} \left(\frac{D_{fish}^{eff}}{D_1}\right)^{1.8} \quad (2)$$

Where, n_s is the saturation water vapour density at temperature T (Foken, 2021), h_r the relative humidity, v the air velocity and

D_{fish}^{eff} is the effective average fish length. C_1 is an empirical constant that was determined by Hisatake et al. (1995) to be 4 mg h^{-1} for a wind velocity $v_1 = 1 \text{ m s}^{-1}$ and dimension $D_1 = 1 \text{ cm}$, with the scaling laws for air velocity v (0.9) and dimension D (1.8) being determined experimentally as well.

In the decreasing drying-rate period, drying is most likely limited by the diffusion of residual moisture from the interior tissue of the fish. In this regime, the moisture content M is modelled by a form of Newton's model for thin layer drying (Oduor-Odote et al., 2010):

$$M = M_0 \exp(-kt) \quad (3)$$

Where, k is the drying rate constant, and t the time. The drying rate dM/dt in the falling rate phase is therefore:

$$\frac{dM}{dt} = -kM_0 \exp(-kt) \quad (4)$$

From a meta-analysis of the available literature (Abdulmjid et al., 2015; Oduor-Odote et al., 2010; Mujaffar and Sankat, 2005, 2014), k was found to have no significant dependence on the relative humidity, but to follow a linear relationship with temperature:

$$k = 0.0030 \text{ h}^{-1} \text{ K}^{-1} T - 0.7805 \text{ h}^{-1} \quad (5)$$

Equations 4 and 5 as well as the environmental conditions determined on the days of the experiments (temperature, humidity and estimated wind velocity) for the ambient (outside) simulations were utilised to model ambient drying.

In order to simulate drying in the heat pump system, we took the following additional effects into account. First and foremost, the heat pump raised the temperature of the air within the enclosure by approximately 15°C . We therefore modelled the air temperature within the tent as being the outside air temperature plus 15°C . The main effect of this elevated temperature is a corresponding reduction of the relative humidity h_r by c. 20%.

Air speed inside the tent is driven by the heat-pump fan at approximately 1 ms^{-1} at the location of the fish, compared to the prevailing conditions outside of approximately 2 ms^{-1} (windspeeds obtained from weather data for the location and time of the experiment (Hersbach et al., 2022)). In addition, we took into account that the evaporated moisture increases the humidity in the tent while moisture is furthermore continuously removed via the intake of fresh air and the ejection of moist air from the enclosure. For our experiment in the small, prototype enclosure, this led to a minor increase in relative humidity of only about 1%, but increased relative humidity within the enclosure may become an important factor in an upscaled version. The aforementioned model was implemented in Matlab Simulink.

RESULTS

Experiment 1: Enclosure + heat pump versus open air

February 24th started as an overcast day with nil wind. At the start of the experiment, at around 8 a.m. local time, air temperature was c. 23°C outside and 26°C inside the

enclosure. Relative humidity was similar inside and out (c. 75%). Fish had a mean length of 25.1 mm (range 18 to 36 mm, mode 25 mm; Figure 4) which, according to length-mass relationships in Yongo et al. (2016) would equate to a mean mass of c. 0.10 g per fish.

Temperature outside the enclosure climbed steadily to a maximum of about 38°C at around 1 p.m., and relative humidity fell to a low of about 40% by that time (Figure 5). Temperature inside the enclosure rose to a maximum of around 52°C by noon, with relative humidity dropping to 30% by that time (Figure 5). The mass of the fish samples inside the enclosure dropped rapidly hour by hour, reaching a floor of around 22 g after about 4.5 h. The fish outside lost mass (water) more slowly, reaching a low of around 24 g after 7 h when the experiment ceased. Mass-loss trajectories are as shown in Figure 5.

Mean final oven-dry sample masses were 20.00 g taking this to be 0% moisture content, the mean final mass of the fish dried with the heat pump in Experiment 1 of 21.12 g equated to a moisture content of 5.29%. The moisture content of the fish dried outside as the control in Experiment 1 equated to a moisture content of 17.07%. Oven-dried (at 100°C) samples left in the open post drying for 4.5 h increased in mass by between 0.61 and 0.87 g (mean 0.72 g), increasing moisture content from 0% to a mean of 3.45%.

Figure 6 shows the output of our Matlab Simulink model under the same environmental conditions as Experiment 1 (Figure 5B and C). The physical model captures the drying characteristics outside and inside the enclosure qualitatively and quantitatively. The transition from the constant-rate drying regime to the decreasing-rate drying regime is evident after c. 4 h in the tented enclosure and c. 6.5 h under ambient conditions outside, and modelled drying times are consistent with the experimental data. This close agreement provides evidence that the model captures well all the relevant experimental parameters and processes. We are therefore confident that the model provides a good basis for the optimisation of the system under constrained energy budgets.

Experiment 2: Enclosure + fan versus open air

February 25th started as a bright day with clear sky and a light wind. At the start of the experiment, at around 8.30 a.m. local, air temperature was c. 26°C outside and 29°C inside the enclosure. Relative humidity was 65% outside and 70% inside. Fish available for Experiment 2 were larger than those landed the previous day, with a mean length of 28.3 mm (range 17 to 42 mm, mode 30 mm, equivalent to a mean mass of c. 0.15 g; Figure 4). Temperature outside climbed steadily to a maximum of about 39°C at around 1 p.m., and relative humidity fell to a low of about 36% by that time (Figure 5), and wind

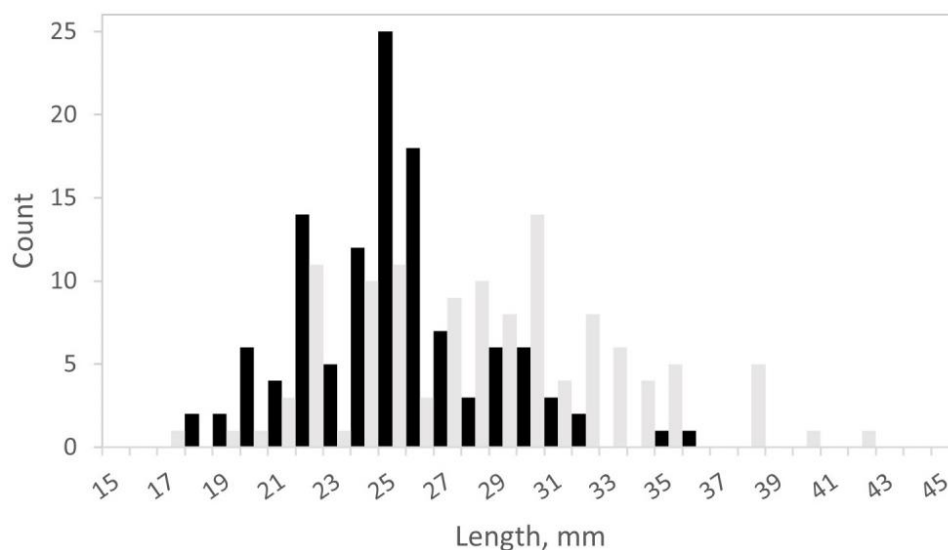


Figure 4. Length distributions of fish used in Experiment 1 (black bars) and Experiment 2 (grey bars). N = 117 fish in both plots.
Source: Authors

velocity increased through the day.

