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Microbes and Dietary Components of Selected Commonly Consumed Foods

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ABSTRACT

Various types of bacterial isolates were obtained from commonly consumed food sources, viz rice and beans in some Akoko communities, Nigeria and the world at large. The bacterial isolates were identified by standard microbiological methods. Proximate analysis was carried out according to the procedure of association of official Analytical Chemists to determine amount of components such as fat content, protein content, moisture content and ash content in food samples. Bacterial isolates encountered during the study includes Clostridium spp. (4), Vibrio cholera (1), Bacillus spp. (3), Escherichia coli (2), Staphylococcus aureus (2), Shigella spp. (2), Pseudomonas spp. (2), Streptococcus spp. (2), and Micrococcus spp. (6). Proximate analysis determined, shows the values of food components (in percentages, %) for rice and beans as, Moisture content (11.76, 13.96), Ash content (0.68, 2.52), Crude Fat (3.00, 6.65), Crude protein (15.67, 21.58) Crude fibre (1.44, 0.32) and Carbohydrate content (67.45, 54.97). This study helps to investigate field samplings of the staple food studied in that microbes isolated from the commonly consumed food (rice and beans) in this part of Nigeria and world at large, were used as a measure to determine level of contamination and possible microflora that may inhabit these food sources for possible industrial applications. Similarly, the proximate analysis done helps to determine dietary value as well as food safety. Hence, these food sources should be properly handled coupled with appropriate storage based on their nutritional value and liability of contamination at difference stages of their processing from the raw source.

Keywords
Bacteria, Dietary, Epidemiology, Nigeria, Rice, Beans.

Introduction and Literature Review

Microbiological analysis of food helps to determine the microbial load and nature of microorganisms that inhabit or contaminate food, including those that cause food spoilage. "Good" bacteria, however, such as probiotics, are becoming increasingly important in food science. In addition, microorganisms are essential for the production of foods such as cheese, yogurt, bread, beer, wine and, other fermented foods [1]. Food analysis is helpful to determine properties of foods and their constituents. This information on different characteristics of foods, including their composition, structure, physiochemical characteristics and sensory attributes enhances food safety.

Food safety is a major focus of food microbiology. Pathogenic bacteria, viruses and toxins produced by microorganism are all possible contaminants of food. However, microorganisms and their products can also be used to combat these pathogenic microbes. Probiotic bacteria, including those that produce bacteriocins, can kill and inhibit pathogens. Alternatively, purified bacteriocins such as nisin can be added directly to food products. Finally, bacteriophages, viruses that only infect bacteria, can be used to kill bacterial pathogens. Thorough preparation of food, including proper cooking, eliminates most bacteria and viruses. However, *toxins* produced by contaminants may not be liable to change to non-toxic forms by heating or cooking the contaminated food [2].

Recently, new analytical techniques have been introduced and the information about food composition is rapidly expanding. However, the system of proximate analysis still forms the basis for the statutory declaration of the composition of foods in Europe. This system of analysis divides the food into six fractions:

moisture, ash, crude protein, ether extract, crude fibre and nitrogen-free extractives. Most of these analytical procedures in food are described by Károly [3].

A microbiological culture, or microbial culture, is a method of multiplying microbial organisms by letting them reproduce in predetermined culture media under controlled laboratory conditions. Microbial cultures are used to determine the type of organism, its abundance in the sample being tested, or both. It is one of the primary diagnostic methods of microbiology and used as a tool to determine the cause of infectious disease by letting the agent multiply in a predetermined medium [4].

Rice and beans is a versatile grain that is a major part of several ethnic cuisines. It provides complex carbohydrates with only 100c calorie in one-half cup serving, according to the U.S. Department of Agriculture (Arie). Food crop systems are community of plants which are managed to obtain food, profit, satisfaction or, most commonly a combination of this goal. Beans and rice are a staple food in many cultures around the world. It provides several important nutrients, and is widely available. In many areas, rice and beans are often served side by side rather than mixed. Either way, they may be considered a meal, frequently with a topping of meat or chicken. Meat or other ingredients are sometimes placed atop rice and beans or (less often) mixed into it [5].

