

MICROBIOLOGICAL ANALYSIS OF KUNUN-ZAKI: A FERMENTED MILLET DRINK IN BENIN CITY, EDO STATE, NIGERIA

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ABSTRACT

Microbiological and physicochemical analyses were carried out on samples of fermented millet drink Kunun Zaki. Fifteen samples were obtained from Ikpoba Hill Market and Aduwawa Quarters. The microbiological analysis was carried out using the standard plate count technique to determine the total microbial population. The mean count of bacteria and fungi was 2.57×10^7 cfu/ml and 0.98×10^7 cfu/ml respectively. Microorganisms identified were *Lactobacillus* sp, *Bacillus* sp, *Staphylococcus aureus*, *Streptococcus* sp, *Escherichia coli*, *Pseudomonas* sp, *Mucor* sp and *Fusarium* sp. The mean value of the pH and titrable acidity was 4.26 ± 0.09 and 2.73 ± 0.08 ml 0.1M NaOH respectively. The moisture content was high with a mean of 85.90 ± 0.95 and mean solid content of 14.1 ± 0.95 . This study has shown that kunun-zaki sold in Ikpoba Hill Market and Aduwawa Quarters is highly contaminated with microorganisms. Practices of good hygiene are therefore necessary in an environment where kunun-zaki is produced, stored, prepared and packaged.

Keywords: Benin City, fermented millet, kunun-zaki, microbiological, physicochemical

INTRODUCTION

Food borne intoxication and poisoning is common in man and local beverage and drinks may be common sources of infection (WHO, 1996). The consumption of these local drinks is of public health significance. Hence, local drinks may serve as vehicles for food-borne pathogens such as *Staphylococcus*, *Salmonella*, *Shigella*, *Listeria*, *Escherichia* etc. Millet is one of the main cereal staples of West Africa. There are various species of millets found and they include Bulrush millet (Nigeria millet), *Pennisetum typhoides* in America, popularly known as Pearl millet, *Pennisetum americanum*, foxtail millet (*Setaria italica*) and finger millet (*Eleusine coracana*). The grain had its origin in Central America and West

Asia (Efiuvwevwe and Akoma, 1995). They are widely grown in Ghana, Cameroon and throughout the Savannah zone of Nigeria such as Bauchi, Sokoto, Katsina and Kano States (Ònuorah *et al.*, 1987). The quality of millet determines its use, if the grain is to be used as seed for planting, it should be pure, have a good yield and be free from disease and insect pest. If the grains are to be eaten by man or livestock, a high protein content is desirable and it must taste good (Oranusi *et al.*, 2003). Kunun-zaki is an indigenous fermented non-alcoholic beverage that is widely consumed for its thirst quenching properties. Though consumed throughout the year, it is extensively consumed during the dry season. The drink can also be produced

from fermented sorghum, guinea-corn and maize (Amusa and Odunbaku, 2008).

It is a popular drink with characteristic sweet-sour taste and fermented cereal drink, it is consumed both in rural and urban areas of Northern Nigeria and enhances lactation in nursing mothers, increase libido, sustain erection and increase sperm count (Amusa and Ashaye, 2009). Other food products derived from these cereals include; malted alcohol known as 'Oyokpo' 'pito' or 'burukutu' (Ekanem *et al.*, 2018; Innocent *et al.*, 2011). Like other grains, maize and millet contain essential nutrients such as vitamins A, B and C, minerals like potassium, zinc, anti-diabetic, anti-diuretic and anti-cancerous compound which are useful in treatment of diseases like diabetics, cancer and urogenital tract infections (Amusa and Odunbaku, 2008).

Organisms usually associated with millets grains include *Aspergillus* sp, *Penicillium* sp., as well bacteria like *Bacillus* sp, *Staphylococcus aureus* and *Lactobacillus* sp. (Elmahmood and Doughari, 2007) which predispose consumers to food borne infection and diseases. In a study on safety and quality evaluation of street foods sold in Zaria, Nigeria, Umoh and Odo (1999) found the mean aerobic counts for kunu ranged from 3.67 ± 0.67 to $4.29 \pm 1.14 \log_{10}/g$ with those sold by mobile food sellers having a significantly higher mean ($4.29 \pm 1.14 \log_{10}/g$) than that sold by stationary food sellers ($3.67 \pm 0.4 \log_{10}/g$) with *Bacillus cereus* and *Staphylococcus aureus* being the major bacteria isolated. Oranusi *et al.* (2003) also worked on the hazards and critical control points of kunu-zaki, in Northern Nigeria and discovered that *S. aureus* contamination in all the samples after pitching increased to $2.90 \log_{10}cfu \text{ ml}^{-1}$ while *Bacillus* counts increased from 1.69 to $4.36 \log_{10}cfu \text{ ml}^{-1}$.

