

*Full Length Research Paper*

## **Evaluation of starter culture fermented sweet potato flour using FTIR spectra and GCMS Chromatogram**

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**Starch is the major component of cereal grains and starchy foods, and changes in its biophysical and biochemical properties (such as, amylose, amylopectin, pasting, gelatinization, viscosity) will have a direct effect on its end use properties (such as, bread, malt, polymers). *Lactobacillus brevis* and *Debaromyces polymorphous* earlier obtained from fermented sweet potato broth were used to ferment sweet potato and these starter cultures broke down the carbohydrate (starch) to produce alcohol, organic acid and carbon dioxide (CO<sub>2</sub>). The study identified that starter cultures *L. brevis* and *D. polymorphous* fermented the sweet potato thereby breaking down the carbohydrate (starch) to produce alcohol, organic acid and CO<sub>2</sub> hence lactic acid fermentation occurred. Fourier Transform Infrared Spectroscopy (FTIR) and Gas Chromatography Mass Spectrometry (GCMS) were used to identify the chemical properties of starter culture fermented sweet potato flour. The FTIR spectra showed peaks at 3322.15, 3298.87, 3292.59, 3279.59 and 3274.59 cm<sup>-1</sup> for the raw sweet potato, starter culture fermented sweet potato flour at various periods (24, 48 and 72 h) and spontaneous fermented sweet potato (control) respectively. The peaks at 2930, 2928.10, 2930.33, 2929.48, 2929.31 and 2927.29 cm<sup>-1</sup> are attributed to C–H bond stretching. Functional groups such as hydroxyl, aldehydes, alcohol and carboxyl were detected in the fermented samples. The GCMS analysis detected the presence of alcohol such as ethanol, butanol etc., and carboxylic acid such as hexadecanoic acid, octadecadienoic acid etc. They were produced *in situ* from the fermentation process and this can serve as antioxidants, help inhibit spoilage organisms and serve as preservatives, thereby increasing shelf life of the product.**

**Key words:** Sweet potato starch, fermentation, FTIR, GCMS.

### **INTRODUCTION**

Carbohydrate is a class of chemical compounds that consists of carbon, oxygen and H<sub>2</sub> (Kim et al., 2007). It

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includes sugars, starch and cellulose. These compounds are classified as monosaccharides (such as, glucose, fructose), disaccharides (such as, sucrose, lactose) or polysaccharides (such as, starch, cellulose) (Kim et al., 2007). All carbohydrate compounds have been used as a source of biomass and a large number of microorganisms use them as energy source as a result, carbohydrates are essential for maintaining life (Kim et al., 2007). Glucose is the key compound as most life systems are built around it.

Starch is a carbohydrate consisting of glucose compounds joined to form a polysaccharide (Dias et al., 2008). It is a plant natural energy source that is most abundant and valuable which needs to be converted to simple sugars before it can be utilized as a carbon source (Yoo and Jane, 2002; Mosier et al., 2005; Yang et al., 2006; Gray et al., 2006).

The starch stored in the seeds and tubers of various agricultural crops including maize, wheat, rice, barley, potato and cassava provide the main sources of energy in the human diet (Evers et al., 1999; Perez et al., 2009; Perez and Bretoft 2010, Schwartz and Whistler, 2009). Starch is the major component of cereal grains, and changes in its biophysical and biochemical properties are related to the amount and ratio of amylose and amylopectin, which influences and affect properties such as viscosity, gelatinization, that will determine its end use properties (such as, bread, malt, beer, polymers) (Evers et al., 1999; Schwartz and Whistler, 2009; Willett, 2009).

Starch functions mainly as a carbohydrate source for the growing plant (such as, for germinating seeds and leaf tissue development) and is consequently the primary source of stored energy in the plant. Depending on the plant, starch can be found in a variety of tissues, including leaves, tubers, fruits, and seeds. It is the primary source of stored energy in cereal grains. Although the amount of starch contained in grains varies, it is generally between 60 and 75% of the weight of the grain and provides 70 to 80% of the calories consumed by humans worldwide.

It consists of two  $\alpha$ -glycan bipolymers, namely, amylose and amylopectin (Yang et al., 2006; Dias et al., 2008; Shariffa et al., 2009). Amylose is a more linear glucose polymer consisting of 200 to 20000 glucose units forming a helix shape; while amylopectin is a highly branched molecule of 10-15 nm in diameter and 200-400 nm long (Yoo and Jane, 2002; Yang et al., 2006; Shariffa et al., 2009).

Amylopectin consist of D-glucopyranose monomers linked to either  $\alpha$ -(1,4) or  $\alpha$ -(1,6) glucosidic bonds (Yang et al., 2006). The joined monomers of  $\alpha$ -(1,4) results in a linear chain; however,  $\alpha$ -(1,6) bond serves as a glue that joins together the linear chains (Yang et al., 2006). Amyloses consist of linear glucan connected via  $\alpha$ -(1,4) bonds (Lesmes et al., 2009). Starches contain about 17 to 28% of amylose (Matveev et al., 2001). Microbial enzymes (Wang et al., 2008) easily hydrolyze these bonds.

Root vegetables are plant roots used as vegetable, they are generally storage organs enlarged to store energy in form of carbohydrates, starch root vegetable

are important staple food particularly in tropical regions overshadowing cereals throughout much of west Africa, Central Africa, they are used directly or mashed to make fufu or poi. Storage roots can be categorised in bulb, rhizome and tubers. Examples of tuberous root include desert yam (*Ipeoma costata*), sweet potato (*Ipeoma batatas*), cassava (*Manihot esculenta*), etc. Sweet potato carbohydrate has been reported to contain pectin substances, lignin, cellulose and hemicellulose, which are all converted to simple sugars when fermented (Yokoi et al., 2001).

Sweet potato has been processed into prickles and consumed as lacto-juices by processing it with lactic acid bacteria as the fermenting organism and the juice produced has been reported to be very rich in minerals and vitamins (Smita et al., 2007; Panda and Ray, 2007). Sweet potato has also been processed into chips in much the same way as Irish potato (Brigato et al., 2010; Hagenimana and Owori, 1998). It can also be eaten boiled, fried and in roasted form. In addition, it can be sliced, dried in the sun and ground to give flour that remains in good condition for a long time (Wheatly, 2009). Sweet potato can be fermented, dried and milled into flour. Fermentation is the conversion of carbohydrate into alcohols and short chain fatty acids by enzymes of microorganisms (Silva et al., 2008; Yuan et al., 2008). It is a basis of many biological products which involves a process of chemical reactions with the use of microbes such as bacteria, yeast and filamentous fungi (Huang and Tang, 2007; Fortman et al., 2008). The primary benefit of fermentation is the conversion of sugars and other carbohydrates to usable products. As stated earlier, not all bacteria can readily use starch as their energy and carbon source (Nigam and Singh, 1995). This means that some starches need to be broken down to simple fermentable sugars so they can be utilized by bacteria (Nigam and Singh, 1995). On the other hand, sweet potato is rich in  $\beta$ -amylase, which converts long chained starch into readily used maltose units making it a good energy and carbon source for bacteria (Yoshida et al., 1992; Brena et al., 1993; Cudney and McPherson, 1993; Nigam and Singh, 1995). Starter cultures are living microorganisms of defined combination used for fermentation purposes. They help to elicit specific changes in the chemical composition, nutritional value and sensorial properties of the substrate (Opere et al., 2012) and they are generally recognised as safe (Aguirre and Collins, 1993). Moreover, their properties are as follows: they are harmless, initiate and control the fermentation process, typical for product, help in rapid acid formation, and help protect against spoilage organisms. Starter cultures are cheaply reproducible in large amount; they also help provide desirable sensory properties and assist in reducing fermentation period. Ajayi et al. (2016) have done work on the fermentation of sweet potato into flour using starter culture.

