

Microbial Assessment of Zobo Drink Sold in Some Locations in Yenagoa Metropolis, Nigeria

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Abstract

This study investigated the microbial quality of zobo drink sold in some major markets area of Yenagoa metropolis, Bayelsa state, Nigeria. Triplicate sample of zone was obtained from 5 locations in Yenagoa metropolis, Bayelsa state, Nigeria. The TCFUml⁻¹ of the samples was assessed using standard microbiological procedure. Results showed that total heterotrophic bacteria counts (4.16 - 5.71 Log cfu/ml), total coliform counts (1.66 - 2.63Log cfu/ml) and total fungi counts (2.64 - 4.58 Log cfu/ml) was significantly different ($P < 0.05$) among the different locations for each of the microbial parameter studied. The microbial counts were observed to be within limits specified by Food and Agricultural Organization, Food Quality Check Programme and International Commission on Microbiological Specifications for Foods. Furthermore, the presence of coliforms suggested likely fecal contamination. The tentatively identified microbial species include *Escherichia coli*, *Staphylococcus aureus*, *Bacillus*, *Enterobacter*, *Micrococcus* and *Proteus* species (bacteria), *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* and *Fusarium* (fungi). Among the isolates, *S. aureus* appeared in all the samples which is an indication of poor handling processes. Hence, there is the need for improve handling processes during processing and storage of zobo sold in the metropolis.

Keywords: Food Drink; Health Concern; Microorganisms; Yenagoa Metropolis; Zobo

Introduction

Different kinds of ready to eat foods abound in Nigeria. Typically, they exist as solid foods and drinks. Some notable food drink such as zobo has gained prominence in Nigeria irrespective of ethnicity and socioeconomic class [1-3]. Like most drinks that are processed traditionally, zobo drink could be a means of transmitting pathogenic microbes which could cause disease condition especially the ones prepared under unhygienic condition.

Typically, zobo is prepared from the calyces of *Hibiscus sabdariffa* [1,2,4-11]. According to Adanlawo and Ajibade [12], Ilondu and Iloh [13], two varieties of *H. sabdariffa* exist in Nigeria including red/brown and green variety, and they are distributed into different environment including Northern guinea and Sudan savanna (brown/red variety) and Southern Guinea savanna (green variety).

H. sabdariffa have varying economic importance. They are used in preparation of other drinks in other part of Africa including Senegal (bissap), Egypt (drink of the Pharaohs), Sudan (tea Karkade), Mali (da Bilenni) [1,14], Zobo (Nigeria). In Nigeria, zobo are called several names depending on the location and tribe. Furthermore, *H. sabdariffa* is used for the production fermented food called Furundu (Sudan), dawadawa botso (Niger), datou (mali) and Mbuja (Cameroun) [1,15-17].

Zobo drink is produced in both Northern and Southern region. The drink is mostly prepared in condition lacking quality control measures in Nigeria. In addition, preservation are some the major challenging confronting the shelf life of the drink. Several microbial species belonging to several genera (viz: *Bacillus*, *Streptococcus*, *Staphylococcus*, *Aeromonas*, *Corynebacterium*, *Veillonella*, *Micrococcus*, *Pseudomonas*, *Enterococcus*, *Leuconostoc*, *Lactobacillus*, *Enterococcus*, *Escherichia*, *Proteus* (bacteria), *Aspergillus*, *Penicillium*, *Saccharomyces*, *Lactobacillus*, *Geotrichum*, *Fusarium*, *Alternaria*, *Candida* and *Mucor* have been reported including in Zobo drinks sold in different locations in Nigeria [1,2,7,8,18,19]. Majority of microorganisms of these genus have been implicated to cause zobo spoilage and disease condition especially in immunocompromized individuals. For instance Bankole., *et al.* [20], listed *Bacillus*, *Streptococcus*, *Staphylococcus*, *Leuconostoc*, *Lactobacillus*, *Aspergillus*, *Penicillium*, *Geotrichum*, *Fusarium* and *Alternaria* species as potential microbes that could deteriorate zobo drink. Umaru., *et al.* [5] stated that consumption of local beverages could be a potential source of transfer of zoonotic and foodborne pathogens including Staphylococcosis, Salmonellosis, Brucellosis, Tuberculosis, Shigellosis, Listeriosis, *E. coli* infections etc.

