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EFFECT OF CASSAVA/SOY FLOUR SUPPLEMENTATION ON THE CHEMICAL, FUNCTIONAL AND BAKING QUALITIES OF BREAD

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ABSTRACT

The study evaluated the suitability and acceptability of cassava composite bread in order to enhance the utilization of cassava flour for production of baked goods which will help alleviate the dependency of developing nations on imported wheat. Flour samples were processed using standard methods. Effect of supplementing wheat flour with cassava flour up to 40% level and 20% soy flour on the physico-chemical, functional, sensory and anti-nutrient properties of the flour samples were studied. Baking qualities and microbial evaluation of wheat/cassava/soy bread was also carried out. Thirteen different bread samples were produced from composite flour consisting of wheat, cassava and soybean flours in the range of following proportions: 60-100%, 10-40% and 10-20% respectively. The protein content decreased as the cassava flour increased, but increased as soy flour was increased. Fibre content increased with increase in the cassava and soy flour ranging from 0.30% to 1.12%. The fat content decreased with increase in cassava flour but increased with a increase in soy four ranging from 2.23% to 3.40% for sample without soybean flour, 6.61% to 9.06% for samples with soybean flour. It was a similar trend for carbohydrate. Sedimentation value of the flour ranged from 11 to 22%, falling Number ranged from 413 to 424 s while the damage starch ranged from 93.01 to 94.54.Oven spring and specific volume of breads decreased with increase in cassava supplementation. Bread baked with up to 20% cassava and 20% soy flour was not significantly different in most sensory attributes from the control sample.

Keywords: Baking, bread, cassava, soybean, supplementation

INTRODUCTION

Bread is an important staple food in Nigeria. It is however, relatively expensive, being made from imported wheat that is not cultivated in the tropics for climatic reasons (Olaoye et al., 2006).The major or mandatory ingredients in bread making are flour, water and yeast (Akobundu and Osuji, 2006). The flour should have good amylase activity, the moisture content should be less than 14% and the colour or appearance should be satisfactory (Giami et al., 2004).

Bread and other baked products are convenient food items that can be fortified with protein for people suffering from protein malnutrition especially children. Fortification of wheat flour with legumes or oilseed flour and carbohydrate rich food resources would further enhance the utility of local raw materials and improve the nutritive composition of composite bread.

The idea of substituting part of wheat with other starchy crops is not new. It would therefore advantageous if imported wheat could be reduced or even eliminated and the demand for baked products such as bread could meet the use of domestically grown products other than wheat.

Several studies have been carried out to find ways of partially substituting wheat flour with other sources of flour or replacing wheat altogether (Bokanga, 1995). With the constant increasing consumption of bread and other baked products in many countries, the composite flour programme promises to save significant amount of foreign exchange, provide a traditional nutritious food to more people at lower cost and to utilize indigenous crops to a greater extent. On the light of this, cocoyam, cassava, taro and other tubers crops have been found to be an alternative source of major raw materials for bread making (Edward, 1974; Giami et al., 2004).

The use of cassava flour for production of baked goods if feasible would help to lower the dependency of developing nations on imported wheat. Cassava (Manihot esculenta Crantz) is a major root crop in the tropics and its starchy roots are significant source of calories for more than 500 million people worldwide

(FAO, 2000; Mroso, 2003), it is also a perennial crop, it grows well in the tropical poor soil and can withstand drought. In spite of its high cyanide content, cassava products with encouraging international market efforts are being made to achieve the quality standards especially if processed to reduce its cyanide content (Rosling, 1994; FAO, 2004).

The soybean is a species of legume native to East Asia, widely grown for its edible bean which has numerous uses. Soy flour refers to soybeans ground finely enough to pass through a 100-mesh or smaller screen where special care was taken during desolventizing to minimize denaturation of the protein. This study is aimed at assessing the quality of wheat-cassava composite flours, determining the baking properties and acceptability of bread from the composite flour.