Fourier transforms infrared (FTIR) spectroscopy is one of the most important and emerging tool used for analysing functional groups present in test samples. This technique is rapid and sensitive with a great variety of sampling techniques. FTIR is a rapid, non-

destructive, time saving method that can detect a range of functional groups and is sensitive to changes in molecular structure. FTIR provides information on the basis of chemical composition and physical state of the whole sample (Cocchi et al., 2004). In addition the sensitivity and accuracy of FTIR detectors along with wide variety of software algorithms have dramatically increased the practical use of infrared for quantitative analysis (Dowell et al., 2006). FTIR works because of functional groups and provide information in the form of peaks.

GCMS combines the features of gas chromatography and mass spectrometry to identify different substances within a test sample. Gas chromatography portion separates the chemical mixture into pulses of pure chemicals and the mass spectrometer identifies and quantifies the chemicals. It reveals the compounds eluted at different retention times with mass spectra corresponding to compounds present (Siong et al., 2014).

This study aimed at investigating the functional groups of the Starter culture fermented sweet potato flour using Fourier transform infrared (FTIR) spectroscopy as well as reporting the effect of the functional group on the products, the chemical compounds present in the starter culture fermented sweet potato flour will be detected using the GCMS.

## MATERIALS AND METHODS

### Sourcing of raw materials

Yellow-fleshed sweet potatoes were obtained from Oshodi market (Oshodi), Lagos, Nigeria. The samples were transported to the biotechnology department of the Federal Institute of Industrial Research for immediate use.

### Starter cultures

The starter cultures used were obtained from the biotechnology department of the Federal Institute of Industrial Research Oshodi.

Potato dextrose agar and De man Rogosa Sharpe (MRS) agar were prepared using manufacturers specification and sterilized using the autoclave at 121°C for 15 min. *L. brevis* and *D. polymorphous* stored in MRS and PDA slants were subcultured into freshly prepared MRS and PDA agar plates.

### Preparation of inoculum

This was carried out using the method of Asmahan et al. (2009). *Lactobacillus brevis* were cultivated by streaking on MRS agar plates (Oxoid) and incubated anaerobically at 37°C for 24 h. A colony was picked from each pure culture plate, grown successively in MRS broth before centrifugation at 5000 rpm for 15 min. The pellet was washed in sterile distilled water centrifuged again and redistributed in distilled water. This procedure achieved a culture preparation containing about  $10^9$  colony forming units cfu/ml, checked as viable count on MRS agar. Pure cultures of *D. polymorphous* were cultivated by streaking on Potato dextrose agar (Oxoid), incubated at 37°C for 24 h and the picked colony was inoculated into yeast extract peptone dextrose broth (YEPD) and incubated at 28°C for 24 h. These cultures were centrifuged and washed as described above. This procedure achieved a culture preparation containing  $10^7$  cfu/ml, as viable count on potato dextrose agar.

### Preparation of starter culture fermented sweet potato flour

The sweet potatoes were washed to remove adhering soil particles and peeled. The peeled tubers were chipped into slices (4-5 mm). Starter cultures were prepared and inoculated into the sweet potato, then left to ferment for a period of two days (48 h).

After this period has elapsed, the fermented chips were drained and dried in a cabinet drier (Mitchel, Model SM220H) at 55°C for 9 h and milled into flour ( $\leq 250 \mu\text{m}$ ) using the method of Ajayi et al. (2016) and the starter culture fermented sweet potato flour was produced.

### Preparation of fermented sweet potato flour (control)

The sweet potatoes were washed to remove adhering soil particles and peeled. The peeled tubers were chipped into slices (4-5 mm) and soaked in potable water for a period of two days (48 h).

After this period has elapsed, the fermented chips were drained and dried in a cabinet drier (Mitchel, Model SM220H) at 55°C for 9 h and milled into flour ( $\leq 250 \mu\text{m}$ ) (Oluwole et al., 2012).

### Fourier Transform Infrared (FTIR)

FTIR spectra illustrate absorption bands with characteristic frequency attributed to different functional groups and all spectra were obtained, using a Bruker FTIR CLASS 1 ALPHA. The spectra were collected at a resolution of  $4 \text{ cm}^{-1}$  in the range of 500- 4000 $\text{cm}^{-1}$ . Each spectrum was ratiomed against a fresh background spectrum recorded from the bare crystal. Prior to collection of each background spectrum, the crystal was cleaned with absolute ethanol to remove any residual. Each sample was scanned in triplicate.

### Gas chromatography mass spectrometry (GCMS)

Ten grams of samples were dissolved in 15ml of ethanol. The sample was analysed on a Shimadzu GC-MS system model QP2010, with a medium polarity capillary column SLB-5ms supelco column (length 30.0m x thickness 0.20 mm x Diameter 0.20 mm), with helium as the carrier gas. Column oven temperature was at 40°C, injection temperature was at 250°C, injection mode split (10:1), temperature program was 40°C (Hold 3 min) 9°C/min to 290°C (Hold 6mins), MS ion source temperature at 200°C interface temperature at 250°C. Detector voltage = Relative to the tuning result solvent cut time 4min, Acquisition mode-scan, scanning range 40-550 m/z. One microlitre of the sample was injected using splitless injection with injector temperature 300°C according to the following scheme: 50°C for 2min with 10°C/min up to 300°C. The final temperature was held for 10 min. The total runtime for each sample was 37min. For MS detection, electron ionization with 70 eV was applied and mass fragments were detected between 40 and 500 m/z. The ion source temperature and transfer line temperature were 200°C and 300°C, respectively. The detector was activated after 5min.

## RESULTS

The FTIR Spectra were recorded in regions below 800  $\text{cm}^{-1}$ , 500  $\text{cm}^{-1}$  (the fingerprint region), the region between 2,800 and 3,000  $\text{cm}^{-1}$  (C-H stretch region), and finally the region between 3,000 and 3,600  $\text{cm}^{-1}$  (O-H stretch region) (Table 1).

The O-H stretching for the raw sweet potato occurred at 3322.15  $\text{cm}^{-1}$ . The peaks at 2928.10  $\text{cm}^{-1}$  was observed as a result of C-H bond stretching. The peaks at 1097  $\text{cm}^{-1}$

Table 1. GCMS Peak report for raw sweet potato (G).

