# Editorial Board

**Global Journal of Medical Research**

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Pharmaceutical Drugs and the Human Energy System (Biofield)

By Seema Bhattessa

Abstract- What is Human Life-Force Energy and What Type of Proof Do We Have?: The concept of subtle human energy fields, or life-force energy, has been recognised and woven into traditional healing systems for millennia. Traditional Chinese Medicine (TCM) describes an intricate network of energy meridians through which this energy, known as “chi” circulates. And, in the traditional Indian system of Ayurveda, the human energy field takes the form of energy vortexes called “chakras”, through which energy, known as “prana” travels.

Accumulating evidence for the existence of these and other subtle, spatially-oriented and biologically-generated, human energy fields has been demonstrated through objective testing methods. As a result, in 1992, the term “biofield” emerged to describe this energy. Biofield Energy is defined as “a massless field, not necessarily electromagnetic, that surrounds and permeates living bodies and affects the body.”[1]

Keywords: biofield; biofield science; biofield research; drugs; pharmaceutical drugs; prescription medicine; human energy system; energy flow; manipulation of the biofield.

GJMR-B Classification: NLMC Code: QV 55

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Pharmaceutical Drugs and the Human Energy System (Biofield)

Seema Bhattessaa

Abstract- What is Human Life-Force Energy and What Type of Proof Do We Have?: The concept of subtle human energy fields, or life-force energy, has been recognised and woven into traditional healing systems for millennia. Traditional Chinese Medicine (TCM) describes an intricate network of energy meridians through which this energy, known as “chi” circulates. And, in the traditional Indian system of Ayurveda, the human energy field takes the form of energy vortexes called “chakras”, through which energy, known as “prana” travels.

Accumulating evidence for the existence of these and other subtle, spatially-oriented and biologically-generated, human energy fields has been demonstrated through objective testing methods. As a result, in 1992, the term “biofield” emerged to describe this energy. Biofield Energy is defined as “a massless field, not necessarily electromagnetic, that surrounds and permeates living bodies and affects the body.”[1]

Biofields are thought to function as a communication conduit that formats and relays information between physiological systems and enables near-instantaneous response times.[2] This inclusive approach affirms core concepts within traditional healing systems, while inviting a more expansive model of living systems than is currently offered by biological science.[3] Through the biofield concept, biological systems are integrated both internally and within their environments across time and space. These energetic relationships increase in scale from subatomic and organismic, to interpersonal and cosmic levels.[2] Potential evidence of the latter can be found in the correlation between solar storms and increased rates and severity of myocardial infarctions.[1,4]

Several distinct biofields have been identified, some of which have been exploited for diagnostic and therapeutic uses, and more are being posited. A few of these include:

a) Electromagnetic Fields

One of only 4 forces in nature – along with gravity, and strong and weak nuclear forces, electromagnetism is the most important natural force at work in living systems. [1] Electromagnetic fields (EMFs) generate electrocardiograms and electroencephalograms that have been a standard of medical diagnostics for decades. More recently, biofield investigations show that subtle magnetic fields generated by the heart radiate outwards from the body. This energy is transmissible to nearby individuals, producing a synchronisation of EEG waves between the source and recipient.[1,2]

b) Biophotons

Also known as ultraweak photon emissions (UPE), cellular glow, or ultraweak bioluminescence, this biofield energy can be detected both in cell cultures and on the body surface via photomultipliers.[4]

Biophoton generation patterns have been associated with fluctuations in cerebral blood flow and

Keywords: biofield; biofield science; biofield research; drugs; pharmaceutical drugs; prescription medicine; human energy system; energy flow; manipulation of the biofield.

I. What is Human Life-force Energy and What Type of Proof do we have?

The concept of subtle human energy fields, or life-force energy, has been recognised and woven into traditional healing systems for millennia. Traditional Chinese Medicine (TCM) describes an intricate network of energy meridians through which this energy, known as “chi” circulates. And, in the traditional Indian system of Ayurveda, the human energy field takes the form of energy vortexes called “chakras”, through which energy, known as “prana” travels.
metabolism. High levels of UPEs have been measured from reactive oxygen species indicating that biophotons may prove to be a sensitive marker for metabolic stress.

c) Cellular Membrane Potentials and Charged Particles

Taken together, the combined effects of transmembrane potentials with the oscillatory frequencies of molecules, and charged particles across an entire organism, create what has been described as a ‘bioelectric scaffold’ through which stem cell development, tissue repair, organ regeneration, and homeostatic processes are facilitated and coordinated.

d) Sub-EMF Level Electron Flux Fields

The possibility of a sub-electromagnetic energy field has been raised via experimental evidence, which may support the phantom limb phenomenon reported by amputees. The evidence stems from energy imaging in plants, which confirms that when leaves are cut the energy of the cut portion is retained as a precise and accurate phantom. Moreover, leaf phantoms conduct electrical current and interact with electromagnetic fields.

e) Biofield Receptor Systems

Where there is an effector there is a receptor, and the search for biofield receptor systems to date, has produced evidence for three such categories:

1. Molecular-level
   - DNA transcription is modulated by low-frequency EMFs.

2. Charge flux sites
   - Voltage-gated calcium channels are affected by the presence of low-frequency EMFs.

3. Endogenous EMFs
   - Gap junctions constitute a bioelectric intracellular communication system that guides tissue development and maintenance.

Additionally, a fourth proposed concept for a biofield receptor system integrates all of the above, and revolves around the fascial network of collagen fibres and water molecules, that surrounds organs and tissues. Collagen fibres are capable of conducting and modifying photon pulses and provide a form of surveillance, alongside the immune and nervous systems. Water molecules contribute to this receptor system via local synchronisation of electron spin, causing the collagen matrix in that area to act as a frequency-specific electromagnetic receptor system.

f) Clinical Evidence of Biofields

In vitro and in vivo published studies dating back to the 1960s demonstrated the ability of biofield energy to facilitate wound healing and inhibit cancer cell viability. More recently, an abundance of clinical data provides illuminating (and increasingly irrefutable) evidence for the existence of human biofield energy, such as the following reports:

- An intriguing 2015 study reported that sceptical volunteers given a “crash course” in energy healing cured laboratory mice injected with lethal doses of cancer cells.
- In a 2016 experiment, Korean scientists produced demonstrable evidence for the existence of acupuncture points. In that study, dye injected into acupuncture points formed visible lines, while the same effect was not observed at non-acupuncture points.
- A newly developed biofield device, which delivers direct current through a saline water immersion foot bath, produced improvements in red blood cell morphology and CO2 transport in healthy participants in a 2018 study.
- To date, over 15 clinical trials have been conducted on various biofield therapies for cancer patients, with reports of reduced pain and fatigue and improvements in biomarkers.

- Intrinsic and Extrinsic Factors that Affect the Human Energy System

The energy values of substances both intrinsic and extrinsic to the human body have varying effects on biofield energy. These include frequencies we generate ourselves, such as our thoughts and emotions, energy from substances we intentionally bring into our own personal biofields, and natural and manmade energy fields in our environment that affect us energetically in a multitude of ways.

Basic nutritional elements have been shown to shift the present energy within the body as they are absorbed and assimilated. Electrical conductance on the skin surface at acupuncture points has been shown to shift in response to glucose ingestion, revealing a fluctuating pattern of energy shifting through the meridians, as glucose is ingested and assimilated. Thoughts, i.e. the electrical activity of cognitive function, affect the central and autonomic nervous systems, contributing to the unique electromagnetic field of an individual, or aura. The aura radiates outwards, carrying with it information about physiological, mental, and emotional status of the individual.

h) Vibratory Signatures and their Effects on the Biofield

All living and non-living things have a unique vibratory rate that can be measured and quantified in Hertz (Hz). Living beings have higher rates of vibration while non living objects have little to no vibratory rate. The foods we consume contribute their own inherent rates of vibration to our own, raising or lowering the overall rate accordingly. In general, highly
processed, pesticide-laden foods have relatively lower vibratory rates, whilst fresh, organic plant foods, and fermented and raw sprouted foods offer the highest rates.\cite{14,16}

Similarly, light energy in the visible spectrum is thought to exert a range of effects based on the vibratory rate of each colour. In Ayurveda, colours within the lower frequency ranges correlate to lower chakras, and influence functions such as survival and sexuality, while higher frequency colours correlate to higher functions: communication, love, intuition, and spirituality.\cite{16}

\textbf{i) Energetic Manipulation of Biofields}
Numerous therapeutic systems and medical practices have been found to have a beneficial influence on human biofields, while other forms of energy have notably detrimental or even mixed effects.

\textit{Examples include:}
\begin{itemize}
\item **Aromatherapy**
Plant essential oils have among the highest recorded vibrational rates. Oils at the lower end of the range are used to heal physical illnesses, those in the middle of the range support emotional health, and those in the upper-frequency ranges are thought to promote spiritual growth.\cite{17}

\item **Grounding**
This is a practice that uses the Earth’s strong negative charge to supply electrons to the body. Grounding is practised by placing one’s bare feet on the ground, and is thought to neutralise free radicals, in essence, acting as an antioxidant.\cite{18} Published research has reported improvements in sleep, pain, stress response, immune function, and blood sugar regulation.\cite{6}

\item **Pulsed EMF**
Embryonic development, tissue regeneration, and other processes requiring biological organisation are thought to occur through energy field phenomena. Pulsed EMF therapy has been used to regenerate limbs in salamanders, and is being investigated as a potential treatment for healing bone fractures.\cite{2}

\item **Wireless Technology**
Counter to prevailing public health doctrine, detrimental non-thermal health effects of nonionising EMFs from cell phones and other wireless technology have been well-documented in the literature.\cite{1,2} Studies show neurodevelopmental, neurodegenerative, cognitive, behavioural, endocrine, immune, and genotoxic effects.\cite{19} The pervasive use of wireless devices, and the imminent deployment of 5G, raise concerns about the effects of our increasingly electromagnetic environment on the health of humans and the environment as a whole.\cite{20}

\item **Prescription Medicines**
Pharmaceutical drugs interact in various ways with biofields, some beneficial and others harmful.
\begin{itemize}
\item On the potentially beneficial side, certain drugs have been found to act synergistically with bio-magnetic fields, increasing response to drugs by several-fold.\cite{20} Drug delivery systems are being developed around this effect using magneto-electric nanoparticles designed to manipulate membrane electric fields to improve absorption- a process known as electroporation.\cite{21} Currently, however; though highly effective, its use is limited by the inability of cells to reverse the process and reseal the cell membrane.\cite{22}

\item Pharmaceuticals may also exert a range of adverse mental and emotional effects through their interactions with the biofield. Empirical reports exist in which patients describe shifts of consciousness from taking certain prescription medications; in some instances, a fragmented, disconnected sense of self and in others an ecstatic sense of unity and wholeness, either scenario being an unsettling departure from reality. Attempts to recreate such sensations have pointed toward over-inhibition or over-activation of the anterior insula which serves an important role in maintaining an individual’s sense of self.\cite{23}
\end{itemize}

\textit{j) My Professional Experience and a Call to Action}
In my professional experience as a pharmacist and holistic energy therapist, I have observed, at times, the biofield effects of pharmaceutical drugs in my patients. This has occurred most notably in patients on hormone replacement, or other endocrine therapies, in the form of a certain sluggishness in chakra energy. In these instances, as well as where the source stems from other types of physical, mental, or emotional trauma, working with the client’s energy field often helps improve the flow of stagnant energy, heal auric tears and remove any blockages that may have developed as a result of health issues, stress and anxiety, depression and other life experiences.

Restoring the balance and the flow of the energy system is a subtle healing process that is not always obvious, but often recognised by others, and expressed in the way that they respond towards you. I have also used energy therapy to help strengthen a client’s vulnerability to adverse situations, or relations with others, in the workplace and at home. In this way, the energy system affects, and is affected by, our behaviour. Biofield therapeutics can act as an early intervention for behavioural health, a self-help tool, and a social investment.

Biofield science is growing rapidly, particularly in the fields of psychiatric and neurodegenerative disorders. However, funding usually favours industry applications, and is lacking in basic science and clinical
applications research. Amongst the most urgent areas requiring further study, researchers have identified the lack of information on the effects of dose, delivery mode, and type of therapy on clinical outcomes. With this brief synopsis on biofield science, I invite my colleagues within the medical, health, and energy-healing spheres to further investigate these important topics. I am seeking to collaborate directly with interested professionals, and to conduct and publish studies that will contribute to the growing body of knowledge about the human energy field.

Conflict of Interest

The author(s) declare that they have no conflict of interest.

References Références Referencias

Risk Factors for Oropharyngeal Colonization with Multidrug-Resistant Bacteria in a Brazilian Hospital

By Dayane Otero Rodrigues & Deyse Silva Câmara

Universidade Federal do Oeste da Bahia

Abstract- The objective of this study was to describe the risk factors and etiology (pathogen species and their antimicrobial susceptibility, and identifying of multidrug-resistant microorganisms-MDR) for the oropharyngeal colonization in a Brazilian hospital. A total of 39 patients were analysed, and presented media age of the 57,7 years and media of the duration of hospitalization the 9,2 days. Streptococcus sp. (39,1%) and Staphylococcus aureus (18,9%) were the mains pathogens of clinical significance detected. This study showed high rates of isolated MDR bacteria, which included methicillin-resistant Staphylococcus aureus (MRSA) (71,4%). The use of the antibiotics, the advanced age and the previous hospitalization were the significant risk factors for the oropharyngeal colonization with MDR bacteria in the statistical analysis. These results reinforce the need for a revised protocol for regulation of antibiotic dispensing, and attention for this population profile, that can develop healthcare-associated infections (HAI) from oropharyngeal colonization with MDR bacteria.

Keywords: risk factors, pathogens, multidrug-resistant, healthcare-associated infections (HAI).

GJMR-B Classification: NLMC Code: QV 37.5
Risk Factors for Oropharyngeal Colonization with Multidrug-Resistant Bacteria in a Brazilian Hospital

Oropharyngeal Colonization with Multidrug-Resistant

Dayane Otero Rodrigues a & Deyse Silva Câmara a

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I. Introduction

A resistência bacteriana é um problema que preocupa o mundo inteiro, diante da velocidade com que avança e da ameaça que representa para a medicina moderna, causando restrição das opções terapêuticas para as infecções causadas por patógenos multirresistentes (MDR) [1,2].

No ambiente hospitalar esses microorganismos MDR são mais frequentes. Esse predomínio é decorrente da combinação de múltiplos fatores, com destaque para o uso indiscriminado de antibióticos, a condição clínica dos pacientes, a realização de procedimentos invasivos, a presença de doenças de base predisponentes à imunossupressão, como diabetes melitus, neoplasias, doença pulmonar obstructiva crônica, dentre outras [3,4].

Ademais, a presença no ambiente hospitalar de patógenos MDR pode resultar em infecções de difícil controle, que requerem a utilização de regimes de antibióticos de espectro maiores [5]. Neste contexto, destacam-se as Infeções Relacionadas à Assistência à Saúde (IRAS) conhecidas popularmente por Infeções Hospitalares, e que estão associadas a elevados custos no tratamento e ao aumento das taxas de morbimortalidade entre os pacientes [5].

Tratando-se da microbiota da orofaringe, sítio alvo desta pesquisa, sabe-se que essa pode ser colonizada transitoriamente, em indivíduos saudáveis, por patógenos como Streptococcus pneumoniae, Streptococcus pyogenes, Haemophilus influenzae, Neisseria meningitidis, Enterobacterias e Staphylococcus sp., esse último é mais presente na microbiota nasal [6]. Também é comum a presença de methicillin-resistant Staphylococcus aureus (MRSA) colonizando o trato respiratório de pacientes hospitalizados, sendo um dos mais frequentes causadores de pneumonias associadas à ventilação mecânica (PAV), já relatado como preditor de reincidente e mortalidade nos casos de pacientes internados em UTI [7].

A aspiração de patógenos da orofaringe é a via primária pela qual as bactérias podem alcançar os pulmões, causando pneumonia [8], caracterizando a colonização da mucosa da orofaringe por bactérias MDR como fator de risco importante para o desenvolvimento de pneumonia hospitalar. Dados científicos demonstraram essa associação entre a colonização da orofaringe e o desenvolvimento de PAV por S. aureus, sendo que dos 346 pacientes, 36,4% apresentaram a orofaringe colonizada, e destes, 63,5% por MRSA, com risco de pneumonia para todos os casos [9].

A pneumonia é a infecção mais frequente entre as IRAS. As taxas de incidência variam de 17 a 40% nos pacientes em UTIs, sendo a maioria associada à ventilação mecânica, ocorrendo em 9% a 24% dos pacientes intubados por mais de 48 horas. Além de estar associada ao aumento da morbimortalidade entre os pacientes de hospitalização prolongada [10].

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Apesar dos casos de infecções por microrganismos MDR serem documentadas com maior frequência em unidades de cuidados intensivos, todas os cenários de práticas de saúde podem ser afetados pelo surgimento e transmissão de microrganismos MDR [11]. Logo é justificável o desenvolvimento desta pesquisa, descrevendo os fatores de risco e a etiologia (espécies patogênicas e sua suscetibilidade antimicrobiana e identificação de microrganismos MDR) para a colonização orofaríngea por patógenos potencialmente causadores de IRAS em um hospital brasileiro.

II. Materials and Methods

Foi realizado um estudo prospectivo que investigou a incidência de bactérias MDR isoladas da mucosa da orofaringe de pacientes clínicos e cirúrgicos internados na enfermaria de adultos do Hospital Municipal Eurico Dutra (HMED) no município de Barreiras, Bahia, no período de jun/2018 e set/2018.

O HMED é um hospital geral de média complexidade, que presta serviços como de apoio diagnóstico e terapêutico, atendimento emergencial e ambulatorial. Os pacientes são admitidos por demanda espontânea e referida da cidade de Barreiras e região. O hospital possui um total de 10 leitos na enfermaria médica e 29 leitos na enfermaria cirúrgica, com lotação de 70% destes, aproximadamente; sendo que os pacientes permanecem internados em torno de uma semana.

A coleta dos dados demográficos e fatores de risco intrínsecos e extrínsecos foi realizada através da análise dos prontuários médicos e entrevista aos pacientes.

