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Invasive Bacterial and Malaria

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} Highlights }

Distortion Product Otoacoustic

Retrospective Review of Records

Discovering Thoughts, Inventing Future

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Improving the Quality of Life of Patients with Back Pain

By Yulduz M. Isamukhamedova & Adiba A. Usmankhodjaeva

Abstract- This paper discusses improving the quality of life of patients with various pains on their back, depending on the sex, age, and nature of the pain syndrome. 130 patients (46 men and 84 women) with vertebrogenic back pain were examined and were on ambulatory treatment in the neurological department of TMA. Age grade was from 20 to 55 years. According to the results of the examination, all patients were divided into three groups, depending on the presence or absence of pain syndromes.

Keywords: pain syndrome, quality of life, patients, chronic pain, acute pain.

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Improving the Quality of Life of Patients with Back Pain

Yulduz M. Isamukhamedova ^α & Adiba A. Usmanhodjaeva ^ο

Abstract- This paper discusses improving the quality of life of patients with various pains on their back, depending on the sex, age, and nature of the pain syndrome. 130 patients (46 men and 84 women) with vertebrogenic back pain were examined and were on ambulatory treatment in the neurological department of TMA. Age grade was from 20 to 55 years. According to the results of the examination, all patients were divided into three groups, depending on the presence or absence of pain syndromes.

Keywords: pain syndrome, quality of life, patients, chronic pain, acute pain.

I. INTRODUCTION

One of the most pressing problems of medicine is pain syndromes, which are a heterogeneous group of common conditions, the medical and social significance of which is difficult to overestimate^[1,2,3]. The reason for patients to seek medical help is often the emergence or intensification of pain^[4, 5]. The most common cause of back pain is dystrophic spinal lesions^[6]. According to experts of the International Association for the Study of Pain, pain lasting more than 3 months is considered chronic^[7,8]. It is the relief of chronic pain in the back is the main task in the treatment of this category of patients. At the same time, in 80% of patients, the pain disappears under the influence of treatment within a month, but in the rest, they take a chronic course. Despite significant advances in the development of issues of pathogenesis, diagnosis, and treatment of neurological manifestations of lumbar osteochondrosis, many aspects of this pathology remain poorly understood. Of particular importance is the problem of outpatient treatment due to the fact that the methods of therapeutic measures used in most medical institutions are adapted exclusively to the inpatient stage^[9]. Such a template approach, not taking into account the stage, etiological and pathogenetic factors, peculiarities of cyanogenetic reactions in a particular patient leads to a breakdown of compensatory reactions and worsens the results of rehabilitation activities^[3, 10]. The lack of sufficiently effective care for patients with diseases of the spine, usually proceeding chronically, with alternating remissions and exacerbations, leads to a loss of confidence in the doctor. According to a number of

experienced clinicians, the passivity of the doctor is unacceptable, as it can lead to the psychosocial death of the patient long before his biological death^[1, 8]. In this regard, it is of interest to study the various components of the quality of life for vertebral back pain. Their analysis will make it possible to rationalize medical tactics for this pathology, which determined the goal and objectives of this study.

The purpose of the study is to assess the quality of life of patients with back pain, depending on gender, age and nature of pain.

II. MATERIAL AND METHODS

The object and subject of the study were 130 patients (46 men and 84 women) with vertebral back pain and were on outpatient treatment in the neurological department of TMA. Patients with acute and chronic back pain between the ages of 20 and 55 years.

III. RESULTS

According to the survey results, all patients were divided into three groups depending on the presence or absence of pain syndromes.

Table 1: Treatment of Patients with Back Pain

	Acute Pain		Chronic Pain		Total
	Men	Women	Men	Women	
Once	10	18	11	13	52 (40%)
On Several Times	9	16	9	21	55 (42,3%)
Regularly	3	6	4	10	23 (17,6%)
All	22	40	24	44	130 (100%)

1st Group consisted of patients whose pain was once.

Group II patients - having pain less than 3 months.

Group III included individuals with algic manifestations that met the criteria for chronic pain.

The frequency of pain in patients of the second group in 40% of cases was once a month or was once. In persons of the third group, the most frequent pain occurred daily (17.6%).

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Localization of pain syndrome, for the most part, was traditional - cephalgia or dorsalgia. Attention was drawn to the fact that these algic violations were not isolated in most cases. Thus, in the group with acute pain, only 29 (16.9%) respondents complained of pain in the two other zones and only 5 (2.9%) in three or more respondents. In the group with chronic pain disorders, almost every third respondent indicated two localizations - 32 people (32.3%), and three or more - 16 people (16.2%). To relieve pain, the subjects took analgesics: irregularly - 41.4% of men and 57.5% of women, regularly - 10.3% and 24.6%, respectively.

Table 2: Pain Relief for Patients

Investigated Parameters	Index	
	Men	Women
Rest or Sleep (% of Patients)	79,5	85,2
Sleep with Sleeping Pills (% of Patients)	30,1	49,5
Awakening due to Pain (% of Patients)	25,1	58,5

Relief or sleep helped to relieve pain - 79.5% of men and 85.2% of women, sedatives - 30.1% of men and 49.5% of women. Patients awoke because of the pain of 25.1% of men and 58.5% of women. Thus, women regularly resorted to more painkillers than men.

IV. CONCLUSION

As a result of the study, it was found among people who did not currently seek medical help for pain syndromes, 47.7% suffered from acute and 52.3% chronic algic disorders. Significant gender differences were revealed both in the clinic and in the attitude of patients to pain syndromes. Both acute and chronic pain syndromes are more often recorded in women, the percentage of women in groups with chronic pain is higher than in groups with acute pain, although the difference is within the margin of error - 30% and 33%, respectively. But, with acute pain syndromes, men were almost 2 times less likely to be referred for examination and treatment (44%) than women (66%). A comparative analysis of the ways to relieve pain revealed that women more often (49.5%) than men (30.1%) use both drug and non-drug methods (85% and 79.5%, respectively).

Contributors

Y. I. conceived and designed the study. Y. I. is the Principal Investigator and the study statistician who prepared the analyses. A. U. wrote the protocol, the Chief Investigator for the Y. I. All authors provided input and approved the final version.

Declaration of Interests

Authors declare that there is no competing interest.

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Distortion Product Otoacoustic Emissions in Children with Autism Spectrum Disorder

By Marlene Escher Boger, Juliana Siqueira, Cleybson Araujo, Alex Maciel
& Denise Torquato

Abstract- Introduction: To study the hearing of children with Autism Spectrum Disorder (ASD) is often a challenge due to the difficulty of interaction, attention, perception and memory that make it difficult to perform subjective tests, such as conventional audiometry. It is believed that objective exams such as Otoacoustic Emissions ensure greater reliability on the results.

Objective: To evaluate otoacoustic emissions in children with Autism Spectrum Disorder.

Method: This is a cross-sectional descriptive study, in which the participants were children diagnosed with Autism Spectrum Disorder, aged between 4 and 11 years. The audiological evaluation was performed using distortion product otoacoustic emissions (DPOAE) to assess cochlear functioning, more specifically functioning of the outer hair cells.

Results: When evaluating the function of outer hair cells in children with ASD, the findings reveal that 55.6% of the children presented alterations in the functioning of the outer hair cells of the cochlea.

Keywords: hearing, autistic disorder, speech, language and hearing sciences.

GJMR-K Classification: NLMC Code: WL 705



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Distortion Product Otoacoustic Emissions in Children with Autism Spectrum Disorder

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Abstract- Introduction: To study the hearing of children with Autism Spectrum Disorder (ASD) is often a challenge due to the difficulty of interaction, attention, perception and memory that make it difficult to perform subjective tests, such as conventional audiometry. It is believed that objective exams such as Otoacoustic Emissions ensure greater reliability on the results.

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Results: When evaluating the function of outer hair cells in children with ASD, the findings reveal that 55.6% of the children presented alterations in the functioning of the outer hair cells of the cochlea.

Conclusion: The DPOAE test proved to be efficient for evaluating children with ASD, but the need to use other tests to determine a safe diagnosis is recognized.

Keywords: hearing, autistic disorder, speech, language and hearing sciences.

1. INTRODUCTION

Autism Spectrum Disorder (ASD) is a complex behavioral syndrome with multiple causes that combine genetic and environmental factors which cause alterations on the motor, psychological and neurological development, thus, jeopardizing the language, the cognition and social communication of the individual¹⁻². Besides, other characteristics such as stereotypical movements, variable intelligence standards and vulnerability can also be observed³. ASD manifests prematurely before the child is 36 months old⁴⁻⁶. Its etiology still hasn't been found and its diagnosis is defined by the set of displayed symptoms^{7,2}.

In the late years, autism has reached a considerable expression. There's a worldwide estimation of 70 cases for 10,000 inhabitants, and the incidence rate seems to be higher for males⁷. In Brazil, about 27.2

cases of autism were reported for 10,000 inhabitants. However, there doesn't seem to be any known association with racial, social, economic or cultural aspects⁸.

As to the linguistic aspects, children diagnosed with ASD manifest alterations on the socioemotional development, displaying difficulty to establish a normal conversation, reduced share of interests, emotions of affection, beside the difficulty to begin or respond to social interactions. They also display difficulty at nonverbal communication employed in social interaction, alternating, for example, between verbal and nonverbal communication. Other characteristics, such as difficulty to develop, maintain and understand relationships, adjust their behavior to fit diverse social contexts and share imaginative games are also observed in children with ASD⁹. Souza and collaborators (2009) consider that the alterations on language in individuals with ASD usually comprise delay or failure on the development of language, not compensated by gestures or mimics; failure in responding to the communication of others; relative failure in beginning or maintaining communication exchange; usage of stereotypical and repetitive language; idiosyncratic usage of words and abnormalities on speech prosody¹⁰.

As to audition, ASD bearers usually display hypersensitivity or hyposensitivity to sound stimulation. Their visual-spatial processing seems to be whole, though¹¹⁻¹². Lately, some studies have evaluated the audition of children ASD, and some controversies have arisen. In 2014, researchers evaluated children with ASD and concluded that the audio logical findings are compatible with normality both at behavioral evaluation and electrophysiological evaluation of audition¹³. In 2016, other researchers identified an alteration on the internal ear of children with ASD that can impact on their ability to recognize speech. The results concluded that the otoacoustic emission (OAE) test can be used to identify children under risk of autism at premature age. That study investigated the audition of children aged between 6 and 17 years old. About half of them were diagnosed with ASD. It was found that children with ASD face difficulty to hear specific frequencies (1-KHz) that are important for the discourse of speech processing. They also found correlation between the cochlear impairment and the gravity of the symptoms of autism¹⁴.

Evaluating the audition of children with ASD is often a challenge. Such evaluation can be done through

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objective and subjective exams. Patients with ASD, for having difficulties regarding interaction, attention, perception and memory, make the application of subjective exams difficult and, thus, answers may be mistaken for auditory loss, because they depend on the participation of the individual. Thus, objective exams such as the Brainstem Auditory Evoked Potential (BAEP) and Otoacoustic Emissions (OAE) assure more trustable results¹⁵⁻¹⁶.

Otoacoustic emissions are the sound energy captured by the acoustic meatus, produced by the contraction of the external hair cells of the cochlea. They can be either spontaneous or evoked. Evoked OAE are classified in three types that vary according to the stimulus: Transient, Distortion Product and Stimulus-frequency. In this study, we chose to use the Distortion Product Otoacoustic Emissions (DPOAE). It's a quick, painless, non-invasive procedure that evaluates the answers obtained by cochlear hair cells to the simultaneous acoustic stimulation of two pure tones. It can detect cochlear lesions, from the basal coil to the apical coil¹⁷.

Considering the controversies in the literature on the communication abilities of children with autism spectrum disorder, how much damages to the hearing function are related to those difficulties, and the possibility of an objective evaluation of the cochlear function of such subjects, the goal of this study is to evaluate DPOAE in children with Autism Spectrum Disorder.

II. METHODS

It's a transversal descriptive study whose participants were children diagnosed with autism spectrum disorder, aged between 4 and 11 years old. The evaluated population was selected through convenience sample and it was composed only by those subjects that accepted to participate in the research. Those responsible for the participants were informed about the methodological procedures and signed the Consent Form (Attachment 1) before any procedure was conducted.

The data collection was conducted at Clínica Escola de Fonoaudiologia do Centro Universitário Planalto do Distrito Federal (Uniplan), and the sample comprised patients that attend the Supervised Attendance of Child Language of that Institution, diagnose with ASD.

In order to select that sample, the following exclusion criteria were previously defined: presence of other associated impairments, diagnosed by an expert team (psychiatrist and neurologist, speech therapists and psychologists), audio logical risk factors, such as prematurity and usage of ototoxic medicine, and presence of alterations on the middle ear, observed in meatoscopy.

