The Pharma Innovation Journal 2021; 10(10): 1704-1710

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2021; 10(10): 1704-1710
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www.thepharmajournal.com
Received: 09-07-2021
Accepted: 22-08-2021

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Impact of Freeze-drying on physiochemical, structural, and mechanical properties of ginger (*Zingiber officinale*)

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Abstract
Ginger (*Zingiber officinale*) is the famous spice and ayurvedic medicine used worldwide. The effect of Freeze-drying (FD) on the physical, chemical and mechanical properties of ginger slices is analyzed. The fresh rhizomes are collected, cleaned, and sliced into desired sizes, and dried using a freeze dryer (FD). The dried slice has a total color difference of 9.73±0.14 and has a higher whiteness index and hue angle, inhibit a lower chrome value, yellowness index. The rehydration ratio of FD slices was calculated as 7.80±1.06. the dried slices have very minimum volumetric shrinkage, SEM was used to analyze the structure of FD dried slices. The hardness of FD slices is identified as 6.57±0.34, which have significantly different from fresh samples. The volatile and nutritional composition of slices is analyzed using GC-MS and FT-IR.

Keywords: Freeze-drying, physiochemical properties, volatile compounds, structural changes, texture

1. Introduction

Ginger (*Zingiber officinale*) is mostly used as a spice in different parts of the world. Also, it is used as a medicine in ayurvedic science to cure diseases like nausea, vomiting, diabetes, migraine headache, and other conditions (Gupta & Sharma, 2014) [6]. Spices are valued based on their aroma, color, taste, functional and medicinal properties. Most fresh spices have a moisture content of 80-90%, which is prone to microorganisms. To preserve or store these spices primary step taken was drying, which helps to reduce the water level in the spices, also helps to improve the flavor and oil content in most of the spices.

The major concern of the current food industry is to produce commodities with high sensory, nutritional and functional attributes. In the case of drying the functional and sensory properties of the end, products are varied based on the drying type and process parameters, and the nutritional characteristics are get altered. In some cases, it provides positive results like enhanced flavor, high shelf life, on the other hand, the negative impact of drying was identified like color change, nutritional loss at high temperature, structural changes. To reduce these disadvantages hybrid dryers, vacuum dryers, heat pump dryers are identified.

A freeze dryer (FD) is the type of conventional dryer, where drying has taken place at a very low temperature. Which can be well suitable for spice drying, since they are sensitive to heat (Jin *et al*., 2017) [9]. FD is also known as lyophilization, the principle of removal water in FD is different from other drying processes. Hence, here water removal has taken place by sublimation, but in other dryers, it has taken place by evaporation of water molecules. In an FD dryer the vapor pressure difference between ice and surrounding air act as a driving force for the sublimation process. But in other dryers differences in heat energy play a role as a driving force, that's why FD operates at low temperatures. In FD drying of samples taken place at two stages, in the first stage, the unbound water molecules in the samples are crystallized by freezing, in the second stage, the crystallized samples are vapourised by the pressure and temperature combination at the sublimation (0^{o} and 6.1 mbar for ice) point (Harguindeguy *et al*., 2019) [7]. So FD can be an alternative drying technique to preserve the nutritional quality of the spices.

The objective of this work is to identify the physical, mechanical, and chemical changes occurring during the freeze-drying of ginger. The results can be helpful to understand the advisable effect of freeze-drying on the spices (ginger).

2. Materials and methods

2.1 Sample preparation

Newly harvested fresh ginger rhizomes are brought from a local market in Thanjavur, Tamil Nadu. They are cleaned, washed, and peeled. The peeled rhizomes are slice into uniform shapes and sizes. Now the slices are used for experimental purposes.

2.2 Freeze dryer

The prepared fresh slices are stored in -20 ºC (MAC MSW 135, India) for 24 hrs for freezing, and then they have undergone freeze-drying (model: penguin classic - BENCHTOP). The pressure of the chamber was maintained at 50 Pa. The dried samples are taken for the experimental study (Silva-Espinoza *et al*., 2020) [14]. (mention FD temperature, pressure).