O estudo foi autorizado pelo Comitê de Ética em Pesquisa com Seres Humanos (CAEE: 84427518.5.0000.8060)

a) Coleta das amostras clínicas

As amostras da orofaringe foram coletadas semanalmente, no turno da manhã e de forma asséptica, por meio de fricção de swabs estéreis na porção da faringe atrás da cavidade oral, seguindo-se ao seu acondicionamento em tubos estéreis com Brain Heart Infusion (BHI, Oxoid, Basingstoke, Hampshire, England) e transporte ao laboratório de Microbiologia da UFOB.

b) Procedimentos laboratoriais

As amostras foram incubadas em estufa à 37 °C por 24 h, e sequencialmente subcultivadas na superfície do Mannitol Salt Agar, MacConkey, e Pseudomonas para isolar S. aureus, coagulase-negative staphylococci (CoNS), e Gram-negative bacilos, seguindo-se à realização de testes microbiológicos clássicos [5]:

- S. aureus and CoNS: fermentation of Mannitol Salt Agar, Gram staining, catalase test, coagulase test, DNase Agar;
- Enterobacteriaceae family: growth on MacConkey agar, cytochrome oxidase, Gram staining, lactose fermentation (Triple Sugar Iron Agar - TSI), biochemical tests (B Enterokit Probac of Brazil);
- Non-fermenter Bacilli (Pseudomonas aeruginosa): growth on Pseudomonas agar, odor, colony morphology, cytochrome oxidase, growth at 42°C, Gram staining, oxidation in Hugh Leifson medium;

The bacterial strains were then tested for their antimicrobial susceptibility in vitro by the agar diffusion technique as recommended by the Clinical and Laboratory Standards Institute [12]. Bacterial strains were considered as multidrug-resistant (MDR) if showing resistance to at least three classes of antimicrobials [1,13] and associated phenotypes. Some examples included MRSA, Enterobacteria resistant to 4th generation cephalosporins and quinolone-resistant P. aeruginosa, according to definitions of the European Center for Disease Prevention and Control [1].

Os dados epidemiológicos, microbiológicos, e os fatores de risco foram analisados através do programa Stata 14 utilizando- se os testes de exato de Fisher, Qui-quadrado e "t" de Student, e pelo modelo da regressão logística na análise multivariada.

III. Results

Durante o período da pesquisa foram coletadas amostras da orofaringe de 39 pacientes, com um total de 74 microrganismos isolados e identificados.

Em relação à etiologia, os microrganismos mais frequentes isolados das amostras clínicas foram Streptococcus spp. (39,1%), e S. aureus (18,9%), seguidos pela Família Enterobacteriaceae (17,5%) (Tabela 1).
Tabela 1: Frequência de microrganismos identificados na microbiota da orofaringe de pacientes internados na enfermaria do Hospital Municipal Eurico Dutra em Barreiras-BA.

<table>
<thead>
<tr>
<th>Tipo de Microrganismo</th>
<th>Frequência (N)</th>
<th>(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cocos Gram Positivos</td>
<td>47</td>
<td>63,5</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>14</td>
<td>18,9</td>
</tr>
<tr>
<td>Staphylococcus coagulase negativo</td>
<td>2</td>
<td>2,7</td>
</tr>
<tr>
<td>Streptococcus sp</td>
<td>29</td>
<td>39,1</td>
</tr>
<tr>
<td>Enterococcus sp.</td>
<td>2</td>
<td>2,7</td>
</tr>
<tr>
<td>Bacilos Gram-negativos</td>
<td>17</td>
<td>22,9</td>
</tr>
<tr>
<td>Família Enterobacteriaceae</td>
<td>13</td>
<td>17,5</td>
</tr>
<tr>
<td>Pseudomonas sp.</td>
<td>4</td>
<td>5,4</td>
</tr>
<tr>
<td>Bacilos Gram-positivos</td>
<td>3</td>
<td>4,0</td>
</tr>
<tr>
<td>Fungos</td>
<td>7</td>
<td>9,4</td>
</tr>
<tr>
<td>Candida sp.</td>
<td>7</td>
<td>9,4</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>74</strong></td>
<td><strong>100,0%</strong></td>
</tr>
</tbody>
</table>

Os resultados do perfil de resistência aos antimicrobianos das 50 amostras bacterianas testadas, considerando as perdas de amostras, estão apresentados na Tabela 2. As amostras de S. aureus apresentaram as maiores taxas de resistência, 57,1% das cepas foram resistentes à claritromicina, 50% à rifampicina, 42,8% à sulfonamidas e 35,7% à ciprofloxacina, tetraciclina e gentamicina, com destaque para a taxa de MRSA, cepas resistentes a todos os antibióticos do grupo dos beta-lactâmicos (71,4%). Altas taxas de resistência na Família Enterobacteriaceae também foram encontradas, com 81,8% de resistência à cefoxitina, e 75% de resistência à cefoxitina dentre as amostras de Pseudomonas sp. (Tabela 2).

Tabela 2: Perfil de resistência aos antimicrobianos dos microrganismos da microbiota da orofaringe dos pacientes internados na enfermaria do Hospital Municipal Eurico Dutra em Barreiras-BA.

<table>
<thead>
<tr>
<th>Antimicrobianos</th>
<th>S. aureus N (%)</th>
<th>SCN N (%)</th>
<th>Streptococcus N (%)</th>
<th>Família Enterobacteriaceae N (%)</th>
<th>Pseudomonas N (%)</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>OXA</td>
<td>10 (71,4)</td>
<td>1 (50,0)</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>14 (28,0)</td>
</tr>
<tr>
<td>CIP</td>
<td>5 (35,7)</td>
<td>1 (50,0)</td>
<td>NT</td>
<td>3 (27,2)</td>
<td>1 (25,0)</td>
<td>10 (20,0)</td>
</tr>
<tr>
<td>CFO</td>
<td>10 (71,4)</td>
<td>1 (50,0)</td>
<td>NT</td>
<td>9 (81,8)</td>
<td>3 (75,0)</td>
<td>25 (50,0)</td>
</tr>
<tr>
<td>SULFA</td>
<td>7 (42,8)</td>
<td>1 (50,0)</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>8 (16,0)</td>
</tr>
<tr>
<td>TET</td>
<td>5 (35,7)</td>
<td>2 (100)</td>
<td>9 (47,3)</td>
<td>NT</td>
<td>1 (25,0)</td>
<td>17 (34,0)</td>
</tr>
<tr>
<td>RIF</td>
<td>7 (50,0)</td>
<td>0</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>7 (14,0)</td>
</tr>
<tr>
<td>GEN</td>
<td>5 (35,7)</td>
<td>1 (50,0)</td>
<td>NT</td>
<td>3 (27,2)</td>
<td>0</td>
<td>9 (18,0)</td>
</tr>
<tr>
<td>CLA</td>
<td>8 (57,1)</td>
<td>1 (50,0)</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>9 (18,0)</td>
</tr>
<tr>
<td>AMP</td>
<td>10 (71,4)</td>
<td>1 (50,0)</td>
<td>1 (5,2)</td>
<td>4 (36,3)</td>
<td>3 (75,0)</td>
<td>22 (44,0)</td>
</tr>
<tr>
<td>VAN</td>
<td>NT</td>
<td>NT</td>
<td>2 (10,5)</td>
<td>NT</td>
<td>NT</td>
<td>2 (4,0)</td>
</tr>
<tr>
<td>CPM</td>
<td>NT</td>
<td>NT</td>
<td>8 (42,1)</td>
<td>7 (63,6)</td>
<td>1 (25,0)</td>
<td>16 (32,0)</td>
</tr>
<tr>
<td>ATM</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>8 (72,7)</td>
<td>1 (25,0)</td>
<td>9 (18,0)</td>
</tr>
<tr>
<td>POL</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>AZI</td>
<td>NT</td>
<td>NT</td>
<td>9 (47,3)</td>
<td>NT</td>
<td>NT</td>
<td>9 (18,0)</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>14 (100,0)</strong></td>
<td><strong>2 (100,0)</strong></td>
<td><strong>19 (100,0)</strong></td>
<td><strong>11 (100,0)</strong></td>
<td><strong>4 (100,0)</strong></td>
<td><strong>50 (100,0)</strong></td>
</tr>
</tbody>
</table>

SCN: Staphylococcus coagulase negativo; OXA: Oxacilina; CIP: Ciprofloxacino; CFO: Cefoxitina; SULFA: Sulfonamidas; TET: Tetraciclina; RIF: Rifampicina; GEN: Gentamicina; CLA: Claritromicina; AMP: Ampicilina; VAN: Vancomicina; CPM: Cefepime; ATM: Aztreonam; POL: Polimixina B; AZI: Azitromicina; NT: Não testado.

Fonte: dados do pesquisador.
Entre os pacientes incluídos na pesquisa, 60% estavam colonizados por patógenos MDR. As frequências das características e fatores de risco dos pacientes para a colonização por bactérias MDR estão apresentadas na Tabela 3. Após a realização da análise estatística dos dados, o uso atual de antibióticos, idade >50 anos, uso prévio de antibióticos, história de hospitalização prévia, estado geral, e internação > 6 dias foram considerados fatores de risco potenciais para aquisição de bactérias MDR e foram analisadas no modelo de regressão logística.

Tabela 3: Associação entre as características demográficas e fatores de risco dos pacientes internados na enfermaria do Hospital Municipal Eurico Dutra em Barreiras-BA e a presença de bactérias multirresistentes.

<table>
<thead>
<tr>
<th>Características e fatores de risco dos pacientes</th>
<th>Pacientes (n=30)</th>
<th>p valor</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Com multirresistência (n=18) N (%)</td>
<td>Sem multirresistência (n=12) N (%)</td>
</tr>
<tr>
<td>Idade (media)</td>
<td>61,7 ± 20,5</td>
<td>51,7 ± 17,7</td>
</tr>
<tr>
<td>Sexo</td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>6 (33,3)</td>
<td>3 (25,0)</td>
</tr>
<tr>
<td>M</td>
<td>12 (66,7)</td>
<td>9 (75,0)</td>
</tr>
<tr>
<td>Estado geral</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bom</td>
<td>13 (72,2)</td>
<td>8 (66,7)</td>
</tr>
<tr>
<td>Ruim</td>
<td>5 (27,8)</td>
<td>4 (33,3)</td>
</tr>
<tr>
<td>Tempo de internação</td>
<td></td>
<td></td>
</tr>
<tr>
<td>≥ 6 d</td>
<td>13 (72,2)</td>
<td>6 (50,0)</td>
</tr>
<tr>
<td>&lt; 6 d</td>
<td>5 (27,8)</td>
<td>6 (50,0)</td>
</tr>
</tbody>
</table>

A análise final da regressão logística mostrou que uma idade superior a 50 anos, o uso atual de antibióticos e história de hospitalização prévia foram fatores de risco estatisticamente significantes para a colonização por bactérias MDR (p≤0,05) (Tabela 4).

Tabela 4: Fatores de risco para colonização por bactérias multirresistentes nos pacientes internados na enfermaria do Hospital Municipal Eurico Dutra em Barreiras-BA após análise de regressão logística.

<table>
<thead>
<tr>
<th>Fator de risco</th>
<th>Odds Ratio</th>
<th>95% CI</th>
<th>p valor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Estado geral</td>
<td>0. 307</td>
<td>0.026-3.559</td>
<td>0.345</td>
</tr>
<tr>
<td>Uso atual de antibióticos</td>
<td>69.022</td>
<td>1.430-3330.746</td>
<td>0.032</td>
</tr>
<tr>
<td>Uso prévio de antibióticos</td>
<td>0. 102</td>
<td>0.006-1.683</td>
<td>0.111</td>
</tr>
<tr>
<td>História de hospitalização prévia</td>
<td>87.299</td>
<td>1.773-4298.123</td>
<td>0.025</td>
</tr>
<tr>
<td>Idade &gt;50 anos</td>
<td>28.767</td>
<td>1.708-484.431</td>
<td>0.020</td>
</tr>
<tr>
<td>Internação &gt;6 dias</td>
<td>0. 994</td>
<td>0.783-1.262</td>
<td>0.966</td>
</tr>
</tbody>
</table>

A frequência de pacientes colonizados por bactérias MDR em uso de dois ou mais antibióticos foi de 61,1%, com diferença significante quando comparada com o grupo de pacientes não-colonizados por bactérias MDR (33,3%), assim como a frequência do uso de cefalosporinas de 3ª geração e de macrolídeos (Tabela 5).
Tabela 5: Associação entre os regimes de antibióticos dos pacientes internados na enfermaria do Hospital Municipal Eurico Dutra em Barreiras-BA e a presença de bactérias multirresistentes

<table>
<thead>
<tr>
<th>Antibiótico em uso</th>
<th>Com multirresistência (n=18) N (%)</th>
<th>Sem multirresistência (n=12) N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Número de antibióticos ≥ 2</td>
<td>11 (61,1)</td>
<td>4 (33,3)</td>
</tr>
<tr>
<td>Cefalosporina de 3ª geração</td>
<td>9 (50,0)</td>
<td>3 (25,0)</td>
</tr>
<tr>
<td>Fluorquinolonas</td>
<td>7 (38,9)</td>
<td>5 (41,6)</td>
</tr>
<tr>
<td>Macrolídeos</td>
<td>6 (33,3)</td>
<td>1 (8,3)</td>
</tr>
<tr>
<td>Outros</td>
<td>7 (38,9)</td>
<td>5 (41,6)</td>
</tr>
</tbody>
</table>

**IV. Discussion**

Diferentes espécies de *Streptococcus* podem colonizar transitoriamente a orofaringe de pacientes saudáveis [6], dessa forma, era esperado que *Streptococcus* sp. fosse encontrado com maior frequência nesta investigação, como demonstraram nossos resultados. Considerando que os participantes do estudo, em sua maioria, possuíam um bom estado geral e apresentavam poucos fatores de gravidade, não se esperava encontrar uma microbiota muito alterada como seria em pacientes críticos que normalmente permanecem internados em UTI[3]. Estudos que verificaram o perfil microbiológico em pacientes de UTI apontam *P. aeruginosa* e *S. aureus* entre os microrganismos mais frequentes[4,14]. Outro estudo que pesquisou a colonização da orofaringe encontrou cepas de Enterobactérias com maior frequência e relacionou este achado à alta proporção de hospitalizações prévias entre os pacientes colonizados [9].

As altas taxas de resistência antimicrobiana encontradas nesta pesquisa entre as amostras de *S. aureus*, Enterobactérias e *Pseudomonas* sp. se assemelhavam aquelas encontradas na literatura, que reportam esses patógenos como os mais relacionados à resistência antimicrobiana em ambiente hospitalar [1,2].

É importante destacar a alta taxa de MRSA encontrada (71,4%) nos pacientes colonizados, nos alerta à necessidade da reflexão acerca da adesão às medidas de prevenção e controle de infecções na unidade, sendo necessário uma revisão nos protocolos de administração antibiótica, tendo em vista que MRSA é um grande responsável por infecções em ambiente hospitalar[3,15]. O relatório do CDC, que traz um resumo de dados referentes aos anos de 2011 a 2014 dos principais patógenos resistentes aos antimicrobianos associados à IRAS em vários tipos de unidades de saúde, apontam que MRSA compõem 42,4% dos isolados de S. aureus em PAVs [11]. Dados do Hospital das Clínicas da Universidade de São Paulo (2009) mostraram que MRSA representou 60% das cepas de *S. aureus* isolados de hemocultura no hospital[15]. Um outro estudo prospectivo (2009-2010) que avaliou a colonização da orofaringe de pacientes de uma UTI em um hospital brasileiro encontrou uma taxa de 63,5% de MRSA, valor um pouco inferior ao encontrado (71,4%) no presente estudo [9].

A colonização por patógenos MDR é um problema de saúde pública, e está associada ao desenvolvimento de infecções mais graves, de difícil controle e com alta mortalidade [16]. Os resultados relatados nesta pesquisa demonstraram que 60% dos pacientes estavam colonizados por bactérias MDR, e observamos que o uso de antibióticos, assim como idade > 50 anos e história de hospitalização prévia são fatores de risco que estão relacionados a um risco maior de colonização por bactérias MDR.

A média de idade dos pacientes colonizados por microrganismos MDR neste estudo foi de 61,7 ± 20,5 e apresentou associação significativa com a colonização por bactérias MDR, corroborando com a literatura, como relatado em outros estudos [9,17]. Uma revisão sistemática da literatura de 74 artigos relaciona o aumento do risco de resistência bacteriana em pacientes com patologias mais graves, presença de comorbidades e pior condição clínica ao fato desses pacientes estarem mais expostos a procedimentos invasivos e ao uso de antibióticos, terem maior tempo de internação, além de possuírem graus variados de imunocomprometimento[17]. Este contexto pode ser estendido ao paciente idoso, uma vez que esse tipo de paciente está mais propenso a apresentar todas essas condições de gravidade.

A hospitalização prévia indica maior risco de colonização por bactérias MDR justamente devido a maior exposição à microbiota hospitalar, um ambiente que apresenta condições que favorecem o surgimento e disseminação da resistência microbiana, como já visto anteriormente [6, 17-18].

Esta pesquisa mostrou que o uso de antibióticos ocorreu em aproximadamente 87% dos pacientes colonizados, sendo administradas preferencialmente as cefalosporinas de 3ª geração e fluorquinolonas, drogas de amplo espectro. Dessa
forma, a alta frequência de prescrições de antibióticos refletiu em taxas mais elevadas de pacientes colonizados por bactérias MDR neste estudo, assim como relatado em outros estudos sobre o assunto [9,17,19]. Um estudo de coorte americano concluiu que a exposição a antibióticos dentro de 3 meses foi o único parâmetro consistentemente associado aos pacientes com Enterobacterias resistentes aos carbapenemases [20]. Um outro estudo americano realizado em UTI destacou o papel das fluorquinolonas no surgimento de P. aeruginosa MDR [21].

