RSU JOURNAL OF BIOLOGY

AND

APPLIED SCIENCES

ISSN: 2811 – 1451



ABOUT US

Rivers State University Journal of Biology and Applied Science (RSUJBAS) publications is a quarterly, open access, international journal for all academic research in science discipline. Microbiology, botany, zoology, environmental biology, chemistry, physics, mathematics, computer science, biochemistry medical laboratory sciences and other applied science related areas. RSUJBAS is a platform set for elites to influence, contribute and communicate to the global environment through their various academic researches. We synergistically engage our noble effort to contribute to the knowledge development, discoveries and innovations in all fields of study. In RSUJBAS we publish research papers on current academic issues with standard scientific reviews. RSUJBAS publishes original research articles, review articles, case studies, short communications, survey report, comparative studies and many more.

Aims and Scope

Rivers state University Journal of Biology and Applied Sciences aims to publish high quality papers that communicate fundamentals and contemporary discoveries both theoretical and practical. Most importantly RSUJBAS seeks to establish a platform for communicating emerging trends in various discipline such as Microbiology, Botany, Zoology, Environmental Biology, Chemistry, physics, Mathematics, Computer Sciences, Biochemistry, Medical Laboratory, Sciences, and other applied sciences related areas.

Description:

- Area of concentration: All science academic disciplines
- Frequency of publishing: Quarterly
- Mode of publishing: both online and print publication
- Language of publication: English
- Double blinded Review Process
- Zero Level Plagiarism Tolerance

Why Publish with us

Low Article Processing Charge (ACP) to promote the research work Easy and Rapid review process

Instant publication upon acceptance

Dedicated editorial and review team for fast review process

RSUJBAS provides hard copies of publication every quarterly

EDITORIAL BOARD

DR. S.A. WEMEDO

Department of Microbiology Rivers State University

PROF. C. K. WACHUKWU

Department of Medical Laboratory Science Rivers State University

DR. (MRS) N.P. AKANI

Department of Microbiology River State University

PROF.E.C. CHUKWU

Department of Plant Science and Biotechnology Rivers State University

PROF. B.O. GREEN

Department of Plant Science and Biotechnology Rivers State University

PROF. J.N. ONWUTEAKA

Department of Animal and Environmental Biology Rivers State University

DR. (MRS) A. P. UGBOMEH

Department of Animal and Environmental Biology Rives State University

DR. (MRS) E. O. IBEGUDEM Department of Medical Laboratory Science Rivers State University

DR. F U. IGWE Department of Biochemistry Rivers State University

DR. V. I. E. ANIREH Department of Computer Science Rivers State University

DR. N. BOISA Department of Chemistry

ISSN: 2811 - 1451

Rivers State University

DR. N. EBERE

Department of Animal and Environmental Biology Rivers State University

DR. D. O. NGEREBARA Department of Geology Rivers State University

DR. D. MARTHIAS Department of Computer Science Rivers State University

PROF.G. C. AKANI.

Department of Animal AND Environmental Biology Rivers State University

PROF.V.B. OMUBO-PEPPLE Department of Physics Rivers State University

DR. A.D. NWAOBURU Department of Mathematics Rivers State University

DR. A. R. C. AMAKIRI Department of Physics Rivers State University

DR. N. M. NAFO Department of Mathematics Rivers State University

> All Correspondence to Prof Sam Wenedu (Editor -in -Chief) Department of Microbiology, Rivers State University <u>Edictor.ibasya@yoo.com</u>

> > Or

OLUCHI DICKSON

Publication Manager Dicksonoluchi87@gmail.com

CONSULTING EDITORS

Prof. F. o. Oroka

Department of Agronomy Delta State University, Abraka

Naluba. N. Goddy (Ph.D.)

Department of Geography and Environmental Studies Faculty of Social Sciences, Ignatius Ajuru University of Education, Rumuolumeni, P.M.B.5047, Port Harcourt, Rivers State.

Godpower- Echie, G.

Department of Integrated Science Ignatius Ajuru University of Education, Rumuolumeni, Port Harcourt.

GUIDELINE FOR MANUSCRIPTS

Manuscripts should be typewritten on an A4sheet having B1.5=line spacing throughout the text. The margins should be 2B54cm (1 inch) in all sides and page number should be consecutively on the bottom of the page. The manuscript should be written in Times New Romans using '12' font size.

For original research paper, the manuscript should be arranged in the following order: Tittle page, Abstract, Keywords Introduction, Materials and Methods Results, Discussion, Acknowledgement, References, Tables with legends and supplementary materials

The tittle page should contain the title, the name(s) of the author(s), the name(s) and address (es) of the instruction(s) where the work was carried out, including a valid e-mail address from the corresponding author along with telephone numbers. The title of the manuscript should be specific and concise but sufficiently informative.

The Abstract should not exceed 250 words and it should contain brief summary of the findings including brief introduction, methodology, results, and conclusions,

The keywords should have a minimum of five and maximum of seven words.

The introduction should provide a clear statement of the problem and indicates aim of the study citing relevant literature to support background statements.

The Materials and Method should include the methods and methodology of the research.

The results should be presented in the form of tables of figures. It should be presented with clarity and precision. Statements used to present results should be written in the past tense. Detailed interpretation of data should not be included in the results but should be put into the Discussion section.

The Discussion should interpret the results clearly and concisely, and should integrate the research findings of this and past studies on the topic. Highlight the significant/unique findings of the research under conclusion.

The acknowledgment of people, grants or funds should be brief.

LIST OF CONTRIBUTORS

Kingsley- Opara, Ngozi

Research Scholar, Department of Computer Science, Ignatius Ajuru University of Education, rivers State, Nigeria.

Prof. Asagba, Prince Oghenekaro.

Visiting Scholar, Department of Computer Science, University of Education (IAUE), Rumolumeni, Port Harcourt, Rivers State Nigeria. Emial:asagbapince@uniport.edu.ng

Gabriel.B.C., Gabriel M.N.O.Asagba

School of Graduate Studies Ignatius AJURU University of Education ((IAUE), Rumolumeni, Port Harcourt, Rivers State Nigeria. Department of Computer Science <u>Gabrielbariyira@gmail.com</u>, <u>meegabzgmail.com</u>

WAIDOR, Tamaramiebi Keith & ASAGBA, Prince Oghenekaro

Department of Computer Science Faculty of Natural and Applied Sciences Ignatius Ajuru University of Education, Port Harcourt Zalimaxxx@gmail.com

Department of Computer Sciences, University of Port Harcourt, Rivers State Nigeria. <u>Prince.asagba@uniport.edu.ng</u>

Fibersima, Alalibo Ralph

Visiting Scholar, Department of Computer Science, University of Port Harcourt, Rivers State Nigeria. Fiberesima.a.r@outlook.com

Asagba, Prince Oghenekaro.