The current food safety challenges rises slowly over the years and requires strategic

efforts to be controlled (USFDA, 2008). These products are being produced on daily basis for sales in markets, offices, schools, motor parks and as drinks during festivities, weddings and naming ceremonies. The production procedures and sales of these products are carried out under unhygienic conditions which may predisposed them to many pathogens of public health importance. This study seeks to give an in-depth focus into the sources of microorganisms that could contaminate Kunun-zaki and also identify practices that would aim at reducing the microbial load of the beverage.

MATERIALS AND METHODS

Sample Collection

Fifteen samples of kunun zaki drinks were purchased from different sales outlets in Ikpoba hill market and Aduwawa quarters in Benin City, Edo State during May- June 2011. These samples were placed in sterile bags and transported in a cold pack to the laboratory for analysis within one hour of collection. Statistical analysis of the sample results were carried out using descriptive statistics (mean and standard deviation).

Preparation of Media and Samples

1 ml of every Kunun zaki beverage was placed into 9 ml of distilled water and serial dilution was carried out up to 10^{-10} dilution. The culture media used for microbiological analyses which include nutrient agar, MacConkey agar, potato dextrose agar, eosin methylene blue agar and mannitol salt agar, were prepared according to manufacturers' instruction (Gadage *et al.*, 2004).

Isolation of Microorganisms

Using pour plate method, 1 ml from the dilutions were inoculated on Nutrient agar, MacConkey agar, potato dextrose agar for the enumeration of total microorganism, coliform count and enumeration of fungi respectively. After inoculation, Petri dishes

containing Nutrient agar, MacConkey agar and Mannitol salt agar were incubated at 37°C for 24 h (Mannitol salt agar was used for the isolation of *Staphylococcus*), while inoculated plates containing Potato dextrose agar was incubated at 28°C for 5-7 days (Gadage *et al.*, 2004).

Identification and Characterization of isolates

The isolated organisms were characterized and identified based on their cultural, morphological, and biochemical tests (Buchanan and Gibbson, 1974; Gadage *et al.*, 2004). The colonies were sub-cultured, Gram stained and subjected to biochemical tests such as oxidase, catalase, coagulase, urease, citrate, indole and sugar fermentation tests. These results were also checked on PIBWIN (Probable identification of microorganisms).

Physicochemical analysis

pH of the Kunun zaki drinks was measured by dipping the pH electrode into 10 ml of the beverage placed in a beaker, and the reading were recorded. Total titrable acidity was measured as percentage lactic acid by adding 3 drops of phenolphthalein indicator into 10ml of the drink placed in a conical flask and thoroughly shaken. The mixture was then titrated against 0.1 M NaOH (Sodiun hydroxide) to a pink color end point and the titre value was calculated. Moisture and solid content were determined by methods described by Ceese (1995).

RESULTS AND DISCUSSION

The results of this study showed that the mean total viable bacterial and fungal count was 2.51×10^7 cfu/ml and 0.98×10^7 cfu/ml respectively as shown in Table 1.

These results revealed that samples gotten from Aduwawa market (B₂) had highest bacterial count while Sample from Ikpoba hill market (C₁) has the lowest bacterial count. It also showed that samples gotten from Aduwawa market (A₁) had highest fungal count while Sample from Ikpoba hill market (C₃) has the lowest fungal count. This showed that kunun drinks sold in Aduwawa are heavily contaminated.