Nosso estudo encontrou uma frequência alta de pacientes colonizados por bactérias MDR em uso de dois ou mais antibióticos (61,1%) Vs. 33,3% no grupo não colonizado, semelhante ao encontrado (66,7%) por um outro grupo de pesquisadores brasileiros [22]. O uso imprudente de antimicrobianos contribui para a pressão seletiva de cepas resistentes e é um dos fatores de risco modificáveis mais significantes para o aumento acelerado da MDR [20,23]. Dessa forma, é também uma das questões mais importantes a serem revistas quando se pensa em medidas preventivas. Vale mencionar que a incidência de IRAS é diretamente proporcional ao aumento do uso de antimicrobianos de diversas classes, funcionando como um ciclo vicioso que favorece o processo da resistência bacteriana [22]. O controle de microrganismos MDR envolve muitas discussões não só pela crescente evolução da resistência microbiana, mas também pela escassez de opções terapêuticas, sendo que a criação e implantação de programas de manejo de antimicrobianos é um caminho que deve ser seguido [1,24]. Sendo assim, é de extrema importância que esse tema seja debatido dentro dos hospitais, com as equipes de saúde, almejando melhor adesão às estratégias de prevenção e controle. Nesse sentido, cabe ressaltar a importância de trabalhos como este ao subsidiarem dados para a discussão e enfrentamento do problema a nível local. No caso deste estudo, que evidenciou altas taxas de MDR em uma unidade de média complexidade e em pacientes estáveis, se faz necessário a discussão sobre a adesão às medidas de prevenção e controle para evitar a propagação dessas cepas resistentes na unidade. Da mesma forma, é importante repensar estratégias de controle e fiscalização das antibioticoterapias, elaboração de protocolos próprios, baseados no perfil de sensibilidade aos antimicrobianos no hospital, e a utilização de testes microbiológicos para orientar a melhor escolha, uma vez que as terapias empíricas com antibióticos de largo espectro contribuem para a seleção de microrganismos resistentes [9,19], podendo acelerar a disseminação da MDR.

V. Conclusion

Os resultados encontrados neste estudo refletem o panorama mundial de registros cada vez maiores da incidência de microorganismos MDR. A alta taxa de colonização da mucosa da orofarínge por patógenos MDR, como MRSA, relatada neste estudo, é um fator de risco para a ocorrência de IRAS, e reforçam o alerta para a urgência de ações mais efetivas na prevenção e controle da presença desses patógenos MDR na unidade, como a adoção de uma política mais rigorosa do uso de antibióticos.

É prioritário na unidade a discussão e adequação das medidas de prevenção e controle de infecções, priorizando maior atenção à pacientes em uso de antibióticos, assim como com idade > 50 anos e histórico de hospitalização prévia, que são pacientes com fatores de risco que se apresentaram relacionados à colonização orofaríngea por bactérias MDR, evitando-se assim esse tipo de colonização, que é fator de risco importante para o desenvolvimento de pneumonia hospitalar.

A caracterização do perfil de resistência nos patógenos encontrados em pacientes de uma determinada unidade de saúde é necessária para orientar o uso de antibióticos direcionado à realidade local, devendo ser priorizada em todos os hospitais, tendo em vista que o uso consciente dos antimicrobianos é o passo mais importante para o controle da resistência bacteriana. Sendo assim, reforçamos que trabalhos como este, tem grande contribuição como subsídio à equipe hospitalar e devem ser incentivados, de onde sugerimos que novas pesquisas, com números amostrais maiores, continuem a serem realizadas, a fim de ampliarem os dados já encontrados neste estudo.

References Références Referencias

Risk Factors for Oropharyngeal Colonization with Multidrug-Resistant Bacteria in a Brazilian Hospital


Online Survey on the Source of Information, Knowledge, and Perceptions towards COVID-19 among Health Care Workers and Health Students in Nepal: A Comparative Study

By Sagarananda Giri, Sheela Khadka, Saroj Bashyal, Pratiksha Paudel, Parbati Thapa & Naveen Shrestha

Pokhara University

Abstract- Background: The burgeoning cases of COVID-19 are the major concern and challenges across the world. However, there are different drugs on the row for the clinical trial. Misinformation and misguidance from the unreliable source of information, misunderstanding, lack, or inadequate awareness among people, and poor sanitation procedure could lead to the rapid transmission of infection in the community. The basic objective was to study the knowledge and perception of HCWs and students about COVID-19.

Keywords: COVID-19, knowledge, online survey, perceptions, source of information, health care workers.

GJMR-B Classification: NLMC Code: QV 704

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Online Survey on the Source of Information, Knowledge, and Perceptions towards COVID-19 among Health Care Workers and Health Students in Nepal: A Comparative Study

Sagarananda Giri *, Sheela Khadka †, Saroj Bashyal ‡, Pratiksha Paudel §, Parbati Thapa ¶ & Naveen Shrestha ¥

Abstract: Background: The burgeoning cases of COVID-19 are the major concern and challenges across the world. However, there are different drugs on the row for the clinical trial. Misinformation and misguidance from the unreliable source of information, misunderstanding, lack, or inadequate awareness among people, and poor sanitation procedure could lead to the rapid transmission of infection in the community. The basic objective was to study the knowledge and perception of HCWs and students about COVID-19.

Method: A web-based, cross-sectional survey was conducted among HCWs and students from the medical and paramedical field. The question was divided into participant characteristics, awareness on COVID-19, source of information, knowledge about symptoms of COVID-19, different modes of transmission, precautions, risk prevention, and perceptions of COVID-19 in which the items were evaluated by Likert Scale. The obtained data were analyzed using SPSS version 16. The study was conducted following the Checklist for Reporting Results of Internet E-Surveys (CHERRIES) guidelines.

Results: A total of 501 participants were enrolled in the study in which 350 were HCWs, and the majority of the respondents were the pharmacist (34%), followed by medical officers (34%) and paramedic students (30%). Social media was the most common source of information. A significant proportion of the participants 51.9%, responded correctly about the transmission of COVID-19 and 86% to the onset of symptoms. About, 253 (72.3%) HCWs and 112 (74.2%) students had a good level of knowledge on COVID-19. Only 185 (52.9%) HCWs and 77 (51%) of students showed a positive perception towards COVID-19. However, there was no significant association between HCWs and students regarding the knowledge and perceptions of COVID-19.

Conclusion: Accurate information is the requirement in the current global pandemic of COVID-19 to prevent its spread. Strategies should be adapted for proper and accurate information dissemination as more than half of the participants seem to rely on social media in our study.

Keywords: COVID-19, knowledge, online survey, perceptions, source of information, health care workers.

I. Introduction

Coronaviruses are enveloped non-segmented positive-sense RNA viruses belongs to the family Coronaviridae and distributed in humans and other mammals. Six coronavirus species are known to cause human disease. Four viruses; 229E, OC43, NL63, and HKU, are prevalent and typically cause common cold symptoms in immune-compromised individuals. The two other strains; severe acute respiratory syndrome coronavirus (SARS-CoV) and middle east respiratory syndrome coronavirus (MERS-CoV) are zoonotic in origin and have been linked often to fatal illness.

The coronavirus disease 2019 (COVID-19), which was originated in late December 2019, in Wuhan, China, has been declared a public health emergency of international concern by the World Health Organization (WHO). The disease was caused by a member of the family of coronaviruses, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). The spectrum of this disease ranges from mild fatigue, myalgia, fever, dry cough, and dyspnea to severe manifestations like acute respiratory distress syndrome (ARDS), septic shock, Disseminated Intravascular Coagulation (DIC), and acute renal failure. On July 11, 2020, there were over 12 million confirmed cases and more than 5,62,000 deaths globally due to COVID-19.

On July 18, 2020, there were a total of 17,502 confirmed cases and 40 deaths due to COVID-19 in Nepal. There is no proven treatment or vaccination against SARS-CoV-2 so far. Hence, applying preventive measures to control COVID-19 infection is the most critical intervention. Recommended measures to prevent spread infection include frequent hand washing, maintaining physical distance, covering coughs and sneezes with a tissue or inner elbow, and avoid frequent face touch with unwashed hand. Health Care Workers (HCWs) are directly in contact with patients and are exposed to infected cases in health care settings; so
they are expected to be at high risk of infection.10-12 In several instances, misunderstandings among HCWs leads to controlling efforts to provide necessary treatment in vain.13 Misinformation, misunderstanding, lack, or inadequate awareness among people, non-compliance to basic sanitation procedures could lead to the rapid transmission of infection in the community. Therefore, for the effective implementation of preventive measures, it is important to examine the level of the knowledge and perception towards COVID – 19 as well as the source of information among the Nepalese HCW and health students during this global health crisis. The main objective of this study is to study the source of information and knowledge and perception of HCWs and students towards COVID-19.

II. Methods

a) Study Design and Population

A web-based, cross-sectional survey was conducted for a week from June 27 to July 03, 2020, among HCWs and health students from the medical and paramedical field, i.e., Doctor, Pharmacist, Nurse, Dentist and Lab Technician in Nepal. Convenient non-probability sampling was used as interested participants could self-participate.

b) Study Tool

The survey instrument comprised closed-ended questions that were developed in Google forms and took approximately five (5) minutes to complete.14 The question was divided into different section including participant characteristics, awareness on COVID-19, source of information (4 statements/4-point Likert scale: 1 for least used to 4 for most used), knowledge about symptoms of COVID - 19 infected patients, different modes of transmission, precautions and risk prevention (3 items) and perceptions of COVID - 19 (7 items/true or false questions).

Knowledge was assessed by a questionnaire focusing on COVID-19 etiology, signs and symptoms, transmission, and risk prevention. Each response was scored as “1” (correct) and “0” (wrong), with scores ranging from 1 to 7. A cutoff level of ≤4 was considered to indicate poor knowledge about COVID – 19, whereas >4 was considered adequate knowledge about COVID 19.

Perceptions toward COVID-19 were assessed using seven (7) items, and each question was labeled as good (scored as “1”) or poor perception (scored as “0”). Scores ranged from 0 to 7. The participants’ perceptions are classified as good (score >5) or poor (score ≤5).

c) Statistical Analysis

The obtained data were coded, validated, and analyzed using SPSS version 16. Descriptive analysis was applied to calculate frequencies and proportions. The chi-square test was used to investigate the level of association among variables. A p value of less than .05 was considered statistically significant.

d) Ethical Considerations

This study was approved by the Ethical Review Board (ERB) of the Nepal Health Research Council (NHRC). Confidentiality of personal information was maintained throughout the study by making participants’ information anonymous and data secured properly. Eligible HCWs’ and students who participated in this survey were voluntary and were not compensated. Electronic informed consent was shown on the initial page of the survey. The study was performed following the Declaration of Helsinki, as revised in 2013. The study was conducted following the Checklist for Reporting Results of Internet E-Surveys (CHERRIES) guidelines.15

III. Results

a) Socio-demographic Characteristics of the Participants

Of the total 501 participants, most of the participants were HCWs, i.e., 350 (69.9%) illustrated in fig 1 below. Two third of the participants, 334 (66.7%) were female, and 18-24 years, 277 (55.3%) were the most common age group. The highest frequency of participants was from province 4 and 3 accounting to 224 (44.7%) and 115 (23.0%), respectively. All participants were aware of COVID 19. However, only 288 (57.5%) attended lectures and discussions about COVID 19. The socio-demographic characteristics of the participants are presented in Table 1.
**Table 1:** Socio-demographic Characteristics of the Participants

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Total (N=501)</th>
<th>HCWs (n=350)</th>
<th>students (n=151)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male (n=178)</td>
<td>167 (33.3%)</td>
<td>114 (22.8%)</td>
<td>53 (10.6%)</td>
</tr>
<tr>
<td>Female (n=175)</td>
<td>334 (66.7%)</td>
<td>236 (47.1%)</td>
<td>98 (19.6%)</td>
</tr>
<tr>
<td><strong>Age (years)</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18-24</td>
<td>277 (55.3%)</td>
<td>158 (31.5%)</td>
<td>119 (23.8%)</td>
</tr>
<tr>
<td>25-34</td>
<td>212 (42.3%)</td>
<td>182 (36.3%)</td>
<td>30 (6%)</td>
</tr>
<tr>
<td>35-44</td>
<td>7 (1.4%)</td>
<td>6 (1.2%)</td>
<td>1 (0.2%)</td>
</tr>
<tr>
<td>45-54</td>
<td>4 (0.8%)</td>
<td>4 (0.8%)</td>
<td>-</td>
</tr>
<tr>
<td>55-64</td>
<td>1 (0.2%)</td>
<td>-</td>
<td>1 (0.2%)</td>
</tr>
<tr>
<td><strong>Heard about COVID19 (Yes)</strong></td>
<td>501 (100%)</td>
<td>350 (69.9%)</td>
<td>151 (30.1%)</td>
</tr>
<tr>
<td><strong>Attended lectures/discussions of COVID-19 (Yes)</strong></td>
<td>288 (57.5%)</td>
<td>187 (37.3%)</td>
<td>101 (20.2%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Province</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number (%)</td>
<td>22</td>
<td>23</td>
<td>115</td>
<td>224</td>
<td>86</td>
<td>12</td>
<td>19</td>
</tr>
<tr>
<td></td>
<td>(4.4%)</td>
<td>(4.6%)</td>
<td>(23%)</td>
<td>(44.7%)</td>
<td>(17.2%)</td>
<td>(2.4%)</td>
<td>(3.8%)</td>
</tr>
</tbody>
</table>

![Profession of Participants](image)

*Fig. 1: Profession of Participants*
The findings of the study primarily depend on the source, which disseminates the information to the public as well as the participants of our study. The various medium of information such as news media, social media, and official government website has been proactively providing information nowadays. In our study, more than half of the participants depending on social media like Facebook, Twitter, and Instagram as the main source of information about COVID – 19, as shown in Fig.2.

**b) Knowledge about COVID-19**

The knowledge about COVID-19 among HCWs and Health students is presented in Table 2. From our survey, we observed that there is no significant gap in knowledge between HCWs and students. Correct responses about the origin of COVID-19 were obtained from 413 (82.4%) participants, among which 287 (82%) were HCWs, and 126 (83.4%) were students. Most of the participants agreed headache, fever, cough, sore throat, and flu as the symptoms of COVID-19, which lead to pneumonia, respiratory failure, and death. Similarly, most of the participants agreed on supportive care as the current treatment approach for COVID-19. The response related to the mode of transmission, incubation period, and current treatment of COVID-19 were poor in both HCWs and students.
**Table 2: Knowledge about COVID-19**

<table>
<thead>
<tr>
<th>Knowledge</th>
<th>Correct responses</th>
<th>HCWs (n=350)</th>
<th>Students (n=151)</th>
<th>p - value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>COVID-19 is thought to be originated from bats</td>
<td>413 (82.4%)</td>
<td>287 (82%)</td>
<td>126 (83.4%)</td>
<td>0.697</td>
</tr>
<tr>
<td>COVID-19 is transmitted through air, contact, fecal-oral routes</td>
<td>260 (51.9%)</td>
<td>187 (53.4%)</td>
<td>73 (48.3%)</td>
<td>0.296</td>
</tr>
<tr>
<td>Headache, fever, cough, sore throat, and flu are symptoms of COVID-19</td>
<td>431 (86%)</td>
<td>296 (84.6%)</td>
<td>135 (84.6%)</td>
<td>0.152</td>
</tr>
<tr>
<td>The incubation period of COVID-19 is 2 to 14 days</td>
<td>231 (46.1%)</td>
<td>152 (43.4%)</td>
<td>79 (52.3%)</td>
<td>0.067</td>
</tr>
<tr>
<td>COVID-19 leads to pneumonia, respiratory failure, and death</td>
<td>405 (80.8%)</td>
<td>285 (81.4%)</td>
<td>120 (79.5%)</td>
<td>0.609</td>
</tr>
<tr>
<td>Supportive care is the current treatment for COVID-19</td>
<td>319 (63.7%)</td>
<td>229 (65.4%)</td>
<td>90 (59.6%)</td>
<td>0.213</td>
</tr>
<tr>
<td>Hand hygiene, covering nose and mouth while coughing, and avoiding sick</td>
<td>484 (97%)</td>
<td>338 (96.8%)</td>
<td>146 (97.3%)</td>
<td>0.771</td>
</tr>
<tr>
<td>contact can help in the prevention of COVID-19 transmission</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**c) Perception of COVID-19**

The perception of COVID-19 among HCWs and Health students are presented in Table 3. There is no significant gap in perception between HCWs and students. The majority of participants 443 (88.4%), perceived COVID-19 incubation period as 2 to 14 days which is correct, 479 (95.6%) responded that flu vaccination is not sufficient for preventing COVID-19, and 452 (90.2%) felt that eating well-cooked and safely handled meat is safe. Additionally, 486 (97%) of the participants agreed that patients should share their recent travel history with health care professionals, and 498 (99.4%) that washing hands with soap and water can help in the prevention of COVID-19 transmission, however, only 152 (30.3%) participants were aware that COVID-19 is not fatal.

**Table 3: Perception of COVID-19**

<table>
<thead>
<tr>
<th>Perception</th>
<th>Correct responses</th>
<th>HCWs (n=350)</th>
<th>Students (n=151)</th>
<th>p - value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>COVID-19 symptoms appear in 2-14 days</td>
<td>443 (88.4%)</td>
<td>308 (88%)</td>
<td>135 (89.4%)</td>
<td>0.652</td>
</tr>
<tr>
<td>COVID-19 is fatal</td>
<td>152 (30.3%)</td>
<td>102 (29.1%)</td>
<td>50 (33.1%)</td>
<td>0.375</td>
</tr>
<tr>
<td>Flu vaccination is not sufficient for preventing COVID-19</td>
<td>479 (95.6%)</td>
<td>332 (94.9%)</td>
<td>147 (97.4%)</td>
<td>0.211</td>
</tr>
<tr>
<td>During the outbreak, eating well-cooked and safety handled meat is safe</td>
<td>452 (90.2%)</td>
<td>313 (89.4%)</td>
<td>139 (92.1%)</td>
<td>0.364</td>
</tr>
<tr>
<td>Sick patients should share their recent travel history with health care</td>
<td>486 (97%)</td>
<td>338 (96.6%)</td>
<td>148 (98%)</td>
<td>0.420</td>
</tr>
<tr>
<td>professionals</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disinfect equipment and working area in wet markets at least once a day</td>
<td>456 (91%)</td>
<td>322 (92%)</td>
<td>134 (88.7%)</td>
<td>0.242</td>
</tr>
<tr>
<td>Washing hands with soap and water can help in the prevention of COVID-19</td>
<td>498 (99.4%)</td>
<td>349 (99.7%)</td>
<td>149 (98.7%)</td>
<td>0.167</td>
</tr>
</tbody>
</table>

**d) Level of Knowledge and Perception of COVID-19**

The level of knowledge was categorized as poor (≤4) and good (>4). Among all participants, 253 (72.3%) HCWs and 112 (74.2%) students had a good level of knowledge on COVID-19. Similarly, the level of perceptions was categorized in positive (>5) and negative (≤5). Only 185 (52.9%) HCWs and 77 (51%) of students showed a positive perception towards COVID-19. There was no significant difference in knowledge between HCWs and students regarding the knowledge and perceptions of COVID-19. The detail of the level of knowledge and perception of COVID-19 is given in Table 4.
### IV. Discussion

The WHO recognized COVID-19 as a pandemic on March 11, 2020. Globally, the mortality rate of COVID-19 was found to be about 7% progressively spreading among more than 200 countries. Participants had good general knowledge and mixed perceptions about the disease in the current study, and there was no significant difference in knowledge between HCWs and students.