Temperature inside the enclosure rose to a maximum of around 42°C by 1 p.m. (this maximum was c. 10°C cooler and occurred 1 h later than in Experiment 1 with the heat pump), with relative humidity dropping to 39% by that time (Figure 5): humidity inside the enclosure was marginally higher than out, possibly due to evaporation of water from the fish into the confined space of the enclosure. The mass of the fish samples inside the enclosure dropped slightly more rapidly than outside over the first 3 h, but by 4 h samples outside and in had a similar range of masses, with both sets of samples reducing rapidly in mass hour by hour, reaching a floor of around 24 g. Mass-loss trajectories are as shown in Figure 5.

Mean final oven-dry sample masses were 20.17 g. Taking this mass to be 0% moisture content, the mean final mass of the fish dried with the fan in the enclosure in Experiment 2, and outside as the control (both means were 24.13 g), equates to a moisture content of 16.42%.

DISCUSSION

Heat pump versus ambient drying

The water-source heat pump was able to increase the temperature in the enclosure on February 24 2022 to 52°C, or some 14°C above ambient. At the same time, it reduced relative humidity between 10 and 20% relative to ambient conditions to a minimum of around 30%. Fish

outside the enclosure dried on February 24 to a moisture content of around 17.1% after 7 h. Fish inside the enclosure reached this level of dryness around 3 h sooner, and over the 7-h duration of the experiment dried in the enclosure to moisture content of 5.3%. The enclosure and heat pump together clearly provided substantially better drying conditions than those outside on February 24. Indeed, it would have been possible to dry two batches of fish in sequence that day on the single rack in the enclosure to the moisture content achieved on the control rack over the full 7 h.

February 24 started off overcast, but there were sunny intervals through the day, and it did not rain: the drying-time gains provided by the enclosure and heat pump would likely be relatively much greater during the wet season. Furthermore, our control was an elevated rack, with air circulating above and below fish. In practise much drying is on the ground, with evaporation therefore principally from the upper surface only, so likely to be at a slower rate than in our control: the relative gain of the tent compared to standard 'field' drying conditions is therefore likely to be greater than portrayed here. The enclosure provided the added advantage of protecting the drying fish from birds and insects.

Fan versus ambient

Although the fan used in experiment 2 on February 25 was able to blow air over the fish in the enclosure at the same rate as had been achieved by the heat pump, this

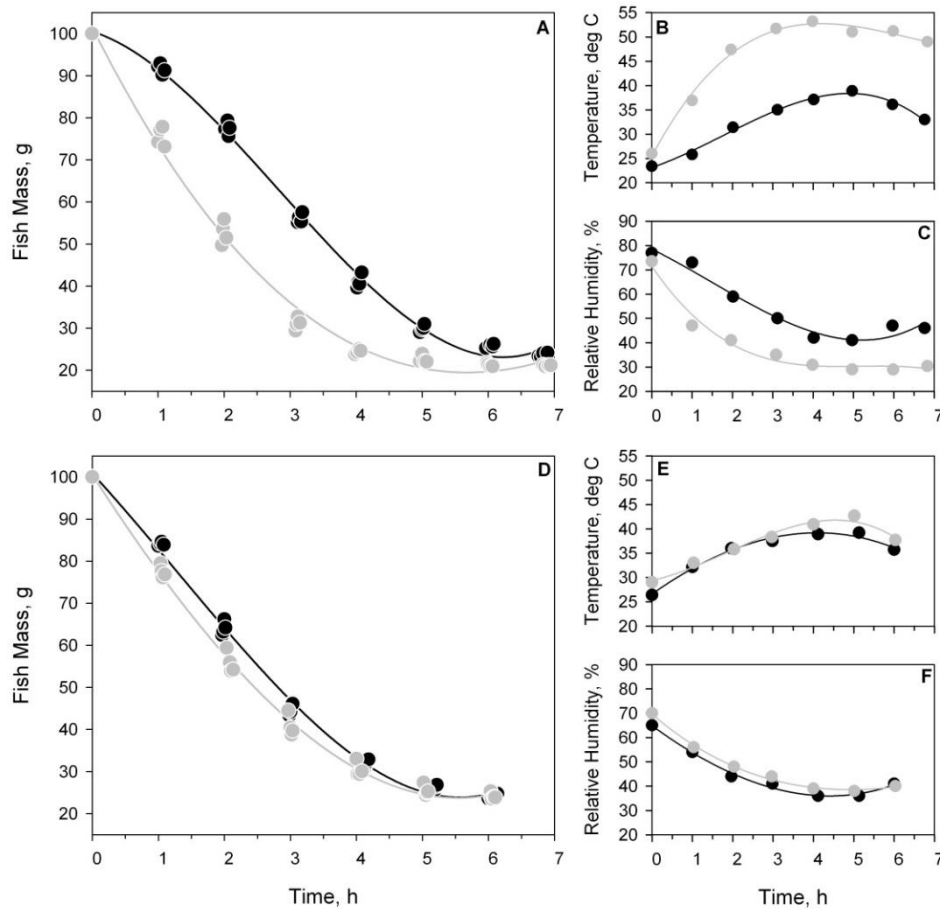


Figure 5. Drying (mass-loss) trajectories, and temperatures and humidities (C) inside and outside the enclosure for Experiment 1 with the heat pump (panels A, B and C) and Experiment 2 with the fan (panels D, E and F). Trajectories inside the enclosure are shown by grey symbols and lines. Trajectories outside in the open air are shown by black symbols and lines. The lines are simple smooths to guide the eye, and are not intended to suggest statistically significant relationships. Drying trajectories tend towards asymptotes slightly above 0% moisture content.

Source: Authors

blown ambient air did not deliver improvements in fish-drying rate or final moisture content compared to the control samples outside (wind velocities outside and air velocities within the tent were of the same order of magnitude). This is consistent with model expectation, given the closely similar temperatures and humidities inside and outside on February 25. Although the fish in the enclosure lost mass initially more quickly than those outside, there was no difference between drying floor or time to drying floor. Fish inside the enclosure were however free of from ants, whereas those outside were not.

Comparison between experiments

The weather on February 25 (Experiment 2) was warmer

and with a stronger breeze than on February 24 (Experiment 1). Fish outside reached the drying floor between 5 and 6 h on February 25, which was about an hour sooner than on February 24, and this was even though fish available to us on February 25 were larger, and hence had a lower surface area to volume ratio than those used on February 24. Evaporation of moisture from the fish will take place from the surface, and moisture from the fish interior will diffuse towards the surface as the surface dries: larger fish with lower surface area to volume ratios would be expected to dry more slowly than smaller fish under the same conditions. The differences in drying between our 2 experiments reveal the likelihood of considerable day-to-day variability: season to season variability should be expected to be considerably more marked.