2.3 Measurement of physical properties

2.3.1 Moisture content (MC) and water activity

The hot air oven method was used to analyze the moisture content of the fresh and dried sample. The fresh samples are kept at 105 ºC for 3hrs to finds out the initial moisture content (Danso-Boateng, 2013) [2]. After the freeze-drying, the moisture content of the samples is calculated in the form of a dry basis using the dry matter content (Equ.2), and it was converted to wed basis using Equ.3.

$$Moisture\ content\ (w.b\%) = \frac{amount\ of\ moisture\ removed}{weight\ of\ the\ sample} \times 100 \quad (1)$$

$$Moisture\ content\ (d.b\%) = \frac{amount\ of\ moisture\ removed}{weight\ of\ bone\ dry\ matter\ of\ the\ sample} \times 100 \quad (2)$$

$$Moisture\ content\ (w.b\%) = \frac{Dry\ basis\ moisture\ content}{1+Dry\ basis\ moisture\ content} \quad (3)$$

To measure the water activity of the fresh and dried samples Aqua lab (dew point water activity meter 4TE, USA) was used.

2.3.2 Surface Color measurement

For analyses, the color of the ginger slices, Hunter lab calorimeter (Colourflex EZ model: 45/0) was used which provides the observation in the three variables as L^*, a^*, and b^* (CIE color components). L^*, a^*, and b^* represent the range from lightness, red-green, and yellow-blue. Before the usage, the calorimeter was calibrated using black and white plates. Samples are filled in the sample cup in the way they fully cover the bottom surface of the cup and then reading are taken. Fresh raw slices are used as a standard. The freeze-dried slices color was measured. Only the surface color of the slices can be measured by colorimeter, replicate are taken in the different positions of ginger slices. The difference in color value was calculated using the following formula,

$$\Delta E = \sqrt{(L_0{}^* - L^*)^2 + (a_0{}^* - a^*)^2 + (b_0{}^* - b^*)^2} \quad (4)$$

The chroma index(ΔC), Browning index (BI), yellowness index (YI), whitening index (WI), and hue angle are calculated using the following formula (Dziki *et al*., 2018) [5],

$$\Delta C = \sqrt{a^{*2} + b^{*2}} \quad (5)$$

$$Hue\ angle = tan^{-1}\left(\frac{b}{a}\right) \quad (6)$$

$$BI = \frac{100(x-0.31)}{0.17} \quad (7)$$

Where,

$$x = \frac{a^* + 1.75L^*}{5.645L^* + a^* - 3.012b^*} \quad (8)$$

$$YI = \frac{142.86b^*}{L^*} \quad (9)$$

$$WI = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^{*2}} \quad (10)$$

Where, $L_0{}^*$, $a_0{}^*$, $and\ b_0{}^*$ represents the value of the fresh sample and L^*, a^*, and b^* represents the value of the dried sample, 142.86 represents the equilibrium coefficient.

2.3.3 Rehydration ratio (RR)

2g of dried ginger slices are immersed in 50ml of distilled water (25ºC) for 2hr. Then, the slices are taken out, the extra surface water is removed by blotting paper then weighed using an electrical weighing balance (WENSAR electronic balance) with ± 0.01 g accuracy. Equ.11 is used to calculate the rehydration ratio of the slices. Where, $X_0$and X_t represents the weight of slices before and after rehydration (Li *et al*., 2019).

$$RR = \frac{X_t}{X_0} \quad (11)$$

2.3.4 Shrinkage

To determine the shrinkage percentage of the dried slices changes in length (L), breath (B), and thickness (T) are used. The volumetric shrinkage (S_v) of dried ginger slices are analyzed by the following Equ.12, where L_0, B_0, and T_0 represent the dimensions of the fresh sample and L, B, and T represent the dimensions of the freeze-dried samples (Seerangurayar *et al*., 2019) [13].

$$S_v = \frac{V}{V_0} = \frac{LBT}{L_0 B_0 T_0} \quad (12)$$

2.3.5 Analysis of microstructural

To study the effect of freeze-drying on the structure of the slices, scanning electron microscopy (Model: TE – SCAN VEGA 3 SEM) was used. Here the middle portion of the dried samples was mounted on the aluminum stubs vertically, then their microphotograph was taken (Izli & Polat, 2019) [8]. Accelerating voltage of 10KV, 3 kx magnification, with a view field of 69.2 µm are used to take the SEM image.