We found that more than half of the participants depended on Social media like Facebook, Twitter, and Instagram as the main source of information about COVID-19. This differs from the findings on previously published studies, where most of the HCWs depended on Government websites and news bulletin to obtain COVID-19 related information. Obtaining information from social media is a major concern because of the difficulty of determining the validity and authenticity of the available information.

Our study highlights that all the HCWs and students are knowledgeable of COVID-19. Majority of the participants had good knowledge of COVID-19 which was similar to the finding of the study conducted in Nepal, China, USA and UK, and Egypt. The present finding suggests that there was inadequate information regarding mode of transmission and incubation period among the participants corresponding to the study done by Bhagavathula et al., but still, in contrast to Farhana and Mannan et al. Regarding the treatment, had the correct responses which were similar to the finding of the study conducted in Nepal, 597 (68.5%) There was no significant gap in knowledge between HCWs and students in our study. However, to further update the knowledge among HCWs and students, there should be a continuous effort from the government and health authorities.

In our study, most of the HCWs and students showed a positive perception regarding COVID-19. Majority of the participants were knowledgeable of 2-14 days incubation period of COVID-19, flu vaccination is not sufficient for preventing COVID-19, eating well-cooked and safety handled meat is safe, sick patients should share their recent travel history with health care professionals, disinfect equipment and working area in wet markets at least once a day and washing hands with soap and water can help in the prevention of COVID-19 transmission. These results are comparable with the study conducted by Bhagavathula et al. and Farhana and Mannan et al. Whereas the correct response for COVID-19 as fatal, accounting to 152 (30.3%), which was low and different from the previous study of Nepal and Bangladesh. To strengthen preventive strategies and raise awareness regarding the COVID-19, the WHO initiated several online training sessions and materials in various languages, which can be utilized to reduce misinformation and misunderstanding regarding the disease.

### V. Conclusion

We identified that there was no significant gap between HCWs and health students regarding the knowledge and perceptions of COVID-19. The global struggle to tackle the COVID-19 pandemics will be successful by ensuring the accurate knowledge and perception among HCWs and the Health students. Strategies should be adapted for effective dissemination of the information regarding COVID-19, among HCWs and students.

### Acknowledgments

The authors would like to thank study participants for their voluntary participation and for providing essential information. The authors also wish to thank Akshaya Srikanth Bhagavathula, PharmD, Institute of Public Health, College of Medicine and Health Sciences, United Arab Emirates University, Abu Dhabi, the United Arab Emirates, for sending questionnaires through airmail, encouragement, and support during this research work.

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Online Survey on the Source of Information, Knowledge, and Perceptions towards COVID-19 among Health Care Workers and Health Students in Nepal: A Comparative Study


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Covid-19: A Review of History, Clinical Presentation, Transmission, Pathogenesis, Diagnosis, Treatment and Prevention

By Amar Prasad Chaudhary, Chiranjibi Sah, Yalda Hashemzadeboneh & Jamuna TR

Rajiv Gandhi University of Health Sciences

Abstract- The outburst of coronavirus disease 2019 (COVID-19) has produced unprecedented challenges in the world which, were seen initially at Wuhan, Hubei Province, China beginning in December 2019. Genomic studies have revealed that the bat might be the primary reservoir of this virus. The symptom of COVID-19 varies from asymptomatic or paucisymptomatic to the clinical condition. The COVID-19 is transmitted through the close contact of infected people via droplet. Real-time Reverse Transcriptase-Polymerase chain reaction (RT-PCR) is considered to be the gold standard for the diagnosis of COVID-19. Many drugs were used for the treatment of this virus, but most of them aren’t effective against it and only help to improve the recovery rate.

GJMR-B Classification: NLMC Code: QV 4

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Covid-19: A Review of History, Clinical Presentation, Transmission, Pathogenesis, Diagnosis, Treatment and Prevention

Amar Prasad Chaudhary *, Chiranjibi Sah *, Yalda Hashemzadeboneh * & Jamuna TR *

Abstract - The outburst of coronavirus disease 2019 (COVID-19) has produced unprecedented challenges in the world which, were seen initially at Wuhan, Hubei Province, China beginning in December 2019. Genomic studies have revealed that the bat might be the primary reservoir of this virus. The symptom of COVID-19 varies from asymptomatic or paucisymptomatic to the clinical condition. The COVID-19 is transmitted through the close contact of infected people via droplet. Real-time Reverse Transcriptase-Polymerase chain reaction (RT-PCR) is considered to be the gold standard for the diagnosis of COVID-19. Many drugs were used for the treatment of this virus, but most of them aren’t effective against it and only help to improve the recovery rate.

I. INTRODUCTION

Coronavirus disease 2019 (COVID-19) outburst were first seen in Wuhan city of China in December 2019 caused by a novel member of coronavirus family, i.e, Severe Acute Respiratory Distress Syndrome Coronavirus-2 (SARS-COV-2) [1, 2]. International Committee on Taxonomy of Viruses (ICTV) officially named it as SARS-COV-2 [3]. The seafood market in Hunan were identified as the source of the SARS-COV-2 virus [4]. As it was the time of the spring festival, visitors have come from various parts of China, so it spread in a different parts of China [4]. With the use of real-time Reverse Transcriptase-Polymerase chain reaction (RT-PCR) researcher identified it caused due to SARS-COV-2 [4]. The cases increased in large number, so the World Health Organization (WHO) declared COVID-19 a Public Health Emergency of International Concern (PHEIC), i.e. Pandemic, in 11-march, 2020. The study of genome sequences of SARS-COV-2 revealed that its genome is 79.5% similar to SARS-COV and 96% to bat SARS-COV [5].

COVID-19 is characterized by fever, cough, fatigue, shortness of breath, pneumonia, and other respiratory tract symptoms and, in many cases, progress to death [5]. In child and adolescents, SARS-COV-2 have mostly caused mild respiratory symptoms rather than severe forms like in adults and old age people, the late manifestation of young adult has shown vasculitis which might be due to post-viral immunological reaction [6]. As of June 21, total confirmed cases worldwide is 8.75 million, 4.33 million people recovered with death mounting to 463,000. Due to the rapid spread of disease, globally, it leads to a shortage of mechanical ventilator, personal protective equipment, and other hospital equipment’s [7]. There are currently no approved antiviral drugs effective against the COVID-19, but few broad-spectrum antibiotics and antivirals are used to improve the recovery rate like doxycycline, oseltamivir, remdesivir, azithromycin, hydroxychloroquine, etc. [1]. Despite the immediate need for information for decision making, data remained limited in COVID-19, which led to a rapid increase in disease and poor health outcomes [7]. There are several reports from China, Italy, and the USA explaining some characteristics of infection but there is very less information regarding the factors associated with hospital stay and severity of disease. But it has been seen old age, heart failure, male sex, chronic kidney disease, and obesity were associated with hospital stay and severity of disease [8].

II. HISTORY AND ORIGIN

Coronavirus belongs to the coronaviridae family in the Nidovirales order [1, 9]. Corona means crown-like spikes on the outer surface of the virus, so it was named as a coronavirus [1]. Coronavirus is very small in size i.e 65-125mm diameter [1]. It is a non-segmented, positive sense, single stranded RNA as nucleic material of 26-32kbs in length [1]. Coronaviruses are important pathogen of birds and mammals [9]. While studying the coronavirus in wild animals has helped found out the greatest diversity of coronavirus in bat and avian species, which suggest these animals are natural reservoirs of the viruses [9]. Molecular clock dating helps to find out the most recent ancestor of these viruses existed around 10000years ago [9]. Human coronavirus reported since 1960 initially by HCOV-229E [10]. In 2003, Guangdong province of China was infected by Symptom of Acute Respiratory Distress Syndrome (SARS) and exhibited pneumonia like symptoms [1]. In 2012, a Couple of Saudi Arabia was infected with another coronavirus strain later it was confirmed Middle East Respiratory Syndrome (MERS) [1]. The coronavirus family is subdivided into four types. They are α coronavirus, β coronavirus, δ coronavirus, γ coronavirus.
Coronavirus [1, 9]. Alpha and beta are exclusively found in mammals, whereas gamma and delta mostly infect birds [9]. Among all types of coronavirus, seven species of coronavirus infects human beings. The four are very common they are [9, 11]

1. 229E (α coronavirus)
2. NL63 (α coronavirus)
3. OC63 (β coronavirus)
4. HKU3 (β coronavirus).

The three less common are [9, 11]

5. MERS-COV (β coronavirus cause Middle East Respiratory Syndrome i.e. MERS)
6. SARS-COV (β coronavirus cause Severe Acute Respiratory Syndrome i.e. SARS)
7. SARS-COV-2. (Nobel coronavirus causes COVID-19)

### III. Clinical Presentation

- **Age**
  
  Most of the cases of COVID-19 age ranges from 30-70 years on an average as the study conducted in China, United Kingdom (UK) and United States of America (USA) and the median age was found to be around 45-58 years [2, 4, 5, 8, 12]

- **Comorbidities**
  
  From the Various studies, it was found that Diabetes mellitus, Hypertension, any form of cardiovascular disease, Chronic Obstructive Pulmonary Disease (COPD) and Chronic Kidney Disease (CKD) as one of the most common comorbidity associated with the hospital admission in the patient and severity of patient [2, 3, 4, 12, 13]

- **Symptoms**
  
  The clinical presentation of disease varies from asymptomatic or paucisymptomatic forms to the clinical conditions. Most of the cases of COVID-19 positive worldwide mostly presents with fever, cough, sputum, shortness of breath, and fatigue [5]. The patient may come with some other symptoms but from the various studies it is found that chances of occurrence of such symptoms are less like headache, wheezing, abdominal pain, confusion, diarrhea, nausea/vomiting, seizures, lymphadenopathy, runny nose, etc. [2, 3, 4, 8, 12, 13]. In children, it was found that the median age was 3.3 years and the most common symptoms were cough, and no feeding or difficulty in feeding [14]. In infants those who were less than 21 months, it was found that fever, cough, shortness of breath was most common [14].

- **Complications**
  
  1. Acute Respiratory Distress Syndrome (ARDS)
     
     A large number of patients admitted in hospitals developed ARDS, and it was seen more in the patient having diabetes mellitus as comorbidity. ARDS is seen worsening in patient more than 65 years old [3, 4, 12].

  2. Myocardial Injury
     
     Myocardial injury includes acute coronary syndrome, heart failure, myocarditis, hypotension or shock, and sepsis. The patient in which the condition is severe the chances of heart failure increases the patient having or not having any cardiovascular disease [4, 13].

  3. Acute kidney injury
     
     The COVID-19 patient undergoing acute kidney injury have an increase in urea and cystatin-C level especially in severe patients [4, 13].

### IV. Transmission

The SARS-COV-2 was originated from Wuhan, China, and spread all over the world through the imported case from China. Based on research, Bats are considered to be the natural host for COVID-19 and Snake and pangolin an intermediate source for the virus [15]. In humans, droplet and close contact are the most common route of transmission of SARS-COV-2 [16]. Transmission occurs through nose, mouth, and eyes [16]. Droplet transmission occurs when a person is in close contact (within 1 meter) with someone who has respiratory symptoms [16]. As per the research conducted by Neeltje van Doremalen, et al. stability of SARS-COV-2 virus was found similar to SARS-COV virus [17]. SARS-COV-2 was more stable on plastic, and stainless steel than copper and cardboard and the viable virus was present up to 72 hours after the application of this virus in these materials under experimental condition (40% humidity, 21°C-23°C) [17]. It was found in copper no viable virus remained after 4 hours and on cardboard after 24 hours [17]. It was seen that SARS-COV-2 was viable in aerosol for 3 hours [17].

### V. Pathogenesis

After the outbreak of the COVID-19 disease, it has led to the loss of a significant amount of lives and effected the quality of life of people. The systematic understanding of pathogenesis of the disease will help a lot in controlling the disease and solving some of the important questions arisen from various studies conducted worldwide. Why old age people are at more risk? [4] Why people with comorbidities like DM, CVD, Hypertension, and CKD are more prone to infection? [2, 13] Why the severity of disease in child and adolescent is seen less than that of adult and old age? [6].

The sign and symptoms of COVID-19 is similar to that of SARS and MERS [18]. The similar pathogenesis of SARS-COV and MERS-COV gives a lot of information to understand the pathogenesis of SARS-COV-2 though COVID-19 is poorly understood.

- **Life cycle of coronavirus**
  
  The life cycle of corona virus consist of five stages, they are attachment, penetration, biosynthesis, maturation, and release [18, 19]. Coronavirus consist of 4 structural protein i.e. Spike (s), Membrane (M),
Envelope (E) and Nucleocapsid (N) [19]. Coronavirus S protein is very important for the penetration of the virus in the host cell [18]. Spike is composed of transmembrane trimetric glycoprotein is above the viral surface [19]. Spike protein consists of two functional subunit S₁ and S₂ [19, 20]. S₁ subunit is responsible for binding to host cell and S₂ subunit is responsible for fusion [19].

The various information suggested that SARS-COV-2, SARS-COV, and MERS bind to ACE2 receptor [18, 19, 21]. The information found from the invitro study of SARS-COV that ciliated cells are primary cells infected in conducting airways and type 2 alveolar cells in lungs which is found in peripheral and sub pleural region of lungs [21].

After binding to the ACE2 receptor, the spike protein undergoes two steps sequential protease cleavage to activate spike protein of SARS-COV and MERS-COV which helps in the fusion [18, 19]. There is also another method for fusion mechanism is SARS-COV i.e. clarithin dependent and independent endocytosis [18].

After the entry of the virus, viral RNA enters into the nucleus for replication, and viral mRNA is used to make viral protein [18, 19]. The newly produced envelope glycoprotein enters in the endoplasmic reticulum and Golgi bodies [18,19]. Then viral particle is formed in endoplasmic golgi intermediate compartment (EGIC) [18,19]. The virus particle fuse with the plasma membrane to release virus [18, 19]

- Host response
  - When the virus enters the body, T-cell mediated response against coronavirus is initiated through antigen presentation cell i.e. dendritic cells and macrophages [18,19]. Antigenic peptides are presented by Major Histocompatibility Complex (MHC) to cytotoxic T-lymphocyte [18, 19]. Due to less research there is no much information regarding type of MHC used by SARS-COV-2 but it is found that MHC I is used by SARS-COV and MHC II is mostly used by MERS-COV [18]. Antigen presentation leads to the development of immunity which is mediated by the virus specific B and T cells [18, 19]. IgG and IgM production takes place in a typical pattern in SARS-COV [18]. IgM remains for 12 weeks and IgG remains for long time [18]. The latest information about COVID-19 patient suggests there is a reduction in CD4⁺ and CD8⁺ with excessive activation status [18, 19]. CD4⁺ activate B cells to promote production of virus specific antibody while CD8⁺ and T cell can kill viral infected cells [19].

VI. Diagnosis

Coronavirus disease 2019 tracking and diagnostic testing are difficult as many patients are asymptomatic or having mild symptoms [16]. The WHO recommends collecting a sample from both upper and lower respiratory tract. The sample is collected through sputum, bronchoalveolar lavage, throat swab, nasopharyngeal swap, endotracheal aspirate and these sample are assessed for the detection of virus [22]. To detect the virus there are currently two major tests are available [23].

- Diagnostic test
  1. Reverse transcriptase Polymerase Chain Reaction (RT-PCR)
  2. Immunological assay

RT-PCR relies mainly on the detection of viral RNA by conversion of RNA to DNA which mainly comprises two enzymatic steps [23, 24]. The first step is to convert RNA to cDNA and next step is to use Taq polymerase which amplifies the cDNA [23, 24]. RT-PCR is considered as the gold standard for the detection of COVID-19 [23, 24]. The immunological assay mostly depends on the detection of antibodies produced by individuals as a respect of exposure to the virus [23, 24].

- Laboratory findings
  - In the cases seen worldwide, there are a large number of changes in the laboratory parameters but some of the major changes in laboratory parameters are lymphopenia, leucopenia, esinopenia, elevated neutrophils, and elevated C-reactive protein. In severe condition patient’s neutrophils, D-Dimer, blood Urea and Creatinine is elevated and lymphocyte is reduced [3, 4, 8, 12, 13].

- Radiological findings
  - When Chest X-ray was done in the patients, most of the patient presented with abnormalities in chest. Ground glass opacification was dominant during the early stage and consolidation present at the later stage, bilateral patches are also seen in the patients sometimes with rounded morphology [3, 4, 13].

VII. Management

Many drugs are used in the treatment of COVID-19 but till now no drugs are found to be effective in its treatment. Most of the drugs are used to improve the mortality rate, recovery rate, treating the sign and symptoms, reducing the prognosis of the disease and prophylaxis. The drugs used in treatment are given below.