Visiting Scholar, Department of Computer Science, University of Port Harcourt, Rivers State Nigeria. <u>Asagba.prince@uniport.edu.ng</u>

Kingsley- Opara, Ngozi

Research Scholar, Department of Computer Science, Ignatius Ajuru University of Education, Rivers State Nigeria. Email: <u>ngoziopara@g.mail.com</u>

Prof Asagba, Prince Oghenekaro.

Visiting Scholar, Department of Computer Science, University of Port Harcourt, Rivers State Nigeria.

PROFILES OF MICROORGANISMS AND DISEASES ASSOCIATED WITH BIOAEROSOLS AND WAYS OF IDENTIFYING THEM.

DISEGHA, G.C.

Department of Microbiology, Rivers State University, Nkpolu-Oruworukwo, Port Harcourt, Nigeria. Email: gabrieldisegha2@gmail.com

&

NRIOR, R.R.

Department of Microbiology, Rivers State University, Nkpolu-Oruworukwo, Port Harcourt, Nigeria.

ABSTRACT

A review of published works related to the profiles of microorganisms in bioarosols was done. It also gives a description of some specific diseases caused by these organisms. Identification methods used for these organisms was also considered. Some key groups of associated microorganisms are bacteria, fungi, viruses, and protozoa. Examples of bacterial organisms and diseases caused by them (in parenthesis) are Mycobacterium tuberculosis (tuberculosis), Streptococcus pneumoniae (Pneumonia); Haemophilus influenzae (meningitis); and Bordetella pertussis (whooping cough). Some fungi related diseases are Microsporum racemosus (Dermatosis - superficial mycosis); Aspergillus fumigatus (Aspergillosis); and Cryptococcus neoformans (Cryptococcosis). Some Viruses associated with bioaerosols and their related diseases are Corona virus (Corona virus infection (COVID-19); Orthomyxoviruses (Influenza); Paramyxovirus (Influenza, measles, mumps, and pneumoniae in neonates); Rhinoviruses and Corona virus (Cold, Rhinitis) and Pox virus (Cowpox). Pneumocystis carrinii, a protozoa is also associated with bioaerosols. Methods of identification of microorgnisms in bioaerosols are both conventional/classical, and advanced. Some methods are manually operated using miniaturized devices while others are automated, and depends on phenotypically expressed traits for their identification. PCR-based methods are now in vogue. The cost implications, using advanced methods is high and poses a challenge to individuals who may be interested to research in related fields. It is therefore recommended that government agencies should make grants available to researchers in need. Most importantly, some microorganisms associated with bioaerosols are pathogenic and poses a serious health challenge, and are epidemic prone. Thus, affordable and rapid means of detection, identification and prevention of diseases implicating these microorganisms is paramount.

Key words: Profile, bioaerosol, diseases, bacteria, fungi, identification.

INTRODUCTION

Biological aerosols also known as Bioaerosols or organic dust, consists of microorganisms (bacterial cells, fungal spores, detritus, cellular fragments, pollen, viruses, protozoa, microalgae and cyanobacter*ia*), microbial products (such as Lipopolysaccharides) and toxins in the air and other volatile organic compounds. In brief, a bioaerosol is an airborne collection of biological materials which may be solid or liquid. Exposure to these agents may cause

different health conditions such as infection, respiratory diseases, allergies, acute toxic effects, nervous system disorders, and possibly cancer (Lighthart, 1997; Crook and Swan, 2001; Douwes and Thorne, 2008). Some sources of bioaerosols are water, soil, waste dump sites, and sewage. These collections of biological materials form a profile of microorganisms responsible for several debilitating and life-threatening human infections and diseases (Obire *et al.*, 2002; Londahl, 2014; National Academies Press, 2017). The distribution of bioaerosols vary in different geopositions due changing environmental and anthropogenic activities with respect to the procedures of operation, ranging from residential, industrial or agricultural activities (Ekaterina & Agranovski, 2018). There is also the concept of sampling efficiency namely physical efficiency, which is the comparison of the amount of the collected particles to the amount of particles in the sampling location or environment, and biological efficiency, which is an estimation of the fraction of microorganisms that remain viable after collection, since some bioaerosol organisms are non-viable (Hogan *et al.*, 2005; Kulkarni *et al.*, 2011).

Microbial Groups of Bioaerosols

Microorganisms associated with bioaerosols are bacteria, fungi, viruses, protozoa and microalgae, and with their microbial products and volatile organic compounds (Srikanth *et al.*, 2008; Londahl, 2014).

In these groups, bacterial species and their diseases caused by them are *Mycobacterium tuberculosis* (Tuberculosis); *Legionella sp.* (Legionnaires disease, Pontiac fever); *Streptococcus pneumonia* (Pneumonia); *Streptococcus sp.* (Scarlet fever, Angina, laryngitis); *Haemophilus influenzae* (Inflammation of upper and lower respiratory system, Menigitis); *Bordetella pertussis* (Whooping cough); *Legionnella pneumophillia* (Pulmonary infections > Legionnaire disease); Norcardia sp. (Norcardiosis); and Corynebacterium diphtheria (Diphtheria) (Burge1990; Nrior, 2020).

Fungal profiles found in bioaerosols with their respective diseases are *Alternaria sp.* (Asthma, rhinitis); *Cladosporium* (Asthma, rhinitis); *Microsporum racemosus* (Superficial mycosis); *Aspergillus fumigatus* (Aspergillosis); *Cryptococcus neoformans* (Criptococcosis) (Nrior & Chioma, 2017).

In the profile of viruses, the following types are prominent in bioaerosols with their diseases in parenthesis: Influenza A and B (Influenza); Rubella virus (Measles); Corona virus (Corona virus infection – COVID 19); Orthomyxoviruses (Influenza); Paramyxovirus (Influenza, measles, mumps and, pneumoniae among new borns); Rhinoviruses and Corona virus (Colds); Pox virus (Cowpox); Adenoviruses (Sorethroat, Pneumonia); Enteroviruses (Meningitis, Pleurdynia); Rhinoviruses, Hantavirus from rodent faeces, and Coronaviruses (chicken pox) (Diglisic *et al.*, 1999; Nrior, 2020). Prozoal organisms found in bioaerosols is *Pneumocystis carrinii* which causes Pneumoniae in humans. Protozoa and algae are also found in bioaerosols and they are implicated in Hypersensitivity pneumonitis (Burges, 1990; Londahl, 2014).

Disease Categories Associated with Bioaerosols.