2.4 Measurement of mechanical property

A textural analyzer was used to find out the deformation in the slices during the applied force. The textural property of the dried slices is analyzed using a texture analyzer model TA.HD plus (stable Micro System, Surrey, UK textural analyzer (P/5 cylinder, pre, and post-test speed 2 mm/sec, 75% strain) (An *et al*., 2015) [1]. A double compression mode test was used to analyze the properties of the sample. The first maximum force of the force-deformation curve is taken as a hardness. Triplicate samples are tested and the hardness values are reported.

2.5 Measurement of chemical properties

2.5.1 Determination of Fourier Transform Infrared Spectroscopy (FT-IR)

An IR-Affinity 1S FTIR spectrometer equipped with a DLAGTS detector was used to perform the molecular level structure analysis of FD ginger powder (Qiu *et al*., 2020) [42]. Ceramic rods are used as an infrared source. The spectra were achieved in the IR range of 4500-500 cm^{-1} with 45 scans at 4 cm^{-1} resolution.

2.5.2 Determination of volatile compounds

1g of dried slices are added with 10 ml of methanol and vortexed for 10 min and stored in a shaker overnight. Then the mixture was placed in the centrifuge at 6000 rpm the supernatant was separated and used for analysis. The extracts were injected in GC-MS, the compounds were identified by the NIST library and matched using the previously published data. The volatile compound's quantities were compared by their peak value (Song *et al*., 2003) [15].

2.6 Statistical analysis

For analysis, all experiments are conducted triplicate times, and the values are given as mean ±SD. 95% confidence levels ($p<0.05$) are taken for analysis. Paired t-test was used for the analysis. SPSS statistics 20.0 (International Business Machines Corp., USA) was used for statistical analysis.

3. Result and discussion

3.1 Effect of freeze-drying on physical characteristics of ginger

3.1.1 Effect of feeze-dying on moisture content and color

The water content of the samples is calculated using the hot air oven method using triplicates. The fresh ginger slices have a moisture content of 82.55±0.3% (wb), and water activity of 0.953±0.043. which have near MC to fruits and vegetables and have higher water activity, it prone to spoilage by microorganisms like bacteria and fungi. The MC content of the FD slices is reduced to 8±0.5%, which was suitable MC for the safe storage of samples. And FD samples have a water activity of 0.392±0.056, during the sublimation, most of the free water is removed it causes the lower water activity of dried slices. Lower water activity helps to preserve the samples from microorganisms.

The Colour of the products is the critical factor when comes to the marketing point of view because it is the first parameter that could attract consumers. The color CIE components (L^*, a^*, and b^*) are shown in Table (1). The total color difference (ΔE) of the fresh and dried have a significant difference ($p<0.05$). The ΔE denotes the ability of the human eye to distinguish the variation of color in the samples (Izli & Polat, 2019) [8]. The calculated total color difference (ΔE) between the samples was >3, which denotes dried slices have a distinct color different from freeze-dried slices. The total color value (E) of the fresh sample and a freeze-dried sample was 64.92±0.53 and 66.06±0.34 respectively. Likewise, chroma, hue angle for fresh and dried samples are 34.41±0.49 and 26.47±0.16; 78.76±0.18 and 80.876±0.07 respectively. This indicates that the total color intensity was reduced by freeze-drying, both fresh and dried samples have near the yellow hues angle. Colour loss during the drying process can be analyzed by different numerical indexes. Normally, yellowness index (YI), whiteness index (WI), and browning index (BI) represent the total degradation by processing, discoloration of the sample during drying, and change in color by enzymatic or non-enzymatic browning during the process. YI, WI, and YI of the samples are shown in Table (1). During freeze-drying, the yellowness of ginger slices gets reduced, and the whiteness index is improved by the discoloration of the slices. It was the same in the cause of freeze-drying of kale (Dziki *et al*., 2018) [5]. Other conventional dryers like tray driers, microwave drier produce samples with a higher browning index. But freeze-dried samples have a low browning index of the samples, it may be because of the very low oxygen present during the FD. The results are correlated with the previous studies carried out on hawthorns, carrots, and orange puree (Duan *et al*., 2017; Nahimana & Zhang, 2011; Silva-Espinoza *et al*., 2020) [4, 11, 14]. Due to the white blush by dehydration and lignifications during drying the whiteness index increased. In the end, freeze-dried samples have a higher whiteness index and lower yellowness and brownness index than fresh samples. The process temperature during FD is very low, it causes fewer physiological changes