While beans are native to the Americas, rice is not. Rice was introduced to the Caribbean and South America by European colonizers at an early date with Spanish colonizers introducing Asian rice to Mexico in the 1520s at Veracruz and the Portuguese and their African slaves introducing it at about the same time to Colonial Brazil. More recent scholarship suggests that African slaves played a more active role in the establishment of rice in the New World, and that African rice was an important crop from an early period [5]. In either case, varieties of rice and bean dishes were a staple dish among the peoples of West Africa, and they remained a staple among their descendants subjected to slavery in the Spanish New World colonies and elsewhere in the Americas [6].

Rice (*Oryza sativa* and *O. glaberrima*) belongs to the subfamily *Bambusoidae*, tribe *Oryzae* [7]. There are two cultivated species: *O. sativa*, Asian origin, *O. glaberrima* of African origin. The former is far more important than the latter. *O. sativa* is differentiated into three races or sub species: Indica, Japonica and Javanica. There appears to be general agreement that *O. sativa* and *O. glaberrima* represent the end point of independent and parallel domestication. It seems likely that both species developed from annual progenitors: in the case of *O.sativa* a species resembling O. nivara, and in the case of *O. glaberrima* one resembling *O. barthii* [8]. Most authorities believe rice was domesticated independently at several locations in south and Southeast Asia. Archaeological evidence was shown in this regard from India dates back to about 500BP, and from Thailand, though this is less certain [9].

Ho concluded 'that while existing a comparative data are in favour of China, the southeastern Asian mainland, and the Indian sub-continent'. From this region, cultivated rice spread to the Yellow River Valley where the Japonica race evolved, and from Korean and Japan. The Indica race spread to Yangtse Valley about 2000BP; the Javanica races developed in Indonesia at a later date [10]. Rice shows a wide range of variation in development pattern through its adaptation to the climatic factors of temperature, depth of flooding and daylenght. Temperature adaptation is illustrated by the importance of crop in the wet, wet-and-dry and cool tropics. Adaptation to depth of flooding and degree of water control has given rice to three cultural classes of rice: Upland rice; wet or Padi rice, grown in water less than 1m deep; and deep water rice, grown in water 1-6m deep [11].

Taxonomy of common beans (*Phaseolus vulgaris*) has been reviewed by Gepts and Debouck [12]. The genus, of New world origin, is in the tribe Phaseoleae and is tentatively regarded as including 55 species, of which five are cultivated. *P. vulgaris*., common beans, *P. coccineus* L., scarlet runner bean, *P. lunatus*., Lima bean, *P. acutifolius* A. Gray, Tepary beean, *P. polyanthus* Greenman, Year-bean. All are diploid (2n=2*=22). There are some genetic affinities between the species: crossing experiments suggests that P. vulgaris, common beans, *P. coccineus*, and *P. acutifolius* are closely related. Classification of types with P. vulgaris is based on determinancy of the main axis, growth habit, crop duration and seed characteristics [13]. Plants may be erect 'brush beans', semi -climbing or climbing; crop duration may vary from 75 days for an early brush type to 270 days for a late climbing type in a cool climate. Evans [14] and Gepts and Debouck [12] have reviewed dispersal: in general, both Central America and Andrean types spread to the same regions of the world, but the former became predominant in lowland South America and the latter in Africa.

There are many possible causes of food poisoning: bacteria, viruses, pesticides, natural toxins, molds, parasites, and more. There are so many types of food poisoning that there are many possible symptoms. Food poisoning is especially dangerous for infants, young children, elderly people, and those with chronic health conditions or weak immune systems [15].

Illness occurs as a result of the consumption of food and water contaminated with enteropathogenic bacteria or viruses which remain alive in the food or water during consumption. These pathogens have the potential to establish and multiply in the digestive tract to cause the illness (e.g, Salmonellosis, Hepatitis A virus infection [16]. The bacteria cells sporulate, colonize, or die and release toxin (s) to produce the symptoms e.g, gastroenteritis [17]. This study helps to investigate field samplings on rice and beans from different markets. Similarly, the natures of microbes that are present in commonly consume food (rice and beans) and the proximate analysis of this food sources will be determined. The result of this study will serve as data base for improving food quality of this commonly consumed foods and as well as a food safety measure to prevent the spread of the disease epidemics.