All those responsible for the children answered to the specific audio logical anamnesis (Attachment 2). The audio logical evaluation began with the otoscopic inspection with the purpose of discarding the presence of structural anomalies and/or the obstruction of the external acoustic meatus that might hinder the conduction of the audio logical exam. Then, the analysis of the distortion product otoacoustic emissions (DPOAE) with the purpose of evaluating the cochlear functioning, more specifically the external hair cells. The Oto Read - Screener equipment of the INTERACOUSTICS brand was used. DPOAE were evaluated through the simultaneous presentation of two different pure tones (F1 and F2), expressed through the reason of 1.22. The intensity parameter L1=65 and L2=55 dB was used, and the cochlear conditions were measured in the frequency of 2 kHz, 3 kHz, 4 kHz and 5kHz. The analyses of DPOAE were conducted through frequency, following the amplitude criteria (SD) higher than -5 dB and the relation between signal/noise (SN) higher than 6 dB. The occurrence of answers to DPOAE on a frequency was considered when the values established in the two foresaid criteria were observed on that frequency. Those that got answers for at least 3 out of the 4 tested frequencies were considered the normality standard for external hair cells functioning, for both the amplitude criterion and the signal-to-noise ratio criterion.

The collected exams were printed during the test. This enables the visualization of the amplitude and signal-to-noise ratio parameters in DPOAE by separate frequencies.

Those responsible for the children were communicated about the result of the exam immediately after its conduction. In case of alteration of the test, they were guided to the complementary Otorhinolaryngological evaluation and BAEP.

The collected data was transported to electronic spreadsheets, through which they got analytical treatment, both central tendency statistics (mode, mean and median) and variance (standard deviation). They were also presented in diagrams and tables, through the usage of the Excel tool.

This work was submitted and approved by the Ethics in Researches with Human Beings Committee at Faculdade Integrada da União Educacional do Planalto Central - FACIPLAC, with register number CAAE 70136017.0.0000.5058 (Attachment 3).

III. RESULTS

First, 17 otoacoustic emission exams were scheduled preceded by meatoscopy and anamnesis. 5 out of this didn't attend, and 3 didn't collaborate for the test conduction, making the evaluation conclusion impossible. By the end, there were 9 children with ASD evaluated in the DPOAE test.

The evaluated subjects had an average age of 6.5 years ($SD \pm 2.4$). Regarding gender, 100% of the children were male (Table 1).

Table 1: Characterization of the Sample according to Age and Gender

Characteristics		Descriptive Statistics	
Age	Mean \pm SD	6,5 \pm 2,4	
	Median	6	
	Minimum	4	
	Maximum	11	
Gender		N	%
Male		9	100
Female		0	0

$\pm SD$ - standard deviation

According to the results obtained in PDOAE, it was observed that 5 out of the 9 evaluated individuals (55.6%) obtained altered results and 4 are within the normality standards. Out of those altered results, only 1 (11.1%) presented alterations in both ears and in all tested frequencies. When each ear was evaluated separately, that variable displayed unilateral alterations of 33.3% on the left ear and 11.1% on the right ear, characterizing failure on the functioning of the external hair cells of the cochlea (Table 2).

Table 2: Prevalence of PDOAE according to Laterality.

PDOAE Prevalence				
Laterality	Normal		Altered	
	N	%	N	%
BE	4	44,4	1	11,1
LE	5	55,5	3	33,3
RE	7	77,7	1	11,1

BE - both ears; LE - left ear; RE - right ear

As to the PDOAE analysis criterion, only 1 individual presented alterations in more than one of the frequencies tested in the signal-to-noise ratio criterion. In the amplitude criterion, 5 individuals displayed alterations on the functioning of the external hair cells, and 4 obtained answers within the normality in both criteria (Table 3).

Table 3: Alterations in PDOAE according to the Criterion Employed

PDOAE Prevalence						
Criterion	Normal		Altered		Total	
	N	%	N	%	N	%
Amplitude	4	44,4	5	55,6	9	100
Signal / Noise	8	88,9	1	11,1	9	100

When PDOAE were analyzed regarding the average amplitudes, the laterality of the ears and the evaluated frequencies, it was observed that, with the increase of frequencies, there is a decrease of amplitudes in the right ear. As to the left ear, the frequency with the highest amplitude is 3kHz (3.3dB)

and the lowest amplitude is on the frequency of 5KHz, which result is lower than -5dB (-6.2dB), showing that the alteration of external hair cells in children with ASD is mainly on that frequency (Table 4).

Table 4: Overall Mean of the DPOAE Amplitudes according to the Laterality (Left and Right Ears) and the Evaluated Frequencies

PDOAE Amplitude				
Freq. (kHz)	RE		LE	
	Mean	$\pm SD$	Mean	$\pm SD$
2	3,7	8,1	1,9	9,4
3	0,0	10,2	3,3	5,2
4	-1,4	9,3	-2,8	9,4
5	-2,7	7,0	-6,2	10,5

$\pm SD$ - standard deviation

Regarding the DPOAE means in the signal-to-noise ratio, the laterality and evaluated frequencies, it was verified that, in both ears and in all the frequencies, the means area higher than 6 dB, a normality criterion (Table 5).

Table 5: Overall Mean of the Signal-To-Noise Ratio of the PDOAE according to the Laterality (Right Ear) and Evaluated Frequencies.

PDOAE Signal-To-Noise Ratio				
Freq.(kHz)	RE		LE	
	Mean	$\pm SD$	Mean	$\pm SD$
2	13,0	6,0	8,2	10,0
3	17,3	8,9	13,7	6,6
4	16,6	9,2	12,9	11,5
5	15,2	8,1	11,2	10,1

$\pm SD$ - standard deviation

IV. DISCUSSION

In this chapter, the results found in the study will be interpreted, and compared to the data of the authors mentioned in the literature revision, whenever it's possible.

The possibility of identifying a cochlear alteration in children with autism spectrum disorder motivated this study, and it motivated several scientists to research through the OAE test. The base of the selection of the distortion products for this research was the possibility of evaluating the cochlear activity in specific frequencies, providing a wide analysis compared to the transient emissions that evaluate the cochlea globally. For the conduction of this test, it's essential that the middle ear is in proper physiological conditions. It is an efficient, quick and objective exam for the differential diagnosis and the monitoring of the audition. However, the Otoacoustic Emissions test is considered a complementary evaluation, and it alone can't diagnose auditory losses. So, complementary

behavioral, electroacoustic and electrophysiological audition exams must be requested.

As to the characterization of the sample, all the evaluated children were male, supporting Volkmar and McPartland (2014), who assert that the incidence rate of OAE seems to be higher for the male gender⁷.

Regarding the auditory characteristics, it was verified that almost half (55.6%) the children displayed altered DPOAE according to the established evaluation criteria, which differs from the study conducted by Romero and collaborators (2014), in which the findings displayed auditory normality in every conducted test, including the otoacoustic emissions¹³.

Besides the prevalence registers, the occurrence of OAE is usually analyzed by a set of criteria. In this research, the amplitude and signal-to-noise ratio criteria, with the purpose of evaluating the occurrence of DPOAE in children with ASD. In this study, the selection of amplitude criterion higher than -5 dB and the signal-to-noise ratio criterion higher than 6 dB proved to be efficient in the detection of alterations of external hair cells in children with ASD. We know that in the clinical practice, some researchers employ more rigid criteria, such as signal-to-noise ratio higher than 8 dB or the absence of answers on a single frequency would be considered an alteration¹⁸. The adoption of these criteria in this study would raise the possibility of even worse results.

When you compare the amplitudes and signal-to-noise ratio amplitudes, you verify that the worst results refer to the amplitude criterion. This data shows that the selection of analysis criteria area extremely important to determine the normality or the alteration of the external hair cells. Gorga and collaborators (1997) assert that the DPOAE amplitude shows a decrease with aggravation of the auditory threshold in individuals with cochlear auditory loss. Thus, the amplitude evinces the real functioning of the cochlea. Therefore, it is an indispensable criterion. Some OAE equipment are already factory-set to consider normality, based only on the signal-to-noise ratio criterion. In this study, we remark that the usage of only one criterion is not enough to establish normality or alteration of the cochlear activity.

We must remark that only the observation of the signal-to-noise ratio of the data presented in this work would raise the percentage of normality of the evaluated children to 88.9%, while with the combination of criteria, signal-to-noise ratio and amplitude, such percentage of normality usually decreases to 44.4%.

When the amplitudes means were analyzed separately, it was observed that the frequency of 5KHz showed the worst means, thus differing with the study of Bennetto and collaborators (2016), which showed that children with ASD have difficulty to hear specific frequencies in the range of 1-2KHz¹⁴. On the other

hand, it supports such research, when it asserts that the OAE test can be used to identify alterations in

One of the aspects that stood out the most in this study was the occurrence of the only individual that presented alterations in both ears, in both criteria, and in every frequency. At the age of two, the child had been evaluated with BAEP and OAE during the phase of diagnostic investigation of ASD, which result, then, was within the normality standards. Currently, at the age of 5, when he was invited to participate in this study, he was evaluated with DPOAE and didn't show cochlear answers. So, he was guided to the conduction of a new BAEP on which severe bilateral auditory loss was confirmed. Such fact evinces the possibility of late auditory loss. Thus, it reinforces the importance of the annual auditory monitoring in children with ASD with the purpose of preventing that late auditory alterations appear and don't be detected, contributing even more for all the difficulties already found in language for that population.

Considering these observations, and admitting the need to advance the studies and researches in this field, we suggest the development of new studies, with the purpose of establish the audiological profile of individuals with ASD, as the suggestion to conduct a case-control study, comparing the audiological profile of children with ASD with children with no complaints, as well as the conduction of complementary exams, such as transient evoked otoacoustic emissions, BAEP and tympanometry, the conduction of longitudinal studies and the study with the highest number of tested frequencies.

V. CONCLUSION

The DPOAE test proved to be efficient to evaluate children with ASD, but we recognize the need to conduct other tests to determine a safe diagnosis.

Conflict of Interests

There's no conflict of interests.

Finance

There was no financing.

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Historical Evidence of “Printer’s Devil” in Pediatric Cancer Literature

By Wilson I. B. Onuigbo

Abstract- “Printer’s devil” is an error introduced during the routine printing of manuscripts. In the author’s experience, this was used to determine the source of such an error with reference to reprints. In this context, since it is known that research profits from the slightest detectable error, this paper points to a printed error which occurred in 1893 in the Transactions of the Pathological Society of London.

Keywords: *printer’s devil, printing, information, error, cancer, history.*

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Keywords: printer’s devil, printing, information, error, cancer, history.

I. INTRODUCTION

In the field of informatics, the printing error is of considerable interest⁽¹⁾. Dubbed “Printer’s Devil,” the author made use of such errors to determine that the magazine, Current Contents, was the innate source in his experience⁽²⁾.

Research, according to Alan Gregg⁽³⁾, “profits from the slightest deviation from the conduct expected from Nature.” It is in this sense that such deviation is equally true of human conduct. Therefore, what of the publications of the medical masters of yester years? Their publications in the Transactions of the Pathological Society of London enamored Willis⁽⁴⁾, the great author of “The Spread of Tumours in the Human Body.” Indeed, he revealed it by frequently citing from them when bolstering historical data. Moreover, Burnet⁽⁵⁾ did advice that in research it is necessary to be aware of the historical antecedents. Consequently, it was while pursuing this apt advice that I came across a documentable printer’s devil!

II. HISTORICAL TEXT

F. C. Turner⁽⁶⁾ furnished a Card Specimen before the London Society. The title was “Medullary sarcoma of both ovaries and of the peritoneum in a child aged 6.” Actually, he began thus:

The specimen consists of the pelvic organs of a female child aged 6. Both ovaries are converted into rounded masses of medullary sarcomatous growth. The right ovary is as large as a full-sized orange. The left ovary is smaller; it was adherent to a large mass of growth surrounding the cæcum and commencement of the ascending colon. The right ovary was free.

Free it was not. Thus, as I will italicize, his discussion also centered on two ovaries. Moreover, he

did add that “Sections of the growth in the ovaries showed the structure of small round-celled sarcoma.”

III. DISCUSSION

The author is persuaded that throwing light on both right⁽⁷⁾ and wrong⁽⁸⁾ historical accounts are good for the growth of scientific knowledge. Thus, as an Editorial has it⁽⁹⁾, the historical perspective of “medical truths” require continuing validation. In sum, errors are worthy of being pointed out. Incidentally, the modern trend is to ensure quality in science editing and publishing⁽¹⁰⁾!

