ASSESMENT OF TUMERIC HERBAL PLANTS FOR THEIR PHYTOCHEMICAL CONTENTS AND INVITRO ASSAY OF THEIR ANTIOXIDANT POTENTIALS

BY

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Abstract

Phytochemical, Antioxidant potentials (DPPH) and proximate composition of Tumeric was investigated. These herbs and their parts were sourced from Kwara State polytechnic campus, Ilorin, Nigeria. The plant parts were grinded with mortar to increase surface area before extraction. The phytochemical screening was carried out using standard methods. The antioxidant activity of plant extracts was determined using 2, 2-diphenyl-1-picrylhydrazyl (DPPH). The. The herb show high antioxidant activities with DPPH is well above 50% even at all the dilution factors, the invitro activities shows that the three herbs can be a good source of antioxidant and could also be used to manufactured drugs for antistress, the extract could be used to alleviate heat stress in broiler chicken. The three plants are well above IC 50 values for antioxidant activities and also contribute to nutritional value of the feed because they bare good sources of protein and energy.

Keywords: Total phenol/flavonoid contents, Free radical, Scavenging, Antioxidant, Cancer and medicinal plants

Introduction

Climate change has brought a lot of problems and questions that are unanswered, one of such is its effect on broiler birds especially in the tropical region and the heat stress resulted to poor growth, which arises from poor feed intake, nutrient utilization, physiological condition, immune function, intestinal ecology, morphology and even mortality therefore heat stress has been an important factor that should greatly be considered in broiler production (Sugiharto et al, 2017). A lot has been done to solve this problem including thermal control pen which is very expensive, nutritional intervention, use of antibiotics and other antioxidant drugs has been tried by scientist and poultry farmers but the expected result was not achieved (Singh et al., 2012). Moreover, heat stress induces oxidative stress in the body, and oxidation of substrates produces free radicals and other reactive oxygen species that can cause serious damage to the cells and organs (Halliwell and Gutteridge, 2018). However, it is well known that antioxidants can be used for elimination or mediation of the adverse effects of heat stress. A biological antioxidant is known as "any substance that, when present at low concentrations compared to those of an oxidizable substrate, significantly delays or prevents oxidation of that substrate" (Halliwell and Gutteridge 2018). Traditional herbal medicines are effective in reducing adverse effect of heat stress due to their antioxidant activity especially now that the use of antibiotics and vetinary drugs are less encouraged in poultry production because of their side effects (Zhang et al, 2017). In this study, both quantitative and qualitative phytochemical screening and invitro assay of the
antioxidant properties and Nutritional content of Tumeric plant

Material and Methods

Pulp of *Tumeric* is obtained from surrounding of Kwara State Polytechnic, Ilorin with 8.7063° N, 4.4622° E. The dried pulp was manually scraped from the seeds and kept in an air-tight container. The seeds and leaves were washed and sun-dried for 10 h separately and then milled into fine powder.

Extraction and phyto-constituents determination

Extraction

The bioactive compounds of the fruit, pulp and leaves of *Adasonia digitata, Pakia baglobosa, Moringa Olifera, Tamarind and Tumeric* were extracted with ethanol to obtain the extracts of the two herbs. Abioye and Ama (2015)

Alkaloids

One milliliter of aqueous extract of the sample was suspended in 1 ml of distilled water and drops of diluted hydrochloric acid (2 M). To the resulting mixture 1 mL Dragendorff's reagent was added. An orange-red precipitate shows the presence of alkaloids . Jongrungruangchok *et al*., (2017)

Flavonoids

To 5 g of the sample, 5 ml of methanol was added and filtered. To a portion of the filtrate, 2 mL of sodium hydroxide solution was added. Three drops of diluted hydrochloric acid were added to the resultant mixture. The appearance of a yellow solution when the sodium hydroxide solution was added which became colourless when dilute hydrochloric acid was added, this confirmed that flavonoids are present He et al., (2018)

Saponins

Two grams of powdered plant material were suspended in 5 mL distilled water. The mixture was vigorously shaken for 1 min. Persistent copious lather for more than 5 min shows the presence of saponins Fahey, (2015)

Phenols

To 1 g of powdered plant material, 2 mL of distilled water was added after which the mixture was filtered. Ferric chloride (10 %) solution was then added to the filtrate dropwise. A dark green color inferred a positive result for the presence of phenols DanMalam et al., 2011

Terpenoids

Five grams of the samples was suspended in chloroform (2 mL) after which 3 ml of concentrated sulfuric acid was added to the resulting solution. The formation of a reddish-brown colored interface showed a positive inference of terpenoids Jimoh and Olatidoye, 2009

Tannins

To 5 g of the sample, 10 ml of distilled water, was added heated, and filtered. One milliliter of FeCl f iltrate. A blueish-green precipitate shows the presence of tannins Hekmat et al., 2015

DPPH free radical scavenging assay

A DPPH solution (0.2 mM) was prepared (0.0078864 g of DPPH in 100 ml methanol). In a microtiter plate, 150 µl of 0.2 mM DPPH was added and followed by a 50 µl of standard (ascorbic acid; , ranging 40 µg/ml to 200 µg/ml) or extract (BO/DBEE/BFWE; ranging 200 µg/ml to 1000 µg/ml). The mixture was left to stand in the dark at 27 ◦C for 30 min. From the resulting solution absorbances were taken against a blank at 517 nm with a SPECTROstar NANO, spectrophotometer. The scavenging activity of the extracts against DPPH radical was calculated as scavenging activity (%) =(DPPHabs – Extractabs/DPPHabs) ×100 Jongrungruangchok et al., 2017