Peak #	R Time	Area	Area %	Height	Height (%)	A/H	Name
1	5.720	1027541	0.27	220079	0.64	4.67	Pyrimidine -2,4(1H,3H) -dione, 5-amino-6-nitro
2	6.268	3010757	0.80	1092925	3.19	2.73	2 -Furanmethanol
3	6.556	677120	0.18	262700	0.77	2.58	Propanoic acid, 2- oxo
4	6.802	2180121	0.58	454271	1.32	4.80	Cycloserine
5	7.150	1176657	0.31	484808	1.41	2.43	Dihydroxyacetone
6	7.406	881086	0.24	487437	1.42	1.81	Glyceraldehyde
7	7.440	2679476	0.72	565822	1.65	4.74	Ethanamine, N- ethyl- N- (1-methylethoxy)meth
8	7.726	112304	0.30	540485	1.58	2.08	6-Oxa-bicyclo[3.1.0] hexan-3-one
9	8.793	324740	0.09	178905	0.52	1.82	4H-pyran-4-one, 2,3- dihydro-3, 5- dihydroxy -6-
10	9.175	8645942	2.31	923865	2.69	9.36	2 -Hydroxy -gamma- butyrolactone
11	9.518	1320939	0.35	234913	0.69	5.62	2-propanol, 1- chloro- 3- (1- methylethoxy
12	9.734	482881	0.13	157144	0.46	3.07	Tetrahydro-4H- pyran-4-ol
13	10.015	492607	0.13	245929	0.72	2.00	Esprocarb
14	10.198	2912455	0.78	707736	2.06	4.12	1, 3-Dioxol-2-one
15	10.390	555860	0.15	167491	0.49	3.32	(3-Methyl-oxiran-2-yl)-methanol
16	10.498	2298739	0.61	666881	1.95	3.45	2,5-Dimethyl-4-hydroxy-3(2H)-furanone
17	10.552	557914	0.15	225743	0.66	2.47	6,7-Dioxabicyclo[3.2.2] nonane
18	10.818	2540952	0.68	662644	1.93	3.80	Cyclopentane, 1-acetyl-1,2-epoxy-
19	11.049	5259236	1.41	816451	2.38	6.44	Cyclopropyl carbinol
20	11.432	253776	0.07	82742	0.24	3.07	Homopiperazine
21	11.592	542159	0.14	161569	0.47	3.36	Hexane, 1, 1- oxybis
22	11.725	2355125	0.63	971901	2.83	2.42	Pentanoic acid, 4- oxo-
23	11.938	7737745	2.07	2812686	8.20	2.75	4H- Pyran-4- one,-2,3- dihydro-3,5- dihydroxy
24	12.203	3353258	0.90	475206	1.39	7.06	2(3H) Furanone, dihydro-4- hydroxy-
25	12.759	1974174	0.53	565909	1.65	3.49	Catechol
26	12.863	1769021	0.47	479487	1.40	3.65	Butanenitrile, 2,3- dioxo-, dioxime, O, O - diacet
27	13.168	1356627	0.36	311275	0.91	4.36	2- Furanmethanol,tetrahydro-5-methyl-
28	13.306	10515375	2.81	3280635	9.57	3.20	5-Hydroxymethylfurfural
29	13.596	1615272	0.43	388747	1.13	4.16	1,2,3- Propanetriol, 1- acetate
30	13.793	1298808	0.35	374957	1.09	3.46	Dimethylmucanoic acid
31	14.936	67094767	17.93	2900347	8.46	23.13	Sec - Butylnitrite
32	17.641	76203093	20.37	2020814	5.89	37.71	Sucrose
33	18.306	2901986	0.78	295502	0.86	9.82	Butyl 2- acetoxycetate
34	17.597	51846108	13.86	3099727	9.04	16.73	1,6-Anhydro-2,4-dideoxy-beta-D-ribohepoxy
35	20.113	87200037	23.30	2246302	6.55	38.75	3- Deoxy- d- mannoic acid
36	23.010	5199882	1.39	1042633	3.04	4.99	n - Hexadecanoic acid
37	23.151	703283	0.19	425723	1.24	1.65	Scopoletin
38	23.344	465000	0.12	310018	0.90	1.50	Hexadecanoic acid, ethyl ester
39	24.832	4985304	1.33	913118	2.66	5.44	9, 12- Octadecadienoic acid (Z, Z) -
40	25.102	605346	0.16	264178	0.77	2.29	12 - Methyl- E- 2, 13-octadecadien-1- ol
41	26.656	4610547	1.23	1353688	3.95	3.41	1-Benzoyl-2-t-butyl-3-methyl-5-vinylimidazoli
42	28.399	304422	0.08	105130	0.31	2.90	6H - Pyrazolo [1, 2-a] [1,2,4,5] tetra-zine, hexahydr
43	28.557	188310	0.05	98405	0.29	1.91	Hexanol
44	29.915	715744	0.19	131722	0.38	5.43	Z, Z- 10, 12- Hexadecadien- 1- ol acetate
45	34.066	238334	0.06	76775	0.22	3.10	alpha- Tocopheryl acetate
		374181566	100.00	34285425	100.00		

and 1019  $\text{cm}^{-1}$  were assigned as the C–O bond stretching (Table 2). This indicates that compounds belonging to hydroxyl group, hydrocarbon and aldehydes group are present in the raw sweet potato.

GCMS peak report reveals the compounds eluted at different retention times with mass spectra

corresponding to compounds present. Figure 1 shows the GCMS chromatogram of raw sweet potato sample from 5.72 to 34.066 s. It showed 45 peaks, compounds such as pyrimidine-2,4(1H,3H) dione was detected with a retention time of 5.72 s and an area percentage of 0.27%, propanoic acid had the retention time of 66.556

**Table 2.** GCMS Peak report for starter culture fermented sweet potato flour 24 h (H).

Peak #	R. Time	Area	Area %	Height	Height %	A/H	Name
1	5.043	407371	0.54	253520	2.70	1.61	Fornamide, N - mmet hoxy-
2	5.245	316923	0.42	178489	1.90	1.78	(S)-(+)-1, 2- Propanediol
3	7.118	23036722	30.71	1491815	15.87	15.22	L - Lactic acid
4	9.032	2009458	2.68	228988	2.44	8.78	1,2,3,4 - Butanetetrol, [S-(R*,R*)]-
5	11.789	402293	0.54	175872	1.87	2.29	4H- Pyran-4-one, 2,3-dihydro-3,5- dihydroxy-6-
6	13.238	416022	0.55	197832	2.10	2.09	5- Hydroxymethylfurfural
7	14.241	20559512	27.40	1087754	11.57	18.90	Oxirane,(propoxymethyl)-
8	15.175	189501	0.25	94701	1.01	2.00	Phenol,2,6- dimethixy
9	16.340	322572	0.43	161965	1.72	1.99	Benzeneethanol
10	16.679	3480912	4.64	375027	3.99	9.28	1,3-propanediol, 2-(hydroxymethyl) -2-nitro-
11	17.373	85920	0.11	43809	0.47	1.96	Propanediol acid, propyl-
12	17.789	354609	0.47	50080	0.53	7.08	Erythritol
13	18.162	294498	0.39	104520	1.11	2.82	n- Decanoic acid
14	18.558	1777004	2.37	285223	3.03	6.23	Diethyl Phthalate
15	18.999	1142058	1.52	201352	2.14	5.56	D -erythro - Pentose, 2- deoxy-
16	23.025	6648837	8.86	1508499	16.05	4.16	n -Hexadecanoic acid
17	24.357	170486	0.23	83154	0.88	2.05	Decanoic acid, ethylester
18	24.848	6196913	8.26	1082209	11.51	5.93	9,12 - Octadecadienoic acid (Z,Z)-
19	25.123	2650278	3.53	364339	3.88	6.86	Octadecanoic acid
20	26.295	79665	0.11	40134	0.43	1.98	Pentanal
21	26.632	319090	0.43	125222	1.33	2.55	2,4-Di-tert- butyl phenyl benzoate
22	26.730	327804	0.44	98670	1.05	3.32	Oxirane, dodecyl-
23	28.406	838178	1.12	308627	3.28	2.72	Hexadec anoic acid, 2- hydroxy-1- (hydroxymeth
24	28.560	414349	0.55	1522901	1.62	2.72	trans-2- Dodecene-1-ol
25	29.928	1497026	2.00	316816	3.37	4.73	Z,Z- 3, 13- Octadecedi-1-ol
26	30.867	710752	0.95	320106	3.41	2.23	Squalene
27	33.517	374032	0.50	67679	0.72	5.53	Carbamic acid, N-[10, 11- dihydro-5- (2 methyla
		75022785	100.00	9398692	100.00		

s and an area percentage of 0.018% and sucrose had a retention time of 17.641s and an area percentage 20.37%.