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Hair Growth Promoting Potential of Consciousness Energy Healing Treatment in Human Dermal Papilla Cells

By Alice Branton & Snehasis Jana

Abstract- Alopecia is a common disorder related to hair fall, observed in all age's peoples around the world. Although several medical approaches are available, however those are insufficient to mitigate or symptomatic relief from these types anomalies. Hence, it is highly essential to establish an alternative treatment strategy to increase hair proliferation. For this context, the current experiment was conducted to investigate the potential of the Consciousness Energy Healing (The Trivedi Effect®) Treatment to the test items (DMEM) in human follicular dermal papilla culture cells for the assessment of hair cell growth and development. The test item was divided into two parts. One part was denoted as the untreated DMEM group without any Biofield Energy Treatment, while the other part was defined as the Biofield Energy Treated DMEM group, which received the Biofield Energy Healing Treatment by a renowned Biofield Energy Healer, Alice Branton. The experimental results showed that cell proliferation was significantly increased by 219.30% in the Biofield Energy Treated DMEM group as compared to the untreated DMEM group. The results demonstrated that the Biofield Energy Healing Treatment significantly increased the proliferation of human hair follicle dermal papilla cells.

Keywords: consciousness energy healing, the trivedi effect®, dermal papilla cells, skin health, hair health, alopecia.

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Hair Growth Promoting Potential of Consciousness Energy Healing Treatment in Human Dermal Papilla Cells

Alice Branton ^α & Snehasis Jana ^σ

Abstract- Alopecia is a common disorder related to hair fall, observed in all age's peoples around the world. Although several medical approaches are available, however those are insufficient to mitigate or symptomatic relief from these types anomalies. Hence, it is highly essential to establish an alternative treatment strategy to increase hair proliferation. For this context, the current experiment was conducted to investigate the potential of the Consciousness Energy Healing (The Trivedi Effect[®]) Treatment to the test items (DMEM) in human follicular dermal papilla culture cells for the assessment of hair cell growth and development. The test item was divided into two parts. One part was denoted as the untreated DMEM group without any Biofield Energy Treatment, while the other part was defined as the Biofield Energy Treated DMEM group, which received the Biofield Energy Healing Treatment by a renowned Biofield Energy Healer, Alice Branton. The experimental results showed that cell proliferation was significantly increased by 219.30% in the Biofield Energy Treated DMEM group as compared to the untreated DMEM group. The results demonstrated that the Biofield Energy Healing Treatment significantly increased the proliferation of human hair follicle dermal papilla cells. Therefore, the Consciousness Energy Healing (The Trivedi Effect[®]) Treatment could be useful as a hair growth enhancer against different skin injuries like actinic keratosis, necrotizing fasciitis, sebaceous cysts, diaper rash, decubitus ulcer and hair-related disorders like androgenetic alopecia, telogen effluvium, trichodystrophy, alopecia areata, etc.

Keywords: consciousness energy healing, the trivedi effect[®], dermal papilla cells, skin health, hair health, alopecia.

1. INTRODUCTION

The hair follicle consists of mainly two types of components such as epithelial components that included the matrix and outer root sheath, and the dermal components, which included the dermal papilla and the connective tissue sheath^[1]. There were three distinct stages of hair growth, which occurs during cellular proliferation like an active phase (anagen), an intermediate regressive (catagen), and a resting phase (telogen)^[2,3]. Literature explored that generally loss of 50-100 hairs per day considered as hair fall disorders like alopecia. Modern trends of fast globalization and indiscriminate grow of industrialization led to the

development of modern socio-cultural factors such as excessive stress, changes in social activities and lifestyle that can aggravate alopecia^[4]. The exact cause of hair fall is still unknown; however, researcher found some possible reasons those are responsible for alopecia. It may be due to genetic factors, more secretion of androgens, excessive stress, blood circulation disorders, nutrient deficiencies, diet, smoking, drinking, and endocrine disorders^[5]. From the literature, it was reported that about 30% males (age more than 30 years) and 50% males (age more than 50 years) are suffered by hair loss-related disorders^[6]. Moreover, literature also reported that females are less sufferer to this problems, however few women found with alopecia areata *i.e.*, patches of baldness^[7]. Numerous literature including United State - Food and Drug Administration (US-FDA) has been reported the hair growth promotion activity of minoxidil^[8,9]. In recent years, several scientific reports have revealed the useful effects of Energy Therapy, which have shown to enhance the immune function in cases of cervical cancer patients *via* therapeutic touch^[10], massage therapy^[11], etc. Complementary and Alternative Medicine (CAM) therapies are now accepting as preferred models of treatment, among which Biofield Therapy (or Healing Modalities) is one approach that has been reported to have several benefits to enhance physical, mental and emotional human wellness. However, as per the data of 2012 from the National Health Interview Survey (NHIS), which indicated that most of the Americans have been used dietary supplements as a complementary health approach as compared with other practices in past years. The National Center of Complementary and Integrative Health (NCCIH) has recognized and accepted Biofield Energy Healing as a CAM health care approach in addition to other therapies, medicines and practices such as natural products, deep breathing, yoga, Tai Chi, Qi Gong, chiropractic/osteopathic manipulation, meditation, massage, special diets, homeopathy, progressive relaxation, guided imagery, acupressure, acupuncture, relaxation techniques, hypnotherapy, healing touch, movement therapy, pilates, rolfing structural integration, mindfulness, Ayurvedic medicine, traditional Chinese herbs and medicines, naturopathy, essential oils, aromatherapy, Reiki, and cranial

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sacral therapy. Human Biofield Energy has subtle energy that can work effectively^[12]. CAM therapies have been practised worldwide with reported clinical benefits in different health disease profiles^[13]. This energy can be harnessed and transmitted by the experts into living and non-living things via the process of Biofield Energy Healing. The Trivedi Effect®- Consciousness Energy Healing Treatment has been reported with a significant revolution in the physicochemical properties of metals, chemicals, ceramics, and polymers^[14-16], improved agricultural crop yield, productivity, and quality^[17,18], transformed antimicrobial characteristics^[19-21], biotechnology^[22,23], improved bioavailability^[24-26], skin health^[27,28], nutraceuticals^[29,30], cancer research^[31,32], bone health^[33-35], human health and wellness. Considering the promising benefits of an alternative natural therapies-based literature information and importance of Biofield Energy Healing Treatment on various fields, the authors sought to evaluate the impact of the Biofield Energy Treatment (The Trivedi Effect®) on the test item (DMEM) for hair cells growth activity using standard assay in human follicular dermal papilla cells.

II. MATERIALS AND METHODS

a) Chemicals and Reagents

Dulbecco's Modified Eagle Medium (DMEM) and fetal bovine serum (FBS) were obtained from Gibco, India. Minoxidil sulphate (positive control) was purchased from Clearsynth Labs Ltd., Mumbai. Antibiotics solution (penicillin-streptomycin) was procured from HiMedia, India. Other chemicals used in this study were analytical grade obtained from India.

b) BrdU Incorporation Cell Proliferation Assay in HFDPs

The human follicular dermal papilla cells (HFDPs) in DMEM supplemented with 10% FBS were counted using a hemocytometer and a single cell suspension was prepared. The single cell suspension was seeded at a density of 800 cells/well in a fresh DMEM supplemented with 10% FBS in 96-well plates. Then, the cells were incubated in a CO₂ incubator for 24 hours at 37°C, 5%CO₂, and 95% humidity. After 24 hours of incubation, the medium was replaced with a fresh DMEM supplemented with 0.1% FBS. Further, after 24 hours, cells were treated with the test items and positive control (minoxidil sulphate). After incubation for 48 hours, the effect of the test items on cell proliferation was assessed by bromodeoxyuridine (BrdU) incorporation using colorimetric ELISA kit. For that, 10 µL of BrdU solution was added per well and the cells were incubated for 90 minutes at 37°C. After incubation, the medium was removed from each well by gentle pipetting. About 200 µL of a FixDenat solution was added to each well. After incubation, cells were incubated for 30 minutes at room temperature (RT)

(15-25°C). The FixDenat solution was removed by gentle pipetting. After incubation, 100 µL of anti-BrdU-POD (peroxidase) solution was added to each well. Then, the cells were incubated for 90 minutes at RT (15-25°C). After incubation, the anti-BrdU-POD solution was removed by gentle pipetting. Each well was washed 3 times using 200 µL of washing solution. About 100 µL of substrate solution was added to each well. Cells were incubated for 30 minutes at RT (15-25°C). After incubation, the absorbance of each well was measured at 370 nm.

Cellular proliferation was determined as following Equation (1):

$$\% \text{ Cellular proliferation} = \{(B - A)/A\} \times 100 \quad (1)$$

Where, A = OD of Untreated DMEM wells

B = OD of cells treated with the test item / positive control.

c) Experimental Design

The experimental groups composed of group 1 (G-I) with DMEM medium defined as the untreated DMEM group. Group 2 (G-II) contained positive control (minoxidil sulphate) at various concentrations. Further, group 3 (G-III) included the Biofield Energy Treated DMEM group.

d) Biofield Energy Healing Strategy

The test item, DMEM was divided into two parts. First part did not receive any sort of treatment and defined as the untreated DMEM group. The second part was treated with the Biofield Energy Treatment by a renowned Biofield Energy Healer (The Trivedi Effect®), Alice Branton remotely for ~5 minutes under laboratory conditions and coded as the Biofield Energy Treated DMEM group. Healer in this study never visited the laboratory in person, nor had any contact with the test items (DMEM medium). Further, the untreated DMEM group was treated with a "sham" healer for comparative purposes. The "sham" healer did not have any knowledge about the Biofield Energy Treatment. After that, the Biofield Energy Treated and untreated test items were kept in similar sealed conditions for experimental study.

e) Statistical Analysis

All the values were represented as Mean ± SEM (standard error of mean) of three independent experiments. The statistical analysis was performed using SigmaPlot statistical software (v11.0). For two groups comparison student's *t*-test was used. For multiple group comparison, one-way analysis of variance (ANOVA) was used followed by post-hoc analysis by Dunnett's test. Statistically significant values were set at the level of $p \leq 0.05$.

III. RESULTS AND DISCUSSION

a) BrdU Incorporated Cell Proliferation of Dermal Papilla Cells

The effect of the test item son Biofield Energy Treatment and the percent of cellular proliferation of DPCs is presented in Figure 1. The immortalized human follicular dermal papilla cells suspension were treated with the positive control and test item (DMEM). Topical application of minoxidil is a well-established therapeutic for various types of hair growth-related disorders like alopecia^[36]. The untreated DMEM group exhibited 100% cells proliferation of DPCs. Additionally, the positive control, minoxidil showed 68.57%, 187.14% ($p \leq 0.001$), and 230.95% ($p \leq 0.001$) increase the cellular proliferation of DPCs at 0.001, 0.01, and 0.1 μM , respectively in a concentration-dependent manner compared to the untreated DMEM group. Moreover, the percent proliferation of DPCs was significantly ($p \leq 0.001$) increased by 219.30% in the Biofield Energy Treated DMEM group with respect to the untreated

DMEM group (Figure 1). For the study of hair follicle biology, human hair growth *in vitro* model is used as a prototype^[37]. Hair follicles undergo different cycles of growth (anagen), regression (catagen), quiescence (telogen), and regeneration^[38]. The current experimental results demonstrated that Biofield Energy Treatment significantly increased the proliferation of dermal papilla cells (DPCs). Further, DPCs are responsible for the regulation of hair follicles development and periodic regeneration^[5,36,39,40]. Also, the DPCs can generate a signal that regulates the behaviour of keratinocytes in the follicle during the hair cycle^[41,42]. Apart from this, Wnt/ β -catenin signaling pathway plays a critical role to initiate generation of hair follicle *via* stimulation of keratinocytes^[43,44]. Based on the literature and findings of this study, it is assumed that the increment of DPCs due to the impact of The Trivedi Effect[®] that could be due to the activation of Wnt/ β -catenin signaling pathway and regulation of keratinocytes during hair growth and development.