Table 1: color and rehydration property of FD ginger slices.

Sample Type	L^*	a^*	b^*	Chrome (C)	Hue angle (°)	Yellowness index (YI)	Whiteness index (WI)	ΔE	Rehydration Ratio (RR)
Fresh	55.05±0.32^b	6.71±0.03^a	33.75±0.50^a	34.41±0.49^a	78.76±0.18^b	87.60±0.80^a	43.38±0.05^b	-	-
FD	60.53±0.31^a	4.20±0.02^b	26.13±0.16^b	26.46±0.16^b	80.88±0.07^a	61.67±0.25^b	52.48±0.19^a	9.73±0.14	7.80±1.06

* The means in the column at different superscript were significantly different at $p<0.05$.

3.1.2 Rehydration behavior of freeze-dried slices

The rehydration process helps to understand the chemical and physical changes that occur in the samples during the drying process. Also, it is used to measure the ability to reobserve the water during the processing like cooking. In the case of FD takes a comparatively higher time to get the desired moisture level of products than the other conventional drying methods. Here it needs time to obtain the sublimation condition then the moisture gets removed. The final dried samples have very less moisture content in them, this catalyzes the rehydration process of FD slices. When the ginger slices were dried used other dryers the noted rehydration ratio (RR) was 3 to 6.2 (Izli & Polat, 2019) [8], also it takes more time for rehydration. In the case of FD, the slices absorb the maximum amount of moisture in the initial stage itself also have the maximum RR of 7.80±1.06. It denotes that after rehydration the slices can able to get their original physical parameters. But it can’t get its 100% original quality, hence the structure and nutritional composition can be modified during drying.

3.1.3 Effect of freeze-drying on Shrinkage and microstructure of dried slices

The volumetric shrinkage of dried slices is expressed as a ratio of dried sample volume to the original volume of slices. The calculated volumetric shrinkage (Sv) of FD slices is 0.9±0.06, which is very low. And the changes in individual breadth, width, and thickness of samples have no significant difference with a fresh one. When the agricultural commodities are subject to drying, the moisture removal has taken place by diffusion of moisture from the surface and inter tissue of the sample. The second stage creates a lot of stress in the sample it leads to the shrinkage of samples. In FD the external stress created was very low, it may be because of the sublimation process. It results in a very less volumetric shrinkage ratio in the samples. Other kinds of dryers have gone up to a shrinkage ratio of 30 to 68%, but FD only goes up to 7 to 10%.

Microstructure changes during the were analysed SEM as shown in Fig (1). The crystalline structure of the ginger slices is completely collapsed, the cell wall got ruptured, the starch material is scattered, and tissue enlargement by vapourization (Izli & Polat, 2019) [8]. The same kind of results was obtained by (Wang *et al*., 2019) [17] during vacuum drying (VD) of ginger slices. It may be because the vacuum generated in both FD and VD, destroys the structure of the sample and increase the starch grain distribution. In addition to that ice crystals forming during the freeze-drying rupture the cell wall (Silva-Espinoza *et al*., 2020) [14]. The final porous dried sample can improve the rehydration and fracturability of the dried samples.