In many earlier reports, the pH was 4.3 (Ekanem *et al.*, 2018); 3.80 and 3.99 reported by Innocent *et al.* (2011), 2.42 to 3.83 recorded by Otaru *et al.* (2013), 5.25 to 5.65 reported by Amusa and Ashaye, (2009). The acidity of the kunu drinks may be due to the presence of some bacteria which help in acid fermentation of the kunu products (Ekanem *et al.*, 2018). The results of the investigation showed that the samples of Kunun-zaki contained a fairly large microbial population. The high microbial densities could be related to the fact that usually a heterogeneous population of microorganism are usually involved in the fermentation process and also that foodstuffs are also susceptible to microbial contamination during the processing and storage. The microorganisms isolated from the samples include *Lactobacillus sp*, *Staphylococcus aureus*, *Bacillus sp*, *Streptococcus sp*, *Escherichia coli* and *Pseudomonas sp*, *Aspergillus*, *Mucor*, *Rhizopus*, *Pencillium* and *Fusarium*. The results correlate with that of Amusa and Odunbaku (2008); Oyenuga *et al.*, (2003); Elmahomood and Doughari (2007). Sources of the organisms may be traced to the cereals as had been reported by

Efiuvwevwere and Akoma (1995). The presence of *Lactobacillus* indicates that

contracted by sneezing or by picking of the nostrils by food handlers. *Staphylococcus*

Table 1. Mean Total Viable Counts of Both Bacterial and Fungal Isolates (CFU/ml)

Sample	Bacteria count	Fungal count	Location
A ₁	1.8x10 ⁷	6.00x10 ¹¹	Aduwawa
A ₂	2.56x10 ⁷	12.0x10 ⁹	Aduwawa
A ₃	2.00x10 ⁹	7.00x10 ⁷	Aduwawa
A ₄	1.15x10 ⁹	6.50x10 ⁹	Aduwawa
A ₅	2.08x10 ⁷	12.0x10 ⁷	Aduwawa
B ₁	1.96x10 ⁷	5.50x10 ⁹	Aduwawa
B ₂	5.60x10 ¹¹	20.0x10 ⁷	Aduwawa
B ₃	2.36x10 ⁹	10.5x10 ⁹	Aduwawa
B ₄	1.34x10 ⁹	7.5x10 ⁷	Ikpoba Hill
B ₅	8.70x10 ⁹	15.0x10 ⁷	Ikpoba Hill
C ₁	1.72x10 ⁷	4x10 ¹¹	Ikpoba Hill
C ₂	1.28x10 ⁹	11.0x10 ⁷	Ikpoba Hill
C ₃	1.05x10 ⁹	6.00x10 ⁷	Ikpoba Hill
C ₄	2.51x10 ⁹	9.61x10 ⁷	Ikpoba Hill
C ₅	1.47x10 ⁹	14.0x10 ⁹	Ikpoba Hill
Total Mean count	2.51x10⁷	0.98x10⁷	

kunun-zaki is a lactic acid bacteria fermented beverage and it is not unexpected because they help in fermentation process. *Lactobacillus* in food samples tend to dominate by preventing other pathogenic microorganisms from surviving in the beverage, and their ability to produce lactic acid reduces the pH of food medium. Most food poisoning organisms cannot tolerate low pH, therefore the isolation of *Lactobacillus* favors this fact. Efiuvwevwere and Akoma (1995) reported the presence of some bacteria including *Lactobacillus spp* in kunun drink. The presence of *Staphylococcus aureus* indicate contamination from handlers as *Staphylococcus aureus* is a normal flora of the skin, nose, throat, palms, hairs and mucus membrane and a common etiological agent of septic arthritis (Charles et al., 2005; Emmanuel-Akerele and Uchendu, 2021). The contamination could have been

aureus can also produce Staphylococcal bacteraemia and abscesses in cell during food infection. *Escherichia coli* in food is an indication of faecal contamination of product. However, *E. coli* is a normal flora of the intestinal tract of man, presence of it in excess could lead to gastroenteritis and bacterial diarrhea disease (Emmanuel-Akerele and Uchendu, 2021). This fail to agree with the WHO standards which suggested that water that contain >10coliforms/100ml or one *E. coli*/100ml with or without other coliforms is unsatisfactory for human consumption. *Streptococcus sp.* may also have been enumerated from the beverage as a result of the handlers, since it is also normal flora of the throat and the buccal activity. The presence of *Bacillus* could render a beverage unsuitable for human consumption (Innocent et al., 2011). It is possible that the

contamination by this pathogen may have occurred during sieving and packaging, as most of the people involved in the production, packaging and hawking do not take necessary precautions, and so such

environmental contamination. The pathogenic microorganisms isolated exceed permissible limit (Efiuvwevwere and Akoma, 1995; USFDA, 2008). Production of kunun drinks should be done under hygienic