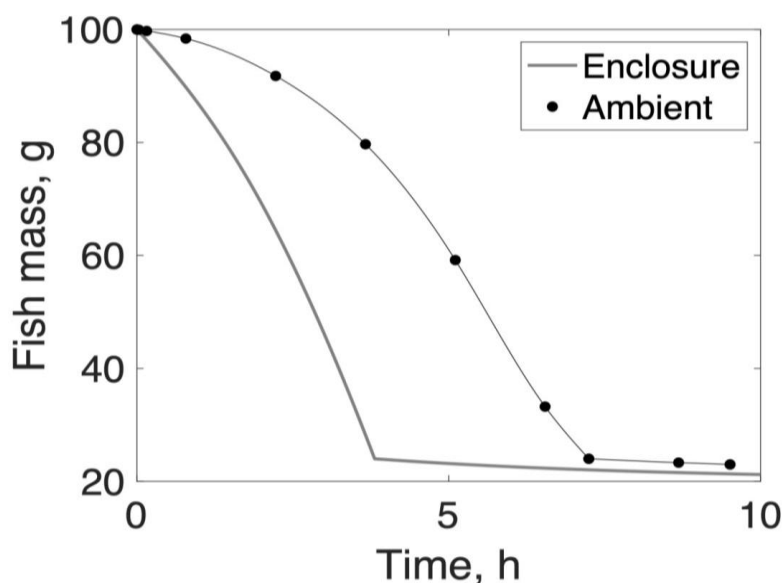


Figure 6. Simulated drying (mass-loss) trajectories for conditions prevailing during Experiment 1. Trajectories equivalent to inside the enclosure are in grey, and outside the enclosure in black.
Source: Authors

Modelling evaporation and drying

Given that evaporation is a function of temperature, air-flow and relative humidity (Hisatake et al., 1995), it is instructive to examine from a physics standpoint where energy should most effectively be invested to maximise drying. Given a limited electrical energy input (limited by the size of the photovoltaic array used to generate solar electricity, and the capacity of batteries to store it), should the available energy budget be directed towards raising temperature or increasing the rate of air flow, or some combination? The heat pump generated an air displacement of approximately $2,000 \text{ m}^3 \text{ h}^{-1}$ which, for a standalone fan of similar capacity requires about 160 W. If this energy was doubled (to 320 W) it would result to first order in a 40% increase in air flow. Providing the same amount of additional energy to the heat pump would increase the air temperature by $\sim 2^\circ\text{C}$. Figure 7 shows the behaviour of the model in these two scenarios, that is; (a) increasing the air velocity by 40% (blue) or (b) increasing the temperature of the air by 2°C (red) relative to the conditions in the tent during Experiment 1 (grey). It is evident that under these conditions, additional energy expended on increasing the air velocity will decrease the drying time more than twice as much than expanding the same energy on increasing the air temperature.

Furthermore, we ran a simulation for weather data with hourly resolution available for the whole year via the ERA5 climate database (Hersbach et al., 2022). This revealed that it was possible to dry fish to the 'safe'

moisture content of 15% dry weight in less than 10 h using the prototype drying enclosure on every day of the year, including during the rainy season (when higher humidity and lower temperature would be expected). Initial calculations suggest that similar levels of performance could be achieved in an upscaled version suitable for processing 50 kg of fish.

Potential for increasing energy efficiency

The results suggest that elevated temperature is important for accelerated drying. Our basic swimming-pool heat pump was able to achieve temperatures in the enclosure of around 55°C . Above about 60°C fish begin to 'cook', so rather than trying to deliver substantially more heat, improved efficiency in capture of heat from water should be sought. We used a basic, cheap (£1,000) and rather inefficient water-source heat pump for this proof-of-concept study. The heat pump had a specified maximum coefficient of performance (COP) of 6. It was observed that, under full load, the water temperature dropped by 1°C from inlet to outlet across the heat pump at a flow rate of 2 m^3 per hour. This indicates that further efficiency could have been achieved if larger water pump was used (ideally the temperature drop across the heat pump should be negligible, which can be achieved by greater water flow). An 'off the shelf' heat pump with a specified maximum COP of 15 can be purchased for around £2,500. This would deliver over

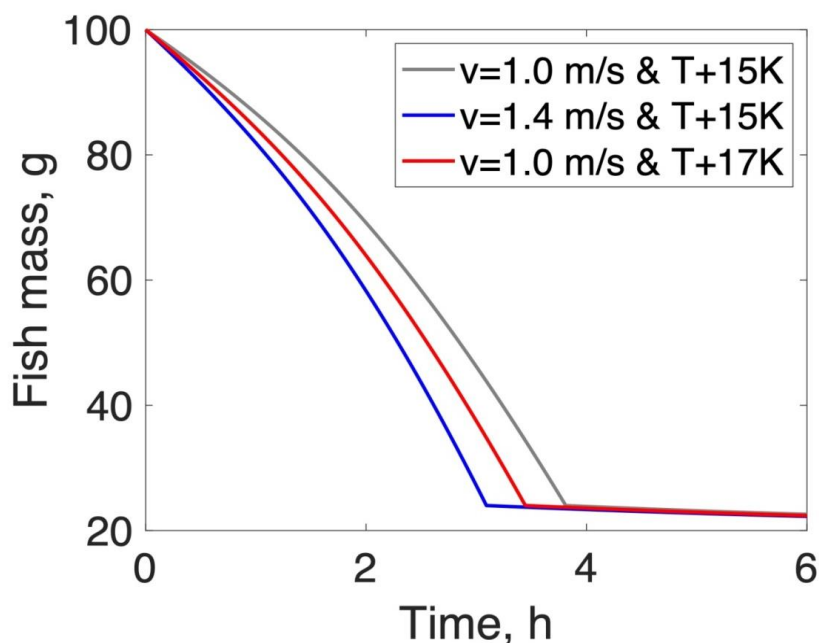


Figure 7. Modelled fish drying as a function of time for the conditions in Experiment 1 (grey), a temperature raise of 2°C (red) or alternatively wind velocity increase of 40% (blue).
Source: Authors

double the heat output for the same electrical input. Calculations indicate that a heat pump specifically designed for our application could have bettered the COP of 15 by an additional ca. 25%. Air-source heat pumps are much less energy efficient than water-source heat pumps; this is due to the inherently variable temperature of the source air and its lower density and lower heat capacity. Ground source heat pumps have similar energy efficiency to water source heat pumps because the temperature of these two sources is relatively constant and their heat capacity is high. Ground source heat pumps would add flexibility in cases where drying operations were carried out away from Lake Victoria.

Further improvements in drying rate could also perhaps be achieved by drying the air before blowing it over the fish: this air drying could be achieved by a condensing heat pump, but the condensing process would cost energy. In our setup with a tent erected on grass, we may have been able to achieve a reduction in humidity in the tent by placing a groundsheet on the floor: some of our heat energy probably served to dry the NaFiRRI lawn rather than the fish!