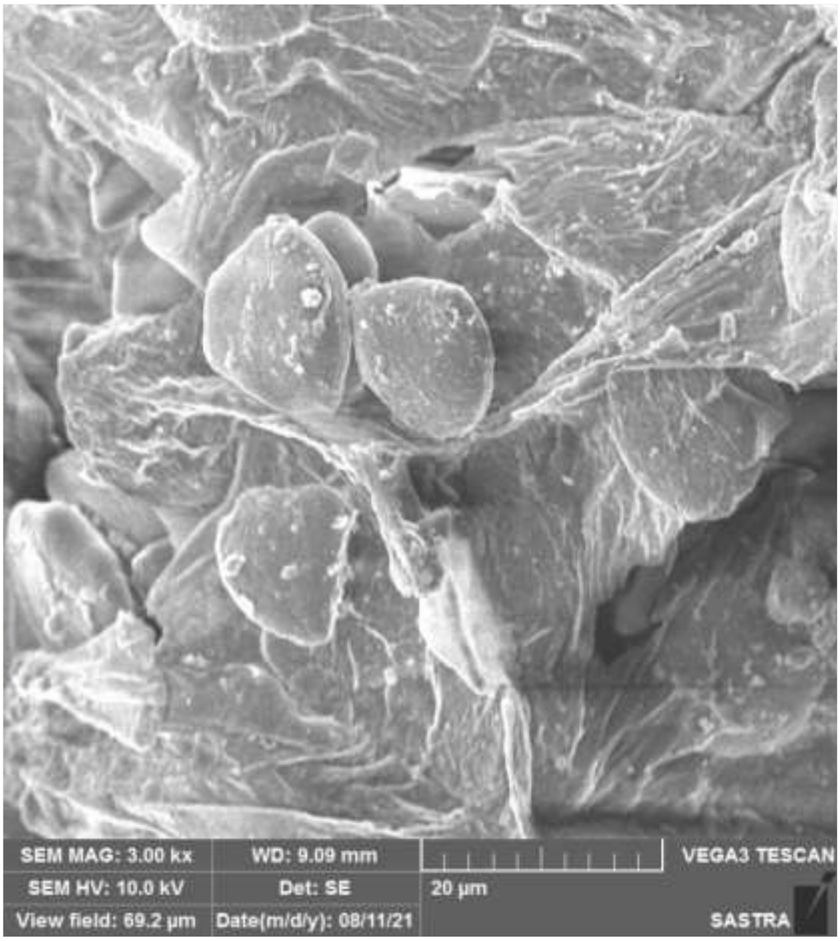

Fig 1: Morphologies of freeze-dried (FD) ginger slices.

3.2 Effect of freeze-drying on mechanical properties

The mechanical property of ginger slices was analyzed using a textural analyzer, the results were shown in Table (2). The first maximum peak of the force-deformation curve is the hardness of the sample, this helps to analyze the hardness of the freeze-dried sample (Thuwapanichayanan *et al*., 2011) [16]. The FD samples have higher hardness than fresh samples because the removal of moisture improves the force required for the deformation. In addition to that fracturability of the sample gets increased. When compare the FD slices to the previous studies in the other driers, FD has very little hardness and fracturability value, hence it contains a more porous structure. The chewiness of FD samples is higher than fresh samples. It shows force require to chew the FD slices are comparatively higher than fresh samples.

Table 2: Effect of freeze-drying (FD) on hardness, fracturability, and chewiness of ginger slices.

Sample Type	Hardness	Fracturability	Chewiness
Fresh Slices	3.096 ± 0.678^{b}	4.044 ± 0.244^{b}	0.704 ± 0.003^{b}
FD Slices	6.580 ± 0.338^{a}	6.691 ± 0.294^{a}	1.444 ± 0.291^{a}

The means in the column at different superscripts were significantly different at $p<0.05$.

3.3 Effect of freeze-drying on chemical properties

3.3.1 FTIR molecular structural profile

The FTIR spectrum was shown in Fig (2), the strong bond absorption was found in the wavelength range of 1000-1750 cm^{-1}. This absorption wavelength may be due to the vibration of O-H and the stretching of the N-H group during analysis. Based on the absorption molecules present in the sample were analyzed. In this manner, at 3000-3500 cm^{-1} amino groups were identified, at 2926 cm^{-1} the absorption due to the CH_2 group, C=O absorbs the IR at 1735 cm^{-1} wavelength. Most of the aromatic compounds C=C are identified at 1514 cm^{-1} wavelength. OH and phenolic groups involved with the wavelength range of 1310-1410 cm^{-1}. Strong absorption at 1629 indicates that the presents of protein compounds (Qiu *et al*., 2020) [42]. The FD samples are shown to have protein, phenolic and aromatic compounds.