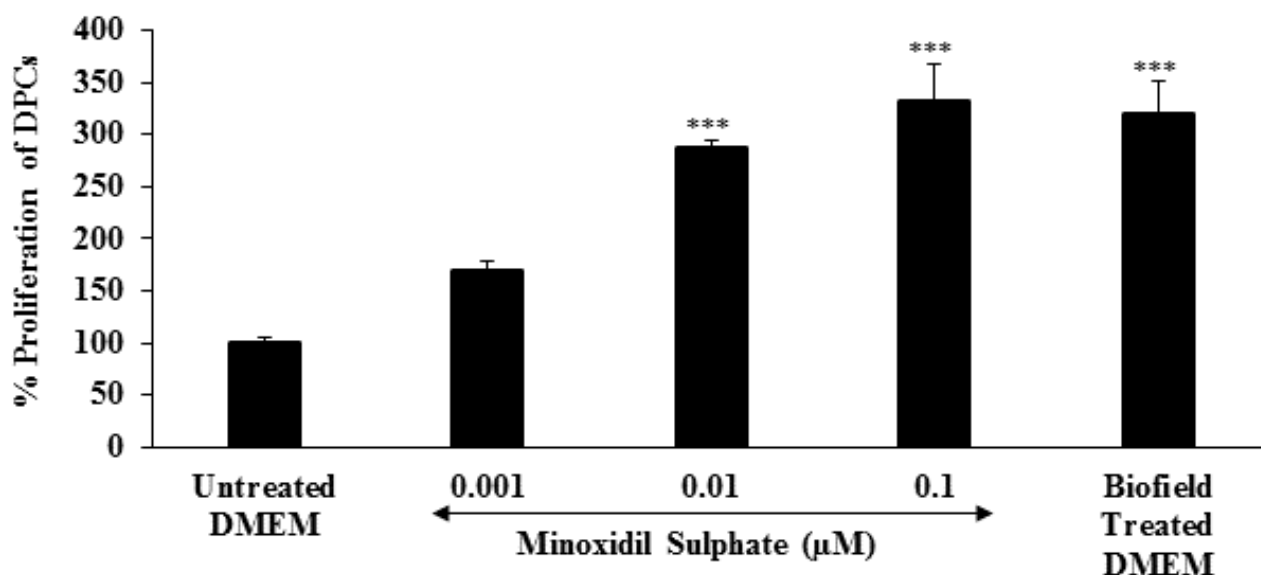


Fig. 1: Effect of the test samples on hair growth regarding dermal papilla cells (DPCs) proliferation after 48 hours of treatment in immortalized human follicular dermal papilla cell line (HFDPC). All the values are represented as mean \pm SEM of three independent experiments. *** $p \leq 0.001$ vs. untreated DMEM group.

The representative photo images signified the intensity of proliferative DPCs after treatment with the Biofield Energy Treated test item (DMEM) in HFDPCs (Figure 2). Overall, data suggested that the Biofield Energy Treatment significantly improved the growth and proliferation of human dermal papilla cells, which is due to the Biofield Energy Healing (The Trivedi Effect[®]). Based on that it is concluded that the Consciousness Energy Therapy could be beneficial to maintain a steady-state proliferation of hair follicles.

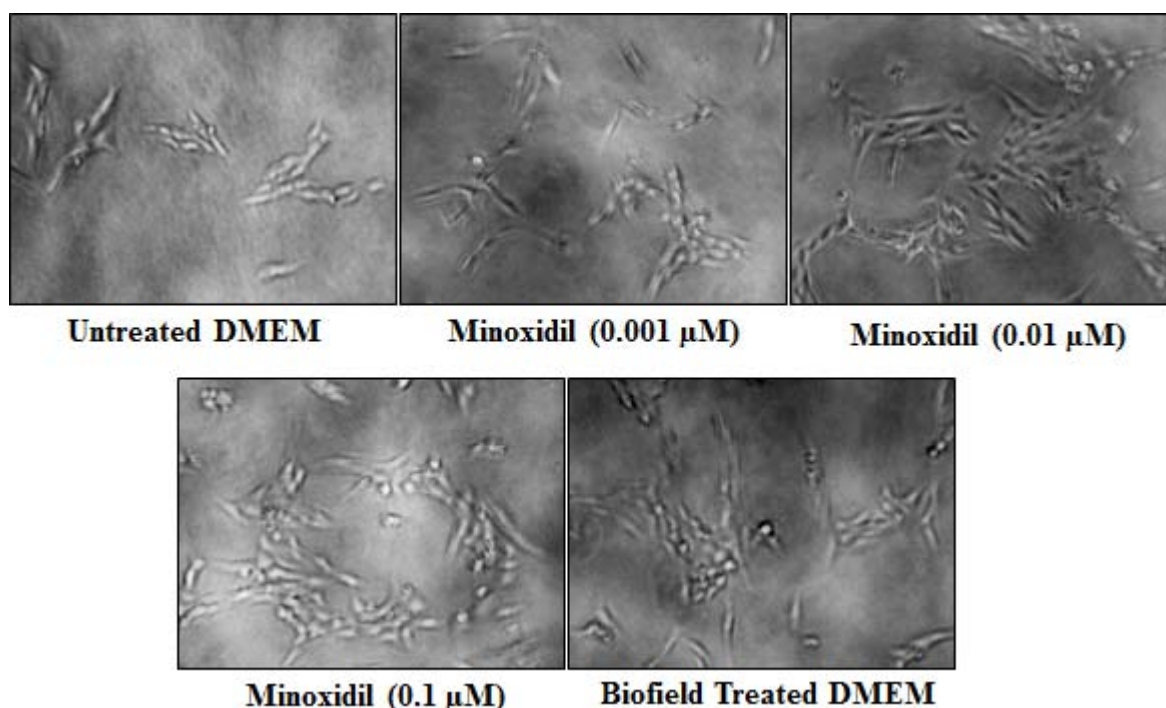


Fig. 2: Effect of Biofield Energy Treated DMEM on the proliferation of dermal papilla cells (DPCs) in human follicular dermal papilla cell line (HFDPC) and the representative images of different treatment groups.

IV. CONCLUSIONS

The study findings was observed that the Biofield Energy Treated test item (DMEM) group showed a significant ($p \leq 0.001$) increase the percent of dermal papilla cells (DPCs) by 219.30% in human follicular dermal papilla cells (HFDPCs) *in vitro*. In conclusion, The Trivedi Effect® - Consciousness Energy Healing Treatment might act as a hair growth promoter, and it can be used as a complementary and alternative treatment for the prevention of various types of skin and hair-related disorders viz. necrotizing fasciitis, actinic keratosis, sebaceous cysts, diaper rash, decubitus ulcer, androgenetic alopecia, telogen effluvium, trichodystrophy, alopecia areata, etc. Besides, it could be useful to improve cell-to-cell communication, normal cell growth, cell differentiation, neurotransmission, cell cycling and proliferation, hormonal balance, skin health, immune and cardiovascular functions. Moreover, it can also be utilized in organ transplants (*i.e.*, kidney transplants, liver transplants and heart transplants), hormonal imbalance, aging, and various immune-related disease conditions such as Ulcerative Colitis (UC), Alzheimer's Disease (AD), Dermatitis, Irritable Bowel Syndrome (IBS), Asthma, Hashimoto Thyroiditis, Pernicious Anemia, Sjogren Syndrome, Multiple Sclerosis, Aplastic Anemia, Hepatitis, Diverticulitis, Graves' Disease, Dermatomyositis, Diabetes, Myasthenia Gravis, Parkinson's Disease, Atherosclerosis, Systemic Lupus Erythematosus (SLE), stress, etc. with a safe therapeutic index to improve overall health and Quality of Life.

Abbreviations:

CAM: Complementary and Alternative Medicine.
DPCs: Dermal papilla cells.
DMEM: Dulbecco's Modified Eagle Medium.
FBS: Fetal bovine serum.
BrdU: Bromodeoxyuridine; POD: Peroxidase.

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Correlation of Lymph Node Density and Recurrence in Carcinoma Penis: A Perspective on Optimal Management and Unmet Needs from a Tertiary Care Centre

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Abstract- Introduction: Penile carcinoma is a devastating urological neoplasm in which the most assertive prognostic variable is the presence of lymph node involvement. Multiple studies in the current literature have shown that the number of positive lymph nodes predict recurrence free survival (RFS) and overall survival (OS). The objective of this present retrospective analysis is to authenticate the use of LND as a predictor of RFS and OS after ILND.

Methods: Our institutional penile cancer database was analyzed for patients who underwent ILND from January 2000 to May 2008. Survival analysis was performed using the Kaplan- Meier method to determine RFS and OS. Two-sided p values <0.05 were considered significant.

Keywords: inguinal lymph node dissection, lymph node density, squamous cell carcinoma penis, overall survival, recurrence free survival, penectomy.

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Correlation of Lymph Node Density and Recurrence in Carcinoma Penis: A Perspective on Optimal Management and Unmet Needs from a Tertiary Care Centre

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Abstract- Introduction: Penile carcinoma is a devastating urological neoplasm in which the most assertive prognostic variable is the presence of lymph node involvement. Multiple studies in the current literature have shown that the number of positive lymph nodes predict recurrence free survival (RFS) and overall survival (OS). The objective of this present retrospective analysis is to authenticate the use of LND as a predictor of RFS and OS after ILND.

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Results: Patients with complete follow up (n=33) were analyzed for clinico-pathological characteristics. LND≥40% was significantly predictive of poor recurrence free survival (Mantle Cox value: 13.609, p=.0005). RFS was significantly lower for patients with LND≥40% (mean survival: 46.22 month vs. 85.79 months, p=.001). Likewise, overall survival was lower in patients with LND≥40% but did not reach significant level (mean survival: 72.48 month vs. 85.30 months, p=.246).

Conclusion: In multivariate Cox regression model continuously coded variables age (omnibus coefficient (OC) value: 1.108, p=.041), no. of positive lymph nodes removed (OC value: 3.681, p=.023), total no. of lymph nodes removed (OC value: 0.438, p=.014) were significant predictors of RFS but not OS in cases with LND ≥ 40%. Recent developments in penile cancer may alter the prognostic significance of our LND cutoff of 40%.

Keywords: inguinal lymph node dissection, lymph node density, squamous cell carcinoma penis, overall survival, recurrence free survival, penectomy.

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1. INTRODUCTION

Squamous cell penile carcinoma is a belligerent urological neoplasm in which the most assertive prognostic variable is the presence and expanse of lymph node involvement. Therefore, inguinal lymph node dissection (ILND) as a marker of pathological staging is an undeniable fact in the present scenario of uncompromising oncological efficacy. ILND guides adjuvant therapy and offers therapeutic benefit^[1,2,3] Currently prognostication of patients with inguinal and pelvic lymph node involvement is based on TNM staging which distributes the patients into 3 different categories based on: 1) the number of positive lymph nodes; 2) the location of positive lymph nodes; and 3) the presence of extranodal extension.^[4,5] When defined initially, the staging criterion was remarkably inconspicuous for providing details about the span and plenum of lymphadenectomy.^[6] Multiple studies in the current literature have shown that the number positive lymph nodes predicts RFS and OS.^[7,8] These studies on the other hand did not account for the span of ILND done, blurring the real extent of lymph node involvement. The lymph node ratio has been defined as the ratio of positive lymph nodes to the total number of lymph nodes isolated, incorporating both a marker of extend of lymph node dissection and the nodal disease load in a single quantitative variable. Role of LND has been proven beyond any doubt in the management of MIBC^[9,10] but the extrapolation of same in patients with squamous cell penile cancer has not been extensively researched upon; to our knowledge currently there is paucity of literature and lack of RCT's with sufficient evidence strength in this part of the world (NE part of India). Additionally there has been a significant disagreement in accurate LND cut off for differentiating poor versus favorable survivability ranging from 6.7- 33 %.^[11-14] The incoherence may be explained by a significant variation in the total number of LN removed in past series, as well as different statistical models used to achieve the cutoff value. The wide array of LN removed from pelvic lymph node stations in addition to the inguinal lymph nodes and simultaneous

addition of these positive lymph nodes in calculating LND skewed the LND calculation. The objective of this present retrospective analysis was to authenticate the use of LND as a predictor of RFS and OS after ILND. We equate our results and analysis with the recent studies in different parts of the world in an effort to validate the variation in cutoff LND and identify how different histopathological and biochemical parameters and statistical rationales affected the results.

II. METHODS

Our institutional penile cancer database was analyzed for patients who underwent ILND (n=33) in a period between January 2005 to May 2013. Clinical and pathologic characteristics including LND and total number of positive lymph nodes (LNs) were analyzed to determine impact on recurrence free survival (RFS) and overall survival (OS). LND or the percent of positive LN out of total LN's, was calculated as both a continuous and categorical variable at varying thresholds. Demographic and pathologic variables were examined to determine impact on RFS and OS. LND categories were defined using the minimum p value approach according to Mazumdar and Glassman analysis^[15] to determine the most significant threshold. Patients underwent bilateral modified templates ILND in all cases. In patients with non-palpable nodes, a superficial dissection above the fascia lata was performed. In cases with palpable lymphadenopathy a deep dissection (below fascia lata) was performed. All lymph nodes were completely embedded prior to pathologist analysis. All specimens were reviewed by senior urological pathologist for issues relating to grade, stage and margins. Descriptive statistics were used to summarize patient characteristics and pathologic features. Continuous variables were compared with the Fisher's exact tests and categorical variables with the chi-square test. Survival analysis was performed using the Kaplan- Meier method to determine RFS. The log rank test was used to compare survival curves. Overall survivals (OS) was calculated from the date of surgery to death from any cause or last follow up. RFS was calculated from the date of surgery to local or distant recurrence or death from cancer. All patients were prescribed a follow-up regimen based on the National Comprehensive Cancer Network guidelines with physical exam every 3-6 months, depending on nodal stage. All statistical analysis was performed with SPSS version 21. Two-sided p values <0.05 were considered significant.