MATERIALS AND METHODS

The cassava roots, wheat soybean were obtained from research farm, Federal University of Technology, Akure while other recipes for bread production were purchased from King's Market Akure, Ondo State, Nigeria.

Processing of Cassava Flour

The fresh cassava roots were processed into flour using a method described by Oni, et al. (2012). The cassava roots were peeled manually with a sharp knife, washed and grated in a locally fabricated mechanical grater It was thereafter packed into Hessian sack and dewatered by pressing in a mechanical press (Addis Engineering Nigeria, Limited) to dewater the mesh. The dewatered lump was pulverised with hands and sifted on a local raffia made sieve of mesh (0.3cm x 0.3cm) mounted on a rectangular wooden frame $40cm^2$ to remove the fibres. The sifted cassava meal obtained was allowed to dry in a cabinet dryer. The dried meal was milled and sieved with a fine mesh (200µm) and later packaged in High Density Polyethylene film and kept under refrigerated storage until ready for further analysis.

Processing of Soybean Flour

According to the method of Oluwamukomi et al. (2005), 1 kg of soy bean (Glycine max) were sorted and washed and then boiled in water at 100^oC for 30 minutes. It was dehulled manually, dried with a cabinet dryer, milled with an attrition mill. The flour was packaged in high density polyethylene and stored until ready for further use.

Bread Production.

570g of composite flour sample was weighed along with the required amount of water (about 360ml), along with 16g of fat, 10g of salt, 8g yeast and 16g sugar to obtain dough, which was kneaded on a pastry-board to smoothen it. The dough was initially fermented for 2 hours at 30^0C before being subsequently knocked back by kneading to expel carbon dioxide and tighten-up the dough to improve the texture of the final product. The secondary fermentation also lasted for 2 hours at 30^0C. The dough were then sized and moulded into the baking pans for final proving at 30^0C for 2 hours. Dough baking was carried out in the oven at a temperature of 230^0C for 25 minutes and allowed to cool (Ihekoronye and Ngoddy, 1985). This process was carried out on 13 different bread samples with composite flour of different proportions.

Proximate compositions of composite flour:

This was determined according to the standard method of AOAC (2005). Fat analysis was determined using soxhlet extractor, Moisture was also determined using Air- oven method as well as Ash was determined. The crude protein was determined by multiplying the total nitrogen by the total nitrogen by 6.25. The carbohydrate was obtained by difference. The pH was measured with a pH meter (Meter Teledo MP: 220) while the titratable acidity was determined using the method of Oshodi (1992). pH was determined using pH meter (model number Jenway 350).

Functional Properties of composite flour:

Water and oil absorption properties of the composite flour were determined using the method of Sathe et al. (1981). Foaming capacity and stability were determined according to the methods described by Desphande et al. (1982).The method of Onwuka (2004) was used to determine the bulk and loosed densities of the flour samples while Okaka and Potter(1977) method with some modifications was used for determining the swelling capacity. Determination of sedimentation value was carried out according to International Association for Cereal Science and Technology (ICC) standard, (2001). Solubility was determined by the method of Sathe et al., (1982) with some modifications.

Falling Number Determination of damage starch was determined by adding an enzyme to hydrolyse the starch granules to glucose. The reaction was observed using iodine solution to check for the starch available. Determination of Hydrogen Cyanide (HCN) was determined using alkaline pikrate colorimetric method by Balagopalan et al. (1998). Phytate Determination was determined by the method of AOAC (2000).

Baking Test on Bread

After baking, bread loaves were cooled for 1 hour at room (ambient) temperature and subjected to the baking test analysis for Loaf volume, Specific loaf volume, and Oven spring. The bread volume was determined using a graduated cylinder 500 ml capacity and a wooden box large enough to contain the loaf of bread in such a manner that the top surface of the loaf remains about 1.5 cm below the surface of the box.

