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#### ABSTRACT

Cloves are a group of aromatic plants that are used widely due to their positive effects on the growth and health of poultry, probably as a result of their immune-stimulatory properties. Clove (*Syzygium aromaticum*) has attracted attention due to its potent antioxidant activities standing out among other spices. The present study was carried out to determine the antioxidant activity of clove bud using ascorbic acid as standard and 2, 2-diphenyl-1-picrylhydrazyl (DPPH) as free radical. The DPPH inhibition activity was carried out using the standard method. The DPPH inhibition activity of the clove extract was observed to increase from 18.17% to 65.12% with increasing concentration of the clove extract from 0.2 to 1.0 mg/ml. The clove bud extract was observed to have DPPH inhibition activity and exhibited a dose-dependent trend. The IC<sub>50</sub> value for the clove and ascorbic acid were 0.725 and 0.320 respectively. Clove bud has the potential to serve as a natural substitute for synthetic antioxidant feed additives.

KEYWORDS: *Syzygium aromaticum*; natural antioxidant; feed additive; free radicals

#### INTRODUCTION

Spices are mostly used for seasoning and flavouring in food preparation. Clove is an ancient spice, that is obtained from the scented dried flower bud from the clove tree. It is an invaluable multipurpose spice that possesses a strong spicy aroma (Hussain *et al.*, 2017). The major bioactive volatile compound in clove bud is eugenol, this may one of the active substances in clove bud responsible for its antioxidant activity (Kamatou *et al.*, 2012; Gaspar *et al.*, 2018).

Clove bud is a good source of phenolic compounds such as eugenol, eugenol acetate, and gallic acid. The presence of these substances has been linked with their antimicrobial and antioxidant property (Cortés-Rojas *et al.*, 2014).

There is a rising need for the replacement of synthetic antioxidants with natural sources that are easily available and safe. Therefore, the present research work was carried out to evaluate the antioxidant potential of the clove bud and the possibility of its inclusion as a natural food and feed additive.



## MATERIALS AND METHODS

### Source of Experimental Test Material

Clove bud (*Syzygium aromaticum*) was purchased from Kure ultra-modern market Minna, Niger state. The clove bud was sun-dried for three days and was thereafter ground to powder and stored in plastic containers.

### Plant Extract Preparation

The extract preparation of the dried clove bud was carried out by weighing one gramme (1g) of the grounded sample into a conical flask. Afterwards, 100ml of ethanol was measured and then added to the weighed sample in the flask. Extraction was performed for 40 minutes with the use of a digital 4 holes water bath (Model: E-Track England) at 70 degrees. The resultant extract was allowed to cool at room temperature and then filtered using a Whatman filter paper (No. 1).

### DPPH (2,2-Diphenyl-1-Picrylhydrazyl) Radical Scavenging Assay

In this experiment, the free radical used to evaluate the antioxidant activity of clove bud was 2, 2-diphenyl-1-picrylhydrazyl (DPPH), in line with the procedure described by Mukherjee *et al.* (2011) with slight changes. 2, 2-diphenyl-1-picrylhydrazyl of 100  $\mu$ M concentration was dissolved in methanol to a final concentration of 0.03mM. Serial dilutions were made to determine the IC<sub>50</sub>. In 96-well microplate total volume was 100  $\mu$ l which was consisting of 90  $\mu$ l of DPPH solution and 10  $\mu$ l of the test solution. The contents were mixed and incubated for 30 minutes at 37°C. An ultraviolet spectrophotometer was used to determine the absorbance at 517 nm. Ascorbic acid was used as the standard antioxidant. All readings were taken in triplicate and the mean values were then recorded. A decrease in absorbance indicated increased radical scavenging activity which was determined by the following formula:

$$\% \text{ Inhibition} = \frac{A_c - A_t}{A_c} \times 100$$

A<sub>c</sub> = Absorbance of control

A<sub>t</sub> = Absorbance of the test sample

## RESULTS AND DISCUSSION

The DPPH free radical scavenging activity of clove bud in the present study ranged from 19.65% to 67.23%. The inhibition activity of the clove bud against free radicals increased with increasing

levels of its extract concentration from 0.2mg/ml to 1.0mg/ml. Implying that the inhibition activity of the clove bud was directly proportional to the level of its extract concentration. The anti-free radical activity was expressed as IC<sub>50</sub> (mg/ml), which is defined as the extract concentration necessary to scavenge 50% DPPH free radicals (Selles *et al.*, 2020). The IC<sub>50</sub> value of 0.725mg/ml was obtained for the clove bud extract, whereas that of ascorbic acid was 0.320mg/ml. These values are the concentrations needed to achieve a radical scavenging outcome of 50 per cent. The inhibition of DPPH was 67.23% at the concentration of 1.0mg/ml of clove bud extract, this value falls within the range reported by Ahmed (2016), although at a lower concentration.