Gains to be made by improved drying

Post-harvest loss due to spoil following inadequate drying - which may exceed 50% of landed fish by mass - has

been identified as one of the major impediments to community Silver cyprinid processors (LVFO 2016b). Although a moisture content of only 15% is desired for long-term preservation (<15% moisture content is required to stop the growth of mould, and <25% content required to stop the growth of bacteria; Oduor-Odote, 2010), this low level might not be required if fish are to be eaten locally quite soon after capture. Indeed, economic analyses presented by LVFO (2016b) suggests a wet-to-dry weight conversion ratio of 1:0.4, which equates to a 50% moisture content (from our measurements here, 1 kg of fresh fish would dry fully to 0.2 kg; 0.4 kg of product from 1 kg of fresh fish would therefore contain 0.2 kg of desiccated fish and 0.2 kg water). In the conditions provided in our enclosure by the heat pump, 50% moisture content was achieved in around 2 hours (Figure 5). Such rapid processing would enable dryers to process multiple batches of fish per day. In the wet seasons, when fish are abundant, open-air drying typically takes 2.5 days (LVFO, 2016b): reducing the drying time by use of renewable energy could reduce wastage, particularly in the wet seasons, deliver more fish of a quality suitable for human consumption, and improve value along the entire catch and processing chain.

Although drying to 15% moisture content by mass is apparently not always required (indeed, perhaps not desired by dryers since 'dried' fish are sold by weight, and dryer fish are lighter), very dry fish that can be stored

for long periods - and hence transported substantial distances from Lake Victoria - can command a high price. Mukene Industries, for example, ask UGX 7,000 per kg for clean, high-quality air-dried product, which is more than double the beach-dried price. In some circumstances, then, drying to 15% could be a sensible business choice. With access to sufficient space in a heat-pump-driven facility, dryers could dry fish to desired levels in wet seasons and dry seasons, and make choices around product quality, longevity or value - in the security of renewable energy- to optimise food availability or income.

Nutritional quality

This study did not include any measures of nutritional quality of fish. A useful avenue for future research would be analytical comparison of fish dried in the open and fish dried in the enclosure. This could provide reassurance that gains made in reduced drying time, and reduction of spoil-rate (in the wet season), were not negated by reductions in nutritional quality. It is unlikely, however, renewable-energy-mediated drying would impact nutritional quality adversely. Studies on the effects of four different artisanal processing methods (smoking, salting, sun-drying and deep-frying) on the lysine content of Silver cyprinid found that only deep-frying reduced the content of this essential amino acid significantly (Margaret and Edgar, 2016). Deep-frying exposes fish to temperatures around 180°C and, using methods employed in Uganda, temperatures during smoking may exceed 120°C (Margaret and Edgar, 2016): lysine losses increase markedly with temperature, exceeding 80% at 120°C. In our experiments, by contrast, temperatures with the heat pump did not exceed 55°C, and with the fan alone did not exceed 45°C. The fact that use of the enclosure produced dried fish with a very low final moisture contents may actually result in *increased* lysine content weight for weight. Other work has reported moisture-loss leading to concentration of protein (Akinwumi, 2014).

Conclusion

Our experiments have shown that considerable gains in drying rate and final moisture content can be achieved in the dry season using an enclosure and water-source heat pump compared to ambient conditions outdoors. Improvements in the wet season are likely to be much better. Individual fish dryers typically work an area of few tens of square m, and are able to dry 1 batch of fish per day in the dry season. An agricultural polytunnel could be used to cover an area greater than this, and use of heat pumps could enable multiple batches of fish to be dried

per day. The quality of the product (in terms of contamination) would likely be higher than for product dried outside on the ground. This would lead to increased availability of fish for human consumption, add value to the product, and bring better income for fishers and fish-dryers. Application of the sort of sustainably powered technology demonstrated here could improve food security, nutrition and economic wellbeing around Lake Victoria. Furthermore, since small pelagic fish are caught from many African lakes (e.g. Malawi, Tanganyika, Mweru, Kivu, Kariba) (Kolding et al., 2019), this technology could bring considerable benefits across Africa.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

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Full Length Research Paper

Phytochemical characteristics and consumer acceptability of watermelon (*Citrullus lanatus*) wine

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Wine, a fruit-based fermented and non-distilled product, containing most of the nutrients in the raw fruit. Wine production, particularly grape wine, is one of the oldest technologies in the history of man. Other fruits such as pineapple, have been used for wine production. The research was conducted to determine the phytochemical characteristics and consumer acceptability of watermelon wine produced using four commercial *Saccharomyces cerevisiae* strains (Lalvin EC 1118, Lalvin QA 23, Redstar star rouge and Redstar premier classique). The data obtained on the phytochemical and free radical scavenging activities were analyzed using Tukey Pair Wise comparison at 95% confidence level for statistical difference. Sensory data was translated into means and ranked. The average mean of total phenols ranged in the young wine samples from 93.36 to 105.33 GAEmg/g) and that of the matured samples wine from 156.08 to 275.99 GAEmg/g). Flavonoids was 35.29 to 52.48 and 34.72 to 58.38 in the young and mature wines, respectively. The mean antioxidant activity and antioxidant capacity ranged between 1.28 to 13.59; 79.14 to 246.72 in the young wine samples and that of the matured wine samples 1279.93 to 1914.02 and 808.62 to 2219.37, respectively. Any of the four commercial yeast strains used in this study was suitable for watermelon winemaking. However, it evinced that the Redstar premier classique produced the most desirable sensory attributes, followed by the Lalvin EC 1118 yeast strains

Key words: *Sacchromyces cerevisiae*, wine, antioxidant, phytochemicals, scavenging activity, watermelon.

INTRODUCTION

Wine, a fruit-based fermented and a non-distilled product contains most of the nutrients present in the raw fruit. Therefore, fruits are important sources of compounds demonstrating beneficial effects on human health, of which one of the most attention-grabbing compounds for researchers today is dietary polyphenol (Mojzer et al., 2016). These dietary polyphenols are termed antioxidants

because they can scavenge radicals in the biological system (Zhou et al., 2016). However, in the raw form of fruits, polyphenols are strongly bonded together, making it less soluble and accessible when eaten raw. In contrast, in wine, polyphenols are present in the soluble form rendering them biologically accessible (Mojzer et al., 2016).

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Wine production involves the metabolism of soluble sugars (glucose and fructose) into ethanol and carbon dioxide. Wine is a rich source of most polyphenols. Antioxidant characteristics of red wine have been linked to the materials phytochemical profile (Lingua et al., 2016). Wine production is one of the oldest technologies in the human era (Morrison and Rabelotti, 2017). Nevertheless, wine gained popularity in the 17th century due to the effective use of sulphur in the sterilization of storage barrels which paved the way for the production of highly recommended wines and their associated increased shelf-life capacities (Raposo et al., 2018). Furthermore, the clinical benefits of wine, such as the prevention of cardiovascular diseases and cancer, influenced consumers to shift from consumption of other alcoholic beverages such as beer to wine (Pavlidou et al., 2018). Hence, it is now one of the most famous drinks in the world (Morrison and Rabelotti, 2017).