Several studies have been carried out with regard to microbial density found in zobo drink consumed in Nigeria including Kano metropolis, Kano state [21], market in Aba, Abia State [11], Jos metropolis, Plateau state [22], Awka metropolis, Anambra state [23], some markets, Osun state [10], Ibadan metropolis, Oyo state [24], market, Abakaliki, Ebonyi state [9], markets, Umuahia, Abia state [25]. But information about the quality of zobo sold in Yenagoa metropolis appears scanty in literature. Hence this study aimed at assessing the microbial population found in zobo drink sold in Yenagoa metropolis, Bayelsa state, Nigeria.

Materials and Methods

Study Area

The study area was carried out in Yenagoa metropolis, Bayelsa state capital. The study has two predominant seasons wet season (April to October with intermittent break in August called August break) and dry season (November to March of the following year) [26]. The relative humidity is about 60 - 95% during the wet season and 50 - 80% during the dry season. The temperature in the area is $30 \pm 6^{\circ}\text{C}$ all year round. The study area is one of the growing cities in Nigeria with population significantly exceeding what is estimated during the last national census. Like other cities in Nigeria, several activities take place in Yenagoa metropolis, Bayelsa state. The region has several tributary Nun River (a major river in Bayelsa state), and Epie creek run through Igbogene to Government house, aligning the major road (Mbiama-Yenagoa road) [27-29].

Field Sampling

Zobo samples were purchased in five major market areas of Yenegoa metropolis including Akenfa, Etegwe Junction, Agudama-Epie, Igbogene and Opolo. The samples were purchased in triplicate. The samples were transported to the laboratory in ice box and preserved at 4°C prior to analysis.

Microbial Counts Enumeration

The enumeration of the microbial population in the zobo drinks were carried out using four different media; three for bacteria (including Nutrient Agar, which is used to enumerate obligate and facultative bacteria; MacConkey Agar were used for the enumeration of bacteria of the *Enterobacteriaceae* including coliforms; Salmonella-Shigella Agar were used to enumerate *Salmonella* and *Shigella*) and one for fungi (Potatoes Dextrose Agar was used to enumerate fungi). All the media were prepared according the manufacturers' guide. The samples were serial diluted and pour plate method previously described by Pepper and Gerba [30], Benson [31] was adopted in the study. Approximately 1 ml of the serial diluted samples was plated in the petri dish meant for the different media. The samples meant for bacteria (Nutrient Agar, MacConkey Agar and Salmonella-Shigella Agar) were incubated inverted at 37°C for 24 - 48 hours, while the Agar plate meant for fungal counts (i.e. containing Potatoes Dextrose Agar) was incubated inverted at 30°C for 3 - 5 days. After incubation, the resultant colonies were counted and expressed in colony forming units per millilitre (cfu/ml) of the zobo samples

Microbial identification

Bacteria Identification

The biochemical tests were carried out using the guide of Cheesbrough [32] and Benson [31]. All the bacteria isolates were cultured on Nutrient Agar prior to use for biochemical tests (viz. gram reaction, motility, indole, catalase, coagulase, urease, citrate). The resultant characteristics were compared with those of known taxa using Bergey’s Manual of Determinative Bacteriology by Holt, *et al.* [33] and the scheme of Cheesbrough [32]. Based on gram reaction, the gram positive cocci organisms were streaked onto Mannitol Salt Agar plate and incubated at 37°C for 24 hours. The presence of yellowish pigments in Mannitol Salt Agar indicates *Staphylococcus aureus*. The pure cultures from MacConkey agar were first streaked in nutrient agar before being streaked again in Levine’s Eosin Methylene Blue (EMB) Agar and incubated at 37°C for 24 hours. The presence of small nucleated colonies with greenish metallic sheen indicates *E. coli* [31]. Furthermore, isolates were streaked in blood agar, and after incubation the presence of swarming characteristics indicates *Proteus* species.

Identification of fungi isolates

The scheme of Pepper and Gerba [30] and Benson [31] was adopted for the microscopic and macroscopic approach of mold identification. The macroscopic/colonial characteristics were carried out based on the morphology, while the microscopic identification was carried out using Lactophenol cotton blue stain on a smeared isolates. The resultant characteristics based on microscopic examination was compared with the guide provided by Ellis, *et al.* [34] and Benson [31], while the macroscopic morphology were similarly compared with the guide provided by Benson [31].