Materials and Methods

Sample collection

Sample (Rice and beans) were purchased from three different market which are Akungba, Oka, and Ikare market from the rice and beans seller. The grains of rice are white in colour and while the beans seed is also white in colour. The rice grain (*Oryza sativa*) is brownwish in colour if the grain has not been milled to remove the shaft. Rice have variable colours, some are whitish, yellowish or brown. Rice has variable shapes which is either long in structure or short. Beans on the other hand taken for analysis is the common white beans called (*Phaseolus vulgaris*). The beans seed are variable in size depending on where it is grown. Some are very big and white in colour, while some are small beans that were grown in Katstina state and in Kano state. All these samples was collected and taken for analysis.

In all the methods of sample collection, three different samples of both rice and beans was collected from three different markets and thus making an overall of six samples in all. During collections, the samples to be taken were collected into a sterile universal container which was labeled to indicate series of sample taken from each food lot and the name of the market. The collected sample was then brought to the microbiology laboratory of Adekunle Ajasin University Akungba-Akoko for microbial analysis. Materials including glass wares and culture media used for this study were aseptically sterilized.

Sampling

Both samples of rice and beans were taken each from labeled container indicating the food lot and market source from which it was purchased and was milled into powdery form differently inside an electric blender for analysis. 9mls of distilled water was equally pipette into 14 test tubes, starting from 10^1-10^7 for the rice sample analysis and equally 10^1-10^7 for the beans sample which was then sterilized prior to serial dilution. 1g of the grounded cotyledon from each food sample was dispensed into the first dilution factor of 10^1 which was shaken properly to form homogenous solution. 1ml of the sample in the test tube was transferred into the second test tube with a sterile syringe till it got to the seventh dilution for both the grounded rice and beans sample.

Isolation Procedure

For bacteria cultivation, 1ml each was aseptically pipette from dilution (10^{-5} and 10^{-7}) by flaming the mouth of the test tube before and after pipetting into sterile Petri-dish each near the Bunsen burner's flame. About 20ml of prepared sterile molten nutrients agar (NA) was aseptically poured on the inoculums in each petri-dish (pour plate method). The dishes were swirled gently to mix the inoculums with the agar and allowed to solidify.

The plates were incubated at 37⁰C for 24 hours. After incubation, the plates were examined microscopically for bacteria growth and the bacteria colonies observed were counted using the colony counter. Selection of various colonies was done and they were sub-cultured to obtain pure cultures. The pure cultures were finally preserved on agar slants in a refrigerator of about 50⁰C for further tests.

Identification of bacteria isolates

The organism were observed and grouped primarily by colonial characteristics on the plates. The selected isolates were sub-cultured, subjected to some biochemical test and identified by standard microbiological methods.

Proximate analysis of dried rice and beans

Proximate analysis was carried out according to the procedure of association of official Analytical Chemists. Proximate analysis reveals the percentage of the classes of food present in a food sample such as fat content, protein content, moisture content, ash content.

Determination of moisture content

Five grams (5g) of grounded rice and beans was weighed differently into a clean and well labeled evaporating dish which has been earlier weighed as W_1. The evaporating dish and the samples re-weighed as (W_2). The sample was then stored and placed in a well-ventilated oven at 100-105⁰C with the lid of desiccator removed. After 24 hours of drying, the sample was then collected from the oven and weighed on a weighing balance after the samples have been allowed to cool. The loss in weight between the initial and the final sample was recorded as the moisture content.

Moisture content is given by the formula:

$$\%\ \text{Moisture content} = \frac{W_1 - W_2}{W_1 - W_0} \times 100$$

Determination of Ash Content

The crucible was first washed, rinsed with distilled water, dried in the oven flow to cool in a desiccator. The crucible was weighed and recorded (W_1) using a mettle balance. 1g of the sample was weighed into the crucible measured and recorded as (W_2), which was the placed inside the muffle furnace. It slowly increases its temperature from 200⁰C-450⁰C for about 3 hours to avoid incomplete ashing. The sample was left inside the furnace until it changes to whitish colour. The crucible was the removed and placed in the desiccator. The crucible was brought and reweighed.