III. RESULTS

Thirty three patents (median age 43 ± 12.28 yrs) with complete follow up (median follow up of 64 ± 22.02 months) were analyzed for clinicopathological characteristics. Maximum number of cases were seen in

the age group of 41 to 50 yrs (n=13, 39.4%) followed by 31 to 40 yrs age group (n=8, 24.2%). The median age was 44 ± 10.26 yrs in the Group A (LND<40%) as compared to 42 ± 14.56 yrs in the group B (LND \geq 40%) and was found to be significant in the final analysis (t=2.34, p=.007). History of multiple sexual partners was maximally present in LND sub group of .31 to .40 (n=5, chi square coefficient=6.46, p=.263). This result was not significantly associated with the same (Pearson's R=.269, p=.129). Phimosis was mostly seen in the LND subgroup of .31 to .40 (n=7, chi square coefficient=10.05, p=.034, Pearson's R=.363, p=.038). Pathological stage was pT2 in 17 cases (51.5%), high grade T1 in 9 cases (27.3%) and pT3 in 7 cases (21.21%). Tumor grade was 5 (15.15%) in G1, 15 (45.45%) in G2 and 13 (39.39%) in G3. Complete blood analysis was analyzed in both the groups with a NLR ratio of 1.8 vis a vis 3.3 in the 2 groups and the higher values being associated with lower RFS (p.009). This result echoed the analysis of a recent study completed in 2016 which showed a high NLR (≥ 2.82) to be associated with significantly poor CSS (p=.023) than those with a low NLR (Increased neutrophil-to-lymphocyte ratio is associated with disease-specific mortality in patients with penile cancer.^[16] None of the patients undergoing ILND had fixed LN's on physical examination and also none of the patients in the study underwent neo adjuvant chemotherapy. Overall 13 (39.39%) patients underwent partial penectomy and 20 (60.61%) patients underwent total penectomy. Primary tumor area (measured in CECT W/A & pelvis) in the group A and B were comparable but not found to be significant in the final analysis (p=.335). LND threshold was calculated and patients were further analyzed into two groups of LND<40% and LND \geq 40%. Lymph node area (measured in CECT abdomen pelvis) was calculated (Table 1) in the group A and B but was not found to be significant in the final analysis (p=.597). Phimosis, T stage, recurrence and LVI were the significantly associated (Table 2) categorical variables along with significant correlation coefficients. On the other end of the spectrum were age, N/C ratio, total and positive no. of LN's (Table 1) extracted which were again significantly associated with the LND groups. LND threshold was calculated and further analyzed into two groups of LND<40% and LND \geq 40%. Maximum number of cases with recurrence was seen in LND \geq 40% (n=6) and was seen in only 2 cases with LND<40%. LVI was significantly associated and correlated with LND \geq 40% and was one of the factors determining recurrence.

Table 1: Clinicopathological characteristics of 33 men undergoing ILND stratified by LND $\geq 40\%$. Values are presented as median (interquartile range), equal variance not assumed (ILND, Inguinal lymph node dissection; LND, lymph node density)

Characteristics	LND < 40% (n = 24)	LND $\geq 40\%$ (n = 9)	p Value
Age	44 (35-50)	42 (39-53)	.007
Primary Tumor Area (CECT Pelvis)	11.62 (6.5-23.90)	13.30 (10-27.90)	.335
Lymph Node Area (CECT Pelvis)	1.8 (1.35-2.35)	2.8 (2.5-4.4)	.597
Neutrophil / Lymphocytic Ratio	1.8 (1.32-2.35)	3.3 (2.05-4.30)	.009
Serum Calcium	9.2 (8.9-9.47)	8.40 (8.25-8.95)	.918
Lymph Nodes Removed	16.5 (12.5- 21.75)	22 (13.5-23.50)	.032
Positive Lymph Node Yield	2.5 (1-3.75)	8 (5-10)	.021

Table 2: Analysis of categorical variable with LND threshold in the 2 groups

Variable	Chi square Coefficient	P value	Pearson R	P value	<40%/≥40% + Cases / Total
Phimosis	4.416	.042	.354	.043	12 / 24 ; 8 / 9
MSP	.733	.392	.149	.408	Non Sig.
CECT (N)	3.417	.065	.322	.068	Non Sig.
CECT (E)	2.068	.150	.250	.160	Non Sig.
Recurrence	12.128	.001	.606	.001	2/24 ; 6/9
T Stage	6.563	.038	.446	.009	9 / 12 / 13; 0 / 5 / 4
Grade	.416	.519	.112	.534	Non Sig.
LVI	6.694	.018	.686	.017	6 / 24 ; 7 / 9
PNI	1.354	.245	.203	.258	Non Sig.

The median (IQR) LN yield in the Group A was 16.5 (12.5-21.75) and 22 (13.5-23.50) respectively and was found to be significant in final analysis ($p=.032$). Similarly the median positive (IQR) LN yield in the Group A was 2.5 (1-3.75) and 8 (5-10) respectively which was significant at the same time ($p=.021$). Detailed pathological examination of positive LN's showed the presence of ENE (extra nodal extension) in 16 (48.48%) patients. This interpretation vehemently changed pathological staging, leading to a migration from lower pN1 categories to higher pN3 category. pN1, pN2 and pN3 disease was observed in 26 (21.21%), 10 (30.30%), and 16 (48.48%) patients, respectively according to the AJCC 8th edition. After stratification according to LND, 5 year CSS rate was 79.1% in patients with LNR<40% vs 31.9 in patients with LNR $\geq 40\%$ (log-rank test: $p<0.001$). In univariable Cox regression models, continuously coded (95% CI: 1.03-1.07; $p=0.003$) but categorically coded LND (95% CI: 2.80-7.48; $p<0.51$) was significant predictors of CSS (Figure 3). Furthermore, in multivariable Cox regression analysis, categorically coded LNR was found to be an independent predictor of CSS ($p\leq 0.001$). Patients with a LND $\geq 40\%$ had from a 2.51- to a 4.08-fold higher probability of dying from their disease compared with patients with a LNR<40%. The inclusion of categorically coded LND in multivariable analysis led to the highest predictive accuracy. Leaving aside the strict prognostic paradigm, we also investigated the most useful LND mark that may help the urologist to patients at various degree of recurrence, in order to contemplate adjuvant management after ILND; nonetheless, the limited number of cases

prevented us from achieving a threshold that could efficiently distinguish between cases with low probability of recurrence and that at higher risk of disease related events. For instance, a LNR threshold of 15% allowed us to divide patients into two groups, where 5-year RFS rates of 86% and 44% were associated with a LND <15% and $\geq 15\%$, respectively. Nonetheless, this effort did not reach significance ($p=0.073$). LND $\geq 40\%$ was significantly predictive of recurrence free survival (Log Rank via Mantle Cox) value: 13.609, $p=.0005$). RFS was significantly lower for patients with LND $\geq 40\%$ (mean survival: 46.22 month vs. 85.79 months, $p=.0001$, Figure 1). Likewise, overall survival was lower in patients with LND $\geq 40\%$ but it did not reach significant level (mean survival: 72.48 month vs. 85.30 months, $p=.246$, Figure 2). Actuarial 5 year RFS for the entire group was 75.75% while the actuarial 5 year overall survival for the same group was 84.85%. In multivariate Cox regression model continuously coded variable age (omnibus coefficient value: 1.108, 95% CI: 1.004-1.223; $p=.041$, Figure 3), no. of positive lymph nodes removed (omnibus coefficient value: 3.681, 95% CI: 1.198-11.311; $p=.023$), total no. of lymph nodes removed (omnibus coefficient value: 0.438, 95% CI: 0.226-0.849; $p=.014$) were significant predictors of RFS.

Table 3: Showing different author experience with disease specific and recurrence free survival in carcinoma penis cases with ILND at various LND thresholds

Previous Study	Inference
Lughezzani et al [$> 22\%$]	Overall 5 yr CSS 65.2% vs. 9.6%, continuous coded variable LND most accurate predictor of CSS.
Ball et al [$> 15\%$]	Median RFS: 62 mo. vs 6.3 mo; Median OS: 73.6 mo. vs 6.3 mo, 5 yr DSS 81.2% vs 24.4%.
Zhu et al [6.7 - 33%]	5-year DSS in patients with LND of 6.7% or less was 91.7%, while only 23.3% in those with LNR greater than 6.7%.
Present Study [$\geq 40\%$]	LND stratified 5 Yr RFS 33.3% (LND $\geq 40\%$) Vs 91.7% (LND $< 40\%$).

IV. DISCUSSION

Loco-regional spread in the inguinal lymph nodes is the most prognostic factor for survival in penile cancer.^[17,18,19] In this contemporary study, we showed that the LND could muster a role of prognostic criterion in the present population of surgically managed patients with carcinoma penis and LNM. Recently, LND emanated as the only independent predictor of CSS, with a favorable prognostic ability as compared to the 6th and 7th TNM staging systems. This signifies the fact that LND encompasses tumor burden (number of positive lymph nodes) and the extent of ILND (number of lymph nodes removed) into a single variable. Thus, besides tumor volume, LND can be considered as a marker of surgical efficacy. The LND may be a significant marker of survival both as a categorically and a continuously coded variable. In the current retrospective study, we corroborate the observation that LND is a predictor of worse outcome. Previous studies have proved that LND levels varied widely in the cutoff used to stratify worse prognosis. Present study identified that a LND $\geq 40\%$ was associated with worse outcome. The analysis further validated the role of LND use as a prognostic tool for predicting recurrence free survival thus validating LND role for clinical purpose. More significantly, we found that continuously coded LND, age, total number of positive LN removed and total number of LN removed outperformed categorically coded LND, phimosis and MSP in multivariate Cox proportional hazard model for RFS. Lymph node burden, representing the total number of positive LN has shown to be associated with worse prognosis in multiple previous studies. Pandey et al. identified a 75.6%, 8.4%, and 0%, 5-year survival with patients who had 1-3, 4-5, and >5 nodes, respectively.^[20] Nevertheless, in this study we did find an association between total positive nodes and total number of LN's removed with RFS but not with OS. Likewise in other solid tumors, LND has been proved to be a superior prognostic tool than LN number for carcinoma penis.^[21-23] Literature investigating LND has identified a survival cutoff, which has ranged widely from 6.7%-33% to classify favorable versus poor prognosis. Our values slightly exceed the previous cutoff values and show RFS was significantly worse with LND $\geq 40\%$. Our results echoed the work of Li et al. and

Lughezzani et al. that identified LND cutoffs of 16% and 22%, respectively, to differentiate poor versus favorable prognosis. Li and his analysis on 71 node positive cases projected that the 16% LND mark separated a 5-year disease-specific survival (DSS) of 81.2% and 24.4% which in multivariate analysis was also independently associated with worse DSS with a HR of 4.31. These outcomes are similar to our 5-year RFS outcomes of 91.7% for patients with LND $< 40\%$ vs 33.7% for patients with LND $\geq 40\%$. Likewise, Lughezzani and team singled out on multivariate analysis that patients with LND $\geq 22\%$ had a 4.55-fold worse CSS. These studies matched our HR of 5.5 for RFS in our multivariate analysis when using a LND $\geq 40\%$. Interestingly, a higher LN threshold of $\geq 40\%$ was not associated with significant OS in our study, but this was likely due to the small number of patients with LND $\geq 40\%$ (n=9). A recent survey investigated the role of LND as a marker of survival, suggesting that its prognostic competence may be superior then the TNM staging system.^[24] Nevertheless their analysis stem from a small patient population (n=45) with lymph node-positive carcinoma penis. The authors applied the median LND to stratify survival results but it is not the most correct method to determine a bench mark for clinical analysis. Recently a study validated the prognostic role of the LND in a medium sized population of 60 patients with carcinoma penis treated between 1990 and 2008, using RFS as their primary endpoint.^[25] The authors predicted that 7th TNM staging system and LND were the most accurate models and that it significantly differed from TNM criteria alone when compared with the previous models. To our misfortune, the limited number of patients included in the present analysis bars us from determining an additional threshold to significantly distinguish between patients with very low 5-year overall survival rates and individuals at higher risk of disease-related events. Another recent study, using patients from the SEER database, calculated a LND intersect of 33%, but a significant proportion of their cases had insufficient ILND. While their median lymph nodes removed (16 LN) was slightly greater than ours (22 LN, including both groups), the IQR of 13.5 to 23.50 LN highlighted the adequacy of ILND which would validate LND calculations as well as supplement survival statistics. In patients with bulky

disease, nodes can be conjoined and an accurate estimation cannot be made out so our study group did not include any case with matted nodes that could affect our analysis. As per our results, a LND threshold of 40% provided the most precise discrimination of the aftereffect of patients with lymph node positive carcinoma penis. Specifically, the 5-year CSS rates was 77.3% in LND <40 % vs 33.9 % in patients with LNR $\geq 40\%$ ($p < 0.01$). In the aftermath of recent analysis we discovered some limitations that may have affected the present study. Although our series ($n=33$) effectively compares with the two previous studies on carcinoma penis, we must recognize that our numbers remain relatively small, which may limit the prognostic significance of our findings.