Diseases associated with bioaerosols are due to exposure to bioaerosols. The incidences are due to a combination of allergens, toxins and microorganisms. Three distinguishing diseases associated with bioaerosols are infectious diseases, respiratory diseases and cancer (<u>Turjanmaa</u>, <u>1987</u>; <u>Charous *et al.*</u>, <u>1994</u>).

Infectious diseases

Infectious diseases arise from viruses, bacteria, fungi, protozoa and helminthes, and involve the transmission of an infectious agent from a reservoir to a susceptible host through direct contact, a common vehicle, airborne transmission or vector-borne transmission. Diseases caused by microorganisms associated with bioaerosols may be attributable to occupationspecific exposures, such as in health workers (tuberculosis, winter stomach flu, measles). For farmers, abattoir workers, veterinarians diseases of common occurrence are Q-fever, swine influenza and anthrax) and forestry workers (tularaemia). Gathering or clustering of people in the workplace such as in the case of market places, office, military or schools may experience influenza, winter stomach flu, Tuberculosis and Corona infection) (Driver et al., 1994; Van den Ende et al., 1998; Srikanth et al., 2008; NCDC, 2020). Other bioaerosol transmitted infections are (particularly *Legionella pneumophila*) caused by exposure to Legionellae. They become airborne often as a result of active aerosolizing processes (Pastoris et al., 1997; Brown et al., 1999). Aloso, inhalation of fungal spores in the course of handling decaying matters, faeces, composts or soils can result in several mycoses. They include adiaspiromycosis, aspergillosis, blastomycosis, coccidioidomycosis and histoplasmosis. (MMWR, 1993; NIOSH, 1997). Thus, high-risk occupations for occupational infectious diseases due to bioaerosol exposure include farmers, veterinarians, health care workers and biomedical workers studying infectious agents.

Respiratory diseases

<u>Bernstein et al. (1999)</u> explains that respiratory, dust-aerosol associated diseases range from acute mild conditions to severe chronic respiratory diseases that require specialist medical attention. Respiratory diseases due to bioaerosls result specifically from exposures to toxins, pro-inflammatory agents or allergens.

Cancer

Cancer can be caused by a variety of factors including oncogenic viruses and other biological agents. Notably the only clearly established non-viral biological occupational carcinogens are the mycotoxins. Hayes *et al.*, (1984) reported that aflatoxin from *Aspergillus flavus* was capable of causing liver cancer. These occur in industries in which mould-contaminated materials are handled (Anonymous, 1998).

Viruses are considered as the most common cause of infectious diseases acquired within indoor environments (<u>Brankston *et al.*</u>, 2007), and many nosocomial infections are due to respiratory and enteric infections of viruses (<u>Belliot *et al.*</u>, 2014; <u>Bruijning *et al.*</u>, 2012, <u>Kambhampati *et al.*, 2015, <u>Rhinehart *et al.*, 2012). Recent examples of particular concern are emerging infectious diseases such as Severe Acute Respiratory Syndrome (SARS) coronavirus (SARS-</u></u>

CoV)– *virus* identified with outbreak in 2003, the outbreak of Ebola Virus Dissease (EVD) in West Africa in 2014-15, and the on-going Middle East Respiratory Syndrome Coronavirus (MERS CoV) outbreaks in the Middle East since 2012, where many health care workers (HCWs) were infected and acted as the amplifiers for the spread of the disease to the community, and the Corona Virus infection (COVID 19) which became a pandemic and affected virtually all countries in the world from March, 2019 to June, 2020, with implied extension towards the second half of 2020. (Ansumana *et al.*, 2017; Ho *et al.*, 2003; Ki, 2015; Shears and O'Dempsey, 2015; Nrior & Dumbor, 2019; WHO, 2020).

Bioaerosol organism detected by qPCR analysis of aerosol samples.

Quantitative Polymerace Chain Reaction (qPCR) approach has discovered various kinds of fungal and bacterial species. These include both culturable and unculturable species of bacteria, fungi and viruses. According to (Kaarakainen et al., 2011), fungal organisms associated with bioaerosols include Aspergillus fumigatus/ Neosartorya fischeri. Aspergillus niger/awamori/foetidus/phoenicis, Aureobasidium pullulans, Cladosporium cladosporioides, herbarum, Cladosporium sphaerospermum, Eurotium Cladosporium amstelodami/ chevalieri/herbariorum /rubrum/repens, Epicoccum nigrum Penicillium brevicompactum/stoloniferum, Penicillium chrysogenum, Penicillium variabile, Wallemia sebi, Cladosporium spp. Streptomyces spp., and Gram-positive and –, Gram-negative. Several of which are implicated in diverse infectious diseases, posing serious health hazards to humans (Kaarakainen et al., 2011). Some of the microbial bioaerosols are culturable while others were unculturable, so could only be detected using metagenomic approache. Some bacterial and fungal species detected through metagenomic studies include *Micrococcus*. Nocardiopsis, Paracoccus, Streptomyces, Scopulariopsis, Pseudonocardia, Staphylococcus, Enhydrobacter, Methylobacterium, Corynebacterium, Sphingomonas, Acinetobacter, Bacillus, Kocuria, Massila and Nocardioides. Some fungi in indoor and outdoor air identified at the phylum and class levels discovered through metagenomic insight are given on Table 1 (Shin et al., 2015).

Dothideomycetes	Eurotiomycetes	Sordariomycetes
Leotiomycetes	Pezizomycetes	Saccharomycetes
Orbiliomycetes	Lecanoromycetes	Arthoniomycetes
LIchinomycetes	Unclassified Ascomycota	Agaricomycetes
Tremellomycetes	Exobasidiomycetes	Waalemiomycetes
Ustilaginomycetes	Agaricomycotina	Agaricostilbomycetes
Dacrymycetes	Atractiellomycetes	Tritirachiomycetes
	Chytridiomycetes	Unclassified Glomeromycota.
	Unclassified Eukarya	

Table 1: Fungi in indoor and outdoor air identified at non-specific levels

Source: Shin et al, (2015).

Bioaerosols are generated due to anthropogenic activities. Such activities include residential, industrial, agricultural and laboratory experimentation, Bioaerosols can be generated in the laboratory by a number of different methods utilizing either dry or wet procedures. Dry dispersion methods are predominantly utilized for fungal aerosol generation. Fungal spores can be detached and dispersed directly from their culture medium by vibration (Scheermeyer and Agranovski, <u>2009</u>), brushing or establishment of airflow across the surface of the agar plate (Jung *et al.*, <u>2009</u>; Zhen *et al.*, <u>2014</u>).