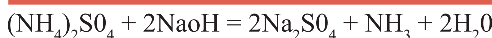
$$\%\ \text{Ash} = \frac{W_3 - W_1}{W_2 - W_1} \times 100$$

Determination of Crude Protein

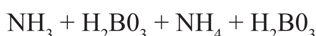
The process involves four stages-Digestion, Distillation, Absorption and titration.

Digestion - 1.0g of dried samples was put inside Kjeldahl flask and 1g of catalase reagent was weighed and added. Sulphuric acid was then added and the substrate was heated for 1hour. Colour change from black to green indicates proper digestion of substrate.

Distillation - 3ml of 2% boric acid was poured into 100ml conical flasks and 3drops of indicator was added with 0.19g bromocresol green plus methyl red was added to the digestion solution. 5mls of sample rich in nitrogen was then pipette and add 10ml 40% NaOH tight the joints and distilled about 50ml into the receiving flask. This procedure involves the use of Markham apparatus.



Absorption - Boric acid was used to trap the liberated ammonia to form ammonium borate.



Titration stage - The received ammonia trapped in form of ammonium borate was then titrated against hydrochloric acid of known concentration to reduce the nitrogen concentration.

Colour changes from pink to blue
% Crude protein = % Nitrogen xx 6.25.

Determination of Crude Fat

This process depends on sparingly solubility of lipid in water. Fat is extracted using solvent such as petroleum ether or N- Hexane. A thimble was then weighed (W_1), the dried sample was then poured into the thimble and weighed again (W_2) and 500ml round bottom flask was then weighed (W_5). Petroleum ether was the poured into the round bottom flask and the soxhlet extractor with a reflux condenser was the fixed and adjusted to with heat source. When the petroleum ether has siphoned over the barrel, the crucible was removed and petroleum ether was distilled from the flask. The fat was then dried in an oven and the constant weighed was then cooled and weighed (W_5).

The % crude Fat was measured by

$$\% \text{ Fat} = \frac{W_2 - W_5}{W_2 - w_1} \times 100$$

Determination of Crude Fibre

2g of defatted samples were weighed into a 500ml conical flask and recorded as (W_1). 200ml of boiling 1.2% H_2SO_4 was added. The solution was boilt gently for 30minutes under specified conditions, filtered through a muslin cloth stretched over $9cm^3$. Buchner funnel and scraped back into the flask with scapula 200ml of boiling 1.25%. NaOH was added and solution was built gently for 30 minutes. These were again washed thoroughly with hot distilled water and were rinsed once with 10% HCL and twice industrial 100% Ethanol. The residues were rinsed finally three times with petroleum ether. Residues were transferred into pre-weighed crucible labeled W_2 and dried in an oven at 105^oC, cooled in a desiccator and weighed W_1. Tis was later transferred into a muffle furnace at about 300^oC for about 30 minutes, removed from the furnace and the cooled in a desiccator, incinerated and weighed as W_4.

$$\% \text{ Crude fiber} = \frac{W_3 - W_4}{W_2 - w_1} \times 100\%$$

Determination of Carbohydrate Content

The most common approach of carbohydrate content of food is used in the difference between the total predominant content in percentage and one hundred.
% Carbohydrate – 100 (% ash + % crude protein + % fat + % crude fibre + % moisture).

Result

Twenty four bacteria were isolated from the rice and beans sample taken for analyses. Nine genera of the bacteria isolated and identified include four Clostridium spp., one genera of Vibrio cholera, three genera of Bacillus spp., two genera of Escherichia coli, two genera of Staphylococcus aureus, two genera of Shigella spp., two genera of Pseudomonas spp., two genera of Streptococcus spp., and six genera of Micrococcus spp.. The identification and characterization of bacteria isolate were based on their cultural, morphological and biochemical characteristics (Table 1 and 2). The frequency of occurrence of these organisms was also determined for possible industrial applications and epidemiological reasons (Table 3). Furthermore, proximate analysis value of the grounded samples of rice and beans taken for analysis (Table 4).