According to the European Union, wine is legally classified as the fermented juice of grapes; however, several plant fruits can be fermented to produce the alcoholic beverage (Saranraj et al., 2017), by using yeast strains (Saranraj and Ramesh, 2019). Different fruits can be used for wine production due to their sugar content, which is the main substrate for alcoholic fermentation. For instance, one of the widely known non-grape fruit wine is apple fruit wine (cider) (Saranraj et al., 2017). However, non-grape fruit must with little natural sugar could be seeded with table sugar to spur the fermentation process (Saranraj et al., 2017).

In the development of novel foods, fruits are mostly seen as significant rich sources as they contribute to the sensory characteristics in foods and serve as bioactive compounds such as vitamins, dietary fibre and polyphenols (Saranraj and Ramesh, 2019). Unfortunately, most fruits have a shorter shelf-life at the farm gate and on the market shelves. The postharvest loss of fruits even worsens in exportation; fruits that do not meet export regulations are left to waste, resulting in financial loss to the farmers (Yusufu et al., 2018). Saranraj et al. (2017) opined that oenology and other alcoholic beverage drinks industries are the recommended 'bail out' measures to salvage the menace in fruit production farms. Fruits such as apple, pear and strawberry, cherries, plum, banana, pineapple, oranges, cucumber, watermelon and guava have been used for home-making wine (Velić et al., 2018).

Watermelon (*C. lanatus*) from the Cucurbitaceae family harbours quite several bioactive compounds besides vitamins A and C, which are available in most fruits (Romdhane et al., 2017). Watermelon is well known for its ability of "thirst quenching", it is also a rich source of lycopene (Romdhane et al., 2017). Earlier clinical studies reported that lycopene has antioxidant activity and free scavenging property on biological systems (Przybylska, 2020). The nutritional profile of watermelon reported by Wong et al. (2016), revealed that carbohydrates, sodium,

vitamins, minerals, fatty acids, and amino acids constitute the important nutrient content of watermelon. Wang et al. (2018) compared fermented and unfermented watermelon juice and reported that fermented juice showed strong antioxidant activity. Similarly, blended watermelon juice and ginger extract fermented into wine revealed that watermelon wine demonstrates improved scavenging activity, hence a promising raw material for fruit winemaking (Yusufu et al., 2018). Despite the numerous promising fruits for wine-making, watermelon has not attracted much attention for wine making from non-grape juice.

The roles of fruits in human nutrition have led most nutritionists to recommend their daily consumption; in line with that, fruit production within the tropics has substantially increased, resulting in postharvest losses in a chunk of the produce. Moreover, the short shelf-life of fruits continues to be the major challenge (Mundaragi and Thangadurai, 2017) that bedevils fruit production worldwide, particularly in developing countries such as Ghana. Postharvest losses in developing countries like Ghana is projected at 20 to 50% of fruits, vegetables, and root and tubers annually (Yakubu et al., 2018), and that of watermelon is not different. Considering the percentages of postharvest loss in Ghana, it shows that it is not only a wastage of food products but also a waste of investment and a burden on the economy as the country spends more to import the same fruits from neighbouring countries such as Burkina Faso, especially at their off-season (Addo et al., 2015).

To stem challenges confronting the fruit production sector, efforts have been made to use fruits for alcoholic beverage production (Saranraj and Ramesh, 2019). Therefore, using watermelon juice for wine production could serve as one of the additional attractive measures to help curb the postharvest loss of the fruit. Considering the nutrient composition of watermelon, its wine is hypothesized to exhibit clinical benefits as other wines (Umeh et al., 2021). Furthermore, food or beverage is worth producing, distributing or marketing when its organoleptic characteristics are accepted (Raposo et al., 2018). Therefore, this study aimed to evaluate the polyphenols, antioxidant activities and sensory attributes of watermelon wine before and after storage by exploring different commercial yeast strains.

MATERIALS AND METHODS

Experimental design and sample preparation

Fresh whole watermelon (Charleston grey) varieties were purchased from the Tamale central market in the Northern Region of Ghana. A 3x4 factorial experimental design was used for the research. The factors include the maturation period (three months) and yeast type (four strains). The watermelon juice was fermented at 25°C with 3 grams of the four differently sourced yeast strains (Lalvin EC 1118, Lalvin QA 23, Redstar premier classique and Redstar premier rouge). The finished wines were then matured with

oak chips (French oak chips) in glass bottles for three months (90 days). All four (4) treatments (the yeast strains) were in triplicates. The production and sensory evaluation of the wine samples were done at the Family and Consumer Science Food laboratory, University for Development Studies. The phytochemical quantification was carried out at the Cocoa Research Institute of Ghana (CRIG), Tafo.

Juice preparation

The sample preparation was done following modification by Darman et al. (2010). First, all containers and equipment were thoroughly sterilized (with 1% sodium metabisulphite), and fruits were processed under aseptic conditions. Next, the watermelons were washed using potable water and rinsed with 1% sodium metabisulphite. Instead of grating and mixing with water before filtration, as stated in the above protocol, the watermelon fruits were cut into pieces separating the red flesh from the rind and seeds. The red flesh (without the seeds) was extracted—by manually squeezing out the juice, and clear juice was obtained by filtration with a sterilized filter cloth. Approximately 20 l of the juice was extracted for the experiment. Thus five (5) liters of juice was used for each treatment. The clear juice was then transferred to the fermenting drum (bioreactor). Approximately 1 g of tartaric and citric acids each was added to the clear juice per litre to adjust the pH from 5.15 to 3.5; the must also was chaptalized with table sugar to increase the °Brix from 6 to 20 concentration. However, following protocol, no acid (tartaric and citric acids) was used to adjust pH level of the must. Also, sugar cane liquid was used for the juice chaptalization in the protocol. Sulfiting was employed with a pill of Campden tablet pre-dissolved in 3 ml of distilled water and added to the five (5) liters of clear juice for sterilisation. The set-up was left on the laboratory bench for 24 h at room temperature (25°C) before being pitched with the yeasts.

Yeasts conditioning, pitching and fermentation

The modified protocol by Darman et al. (2010), coupled with the manufacturer's protocol, was followed for the starter culture conditioning. The yeasts were used in the rehydrated form as the fermentation agent. Averagely, 3 g of each yeast was rehydrated in an aliquot (30 ml) of the must to make a slurry and allowed to stand for about 10 min before being pitched with the fermentable must. The fermentation proceeded for 7 days at room temperature (25°C) in airtight high-density polyethylene (HDPE) plastic fermenters.

Wine clarification and fining

After seven days, the fermentation process was terminated by flocculating with a yeast cell agent (Bentonite). The bentonite slurry was prepared by mixing 3 tablespoons of the bentonite with a pint of warm water (45°C) in a saucepan per the manufacturer's protocol. Averagely, two tablespoons of bentonite slurry was added to five litres of the wine and stirred vigorously while adding the slurry. According to the manufacturer's protocol, the lees was allowed to settle for about 12 h. After 12 hrs, the wine sample was siphoned. The wine was racked twice at 12 hrs before bottling and maturation. Before the bottling and maturation, samples were taken for phytochemical analyses and consumer acceptability evaluations.