Statistical analysis

Statistical Package for Social Sciences software version 20 was used for the statistical analysis. Descriptive statistics i.e. mean ± standard error values were expressed for log transformed microbial counts. One-way analysis of variance was carried out at P = 0.05, and Tukey HSD statistics was used for mean separation.

Results and Discussion

The microbial population of zobo drink sold in some major market areas of Yenagoa metropolis, Bayelsa state, Nigeria is presented in table 1. The microbial population of total heterotrophic bacteria counts in the various locations ranged from 4.16 - 5.71 Log cfu/ml, being not significantly different (P > 0.05) among the various markets apart from Etegwé and Igboegene that were also not significantly different (P > 0.05). The total coliform counts ranged from 1.66 - 2.63Log cfu/ml, being not significantly different (P > 0.05) apart from the samples collected from Opolo. The total fungi counts ranged from 2.64- 4.58Log cfu/ml, being not significantly different (P > 0.05) except for samples obtained from Opolo.

Location	Total Heterotrophic Bacteria counts, Log cfu/ml	Total coliform counts, Log cfu/ml	Total Fungi, counts, Log cfu/ml	Salmonella-Shigella counts, cfu/ml
Akenfa	4.40 ± 0.19a	2.62 ± 0.15b	3.53 ± 0.28abc	ND
Etegwé Junction	5.41 ± 0.17b	2.38 ± 0.20ab	3.91 ± 0.41bc	ND
Agudama-Epie	4.48 ± 0.17a	2.53 ± 0.23ab	3.08 ± 0.29ab	ND
Igbogene	5.75 ± 0.05b	2.56 ± 0.24b	4.58 ± 0.13c	ND
Opolo	4.16 ± 0.10a	1.66 ± 0.08a	2.64 ± 0.06a	ND

Table 1: Microbial density of zobo drink sold in some major market area of Yenagoa Metropolis, Bayelsa state, Nigeria.

Data are expressed as mean ± standard error (n = 3); Different letters (a, b, c) across the row indicate significance variation (P < 0.05) according to Tukey HSD statistics; ND: Not Detected

The various microbial parameters showed significant difference ($P < 0.05$). This could be associated to changes in handling processes prior to purchase. The quality of the packaging material (emptied used soft drink and water) could also contribute to variation in the drink. Furthermore handling and storage period could also account for the variation that exists among the different locations. This is because some microbes have a very short period of proliferation under suitable environmental condition [35]. Furthermore, instances of no variation between means of some zobo samples obtained from two or more locations using tukey test could be due similarity in productions and packaging processes, and hygienic status of vendors. As observed during the collection of samples a zobo vendor was seen hawking the fruit drink in two different markets area. This observation have been previously made by Izah., *et al.* [36] among vendor that sold prepared fruits (pineapple, paw-paw and water melon) in Yenagoa metropolis, Nigeria. However, the microbial population of the drink is within aerobic plate count for zobo (10^4) [10] apart from few instances i.e. Etegwe and Igbogene for total heterotrophic bacteria counts. The density were within the limit (10^5) recommended by Food and Agricultural Organization [21,37], Tolerable limits ($10^4 - 10^5$) for ready- to-eat food [1,38-41] and lower than the limit of 10^6 for Food Quality Check Programme limits [23,42].

Like water, coliform is not supposed to be found in zobo. As such, their occurrence suggests contamination. The findings of this study have some similarity with previous study. For instance, Bukar., *et al.* [21] reported total viable bacterial counts in the range of $< 30 - 1.23 \times 10^4$ cfu/ml in zobo sold in Kano metropolis, Kano state. Ezeigbo., *et al.* [11] reported total viable counts ($0.3 - 4.4 \times 10^6$ cfu/ml), total coliform ($0.1 - 6.5 \times 10^5$ cfu/ml) in zobo sold in Market in Aba, Abia State, Southeast Nigeria. Zumbes., *et al.* [22] reported total viable counts ($5.20 - 7.70$ cfu/ml), total coliform ($\times 10^4$ cfu/ml) in zobo sold in Jos metropolis, Plateau state. Anagu., *et al.* [23] reported total viable bacterial counts in the range of $3.0 \times 10^2 - 1.0 \times 10^5$ cfu/ml in zobo sold in Awka metropolis, Anambra state. Risiquat [10] reported total viable bacterial counts in the range of $1.2 \times 10^2 - 1.2 \times 10^6$ cfu/ml in zobo sold in Markets, Osun state. Slight variations that exist in the findings of this study when compared with previous studies could be due to handling period, quality of the materials used for production and hygienic status of the processors and vendors.