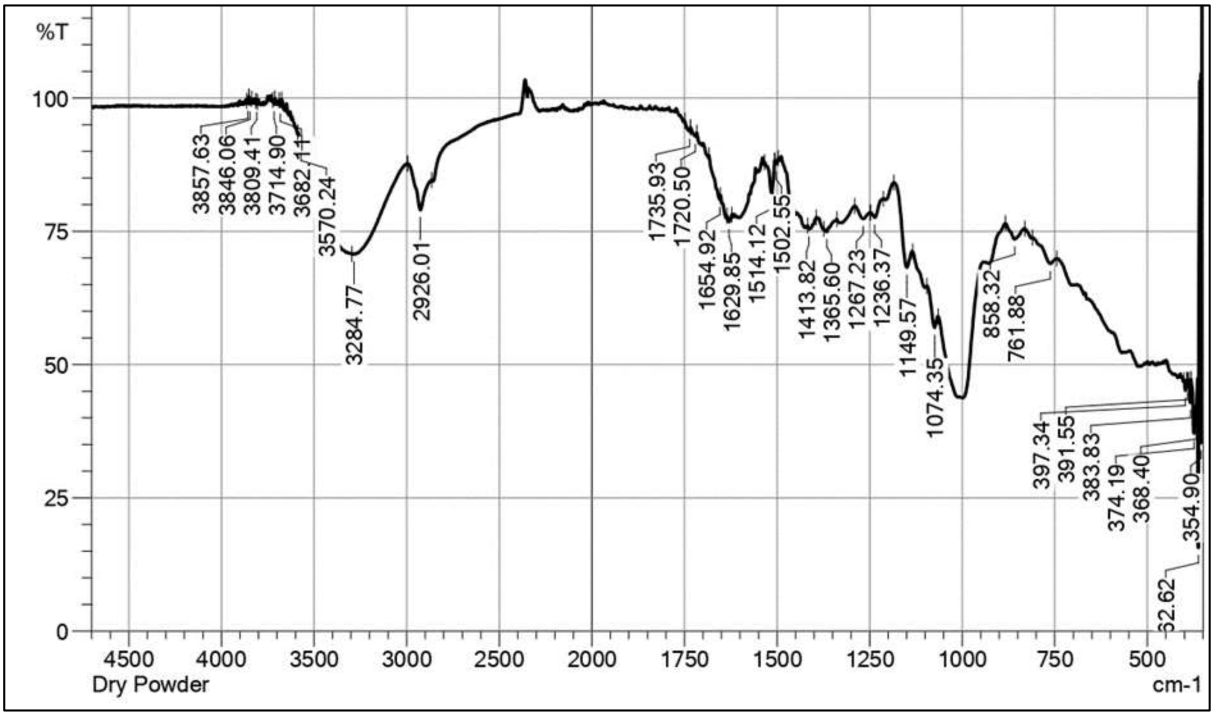

Fig 2: Spectra of ginger slices dried by FD in the frequency range of 4500-500 cm^{-1}.