A few non RCT's have reported on the efficacy of multidrug chemotherapy in advanced carcinoma penis.^[26,27] Recently, a 10% complete pathological remission rate has been reported in a study with carcinoma penis undergoing neoadjuvant treatment.^[28] These observations support the use of chemotherapy in patients with high disease burden. However, the potential curative intent of chemotherapy has not been validated and RCT's are required to ascertain the effect of chemotherapy in delaying disease advancement. Our study was constrained by the retrospective model as well as the small size of our patient pool and may limit the prognostic implication of our analysis. The extent of patients undergoing ILND represents another catch in the management of carcinoma penis. A more extended ILND increases the number of lymph node yield and a wide spectrum ILND may affect the LND, although the curative footprint of pelvic lymph node dissection is still debatable. In addition, we relied on a single genitourinary pathologist who examined all lymph node specimens which may be source of bias.

V. CONCLUSION

The evolving treatment of carcinoma penis may change the prognostic significance of our LND cutoff, and additional studies are needed to validate this finding in other cohorts. The present retrospective analysis represents the largest single-institutional attempt to validate the prognostic value of the LND in patients with squamous cell carcinoma penis. It's an endeavor to discover optimal mechanisms for the management of patients with LN metastasis with carcinoma penis. Although our analysis awaits independent validation, it supports the use of the LND in clinical practice as an accurate prognostic factor. As the final target should be the individualization of therapy according to risk stratification in patients with LND from penile cancer undergoing radical surgery, we still need to identify and validate further accurate and clinically useful prognostic parameters. We propose that optimal cutoff point for LND and its prognostic significance can be confirmed

by multivariate analysis. Furthermore LND and contemporary TNM staging have emerged as a powerful predictive factor for RFS but there is still an unmet need to identify further accurate and clinically useful prognostic parameters.

Abbreviations:

ILND: Inguinal Lymph Node Dissection.

LND: Lymph Node Density.

OS: Overall Survival.

RFS: Recurrence Free Survival.

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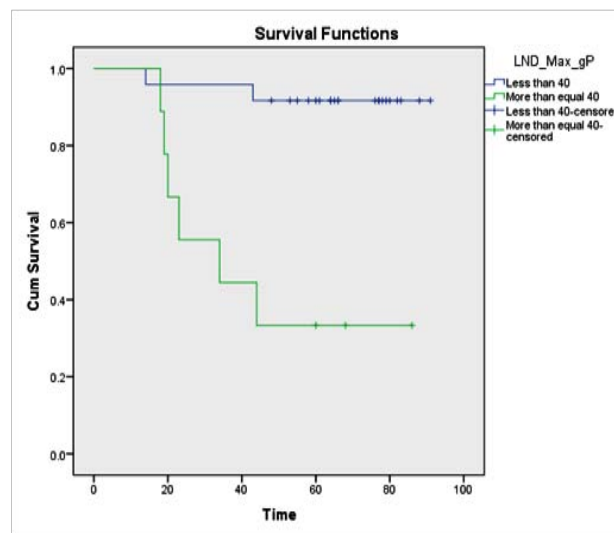


Fig. 1: Kaplan - Meier survival curve showing RFS rates in the overall population of patients with penile cancer and LN metastasis (n=33)

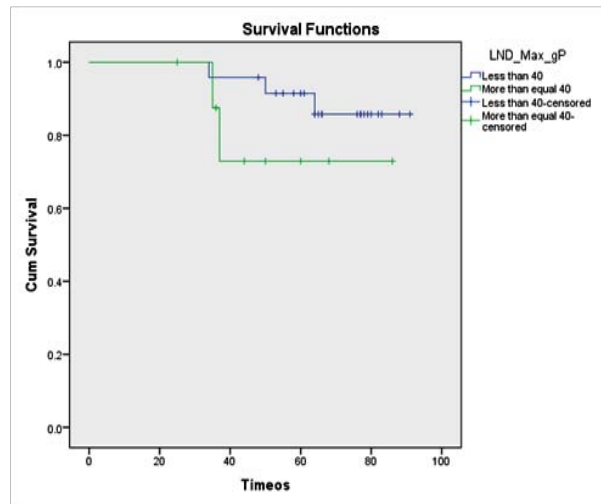


Fig. 2: Kaplan - Meier survival curve showing OS rates in the overall population of patients with penile cancer and LN metastasis (n=33)

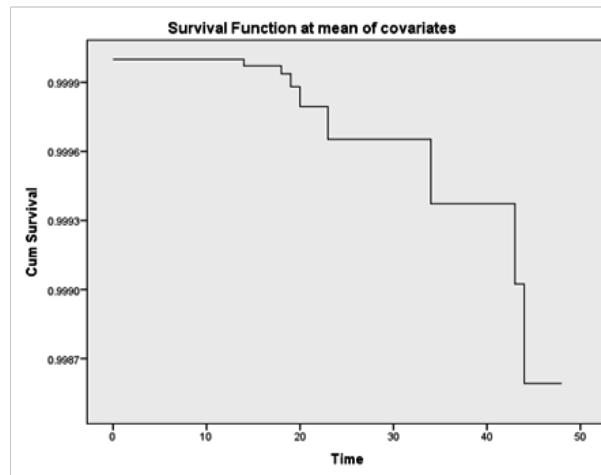


Fig. 3: Multivariate Cox regression analysis curve showing predictors for RFS rates in the overall population of patients with penile cancer and LN metastasis (n=33)



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Assessment of the Level of Adaptation and Disadaptation of Athletesbody to Physical Loads

By Yulduz N. Yusupova & Abdugafur A. Khadjimetov

Abstract- Data of blood indicators, oral fluid and lactate in rowers on canoes, canoes at rest and under the influence of complex of physical activities of different directions are given in this study. The study involved canoeists and canoes (21 athletes, men and women, aged 19-25 years), who are at the general preparatory stage of the preparatory training period. The dynamics of the content of histamine in the oral fluid and blood as well as the content of lactate in the blood were investigated. The results of the study indicate the degree of adaptation of the athlete to physical activity.

Keywords: sport, physical activity, adaptation, histamine, lactate.

GJMR-K Classification: NLMC Code: WD 705



ASSESSMENT OF THE LEVEL OF ADAPTATION AND DISADAPTATION OF ATHLETES BODY TO PHYSICAL LOADS

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Assessment of the Level of Adaptation and Disadaptation of Athletesbody to Physical Loads

Yulduz N. Yusupova ^α & Abdugafur A. Khadjimetov ^σ

Abstract- Data of blood indicators, oral fluid and lactate in rowers on canoes, canoes at rest and under the influence of complex of physical activities of different directions are given in this study. The study involved canoeists and canoes (21 athletes, men and women, aged 19-25 years), who are at the general preparatory stage of the preparatory training period. The dynamics of the content of histamine in the oral fluid and blood as well as the content of lactate in the blood were investigated. The results of the study indicate the degree of adaptation of the athlete to physical activity.

Keywords: sport, physical activity, adaptation, histamine, lactate.

I. INTRODUCTION

The main goal of top-level sports is to achieve the highest possible sports results in sports competitions, competitions of various levels, and, of course, at the Olympics. Any athlete's highest achievement is not only personal for him, but also becomes a national treasure, as records and victories in major international competitions contribute to maintaining and strengthening the country's authority in the international arena.

One of the most important problems of modern sports science and practice is the problem of adaptation to muscular activity. It is known that any adaptation is the output of a biosystem to a new level of homeostasis. In this case, the first rebuilt regulatory mechanisms. Only after this, physiological or morphological changes occur^[1,2,3]. Constantly increasing amounts of training loads can cause a number of violations of the functional state of athletes, lead to overstrain of body systems, increase injuries, reduce the level of athletic performance, as well as reduce the duration of performances at the stage of conservation of sports achievements^[4,5]. To prevent these phenomena, constant monitoring of the functional state of athletes is required using a number of methods that allow assessing readiness to perform significant loads, speed of regenerative processes, efficiency of functioning of various physiological systems, degree of mobilization and use of reserve capabilities of the body, orientation and efficiency of training effects of loads. Since the state of the adaptive capabilities of the body of the rowers in a canoe and canoe at the general preparatory stage of the preparatory period contributes to the formation of a

functional base that ensures the implementation of large volumes of special work. In turn, monitoring the capabilities of the body during stress loads allows you to evaluate the effectiveness of the training process at the subsequent stages of preparation, which makes this study relevant^[6]. The commonly used laboratory methods (determination of lactate, glucose, urea, etc.) involve the determination of the final or intermediate products of a particular type of metabolism and do not give an idea of the regulatory processes in the body. Quite simple methods are needed that allow in a short time to give an objective conclusion about the degree of adaptation of the athlete's body to physical exertion.

The purpose of this study is to study the dynamics of free histamine and lactate in biological fluids at rest and after exercise in rowers in canoes and canoes at the general preparatory stage of the preparatory training period.

II. MATERIAL AND METHODS

The studies were carried out in the Republican Scientific and Practical Center for Sports Medicine with the participation of 24 paddlers in canoes and canoes (men and women, age 19-25 years old, sports qualifications MS, MSIC) are in the general preparatory stage of the preparatory training period. The control group consisted of 16 functionally healthy students.

To determine histamine from athletes, they collected 1 ml of oral liquid in a centrifuge tube with 4 ml of 10% trichloroacetic acid. Then in the centrifugate was determined the level of histamine. Determination of histamine in various biological fluids (serum and oral fluid) was performed by gas-liquid chromatography with a Thermo Scientific TSQ 8000 EVO mass spectrometer GC-MS / MS.

In parallel, at rest and after exercise, the blood lactate content was determined using the ROSH biochemical analyzer COBAS-311 using reagents of the same company.

To assess the functional state of the cardiovascular system at rest and after exercise, the heart rate (HR) was determined - by palpation on the radial artery or by means of an electrocardiograph, blood pressure (DC) - by auscultatory method. Testing was performed on the Treadmill runway (Germany). Gas analysis of exhaled air was carried out with the MetaMax high-speed automatic gas analyzer. Pulse modes were recorded using a Sport Tester heart rate monitor. The

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obtained experimental data were processed on a computer using the generally accepted methods of mathematical statistics.

III. RESULTS

The prevalence of allergic diseases among the population of the planet, which according to WHO data currently makes up about 40%, cannot but concern athletes of the highest achievements. As shown by our studies in athletes of higher achievements who suffered from recurrent symptoms of the nose (nasal congestion, rhinorrhea, etc.), an allergic reaction (AR) was identified. Thus, according to foreign data, it was found that more often the AR was detected in water sports (swimming, rowing, diving), in comparison with waterless sports. In such susceptible individuals, cold dry air leads to the release of mediators from the fat (histamine, tryptase) and epithelial cells, which may be secondary to the increased osmolality of mucous secretions.

As can be seen from the results of the study, in the process of stress in athletes rowing on kayaks and canoes, an individual "histamine profile" is formed, characterized by an increased content of free histamine in the body. In this situation, the medical examination and electrocardiography did not detect any abnormalities in the health status of this group of athletes with consistently high concentrations of histamine in biological fluids. Obviously, a high level of free histamine in the blood in this case represents a long-term sustainable adaptation to regular physical exertion. This is also true in relation to the adaptive increase in the level of histamine in the athlete's oral

fluid. It is possible that a high level in the body of athletes (formed as an adaptation to high physical exertion) in the future may lead to the development of an allergic disease.