Various functional groups such as hydroxyl group, carboxyl group were also observed in Figure 2. The peak at 3298.87  $\text{cm}^{-1}$  is attributed to O–H stretching. The O–H stretching was also observed and after 24h fermentation using starter cultures it reduced to 3298.87 $\text{cm}^{-1}$ . The 2930.33  $\text{cm}^{-1}$  peak observed is attributed to C–H bond stretching. The peaks at 1412.94  $\text{cm}^{-1}$  is attributed to the bending modes of O–H.

The GC/MS analysis of the starter culture fermented sweet potato for 24 h of wheat extract showed the presence of 27 compounds corresponding with retention time from 5.043 to 33517 s. The most intensive peak was the lactic acid with an area percentage of 30% and a retention time of 7.118 s. The chromatogram had the following organic acids identified: octadecanoic acid at 25.123 s with an area percentage of 3.53%. n-Hexadecanoic acid at 23.025 s with an area percentage of 8.86% and oxirane had an area percentage of 27.40% and retention time of 14.241 s. The presence of lactic acid in the starter culture fermented sweet potato flour will help serve as a biopreservative, extend shelf life, and give a tart and

tangy flavour (Schnurer et al., 2005; Cizeikiene et al., 2013).

The absorbance at 3292.59  $\text{cm}^{-1}$  is attributed to O–H stretching; while 2929.48  $\text{cm}^{-1}$  is attributed to C–H bond stretching. The peaks at 1412.83  $\text{cm}^{-1}$  is attributed to the bending modes of O–H. The various functional groups detected are similar to those obtained in the 24 h fermented sweet potato flour.

Table 3 shows the chromatogram of starter culture fermented sweet potato flour after 48 h with 24 peaks. Lactic acid was detected and it also had the most intensive peak after 48 h fermentation using starter cultures (Figure 3), it had an area percentage of 55.53% and a retention time of 7.378 s, oxirane was detected at 14.117 s with an area percentage of 11.70%. Other compounds such as 9,12-octadecanoic acid (Z,Z) are polysaturated fatty acid which are anti-inflammatory, hypocholesterolemic, and cancer preventive according to Adeoye-Issijola et al. (2018) were detected with an area percentage of 10.265% and a retention time of 24.850 s. At 23.026 s, n-Hexadecanoic acid was detected with an area percentage of 11.87%.

The absorbance spectra obtained from Figure 4 shows O–H stretching, C–H bond stretching and O–H bending.

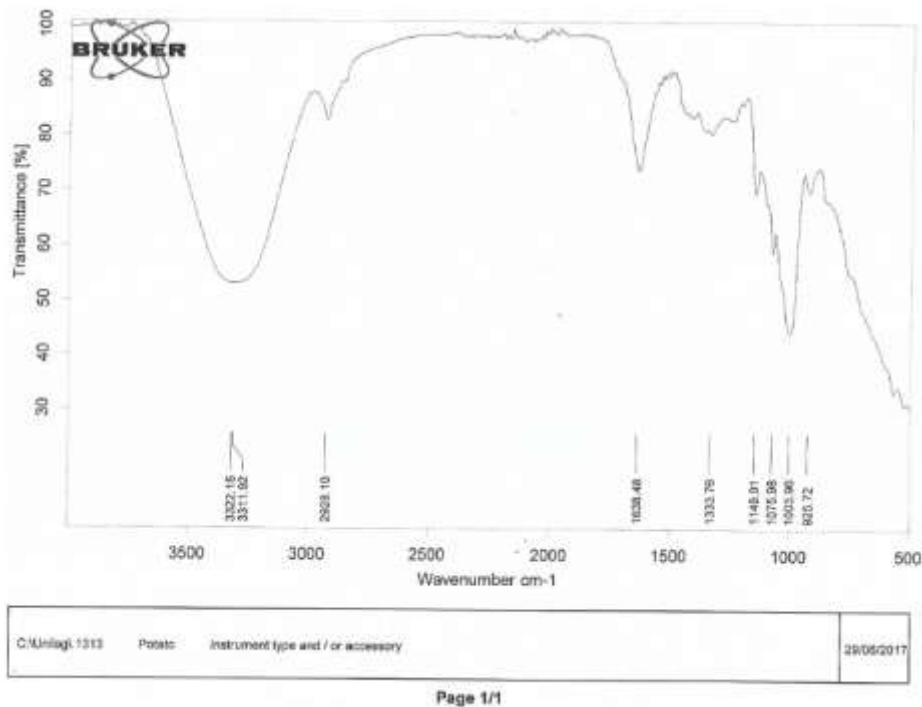


Figure 1. FTIR absorbance spectra for raw sweet potato (G).

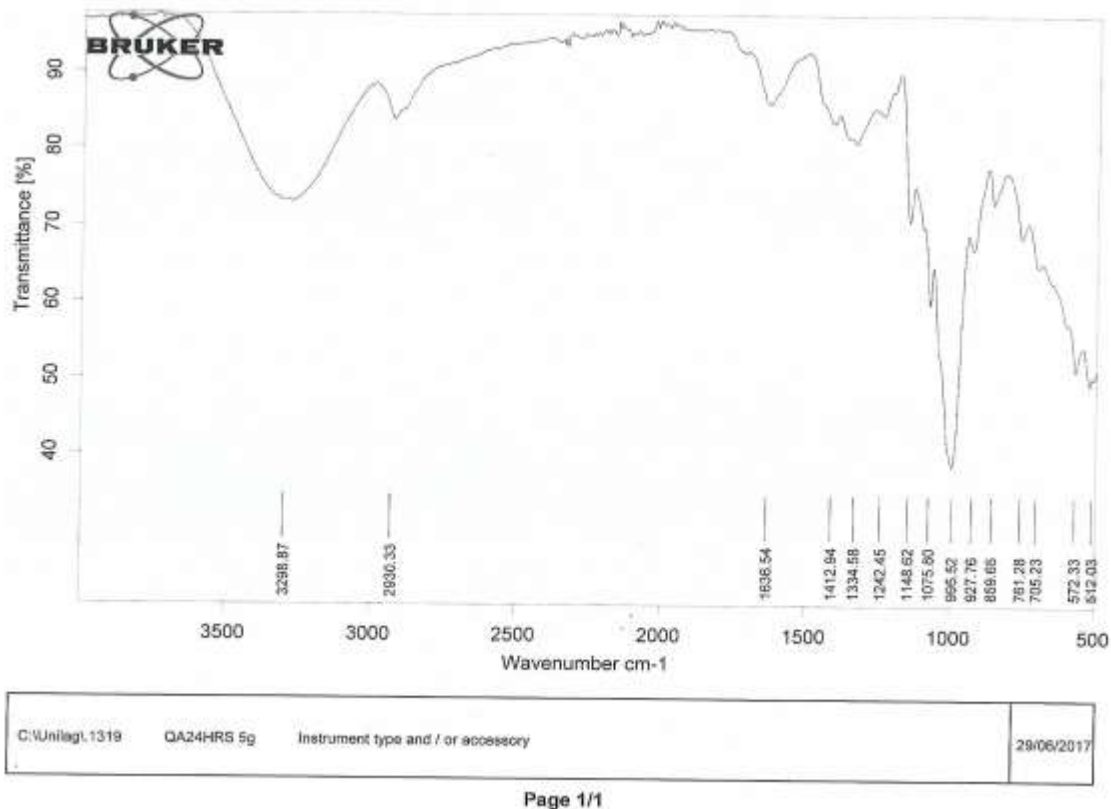


Figure 2. FTIR absorbance spectra for Starter culture fermented sweet potato flour 24 h (h).