When the bread was placed in the box, the millet grains were displaced and the volume of the grains in the box was read on the cylinder. This corresponds to the volume of the loaf and the specific volume (volume to mass ratio) was calculated. Weight of loaf samples were taken and specific loaf volume was determined by dividing the loaf volume by its corresponding loaf weight. Oven spring was determined from the difference in height of dough before and after baking.

Sensory Evaluation: This was performed two hours after baking to evaluate loaf appearance, crust colour, crumb colour, taste/flavour and overall acceptability of the bread sample. The bread samples were sliced into pieces of uniform thickness and served with water. Twenty panel members (familiar with quality attributes of local bread) were randomly selected from students and staff of the Department of Food Science and Technology, to perform the evaluation. Panelists will evaluate bread samples on a 9 point hedonic scale quality analysis (Larmond, 1997) with 9 = liked extremely, 8 = liked very much, 7 = liked, 6 = liked mildly, 5 = neither liked nor disliked, 4 = disliked mildly, 3 = disliked, 2 = disliked very much and 1 = disliked extremely.

Microbiological Determination

The aerobic plate count was carried out on different bread samples using the method of Olaoye et al.(2006)., 10g of each sample were taken aseptically and homogenized in 90ml sterile distilled water, in a blender for about 2 minutes. The serial dilution was done, by taking 1ml of homogenate into 9ml of sterile water, dispensing in test tubes. 1ml of each dilution factor was pour plated in sterile petri dishes, using the nutrient Agar, incubated at 37^0C for 24 hours. Counts of visible colonies were made and expressed in Cfu/g (colony forming unit per gram) sample. The procedures were repeated for fungi, using (Eosin Methylene Blue) culture and 1ml of acetic acid (to prevent bacterial growth) was added and count of visible colonies done after incubating for 72 hours and expressed in Cfu/g (colony forming unit per gram) sample.

Statistical Analysis: Triplicate samples from the analysis were subjected to statistical analysis by analysis of variance (ANOVA) using SPSS version 16 computer program. Means were separated using Duncan New Multiple Range technique (Steel et al., 1997).