Table 1: Morphological characteristics of bacterial isolate from rice and beans samples.

Isolates	Colour	Shape	Edge	Optical	Surface	Probable organism
AkgR1	White	Spherical	Irregular	Opaque	Smooth	*Clostridium* spp.
AkgR2	Yellow	Spherical	Entire	Translucent	Smooth	*Micrococcus* spp.
AkgR3	Cream	Spherical	Entire	Opaque	Smooth	*Pseudomonas* spp.
AkgR4	White	Flat	Entire	Opaque	Rough	*Escherichia coli*
AkgB1	White	Spherical	Irregular	Opaque	Smooth	*Clostridium* spp.
AkgB2	Yellow	Spherical	Entire	Translucent	Smooth	*Micrococcus* spp.
AkgB3	Cream	Comma	Irregular	Transparent	Smooth	*Vibrio* spp.
AkgB4	Creamy	Rhizoid	Irregular	Opaque	Smooth	*Bacillus* spp.
IkR1	Creamy	Round	Irregular	Opaque	Smooth	*Streptococcus* spp.
IkR2	Cream	Spherical	Irregular	Opaque	Smooth	*Streptococcus* spp.
IkR3	White	Spherical	Irregular	Opaque	Smooth	*Clostridium* spp.
IkR4	Yellow	Circular	Entire	Translucent	Smooth	*Micrococcus* spp.
IkB1	Greyish	Flat	Entire	Opaque	Rough	*Escherichia coli*
IkB2	Cream	Rhizoid	Regular	Opaque	Smooth	*Bacillus* spp.
IkB3	Yellow	Spherical	Entire	Translucent	Smooth	*Micrococcus* spp.
IkB4	Creamy	Round	Irregular	Opaque	Smooth	*Bacillus* spp.
OkaR1	Cream	Spherical	Irregular	Translucent	Smooth	*Staphylococcus* spp.
OkaR2	White	Round	Regular	Opaque	Smooth	*Clostridium* spp.
OkaR3	Creamy	Spherical	Entire	Opaque	Smooth	*Pseudomonas* spp.
OkaR4	Red	Spherical	Entire	Translucent	Smooth	*Micrococcus* spp.
OkaB1	White	Circular	Entire	Translucent	Smooth	*Shigella* spp.
OkaB2	Greyish	Circular	Entire	Translucent	Smooth	*Shigella* spp.
OkaB3	Yellow	Round	Entire	Translucent	Smooth	*Micrococcus* spp.
OkaB4	White	Round	Irregular	Translucent	Smooth	*Staphylococcus* spp.

Key: Akg- Akungba, R_1-R_4-Isolated organism from rice, B_1-B_4- Isolated organism from beans, Ik- Ikare, Oka- Oka Akoko.

Table 2: Biochemical characteristics of the isolates.