The absorbance at  $3279.59\text{ cm}^{-1}$  is attributed to O–H stretching while C–H<sub>stretching</sub> occurred at  $2929.31\text{ cm}^{-1}$  (Table 4). The peaks at  $1414.11\text{ cm}^{-1}$  is attributed to the bending modes of O–H.

The chromatogram for 72 h starter culture fermented sweet potato flour is showed in Table 5 and Figure 5. Compounds such as n-Hexadecanoic acid with area percentage of 11.875% with a retention time of 23.026

**Table 3.** GCMS Peak report for starter culture fermented sweet potato flour 48 h (H).

Peak #	R. Time	Area	Area %	Height	Height %	A/H	Name
1	5.232	932784	0.86	473537	3.70	1.97	(S)-(+)-1,2 - Propanediol
2	7.378	60375505	55.53	2912673	22.76	20.68	L- Lactic acid
3	9.052	1980434	1.82	170548	1.33	11.61	1,2,3,4- Butanetetrol,[S-(R*,R*)]-
4	14.117	12717260	11.70	874627	6.83	14.54	Oxirane, (Propoxymethyl)-
5	15.180	196411	0.18	100516	0.79	1.95	Phenol, 2,6- dimethoxy
6	15.514	196211	0.18	54044	0.42	3.63	Butanal, 3- hydroxy
7	16.336	188766	0.17	87901	0.69	2.15	Methanimidane, N, N- dimethyl-N'-phenyl
8	16.586	1416787	1.30	240524	1.88	5.89	1,3- Propanediol, 2- (hydroxymethyl) -2- nitro
9	17.712	260612	0.24	78974	0.62	3.30	1- Imidazolidine carboxaldehyde, 5- hydroxy-2,4
10	18.165	464043	0.43	147730	1.15	3.14	undecanoic acid
11	18.563	842982	0.78	191940	1.50	4.39	Diethyl Phthalate
12	18.954	369712	0.34	87169	0.68	4.24	Butanal, 3-methyl -
13	22.132	185140	0.17	89378	0.70	2.07	1- Undecanol
14	23.031	7858036	7.23	211210	16.50	3.72	n- Hexadecanoic acid
15	23.346	590259	0.54	222690	1.74	2.65	Ethyl tridecanoate
16	24.854	9832442	9.04	2098048	16.39	4.69	9, 12 -Octadecadienoic acid (Z, Z)-
17	25.107	3374356	3.10	509313	3.98	6.63	Z,Z- 8, 10 Hexadecadien-1-ol
18	26.614	507526	0.47	259537	2.03	1.96	1-(2- Tetrahydrofurylmethyl)piperidine
19	28.032	352962	0.32	128807	1.01	2.74	9,12- Octadecadienal
20	28.405	1081626	0.99	407600	3.19	2.65	Hexadecanoic acid, 2-hydroxy-1- (hydrometh)
21	28.561	596971	0.55	188941	1.48	3.16	Tridecanal
22	29.925	2653622	2.44	606240	4.74	4.38	9,12- Octadecadienoic acid (Z,Z)-, 2- hydroxy-1
23	30.866	1446910	1.33	697038	5.45	2.08	Squalene
24	33.512	297818	0.27	58459	0.46	5.09	Carbamic acid, N-[10,11- dihydro-5-(2-methyla
		108719175	100.00	12797444	100.00		

s was detected. 9,12 octadecanoic acid (Z,Z) had a retention time of 24.850 s and an area percentage of 10.26% was detected. The chromatogram had 26 peaks and the most intensive peak was lactic acid with an area percentage of 58.645% and a retention time of 7.295 s. n-Hexadecanoic acid (palmitic acid) is a fatty acid which is antioxidant, antibacterial, anti-inflammatory, cancer preventive amongst other (Adeoye-Isijola et al., 2018).

In Figure 5 3274.59  $\text{cm}^{-1}$  could be attributed to O-H stretching. 2927.29  $\text{cm}^{-1}$  are attributed to C-H bond stretching. The peaks at 1410.41  $\text{cm}^{-1}$  was attributed to the bending modes of O-H.

The chromatogram of the control (spontaneously fermented sweet potato flour) was observed in Figure 5 with 22 peaks. It also had lactic acid has the most intensive peak with an area percentage of 28.71% at 6.92 s retention time, octadecanoic acid (Z,Z) was also detected at 24.843 s with an area percentage of 10.64%. n-Hexadecanoic acid was also detected with an area percentage of 11.8755% at a retention time of 23.026 s (Table 6).

## DISCUSSION

The peaks obtained were at 3322.15, 3311.92, 2928, 1638.48  $\text{cm}^{-1}$  for the raw sweet potato, 3298.87,

2930.33, 1636.54, 1412.94 and 995.52  $\text{cm}^{-1}$  for the starter culture fermented sweet potato flour for 24 h. Then, 3292.59, 2929.48, 1637.64, 1412.83 and 994.41  $\text{cm}^{-1}$  for starter culture fermented sweet potato flour for 48 h 3279.59, 2929.31, 1636.20, 1414.11, 994.75  $\text{cm}^{-1}$  the starter culture fermented sweet potato flour for 72 h. Also, 3274.59, 2927.29, 1635.65, 1414.41 and 995.42  $\text{cm}^{-1}$  for the spontaneously fermented sweet potato flour for 72 h. This is in line with earlier works carried out on starch contents spectra of starch were recorded using FT-IR, key bands by Belton et al. (1991). Spectra were recorded in regions below 800  $\text{cm}^{-1}$ , 500  $\text{cm}^{-1}$  (the fingerprint region), the region between 2,800 and 3,000  $\text{cm}^{-1}$  (C-H stretch region), and finally the region between 3,000 and 3,600  $\text{cm}^{-1}$  (O-H stretch region). Fourier transform infrared (FTIR) spectroscopy is a tool used to differentiate between patterns of amylose in different granule. Peaks near 3500, 3000, 1600, 1400, 1000, 800, described the IR spectrum of starch samples and Manley et al. (2002) to determine the presence of moisture used 500  $\text{cm}^{-1}$  as seen in the study by Zeng et al. (2011) and it. Moisture contents were also on mid infrared range. Peaks for water were observed on 1,640 and 3,300  $\text{cm}^{-1}$ . The absorption is done on the base of functional groups H and OH. The absorption spectra of starter culture fermented sweet potato flour and the control shows strong peaks in the same region indicating the presence of moisture in the