3.3.2 Volatile profile

To analyze the volatile compounds in the dried (FD) ginger slices GC-MS was used. More than 30 peaks were identified in different retention times, in that only 17 compounds were identified by MS. Volatile compounds present in dried ginger slices are presented in Table (3). The important compounds identified by GC-MS are Bicyclo[3.1.1]heptane, 2,6,6-trimethyl-, (1.alpha.,2.beta.,5.alpha.), 2,6-Octadienal, 3,7-dimethyl-, Naphthalene, decahydro-4a-methyl-1-methylene-7-(1-methyl ethenyl)-, 1H-Cycloprop[e]azulene, decahydro-1,1,7-trimethyl-4-methylene-, [1aR-(1a.alpha.,4a.alpha.,7.alpha.,7a.beta.,7b.alpha.)]-, Phellandrene, 3,5-Octadiene, 2,2,4,5,7,7-hexamethyl-. Alcohols, ketones, and alkenes occupied are the most compounds identified. β-Phellandrene are the main volatile compounds present in most of the ginger species, and it was present in FD slices. Some new chemical compounds are analyzed compare to previous studies, it may be because of different geographical origins and solvent extraction techniques. The aroma of dried slices is specially controlled by α-farnesene, β-myrcene, β-phellandrene, and borneol. In this α-farnesene is the primary compound responsible for the flavor of the FD slices, and β-myrcene, β-phellandrene plays a secondary role to produce a flavor. Borneol was not identified by GC-MS in the FD samples (Song *et al*., 2003) [15]. Compare to the fresh samples the dried samples have a distinct flavor, because of the increase in volatile compounds such as 1,3-Cyclohexadiene, 5-(1,5-dimethy l-4-hexenyl)-2-methyl-, [S-(R*,S*)]-, α-farnesene, 3,5-Octadiene, 2,2,4,5,7,7-hexamethyl-, (E,Z)-, and 2,6-Octadienal, 3,7-dimethyl-. It was probably because of the formation of short-chain alkenes and the isomerization of compounds (Ding *et al*., 2012) [3]. On the other hand moisture reduction and cell wall damage during drying helps to release the volatile compounds. In other drying methods, due to the presents of oxygen some volatile compounds get degrades, this was not happening in FD slices.so, FD can be recommended to get a better flavor of ginger slices.

Table 3: Volatile compounds in freeze-dried ginger slices detected by GC-MS.

SI. No	RT	Compound name	% Peak area
1	5.775	Phellandrene	4.28
2	8.208	Bicyclo[3.1.1]heptane, 2,6,6-trimethyl-, (1.alpha.,2.beta.,5.alpha.)	1.22
3	8.631	2,6-Octadienal, 3,7-dimethyl-	1.81
4	11.319	1H-Cycloprop[e]azulene, decahydro-1,1,7-trimethyl-4-methylene-, [1aR -(1a.alpha.,4a.alpha.,7.alpha.,7a. beta., 7b.alpha.)]-	1.09
5	11.464	Benzene, 1-(1,5-dimethyl-4-hexenyl)-4-methyl-	5.30
6	11.530	1,6-Cyclodecadiene, 1-methyl-5-met hylene-8-(1-methylethyl)-,	1.20
7	11.619	1,3-Cyclohexadiene, 5-(1,5-dimethy l-4-hexenyl)-2-methyl-, [S-(R*,S*)]-	15.66
8	11.664	1,3,6-Octatriene, 3,7-dimethyl-, (E)-, alpha farnesene	4.36
9	11.741	Naphthalene, 1,2,3,4,4a,5,6,8a-oct ahydro-7-methyl-4-methylene-1-(1-m ethylethyl)-	5.44
11	11.852	(+)-Epi-bicyclosesquiphellandrene	0.87
12	11.919	1,3-Cyclohexadiene, 5-(1,5-dimethy l-4-hexenyl)-2-methyl-, [S-(R*,S*)]-	6.27
13	12.186	Naphthalene, decahydro-4a-methyl-1-methylene-7-(1-methylethenyl)-, [4aR-(4a.alpha.,7alpha.,8a.beta.)]	0.81
14	12.830	1,6,10-Dodecatriene, 7,11-dimethyl-3-methylene-, (E)-	0.94
15	13.130	3,5-Octadiene, 2,2,4,5,7,7-hexamethyl-, (E,Z)-	14.55