RESULTS AND DISCUSSION

The result of the proximate composition of soy enriched wheat-cassava composite flour samples is presented in Table 1. The moisture content of 100% wheat flour was 11.27% while composite samples ranged from 11.13% to 11.73% for samples without soybean flour and 10.13% to 11.20% for samples with soybean flour. Since different food materials have different capacity for absorbing/retaining moisture which may exist as occluded or absorbed water. Therefore it can be deduced that even at the high baking temperature, some moisture will be found in the samples as observed during the study (Eddy, 2004; James, 1984). The crude protein content of 100% wheat flour was 13.69% and composite bread samples ranged from 9.20% to 13.55% for samples without soybean flour and 11.72% to 16.26% for samples with soybean flour. The protein content decreased as the cassava flour increased, although, there was significant increment in samples with soybean flour. Generally, the protein content of all the samples were relatively low because wheat and cassava are poor sources of protein. The amount of protein required by humans is between 30 to 40% and bread of good quality should have at least 10% protein content (Eke et al., 2013; Oyenuga, 1992).Generally, the fat content of the bread decreased in the order of supplementation ranging from 2.23% to 3.40% for samples without soybean flour, 6.61% to 9.06% for samples with soybean flour and 2.80% for 100% wheat flour. However, there was a slight significant difference between the fat contents of 100% wheat flour and the composite flour without soybean flour and significant difference for samples with soybean flour. The amount of fat content that should be present is between 2 to 5% as reported by Eke et al. (2013). Fibre content of 100% wheat-flour was 0.30% which is low compare to the fibre content of the composite bread samples ranging from 0.30% to 1.12% for samples without soybean flour and 0.30% to 1.93% for samples with soybean flour which were higher than those of 100% wheat bread as the level of supplementation increases. The minimum amount of fibre that should be present is 0.5% (Eke et al., 2013). Fibre contributes to the health of the gastrointestinal system and metabolic system in man (Bub et al., 2003). Generally, the ash content of 100% wheat flour is 1.41%, for composite flour without soybean flour range from 1.68% to 1.79% and 1.67% to 1.79% for flour samples with soybean flour and the amount of ash that should be present in bread is about 0.6%, composite flour increases as the level of supplementation increases implying that the inorganic nutrients in the composite flour is richer than that of wheat flour. The carbohydrate of wheat flour sample is 81.80% with higher values from 81.04% to 85.66% obtained in composite flour without soybean flour and lesser values from 72.48% to 78.64% in samples with soybean flour. This observation may be attributed to the high content of carbohydrate in cassava. Carbohydrate supplies quick source of metabolized energy and assists in fat metabolism. Table 2 shows the results for functional properties of the flour samples. The bulk density was highest for wheat flour of about 0.84g/ml, while the bulk density of composite flour which ranges from 0.81g/ml to 0.75g/ml for samples without soybean flour and 0.75g/ml to 0.78g/ml for samples with soybean flour was reducing as composition of flour sample increases. The bulk density is influenced by particle size and the density of the flour and is important in determining the packaging requirement and material handling. Water absorption capacity of composite flours decreased significantly ($p \leq 0.05$). 100% wheat flour was 2.27g/ml while composite flour samples ranged from 2.2g/ml to 1.60g/ml, on addition of cassava and soybean flour in different proportion. The water absorption capacity is a function of water holding ability of the flour sample and is desirable in food systems to improve yield and consistency. The water absorption capacity for 100% wheat flour was2.27% while flour samples without soybean flour, the values ranged from 1.60 to 2.20% and for samples with soy bean flour ranged from 1.60 to 2.20%. The water absorption capacity of this effect was probably due to loose association of amylose and amylopectin in the native granules of starch and weaker associative forces maintaining the granules structure Oil absorption capacity of composite flours also decreased significantly ($p \leq 0.05$) on addition of cassava and soybean flour in varying proportions. Oil absorption capacity for 100% wheat flour was 2.53g/ml while samples without soybean flour ranged from 2.07g/ml to 2.84g/ml and 1.53g/ml to 2.15g/ml for samples with soybean flour. The result of the swelling capacity showed that there significant difference between the 100% wheat flour and the composite flour at ($p \leq 0.05$), 100% wheat flour 12.33ml, composite flour samples without soybean flour ranged from 11ml to 13.00ml and 12.00ml to 15.00ml for sample with soybean flour. The swelling capacity is an indication of presence of amylase which influences the quantity of amylose and amylopectin present in wheat flour. The swelling capacity of flour granules is an indication of the extent of associative forces within the granule while its variation indicates the degree of exposure of the internal structure of the starch present in the flour to the action of water. The emulsion capacity of the composite flours varies in different proportion. The samples without soybean flour ranged from 58.35% to 78.95% and 59.39 to 89.59% for samples with soybean flour. The emulsion capacity of 100% wheat flour was61.73%.According to Lin et al. (1974), the emulsion capacity (EC) of cassava flour falls within the range of 8.3 to 32.5%. Data on foaming capacity and stability of wheat-cassava and soybean flour blends suggest that the foaming capacity of the blends ranged from 3.33 to 14.0% for samples with soybean flour and 8.67 to 21.33% for samples without soybean flour compared with 100% wheat flour which had 28.67%. Similarly, foam stability (FS) ranged from 85.18 to 95.06% for samples without soybean flour and 82.64 to 96.78% for samples with soybean flour. The blends depicted high foam stability and may find application in baked and confectionery products. Products' ability to foam is related to the rate of decrease of the surface tension of air/water interface caused by absorption of protein molecules. The result of sedimentation is presented in Table 3. The wheat cassava composite flour samples ranged from 11 to 22% for samples with soybean flour and 20 to 26% for samples without soybean flour compared with 100% wheat flour which had 20%. Sedimentation is the sinking of solid in liquid and measures the volume of the swollen gluten protein.