- Post exposure prophylaxis
  - Normally Hydroxychloroquine, an antimalarial drug was used for the post-exposure prophylaxis but trial conducted by D.R. Boulware, et al. by administering 800mg once followed by 600mg after 6-8hours of first dose, then 600mg for 4 days once daily showed it don’t prevent illness after high risk or moderated risk exposure to COVID-19 [25].
In the treatment of the COVID-19 patient but as per the randomized, controlled, open-label trial involving a hospitalized adult patients with COVID-19 positive found no benefits when given in 1:1 ratio (400mg:100mg) twice daily for 14 days [30].
• Avoid visiting markets, large events and mass gathering [35].
• Wash the hands regularly and thoroughly with soap and water at least for 20 seconds or use alcohol based hand sanitizer. (Contains at least 60% alcohol)[16, 35].
• Contaminated hands can transfer the virus from hand to eye, nose and mouth, so avoid touching these organs with unwashed hands [16].
• Maintain the social distance i.e distance of 1 meter (3 feet) [16]  

IX. Conclusion

Since the outbreak of COVID-19 lot of research have been carried out most of them focused mostly in epidemiology, clinical presentation, diagnosis and treatment. More research is required to understand the pathogenesis and treatment which will help a lot to eradicate the disease. Currently, the best measures to fight against the COVID-19 is to follow the preventive measures as no specific medications are available nor the vaccine.

References Références Referencias

Phenolic Profile, Anti-Inflammatory and Diuretic Properties of Asplenium Ceterach Tested on Albino Mice and Wistar Albino Rats

By Mostapha Bachir Bey, Rekia Sidhoum, Latifa Halli, Othmane Yalaoui & Asma Belkadi

University of Bejaia

Abstract- Asplenium ceterach is a fern used in traditional medicine against kidney stones and as a diuretic agent. Thus, we set as objective of this study is the determination of the phenolic profile as well as the study of anti-inflammatory and diuretic properties of Asplenium ceterach (Rustyback). Phenolic composition is achieved by High-performance liquid chromatography, the anti-inflammatory activity is determined in albino mice, and the diuretic activity in Wistar albino rats. The results of phenolic profile show an overall concentration of 3926 μg/g DM of Asplenium ceterach, of which 3/4 are represented by chlorogenic acid. This plant is endowed with an interesting plantar anti-inflammatory property and a topical anti-inflammatory comparable to that of ibuprofen. Asplenium ceterach expresses an excellent diuretic activity significantly higher that of Furosemide. This study confirms the traditional use of the infusion of Asplenium ceterach as a powerful diuretic and an anti-inflammatory.

Keywords: rustyback; phenolic composition; chlorogenic acid; biological property; in vivo activity.

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1. Introduction

Several researches have intensified on medicinal plants used in therapeutic purposes subsequent the evolution of means and methods of analysis. The special interest for these plants is their ability to synthesize and accumulate micronutrients endowed with preventive and curative properties of innumerable pathologies.

Many substances of plant origin, such as phenolic compounds, terpenoids, and alkaloids are sought for their variables biological activities and therapeutic properties for several diseases including cardiovascular disorders, cancers, and neurodegenerative diseases (1).

The plant which is the subject of this study is Asplenium ceterach (Finger fern and locally called “Tahchicht warman tassa” or “Kessar lahar”) and which is belongs to Aspleniaceae family known as a common plant of rocks and old limestone walls. This perennial plant is characterized by 5-15 cm of length, short strain, thick, and turf. Its leaves are tufted, rolled in lacrosse at their young age, spread, with a short and scaly petiole, narrow oblong, pennatized, short-lobed, ovate-obtuse, whole or crenate, alternate, confluent, thick, glabrous, and green on top, covered under shiny scales first silvery then reddish. The fructification is arranged on the underside of the leaves in elongated, straight, scattered and placed obliquely on the midrib, devoid of indusia. It adapted to drought; in dry weather the leaves curl and the presence of scales disposed in the under face limited evapotranspiration (2).

The medicinal plant Asplenium ceterach is little studied, however, some properties and traditional uses have been reported as its use against kidney stones, diarrhea, neuralgia as well as prostate and gastrointestinal disorders (3). This plant is also endowed with interesting antioxidant potential and antimicrobial and protective activities against DNA damage (4). The infusion prepared with this plant is an excellent diuretic by taking 2 to 3 cups per day (5).

In speaking of diuretic, diuresis is a vital function for the living being by which the organism excretes all the waste resulting from the cellular combustion, once it has kept the necessary substances to feed itself and produce energy, it is considered among the functions of elimination. Many diseases, namely coronary heart disease, arterial disorders, cardiac, and renal insufficiency, are related to hypertension (6,7). Common clinical strategies to reduce blood pressure include the prescription of drug that decreasing arterial resistance and/or reducing cardiac output. Among the medicament most used to promote increased urine volume output, urinary sodium, and which leads to the reduction of blood pressure are diuretics (8).

Inflammation is the response of vascularized tissue to physical, chemical, or biological aggression in order to maintain its integrity. Inflammatory response is a usually beneficial process; its goal is to mobilize the immune system to eliminate the pathogen and repair damaged tissues. In some cases, inflammation may be harmful because of the aggressiveness of the pathogen, its persistence, the site of inflammation, or abnormal regulation of the inflammatory process (9). The use of drugs and some plants reduce inflammation and thus prevent pain.
To our knowledge, no work has been conducted to investigate biological activities of *Asplenium ceterach*. Thus, the objectives of this investigation are the determination of phenolic profile as well as anti-inflammatory and diuretic properties of *Asplenium ceterach*.

II. MATERIALS AND METHODS

a) Plant material

*Asplenium ceterach* was collected from Fenaia Ilmatten municipality (Bejaia department, Algeria) at an altitude of 400-450m during the period from March to April 2017. The aerial part of the harvested plant was dried in the open air during 15 days after which the dried plant was crushed and reduced to a fine powder using an electric grinding mill (Philips, Enapem®) and then sieved using a sieve of mesh size of 250 μm. The resulting powder is kept away from air and moisture.

b) Preparation of plant extracts

Extraction of phenol compounds is carried out by maceration of the *Asplenium ceterach* powder (1 g) using 10 ml of methanol HPLC grade with magnetic stirring during 30 min. The extract is separated from the powder by filtration. The powder is re-extracted two more times using 10 ml with the same solvent. The three recovered filtrates are mixed and the solvent is removed by a rotary evaporator using the temperature of 65 °C until the volume is reduced to 4 ml.

c) Phytochemical screening

Phytochemical screening is a set of tests carried out either on the powder or on the plant infusion to highlight the presence or absence of certain secondary metabolites. Phytochemical screening of *Asplenium ceterach* concerns flavonoids, anthocyanins, leucoanthocyanins, total tannins, catechic tannins, gallic tannins, saponosides, coumarins, free quinones, combined quinines, and alkaloids (10,11).

d) Determination of phenolic compounds

The phenolic profile of *Asplenium ceterach* is determined by high performance liquid chromatography (Waters 2695 Alliance) equipped by a diode array detector (DAD) set at three wavelengths (280, 320, and 350 nm). For the elution of phenolics, a mobile phase consisting of 1% formic acid solution (A) and acetonitrile (B) that used with a flow rate of 1 ml/min. For the stationary phase, it consists on LiChrosorb RP-18 column (250 x 4.0 mm, 5 μm). The volume of extract injected is 10 μl and the system is set at a temperature of 30 °C. The elution gradient used is as follows: 5% (B): during the first two minutes, 02-12 min: 5-95% (B), 12-12.2 min: 95-5% (B), and 5% (B) up to 20 min. Phenolic compounds are identified and quantified by reference to retention times and peak areas of phenolic standards, and the results were expressed as micrograms per gram of dry matter (μg/g DM).

The total phenolic content (TPC) is determined by the Folin-Ciocalteu method according to Singleton and Rossi (12). The TPC is expressed as μg/g DM according to the calibration curve using gallic acid as standard ($y = 11.64x - 0.019, R^2 = 0.998$).

e) Biological properties

i. Animal material

15 Albino female/male mice (weighing between 26-28 g) and 15 Wistar albino male rats (weighing from 150 to 200 g) obtained from the pharmaceutical group company SAIDAL (Algiers, Algeria) are used for anti-inflammatory and diuretic activities. Animals were housed in plastic cages with a 10/14h light/dark cycle at an ambient temperature of 20-24°C. Animals were fed standard diet and water *ad libitum*. All experiments were in compliance with the guidelines for the care and use of laboratory animals published by the US National Institute of Health (NIH publication No 85-23, revised 1985) with approval of SAIDAL ethic committee.

ii. Anti-inflammatory activity

Anti-inflammatory activity is estimated according to Vetriselvan et al. (13). The fifteen mice are randomly divided into three groups (n = 5). The first group received orally 0.5 mL of *Asplenium ceterach* extract (20g of plant powder extracted by 100ml of boiling distilled water per 15min); the second is treated with 0.5 mL of Ibuprofene (a diuretic drug prepared at 200mg/250mL dw) and considered as a positive control; the third group (negative control) received 0.5 mL of distilled water.

Subsequently, the plantar and topical inflammations are induced after 30 min. The plantar inflammation is generated by the injection of carrageenan (25μl) under the plant aponeurosis of the left hind paw of the mouse; the no treated right paw is considered as a control. The topical inflammation is induced by topical application of xylene (10μl) on outer surface of right ear of each mouse; the left ear represents the control.

The mice are sacrificed by cervical dislocation after 4 h of the inflammatory induction. The paws and ears are removed from the same position and weighed in analytical balance immediately. The swelling degree (SD) was estimated as the difference in the weight between treated and untreated limbs and the inhibitory rate was calculated as follows:

\[
\text{Inhibitory rate} \% = 100\times \frac{(\text{SD}_{\text{Control}} - \text{SD}_{\text{Treated}})}{\text{SD}_{\text{Control}}}
\]

iii. Diuretic activity

Diuretic activity is determined according to the method described by de Paula Vasconcelos et al. (14). The 15 Wistar albino male rats were randomly divided into three groups (n = 5). Before treatment all animals were fasting 18 hours. Then, *Asplenium ceterach* extract was administrated by the oral route at dose of 50 mg/kg. The negative control group received the isotonic saline...
(0.9% NaCl, 50 ml/kg) and the positive control group is treated with 50 mg/kg of Furosemide. Urine was collected and measured for each rat and averaged for every group after each hour during 6h and the cumulative urinary excretion (CUE) was calculated.

III. Results and Discussion

a) Phytochemical screening
The results of the phytochemical screening carried out on the infused and the powder of Asplenium ceterach are summarized in Table 1. The tests indicate that the studied plant contains many classes of secondary metabolites, of which leucoanthocyanins and tannins are the most abundant. Free quinones, coumarins, and flavonoids are also present with appreciable levels. However, the phytochemical screening does not reveal the presence of anthocyanins, combined quinones, saponosides, and alkaloids.

<table>
<thead>
<tr>
<th>Metabolites</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>Anthocyanins</td>
<td>-</td>
</tr>
<tr>
<td>Leucoanthocyanins</td>
<td>+++</td>
</tr>
<tr>
<td>Total tannins</td>
<td>+++</td>
</tr>
<tr>
<td>Catechic tannins</td>
<td>+</td>
</tr>
<tr>
<td>Gallic tannins</td>
<td>++</td>
</tr>
<tr>
<td>Saponosides</td>
<td>-</td>
</tr>
<tr>
<td>Coumarins</td>
<td>+</td>
</tr>
<tr>
<td>Free quinones</td>
<td>+</td>
</tr>
<tr>
<td>Combined quinones</td>
<td>-</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>-</td>
</tr>
</tbody>
</table>

(-), Absence of metabolites; (+), low level; (++), medium; (+++), high level.

b) Phenolic profile of Asplenium ceterach
Seven phenolic compounds are identified in studied plant and results are regrouped in Table 2. Chlorogenic acid is the compounds with the higher concentration (2817.72 μg/g DM) which represents ¾ of total phenolics of Asplenium ceterach. This latter contains appreciable content of gallic, vanillic, and coumaric acids as well as low concentrations of quercetin and benzoic acid. Overall, total phenol content of Asplenium ceterach is 3926.19 μg/g DM, indicating the richness of this plant on phenolic compounds.

Table 1: Phytochemical screening results of Asplenium ceterach.

<table>
<thead>
<tr>
<th>Compounds</th>
<th>Retention time (min)</th>
<th>Wavelength (nm)</th>
<th>Peak area (μg g⁻¹ DM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydroxybenzoic acids</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gallic acid</td>
<td>3.484</td>
<td>280</td>
<td>8657895</td>
</tr>
<tr>
<td>Vanillic acid</td>
<td>8.519</td>
<td>350</td>
<td>112851</td>
</tr>
<tr>
<td>Benzoic acid</td>
<td>13.088</td>
<td>280</td>
<td>51459</td>
</tr>
<tr>
<td>Tannic acid</td>
<td>15.091</td>
<td>280</td>
<td>1055650</td>
</tr>
<tr>
<td>Hydroxycinnamic acids</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chlorogenic acid</td>
<td>6.899</td>
<td>320</td>
<td>29407706</td>
</tr>
<tr>
<td>Coumarinic acid</td>
<td>7.950</td>
<td>320</td>
<td>3204610</td>
</tr>
<tr>
<td>Flavonoids</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Quercetin</td>
<td>10.950</td>
<td>280</td>
<td>88687</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 2: Identification and quantification of phenolic compounds of Asplenium ceterach by HPLC-DAD

TPC of Asplenium ceterach is also quantified using Folin-Ciocalteu method. This latter is widely used for the overall quantification of phenolic compounds. It based on the oxido-reduction reaction between the hydroxyl groups of the phenolics and the phosphotungstic and molybdic acids of the Folin-Ciocalteu reagent which it reduction causes the change of its color from greenish-yellow to blue. The result of total phenolic content obtained using Folin-Ciocalteu essay of the studied plant revealed a value of 7466.67 ± 148.49 μg/g DM.
The comparison of TPC results using HPLC and Folin-Ciocalteu reagent indicates that the concentration obtained by the second method is higher than the first one. This could be explained by the fact that Folin-Ciocalteu essay measures through the oxido-reduction reaction all compounds present in the extract that allowing the reduction of this reagent (phenolic compounds, reducing sugars, amino acids, etc.), whereas the chromatographic method is more accurate because it targets the compound of interest.

c) Anti-inflammatory property

The evaluation of the anti-inflammatory activity of *Asplenium ceterach* is conducted by induction of acute inflammation on the paws and ears of the mice using carageenin and xylene, respectively. Inflammation is measured by changes in paw and ear weights following induction or inhibition of edema formation.

**Table 3: Anti-inflammatory activities results of control, Asplenium ceterach, and Ibuprofen 200 mg.**

<table>
<thead>
<tr>
<th>Group</th>
<th>Parameter</th>
<th>Right legs (g)</th>
<th>Left legs (g)</th>
<th>Left ears (g)</th>
<th>Right ears (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>Average weight</td>
<td>0.076</td>
<td>0.109</td>
<td>0.029</td>
<td>0.048</td>
</tr>
<tr>
<td></td>
<td>Standard deviation</td>
<td>0.011</td>
<td>0.011</td>
<td>0.004</td>
<td>0.005</td>
</tr>
<tr>
<td></td>
<td>% of edema</td>
<td>44.33%</td>
<td>64.14%</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Asplenium ceterach</em></td>
<td>Average weight</td>
<td>0.127</td>
<td>0.165</td>
<td>0.032</td>
<td>0.044</td>
</tr>
<tr>
<td></td>
<td>Standard deviation</td>
<td>0.009</td>
<td>0.007</td>
<td>0.004</td>
<td>0.010</td>
</tr>
<tr>
<td></td>
<td>% of edema</td>
<td>29.81%</td>
<td>35.80%</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>% of edema reduction</td>
<td>32.75%</td>
<td>44.18%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ibuprofen 200 mg</td>
<td>Average weight</td>
<td>0.139</td>
<td>0.169</td>
<td>0.159</td>
<td>0.210</td>
</tr>
<tr>
<td></td>
<td>Standard deviation</td>
<td>0.021</td>
<td>0.018</td>
<td>0.027</td>
<td>0.031</td>
</tr>
<tr>
<td></td>
<td>% of edema</td>
<td>21.58%</td>
<td>31.78%</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>% of edema reduction</td>
<td>51.31%</td>
<td>50.44%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The anti-inflammatory activity of *Asplenium ceterach* could be strongly due to the richness of this plant on chlorogenic acid which represents 3/4 of the total content of phenolic compounds. Several studies have shown that chlorogenic acid has anti-inflammatory properties (15,16). Hwang et al. (17) found that chlorogenic acid significantly inhibited not only NO production but also the expression of cyclooxygenase-2 and NO synthase. This compound also attenuated pro-inflammatory cytokines such as IL-1β and TNF-α and other inflammation-related markers. Chlorogenic acid decreased also the endotoxin-induced adhesion of macrophages, the expression level of ninjurin1 (Ninj1), and the nuclear translocation of NF-κB.

d) Diuretic activity

The diuretic activity of *Asplenium ceterach*, tested on Wistar albino rats, is compared to those of the control (0.9% saline solution) and the reference (diuretic drug: Furosemide 40mg). The accumulated volumetric urinary excretion of *Asplenium ceterach* increases continuously during the 6 hours with a great intensity during the first 3 hours and then the excretion rate decreases slightly (Figure 1). For the control, the urinary excretion increases from the 1st to the 2nd hour then resumes continuously from the 3rd hour. Concerning the group treated with Furosemide, the excretion begins slightly; afterwards it becomes very intense between the 2nd and the 4th hours, and stops during the last two hours. The total urinary volumes indicate that the treatment with *Asplenium ceterach* infusion gave the highest diuretic activity, with a volume of 7.1ml, compared to the control (5.2ml) and the reference (5.7ml) which correspond to volumetric urinary excretions of 83.53, 69.32, and 76.00%, respectively.
The diuretic activity has been demonstrated by several other plants such as *Merremia emarginata* and *Hibiscus sabdariffa* which is due to the presence of chlorogenic acid (18,19). During the catabolism of this latter, hippuric acid was formed that can act as diuretic agent but the mechanism of this compounds still unclear (18).

### IV. Conclusions

This study concludes that *Asplenium ceterach* is characterized by a particular phenolic profile with the dominance of chlorogenic acid. Animal tests (mice and rats) have shown that this plant is endowed with interesting anti-inflammatory and diuretic properties.

### Acknowledgments

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### Conflicts of Interest

The authors declare that they have no conflicts of interest regarding this work.