The microbes tentatively identified from the zobo drinks sold in Yenagoa metropolis, Bayelsa state, Nigeria is presented in table 2. The bacteria isolates include *Escherichia coli*, *Staphylococcus aureus*, *Bacillus*, *Micrococcus*, *Enterobacter* and *Proteus* species, while the fungi isolates are *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* and *Fusarium*.

Microbes	Akenfa	Etegwe Junction	Agudama- Epie	Igbogene	Opolo
<i>Escherichia coli</i>	+	-	+	+	+
<i>Bacillus</i> species	-	+	-	+	+
<i>Staphylococcus aureus</i>	+	+	+	+	+
<i>Enterobacter</i> species	-	+	-	-	-
<i>Micrococcus</i> species	-	+	-	+	-
<i>Proteus</i> species	+	-	-	-	-
<i>Aspergillus niger</i>	+	+	+	-	+
<i>Aspergillus flavus</i>	+	+	+	+	-
<i>Penicillium</i> species	+	-	-	+	-
<i>Fusarium</i> species	-	-	-	-	+

Table 2: Microbial characterization found in Zobo drink sold in Yenagoa metropolis, Nigeria.

+: Present; -: Absent (some of the microbes were only found in one of the three samples from each location).

The microbes identified from this study has some similarity with the findings of other authors on zobo drinks sold in different locations in Nigeria including Kano metropolis [21], Aba metropolis [11], Jos metropolis [22], Awka metropolis [23], Ibadan metropolis [24], Abakaliki [9] and Abia state [25].

Furthermore, the occurrence of coliforms (*Enterobacter* species and *E.coli*) may be from packaging materials, water used for processing and washing, handling etc [36]. *Bacillus* and *Micrococcus* may be from contaminant from the environment such as soil and processing equipment. Similarly, the presence *Staphylococcus aureus* may be associated with handling processes [36]. *Staphylococcus aureus* occurred in all the samples unlike other microbes isolated. This suggests that *Staphylococcus aureus* may have the highest occurrence rate in the samples in each of the location. The occurrence of the organisms could put potential consumers of the drink at risk of disease condition especially in immune compromised individuals. Some species of fungi could cause disease condition especially in immunocompromised patients as well. Some of this notable fungi species include *Penicillium*, *Fusarium* and *Aspergillus* species have the tendency to produce toxins that are harmful to human health [43].

Conclusion

In Nigeria, zobo is consumed by wide class of people. Its consumption has gain prominences and are sold in public places including school, hospital, work area, markets, streets etc. Zobo is prepared from the dried calyces of *Hibiscus sabdariffa* which belongs to malvaceae family. This study assessed the microbial population and isolates of Zobo sold in Yenagoa metropolis Bayelsa state, Nigeria. The study found that the microbial density is within various limits including Food and Agricultural Organization, Food Quality Check Programme limits and International Commission on Microbiological Specifications for Foods for ready to eat foods. Furthermore, the presence of coliforms (*Enterobacter* and *E.coli*) is an indication of contamination in the drink. This may have resulted from processing and handling processes and even processing ingredients i.e. spices, water, of *Hibiscus sabdariffa*, packaging containers etc. Hence, care should be put into consideration during processing, handling and storage since they are potential point through which microbes invade the drink.