Bioaerosols are also generated in anthropogenic environments by bursting bubbles: When bubbles from sea waves burst whitecaps becomes efficient producers of marine bioaerosol particles. In residential and industrial environment transfer of microorganism from water to air by bursting bubbles occur frequently during aeration or agitation of liquids (Aziz *et al.*, 2008; Chen *et al.*, 2013; Mannina *et al.*, 2016). Other methods of bioaerosol generation are liquid dispensing and splashing with high probability of bacteria surviving more than one hour after generation process (Joung *et al.*, 2017); High pressure cleaning using high pressure mechanical spray guns and machines applied in various industrial and residential areas for cleaning contaminated surfaces (Seidl *et al.*, 2016).

Transmission of Bioaerosol associated microorganisms/diseases

In nosocomial instances, airborne transmission usually occurs only when an infected subject is coughing, sneezing, or otherwise actively shedding fresh organisms into air close to susceptible individuals. Bio-aerosols can be transmitted either at long distances beyond the patient room environment, or within short distances. Small particle are transmitted to persons in the immediate area near the patient. Viruses like Corona virus (COVID 19), Severe Acute Respiratory Syndrome (SARS), influenza and norovirus are transmitted from patients usually by contact and/or droplet routes, while airborne transmission occurs over a limited distance (Bollin *et al.*, 1985). Centre for Diseace Control (2020) recommends at least 6 feet (2 arms) length physical/social distance from person to person (Legionella may be derived from the environment (Bollin *et al.*, 1985) and others include contaminated food, water, medications (e.g., intravenous fluids) or through vectors (Anaissie *et al.*, 2003).

Transmission routes of infections is not always easily determined in an environment with undefined parameters. Infection by direct contact can occur when infected hosts are in close proximity with a susceptible population. On the other hand, infected hosts can transmit the disease without direct contact. Moreover, many microorganisms, including viruses (<u>Dinsdale et al.</u>, 2008), can remain infectious outside their hosts for prolonged periods of time, and this can lead to infections by indirect contact. For example, a surface can become contaminated by deposited infectious droplets and eventually cause the infection of susceptible hosts coming into contact with it. The probability of airborne transmission of an infectious disease can be determined by conducting epidemiological studies (<u>Pirtle</u> and Beran, 1991) and/or by analyzing the microbiological content of air samples.

Methods of Identification of Microorganisms in Bioaerosols

Sampling and Cultivation of airborne bacteria and fungi follows mostly a passive approach. The passive sampling based on settle plate (sedimentation sampling) method which is preferred mostly in the collection of air borne particles containing microorganisms (Sivagnanasundaram *et al.*, 2019)

Bacteria: Identification of microorganisms from bioaerosols according to conventional methods. In this proposal bioaerosol samples are collected in a specified environment and their cultivation on agar nutrient media. Representative colonies are picked and inoculated unto nutrient agar to obtain pure cultures. The bacterial isolates are identified either gram positive or gram negative based on their reaction with gram stain. The pure cultures are stored as frozen 10%(v/v) glycerol suspensions at -35° C in a refrigerator. This glycerol stock serves as a means for fresh working cultures. Further inoculation of pure cultures unto appropriate media is done to check for consistency. Identification of the isolates are carried out using appropriate identification schemes, which supposes that traditional schemes for identifying a pure culture phenotype characteristics chosen for an identification scheme should be easily determinable by most microbiology laboratories (Krieg, 2001; <u>Duquenn</u>, (2018; Sivagnanasundaram *et al.*, 2019).

Fungi: Isolation and identification of fungi is based on their macroscopic morphology, such as best growth temperature (*e.g.* Growth at 37° C, 40° C, 45° C), growth rate (e.g. colony diameter 5 cm in 15 days), colour or pigmentation on SDA (e.g. white, cream, yellow, brown, pink, grey, black *etc.*), colour/pigmentation on reverse side of plate (e.g. none, yellow, brown, red, black, *etc.*), texture and special features (e.g. glabrous, suede-like, powdery, granular, fluffy, downy, cottony), Surface topography (*e.g.* flat, raised, heaped, folded, domed, radial grooved), septation of hyphae (septate, non-septate), while the microscopic morphologies and identities of the different species of fungal isolates are based on characteristic features of conidiophore, phialides, vesicle, sclerotia, hulle cells, sporangiophore, apophysis, columella, sporangium and rhizoids according to scheme of Cheesbrough, 2002; Kidd *et al.*, 2016; Lindsley, *et al.*, 2017; Nrior, 2020).

Some Settled Dust Collection Devices used for Bioaerosol sampling.

Instead of passive sedimentation method, bioaerosol is also sampled using settled dust collection devices. These include Vacuums, Swabs, wipes, adhesive tapes and contact plates. Their respective methodologies are giving by HUD (2008); Bolanos-Rosero *et al.* (2013); Morey, (2007) and Poletti *et al.*, (1999).

Enumeration of viruses

Prior to molecular PCR-based detection methodologies, cell cultures and serological methods were used as detection methods of viral pathogens. By the use of commercially available immortal cell lines, researchers can screen collected bioaerosols by inoculating cells and observing for modal cytopathic effects (CPEs) such as fusion and lysis of cells (Leland and Ginocchio, 2007).

Sampling and Characterization of Viruses in Bioaerosols

Several methods are available, but only few are mentioned below.

Viral Plaque assay (VPA)

A common method for detecting and quantifying viral pathogens and titres respectively is the viral plaque assay (VPA) (Condit, 2007).

Tissue Culture infectious dose Assay (TCID)

Tissue Culture Infectious Dose assay (TCID) also known as endpoint dilution assay, is also a culture based method of assaying viruses in bioaerosols (Condit, 2007). The procedure selecting cells which are plated at a predetermined concentration in a 96-well format and inoculated with serial dilutions of the collected sample. Predetermined incubation period is set, and then cytopatic effects (CPEs) are closely observed. The TCID50 is defined as the dilution of virus required to infect 50% of the cell culture wells (Reed & Muench, 1938). Based on the number of cells that are infected at the designated virus dilution, viral titers are mathematically calculated. However, this method has its limitation (Reed & Muench, 1938).

Immuofluorescence Antibody (IFA) Assay

Detection and quantification of viral loads is also achieved via direct or indirect immunofluorescence antibody (IFA) assays and are frequently used in combination with cell culture-based methods (Flint *et al.*, 2009; Tortora *et al.*, 2013). Infected cells are by this method combined with a fluorescently-labeled, antigen-specific antibody. Optimization is possible by varying the parameters (Flint *et al.*, 2009; Tortora *et al.*, 2013).

Molecular Genetic Methods

Molecular identification of bioaerosol samples follows a predetermined procedure. Samples are collected in a specified environment and subsequently, cultivation on agar nutrient media (except for viruses). The morphologically different colonies are then selected, and individually transferred into a DNA-free reaction tubes. *PCR Amplification of Purified Bacterial DNA using* 16S rRNA gene region of bacterial genomic DNA is amplified using universal bacterial primers. An amplicon-mixture of each bioaerosol sample is prepared by mixing equimolar DNA amounts of PCR product from each selected colony. Then, the amplicon-mixture is used to generate clone libraries that are sequenced; the gene sequences are finally aligned against the NCBI database for bacterial identification (Schäfer *et al.*, 2017; Disegha & Akani, 2919; Sivagnanasundaram *et al.*, 2019).