### References Références Referencias


Phytochemical Analysis, Antimicrobial and Radical Scavenging Properties of Methanol Extracts of Dracaena Deisteliana (Dracaenaceae) and Sporobolus Indicus (Poaceae)

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Abstract- Dracaena deisteliana and Sporobolus indicus are medicinal plants with broad use in Cameroonian folk medicine to treat several infectious diseases. This study aimed to investigate the phytochemical composition, the antimicrobial and antiradical properties of methanol extracts of the leaves, stem and whole plant of D. deisteliana, and S. indicus. The phytochemical test was undertaken using standard methods. Agar well diffusion was used for sensitivity test while the microdilution method was used to determine the minimum inhibition concentrations (MICs) and the minimum bactericidal/fungicidal concentrations (MBCs/MFCs). The antiradical property of the plant extracts was performed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging assay.

Keywords: Dracaena deisteliana, Sporobolus indicus, phytochemical, antibacterial, antifungal, antiradical.

GJMR-B Classification: NLMC Code: QV 704
Phytochemical Analysis, Antimicrobial, and Radical Scavenging Properties of Methanol Extracts of *Dracaena Deisteliana* (Dracaenaceae) and *Sporobolus Indicus* (Poaceae)

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**Abstract** - *Dracaena deisteliana* and *Sporobolus indicus* are medicinal plants with broad use in Cameroonian folk medicine to treat several infectious diseases. This study aimed to investigate the phytochemical composition, the antimicrobial and antiradical properties of methanol extracts of the leaves, stem and whole plant of *D. deisteliana*, and *S. indicus*. The phytochemical test was carried out using standard methods. Agar well diffusion was used for sensitivity tests, while the minimum inhibitory concentrations (MICs) and the minimum bactericidal/fungicidal concentrations (MBCs/MFCs) were determined through microlitigation methods. The antiradical property of the plant extracts was investigated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging assay. The results revealed the presence of phenols, saponins, terpenoids, anthraquinones, alkaloids, and flavonoids in all the extracts. *D. deisteliana* extracts inhibited the growth of six bacteria strains with the inhibition zones varying from 8.50 ± 0.28 to 18.00 ± 0.00 mm and the MIC values ranging between 6.25 and 100 mg/mL. The leaf extract of *D. deisteliana* exhibited a higher effect on *Klebsiella oxytoca* and *Escherichia coli* with the MIC of 6.25 mg/mL. The stem and whole plant extracts showed similar activities on *Escherichia coli* and *Enterobacter cloacae*. *S. indicus* was most active on *Acinetobacter sp* and *Bacillus cereus*. *D. deisteliana* stem extract exhibited higher activity with EC₅₀ of 491 µg/mL, while *S. indicus* showed an EC₅₀ of 550.5 µg/mL. This study indicates that *D. deisteliana* and *S. indicus* possess antimicrobial and antiradical compounds and provides scientific evidence for their traditional uses to treat several infections.

**Keywords** - *Dracaena deisteliana*, *Sporobolus indicus*, phytochemical, antibacterial, antifungal, antiradical.

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**I. Background**

Infectious diseases remain a serious public health concern worldwide [1]. Despite the significant increase in the comprehension of the pathogenesis and management of infectious diseases, they remain one of the causes of mortality and morbidity, particularly in developing countries [2]. The indiscriminate use of antimicrobials and the poor management of infections lead to a new upsurge in the loss of drugs and the increase of resistant pathogenic microorganisms in recent years [3]. Approximately 700,000 people died yearly due to antibiotic resistance, and an estimated 10 million lives may be at risk by 2050 if nothing is done to solve the problem of antimicrobial resistance [4]. This situation increases the frequency of therapeutic failures and leads to economic liability, coupled with the undesired side effects of synthetic antimicrobials which complicate treatment [5].

During infection, highly reactive free radical and oxygen species are produced. This leads to high oxidative stress, which can provoke cancer, autoimmune, degenerative, and cardiovascular diseases [6]. Synthetic antioxidants widely used in cosmetics, food, and therapeutic industries are being restricted due to their carcinogenicity [7]. At the time, the current steroidal and non-steroidal anti-inflammatory drugs present adverse side effects [8]. The need to challenge these problems, coupled with the limited number of drugs, motivates the intensive searches for novel, effective, and affordable medicines from different sources [9].

Herbal products are extensively used in African traditional medicine to manage various illnesses [10]. Natural products from plants have been recognized as a reservoir of novel drugs with possible new mechanisms of action [11-13]. The use of medicinal plants is increasing worldwide, especially in advanced countries where many people rely on plants as primary healthcare modality due to limited access to modern medicine [15, 16].

*Dracaena deisteliana* belongs to the family of Dracaenaceae, which includes more than 480 species distributed, principally in tropical and sub-tropical...
regions [17]. The resin of *D. deisteliana* is used in Arab medicinal tradition to treat diarrhea, fracture, stomach, intestinal fever, and toothache [18]. In Cameroon, *D. deisteliana* leaf is used to treat infertility [19] and typhoid [20]. The stem is used to treat toothache [21]. In Nigeria, this plant is used to treat cough [22]. The pharmacological properties of *D. deisteliana* include the antileishmanias, anti molluscoidal, antimalarial, antibacterial, and antifungal activities [20, 23]. The phytochemical studies of *D. deisteliana* lead to the isolation of numerous compounds with biological properties [24, 25].

*S. indicus* belonging to the family of Poaceae is a perennial grass that grows in dense tufts. It is represented by approximately 45 species that generally grow in tropical and sub-tropical regions all over the world [26]. The sirup of *S. indicus* prepared with fruits is used to fight chronic diarrhea. The astringent bark decoction of *S. indicus* is a medicine against scabies, ulcers, and dysentery. The leaves and bark are used as a febrifuge [27]. However, phytochemical and biological activities of these plants are less or not investigated. Therefore, this study was undertaken to investigate the phytochemical composition, the antibacterial and antifungal, and the radical scavenging properties of *D. deisteliana* and *S. indicus*.

## II. Materials and Methods

### a) Collection and Identification of Plant Materials

*D. deisteliana* and *S. indicus* plant materials (Figure 1) were collected at Ngousso-Yaounde in the Centre region of Cameroon in April 2012 and May 2013, respectively. The plant identification was made at the Cameroon National Herbarium by comparison with specimen number 55004/HNC (*Dracaena deisteliana*) and 15719/SRF Cam (*Sporobolus indicus*).

![Figure 1: Photographs of selected plants: (A) Dracaena deisteliana and (B) Sporobolus indicus](image)

### b) Preparation of Methanol Extracts

The different parts and the whole plant of *D. deisteliana* and *S. indicus* were air-dried during two weeks at shade at room temperature. The samples were ground separately in a mortar, and 500 g of dried powder of each sample were soaked for 72 h in methanol (1:10 w/v) with constant stirrings. The resulting supernatant was filtered through Whatman no.1 filter paper and concentrated using a rotary evaporator at 55°C. The resultant extracts were transferred into pre-weighed labeled glass vials. The process was repeated twice on the marc to exhaustively extract the plant material. The extraction yield of each plant extract was determined by dividing the total extracted mass by dried plant mass used for extraction. Resultant extracts were air-dried and kept at 4°C for further use.

### c) Phytochemical Screening

Qualitative methods were used to determine different classes of phytochemicals (phenolic compound, tannins, saponins, alkaloids, anthocyanins, terpenoids, glycosides, cardiac glycosides, phlobatannins, and flavonoids), as previously described by Trease and Evans [28], and Sofowora [29].

### d) Antimicrobial Assays

#### i. Microbial Strains and Culture Media

Twelve bacteria including ten Gram-negative (*Enterobacter cloacae*, *Klebsiella oxytoca*, *Klebsiella pneumoniae*, *Morganella morganii*, *Bacillus cereus*, *Escherichia coli*, *Proteus vulgaris*, *Citrobacter freundii*, *Pseudomonas aeruginosa*, *Acinetobacter sp*) and two Gram-positive (*Staphylococcus aureus* and *Streptococcus faecalis*) and four yeasts (*Candida albicans*, *Candida kruzei*, *Candida parapsilosis*, and
**Gentamicin 1 mg/mL** (bacteria) and **Fluconazole 100 mg/mL** (fungi) were used as positive control. The Mueller Hinton broth (MHB) and Sabouraud dextrose broth (SDB) were used as liquid medium for the determination of the minimum inhibitory concentrations (MICs) and the minimum bactericidal/fungicidal concentrations (MBCs/MFCs). Gentamicin 1 mg/mL (bacteria) and Fluconazole 100 mg/mL (fungi) were used as positive control.

### ii. Preparation of Microbial Inoculum

The microbial inoculum was prepared using a direct colony suspension method. Suspensions of bacteria and yeasts were prepared in normal saline from 24 h grown on fresh MHA or SDA at 37°C. The bacterial suspension formed was adjusted with a spectrophotometer to a McFarland standard of 0.5, which is approximately 1.5 x 10⁸ CFU/mL. The turbidity of fungal strains was adjusted to a standard of 0.9 to give 1-5 x 10⁷ CFU/mL. Each suspension was then diluted 1:100 by transferring 0.1 mL of the bacterial suspension to 9.9 mL of sterile MHB while preparing for experiments [30].

### iii. Agar Well Diffusion Assay for Antibacterial Screening

The antibacterial activity was performed using the agar well diffusion method according to the modified Kirby Bauer diffusion technique [31]. The agar plates were swabbed with overnight bacterial suspensions of each strain. Then, wells were bored into the agar medium with heat sterilized 6 mm cork borer. A 75 µL of the methanolic extracts (100 mg/mL) was dispensed into the wells, and the plates were left for 30 min before being incubated for 24 h at 37 °C. Each zone of inhibition around the wells was measured using a vernier caliper.

### iv. Determination of the minimum inhibitory concentrations (MICs) and minimum bactericidal/fungicidal concentrations (MBCs/MFCs) of the plant extracts

The MICs values of the plant extracts for bacteria and yeasts were determined using serial dilution microplate methods [32, 33]. Two-fold serial dilution of the extract (dissolved in MHB or SDB) was made in a 96-wells microplate for a final concentration ranging from 100 to 97.65 x 10⁻³ mg/mL. An equal volume (100 µL) of the 1.5 x 10⁻⁶ CFU/mL bacterial inoculum or 1-5 x 10⁻⁶ CFU/mL fungal inoculum prepared in MHB or SDB was then added. The plates were covered with a sterile plate sealer and then incubated for 24 h at 37°C (48 h for fungi). After incubation, 40 µL of 2,3,5-triphenyltetrazolium chloride 0.01 % w/v (TTC) was added in each well of the plates and incubated for 30 min at 37°C. The MIC, defined as the lowest sample concentration that prevented the growth of the bacteria, was then detected by any observed color change. The MBCs/MFCs of each fraction were determined by sub-culturing the sample (50 µL) taken from the wells without growth during MIC determination to 150 µL of MHB or SDB. The plates were incubated at 37°C for 48 h (72 h for fungi). The MBC (or MFC) was regarded as the lowest concentration of extracts with the absence of growth that prevented the color change of the medium after the addition of TTC as mentioned above.

### e) Free Radical Scavenging Assay

The DPPH (2,2-diphenyl-1-picrylhydrazyl) radical-scavenging potential of *D. deisteliana* and *S. indicus* extracts was determined following a modified method of Brand-Williams et al. [34]. A 10 µL of each extract prepared in methanol at different concentrations was added into 1990 µL of DPPH solution (0.04 mg/mL) in different tubes for final concentrations of 5 µg/mL; 10 µg/mL; 15 µg/mL; 20 µg/mL; 25 µg/mL; 30 µg/mL. After vortexing, the tubes were kept in the darkness at room temperature for 30 min. The absorbance at 517 nm was taken. The percentage inhibition was calculated from [(A₀-A₁)/A₀] x 100, where A₀ is the absorbance of the control at 30 min (DPPH solution), and A₁ is the absorbance of the extract/reference. Ascorbic acid was used as a reference. The inhibition curves were prepared, and EC₅₀ (Efficient concentration of the sample (g) to scavenge 50 % of the DPPH free radical) values were calculated.

### f) Statistical Analysis

Data were represented as mean ± standard deviation (SD) of three replicates and subjected to one way analysis of variance (ANOVA) using the Fisher test at the threshold of *p* < 0.05 with Stat graphics plus 5.0 for windows. Linear regression analysis was used to calculate EC₅₀ values. Microsoft Excel was used to enter and capture data.

### III. Results and Discussion

#### a) Extraction Yields of the Plant Extracts

The plant material was extracted using methanol as solvent. The highest yield was obtained with *D. deisteliana* leaf extract (8.89 %). The least yield of extraction was obtained with *S. indicus* extract (3.46 %) (Table 1). It has been shown that the type of solvent used in extraction procedure determines the success of isolated compounds from the plant material [35]. The yield of extraction of the stem extract of *D. deisteliana* was 5.63, which is higher than 0.95 previously obtained by Kougan et al. [25].
Phytochemical Analysis, Antimicrobial and Radical Scavenging Properties of Methanol Extracts of *Dracaena deisteliana* (Dracaenaceae) and *Sporobolus indicus* (Poaceae)

### Table 1: Yield percentage (%) of different extracts of plants used in the study

<table>
<thead>
<tr>
<th>Plant</th>
<th>Part used</th>
<th>Solvent used</th>
<th>Yield of extraction (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>D. deisteliana</em></td>
<td>Leaf</td>
<td>Methanol</td>
<td>8.89</td>
</tr>
<tr>
<td></td>
<td>Stem</td>
<td>Methanol</td>
<td>5.63</td>
</tr>
<tr>
<td></td>
<td>Whole plant</td>
<td>Methanol</td>
<td>6.37</td>
</tr>
<tr>
<td><em>S. indicus</em></td>
<td>Whole plant</td>
<td>Methanol</td>
<td>3.46</td>
</tr>
</tbody>
</table>

b) **Phytochemical Screening**

The results of the phytochemical screening carried out with the crude extracts of *D. deisteliana* and *S. indicus* are presented in Table 2. Results showed that all extracts are rich in phenols, saponins, anthraquinones, and alkaloids. Terpenoids and flavonoids were present at a moderate level in the stem and whole plant extracts of *D. deisteliana*, while abundant amount was found in the leaf extract. It has been shown that plants belonging to the genus Dracaena contain steroidal saponins and flavonoids [36, 37]. The phytochemical investigations of *D. deisteliana* leaf extract by Kougan et al. [25] reported the presence of steroidal saponins and saponins. Anthocyanins and tannins were found in the crude extracts of *S. indicus*. In a previous study, it has been reported that *S. indicus* is rich in tannins [27]. Several classes of secondary metabolites found in these plant extracts have been reported to possess antimicrobial activities [38-40].

### Table 2: Phytochemical composition of *D. deisteliana* and *S. indicus* extracts

<table>
<thead>
<tr>
<th>Plant constituents</th>
<th><em>D. deisteliana</em></th>
<th><em>S. indicus</em></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Leaf</td>
<td>Stem</td>
</tr>
<tr>
<td>Phenolic</td>
<td>Ferric chloride test</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Potassium dichromate</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>Ferric chloride test</td>
<td>-</td>
</tr>
<tr>
<td>Anthocyanins</td>
<td>Ammonia HCl test</td>
<td>-</td>
</tr>
<tr>
<td>Saponins</td>
<td>Frothing test</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>Alkaline reagent test</td>
<td>+++</td>
</tr>
<tr>
<td></td>
<td>Lead acetate test</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>Tannic acid test</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Mayer’s test</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Wagner’s test</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>Salowski test</td>
<td>+++</td>
</tr>
<tr>
<td>Anthraquinones</td>
<td>Borntrager’s test</td>
<td>+</td>
</tr>
<tr>
<td>Glycosides (Free sugar)</td>
<td>Legal’s test</td>
<td>-</td>
</tr>
<tr>
<td>Cardiac glycosides</td>
<td>Killer Killani test</td>
<td>-</td>
</tr>
<tr>
<td>Phlobatanins</td>
<td>Hydrochloride test</td>
<td>-</td>
</tr>
</tbody>
</table>

+++ = abundant; ++ = moderate; + present; - completely absent

c) **Diameter of Inhibition Zones**

The presence of inhibition zones after incubation showed that the Gram-positive, Gram-negative, and fungi isolates exhibited a varied degree of susceptibility to each of the plant extracts that can be considered as a plant with a broad spectrum of activity (Table 3). Considering the susceptibility of the isolates to *D. deisteliana*, the inhibition zones ranged between 8.5 ± 0.28 (*K. pneumoniae*) and 15 ± 0.57 mm (*K. oxytoca* and *E. coli*) for the leaf extract while no activity was noted on *B. cereus* and *P. aeruginosa*. In the previous report, the leaf extract of *D. deisteliana* exhibited the inhibition zones of 8.5 ± 0.00 mm (80 mg/mL) and 12 ± 0.00 mm (160 mg/mL) on *S. typhi* [20]. The stem extract...
was less active on \( \text{K. pneumoniae} \) with inhibition zone of 11 ± 0.00 mm and exhibited higher activity on \( \text{E. cloacae} \) with inhibition zone of 18 ± 0.00 mm. The inhibition zones varied between 9.5 ± 0.28 (\( \text{K. pneumoniae} \)) and 16.5 ± 0.28 mm (\( \text{E. cloacae} \)) for the whole plant extract. This study provides additional data on the antimicrobial activity of \( \text{D. deisteliana} \). The whole extract of \( \text{S. indicus} \) exhibited inhibition zones varying between 7 ± 0.00 (\( \text{E. coli} \)) and 14 ± 0.43 mm (\( \text{Acinetobacter spp} \)). These results revealed for the first time information on the antimicrobial properties of \( \text{S. indicus} \). Nevertheless, the antibacterial activity of both the plant extracts was less pronounced compared to the standard antibiotic (gentamicin) with inhibition zones varying between 21 ± 00 and 29.67 ± 0.88 mm.