Bibliography

1. Izah SC., et al. "Advances in preservatives and condiments used in zobo (a food-drink) production". *Biotechnological Research* 2.3 (2016): 104-119.
2. Izah SC., et al. "A review of the quality assessment of zobo drink consumed in Nigeria". *ASIO Journal of Microbiology, Food Science and Biotechnology Innovations* 1.1 (2015a): 34-44.
3. Nwafor OE. "Growth inhibitions of three fungal isolates from Zobo drink using sorbic and benzoic acids". *African Journal of Food Science and Technology* 3.3 (2012): 66-72.
4. Adebayo-Tayo BC and Samuel UA. "Microbial quality and proximate composition of dried Hibiscus sabdariffa clyxes in Uyo, Eastern Nigeria". *Malaysian Journal of Microbiology* 5.1 (2009): 13-18.
5. Umaru GA., et al. "Microflora of Kunun-Zaki and Sobo drinks in relation to public health in Jalingo Metropolis, North-Eastern Nigeria". *International Journal of Food Research* 1 (2014): 16-21.
6. Adesokan IA., et al. "Analysis of quality attributes of Hibiscus sabdariffa (Zobo) drinks blended with aqueous extract of ginger and garlic". *African Journal of Food Science* 7.7 (2013): 174-177.
7. Ezearigo OE., et al. "Screening of natural spices for improving microbiological, nutritional and organoleptic qualities of the Zobo drink". *Journal of Applied Biosciences* 76 (2014): 6397-6410.
8. Braide W., et al. "Perspectives in the hurdle techniques in the preservation of a non alcoholic beverage, Zobo". *African Journal of Food Science and Technology* 3.2 (2012): 46-52.

9. Alo MN., *et al.* "Bacteriological examination of locally produced beverage (Zobo) sold in Abakaliki, South-Eastern Nigeria". *International Scientific Research Journal* 4.2 (2012): 58-64.
10. Risiquat RO. "Bacteriology Quality of Zobo Drinks Consumed in Some Parts of Osun State, Nigeria". *Journal of Applied Science and Environmental Management* 17.1 (2013): 113-117.
11. Ezeigbo OR., *et al.* "Bacteriological Assessment of Hawked Sorrel Drink (Zobo Drink) in Aba, South-East Nigeria". *British Microbiology Research Journal* 5.2 (2015a): 146-151.
12. Adanlawo IG and Ajibade VA. "Nutritive Value of the Two Varieties of Roselle (*Hibiscus sabdariffa*) Calyces Soaked with Wood Ash". *Pakistan Journal of Nutrition* 5.6 (2006): 555-557.
13. Ilondu EM and Iloh AC. "Inhibition of Three Fungal Isolates from Sorrel Drink (Zobo) Using Hurdle Technique". *World Journal of Agricultural Sciences* 3.3 (2007): 339-343.
14. Padmaja H., *et al.* "Review on *Hibiscus sabdariffa*-A valuable herb". *International Journal of Pharmacy and Life Sciences* 5.8 (2014): 3747-3752.
15. Parkouda C., *et al.* "Technology and physico-chemical characteristics of Bikalga, alkaline fermented seeds of *Hibiscus sabdariffa*". *African Journal of Biotechnology* 7.7 (2008) : 916-922.
16. Daramola B and Asunni OA. "Nutrient composition and storage studies on Roselle extract enriched deep-fat-fried snack food". *African Journal of Biotechnology* 5.19 (2006): 1803-1807.
17. Tounkara F., *et al.* "Effect of boiling on the physicochemical properties of Roselle seeds (*Hibiscus sabdariffa* L.) cultivated in Mali". *African Journal of Biotechnology* 10.79 (2011): 18160-18166.
18. Egbere OJ., *et al.* "Effects of some preservation techniques on the quality and storage stability of Zobo drink (a Nigerian, non alcoholic beverage from *Hibiscus sabdariffa*)". *Journal of Food Technology* 5.3 (2007): 225-228.
19. Ezeigbo OR., *et al.* "Antimicrobial Effect of Lime Juice Treatment on the Shelf-Life of Zobo Drink". *British Microbiology Research Journal* 6.3 (2015b): 147-153.
20. Bankole YO., *et al.* "Preservation of Zobo Drink (Calyces of *Hibiscus Sabdariffa*) Using Kolanut". *Academic Journal of Interdisciplinary Studies* 2.10 (2013): 17-23.
21. Bukar A., *et al.* "Occurrence of some enteropathogenic bacteria in some minimally and fully processed ready-to-eat foods in Kano metropolis, Nigeria". *African Journal of Food Science* 4.2 (2010): 32-36.
22. Zumbes JH., *et al.* "Enteropathogenic Bacterial Contamination of Some Ready to Eat Foods Sold in Jos Metropolis, Nigeria". *Indian Journal of Applied Research* 3.7 (2014): 456-458.
23. Anagu L., *et al.* "Potential Spread of Pathogens by Consumption of Locally Produced Zobo and Soya Milk Drinks in Awka Metropolis, Nigeria". *British Microbiology Research Journal* 5.5 (2015): 424-431.
24. Amusa NA., *et al.* "Microbiological and nutritional quality of hawked sorrel drinks (soborodo) (the Nigerian locally brewed soft drinks) widely consumed and notable drinks in Nigeria". *Journal of Food, Agriculture and Environment* 3.3-4 (2005): 47-50.
25. Nwachukwu E., *et al.* "Effect of lime juice on the bacterial quality of zobo drinks locally produced in Nigeria". *Research Journal of Microbiology* 2 (2007): 787-791.
26. Izah SC., *et al.* "Climate change: some meteorological indicators and perception of farmers in Yenagoa metropolis, Bayelsa state, Nigeria". *International Journal of Geology, Agriculture and Environmental Sciences* 3.1 (2015b): 56-60.