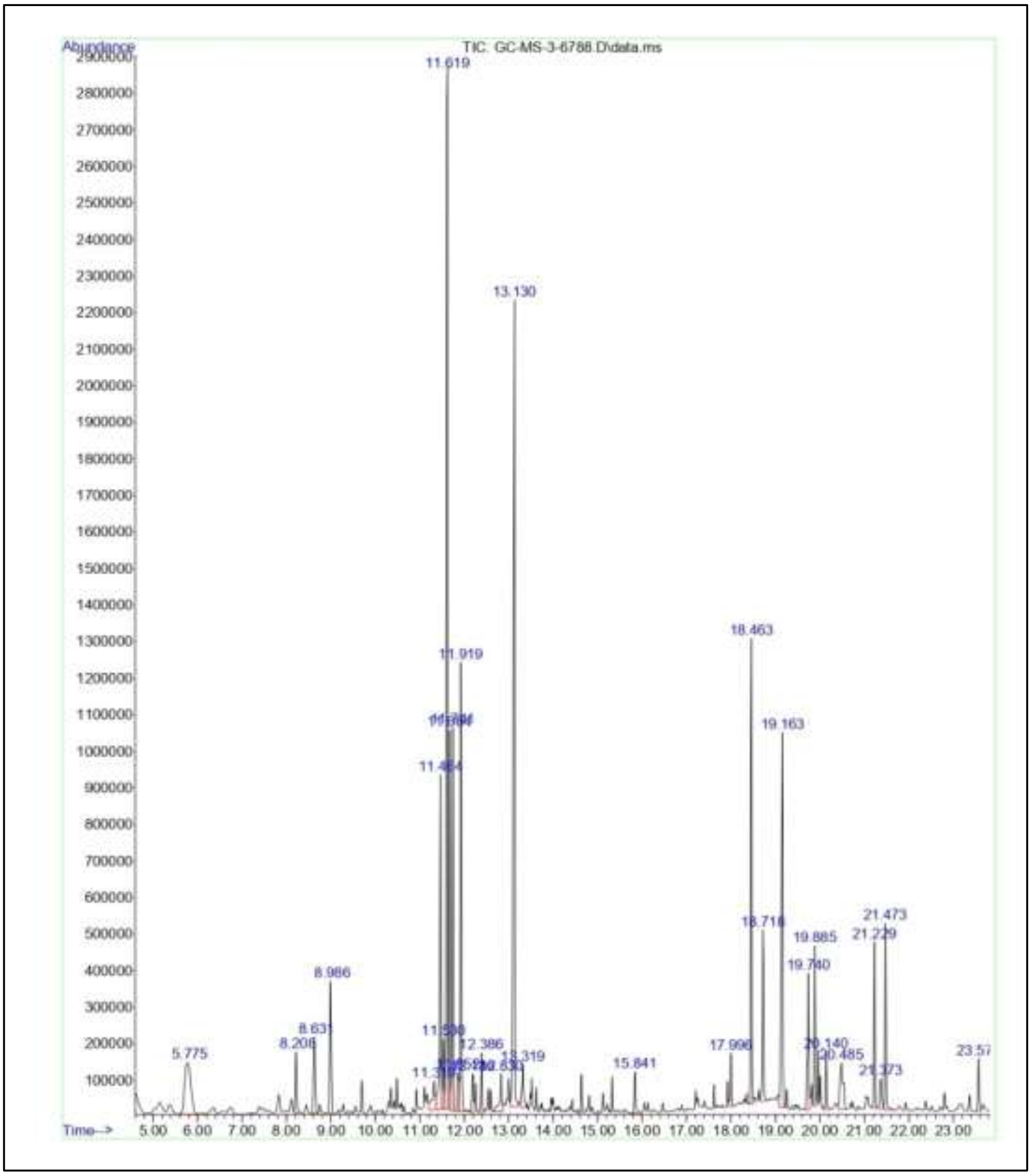

Fig 3: GC-MS chromatogram for FD ginger slices

4. Conclusion

The study has identified the effect of FD on the physical, chemical, and mechanical properties of ginger slices. In the case of color, FD samples have higher hue angle, whiteness index; and lower chrome value and yellowness index. But the browning index of slices is low because of an absence of oxygen during vacuum freezing. The dried slices have a rehydration ratio of 7.80±1.06, and the slices need a very minimum time to obtain the RR. Likewise, the shrinkage occurs during FD was very low, but the cell wall rupture that occurred during FD improved the porosity of the slices. When coming to the mechanical property, FD slices have higher hardness, fracturability, and chewiness values than a fresh sample. The nutritional compounds and volatile compounds are well preserved during the FD. So, FD can be used to preserve the nutritive and flavor components of spices with minimum physical changes in the samples.

Declaration of conflict of interest

The authors declare that there is no conflict of interest.

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