In the majority of the sportsmen surveyed, the lactate content at rest mostly corresponded to the normative one. Nevertheless, in four rowers, the lactate values at rest were significantly higher than the upper limit of the norm and amounted to 2.72 - 3.45 mmol/l, which indicates the activation of the glycol-compensatory reaction in the conditions of oxygen deficiency of the body. It is known that the reaction of the body of athletes to the standard load is one of the indicators of the state of their fitness. In this regard, of undoubted interest was the analysis of the reaction of the body of rowers to the standard work on the content of lactate in the blood. Standard exercise, limited by duration and intensity, caused a different reaction of the body to lactate levels in the blood. The most optimal metabolic reaction is the minimum increase in blood lactate in response to the standard load. Such a reaction was noted in most athletes, but in individual subjects the lactate content increased significantly (even (even to 4.38 mmol/l), which indicates their lower fitness relative to other athletes, providing a lower metabolic response of the body to standard work. After performing the load in steps of increasing power, practically all athletes had the maximum metabolic reaction immediately after it was performed. The lactate content was in the range of 8.6-12.7 mmol / l. The high utilization of lactate observed in the overwhelming majority of rowers indicates their high fitness.

Table 1: The Content of Histamine in the Oral Fluid and Lactate in the Blood Serum of Athletes Rowing on Canoes and Canoes

	Control Group	Resting State	After Standard Load	After Loading Stepwise Increasing Power, 10 S;
Histamine in Oral Fluid ($\mu\text{M} / \text{L}$)	7.83 ± 0.82	8.01 ± 0.56	9.21 ± 0.78	$15.84 \pm 1.32^*$
Histamine in Serum ($\mu\text{M} / \text{L}$)	12.34 ± 0.87	11.67 ± 0.97	12.33 ± 1.05	$19.51 \pm 2.03^*$
Blood Lactate mmol / l	1.56 ± 0.11	2.03 ± 0.19	$2.56 \pm 0.18^*$	$10.76 \pm 1.09^*$

Note: * - significance of differences $P < 0.05$;

When resting - HR-72 beats/min.

After a standard HR load of 130 BP/min.

After a load of stepwise increasing power, 10 sec.- HR-180 BP/min.

IV. DISCUSSION

Numerous studies have shown that intensive training work in athletes leads to a decrease in the level of free histamine in the body. During the rest period, its content returns to its original level, passing through the stage of super restoration. With sufficiently intense loads, an increase in its level compared to the baseline is observed approximately a day after work. Thus, with daily training sessions, it is possible that each

subsequent workout occurs during the super-recovery phase in the histamine system after the previous one. As a result of the cumulative training effect, its content in the body in this case increases. As training improves, the recovery of histamine used in the process starts to occur at a faster rate. In this case, the over-recovery phase ends earlier than the next workout begins. Thus, the impact of regular training loads leads to a steady increase in the level of free histamine in the body of athletes at rest. The accumulation of substances with

such a high biological activity, cannot affect the various aspects of the life of the athlete. On the one hand, a high level of histamine in biological fluids is associated with a number of pathological conditions, such as infectious-inflammatory processes, allergies; on the other hand, there is evidence of better athletic performance in athletes with higher levels of histamine. To clarify this issue, we conducted a study among athletes (rowing and canoeing) the effects of the initial level of histamine in the body on physical performance when working in different modes. As shown by the study, for each mode of operation there is its optimal level of histamine in the oral fluid and in the blood. Thus, the accumulation of free histamine in the body while reducing the oxygen capacity of the blood is aimed at preventing the weakening of the oxidative function, which is possible due to the ability of histamine to increase coronary circulation, dilate blood vessels, increase local blood flow and, thus, improve the blood supply to the heart and other organs and tissues with oxygen. In addition, the revealed numerous relationships between the content of free histamine in biological fluids and the performance of the cardiovascular system (arterial and pulse pressure, heart rate, etc.) confirm that the control effect of histamine on the processes of adaptation to muscle activity is mediated by influence on blood circulation. During intensive muscular work, the body functions under conditions of partial anaerobiosis. Under the influence of regular training and competitive loads, a long-term adaptation of the athlete's body to the conditions of oxygen deficiency occurs. In particular, the body creates a "stock" of histamine, which allows to a certain extent to weaken the effect of hypoxia. Accumulating in the body, histamine inhibits acetyl cholinesterase, thereby increasing the concentration of acetylcholine and activating the parasympathetic division of the autonomic nervous system. Under the influence of increased parasympathetic regulation and the direct action of histamine on the heart and blood vessels, the level of functioning of the circulatory system in a state of rest decreases. As you know, during physical exertion, the strength and heart rate, blood pressure increase, but this increase is limited by the capacity of the heart. Therefore, at low values of these indicators at rest, the range of their possible changes expands, i.e. low level of functioning of the circulatory system at rest creates a reserve for its activation during operation.

The analysis of the staged state of such indicators as histamine and lactate in the blood made it possible to assess the state of fitness of athletes of the studied group, both at rest and after exposure to a complex of loads of different energy orientation. Similar dynamics were observed with respect to histamine in the oral fluid. The results indicate that an individual correction of the athlete's condition is carried out, to give nutritional and pharmacological recommendations

with a view to optimizing it, which can contribute to the enhanced solution of specific tasks facing athletes in the general preparatory stage of the preparatory training period.

V. CONCLUSION

1. The reaction to the standard load on the content of histamine in the oral fluid and blood as well as the content of lactate in the blood made it possible to draw a preliminary conclusion about the degree of fitness of the examined contingent of athletes.
2. Rowers had a prolonged production of histamine and lactate release into the blood for 10 seconds of recovery, which causes its delayed utilization.
3. A positive correlation is noted between the value of the heart rate and the content of histamine and lactate in the blood for 10 s recovery after performing the load in steps of increasing power.
4. Monitoring indicators of histamine and lactate in the blood of athletes at the general preparatory stage of the preparatory training period made it possible to evaluate and outline ways for its correction. Further study of the dynamics of lactate and histamine in the oral fluid and blood during different periods of preparation will improve the effectiveness of training loads and predict athletic performance by optimizing the metabolism of athletes.

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Consent

It is not applicable.

Ethical Approval

All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

Declaration of Interests

Authors declare that there is no competing interests.

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Invasive Bacterial and Malaria Co-Infection and Associated Factors among Pregnant Mothers Attending a Private Tertiary Teaching Hospital in South Western Rural Uganda: Retrospective Review of Records between Feb 2014-Feb 2018

By Collins Atuheire, Nakibuuka Lydia, Mark Agaba, Justine Okello,
Esther Itamba, Hilda Amolo Doreen & Paul Ssajakambwe

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Abstract- Introduction: Malaria and invasive bacterial infection (IBI) adversely affects the prognosis of pregnant women, globally. However, in sub-Saharan Africa, data on IBI and malaria co-infection among pregnant women is scarce, rather most studies look at Malaria and Invasive bacterial infection dis-jointly.

Objectives: We aimed to study the burden of Invasive bacterial and malaria co- infection and associated factors among pregnant mothers attending a private hospital in rural South-Western Uganda.

Methods: A cross-sectional study involving retrospective review of records was carried out. The records about pregnant women attending ante-natal clinic at Kampala International University teaching hospital were collected from February 2014 to February 2018. Waiver of consent was thought from relevant Ethical committees. Checklists were used to capture relevant data and data analysis was done using STATA version 14.2. Descriptive statistics, binary crude and adjusted logistic regression were carried out.

GJMR-K Classification: NLMC Code: WC 195



Strictly as per the compliance and regulations of:



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Invasive Bacterial and Malaria Co-Infection and Associated Factors among Pregnant Mothers Attending a Private Tertiary Teaching Hospital in South Western Rural Uganda: Retrospective Review of Records between Feb 2014-Feb 2018

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Abstract- Introduction: Malaria and invasive bacterial infection (IBI) adversely affects the prognosis of pregnant women, globally. However, in sub-Saharan Africa, data on IBI and malaria co-infection among pregnant women is scarce, rather most studies look at Malaria and Invasive bacterial infection dis-jointly.

Objectives: We aimed to study the burden of Invasive bacterial and malaria co- infection and associated factors among pregnant mothers attending a private hospital in rural South-Western Uganda.

Methods: A cross-sectional study involving retrospective review of records was carried out. The records about pregnant women attending ante-natal clinic at Kampala International University teaching hospital were collected from February 2014 to February 2018. Waiver of consent was thought from relevant Ethical committees. Checklists were used to capture relevant data and data analysis was done using STATA version 14.2. Descriptive statistics, binary crude and adjusted logistic regression were carried out.

Results: Of 367 pregnant women enrolled, the prevalence of Malaria-IBI co-infection was 26% (95%CI 22-31). Being of Muganda tribe was associated with reduced risk of IBI-malaria co-infection as compared to Munyankole; aOR= 0.33, [95% CI 0.11-1.02]. Mothers aged over 28 years were 56% less likely to experience IBI-malaria co-infection as compared to adult mothers below 22 years; aOR=0.44, [95% CI=0.21-0.92]. Those with history of blood transfusion were more likely to have IBI-malaria co- infection compared to those who have

never had history of blood transfusion; aOR=1.65, [95%CI=0.96-2.84].

Conclusions & Recommendations: The prevalence of IBI-malaria co-infection was high among pregnant women attending Kampala International Teaching hospital of Western Uganda. Factors like age, tribe were significant predictors of the co-infection. There is need for emphasis on education of young mothers on issues of malaria prevention. Health care providers should prioritize attention towards young women especially non-indigenous ethnic groups in this setting as well as not undermining the history of blood transfusion regarding risk of IBI-Malaria co-infection.

I. INTRODUCTION

Pregnant mothers with malaria and IBI have been associated with increased risk of adverse birth outcomes such as stillbirths, preterm, intrauterine growth restriction (IUGR) and Low Birth weight (LBW)[1]. Bacterial diseases contribute a big part towards sexually transmitted infection (STI) and reproductive tract infections (RTI) such as *Treponema pallidum*, *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, *Trichomonas vaginalis* and bacterial vaginosis [2]. Also likely bacteria to be found in blood include; *Staphylococcus aureus*, *Salmonella Typhi*, Other *Salmonella* serovars, *Viridans streptococci*, *Streptococcus pneumoniae*, *Brucella* species, *Streptococcus pyogenes*, *Haemophilus influenzae*, *Enterococcus faecalis*, *Pseudomonas aeruginosa*, *Clostridium perfringens* *Klebsiella* strains, Anaerobic streptococci *Escherichia coli*, *Proteus* species, *Bacteroides fragilis*, *Neisseria meningitidis*, *Yersinia pestis* [3]

In Sub-Saharan Africa, data on IBI and malaria co-infection among pregnant mothers is indeed scarce except data on Malaria and Bacteraemia that is independently available among pregnant mothers. The pooled prevalence of malaria among pregnant women in various regions show variation in a study by Chico *et al.*, peripheral malaria 32.0%, and placental malaria 25.8% in East and Southern Africa also peripheral

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malaria 38.2% and placental malaria 39.9% in West and Central Africa[4].

Malaria affects mainly young pregnant mothers. In area with a high endemicity, occupants tend to acquire high immunity with age against *P. falciparum* and it is usually asymptomatic in pregnancy, though the parasite may be present in the placenta and contribute to maternal anaemia [5], this was evidenced from a study by Namusoke *et al.*, at Mulago National referral hospital, Uganda where a prevalence of 9% and 13.9% using peripheral smear and placental histology respectively was obtained and anaemia was the major significant factor[6].

Invasive bacterial infections have not been documented among pregnant women in Uganda.

II. MATERIALS AND METHODS

a) Study design

A cross-sectional study design involving a retrospective review of records was carried out. This study was conducted between the months of June and July 2018 among pregnant women who attended KIU-TH for antenatal services between February 2014 to February 2018.

b) Study area

Kampala International University Teaching Hospital is located in Ishaka town which is a municipality in Bushenyi District. Ishaka is found approximately 62 kilometers west of Mbarara town. Ishaka has a population of 16,646 where females are 8,840 (UBOS, 2014). Kampala International University Teaching Hospital has a bed capacity of 700, providing both out-patient and in-patient services. The catchment areas of Kampala International Teaching Hospital include Bushenyi, Sheema, Rubirizi, Mitooma and other neighboring districts of Western Uganda. The study was conducted in the records office of ante-natal clinic of KIU-TH.

c) Study population

Inclusion criteria: All records of adult pregnant women [from Bushenyi and neighboring districts; and who have stayed in this area for more than 2 years] and who attended KIU-TH for antenatal services from February 2014 to February 2018. Records with missing CBC results as well as results were excluded from the study.

d) Sample Size and Sampling Method

Consecutive enrolment of patients was carried out, that is, file by file of records was sampled consecutively. Sample size was determined using Daniel's formula.

e) Data Collection

Socio-demographic factors and laboratory data were obtained from patients' file. Complete Blood Count (CBC) records were used to determine whether an

individual was invasively infected with bacteria. An individual was considered to have IBI when they had highly elevated neutrophil count (neutrophilia)[3]. A patient was considered malaria positive when they had their Rapid Diagnostic Test (RDT) positive and positive peripheral thick smear. At Kampala International teaching Hospital laboratory, microscopic examination of thick blood smears for malaria parasite was used as a gold standard and a confirmatory test for malarial infection. All preparations and testing were done following the Uganda Ministry of Health standard operating procedures[7].

f) Statistical analysis

Collected data were extracted from patient files onto data check lists and then entered into excel version 2010. Data was then exported and analyzed using STATA 14.2 (Statacorp 4905 Lakeway Drive, College station Texas 77,845 USA) to obtain the prevalence as frequencies and percentage and we constructed respective 95% confidence interval (95% CI). Crude binary and adjusted logistic regression were used to assess the association between malaria, IBI and co-infection (outcomes) and associated factors (predictors). Measures of association, that is; crude odds ratios (cOR) and adjusted odds ratios (aOR) were reported with their respective 95%CI and p-value. Those factors whose p-value ≤ 0.05 were considered statistically significant.

g) Ethical clearance

The fact that the study involved secondary data waiver of consent was sought from the two ethical committees, that is, School of Public Health, College Health Sciences, Makerere University and Research Ethics Committee of Kampala International University-Western Campus in order to conduct research.