Table 2. Characteristics of Bacterial Isolated from the Kunun-Zaki Sample

Characteristics	Description					
Cultural						
Color	Cream	Cream	Cream	Yellow	Cream	Florescent green
Surface appearance	Mucoid	Rough	Dry	Mucoid	Smooth	Rough
Elevation opacity	Umbonated translucent	Slightly convex opaque	Convex opaque	Flat opaque	Convex semi-transparent	Convex opaque
Morphological						
Gram stain	+	+	-	+	+	-
Cell type	Rods	Rods	Rods	Cocci	Cocci	Rods
Cell arrangement	Single	Chains	Single	Cluster	Single	Chains
Motility	-	+		-	+	+
Biochemical						
Catalase	-	+	+	+	-	-
Coagulase	-	-	+	+	-	-
Oxidase	-	-	-	-	-	+
Urease	-	-	-	-	-	-
Citrate	+	-	-	+	-	+
Indole	-	-	+	-	-	-
Sugar fermentation						
Glucose	AG	A	AG	A	A	-
Lactose	A	-	AG	A	A	-
Manitol	A	A	A	A	A	-
Bacteria isolated	<i>Lactobacillus sp</i>	<i>Bacillus sp</i>	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Streptococcus sp</i>	<i>Pseudomonas sp</i>

Key: AG- Acid and gas production; A- Acid production

Table 2 shows the cultural, morphological and biochemical characteristics of the isolates. The bacteria isolated were *Lactobacillus*, *Bacillus sp*, *Escherichia coli*, *Staphylococcus aureus*, *Streptococcus sp* and *Pseudomonas sp*

contamination could be very prominent. *Bacillus* is a spore former and as such the spores were easily distributed and was able to withstand high temperature and pH to fully germinate (Otaru *et al.*, 2013). The organism has the potential of causing an array of infections. The presence of *Pseudomonas* is not of great significance due to their low population. They could have occurred due to

conditions to avoid proliferation and spread of these pathogenic organisms.

The fungal isolates present could be traced right to when the grains were either being harvested or stored. The presence of *Aspergillus*, *Penicillium*, and *Fusarium* in the kunun-zaki samples might not be too surprising as they are known as common spoilage organism of carbohydrate foods as

Table 3. Cultural and Morphological Characteristics of the Fungal Isolate

Isolates	Physical Appearance	Microscopic Observation	Fungi isolated
F ₁	Black powdery threads	Septated mycelium conidiophore septate and arising from foot cell. Bear sterigmata conidia in chains and black coloration	<i>Aspergillus sp</i>
F ₂	Dirty blue powdery growth, hair-line	Septated branched mycelium, septated aerial conidiophores with brush-like spore bearing head with sterigmata bore in clusters	<i>Penicillium sp</i>
F ₃	White thread with surface colored black	Non-septated with aerial sporangiophore. Round columella smooth spores. No stolons and rhizoids	<i>Mucor sp</i>
F ₄	Pure white thick and abundant cotton mycelium	Non-septated with stolons and rhizoids. Sporangiophores arising at the nodes. Sporangia are usually black	<i>Rhizopus sp</i>
F ₅	Cotton-like growth with white coloration	Separated with large canoe-shaped microconida on branched conidiophores	<i>Fusarium sp</i>

well as storage microflora of many cereals including sorghum (Ekanem et al., 2018;

preparation and finally germinate in the finished product. The presence of these fungi

Table 4. pH and Titratable Acidity

Sample	pH	Titratable acidity (ML 0.1M NaOH)	LOCATION
A ₁	4.22	2.90	Aduwawa
A ₂	4.21	2.80	Aduwawa
A ₃	4.22	2.84	Aduwawa
A ₄	4.31	2.80	Aduwawa
A ₅	4.20	2.80	Aduwawa
Mean value	4.23	2.83	
B ₁	4.30	2.75	Aduwawa
B ₂	4.27	2.65	Aduwawa
B ₃	4.21	2.70	Aduwawa
B ₄	4.98	2.66	Ikpoba Hill
B ₅	4.27	2.60	Ikpoba Hill
Mean value	4.21	2.67	
C ₁	4.33	2.68	Ikpoba Hill
C ₂	4.31	2.68	Ikpoba Hill
C ₃	4.33	2.66	Ikpoba Hill
C ₄	4.27	2.72	Ikpoba Hill
C ₅	4.30	2.70	Ikpoba Hill
Mean value	4.33	2.70	
Total mean	4.26±0.09	2.73±0.08	