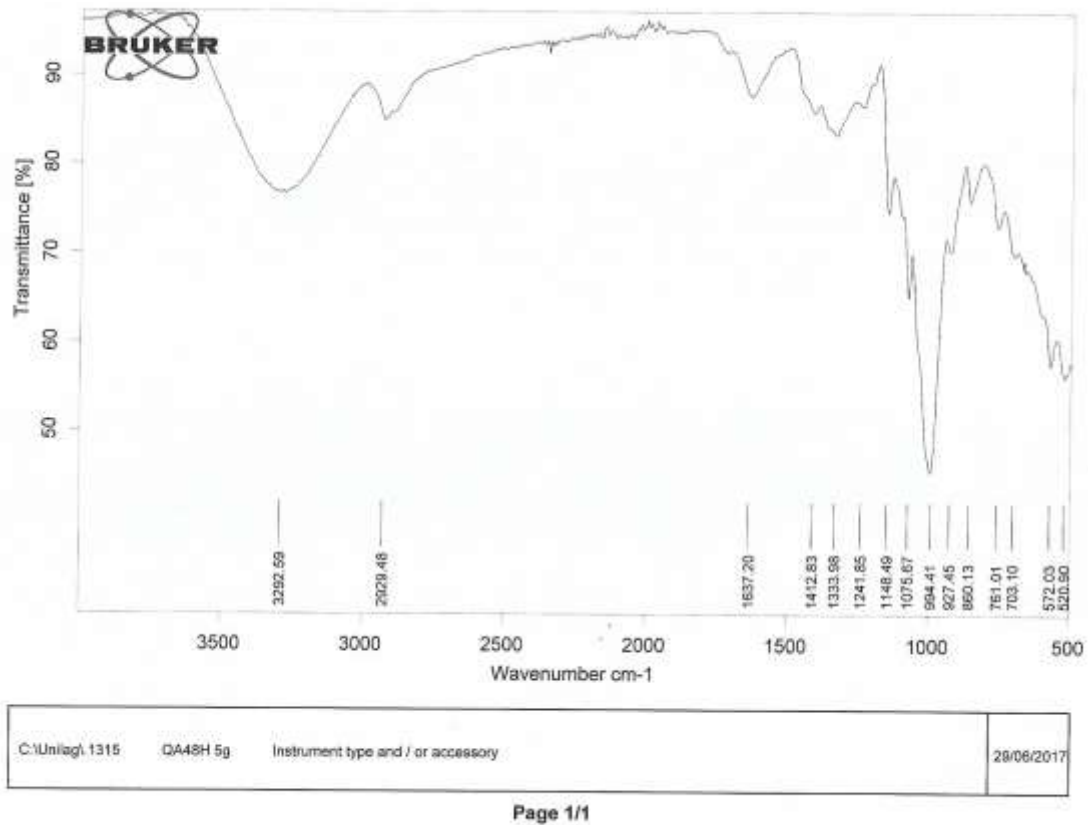


Figure 3. FTIR absorbance spectra for starter culture fermented sweet potato flour 48 h (I).

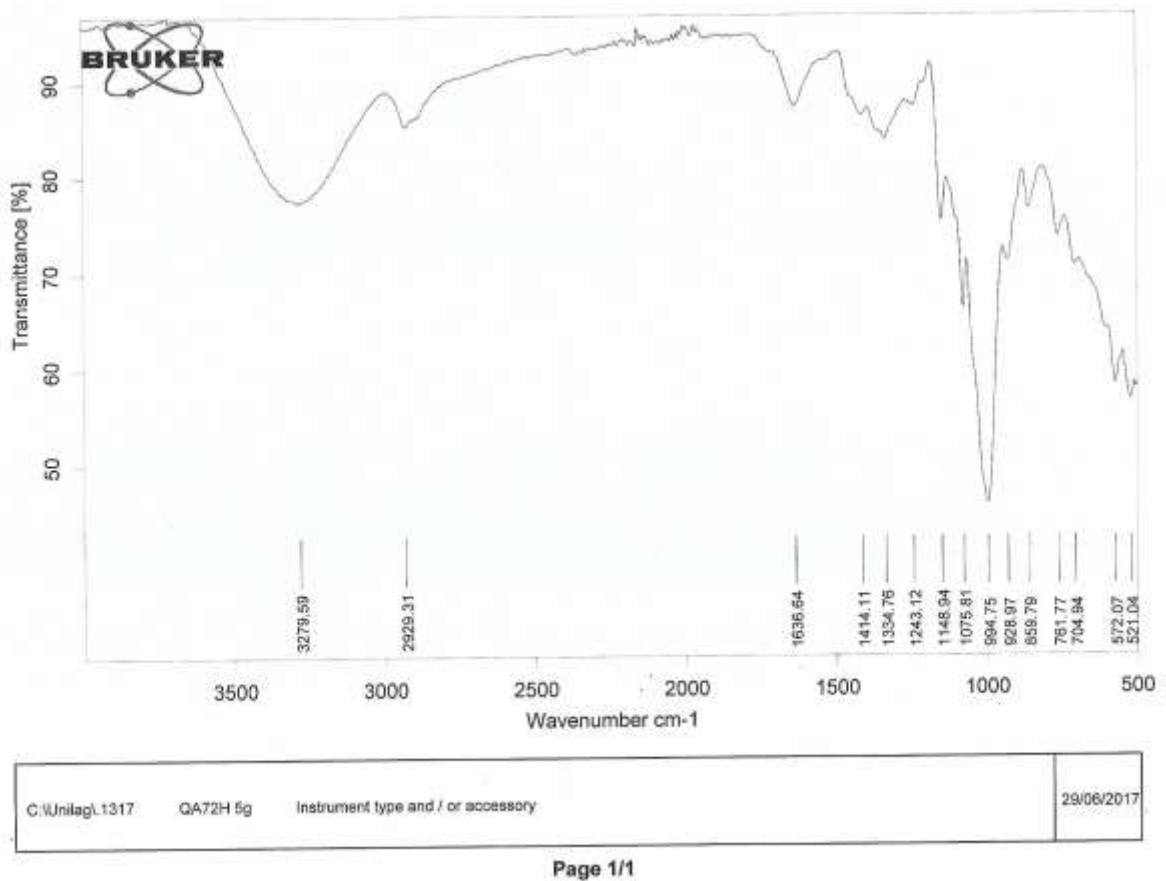


Figure 4. FTIR absorbance spectra for starter culture fermented sweet potato flour for 72 h (J).