Isolate	Cell charactertistics	Gram Stain	Catalase	Glucose	Sucrose	Fructose	Lactose	Mannitol	Oxidase	Coagulase	Probable organisms
AkgR1	Rod	+	_	AG	AG	_	AG	AG	_	_	*Clostridium* spp.
AkgR2	Rod	+	+	_	_	_	_	_	+	_	*Micrococcus* spp.
AkgR3	Rod	_	+	A	_	A	_	A	+	_	*Pseudomonas* spp.
AkgR4	Rod	_	+	AG	_	A	AG	A	_	+	*Escherichia coli*
AkgB1	Rod	+	_	AG	AG	_	AG	AG	_	_	*Clostridium* spp.
AkgB2	Rod	+	+	_	_	_	_	_	+	_	*Micrococcus* spp.
AkgB3	Rod	_	+	AG	A	A	AG	A	+	_	*Vibrio* spp.
AkgB4	Rod	+	+	AG	AG	A	_	A	_	_	*Bacillus* spp.
IkR1	Cocci	+	_	AG	A	AG	AG	A	_	_	*Streptococcus* spp.
IkR2	Cocci	+	_	AG	A	AG	AG	A	_	_	*Streptococcus* spp.
IkR3	Rod	+	_	AG	AG	_	AG	AG	_	_	*Clostridium* spp.
IkR4	Rod	+	+	_	_	_	_	_	+	_	*Micrococcus* spp.
IkB1	Rod	_	+	AG	_	A	AG	A	_	+	*Escherichia coli*
IkB2	Rod	+	+	AG	AG	A	_	A	_	_	*Bacillus* spp.
IkB3	Rod	+	+	_	_	_	_	_	+	_	*Micrococcus* spp.
IkB4	Rod	+	+	AG	AG	A	_	A	_	_	*Bacillus* spp.
OkaR1	Cocci	+	+	AG	AG	A	AG	AG	_	+	*Staphylococcus* spp.
OkaR2	Rod	+	_	AG	AG	_	AG	AG	_	_	*Clostridium* spp.
OkaR3	Rod	_	+	A	_	A	NG	A	+	_	*Pseudomonas* spp.
OkaR4	Rod	+	+	_	_	_	_	_	+	_	*Micrococcus* spp.
OkaB1	Short rod	_	_	A	_	A	_	AG	_	_	*Shigella* spp.
OkaB2	Rod	+	+	_	_	_	_	_	+	_	*Micrococcus* spp.
OkaB3	Short rod	_	_	A	_	A	_	AG	_	_	*Shigella* spp.
OkaB4	Cocci	+	+	AG	AG	A	AG	AG	_	+	*Staphylococcus* spp.

Key: +: Positive reaction, _: Negative reaction, A: Acid production AG: Acid and gas.
Akg: Akungba, R1-R4: Isolated organism from rice, B1-B4: Isolated organism from beans, Ik: Ikare, Oka: Oka Akoko.

Table 3: Frequency of bacterial isolates encountered.

Isolates	Numbers	Percentage (%)
Clostridium spp.	4	16.66
Vibrio cholera	1	4.15
Bacillus species	3	12.50
Escherichia coli	2	8.33
Staphylococcus aureus	2	8.33
Shigella species	2	8.33
Pseudomonas spp.	2	8.33
Streptococcus spp.	2	8.33
Micrococcus spp.	6	25
Total	24	100

Table 4: Proximate analysis of the rice and beans samples.

Proximate Analysis (%)	Rice	Beans
Moisture content	11.76	13.96
Ash content	0.68	2.52
Crude Fat	3.00	6.65
Crude protein	15.67	21.58
Crude fibre	1.44	0.32
Carbohydrate content	67.45	54.97

Discussion and Conclusion

Various types of microbial species were isolated from grounded rice and beans (Tables 1 and 2). They belong to genera of bacteria which consists of *Clostridium* spp (4)., *Vibrio cholera* (1), *Bacillus* spp. (3), *Escherichia coli* (2), *Staphylococcus aureus* (2), *Shigella* spp. (2), *Pseudomonas* spp. (2), *Streptococcus* spp. (2), and *Micrococcus* spp. (6) (Table, 3).

There are more than 250 types of food poisoning. It's no wonder that one in six Americans gets food poisoning every year. The causes of foodborne illness range from *amebiasis* and anthrax to *vibrio* and *yersinia*. They include bacteria, viruses, pesticides, natural toxins, molds, parasites, and more. But most people don't want an encyclopedia of food poisoning (though such things exist). They just don't want to get sick [18].

Botulism is a rare but dangerous type of foodborne illness caused by *Clostridium botulinum*. Foodborne botulism usually is caused by improperly canned food which affects the nervous system [18]. *Salmonella, Cholera, Staphylococcus, Shigella* spp., *Pseudomonas* spp., *Escherichia coli* and *Bacllus* spp. causes about 1 million illnesses every year which is the leading cause of hospitalization and of death due to foodborne illness. Symptoms include fever and abdominal pain, vomiting, and diarrhea [19].