PCR-based method of identifying Viruses from Bioaerosol.

Generally, viruses consist of more genetic material, DNA or RNA than other microorganisms. More general studies have also been conducted (Sigari *et al.*, 2006) based on PCR detection of a number of viruses, especially enteroviruses and reoviruses, from aerosols around a sewage treatment facility. The most generalized detection of viruses, however, lies in the metagenomics studies mentioned above, particularly the study of Dinsdale *et al.* (2008). In this study, viral DNA only, not RNA, was isolated from the smallest-size fraction of a serially filtered environmental sample, and included a wide variety of viruses, phages and prophages.

More effective and rapid detection of viruses in collection media, preferably in real-time, is well-preferred in aerovirology. Some good technologies include loop-mediated isothermal amplification, which has the potential to detect and offer a presumptive identification of a virus under one hour, as demonstrated for influenza virus (Mori and Notomi, <u>2009</u>), and real-time PCR.

Analytical Formula for Direct Sedimentation Methods Culturable Microbial Sample Analysis.

In culturable microorganisms, the number of both bacterial and fungal colonies is enumerated on each agar plate after incubation and the counts are obtained as colony forming units per m^2 area (cfu/m2) (Sivagnanasundaram *et al.*, 2019. A good background information on enumeration of bioaerosol microorganisms can be found in Nrior & Chioma (2017). Analytic method is used to calculate the estimated number of colony forming units from a bioaerosol sample. The standard time for bioaerosol sampling is ten minutes (10 mins) (Nrior, 2020).

For Direct Sedimentation Method:

$$CFU(min - m^2 = \frac{Number \ of Colonies}{Time \ of \ Exposure \ (Min) \ x \ \pi r^2}$$

Where r = radius of media plate (Petri dish) used (in meters).

The above method uses settled plates using prepared Petri dishes with preferred medium for microbial growth.

For Indirect Sedimentation Method:

$$CFU(min - m^2 = \frac{Number \ of Colonies \ x \ VD}{Time \ of \ Exposure \ (Min) \ x \ \pi r^2 x \ VP}$$

Where: r = radius of beaker used in (in meters); VD = Volume of diluent (normal saline 100 ml in 250ml beaker) and VP = Volume plated (usually 0.1 ml aliquot /inoculum) (Nrior and Dumbor, 2019). The indirect method uses beaker with diluents placed for at least 24 hours.

Other Devices used for bioaerosol sampling

This review considers only settle plate (gravitational sedimentation) method for bioaerosol sampling. Settle plates are culture plates containing culture media exposed at sampling locations of interest for passive inoculation of microorganisms. Airborne particles are allowed to settle onto the plates for a specified time. Usually 10 minutes (Nrior, 2020), and the plates are then closed, incubated and inspected for growth (Dyer *et al.* 2004). For purposes of simplicity and efficiency in cost, settle (gravitational sedimentation) plates are more frequently used by researchers in conducting bioaerosol sampling. However, there are other devices or techniques used for bioaerosol sampling. Some of them are given below:

(i)Filters: These are useful for personal bioaerosol sampling because filter-based collectors are small and lightweight and work well with personal sampling pumps (Raynor *et al.*, 2011).
(ii) Impactors: These are described by Hering (2001) and Marple and Olson (2011) as instruments consisting of a series of nozzles (circular- or slot-shaped) and a surface for

impaction (Marple and Willeke, 1976). Air is drawn into the impactor using a vacuum pump, and the air stream flows through the nozzles and toward the impaction surface, where particles are separated from the air stream by their inertia. Larger particles collect on the impaction surface, while small particles that do not impact follow the air stream. The impaction surface typically consists of a greased plate or tape, filter material, or growth media (agar) contained in Petri dishes. In some applications, impactors are not used as collection devices themselves, but rather to remove particles above a certain size before collection or characterization of the downstream aerosol (Figure 1). The most commonly used impactor for sampling airborne culturable bacteria and fungi is the Andersen impactor illustrated below (Andersen ,1958).



Figure 1: Example of impaction mechanism. Existing air stream in the impactor nozzle, quickly changes direction following the arrows. On the left, smaller/ligher particles flow with the air stream and are not collected. Larger particles cannot change direction as quickly due to their higher inertia and collide with the collection surface and are accumulated. (Source: Lindsley *et al.*, 2017)

(iii) Cyclones: A cyclone sampler consists of a circular chamber with the aerosol stream entering through one or more tangential nozzles (Hering, 2001). Clone is similar to an impactor; a cyclone sampler depends upon the inertia of the particle to cause it to deposit on the sampler wall as the air stream curves around inside. See Figure 2



Figure 2: Cyclone aerosol collection. From the inlet, aerosol stream enters the body of the cyclone, the air flow is according to curved interior wall and moves in a spiral pattern. Collision of larger aerosol particles with the inner wall occur due to the inertia of the particles and accumulate. After spiraling downward, the air flow exits through the vortex finder via the center of the cyclone. Source: Lindsley *et al.* (2017).

(iv) Liquid Impingers: Many microorganisms can lose their viability if they are collected onto dry solid surfaces. Impingers often have curved inlets to remove larger particles from the air

stream before collection. Because impingers are essentially another type of inertial collection device, they have a collection efficiency curve and a cut-off diameter like impactors and cyclones. Additives to the collection medium such as proteins, antifoam, or antifreeze aid in resuscitation of bacterial cells, prevent foaming and loss of the collection fluid, and minimize injury to the cells (Chang and Chou, 2011; Cown *et al.* 1957; Dungan and Leytem, 2015).



Figure 3: Impingement. Bioaerosol particles exit the nozzle of the impinger at high velocity and impact the liquid or the bottom surface of the collection vessel. Some types of impingers produce air bubbles in the collection media, which can enhance particle collection, but can damage some types of microorganisms. Source: Lindsley *et al.* (2017).

(v). Wetted-surface bioaerosol samplers (WSBS)

Wetted-surface bioaerosol sampler generates streams of air, which impacts onto a wetted surface with collection media (Kesavan and Sagripanti, 2015; Kesavan *et al.*, 2011). WSBS produce bubbling and agitation, which may not be favourable to some microorganisms (Lin *et al.*, 2000).