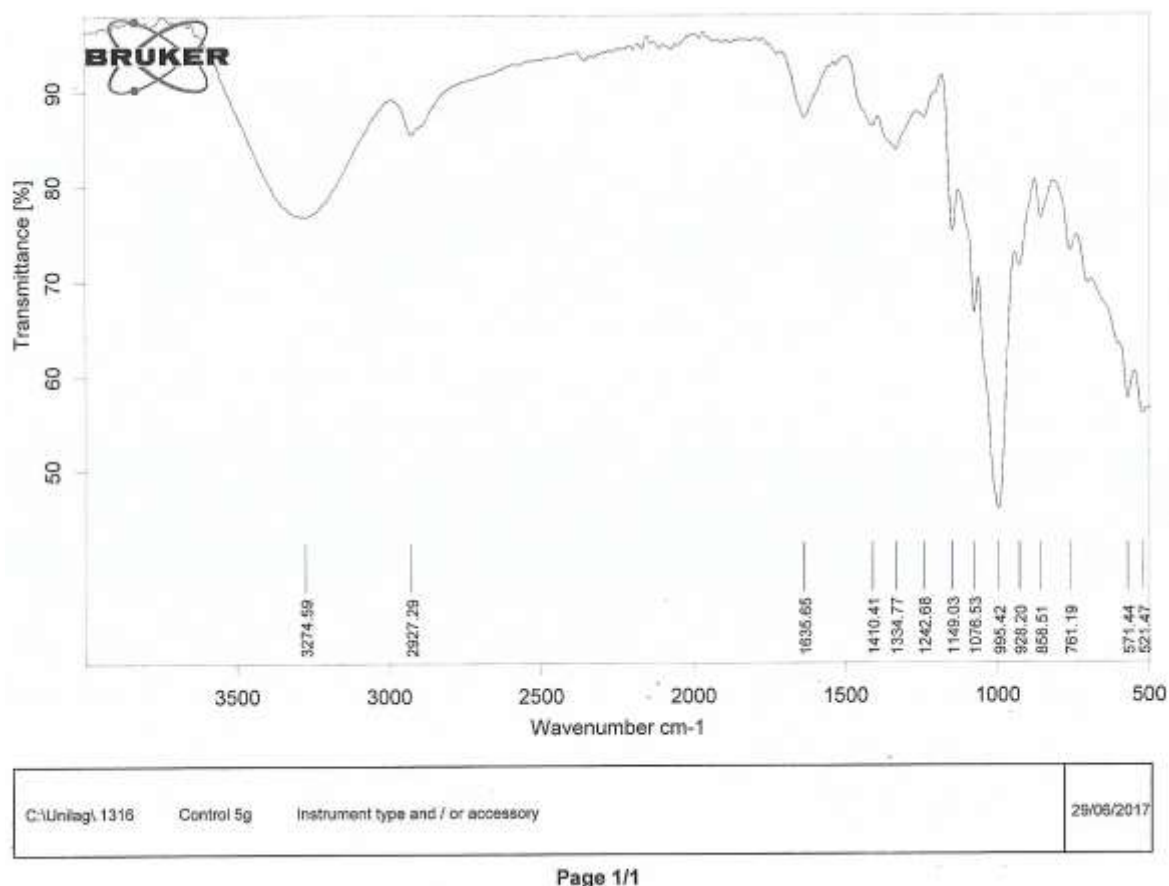


**Table 4.** GCMS Peak Report for starter culture fermented sweet potato flour 72 h (J).

Peak #	R. Time	Area	Area %	Height	Height %	A/H	Name
1	5.235	663461	0.86	351595	3.60	1.89	(S)-(+)- 1,2- Propanediol
2	7.295	44982527	58.64	2582353	26.45	17.42	L- Lactic acid
3	8.298	78983	0.10	51596	0.53	1.53	Ethanamine, 2-propoxy-
4	9.048	1711842	2.23	182475	1.87	9.38	1,2,3,4- Butanetetrol, [S-(R*,R*)]-
5	9.848	271735	0.35	69837	0.72	3.89	Butanoic acid, 2- hydroxy-3- methyl-
6	10.011	58890	0.08	35416	0.36	1.66	Benzeneacetic acid 1- methyl ethylester
7	10.151	81200	0.11	36343	0.37	2.23	Oxirane, 2,3- dimethyl-, cis-
8	13.937	4303934	5.61	559557	5.73	7.69	2- furanol, tetrahydro-2,3- dimethyl -, trans-
9	15.780	68755	0.09	37857	0.39	1.82	2 - Heptanamine, 5- methyl-
10	15.900	58065	0.08	34379	0.35	1.69	3,4 - Hexanedione, 2,2,5- trimethyl-
11	16.332	148684	0.19	65630	0.67	2.27	2- propenoic acid, 3 - phenyl
12	17.690	417392	0.54	128830	1.32	3.24	1,2,3,4- Cyclopentanetetrol, (1.alpha., 2beta., 3.1
13	18.057	31594	0.04	17255	0.18	1.83	Cyclopropyl carbinol
14	18.160	210811	0.27	71411	0.73	2.95	Propanedioic acid propyl-
15	18.564	337334	0.44	115687	1.19	2.92	Diethyl Phthalate
16	18.674	61127	0.08	34664	0.36	1.76	Pentanal
17	23.026	9108301	11.87	2018266	20.68	4.35	n - Hexadecanoic acid
18	23.347	197817	0.26	112642	1.15	1.76	Ethyl tridecanoate
19	24.343	261728	0.34	88641	0.91	2.95	Oxalic acid, allyl pentadecyl ester
20	24.850	7871474	10.26	1756535	17.99	4.48	9, 12- Octadecadienoic acid (Z, Z)-
21	25.109	2179835	2.84	388434	3.98	5.61	9, 9- Dimethoxybicyclo[3. 3. 1] nona- 2,4- dione
22	28.404	507649	0.66	175857	1.80	2.89	Hexadecanoic acid, 2 -hydroxy-1-(hydroxymeth
23	28.562	390981	0.51	129992	1.33	3.01	Heptanal
24	29.562	1156839	1.51	206547	2.12	5.60	2- Methyl- Z,Z-3,13- octadecadienol
25	30.865	982993	1.28	419254	4.29	3.34	2,6,10,14,18- Pentamethyl-2,6,10,14,18- eicosap
26	33.516	569698	0.74	90453	0.93	6.30	Carbamic acid, N -[10, 11- dihydro-5-(2-methyla
		76713649	100.00	9761506	100.00		

**Table 5.** GCMS Peak Report for starter culture fermented sweet potato flour 72 h (J).

Peak #	R. Time	Area	Area %	Height	Height %	A/H	Name
1	5.059	537269	2.06	117343	2.34	4.58	Ethanol, 2-nitro-
2	5.264	140986	0.54	67247	1.34	2.07	Formamide
3	6.920	7496208	28.71	617319	12.33	11.93	L- Lactic acid
4	8.773	212749	0.81	52524	1.05	4.05	Propanedioic acid, propyl-
5	8.849	671411	2.57	102578	2.05	6.55	1,2,3,4 - Butanetetrol, [S-(R*, R*)]-
6	11.331	115365	0.44	53716	1.07	2.15	Phenylethyl Alcohol
7	13.932	4338523	16.62	454674	9.08	9.29	(S)-(-)-1,2,4- Butanetriol, 2- acetate
8	15.177	84153	0.32	60439	1.21	1.39	Phenol, 2,6- dimethoxy-
9	18.156	15142	0.59	50058	1.00	3.08	Propanedioic acid, propyl-
10	23.026	3099692	11.87	1051581	21.00	2.95	n - Hexadecanoic acid
11	23.346	308180	1.18	126965	2.54	2.43	Decanoic acid
12	24.343	116249	0.45	63494	1.27	1.83	2- Heptanamine, 5-methyl -
13	24.843	2779010	10.64	524660	10.48	5.30	9,12- Octadecadienoic acid (Z,Z)-
14	25.102	1304627	5.00	263292	5.26	4.96	9- Octadecyonic acid
15	25.336	183668	0.70	57224	1.14	3.21	Hexanal
16	26.429	150043	0.57	65403	1.31	2.29	2- Heptanamine, 5 -methyl-
17	26.961	173089	0.66	87917	1.76	1.97	Cyclooctyl alcohol
18	28.032	534373	2.05	130463	2.61	4.10	Z, -1,9- Hexadecadiene
19	28.405	751722	2.88	284641	5.69	2.64	Hexadecanoic acid, 2- hydroxy-1-(hydroxymeth
20	29.931	1550434	5.94	288336	5.76	5.38	Z,Z -3, 13- Octadecadien-1-ol
21	30.867	985028	3.77	403555	8.06	2.44	Squalene
22	31.417	419765	1.61	82964	1.66	5.06	Carbamic acid, N -[10,11-dihydro-5-(2-methyla
		26106596	100.00	5006393	100.00		



**Figure 5.** FTIR absorbance spectra for starter culture fermented sweet potato flour for 72 h control (K).

flour. FTIR is one of the most elusive methods for the analysis of moisture. Water absorbs strongly in the infrared region of the spectrum due to its O-H stretching and H bending vibrations however its quantization is frequently complicated by spectral interferences from other OH containing constituents such as alcohols, phenols and hydroperoxides and confounded further by hydrogen bonding effects (Dong et al., 2000).