**Table 3: Inhibition zone (mm) of plant extracts against some bacteria species**

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Leaf</th>
<th>Stem</th>
<th>Whole plant</th>
<th>( \text{S. indicus} )</th>
<th>Gentamicin</th>
</tr>
</thead>
<tbody>
<tr>
<td>( \text{E. cloacae} )</td>
<td>12 ± 1a</td>
<td>18 ± 0.0b</td>
<td>16.5 ± 0.5c</td>
<td>9 ± 0.0d</td>
<td>28 ± 0.0d</td>
</tr>
<tr>
<td>( \text{K. pneumoniae} )</td>
<td>8.5 ± 0.5a</td>
<td>11 ± 0.0b</td>
<td>9.5 ± 0.5a</td>
<td>0 ± 0.0b</td>
<td>30 ± 0.0d</td>
</tr>
<tr>
<td>( \text{S. faecalis} )</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>25 ± 0.0d</td>
</tr>
<tr>
<td>( \text{S. aureus} )</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>27 ± 0.0d</td>
</tr>
<tr>
<td>( \text{K. oxytoca} )</td>
<td>15a</td>
<td>11 ± 1b</td>
<td>14 ± 2a</td>
<td>0 ± 0.0a</td>
<td>27 ± 0.0d</td>
</tr>
<tr>
<td>( \text{Acinetobacter sp} )</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>14 ± 0.75b</td>
<td>23.33 ± 0.57c</td>
</tr>
<tr>
<td>( \text{E. coli} )</td>
<td>15 ± 1a</td>
<td>13 ± 1a</td>
<td>18 ± 0.0b</td>
<td>7 ± 0.0b</td>
<td>23.33 ± 0.57c</td>
</tr>
<tr>
<td>( \text{P. vulgaris} )</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>8 ± 0.0b</td>
<td>21 ± 0.0d</td>
</tr>
<tr>
<td>( \text{P. aeruginosa} )</td>
<td>0 ± 0.0a</td>
<td>12.5 ± 0.5b</td>
<td>11 ± 0.0b</td>
<td>9 ± 0.0a</td>
<td>29.67 ± 0.57c</td>
</tr>
<tr>
<td>( \text{M. morganii} )</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>7 ± 1b</td>
<td>26.67 ± 1.15c</td>
</tr>
<tr>
<td>( \text{C. freundii} )</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>0 ± 0.0a</td>
<td>12 ± 0.0b</td>
<td>23 ± 0.0d</td>
</tr>
<tr>
<td>( \text{B. cereus} )</td>
<td>0 ± 0.0a</td>
<td>12.5 ± 1.5b</td>
<td>14 ± 1b</td>
<td>12.0 ± 0.0b</td>
<td>23 ± 0.0d</td>
</tr>
</tbody>
</table>

The results are expressed as means ± standard deviation of three determinations. Values with different letters in the same line are significantly different at \( p<0.05 \).

d) Minimum Inhibitory Concentrations (MICs) and Minimum Bactericidal/Fungicidal Concentrations (MBCs/MFCs)

In this study, the extracts obtained from both the plant extracts displayed varying antimicrobial activities according to their MICs (6.25-50 mg/mL for bacteria and 1.56-50 mg/mL for yeasts) as reported in Table 4. The leaf and the whole plant extracts of \( \text{D. deisteliana} \) were the most active (MIC = 6.25 mg/mL) against \( \text{E. cloacae} \) and \( \text{E. coli} \) (Gram-negative) while both bacteria had the same degree of susceptibility (MIC = 50 mg/mL) to the stem extract. The whole extract of \( \text{S. indicus} \) (MIC = 6.25 mg/mL) had remarkable activity against \( \text{B. cereus} \) (Gram-positive). The sensitivity of Gram-negative and Gram-positive bacteria could be due to the difference in their membrane morphology [41]. The phospholipidic bilayer of the outer membrane of bacteria is the target of interactions with antimicrobial compounds. Damages on the bacterial membrane could increase permeability to ions, the release of intracellular constituents, deterioration of the enzymatic system of bacteria, and even death [42, 43]. All the tested extracts of \( \text{D. deisteliana} \) were most active on \( \text{C. albicans} \) and \( \text{C. krusei} \) (MIC = 1.56 µg/mL). A similar activity was observed with the leaf and the whole-plant extracts on \( \text{C. parapsilosis} \). \( \text{C. neofarmans} \) had the least susceptibility (MIC = 50 µg/mL) to all the extracts of \( \text{D. deisteliana} \). The antimicrobial activity can be classified as interesting (CMI < 100 µg/mL), moderate (100 < CMI < 625 µg/mL) and weak (CMI > 625 µg/mL) [44, 45]. Therefore, all the plant extracts have weak activity on the tested microorganisms. The weak antibacterial activity exhibited by all the plant extracts could be correlated to the few amounts of secondary metabolites since it has been proven that the concentration, the nature, and the origin of active compounds present in plant extracts may influence the antimicrobial activity [40, 46]. The antimicrobial mechanism of active ingredients may vary with species, chemical composition, cell wall composition, and genetic material of each microorganism [38, 41, 47].

According to Mims et al. [48], the leaves extracts of \( \text{D. deisteliana} \) had a bactericidal effect on \( \text{S. aureus} \), \( \text{K. pneumoniae} \), \( \text{B. cereus} \), and \( \text{K. oxytoca} \). In comparison, the stem extracts had bacteriostatic effect on \( \text{S. aureus} \), \( \text{K. pneumoniae} \), \( \text{E. cloacae} \), and \( \text{E. coli} \). The whole-plant extract of \( \text{D. deisteliana} \) exhibited a bactericidal action on \( \text{S. aureus} \), \( \text{K. pneumoniae} \), and \( \text{B. cereus} \). At the same time, the bacteriostatic effect was observed on \( \text{K. oxytoca} \), \( \text{E. cloacae} \), and \( \text{E. coli} \). The whole-plant extract of \( \text{S. indicus} \) exhibited a bacteriostatic effect on \( \text{K. pneumoniae} \), \( \text{E. coli} \), \( \text{B. cereus} \), and \( \text{P. vulgaris} \) while the bactericidal effect was observed on the rest. All the extracts of \( \text{D. deisteliana} \) have a bactericidal effect on all the yeast strains used in this study.
### Table 4: Inhibition parameters (MICs, MBC/MFCs) of methanol extracts from *D. deisteliana* and *S. indicus* (mg/mL) and reference drugs (µg/mL)

<table>
<thead>
<tr>
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<th>( S. \text{ indicus} )</th>
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<td></td>
<td></td>
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<td>50</td>
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<tr>
<td></td>
<td>MBC</td>
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<td></td>
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<td>MBC</td>
<td>100</td>
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<td>MFC/MIC</td>
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Legend: MIC = Minimal Inhibitory Concentration; MBC/MFC = Minimal Bactericidal/Fungicidal Concentration; Ref* = reference drugs: gentamicin (for bacteria) and fluconazole (for yeasts); / = Not determined
e) Antiradical Properties of Plant Extracts

The free radicals scavenging properties of the plant extracts are reported in Figure 2. The crude extracts of *D. deisteliana* and *S. indicus* exhibited radical scavenging properties in concentration-dependent manners. The inhibition percentages of the stem (26 %) and the whole plant extract of *D. deisteliana* (28 %) and *S. indicus* (24 %) were most pronounced than that of the leaf extract of *D. deisteliana* (14 %) at the concentration of 1 mg/mL. The higher inhibition percentage was observed with the stem extract of *D. deisteliana* (62 %) at the concentration of 5.5 mg/mL. It can be observed that the DPPH activity *D. deisteliana* and *S. indicus* were found to be increasing in concentration-dependent manner.

*Figure 2:* DPPH free radical scavenging activities of plant extracts. L: leaf; S: stem; WP: whole plant.

From each graph, the EC_{50} of each extract was determined. The EC_{50} is the concentration of the samples, which scavenges 50 % of free radicals. Figure 3 shows the scavenging activity of the crude extracts of *D. deisteliana* and *S. indicus* in comparison with that of ascorbic acid. The EC_{50} obtained showed that among the crude extracts of *D. deisteliana*, the leaf extract exhibited the lowest activity (646.75 µg/mL), while the stem extract had the higher one (491 µg/mL). The whole plant extract of *S. indicus* had an EC_{50} of 550.5 µg/mL, while the EC_{50} value of the standard was found to be 411 µg/mL. Numerous previous studies show the correlation between antiradical activity and the phenolic compounds [49]. These studies have confirmed that the phenolic compounds contribute significantly to the antioxidant activity [50]. The antiradical activity depends on the content in phenolic compounds that give up hydrogen to the free radicals and interrupt the chain of lipid oxidative reaction in the first step of inhibition [51]. This higher efficiency of the phenolic compounds to scavenge free radicals like singlet oxygen, superoxide, and hydroxyl radicals is due to their hydroxyl phenolic group [52]. Flavonoids and tannins found in these plant extracts possess a large spectrum of antiradical properties [53]. However, these activities may be due to the synergistic action of the chemical compounds presents in the extracts [54].
IV. CONCLUSION

Overall, the phytochemical screening of the crude extracts of *D. deisteliana* and *S. indicus* revealed the presence of several classes of secondary metabolites with known antimicrobial and antioxidant activities. The three extracts of *D. deisteliana* have bactericidal activity on *K. pneumoniae* and *S. aureus* and all the fungi tested. *S. indicus* have bactericidal activities on all the bacteria tested. All the plant extracts exhibited weak antibacterial activity. Nevertheless, these results support their traditional uses for the treatment of infections. *D. deisteliana* and *S. indicus* possess' important antiradical activities and can be used to scavenge free radicals. Further studies are still required, namely to isolate active ingredient from these plants to increase their activities and elucidate their potential mechanism of action.

**Ethics approval and consent to participate**
Not applicable

**Consent for publication**
Not applicable

**Availability of data and material**
The datasets used and analyzed during the current study are available from the corresponding authors on reasonable request.

**Competing interests**
The authors declare that they have no competing interests.

**Funding**
None

**Author’s contributions**
RNK and CTM designed the study, supervised experiments, and critically revised the paper and intellectual content. ADA carried out the experiments, analyze data, and wrote the manuscript. All authors read and approve the final manuscript.

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Preferred Author Guidelines

We accept the manuscript submissions in any standard (generic) format.

We typeset manuscripts using advanced typesetting tools like Adobe In Design, CorelDraw, TeXnicCenter, and TeXStudio. We usually recommend authors submit their research using any standard format they are comfortable with, and let Global Journals do the rest.

Alternatively, you can download our basic template from https://globaljournals.org/Template

Authors should submit their complete paper/article, including text illustrations, graphics, conclusions, artwork, and tables. Authors who are not able to submit manuscript using the form above can email the manuscript department at submit@globaljournals.org or get in touch with chiefeditor@globaljournals.org if they wish to send the abstract before submission.

Before and during Submission

Authors must ensure the information provided during the submission of a paper is authentic. Please go through the following checklist before submitting:

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2. Authors must accept the privacy policy, terms, and conditions of Global Journals.
3. Ensure corresponding author’s email address and postal address are accurate and reachable.
4. Manuscript to be submitted must include keywords, an abstract, a paper title, co-author(s’) names and details (email address, name, phone number, and institution), figures and illustrations in vector format including appropriate captions, tables, including titles and footnotes, a conclusion, results, acknowledgments and references.
5. Authors should submit paper in a ZIP archive if any supplementary files are required along with the paper.
6. Proper permissions must be acquired for the use of any copyrighted material.
7. Manuscript submitted must not have been submitted or published elsewhere and all authors must be aware of the submission.

Declaration of Conflicts of Interest

It is required for authors to declare all financial, institutional, and personal relationships with other individuals and organizations that could influence (bias) their research.

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- Findings
- Writings
- Diagrams
- Graphs
- Illustrations
- Lectures
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2. Drafting the paper and revising it critically regarding important academic content.
3. Final approval of the version of the paper to be published.

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The corresponding author should mention the name and complete details of all co-authors during submission and in manuscript. We support addition, rearrangement, manipulation, and deletions in authors list till the early view publication of the journal. We expect that corresponding author will notify all co-authors of submission. We follow COPE guidelines for changes in authorship.

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Unless specified in the notification, the Editorial Board’s decision on publication of the paper is final and cannot be appealed before making the major change in the manuscript.

Acknowledgments

Contributors to the research other than authors credited should be mentioned in Acknowledgments. The source of funding for the research can be included. Suppliers of resources may be mentioned along with their addresses.

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Global Journals is in partnership with various universities, laboratories, and other institutions worldwide in the research domain. Authors are requested to disclose their source of funding during every stage of their research, such as making analysis, performing laboratory operations, computing data, and using institutional resources, from writing an article to its submission. This will also help authors to get reimbursements by requesting an open access publication letter from Global Journals and submitting to the respective funding source.

Preparing your Manuscript

Authors can submit papers and articles in an acceptable file format: MS Word (doc, docx), LaTeX (.tex, .zip or .rar including all of your files), Adobe PDF (.pdf), rich text format (.rtf), simple text document (.txt), Open Document Text (.odt), and Apple Pages (.pages). Our professional layout editors will format the entire paper according to our official guidelines. This is one of the highlights of publishing with Global Journals—authors should not be concerned about the formatting of their paper. Global Journals accepts articles and manuscripts in every major language, be it Spanish, Chinese, Japanese, Portuguese, Russian, French, German, Dutch, Italian, Greek, or any other national language, but the title, subtitle, and abstract should be in English. This will facilitate indexing and the pre-peer review process.

The following is the official style and template developed for publication of a research paper. Authors are not required to follow this style during the submission of the paper. It is just for reference purposes.
Manuscript Style Instruction (Optional)

- Microsoft Word Document Setting Instructions.
- Font type of all text should be Swis721 Lt BT.
- Page size: 8.27” x 11”", left margin: 0.65, right margin: 0.65, bottom margin: 0.75.
- Paper title should be in one column of font size 24.
- Author name in font size of 11 in one column.
- Abstract: font size 9 with the word “Abstract” in bold italics.
- Main text: font size 10 with two justified columns.
- Two columns with equal column width of 3.38 and spacing of 0.2.
- First character must be three lines drop-capped.
- The paragraph before spacing of 1 pt and after of 0 pt.
- Line spacing of 1 pt.
- Large images must be in one column.
- The names of first main headings (Heading 1) must be in Roman font, capital letters, and font size of 10.
- The names of second main headings (Heading 2) must not include numbers and must be in italics with a font size of 10.

Structure and Format of Manuscript

The recommended size of an original research paper is under 15,000 words and review papers under 7,000 words. Research articles should be less than 10,000 words. Research papers are usually longer than review papers. Review papers are reports of significant research (typically less than 7,000 words, including tables, figures, and references)

A research paper must include:

a) A title which should be relevant to the theme of the paper.
b) A summary, known as an abstract (less than 150 words), containing the major results and conclusions.
c) Up to 10 keywords that precisely identify the paper’s subject, purpose, and focus.
d) An introduction, giving fundamental background objectives.
e) Resources and techniques with sufficient complete experimental details (wherever possible by reference) to permit repetition, sources of information must be given, and numerical methods must be specified by reference.
f) Results which should be presented concisely by well-designed tables and figures.
g) Suitable statistical data should also be given.
h) All data must have been gathered with attention to numerical detail in the planning stage.

Design has been recognized to be essential to experiments for a considerable time, and the editor has decided that any paper that appears not to have adequate numerical treatments of the data will be returned unrefereed.

i) Discussion should cover implications and consequences and not just recapitulate the results; conclusions should also be summarized.
j) There should be brief acknowledgments.
k) There ought to be references in the conventional format. Global Journals recommends APA format.

Authors should carefully consider the preparation of papers to ensure that they communicate effectively. Papers are much more likely to be accepted if they are carefully designed and laid out, contain few or no errors, are summarizing, and follow instructions. They will also be published with much fewer delays than those that require much technical and editorial correction.

The Editorial Board reserves the right to make literary corrections and suggestions to improve brevity.
It is necessary that authors take care in submitting a manuscript that is written in simple language and adheres to published guidelines.

All manuscripts submitted to Global Journals should include:

**Title**
The title page must carry an informative title that reflects the content, a running title (less than 45 characters together with spaces), names of the authors and co-authors, and the place(s) where the work was carried out.

**Author details**
The full postal address of any related author(s) must be specified.

**Abstract**
The abstract is the foundation of the research paper. It should be clear and concise and must contain the objective of the paper and inferences drawn. It is advised to not include big mathematical equations or complicated jargon.

Many researchers searching for information online will use search engines such as Google, Yahoo or others. By optimizing your paper for search engines, you will amplify the chance of someone finding it. In turn, this will make it more likely to be viewed and cited in further works. Global Journals has compiled these guidelines to facilitate you to maximize the web-friendliness of the most public part of your paper.

**Keywords**
A major lynchpin of research work for the writing of research papers is the keyword search, which one will employ to find both library and internet resources. Up to eleven keywords or very brief phrases have to be given to help data retrieval, mining, and indexing.

One must be persistent and creative in using keywords. An effective keyword search requires a strategy: planning of a list of possible keywords and phrases to try.

Choice of the main keywords is the first tool of writing a research paper. Research paper writing is an art. Keyword search should be as strategic as possible.

One should start brainstorming lists of potential keywords before even beginning searching. Think about the most important concepts related to research work. Ask, “What words would a source have to include to be truly valuable in a research paper?” Then consider synonyms for the important words.

It may take the discovery of only one important paper to steer in the right keyword direction because, in most databases, the keywords under which a research paper is abstracted are listed with the paper.

**Numerical Methods**
Numerical methods used should be transparent and, where appropriate, supported by references.

**Abbreviations**
Authors must list all the abbreviations used in the paper at the end of the paper or in a separate table before using them.

**Formulas and equations**
Authors are advised to submit any mathematical equation using either MathJax, KaTeX, or LaTeX, or in a very high-quality image.

**Tables, Figures, and Figure Legends**
Tables: Tables should be cautiously designed, uncrowned, and include only essential data. Each must have an Arabic number, e.g., Table 4, a self-explanatory caption, and be on a separate sheet. Authors must submit tables in an editable format and not as images. References to these tables (if any) must be mentioned accurately.
Figures

Figures are supposed to be submitted as separate files. Always include a citation in the text for each figure using Arabic numbers, e.g., Fig. 4. Artwork must be submitted online in vector electronic form or by emailing it.