Maturation and compositional data collection

After the thorough fining of the wine, the samples were filled into sterilized glass bottles and tightly corked. The bottled wine samples

were then kept for three months in a dark cabinet in the Family and Consumer Science Food laboratory to mature. The data collection of this study was done in three phases: The first phase of data was obtained on the Young wine (before maturation) on the phytochemical contents, antioxidant characteristics and consumer acceptability. The second phase of data (phytochemical contents and antioxidant characteristics) was done when the wines were subjected to ageing for three months at room temperature (25°C), of which aliquots of the samples were drawn every 30 days' interval for phytochemical quantification. Finally, the last phase of data (phytochemical and consumer acceptability) was obtained after the maturation period (3 months).

Phytochemical determination

The phytochemical contents quantified were as follows: Phenolics, anthocyanins, flavonoids and antioxidant characteristics as described following. Also, the sensory attributes evaluated include colour, aroma, taste, mouthfeel and overall acceptability.

Determination of total phenolic content

The phenolic content of the wine was quantified using the Folin-Ciocalteu procedure published (Prior et al., 2005). About 0.5 ml of the wine samples was mixed with 0.5 ml of Folin-Ciocalteu phenol reagent (Sigma Chemical Co., St. Louis, Mo., U.S.A.) and 7.5 ml of deionized water. The mixture was held at room temperature for 10 min, and 1.5 ml of 20% sodium carbonate (w/v) was added. The mixture was then heated at 30°C in a water bath for 20 min and cooled in an ice bath before measuring the absorbance at 760 nm using the UV/Vis spectrophotometer (JENWAY). A standard curve of gallic acid (ranging from 0 to 100 mg/l) was prepared, and the results were determined from a regression equation;

$$(\text{phenolic concentration} = 0.0097x \text{ absorbance} + 0.0082; R^2 = 0.9977)$$

was then expressed as mg gallic acid equivalents per litre of wine (GAE/mg/g).

Determination of total flavonoid content

The total flavonoid content of the watermelon wine was quantified by a colourimetric method followed by Jia et al. (1999). Approximately 125 µl of the wine was added to 75 µl of 5% Sodium Nitrite (NaNO₂) solution, and the mixture was allowed to stand for 6 min. Afterwards, 150 µl of aluminium trichloride (10%) was added and then incubated for 5 min, followed by an addition 750 µl of NaOH (1 M). The final volume of the mixture was then adjusted to 2500 µl with distilled water. The mixture was allowed to incubate for another 15 min before the absorbance was read at 510 nm wavelength against a prepared blank using a spectrophotometer. The flavonoid content was determined using a standard curve of catechin (0 to 100 mg/l), and the results were determined from a regression equation;

$$(\text{Flavonoid concentration} = 0.007x \text{ absorbance} - 0.0016; R^2 = 0.9956),$$

was expressed as mg catechin equivalents per litre of wine (CTE/mg/g).

Total anthocyanin content determination

The collaborative study methodology published by Lee et al. (2005)

was followed to quantify the total anthocyanin content. The method employs pH differential on the structural change of the anthocyanin chromophore between pH values of 1.0 and 4.5. Anthocyanins have a maximum absorbance at a wavelength of 510 nm at a pH of 1.0. The coloured oxonium form predominates at a pH of 1.0, and the colourless hemiketal forms at a pH of 4.5. An aliquot of the wine (1ml) was added to 4 ml of buffer (KCl, 0.025 mol/l) with a pH of 1.0. The pH was adjusted with HCl (0.20 mol equi/l). One millilitre of the wine was also placed into a 10 ml volumetric flask, and 4 ml of the buffer (CH₃CO₂Na, 0.4 mol/l) with a pH of 4.5 was added. The pH was adjusted with HCl (0.20 mol equi/l). Absorbance was measured with a spectrophotometer at 510 and 700 nm. The total anthocyanin content (TAC) was calculated as the milligram equivalent of cyanidin 3-glucoside per litre as follows:

$$\text{TAC} = (A \times \text{MW} \times \text{DF} \times 100) / (\epsilon \times L) \times 10$$

Where, A = (A_{520 nm} - A_{700 nm}) pH 1.0 - (A_{520 nm} - A_{700 nm}) pH 4.5; Molecular weight (MW) = 449.2 g/mol; Dilution factor (DF) = 10; L (path length in cm) = 1; ϵ (molar extinction coefficient) = 26,900 l/mol cm

Determination of antioxidant activity-DPPH assay

The antioxidant activity of the wine was determined using the 1, 1-diphenyl-2-picrylhydrazyl (DPPH) assay described by Yu et al. (2002). Approximately 1 ml of the wine sample each was mixed vigorously with 3 ml of 0.06 μ M DPPH dissolved in methanol and was incubated at 25°C in darkness for 20 min instead of the 60 min as used in the stated protocol. Afterwards, the decrease in the absorption of DPPH after the wine sample was measured at 517 nm instead of 515 in the stated protocol. The DPPH radical scavenging activity (RSA) was calculated as follows:

$$\% \text{ RSA} = \frac{[\text{Abs blank} - \text{Abs sample}]}{\text{Abs blank}} \times 100$$

Where; Abs sample is the absorbance of the sample (wine), and Abs blank is the absorbance of the control (Distilled water)

Total antioxidant capacity determination

The antioxidant capacity of the treatments were determined using the protocol by Prieto et al. (1999). First, an aliquot of the watermelon wine (500 μ l, diluted at 1:20) was combined with 5 ml of reagent solution (0.6 M H₂SO₄, 28 mM NaH₂PO₄, and 4 mM ((NH₄)₂MoO₄) in a 15 ml test tube. The reaction mixture was incubated at 95°C for 90 min and then cooled at room temperature, and absorbance was measured at 695 nm. The total antioxidant capacity (TAC) was expressed as tannic acid equivalent (TAE).

Sensory evaluation of watermelon wine

Sensory analysis was conducted on wine samples from Lalvin EC 1118 wine, Lalvin QA 23 wine, Redstar premier rouge wine and Redstar premier classique wine following fermentation and ageing. The sensory evaluation was conducted on the following attributes; aroma, colour, taste, mouthfeel (astringency) and overall acceptability using a five-point hedonic scale described by Stone and Sidel (2004) and shown on the scale as 5 representing like very much, 4 representing like, 3 representing neither like nor dislike, 2 representing dislike and 1 representing dislike very much. Fifty-six (56) untrained panellists familiar with wine were used for the sensory evaluation. The untrained panellists were between 18 and 45 years comprising students (50) and staff (6) at the

Nyankpala campus.

Statistical analysis

The data obtained was subjected to a two-way analysis of variance (ANOVA) using Genstat eighteenth edition and Tukey Pair Wise comparison used for mean separation at the confidence level of $p < 0.05$.

RESULTS AND DISCUSSION

Phytochemical characteristics of watermelon wine

The phytochemical characteristic of wine is of great concern in enology because of its contribution to clinical studies aside from its sensory influence. The watermelon wine's phytochemical characteristics include total phenolics, anthocyanins, flavonoids, and antioxidant activity quantified before and after the maturation/ageing (Table 1).