Food can become contaminated at many points from its origin, for example, a chicken with infected reproductive organs can lay eggs containing salmonella. Pesticides or chemicals could somehow find their way into or onto food. These substances may interfere with the general component and diminish values of food sources (Table 4). Food could be washed or handled in a contaminated environment. Food could be processed in a contaminated facility. Food handlers can contaminate food. This can happen if they are ill, if they don't wash their hands often enough or after using the bathroom, or if they have a cut or sore on their hands [20].

Proper handling of food, washing of hands and facilities such as knives, forks, and spoons can prevent food borne illness. If affected with enteric bacterial infection, treatment is a safe and good control measures. At anyrate, the consciousness of protective handling of our food substances at various levels of production sources, storage and processing for consumption is valuable to sustain wellness of humans in terms of food security.

References

1. Fratamico PM, Bayles DO. Foodborne Pathogens:

Microbiology and Molecular Biology. Caister Academic Press. 2005; ISBN 978-1-904455-00-4.
2. Tannock GW. Probiotics and Prebiotics: Scientific Aspects. Caister Academic Press. 2005. ISBN 978-1-904455-01-1.
3. Károly D. Chapter 3. Analysis of food. Proximate analysis of food. London: Longman. 2011.
4. Gregor R, Donald P. Healthwise, Incorporated (2010-06-28). "Throat Culture". WebMD. 2012.
5. Judith AC. Black Rice: The African origins of Rice Cultivation in Americas. 2010.
6. National Research Council. "African Rice". Lost Crops of Africa: Volume I: Grains. Lost Crops of Africa 1. National Academies Press. 1996-2014; ISBN 978-0-309-04990-0.
7. Clayton WD, Renvoize SA. General Graminum. Grasses of the world. London. 2000.
8. Chang TT. Rice. Oryza sativa and O. glaberrima (Gramineae Oryzae). In evolution of crop plants ed. N.W. Simmonds. London and Newyork: Longman. 1997; 98-104.
9. Gorman C. A prior model and Thai Prehistory: a reconsideration of the beginnings of agriculture in southeastern Asia. In origins of agriculture, ed. C.A. Reed. 2002; 321-355.
10. IRRI. The IRRI Reporter, 1997. Los Banos Philippines: International rice research institute. 1997.
11. Huke R. Geography and climate of rice. In climate and rice, pp.31-50. Los Banos, Philippines: International Rice Research Institute. 2005.
12. Gepts P, Debouck D. Origin, domestication, and evolution of the common bean (P. vulgaris L.). In common beans: Research for Crop Improvement, ed. A. Van Schoonhoven and O. Voysest. 1991; 7-53.
13. Voysest O, Dessert, M. (1991). Bean cultivars: Classes and commercial seed types. In common Beans: Research for crop improvement, edn. Wallingford, U.K: (AB International/ Centro Internacional de Agricultura Tropical. 119-162.
14. Evans AM. Beans. Phaseolus species. (Legumionosae-papilionatae).in evolution of crop plants, edn. N.W Simmonds. London Longman. 1986; 168-172.
15. Lampel KA, Al-Khaldi S, Cahill SM. Foodborne Pathogenic Microorganisms and Natural Toxins Handbook, second edition. Rockville, MD: Food and Drug Administration. 2012.
16. Nyachuba DG. Foodborne illness: Is on the rise?, Nutr. 2010; 68: 257-259.
17. Newell DR. Food-borne diseases: The challenges of 20 years ago still persist while new ones continue to emerge, International Journal Food Microbiology. 2010; 139: S3-S15.
18. Painter JA, Hoekstra RM, Ayers T, et al. Attribution of foodborne illnesses, hospitalizations, and deaths to food commodities by using outbreak data, United States. 2013; 1998-2008.
19. U.S. Food and Drug Administration. Center for Food Safety and Applied Nutrition. Rockville (MD): U.S. Food and Drug Administration. Foodborne illnesses: what you need to know. 2013.
20. U.S. Centers for Disease Control and Prevention. National Center for Emerging and Zoonotic Infectious Diseases, Division of Foodborne, Waterborne, and Environmental Diseases [Internet]. Atlanta (G.A): Centers for Disease Control and Prevention. Botulism General Information Frequently Asked Questions. 2010.

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