CONCLUSION:

This study has made some exposition on the profile of microorganisms associated with bioaerosols. It was noted that the predominant microbial groups of bioaerosol are fungi, bacteria and viruses, while protozoa and microalgae were present in low numbers. Some disease categories caused by specific organisms or combination of them are infectious diseases, and respiratory diseases expressed on humans in varying degrees, depending on the measure of exposure. Sources of micro-bioaerosols and transmission routes are critical for contact of microorganisms and their consequent infection on susceptible hosts. Some sampling devices are also mentioned which are used in bioaerosol researches. Detection and identification of bioaerosol microorganisms used to be carried out using cultural and biochemical methods. But recently, the trend has changed to molecular approaches using different advanced method, including the real time PCR and other analytical techniques. The use of advanced techniques enhances the detection of microoganisms including non-culturable species via metagenomic

approaches, is a great achievement. However, cost of advanced techniques is high and not easily affordable by research students, and even some Research Institutes.

It is therefore recommended that government agencies such as TETFUND, World Bank and non-governmental establishments should make grants available to researchers in need of such equipment and devices. Most importantly, some microorganisms associated with bioaerosols are pathogenic and are of significant health challenge, and are epidemic prone. Thus, affordable and rapid means of detection, identification and prevention of diseases and epidemics from bioaerosols associated microorganisms is paramount.

ACKNOWLEDGEMENT

We acknowledge with gratitude the various authors whose works are used and cited in this review. Their detailed reports actually widened the horizon of this article. Thanks also due to Dr. R.R. Nrior for his encouragement in partnership and provision of materials which served as guideline. We also wish to appreciate Professor O. Obire who initially introduced the corresponding author to the culture of learning how to write academically. Thanks are also due to other members of the academic community in the Rivers State University, Port Harcourt, Nigeria, who have assisted us in diverse ways.

REFERENCES

- Anaissie, E.J., Stratton, S.L, Dignani, M.C., Lee, C.K., Summerbell, R.C., Rex, J.H. *et al.* (2003). Pathogenic molds (including *Aspergillus species*) in hospital water distribution systems: A 3-year prospective study and clinical implications for patients with hematologic malignancies. *Blood*; 101:2542-2546.
- Andersen, A.A. (1958). New sampler for the collection, sizing, and enumeration of viable airborne particles. *Journal of Bacteriology*, 76(5):471-484.
- Anonymous. (1998) Respiratory health hazards in agriculture. Am. J. Respir. Crit. Care Med., 158: S1–76.
- Ansumana, R., Keitell, S., Roberts G.M., Ntoumi F., Petersen E., Impolite G., and Zola (2017)
 A. Impact of infectious disease epidemics on tuberculosis diagnostic, management, and prevention services: experiences and lessons from the 2014-2015 Ebola virus disease outbreak in West Africa. *International Journal of Infectious Diseases*, 56:101–104.
- Aziz, H.A., Adlan, M.N. and Ariffin, K.S. (2008). Heavy metals (Cd, Pb, Zn, Ni, Cu and Cr (III)) removal from water in Malaysia: post treatment by high quality limestone. *Journal* of Bioresource Technology, 99:1578–1583.
- Belliot G., Lopman B.A., Ambert-Balay, K. and Pothier P. (2014). The burden of norovirus gastroenteritis: an important foodborne and healthcare-related infection. *Clinical Microbiology and Infection*, 20(8):724–730.
- Bernstein, I.L., Chan-Yeung, M. Malo, J.L. and Bernstein, D.I. (eds). (1999). Asthma in the workplace, 2nd edn., New York, NY: Marcel Dekker.
- Bolaños-Rosero, B., Betancourt, D., Dean, T. and Vesper, S. (2013). Pilot study of mold populations inside and outside of Puerto Rican residences. *Aerobiologia*, 29(4):537-543.

- Bollin, G.E., Plouffe, J.F., Para, M. F. and Hackman, B. (1985) Aerosols containing *Legionella pneumophila* generated by Shower Heads and Hot-Water Faucets. *Journal of Applied and Environmental Microbiology*, 50:1128-1131.
- Brankston, G., Gitterman, L., Hirji, Z., Lemieux, C. and Gardam, M. (2007). Transmission of Influenza A in human beings. *Lancet Infectious Diseases*, 7(4):257–265.
- Brown, C.M., Nuorti, P.J., Breiman, R.F. *et al.* (1999). A community outbreak of Legionnaires' disease linked to hospital cooling towers: an epidemiological method to calculate dose of exposure. *Int. J. Epidemiol.*, 28: 353–9.
- Bruijning, P., Quach C. and Bonten, M. (2012). Nosocomial rotavirus infections: a metaanalysis. *Pediatrics*, 129(4): e1011–e1019.
- Burges, H. (1990). Bioaerosols Prevalence and health effects in the indoor environment. *The Journal of Allergy and Clinical Immunology*, 88(9): 1-15.
- Centre of Disease Control (2020). What is social distancing? Centre For Disease Control and Prevention. <u>https://www.cdc.gov/coronavirus/2019-ncov/prevent-getting-sick/social-distancing.html</u>
- Chang, C.W. and Chou, C. (2011). Assessment of bioaerosol sampling techniques for viable *Legionella pneumophila* by ethidium monoazide quantitative PCR. *Aerosol Sci Technol;* 45(3):343-351.
- Charous, B.L., Hamilton, R.G., and Yunginger, J.W. (1994) Occupational latex exposure: characteristics of contact and systemic reactions in 47 workers. *J Allergy Clin Immunol.*, 94: 12–8.
- Cheesebrough (2002). *District laboratory practice in tropical countries* (Part 2). Cambridge: Cambridge University Press.
- Chen, W.H., Yang, W.B., Yuan. C.S., Yang, J.C. and Zhao, Q.L. (2013). Influences of aeration and biological treatment on the fates of aromatic VOCs in wastewater treatment processes. *Journal of Aerosol Air Quality Research*. 13:225–236.
- Condit, R.C. (2007). Principles of virology. In: Knipe D.M., Howley, P.M., eds. *Fields virology*. Philadelphia: Lippincott, Williams & Wilkins.
- Cown, W.B. Kethley, T.W. and Fincher, E.L. (1957). The critical-orifice liquid impinger as a sampler for bacterial aerosols. *Appl Microbiol;* 5(2):119-124.
- Crook, B. and Swan, J.R. (2001). Bacteria and other bioaerosols in industrial workplaces. Microorganisms in Home and Indoor Work Environments; Diversity, Health Impacts, Investigation and Control. Flannigan B., Samson R.A., Miller J.D. (Editors), 69–82.
- Dinsdale, E.A., Edwards, R.A., Hall, D., et al. (2008). Functional metagenomic profiling of nine biomes. *Nature*, 452, 629-632.
- Diglisic, G., Rossi, C.A., Doti, A. and Walshe, D.K. (1999). Seroprevalence study of Hantavirus infection in the community based population. *Md Med. J.*, 48:303-306.
- Disegha, G.C. and Akani, N.P. (2019). Bioinformatic method for fungi identification. *Journal* of Biology and Genetic Research, 5 (1): 1-14.
- Douwes, J. and Thorne, P.S. (2008). Bioaerosols in: *International Encyclopaedia of Public Health*, 2nd Ed., London: Elsvier, pp. 107 121.
- Driver, C.R., Valway, S.E., Morgan, W.M., Onorato, I.M., and Castro, K.G. (1994). Transmission of *Mycobacterium tuberculosis* associated with air travel. *J. Am. Med. Assoc.*, 272: 1031–5.