Preparation of Electronic Figures for Publication

Although low-quality images are sufficient for review purposes, print publication requires high-quality images to prevent the final product being blurred or fuzzy. Submit (possibly by e-mail) EPS (line art) or TIFF (halftone/photographs) files only. MS PowerPoint and Word Graphics are unsuitable for printed pictures. Avoid using pixel-oriented software. Scans (TIFF only) should have a resolution of at least 350 dpi (halftone) or 700 to 1100 dpi (line drawings). Please give the data for figures in black and white or submit a Color Work Agreement form. EPS files must be saved with fonts embedded (and with a TIFF preview, if possible).

For scanned images, the scanning resolution at final image size ought to be as follows to ensure good reproduction: line art: >650 dpi; halftones (including gel photographs): >350 dpi; figures containing both halftone and line images: >650 dpi.

Color charges: Authors are advised to pay the full cost for the reproduction of their color artwork. Hence, please note that if there is color artwork in your manuscript when it is accepted for publication, we would require you to complete and return a Color Work Agreement form before your paper can be published. Also, you can email your editor to remove the color fee after acceptance of the paper.

Tips for Writing a Good Quality Medical Research Paper

1. Choosing the topic: In most cases, the topic is selected by the interests of the author, but it can also be suggested by the guides. You can have several topics, and then judge which you are most comfortable with. This may be done by asking several questions of yourself, like "Will I be able to carry out a search in this area? Will I find all necessary resources to accomplish the search? Will I be able to find all information in this field area?" If the answer to this type of question is "yes," then you ought to choose that topic. In most cases, you may have to conduct surveys and visit several places. Also, you might have to do a lot of work to find all the rises and falls of the various data on that subject. Sometimes, detailed information plays a vital role, instead of short information. Evaluators are human: The first thing to remember is that evaluators are also human beings. They are not only meant for rejecting a paper. They are here to evaluate your paper. So present your best aspect.

2. Think like evaluators: If you are in confusion or getting demotivated because your paper may not be accepted by the evaluators, then think, and try to evaluate your paper like an evaluator. Try to understand what an evaluator wants in your research paper, and you will automatically have your answer. Make blueprints of paper: The outline is the plan or framework that will help you to arrange your thoughts. It will make your paper logical. But remember that all points of your outline must be related to the topic you have chosen.

3. Ask your guides: If you are having any difficulty with your research, then do not hesitate to share your difficulty with your guide (if you have one). They will surely help you out and resolve your doubts. If you can't clarify what exactly you require for your work, then ask your supervisor to help you with an alternative. He or she might also provide you with a list of essential readings.

4. Use of computer is recommended: As you are doing research in the field of medical research then this point is quite obvious. Use right software: Always use good quality software packages. If you are not capable of judging good software, then you can lose the quality of your paper unknowingly. There are various programs available to help you which you can get through the internet.

5. Use the internet for help: An excellent start for your paper is using Google. It is a wondrous search engine, where you can have your doubts resolved. You may also read some answers for the frequent question of how to write your research paper or find a model research paper. You can download books from the internet. If you have all the required books, place importance on reading, selecting, and analyzing the specified information. Then sketch out your research paper. Use big pictures: You may use encyclopedias like Wikipedia to get pictures with the best resolution. At Global Journals, you should strictly follow here.
6. Bookmarks are useful: When you read any book or magazine, you generally use bookmarks, right? It is a good habit which helps to not lose your continuity. You should always use bookmarks while searching on the internet also, which will make your search easier.

7. Revise what you wrote: When you write anything, always read it, summarize it, and then finalize it.

8. Make every effort: Make every effort to mention what you are going to write in your paper. That means always have a good start. Try to mention everything in the introduction—what is the need for a particular research paper. Polish your work with good writing skills and always give an evaluator what he wants. Make backups: When you are going to do any important thing like making a research paper, you should always have backup copies of it either on your computer or on paper. This protects you from losing any portion of your important data.

9. Produce good diagrams of your own: Always try to include good charts or diagrams in your paper to improve quality. Using several unnecessary diagrams will degrade the quality of your paper by creating a hodgepodge. So always try to include diagrams which were made by you to improve the readability of your paper. Use of direct quotes: When you do research relevant to literature, history, or current affairs, then use of quotes becomes essential, but if the study is relevant to science, use of quotes is not preferable.

10. Use proper verb tense: Use proper verb tenses in your paper. Use past tense to present those events that have happened. Use present tense to indicate events that are going on. Use future tense to indicate events that will happen in the future. Use of wrong tenses will confuse the evaluator. Avoid sentences that are incomplete.

11. Pick a good study spot: Always try to pick a spot for your research which is quiet. Not every spot is good for studying.

12. Know what you know: Always try to know what you know by making objectives, otherwise you will be confused and unable to achieve your target.

13. Use good grammar: Always use good grammar and words that will have a positive impact on the evaluator; use of good vocabulary does not mean using tough words which the evaluator has to find in a dictionary. Do not fragment sentences. Eliminate one-word sentences. Do not ever use a big word when a smaller one would suffice. Verbs have to be in agreement with their subjects. In a research paper, do not start sentences with conjunctions or finish them with prepositions. When writing formally, it is advisable to never split an infinitive because someone will (wrongly) complain. Avoid clichés like a disease. Always shun irritating alliteration. Use language which is simple and straightforward. Put together a neat summary.

14. Arrangement of information: Each section of the main body should start with an opening sentence, and there should be a changeover at the end of the section. Give only valid and powerful arguments for your topic. You may also maintain your arguments with records.

15. Never start at the last minute: Always allow enough time for research work. Leaving everything to the last minute will degrade your paper and spoil your work.

16. Multitasking in research is not good: Doing several things at the same time is a bad habit in the case of research activity. Research is an area where everything has a particular time slot. Divide your research work into parts, and do a particular part in a particular time slot.

17. Never copy others’ work: Never copy others’ work and give it your name because if the evaluator has seen it anywhere, you will be in trouble. Take proper rest and food: No matter how many hours you spend on your research activity, if you are not taking care of your health, then all your efforts will have been in vain. For quality research, take proper rest and food.

18. Go to seminars: Attend seminars if the topic is relevant to your research area. Utilize all your resources.

19. Refresh your mind after intervals: Try to give your mind a rest by listening to soft music or sleeping in intervals. This will also improve your memory. Acquire colleagues: Always try to acquire colleagues. No matter how sharp you are, if you acquire colleagues, they can give you ideas which will be helpful to your research.
20. **Think technically:** Always think technically. If anything happens, search for its reasons, benefits, and demerits. Think and then print: When you go to print your paper, check that tables are not split, headings are not detached from their descriptions, and page sequence is maintained.

21. **Adding unnecessary information:** Do not add unnecessary information like "I have used MS Excel to draw graphs." Irrelevant and inappropriate material is superfluous. Foreign terminology and phrases are not apropos. One should never take a broad view. Analogy is like feathers on a snake. Use words properly, regardless of how others use them. Remove quotations. Puns are for kids, not grunt readers. Never oversimplify: When adding material to your research paper, never go for oversimplification; this will definitely irritate the evaluator. Be specific. Never use rhythmic redundancies. Constractions shouldn’t be used in a research paper. Comparisons are as terrible as clichés. Give up ampersands, abbreviations, and so on. Remove commas that are not necessary. Parenthetical words should be between brackets or commas. Understatement is always the best way to put forward earth-shaking thoughts. Give a detailed literary review.

22. **Report concluded results:** Use concluded results. From raw data, filter the results, and then conclude your studies based on measurements and observations taken. An appropriate number of decimal places should be used. Parenthetical remarks are prohibited here. Proofread carefully at the final stage. At the end, give an outline to your arguments. Spot perspectives of further study of the subject. Justify your conclusion at the bottom sufficiently, which will probably include examples.

23. **Upon conclusion:** Once you have concluded your research, the next most important step is to present your findings. Presentation is extremely important as it is the definite medium though which your research is going to be in print for the rest of the crowd. Care should be taken to categorize your thoughts well and present them in a logical and neat manner. A good quality research paper format is essential because it serves to highlight your research paper and bring to light all necessary aspects of your research.

**Informal Guidelines of Research Paper Writing**

**Key points to remember:**
- Submit all work in its final form.
- Write your paper in the form which is presented in the guidelines using the template.
- Please note the criteria peer reviewers will use for grading the final paper.

**Final points:**

One purpose of organizing a research paper is to let people interpret your efforts selectively. The journal requires the following sections, submitted in the order listed, with each section starting on a new page:

**The introduction:** This will be compiled from reference matter and reflect the design processes or outline of basis that directed you to make a study. As you carry out the process of study, the method and process section will be constructed like that. The results segment will show related statistics in nearly sequential order and direct reviewers to similar intellectual paths throughout the data that you gathered to carry out your study.

**The discussion section:**

This will provide understanding of the data and projections as to the implications of the results. The use of good quality references throughout the paper will give the effort trustworthiness by representing an alertness to prior workings.

Writing a research paper is not an easy job, no matter how trouble-free the actual research or concept. Practice, excellent preparation, and controlled record-keeping are the only means to make straightforward progression.

**General style:**

Specific editorial column necessities for compliance of a manuscript will always take over from directions in these general guidelines.

**To make a paper clear:** Adhere to recommended page limits.
**Mistakes to avoid:**

- Insertion of a title at the foot of a page with subsequent text on the next page.
- Separating a table, chart, or figure—confine each to a single page.
- Submitting a manuscript with pages out of sequence.
- In every section of your document, use standard writing style, including articles (“a” and “the”).
- Keep paying attention to the topic of the paper.
- Use paragraphs to split each significant point (excluding the abstract).
- Align the primary line of each section.
- Present your points in sound order.
- Use present tense to report well-accepted matters.
- Use past tense to describe specific results.
- Do not use familiar wording; don’t address the reviewer directly. Don’t use slang or superlatives.
- Avoid use of extra pictures—include only those figures essential to presenting results.

**Title page:**

Choose a revealing title. It should be short and include the name(s) and address(es) of all authors. It should not have acronyms or abbreviations or exceed two printed lines.

**Abstract:** This summary should be two hundred words or less. It should clearly and briefly explain the key findings reported in the manuscript and must have precise statistics. It should not have acronyms or abbreviations. It should be logical in itself. Do not cite references at this point.

An abstract is a brief, distinct paragraph summary of finished work or work in development. In a minute or less, a reviewer can be taught the foundation behind the study, common approaches to the problem, relevant results, and significant conclusions or new questions.

Write your summary when your paper is completed because how can you write the summary of anything which is not yet written? Wealth of terminology is very essential in abstract. Use comprehensive sentences, and do not sacrifice readability for brevity; you can maintain it succinctly by phrasing sentences so that they provide more than a lone rationale. The author can at this moment go straight to shortening the outcome. Sum up the study with the subsequent elements in any summary. Try to limit the initial two items to no more than one line each.

**Reason for writing the article**—theory, overall issue, purpose.

- Fundamental goal.
- To-the-point depiction of the research.
- Consequences, including definite statistics—if the consequences are quantitative in nature, account for this; results of any numerical analysis should be reported. Significant conclusions or questions that emerge from the research.

**Approach:**

- Single section and succinct.
- An outline of the job done is always written in past tense.
- Concentrate on shortening results—limit background information to a verdict or two.
- Exact spelling, clarity of sentences and phrases, and appropriate reporting of quantities (proper units, important statistics) are just as significant in an abstract as they are anywhere else.

**Introduction:**

The introduction should "introduce" the manuscript. The reviewer should be presented with sufficient background information to be capable of comprehending and calculating the purpose of your study without having to refer to other works. The basis for the study should be offered. Give the most important references, but avoid making a comprehensive appraisal of the topic. Describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will give no attention to your results. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here.
The following approach can create a valuable beginning:

- Explain the value (significance) of the study.
- Defend the model—why did you employ this particular system or method? What is its compensation? Remark upon its appropriateness from an abstract point of view as well as pointing out sensible reasons for using it.
- Present a justification. State your particular theory(-ies) or aim(s), and describe the logic that led you to choose them.
- Briefly explain the study's tentative purpose and how it meets the declared objectives.

**Approach:**

Use past tense except for when referring to recognized facts. After all, the manuscript will be submitted after the entire job is done. Sort out your thoughts; manufacture one key point for every section. If you make the four points listed above, you will need at least four paragraphs. Present surrounding information only when it is necessary to support a situation. The reviewer does not desire to read everything you know about a topic. Shape the theory specifically—do not take a broad view.

As always, give awareness to spelling, simplicity, and correctness of sentences and phrases.

**Procedures (methods and materials):**

This part is supposed to be the easiest to carve if you have good skills. A soundly written procedures segment allows a capable scientist to replicate your results. Present precise information about your supplies. The suppliers and clarity of reagents can be helpful bits of information. Present methods in sequential order, but linked methodologies can be grouped as a segment. Be concise when relating the protocols. Attempt to give the least amount of information that would permit another capable scientist to replicate your outcome, but be cautious that vital information is integrated. The use of subheadings is suggested and ought to be synchronized with the results section.

When a technique is used that has been well-described in another section, mention the specific item describing the way, but draw the basic principle while stating the situation. The purpose is to show all particular resources and broad procedures so that another person may use some or all of the methods in one more study or referee the scientific value of your work. It is not to be a step-by-step report of the whole thing you did, nor is a methods section a set of orders.

**Materials:**

*Materials may be reported in part of a section or else they may be recognized along with your measures.*

**Methods:**

- Report the method and not the particulars of each process that engaged the same methodology.
- Describe the method entirely.
- To be succinct, present methods under headings dedicated to specific dealings or groups of measures.
- Simplify—detail how procedures were completed, not how they were performed on a particular day.
- If well-known procedures were used, account for the procedure by name, possibly with a reference, and that's all.

**Approach:**

It is embarrassing to use vigorous voice when documenting methods without using first person, which would focus the reviewer's interest on the researcher rather than the job. As a result, when writing up the methods, most authors use third person passive voice.

Use standard style in this and every other part of the paper—avoid familiar lists, and use full sentences.

**What to keep away from:**

- Resources and methods are not a set of information.
- Skip all descriptive information and surroundings—save it for the argument.
- Leave out information that is immaterial to a third party.
Results:
The principle of a results segment is to present and demonstrate your conclusion. Create this part as entirely objective details of the outcome, and save all understanding for the discussion.

The page length of this segment is set by the sum and types of data to be reported. Use statistics and tables, if suitable, to present consequences most efficiently.

You must clearly differentiate material which would usually be incorporated in a study editorial from any unprocessed data or additional appendix matter that would not be available. In fact, such matters should not be submitted at all except if requested by the instructor.

Content:
- Sum up your conclusions in text and demonstrate them, if suitable, with figures and tables.
- In the manuscript, explain each of your consequences, and point the reader to remarks that are most appropriate.
- Present a background, such as by describing the question that was addressed by creation of an exacting study.
- Explain results of control experiments and give remarks that are not accessible in a prescribed figure or table, if appropriate.
- Examine your data, then prepare the analyzed (transformed) data in the form of a figure (graph), table, or manuscript.

What to stay away from:
- Do not discuss or infer your outcome, report surrounding information, or try to explain anything.
- Do not include raw data or intermediate calculations in a research manuscript.
- Do not present similar data more than once.
- A manuscript should complement any figures or tables, not duplicate information.
- Never confuse figures with tables—there is a difference.

Approach:
As always, use past tense when you submit your results, and put the whole thing in a reasonable order.

Put figures and tables, appropriately numbered, in order at the end of the report.

If you desire, you may place your figures and tables properly within the text of your results section.

Figures and tables:
If you put figures and tables at the end of some details, make certain that they are visibly distinguished from any attached appendix materials, such as raw facts. Whatever the position, each table must be titled, numbered one after the other, and include a heading. All figures and tables must be divided from the text.

Discussion:
The discussion is expected to be the trickiest segment to write. A lot of papers submitted to the journal are discarded based on problems with the discussion. There is no rule for how long an argument should be.

Position your understanding of the outcome visibly to lead the reviewer through your conclusions, and then finish the paper with a summing up of the implications of the study. The purpose here is to offer an understanding of your results and support all of your conclusions, using facts from your research and generally accepted information, if suitable. The implication of results should be fully described.

Infer your data in the conversation in suitable depth. This means that when you clarify an observable fact, you must explain mechanisms that may account for the observation. If your results vary from your prospect, make clear why that may have happened. If your results agree, then explain the theory that the proof supported. It is never suitable to just state that the data approved the prospect, and let it drop at that. Make a decision as to whether each premise is supported or discarded or if you cannot make a conclusion with assurance. Do not just dismiss a study or part of a study as "uncertain."
Research papers are not acknowledged if the work is imperfect. Draw what conclusions you can based upon the results that you have, and take care of the study as a finished work.

- You may propose future guidelines, such as how an experiment might be personalized to accomplish a new idea.
- Give details of all of your remarks as much as possible, focusing on mechanisms.
- Make a decision as to whether the tentative design sufficiently addressed the theory and whether or not it was correctly restricted. Try to present substitute explanations if they are sensible alternatives.
- One piece of research will not counter an overall question, so maintain the large picture in mind. Where do you go next? The best studies unlock new avenues of study. What questions remain?
- Recommendations for detailed papers will offer supplementary suggestions.

**Approach:**

When you refer to information, differentiate data generated by your own studies from other available information. Present work done by specific persons (including you) in past tense.

Describe generally acknowledged facts and main beliefs in present tense.

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Administration Rules to Be Strictly Followed before Submitting Your Research Paper to Global Journals Inc.

*Please read the following rules and regulations carefully before submitting your research paper to Global Journals Inc. to avoid rejection.*

**Segment draft and final research paper:** You have to strictly follow the template of a research paper, failing which your paper may get rejected. You are expected to write each part of the paper wholly on your own. The peer reviewers need to identify your own perspective of the concepts in your own terms. Please do not extract straight from any other source, and do not rephrase someone else's analysis. Do not allow anyone else to proofread your manuscript.

**Written material:** You may discuss this with your guides and key sources. Do not copy anyone else's paper, even if this is only imitation, otherwise it will be rejected on the grounds of plagiarism, which is illegal. Various methods to avoid plagiarism are strictly applied by us to every paper, and, if found guilty, you may be blacklisted, which could affect your career adversely. To guard yourself and others from possible illegal use, please do not permit anyone to use or even read your paper and file.
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