Total phenolic content

The total phenolic content of the watermelon wine varied significantly ($p=0.003$) between treatments in both the young (sample after 7 days of fermentation) and the matured (90 days after initial fermentation) wine. In the young wine, the total phenolic content was statistically higher in the Redstar premier classique sample (105.33 μ g/g) and lower in the Lalvin EC 1118 (98.69 μ g/g). Similar phenolic concentrations were also recorded in the matured wine samples, with the least concentration of the phenolics in the Lalvin EC1118 (156.08 μ g/g) and the highest in Redstar premier classique (275.99 μ g/g). The maturation process impacted the concentration level of the phenolic content in all the treatments (Table 1). The phenolic concentration in the young wines was lower than the phenolic concentrations recorded in the matured wine samples.

This observation was contrary to what was reported by Baiano et al. (2016), that mostly phenolic concentration diminished with oak chip treatment in the course of maturation. Other authors reported that low evolution of anthocyanins and tannins during ageing decreases total phenolic concentration (Tzachristas et al., 2020). However, the samples were completely devoid of anthocyanins (Table 1). The Redstar premier classique and Redstar premier rouge samples recorded significantly higher concentrations in the young and matured wine samples (Table 1). The increase in phenolic concentrations could be linked to the evolution of additional compounds from the oak chips used for wine maturation (Jean-Louis et al., 2020). This disparity in phenolic contents confirms the earlier research that alcoholic medium extracts phytochemicals better (Mojzer et al., 2016).

Table 1. Phytochemical characteristics of watermelon wine.

Sample	Parameter of young and Aged wines									
	Total phenols content (GAEmg/g)		Total anthocyanins content		Total flavonoids content (CTE/mg/g)		%Antioxidant activity		%Antioxidant capacity	
	Young	Aged	Young	Aged	Young	Aged	Young	Aged	Young	Aged
Lalvin EC 1118	98.68 ^b	156.08 ^d	0.00	0.00	52.4	34.72	13.29 ^b	79.14 ^d	1914.02 ^a	2219.37 ^a
Lalvin QA 23	93.36 ^c	186.49 ^c	0.00	0.00	36.75	29.00	13.19 ^c	174.49 ^c	1341.29 ^c	1752.71 ^b
Redstar premier rouge	98.71 ^b	212.01 ^b	0.00	0.00	36.34	41.34	1.28 ^d	181.35 ^b	1793.57 ^b	808.62 ^d
Redstar premier classique	105.33 ^a	275.99 ^a	0.089	0.00	35.29	58.38	13.59 ^a	246.72 ^a	1279.93 ^d	1634.43 ^c
P - value	0.003	0.0001	0.065	0.945	0.184	0.108	0.0001	0.0001	0.0001	0.0001

*Mean values with, different superscripts in the same columns are significantly different.
Source: Authors

Total anthocyanins content

In the young wine, anthocyanins concentration was not detected in all the wine samples except in Redstar premier classique wine sample that recorded a minute concentration (0.089). However, anthocyanin was not detected in the matured wine samples. The absence of anthocyanins could be attributed to the report that yeast cells can adsorb anthocyanins during wine fermentation (Morata et al., 2019). Therefore, treatment did not show a significant difference in the young wines (p=0.065) and the matured/aged samples (p=0.945), respectively, as presented in Table 1 for the anthocyanins concentration.

Total flavonoids content

The mean concentrations of the flavonoids recorded in the young wines were 35.29 in the Redstar premier classique, 52.48 in the Lalvin EC 1118, 36.75 in the Lalvin QA 23 and 36.64 in that the Redstar premier rouge. After the maturation phase, the flavonoids quantified in the wines after the maturation period revealed that the least

mean flavonoids concentration was observed in the Lalvin QA 23 (29.00), while Redstar premier classique (58.38) recorded the highest mean for total flavonoids content. However, Lalvin EC 1118 and Redstar premier rouge recorded 34.72 and 41.34 flavonoids concentration, respectively. The quantified flavonoid content appeared to correlate with the anthocyanins content in the watermelon wine samples. Generally, the flavonoid concentration was low in the watermelon wine samples (Mariya et al., 2020). The flavonoid concentration quantified in watermelon wines could be due to much of the flavonoids concentrated in the rind rather than the pulp since most of the polyphenols are localized in the skin of fruits, as reported by Mariya et al. (2020). Treatment Lalvin EC 1118 and Lalvin QA 23 samples recorded reduced flavonoid content after storage. This is in line with the report of Bala and Kocher (2017), who recorded a decrease in phenolic content in muskmelon wine during bottle storage. However, the trend was the opposite of the Redstar premier rouge and Redstar premier classique samples. Although this opposite flavonoid concentration pattern may be attributed to the yeast species involved, it was noticed

that the reduction in the flavonoid concentration was recorded in the Lalvin species, while the Redstar premier species recorded an increment in flavonoid concentrations.

Antioxidant activity and capacity

The scavenging activity of the watermelon wines showed a significant difference (p=0.001) in both the young and matured wine samples (Table 1). The study revealed an increase in the percentage inhibition activity of the treatment as the samples matured for 90 days (3 months). The least percentage scavenging activity in the young wine was experimentally noticed in the Redstar Premier rouge wine sample (1.28), while the Redstar premier classique recorded the highest potential free radical scavenging activity of 13.59. The percentage scavenging activity of Lalvin EC 1118 (13.59) was statistically higher than that of the percentage scavenging activity recorded in the Lalvin QA 23 (13.19). Again, in the matured samples, Redstar premier classique still recorded the highest percentage of inhibition activity (246.72), followed by the Redstar premier rouge

(181.35) and then the Lalvin QA 23 (174.49), while the least recorded in the Lalvin EC 1118 (79.14). It was indicating that maturation is a significant factor influencing the level of radical scavenging activity in the watermelon wine samples. Generally, the antioxidant activity improved in the young wines from 1.28 to 13.59 to 79.14 to 246.72% after the maturation period (90 days). An increase in the radical scavenging activity observed in the matured wine samples could be attributed to the concentration of the phenolic compounds in the wines. This observation supports reports that wine scavenging ability depends on its phenolic content (Bertelli et al., 2021; Innocent and Matenda 2018).

The samples' antioxidant capacity, which measures the thermodynamic conversion efficiency of an oxidant probe upon reaction with an antioxidant, of the samples was significantly different ($p=0.001$) among the treatments. In the young wine samples, the lowest mean antioxidant capacity was observed in Redstar premier classique (1279.93), while the highest mean percentage (1914.02) antioxidant capacity was recorded in Lalvin EC 1118 sample. The antioxidant capacity of the Redstar premier (1793.57) was significantly higher than that of the Lalvin QA 23 (1341.29). Accordingly, the Lalvin EC 1118 sample retained the greatest mean percentage (2219.37) of antioxidant capacity, while Redstar premier rouge recorded the least mean antioxidant capacity percentage (808.62) in the aged wines. Generally, the percentage antioxidant capacity increased significantly in all the samples across the maturation time (Table 1), indicating maturation time impacted the antioxidant capacity of the wine samples. This increment in antioxidant capacity could be due to adding oak chips during maturation. This affirms the report by Jean-Louis et al. (2020) that extracts from oak (phenolic compounds) increase the antioxidant capacity of matured wines.