- Dungan, R.S. and Leytem, A.B. (2015). Recovery of culturable Escherichia coli O157:H7 during operation of a liquid-based bioaerosol sampler. *Aerosol Sci Technol*; 50(1):71-75.
- Duquenn, P. (2018). On the identification of culturable microorganisms for the assessment of biodiversity in bioaerosols. *Annals of Work Exposures and Health*, 62 (2): 139– 146, https://doi.org/10.1093/annweh/wxx096.
- Dyer, R.L., Frank, J.F., Johnson, B., Hickey, P. and Fitts, J. (2004). Microbiological tests for equipment, containers, water, and air. In: Wehr H.M., Frank J.F, eds. *Standard methods for the examination of dairy products*. Washington, DC: American Public Health Association.
- Ekaterina, M. & Agranovski, I.E. (2018). Sources and mechanisms of bioaerosol generation in occupational environments. *Critical Reviews in Microbiology*, 44 (6): 739 – 758.
- Flint, J.S., Enquist, L.W. and Shalka, A.M. (2009). Virological methods. In: *Principles of Virology*, 3rd edition. Washington, DC: American Society for Microbiology.
- Hayes, R.B., Van Nieuwenhuize ,J.P., Raatgever, J.W. and Kate, F.J.W. (1984). Aflatoxin exposures in the industrial setting: An epidemiological study of mortality. *Food Chem Toxico.*, 22:39-43.
- Hering S.V. (2001). Impactors, cyclones, and other particle collectors. In: Cohen B.S., McCammon C.S. Jr., eds. Air sampling instruments for evaluation of atmospheric contaminants. Cincinnati, OH: American Conference of Governmental Industrial Hygienists.
- Ho, P.L., Tang, X.P. and Seto, W.H. (2003). SARS: hospital infection control and admission strategies, 8: 41 45.
- Hogan, C.J., Kettleson, E.M., Lee, M.H., Ramaswami, B., Angenent, L.T. and Biswas,
 P. (2005) Sampling methodologies and dosage assessment techniques for submicrometre and ultrafine virus aerosol particles. J. Appl Microbiol., 99: 1422–1434.
- HUD (2008). *Vacuum dust sample collection protocol for allergens*. Washington, DC: Department of Housing and Urban Development, <u>http://portal.hud.gov/</u>
- Jonges, M., van Leuken, J., Wouters, I., Koch, G., Meijer, A., Koopmans, M. (2015) Windmediated spread of low-pathogenic Avian influenza virus into the environment during outbreaks at commercial poultry farms. *PLoS ONE*, 10, e0125401.
- Joung, Y.S., Ge, Z. and Buie, C.R. (2017). Bioaerosol generation by raindrops on soil. *Natural Communities*, 8:14668
- Jung, J.H. Chang., H.L., Eun, L., Jun, H.L., Sang, S.K., and Byung, U.L. (2009. Design and characterization of a fungal bioaerosol generator using multi-orifice air jets and a rotating substrate. Journal of Aerosol Science, 40:72–80.
- Kaarakainen, P., Rintala, H., Meklin, T., Päivi, K. P., Hyvärinen, A. (2011).Concentrations and Diversity of Microbes from Four Local Bioaerosol Emission Sources in Finland. *Journal* of the Air & Waste Management Association, 61 ((12): 1382-1392. DOI: 10.1080/10473289.2011.628902
- Kesavan, J. and Sagripanti, J.L. (2015). Evaluation criteria for bioaerosol samplers. *Environ Sci Process Impacts* 17(3):638-645.

- Kesavan, J.S., Schepers, D. and Bottiger, J. (2011). Characteristics of twenty-nine aerosol samplers tested at U.S. Army Edgewood Chemical Biological Center (2000 - 2006). Aberdeen Proving Ground, MD: U.S. Army, http://www.dtic.mil/dtic/tr/fulltext/u2/a540009.pdf.
- Kidd, S., Halliday, C., Alexiou, H. and Ellis, D. (2016). Descriptions of medical fungi (3rd Ed.). National Mycology Reference Centre, Microbiology & Infectious Diseases.
- Krieg, N. R. (2001). Identification of procaryotes. In: Bergey's Manual of Systematic Bacteriology, 2nd Ed., Volume 1. pp. 33-42.
- Kulkarni, P., Baron, P.A. and Willeke, K. (2011). Aerosol Measurement: Principles, Techniques, and Applications. Hoboken, NJ: John Wiley & Sons.
- Leland, D.S. and Ginocchio, C.C. (2007). Role of cell culture for virus detection in the age of technology. *Clinical Microbiology Review*, 20(1):49-78.
- Lighthart, B. (1997). The ecology of bacteria in the alfresco atmosphere. *Fems Microbiol. Ecol.*, 23 (4):263–274. doi:10.1016/S0168-6496(97)00036-6
- Lin, X.J., Reponen T, Willeke, K., Wang, Z., Grinshpun, S.A., and Trunov, M. (2000). Survival of airborne microorganisms during swirling aerosol collection. *Aerosol Sci. Technol.*, 32(3):184-196.
- Lindsley, W.G., Green, B.J., Blachere, F.M., Martin, S.B., Law, B.F., Paul A. ... and Schafer, M.P. (2017). Sampling and characterization of bioaerosols. In: *NIOSH Manual of Analytical Methods* (5th ed.). pp. BA-2 of BA-115.
- Löndahl, J. (2014). Physical and Biological Properties of Bioaerosols. In: Jonsson P., Olofsson G., Tjärnhage T. (eds.) *Bioaerosol Detection Technologies. Integrated Analytical Systems.*, New York, NY: Springer.
- Mannina ,G., Morici, C. Cosenza, A., Di Trapani, D. and Ødegaard, H. (2016). Greenhouse gases from sequential batch membrane bioreactors: a pilot plant case study. *Journal of Biochemical Engineering*, 112:114–122.
- Marple, V.A. and Olson, B.A. (2011). Sampling and measurement using inertial, gravitational, centrifugal, and thermal techniques. In: Kulkarni P, Baron P.A., Willeke K., eds. Aerosol measurement: principles, techniques, and applications. Hoboken, NJ: John Wiley & Sons.
- Marple, V.A.and Willeke, K. (1976). Inertial impactors: theory, design and use. In: Liu, B.Y.H., ed. *Fine particles: aerosol generation, measurement, sampling and analysis*. New York: Academic Press.
- MMWR. (1993). Coccidioidomycosis—United States, 1991–1992. Morbid Mortal Weekly Report, 42 (2): 21–4
- Morey, P.R. (2007). *Microbiological sampling strategies in indoor environments. In: Sampling and analysis of indoor microorganisms.* Hoboken, NJ: John Wiley & Sons, Inc.
- Mori, Y. and Notomi, T. (2009). Loop-mediated isothermal amplification (LAMP): a rapid, accurate, and cost-effective diagnostic method for infectious diseases. *J. Infect. Chemothe*, 15, 62–69.
- National Academies Press (2017). A Research Agenda for Indoor Microbiology, Human Health, and Buildings. Washington (DC): National Academies Press (US);
- NCDC (2020). Guideline on self-isolation. National Centre for Disease Control. https://covid19.ncdc.gov.ng/media/files/SelfIsolationGuideJune2020.pdf