Falling number result shows that wheat cassava composite flour ranged from 413 to 424 seconds for samples with soybean flour and 398 to 424 seconds for samples without soybean flour compared with 100% wheat flour which had 257 seconds. Table 4 presents the sensory properties carried out on bread samples for 100% wheat and composite flour. Aroma is related to taste, a good level of aroma intensity influences taste. The texture of samples ranged with the highest obtained in the control and lowest in the 40% substituted composite bread. This showed that the level of supplementation influences the quality of dough that could provide the texture known for bread. The result for 10% wheat-cassava composite bread sample was comparable to that of the control. This suggests that the quality of bread that can be produced from wheat-cassava flours mixtures depends on the level of substitution. Preference to buy the samples scored a mean that ranged from 7.90 to 8.40 with the highest been recorded for 100% wheat bread, and lowest for 40 and 30% cassava substituted composite breads.

Table 5 shows the result for anti-nutrient for the flour samples which ranged from 0.17 mg/kg to 0.28 mg/kg for samples without soybean flour and 0.17 to 0.40 mg/kg for samples with soybean flour which is minimal dose recommended for human consumption. The result for phytate ranged from 112.82 mg/100g to 169.23mg/100g for samples without soybean flour and 225.64 to 282.05 mg/100g for samples without soybean flour. Results in Table 6 show the baking qualities of the bread. Significant differences in oven spring and specific volume of breads were observed. Incorporation of cassava and soybean flours in the samples decreased the oven spring, loaf volume and specific loaf volume. Reduction in volume and quality as a result of blending wheat flour with more than 10% cassava and soybean flour will lead to reduction of gluten effect on the samples. The microbiological evaluation of the bread sample is presented in Table 7. The fungi and bacteria count for bread samples were 1 x 10^{-4} cfu/ml for WF 80%-CF 20%, WF 70%-CF 30%, WF 70%- CF 30%-SF 20%, WF 60%-CF 40%-SF 20%, and WF 100% (Control). The values for mesophyll aerobes, molds and yeasts were found within the limits indicated by Moreno–Álvarez et al. (2007).

CONCLUSION

Although the proximate composition of the composite flour samples were different from that of control (100% wheat flour) depending on the quantity of cassava and soybean flour added, it has been found that bread baked with 10% composite flour was rated higher than the control especially in aroma, colour, flavour and general acceptability although there was no significant different in most sensory attributes from the control. Bread baked from 30% and 40% composite flour showed low mean score to all the attributes. These results showed that bread produced from 10% composite flour could be a viable alternative to achieve the desired stable economic, food security and health.

Table 1:Proximate Composition of Flour Samples (%d.b)

Sample	Ash (%)	Crude fat (%)	Crude Protein (%)	Crude fibre (%)	Carbohydrate (%)	Moisture (%)
WF 90% CF10%,	1.71^{a}±0.15	3.40^{e}±0.15	13.55^{c}±0.23	0.30^{g}±0.13	81.04^{c}±0.39	11.73^{a}±0.31
WF 80% CF 20%,	1.68^{a}±0.09	3.22^{e}±0.16	11.57^{d}±0.50	0.30^{g}±0.13	83.23^{b}±0.64	11.47^{a}±o.42
WF 70% CF 30%	1.79^{a}±0.12	3.15^{e}±0.68	10.49^{e}±0.30	0.98^{e}±0.14	83.93^{b}±0.507	11.60^{a}±0.40
WF 60% CF 40%	1.79^{a}±0.17	2.23^{f}±0.11	9.20^{f}±0.40	1.12^{de}±0.23	85.66^{a}±0.80	11.13^{abcd}±0.23
CONTROL 100% WF	1.41^{b}±0.02	2.80^{e}±0.61	13.69^{c}±0.26	0.30^{a}±0.12	81.80^{c}±0.98	11.27^{abc}±0.12

Values are means of triplicate readings. Means followed by the same letter of alphabets vertically are not significantly different (p ≤0.05) at 5% level