27. Ben-Eledo VN., *et al.* "Bacteriological Quality Assessment of Epie Creek, Niger Delta Region of Nigeria". *International Journal of Ecotoxicology and Ecobiology* 2.3 (2017a): 102-108.
28. Ben-Eledo VN., *et al.* "Water quality assessment of Epie creek in Yenagoa metropolis, Bayelsa state, Nigeria". *Archives of Current Research International* 8.2 (2017b): 1-24.
29. Seiyaboh EI., *et al.* "Spatial Variation in Physico-chemical Characteristics of Sediment from Epie Creek, Bayelsa State, Nigeria". *Greener Journal of Environment Management and Public Safety* 5.5 (2016): 100-105.
30. Pepper IL and Gerba CP. "Environmental microbiology. A laboratory manual. Second edition". Elsevier academic press (2005).
31. Benson HJ. "Microbiological Applications: Laboratory Manual in General Microbiology". Complete version, 5th edition. McGaraw-Hill, New York (2002).
32. Cheesbrough M. "District Laboratory Practice in Tropical Countries. Low price Edition part 2". Cambridge press, England (2006).
33. Holt JG., *et al.* "Bergey's Manual of Determinative Bacteriology". William and Wilkins Publisher, Maryland. New York (1994).
34. Ellis D., *et al.* "Descriptions of Medical Fungi. Second Edition". Printed in Adelaide by Nexus Print Solutions, Underdale, South Australia (2007).
35. Izah SC and Ohimain EI. "Microbiological quality of palm oil used in Nigeria: Health impacts perspective". *Point Journal of Botany and Microbiology Research* 2.1 (2016): 037-045.
36. Izah SC., *et al.* "Microbial quality of polythene packaged sliced fruits sold in major markets of Yenagoa Metropolis, Nigeria". *Point Journal of Botany and Microbiology Research* 1.3 (2015c): 30-36.
37. Food and Agriculture Organization (FAO). "Manuals of food quality control. FAO food and nutrition paper". United Nation, Rome (1979).
38. International Commission on Microbiological Specifications for Foods (ICMSF). "Microorganisms in Foods 5: Microbiological Specifications of Pathogens" (1996).
39. Olopade BK., *et al.* "Microbiological quality of fermented Cassava (Gari) sold in Ota Ogun State Nigeria". *International Journal of Applied Microbiology and Applied Sciences* 3.3 (2014): 888-895.
40. Kigigha LT., *et al.* "Microbiological quality of fermented Cassava Flakes (Gari) sold in Yenagoa, Metropolis, Nigeria". *Bulletin of Advanced Scientific Research* 1.7 (2015a): 157-160.
41. Kigigha LT., *et al.* "Microbiological quality assessment of suya sold in Yenagoa Metropolis, Nigeria". *Journal of Advances in Biological and Basic Research* 1.5 (2015b): 106-109.
42. "Food Quality Check Programme. Microbiological Recommendations, Food Laboratory, Environmental Microbiology BC Public Health Microbiology and Reference laboratory" (2011).
43. Joseph AD and Adogbo GM. "Processing and Packaging of Hibiscus Sabdariffa for Preservation of Nutritional Constituents". *International Journal of Scientific and Engineering Research* 6.4 (2015): 532-536.

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