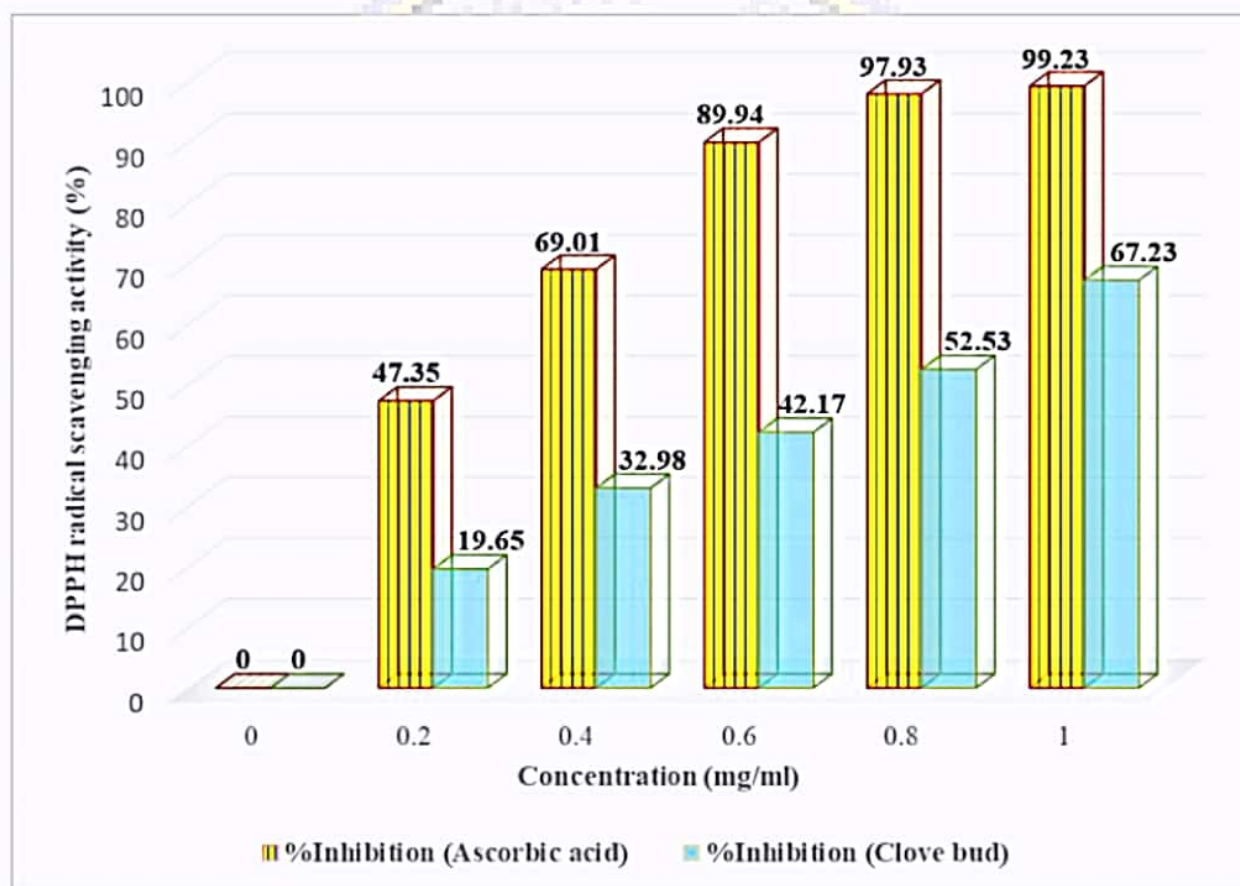


Figure 6: Clove bud antioxidant activity

## CONCLUSION

The results obtained from the present study suggest that the clove bud possesses antioxidant activities and may therefore be used as a potent free radical scavenger and well as a natural preservative in food and feeds. *Syzygium aromaticum* (clove bud) may find a useful application in the management of oxidative stress-related health conditions probably in the future.

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