- NIOSH. (1997) Histoplasmosis: protecting workers at risk. DHHS (NIOSH) Publication no. 97–146. Cincinnati, OH: NIOSH.
- Nrior, R.R. (2020). Advanced techniques in microbiology: categories of major microbial characteristics used for its identification. Monogram. Department of Microbiology, Rivers State University, Port Harcourt, Nigeria. pp. 1 -20.
- Nrior, R.R. and Chioma, I.C. (2017). Black Soot in Port Harcourt: Incidence of pathogenic microbes associated with black soot in indoor aerosols of classrooms in Port Harcourt Nigeria. *IOSR Journal of Environmental Science, Toxicology and Food Technology*, 11 (12): 43-49. www.iosrjournals.org.
- Nrior, R.R. & Dumbor, K.P. (2019). Incidence of Alarming Microbial Pathogens over Refuse Dump Sites and Its 'Harmful Distance' in Port Harcourt, Southern Nigeria. *Journal of Scientific Research & Reports*, 23(3): 1-11.
- Obire O., Nwaubeta, O. and Adue, S.B.N. (2002). Microbial Community of a Waste-Dump Site. *Journal of Applied Science and Environmental Management*, 6(1):78-83.
- Pastoris, M.C., Ciceroni, L., Monaco, R.L., Goldoni, P., Mentore, B., Flego, G., ...and Visca, P. (1997). Molecular epidemiology of an outbreak of Legionnaires' disease associated with a cooling tower in Genova-Sestri Ponente, Italy. *European Journal of Clinical Microbiology and Infectious Diseases*, 16: 883-892. https://doi.org/10.1007/BF01700554
- Pirtle, E. C., and G. W. Beran. (1991). Virus survival in the environment. *Rev. Sci. Technol*.10:733-748.
- Poletti. L., Pasquarella, C., Pitzurra, M. and Savino, A. (1999). Comparative efficiency of nitrocellulose membranes versus RODAC plates in microbial sampling on surfaces. J. Hosp. Infect., 41(3):195-201.
- Raynor, P.C., Leith, D., Lee, K.W. and Mukund, R. (2011). Sampling and analysis using filters.
 In: Kulkarni P, Baron PA, Willeke K, eds. *Aerosol measurement: Principles, techniques, and applications*. Hoboken, NJ: John Wiley & Sons.
- Reed, L.J. and Muench, H. (1938). A simple method of estimating fifty per cent endpoints. *A. J. Hyg.*, 27:493-497.
- Schäfer, J, Weiß, S. and Jäckel, U. (2017). Preliminary Validation of a Method Combining Cultivation and Cloning-Based Approaches to Monitor Airborne Bacteria. Ann. Work Expo. Health; 61: 633–42.
- Scheermeyer, E, and Agranovski, I.E. (2009). Design and evaluation of a new device for fungal spore aerosolization for laboratory applications. *Journal of Aerosol Science*, 40:879–889.
- Seidl, M., Da, G., Ausset, P., Haenn, S., Géhin, E. and Moulin, L. (2016). Evaluating exposure of pedestrians to airborne contaminants associated with non-potable water use for pavement cleaning. *Journal of Environmental Science Pollution Research International*, 23:6091–6101.
- Shears, P. and O'Dempsey, T.J. (2015). Ebola virus disease in Africa: epidemiology and nosocomial transmission. *Journal of Hospital Infection*,90(1):1–9.
- Shin, S.K., Kim, J., Ha, S.M., Oh, H. S., Chun, J. and Sohn, J. (2015). Metagenomic Insights into the Bioaerosols in the Indoor and Outdoor Environments of Childcare Facilities. *PLoS ONE* 10(5): e0126960. https://doi.org/10.1371/journal.pone.0126960

- Sigari, G., Panatto, D., Lai, P., *et al.* (2006). Virological investigation on aerosol from waste depuration plants. *J. Rev. Med. Hyg.*, 47, 4–7.
- Sivagnanasundaram, P., Amarasekara, R.W. K., Madegedara, R.M.D., Ekanayake, A., and Magana-Arachchi, D.N. (2019). Assessment of Airborne Bacterial and Fungal Communities in Selected Areas of Teaching Hospital, Kandy, Sri Lanka. *Journal of <u>BioMed Research International</u>, vol. 2019: (7393926): 11 pages, 2019 doi: https://doi.org/10.1155/2019/7393926*
- Srikanth, P., Sudharsanam, S., and Steinberg, R. (2008). Bio-aerosols in indoor environment: composition, health effects and analysis. *Indian Journal of Medical Microbiology*, 26(4): 302-12.
- Tortora, G.J., Funke, B.R., and Case, C.L. (2013). *Microbiology An Introduction*. Glenview, IL: Pearson Education.
- Turjanmaa K. (1987) Incidence of immediate allergy to latex gloves in hospital personnel. *Contact Dermatitis*; 17: 270–5.
- Van den Ende, J., Lynen L., Elsen, P. *et al.* (1998). A cluster of airport malaria in Belgium in 1995. *Acta. Clin. Belg.*, 53: 259–63.
- WHO (2020). Middle East respiratory syndrome coronavirus (MERS-CoV). <u>https://www.who.int/news-room/fact-sheets/detail/middle-east-respiratory-syndrome-coronavirus-(mers-cov).</u>
- Zhen, H., Han, T., Fennell, D.E., and Mainelis, G. (2014). Release of free DNA by membraneimpaired bacterial aerosols due to aerosolization and air sampling. Journal of *Applied Environmental Microbiology*, 79:7780–7789.