Omonigho and Osubor, 2002). The fungal may produce spores attached to the grains and overcome adverse condition during the

such as *Aspergillus*, *Mucor*, *Fusarium* and *Rhizopus* is associated with spoilage of the beverage (Oyenuga et al., 2003). Some of

these fungal species elicit some toxins which are very hazardous. One of such is *Aspergillus* which produce aflatoxins that are quite harmful and as such their occurrence is undesirable. Table 3 shows the cultural and morphological characteristics of the fungi isolated and they are *Aspergillus sp*, *Penicillium sp*, *Rhizopus sp*, *Mucor sp* and *Fusarium sp*. The pH value of the samples ranged from of 4.21 to 4.30, while the titrable acidity shows the presence of organic acids and it ranged from 2.67 to 2.82ml.

The acidity level of kunun-zaki drinks have been described by several researchers including Efiuwewwere and Akoma (1995) and Amusa and Ashaye (2009), who attributed these to the presence of lactic acid bacteria. The acidity tends to increase with increase in fermentation period resulting into spoilage. Consequently, the low pH value may have encouraged the growth of fungi and

this could be responsible for the species of microorganism isolated. The pH brought about a corresponding increase in the titratable acidity and sour taste flavour of the kunun-zaki drink. The moisture and solid content of the analyzed Kunun-Zaki had overall mean of 85.90±0.95 and 14.1± 0.95 respectively. The low moisture contents could be due to the ease of moisture loss during production (drying resulting from hydrolytic enzyme activities during malting and incubation). The relatively high solid content indicates that the kunun can develop off flavor and colour if stored at room temperature for few days. The presence of all these organisms indicated in this report are of great public health concern as it has passed the permissible limit by WHO (WHO, 1996; Ekanem *et al.*, 2018), and in situations where the beverage is contaminated, quick medical

Table 5. Percentage of Moisture and Solid content present in the Sample

Sample	Moisture content (%)	Solid content (%)	Location
A ₁	86.47	13.53	Aduwawa
A ₂	85.80	14.20	Aduwawa
A ₃	85.85	14.15	Aduwawa
A ₄	85.83	14.17	Aduwawa
A ₅	85.89	14.11	Aduwawa
Mean value	85.97	14.03	
B ₁	87.22	12.78	Aduwawa
B ₂	86.83	13.17	Aduwawa
B ₃	86.70	13.30	Aduwawa
B ₄	86.86	13.14	Ikpoba Hill
B ₅	87.08	12.92	Ikpoba Hill
Mean value	86.94	13.06	
C ₁	84.99	15.01	Ikpoba Hill
C ₂	83.90	16.10	Ikpoba Hill
C ₃	85.25	14.75	Ikpoba Hill
C ₄	85.27	14.73	Ikpoba Hill
C ₅	84.51	15.49	Ikpoba Hill
Mean value	84.78	15.22	
Total mean	85.90±0.95	14.10±0.95	

care should be sought to avoid food poisoning.

CONCLUSION

Since there are no routine hygiene standard techniques for preparation of kunun-zaki such food will always contain an unusual large population of fermentative beneficial organisms and some pathogenic microorganisms. This study has shown that the preparation procedure for kunun-zaki does not completely eliminate microorganisms from the finished products. Storage of the product at room temperature allowed for proliferation of microorganisms and this tends to utilize the kunun-zaki constituents resulting in significant changes in the physicochemical composition (pH and titrable acidity) of the product. The isolation of pathogens as *Staphylococcus*, *Streptococci* and *Aspergillus* could be indicative of health hazards, even when their population has been inhibited to an extent by acid produced by the lactic acid bacteria. In order to reduce the rate of contamination and gently enhance the microbiology qualities of the product the following measures should be adhered to; educate producers and hawkers of the product on good sanitary practice during the preparing and sale of the product; advocate the use of boiled water in washing utensils; treated municipal water should be used during processing and dilution of the processed drinks to avoid contamination with entero-pathogenic bacteria; the processing environment should be hygienic and the packaging materials should be sterilized.