Table 2: Functional Properties of the flour

Sample	Water absorption (g/ml)	Oil Absorption (g/ml)	Foaming Capacity (%)	Foaming Stability (%)	Swelling Capacity Ml	Emulsion Capacity (%)	Bulk Density (g/ml)
WF 90%, CF 10%	2.20^{ab} ±0.00	2.84^{a} ±0.13	21.33^{b} ±2.31	85.18^{e} ±1.41	12.67^{ab} ±0.58	61.48^{ef} ±2.09	0.81^{b} ±0.01
WF 80%, CF 20%	2.10^{bc} ±0.00	2.45^{b} ±0.13	13.33^{cd} ±1.15	89.49^{d} ±1.57	11.00^{b} ±1.00	58.35^{f} ±4.69	0.77^{d} ±0.00
WF 70%, CF 30%	1.60^{e} ±0.00	2.15^{c} ±0.14	12.00^{cd} ±2.00	94.07^{abc} ±2.63	13.00^{ab} ±2.65	72.00^{cd} ±2.00	0.76^{de} ±0.00
WF 60%, CF 40%	1.70^{e} ±0.10	2.07^{cd} ±0.00	8.67^{efg} ±2.31	95.06^{abc} ±2.02	12.67^{ab} ±1.53	78.95^{bc} ±1.00	0.75^{ef} ±0.01
WF 100% (CONTROL)	2.27^{e} ±0.12	2.53^{b} ±0.00	28.67^{a} ±1.15	82.64^{e} ±2.45	12.33^{ab} ±0.58	61.73^{ef} ±1.55	0.84^{a} ±0.02

Values are means of triplicate readings

Means followed by the same letter of alphabets vertically are not significantly different (p ≤0.05) at 5% level.

Table **3: Functional Properties**

Sample	Bulk density (g/ml)	Sedimentation (%)	Water absorption Capacity (g/g)	Oil absorption Capacity(g/g)	Falling Number (sec)
WF 90%, CF 10%	0.646	26	0.94	0.9	398
WF 80%, CF 20%	0.635	24	0.95	0.8	420
WF 70%, CF 30%	0.646	22	0.96	0.81	424
WF 60%, CF 40%	0.641	20	1.2	0.82	424
WF 100% (CONTROL)	0.667	20	1.8	0.89	257

Values are means of triplicate readings

Means followed by the same letter of alphabets vertically are not significantly different ($p \leq 0.05$) at 5% level.

Table 4: Sensory Properties.

SAMPLES	APPEARANCE	AROMA	TASTE	CRUST TEXTURE	CRUMB TEXTURE
WF 100% (CONTROL)	5.35^{cdef}	5.25^{f}	4.75^{f}	4.65^{g}	6.60^{e}
WF 90%, CF 10%.	8.25^{a}	7.55^{a}	7.80^{a}	7.85^{a}	8.00^{a}
WF 80%, CF 20%	5.35^{cdef}	7.00^{ab}	6.70^{bcd}	6.50^{bc}	6.70^{bcd}
WF 70%, CF 30%	5.50^{cdef}	6.45^{bcd}	5.90^{cde}	5.90^{cde}	6.15^{cde}
WF 60%, CF 40%	4.95^{def}	6.60^{abc}	5.25^{ef}	5.70^{cdef}	$6.15d^{e}$

Means followed by the same letter of alphabets vertically are not significantly different ($p \leq 0.05$) at 5% level.