The O-H stretching for the raw sweet potato occurred at  $3322.15\text{ cm}^{-1}$  and after 24h fermentation using starter cultures, it reduced to  $3298.87\text{ cm}^{-1}$ ; at 48 h it was  $3279.59\text{ cm}^{-1}$  and at 72h it was  $3279.59\text{ cm}^{-1}$ . The decrease in the wavelength with increase in time depicts dilution of the crystalline (amylose) region, leading to the breakage of  $\alpha$ -1,4- glycosidic linkage. Consequently, the hydrophilic O-H group will contribute to the increasing amorphous fraction.

The peaks at  $2928.10$ ,  $2930.33$ ,  $2929.48$ ,  $2929.31$  and  $2927.29\text{ cm}^{-1}$  are attributed to C-H bond stretching. The C-H stretching for raw potato, starter culture fermented sweet potato flour at 24 h, 48 h, 72 h were  $2928.10$ ,  $2930.33$ ,  $2929.48$  and  $2929.31\text{ cm}^{-1}$  respectively. These values fall within the same range. This indicates that there was a change in the functional group and this could be the oxidation of an aldehyde group (CHO) during the fermentation.

Supriya et al. (2015) observed that Crude fat of flour samples had peaks at  $1,600\text{ cm}^{-1}$  to  $1,700\text{ cm}^{-1}$  and

$1,550$  to  $1,570\text{ cm}^{-1}$ . The absorption peaks are determined on the basis of C-H bonds. The absorption spectra of starter culture fermented sweet potato flour also show strong peaks in the same region, which indicates the presence of fat in flour. It has been reported that FTIR spectroscopy could be utilized as a quality control method for fat and moisture determination in butter and high-fat products (Van de Voort et al., 1992). Che-Man and Setiowaty (1999) and Rai et al. (2013) also reported similar results.

Supriya et al. (2015) observed absorption bands that are two primary features of the protein, amide I and amide II bands at approximately  $1,660\text{ cm}^{-1}$  and  $1,550\text{ cm}^{-1}$ , respectively. Amide I arises from the stretch of C=O of the peptide group in the protein. Peaks were also observed around this region for the starter culture fermented sweet potato flour as well as the control.

A cursory look at the spectra shows a slight decrease in the wavelength of the carboxyl (C=O stretching) of the  $\alpha$  1,4- and  $\alpha$ 1,6- glycosidic linkage ( $1638.48\text{ cm}^{-1}$  for raw sweet potato,  $1636.54\text{ cm}^{-1}$  after 24 h,  $1637.20\text{ cm}^{-1}$  after 48 h and  $1636.64\text{ cm}^{-1}$  after 72 h). The slight decrease in wavenumber indicates arial oxidation to carboxylic acid such as, acetic acid /ascorbic acid which can serve as preservative to the flour.

The stretching vibration for  $\alpha$  1,4- glycosidic linkage at the amylose region decreased from  $1003.96\text{ cm}^{-1}$  for raw sweet potato to  $994.75\text{ cm}^{-1}$  after 72 h, this will

**Table 6.** Summary of some compounds obtained from GC-MS analysis.

Duration	Compounds	Area (%)
0 h (G)	3- Deoxy-d-mannonic acid	23.3
	Sucrose	20.37
	Sec- Butylnitrite	17.93
	1,6- Anhdr-2,4-dieoxy-beta-D-ribo-hexopy	13.86
	5-Hydroxymethylfufura	2.81
24 h (H)	L-Lactic acid	30.71
	Oxirane ,(Propoxymethyl)	27.4
	n-Hexadecanoic acid	8.86
	9,12-Octadecadienoic acid (Z, Z)	8.26
	1,3-Propanediol,2-(hydroxymethyl)-2-nitro	4.64
	Octadecanoic acid	3.53
	1,2,3,4-Butanetetrol	2.68
	Diethyl phthalate	2.37
L-Lactic acid	55.53	
48 h (I)	Oxirane	11.7
	9,12-Octadecanoic acid	9.04
	n-Hexadecanoic acid	7.23
	Z, Z-8,10-Hexadecadien-1-ol	3.1
	9,12-Octadecadienoic acid	2.44
	1,2,3,4-Butanetetrol	1.82
72 h (J)	L-Lactic acid	58.64
	n-Hexadecanoic acid	11.87
	9,12-Octadecadienoic acid	10.26
	2-Furanol,tetrahydro-2,3-dimethyl-trans-9,9-Dimethoxybicyclo(3.3.1)nona-2,4-dione	5.61
	1,2,3,4-Butanetetrol	2.84
		2.23

further release more glucose unit into the amorphous region thus enhancing the swelling properties of the flour.

The peaks at 1412.94, 1412.83, 1414.11 and 1414.41  $\text{cm}^{-1}$  were attributed to the bending modes of O–H.

The peaks at 1097 and 1019  $\text{cm}^{-1}$  were assigned as the C–O bond stretching. The bands at 1047 and 1022  $\text{cm}^{-1}$  were associated with the ordered and amorphous structures of starch respectively.

The starter culture fermented sweet potato flour had more peaks than the control after 72 h fermentation. It also had higher concentration of lactic acid, which indicates a faster rate of fermentation with the use of starter cultures. The presence of high concentration of lactic acid will also help inhibit the presence of spoilage organisms, the elimination of spoilage organisms and it will serve as preservatives.

Some of the compounds detected by the GCMS include 9,12-octadecadienoic acid; hexadecanoic acid which are antioxidant; ethanol which is used as food preservative and propanoic acid which is widely used as an antifungal agent amongst others Supriya et al. (2015).

Generally, the chemical components were identified in each sample. The purpose of the study was identify the chemical components and compare presence of components in the various sample. Similar chromatograms were also obtained for all the starter culture fermented sweet potato samples. This study has shown that it is possible to compare the chemical compounds of samples with GCMS chromatography.

## Conclusion

The study reveals that sweet potato contain starch (amylose and amylopectin) and sucrose as sugar.

Starter cultures *L. brevis* and *D. polymorphous* fermented the sweet potato thereby breaking down the carbohydrate (starch) to produce alcohol, organic acid and  $\text{CO}_2$  hence lactic acid fermentation occurred.

Functional groups such as hydroxyl, aldehydes, alcohol and carboxyl are present in the fermented samples. Lactic acid fermentation occurred and it caused a shift in some of the functional groups.

The chemical shift indicates that the starter culture fermented sweet potato flour containing compounds

such as carboxylic acids, alcohols, aldehydes, hydroxyl and alkenes.

Starter culture fermented sweet potato flour had a higher concentration of carboxylic acids, alcohols, aldehydes etc. Alcohol and carboxylic acid was produced in situ from the fermentation process, which will help inhibit spoilage organisms moreover, it will also serve as preservatives thereby increasing shelf life of the product.

The FTIR showed a similar spectrum for all the fermented sweet potato flour and it was possible to verify the main organic functions associated, the results corroborate the wide variety of volatile organic compounds identified by the CGMS. The presence of OH group detected by FTIR which was further emphasized by the presence of butanol detected by the GCMS, and the presence of the C=O group detected by FTIR which was also emphasized.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interest.

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