REFERENCES

Amusa, N.A and Odunbaku, O.A. 2008. Microbiological and nutritional quality

of hawked kunun (a sorghum based non-alcoholic beverage/widely consumed in Nigeria). *Pakistan Journal of Nutrition* 8(1): 20-25.

Amusa, N.A. and Ashaye, O.A. 2009. Effects of processing on nutritional, microbiological and sensory properties of kunu-zaki (A sorghum based nonalcoholic beverage) widely consumed in Nigeria. *Pakistan Journal Nutrition* 8(3):288-292.

Buchanan, R.E. Gibbson, N.E. 1974. *Bergey's Manual of Determinative Bacteriology* 8th Edition. William and Wilkins Co. Baltimore, 1286pp.

Ceese, J. 1995. Measuring physical properties of foods. *Food Technology* 49:53-63.

Charles, C.A. Dennis, A.A. and Comfort, U.I. 2005. Growth and heat resistant of *Listeria monocytogenes* in kunun-zaki. A sorghum based beverage. *Journal of Food Processing and Preservation* 29(3-4):278-290.

Efiuvwevwere, B. J. O. and Akoma, O. 1995. The microbiology of kunun-zaki, a cereal beverage from Northern Nigeria made during fermentation process. *World Journal of Microbiology and Biotechnology* 11:491-493.

Ekanem, J.O. Mensah, B.J. Marcus, N.S., Ukpe, B.A. 2018. Microbial Quality and Proximate Composition of Kunu Drinks Produced and Sold in Ikot Ekpene Metropolis, Akwa Ibom State, Nigeria. *Journal of Applied Science and Environmental Management* 22(11): 1713–1718.

Elmahmood, A. M. and Doughari, J.H. 2007. Microbial quality assessment of kunun-zaki beverage solid in Girei town of

- Adamawa State, Nigeria. African Journal of Food Science 011-015.
- Gadage, T. H. Nyanka, L.K. and Mutukumira, A.N. 2004. The occurrence, growth and control of pathogens in Africa fermented food. African Journal of food Agriculture Nutrition and Development 4(1): 1-10
- H. A. Emmanuel-Akerele & F.C. Uchendu. 2021. Microbial assessment of frozen foods sold in Ayobo, Lagos. Bacterial Empire. <https://doi.org/10.36547/be.305>.
- Innocent, O.O., Mariam, Y.O., Blessed, K. and James, T.W. 2011. Microbial evaluation and proximate composition of kunun zaki, an indigenous fermented food drink consumed predominantly in Northern Nigeria. Internet Journal of Food Safety 13: 93- 97.
- Omonigho, S.E. and Osubor, C.C. 2002. Analysis of the microbiological composition of Kunun-zaki quality and physico-chemical during storage at 28±20C. Nigerian Journal of Microbiology 16:43-50.
- Onuorah, S.I., Adesiyun, A.A. and Adekaya, J.O. 1987. Survival and multiplication of Staphylococcus aureus and Escherichia coli in Nigeria cereal drink (Kunun-zaki). Effect of spices pH and temperature. Journal food Agric. 1:31-34
- Oranusi, S.U., Umoh, Y.J. and Kwaga, J.K. 2003. Hazard and critical control of kunun-zaki a non-alcoholic beverage in Northern Nigeria. Food Microbiology 20(10):127-132.
- Otaru, A.J., Ameh, CU; Okafor, JO; Odigure, JO; Abdulkareem, AS 2013. Development, carbonation and characterization of local millet beverage (Kunu). Inter. J. Computational Engineer. Res. 3(4):80-86.
- Oyenuga, S.U., Umoh, Y.J. and Kwaga, J.K. 2003. Nigerian Food and Feeding Stuff, 4th ed. University press, Ibadan. 99pp.
- Umoh J. U. & Odoba M. B. 1999. Safety and evaluation of street foods sold in Zaria, Nigeria. Food Contr. 10:9-14.
- United State Food and Drug Administration 2008. Bad bug book. Foodborne pathogenic microorganisms and natural toxins handbook. Available from: www.cfsan.fda.gov/mow/intro.html.
- World Health Organization. 1996. Guidelines of drinking water quality, Vol. 2. W HO, Geneva.