Consumer acceptability of the watermelon wine

The results of the sensory attributes of wines produced from watermelon fruit are shown in Table 2. The wine samples of both young and matured (aged) were evaluated. Interestingly, the treatments did not show any significant difference ($p > 0.05$) in the sensory attributes evaluated (Table 2). The sensory attributes evaluated on the wine were colour, aroma, taste, mouthfeel and overall consumer acceptability.

Colour

The mean rank of the measured sensory attributes on the young wine samples did not show a significant difference from each other. However, Redstar premier classique sample ranked the highest (63.71), the next ranked sample was the Lalvin QA23 (60.00), followed by Lalvin EC 1113 (52.00), and the Redstar premier rouge recorded

the least rank (50.29). However, considering the matured samples, interestingly, Redstar premier rouge sample scored the maximum (63.11), Lalvin EC1118 ranked second best (60.29), the third highest scored sample was Redstar premier classique (52.50), and the Lalvin QA 23 (50.11) ranked the least in terms of colour for the matured samples. The maturation period, therefore, influenced the sensory characteristics of the wine samples. From the panels' choice of colour among the young wines, Lalvin EC 1118 and Redstar premier rouge were preferred compared to the Lalvin QA 23 and Redstar premier classique. However, the opposite happened after the wines matured; thus, Redstar premier rouge and Lalvin EC 1118 were the most preferred. The colour improvement in the Redstar premier rouge and Lalvin EC 1118 samples agreed with other research reports that sensory properties improve with maturation when exposed to oak products (Tchabo et al., 2017).

Aroma

The mean values obtained for the aroma of young wine samples ranged between 51.93 to 61.95. Though no significant difference was observed in the mean rank of aroma in the young wines ($p=0.0692$), Lalvin QA23 recorded the highest mean rank aroma (61.95), followed by Redstar premier classique (56.80), Redstar premier rouge (55.68) and Lalvin EC 1118 (51.90). Similarly, the aroma concentrations of the samples were not affected by maturation time since there were no significant differences in the mean values of aroma among the samples ($p=0.253$). However, the Redstar premier rouge scored higher (62.98) in terms of an aroma than the Redstar premier classique (59.21), followed by Lalvin EC 1118 (56.82), and the lowest rank was recorded in the Lalvin QA 23 (46.98). The aroma of the wines was similar irrespective of the treatments within the young wines and after maturation. The similarities in the aroma of the wine samples may result from little to no variation in the yeast strains used for the fermentation.

Taste

In the young wine samples, the Redstar premier rouge was ranked first (60.50), followed by Lalvin QA 23 (55.89), Redstar premier classique (55.07) and Lalvin EC 1118 (54.54). For the matured wine, Redstar premier rouge was ranked the highest (64.23), followed by Redstar premier classique (59.32), Lalvin EC 1118 (53.59), and Lalvin QA 23 (48.86) in that order. Maturation factor did not affect the taste of the wine samples because changes in taste mean ranks were almost the same for both young and aged wine. This observation could be linked to the negligible change in apparent °Brix measured during the maturation period. It is discernible that a change in apparent °Brix could

Table 2. Sensory attributes of watermelon wine.

Sample	Means ranks of sensory attributes									
	Colour		Aroma		Taste		Mouthfeel		Overall acceptability	
	Young	Aged	Young	Aged	Young	Aged	Young	Aged	Young	Aged
Lalvin EC 1118	52.00	60.29	51.93	56.82	54.54	53.59	56.20	52.77	47.80	57.02
Lalvin QA 23	60.00	50.11	61.59	46.98	55.89	48.86	49.11	54.96	57.50	52.54
Redstar premier rouge	50.29	63.11	55.68	62.98	60.50	64.23	61.55	67.12	57.11	66.02
Redstar premier classique	63.71	52.50	56.80	59.21	55.07	59.32	59.14	51.14	63.59	50.43
P - value	0.247	0.334	0.692	0.253	0.880	0.264	0.436	0.206	0.275	0.225

*Mean values with the same superscripts in the same columns are not significantly different.
Source: Authors

directly affect the taste of the sample. The maturation time did not greatly influence the taste due to the stability of the ethanol concentration in the samples throughout the maturation period. This observation was earlier reported by Villamor et al. (2013) that the taste of wine could be influenced by its alcohol concentration instability.

Mouthfeel (astringency)

The Redstar premier rouge sample was ranked highest (61.55) as compared to the mean ranks of Redstar premier classique (59.14), Lalvin EC 1118 (56.20) and Lalvin QA 23 (49.11) among the young wine samples as presented in Table 2.

However, astringency in the wine samples decreased after maturation, indicating maturation factor influenced wine astringency content. The astringency level increased in Redstar premier rouge (67.12), followed by Lalvin QA 23 (54.96). However, the astringency levels of Lalvin EC 1118 (52.77) and Redstar premier classique (51.14) declined after the samples matured and hence ranked the lowest (Table 2). Changes in the individual phytochemical compound compositions could explain the instability in the astringency.

Overall acceptability

The mean values for consumer acceptability of young wines ranked from 47.80 to 63.59. Even though the overall acceptability revealed similarities in the sensory characteristics among the wine samples, the Redstar premier classique was ranked the most accepted sample (63.59), followed by Lalvin QA 23 (57.50), Redstar premier rouge (57.11) and the Lalvin EC 1118 (47.80) before maturation. In the matured samples, the Redstar premier rouge was ranked most preferred (66.02), followed by Lalvin EC 1118 (57.02), Lalvin QA 23 (52.54), and Redstar premier classique (50.43). The panelists' choice of the samples at the young and mature stages was the Redstar premier classique ranked highest among the samples.

Conclusion

The study significantly revealed that watermelon is a potential tropical fruit for winemaking based on its high moisture and sugar content. The research further showed that the phytochemical characteristics of the yeast strains exhibited

different patterns during maturation. The phytochemical concentrations were generally higher in the matured wine samples compared to the young wine samples indicating that adding oak chips improves phytochemical contents during maturation. The research findings also showed that anthocyanin was not detected in the watermelon wine samples except the Redstar premier classique, which recorded negligibly low concentration in the young wine. The flavonoid concentrations in the wines were also low, both in the young and matured wines.

The wine samples showed great potential for free radical scavenging and antioxidant activity with the artificial radical reagent (DPPH). The antioxidant activity of the matured wine samples, regardless of the yeast strain, increased as a function of maturation time. The antioxidant capacity of the watermelon wines generally increased in the matured wines except in the Redstar premier rouge, which drastically declined. However, the Lalvin EC 1118 recorded the highest antioxidant capacity among the young and matured wine samples. The antioxidant activity of the matured wines improved due to the addition of oak chips as the main agent for the maturation of the wine samples in the glass bottles.

From the data obtained, it can be concluded that any of the four commercial yeast strains used in this study was suitable for making watermelon wine. However, it manifested that the Redstar premier classique produced the most desirable results, followed by the Lalvin EC 1118 yeast strains. Furthermore, the antioxidant activity and capacity revealed the clinical potential of the watermelon wines.

CONFLICT OF INTERESTS

The authors declared that they have no conflict of interest.

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