Table 5: Anti Nutrient

Sample	Cyanide mg/kg	Phytate mg/100g
WF 90%, CF 10%.	$0.28^{a}\pm0.20$	$169.23^{cde}\pm0.00$
WF 90%, CF10%, SF10%.	$0.17^{ab}\pm0.00$	$263.25^{a}\pm32.57$
WF 90%, CF 10%, SF 20%.	$0.28^{a}\pm0.20$	$282.05^{a}\pm56.41$
WF 80%, CF 20%.	$0.17^{ab}\pm0.00$	$150.43^{de}\pm32.57$
WF 80%, CF 20%, SF 10%.	$0.17^{ab}\pm0.00$	$244.44^{ab}\pm32.57$
WF 80%, CF 20%, SF 20%.	$0.28^{a}\pm0.20$	$263.25^{a}\pm32.57$
WF 70%, CF 30%.	$0.28^{a}\pm0.20$	$131.62^{de}\pm32.57$
WF 70%, CF 30%, SF 10%.	$0.40^{a}\pm0.20$	$225.64^{abc}\pm0.00$
WF 70%, CF 30%, SF 20%.	$0.40^{a}\pm0.20$	$282.05^{a}\pm56.41$
WF 60%, CF 40%.	$0.17^{b}\pm0.00$	$112.82^{e}\pm0.00$
WF 60%, CF 40%, SF 10%.	$0.17^{ab}\pm0.00$	$244.45^{ab}\pm32.57$
WF 60%, CF 40%, SF 20%.	$0.28^{a}\pm0.20$	$263.25^{a}\pm32.57$
WF 100% (CONTROL)	$0.00^{b}\pm0.00$	$188.03^{bcd}\pm32.57$

Means followed by the same letter of alphabets vertically are not significantly different ($p \leq 0.05$) at 5% level.

Table 6: Baking Quality

Sample	Specific loaf Volume (cm^3/g)	Seed displaced by loaf (g)	Loaf volume (cm^3)	Oven spring (cm)
WF 90%, CF 10%.	$2.17^c \pm 0.02$	$4008.33^{cd} \pm 14.43$	$1086.67^d \pm 11.55$	$3.067^a \pm 0.06$
WF 80%, CF 20%.	$2.38^b \pm 0.02$	$4058.33^b \pm 14.43$	$1188.33^b \pm 10.41$	$2.60^c \pm 0.10$
WF 70%, CF 30%.	$2.08^d \pm 0.01$	$3850.00^e \pm 25.00$	$938.33^g \pm 10.41$	$1.90^f \pm 0.00$
WF 60%, CF 40%.	$2.21^c \pm 0.00$	$4050.00^{bc} \pm 50.00$	$1160.00^c \pm 0.00$	$2.00^f \pm 0.00$
WF 100% (CONTROL)	$3.12^a \pm 0.02$	$4425.00^a \pm 25.00$	$1560.00^a \pm 10.00$	$3.13^a \pm 0.12$

Means followed by the same letter of alphabets vertically are not significantly different ($p \leq 0.05$) at 5% level.

Table 7:Microbiological Properties

Sample	fungi colonies (cfu/ml)	e.coli colonies (cfu/ml)	Bacteria colonies (cfu/ml)	staphylococcus colonies (cfu/ml)
WF 90%, CF 10%.	NP	NP	NP	NP
WF 90%, CF10%, SF10%.	NP	NP	NP	NP
WF 90%, CF 10%, SF 20%.	NP	NP	NP	NP
WF 80%, CF 20%.	1×10^{-4}	NP	2×10^{-4}	NP
WF 80%, CF 20%, SF 10%.	NP	NP	NP	NP
WF 80%, CF 20%, SF 20%.	NP	NP	NP	NP
WF 70%, CF 30%.	1×10^{-4}	NP	1×10^{-4}	NP
WF 70%, CF 30%, SF 10%.	NP	NP	1 X 10-4	NP
WF 70%, CF 30%, SF 20%.	1×10^{-4}	NP	1×10^{-4}	NP
WF 60%, CF 40%.	NP	NP	NP	NP
WF 60%, CF 40%, SF 10%.	NP	NP	NP	NP
WF 60%, CF 40%, SF 20%.	1 X 10-4	NP	1.33×10^{-4}	NP
WF 100% (CONTROL)	1×10^{-4}	NP	1.67 X 10-4	NP

Means followed by the same letter of alphabets vertically are not significantly different ($p \leq 0.05$) at 5% level

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