Proximate composition analysis

The nutritional composition such as moisture, ash, protein, fat, fiber, and carbohydrate content of the seeds and fruit pulp were determined using techniques certified by the Association of Official Agricultural Chemists (AOAC) with slight modifications (Nkukwana et al., 2014). Each analysis was done in triplicates. A weighed sample (5 g) was placed in a preheated oven at 105 ◦ C and dried to a contant weight to measure the percentage moisture. The micro-Kjeldahl technique was used to determine the crude protein concentration. Percentage fatin the seeds and fruit pulp were determined by extracting from a 5 g sample in petroleum ether through the Soxhlet extraction method Nkukwana et al., 2014. After digesting a measured amount of the fat-free residue produced from the crude fat determination, the crude fiber was evaluated gravimetrically. With 1.25 % sodium hydroxide and 1.25 % sulfuric acid, the digestion was carried out under reflux. The sample was burned for 10 h at 600 ◦ C in a muffle furnace to ascertain its ash content. Carbohydrate percentages of the various samples were estimated as follows: Carbohydrate (%) = 100 - % yield [moisture + fat + ash + protein + fiber] The Total Energy values of the plant materials were obtained by calculations according to the Atwater factor as shown below: Energy value (kcal) = 4(% crude protein) + 9(% crude fat) + 4(% carbohydrate).

FRAP

1ml of water and 80µl of test bee product sample was pipetted in to a standard 4ml plastic cuvette. 600µl of the incubated FRAP reagent was added to the cuvette, which was briefly inverted to mix the solutions. A reagent blank was also prepared as described above but 80µl of water was added instead of a test sample.
Change in absorbance at 593nm (a result of reduction of the Fe3+-TPTZ complex to the blue Fe2+ TPTZ complex at low pH) was recorded at exactly four minutes using a spectrophotometer. Each test sample dilution was tested in triplicate to allow a mean absorbance to be calculated. A number of dilutions of each bee product were tested allowing dose response curves to be produced.

Results and Discussions

The qualitative phytochemical screening in table 1 shows that turmeric pulp has all of alkanoids, flavonoids, Saponins, Terpenoids while phenol and tannin are absent, the quantitative phytochemicals table 2 shows that alkanol is the highest (9.30) and tannin is the least of them all (0.83), table 3 shows the mineral content with calcium been the highest (11.129) and Copper been the least of them all (0.105), table 5 shows the three vitamins that are concern with heat stress are A, C and E with Vitamin C been the highest (142.048) and Vitamin E is the least (24.606). The DPPH is shown in table 6 and it gives 83.05% as the highest inhibition comparable with Vitamin C that is the standard that has 89.50% inhibition factor higher, this is in line with FRAP that has the highest values at 50 conc (20.803) and least at 3.125 conc (18.372). The proximate composition in table 4 shows that it has a metabolizable energy of 360.08Kcal/kg while the protein content is 19.40% this makes Tumeric a good feed ingredient for both human and livestock.

Table 1: Qualitative Phytochemical screening of Tumeric pulp plant

samples Parameters	Tumeric
Alkaloids (Wagner's test)	++
Flavonoids (Alkaline test)	++
Saponins (Foam test)	++
Phenols (Ferric chloride test)	-
Terpenoids	+
Tannins	-

Table 2: Quantitative phytochemical screening of Tumeric pulp plants

Parameters	Tumeric
Alkaloids	9.30
Flavonoids	4.15
Saponins	3.15
Phenols	1.02
Terpenoids	0.92
Tannins	0.83

Table 3: Mineral content of Tumeric plant (ppm)

Samples	Ca	Zn	Cu	Fe	K	Mg	Na	Cr	Mn	Ph
Tame	11.129	0.457	0.105	3.798	9.031	6.931	2.538	0.182	0.332	2.234

Tame = tameric pulp plant

Table 4: Proximate composition of Tumeric plants

Samples	Moisture (%)	Ash(%)	Crude fibre (%)	Crude protein(%)	Crude fat(%)	Carbohydrate(%)	Energy(kg/kcal)
Tamar	14.75	3.75	0.86	19.40	7.50	53.75	360.08

Tam = tamarind plant

Table 5: Vitamin content of Tumeric pulp plants

Vitamin Samples	A(mg/kg)	E(mg/kg)	C(mg/kg)
Tam	27.647	24.606	142.048

Tam = tamarind plant

Table 6: TABLE OF VALUE FOR DPPH (2,2-Diphenyl-1-picrylhydrazyl) Invitro FOR SOME HERBAL PLANTS
(Abs control = 0.413)

Sample name	Dilution factor	Average Abs	%inhibition
Vitc (Shd	50.000	0.015	89.50
	25.000	0.031	92.49
	12.500	0.054	86.92
	6.125	0.082	80.15
	3.125	0.126	70.22
Tum	50.000	0.070	83.05
	25.000	0.081	80.39
	12.500	0.121	70.70
	6.125	0.133	67.80
	3.125	0.164	60.29

Tum =
Tumeric

FRAP (Ferric reducing antioxidant power)

Conc.	Tumeric	Vit C
3.125	18.372	20.705
6.25	18.960	23.588
12.5	19.745	26.686
25	20.058	29.000
50	20.803	31.980

Tum = Tumeric

Conlusion

i-The herb is good for use for anti oxidant because of their high anti oxidant properties
ii- The herbs are well above IC 150 slope
iii-The herb could be use for drug manufacturing

Recommendation

It could be included in livestock feed for treatment because of its medicinal qualities

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