III. RESULTS AND DISCUSSION

a) Socio-demographic characteristics of pregnant women who participated in the study

Table 1: A total of 367 pregnant women were enrolled. A greater number of pregnant mothers were aged between 21-24 years, 31.3% and with majority being peasants, 60%. About 57% of the mothers had attained secondary school education. Most of these mothers were married 77.1%. Participants belonging to Banyankoletribe were the majority 51% as well as Catholics, 34.9%. About 59% of mothers were multiparous.

Table 1: Showing frequency of socio-demographic characteristics of participants attending antenatal care at KIU-TH, in Ishaka-Bushenyi district.

Variable	Frequency	percentage
Age in yrs		
16-20	80	21.8
21-24	115	31.3
25-29	98	26.7
30-34	55	15.0
>35	19	5.2
Education		
primary	96	37.6
secondary	210	57.2
tertiary	61	5.2
Occupation		
Trader	20	5.4
Teacher	34	9.2
Peasant	220	60.0
other	93	35.0
Tribe		
Banyankole	187	51.0
Baganda	39	10.6
Bakiga	88	24.0
other	53	14.4
Religion		
Catholic	128	34.9
Protestant	103	28.1
Muslim	91	24.8
SDA	45	12.2
Marital status		
Married	283	77.1
Not married	84	22.9
Gravidity		
Prime gravida	152	41.4
multigravida	215	58.6

b) *Prevalence of invasive bacterial infection, malaria and co-infection among pregnant attending antenatal care at KIU-TH, Western Uganda*

Table 2 Of the 367 participants enrolled into the study, 97 (26%) had co-infection of malaria and invasive bacterial infection. About 69% had malaria infection and 57% of the mothers had invasive bacterial infection.

Table 2: Prevalence of invasive bacterial infection and malaria among pregnant women attending antenatal care at KIU-TH, Ishaka-Bushenyi district, Western Uganda

Variable	Frequency	Percentage	95%CI
Confection			
Yes	97	26.4	22.1-31.2
No	270	73.6	68.8-77.9
Malaria infection			
Yes	255	69.4	64.4-74.0
No	112	30.6	26.0-35.5
Invasive bacterial infection			
Yes	209	57.0	51.9-62.0
No	158	43.0	38.0-48.1

c) *Bivariate analysis of factors that influence invasive bacterial and malaria co-infection among pregnant mothers attending KIU-TH, Western Uganda*

Table 3: Pregnant mothers aged over 28 years were 3 times more likely to have malaria and invasive bacterial infection (IBI) compared to those aged 18-21 years, (cOR=3.07, 95% CI1.56-6.03) and also those aged 21-28 years were almost as twice as likely to have IBI-malaria co-infection as adult mothers aged less than 22 years (cOR=1.84, 95%CI0.99-3.43).

Participants with previous history of Blood transfusion were 2 times more likely to have malaria and invasive bacterial infection compared to those who were not previously transfused, (cOR=1.80, 95%CI1.07-3.02). Mothers who belonged to Baganda ethnic group were

77% less likely to have malaria and invasive bacterial infection compared to their counterparts that belonged to Banyankole ethnic group (cOR=0.23, 95%CI0.08-0.68). Individuals who had completed tertiary level of education were 55% less likely to experience malaria and invasive bacterial infection compared to those who had completed primary level of education (cOR=0.45, 95%CI0.20-1.0). Additionally, mothers who attended full antenatal care (n=4 visits) were 26% less likely to have malaria and invasive bacterial infection compared to those who never attended full ANC (n<4 visits), (cOR=0.74, 95%CI0.29-1.89), likewise also those who were married, were less likely to have IBI-malaria co-infection as compared to unmarried mothers, (cOR=0.9395% CI 0.53-1.62).

Table 3: Bivariate and Multivariate analysis of factors affecting invasive bacterial and malaria co infection among pregnant mothers attending KIU TH

Variable n (%)	Co-infection		Bivariate analysis			Multiple analysis		
	No=267	Yes=95	cOR	95%CI	p-value	aOR	95%CI	p-value
Age in years								
18-21	87(83.6)	17(16.4)	1.00			1.00		
22-28	125(73.5)	45(26.5)	1.84	0.99-3.43	0.05	0.72	0.40-1.29	0.27
>28	55(62.5)	33(37.5)	3.07	1.56-6.03	0.001	0.44	0.21-0.92	0.03
Blood transfusion								
No	211(76.5)	65(23.5)	1.00			1.00		
Yes	56(64.4)	31(35.6)	1.80	1.07-3.02	0.03	1.65	0.96-2.84	0.07
Tribe								
Munyankole	124(67.0)	61(33.0)	1.00			1.00		
Muganda	35(89.7)	4(10.3)	0.23	0.08-0.68	0.008	0.33	0.11-1.02	0.05
Mukiga	63(72.4)	24(27.6)	0.77	0.44-1.36	0.37	0.93	0.51-1.69	0.81
Other	45(86.5)	7(13.5)	0.32	0.13-0.74	0.008	0.34	0.13-0.84	0.02
Toilet use								
No	7(58.3)	5(41.7)	1.00					
Yes	260(74.1)	91(25.9)	0.49	0.15-1.58	0.23			
Education								
Primary	65(69.2)	29(30.8)	1.00			1.00		
Secondary	152(72.7)	57(27.3)	0.84	0.49-1.43	0.52	1.12	0.63-1.97	0.70
Tertiary	50(83.5)	10(16.7)	0.45	0.20-1.00	0.05	0.69	0.29-1.62	0.39
Full Antenatal care visits								
No (n<4 visits)	245(73.1)	90(26.9)	1.00					
Yes (n=4 visits)	22(78.6)	6(21.4)	0.74	0.29-1.89	0.53			
Marital status								
Un married	205(73.2)	75(26.8)	1.00					
Married	62(74.7)	21(25.3)	0.93	0.53-1.62	0.79			
Occupation								
Trader	14(70.0)	6(30)	1.00					
Teacher	26(78.8)	7(21.2)	0.63	0.18-2.24	0.47			
Peasant	160(73.1)	59(26.9)	0.86	0.32-2.34	0.77			
Other	67(73.6)	24(26.4)	0.84	0.29-2.42	0.74			
Smoking & alcohol								
No	175(71.7)	69(28.3)	1.00					
Yes	92(77.3)	27(22.7)	0.74	0.45-1.24	0.26			

d) *Multivariate analysis for factors that influence invasive bacterial and malaria co-infection among pregnant mothers attending KIU-TH*

Table 3: After multivariate analysis, participants aged over 28 years were 56% less likely to have malaria and invasive bacterial infection compared to adult mothers aged less than 22 years, (aOR=0.44, 95% CI 0.21-0.92). Mothers belonging to Baganda ethnic group were 67% less likely to have malaria and invasive bacterial infection compared to those of Banyankole origin, (aOR=0.33, 95% CI 0.13-0.84). Participants who had previous history of blood transfusion were about 2 times more likely to have malaria and invasive bacterial co-infection compared to those who were not previously transfused (aOR=1.65, 95% CI 0.96-2.84). Mothers with tertiary education were less likely to be co-infected compared to those with primary education (aOR=0.69, 95% CI 0.29-1.62), however, the association was not statistically significant.

IV. DISCUSSION

Several studies in literature have investigated the burden of malaria and invasive bacterial infection in children living in Sub-Saharan Africa with no study reporting the same in pregnant women [8]. In our study, the prevalence of IBI-malaria co-infection was 26.4%. This is much higher than IBI-malaria co-infection in Sub-Saharan African children [9]. There are no comparable literature concerning IBI-malaria co-infection among pregnant women in Uganda. In many studies Malaria has been responsible for Invasive Bacterial infection. During malaria illness, lysis of red blood cells is possible and Iron released may act as nutrients for bacteria [10-12].

We used Complete Blood Count (CBC) neutrophilia picture to predict invasive bacterial infection since limited records for blood cultures were available which were for the study period set for this study. During bacteraemia, there is usually neutrophilia with left shift [3]. However other causes that can lead to increase neutrophils including viral infections, all kinds of stress, pregnancy, Connective tissue diseases, tissue necrosis, acidosis of various etiologies, (e.g., nephrogenic diabetes insipidus), medications [13]. Hence these may account for the high prevalence obtained in this study.

The prevalence of malaria in our study was high, 69.4%. Malaria burden in this area could be due to much vegetation cover that facilitates mosquito breeding [14]. Our findings about malaria burden are higher than those obtained among pregnant women in Mulago national referral hospital Kampala [6]. The discrepancy is due to several factors including poorer coverage on insecticide mosquito treated Mosquito nets as well as health education regarding malaria prevention [15].

The prevalence of Invasive Bacterial infection among pregnant women was 57%. This could be attributed to poor hygiene practices in semi-urban setting. The elevated IBI reported could be an over estimation since classifying IBI was based on raised neutrophils in Blood under febrile conditions, neutrophils may not be necessarily due to Bacteremia [16, 17]. As envisaged, this study area is endemic with malaria that contributes to Invasive Bacterial Infection through several mechanisms such as increase in the permeability of the gut [18]. This enables the breakdown of the gut-blood barrier providing a way for invasion by bacteria. Also sequestration of the Parasite has been demonstrated in the intestines of patients suffering fatal cerebral malaria [19].

In our study, several factors were found to be associated with Malaria-Invasive bacterial infection among pregnant mothers in Kampala International University-Teaching Hospital.

In this study, it was found that pregnant women with tertiary level of education were less likely to have IBI-malaria co-infection as compared to those with primary education. This can be explained by insufficient knowledge, low income and economic status of primary education holders and hence cannot afford timely medical care. Lack of timely medical care often complicates bacterial infection to invasive stages [15, 19, 20].

Mothers aged 18-21 years were more likely to contract Malaria-Invasive bacterial co-infections as compared to mothers aged 28 years and above. This may have been attributed to low knowledge and laxity of young mothers concerning hygiene and other preventive methods of bacterial and malaria infections. Young mothers in this setting tend to go night clubbing with their non-pregnant peers and eventually get mosquito bites. Also, most young mothers of this age group in Bushenyi-Ishaka have no official husbands that could support them financially and so do indulge in hard and unhygienic activities that have potential of predisposing them to Malaria and bacterial infections simultaneously.

Pregnant mothers with previous history of blood transfusion were more likely to experience IBI-Malaria co-infection compared to those that had not had previous history of IBI-malaria co-infection. In the study setting, malnutrition is on a higher tide among pregnant women and anemia is not uncommon in the pregnant mothers that in most cases calls for blood transfusion. Transfused patients in this area may not recover well from anemia since household food basket is constant. Persistent anemia facilitates lowered immunity and hence capable of propagating Malaria-IBI co-infection [21].

Also mothers who attended all the antenatal care visits throughout the period of pregnancy were less likely to get IBI-malaria co-infection compared to those who attended less than the minimum recommended

(i.e less than 4 visits). During these antenatal visits, patients receive health services such as IPT and health education regarding prevention methods about malaria and bacterial diseases during pregnancy[4].

Furthermore, our study shows that the co-infection was associated with tribes/ethnicity. Baganda are less likely to be diagnosed with Malaria-IBI infection as compared to Banyankole tribe. Baganda migrate from central Uganda to Bushenyi (study area) to conduct business and so majority of them stay in towns not surrounded by potential breeding places for mosquito. Most Banyankole women stay in remote villages where mosquito bites are not uncommon.

V. WEAKNESSES AND STRENGTHS

Being secondary data, critically important variables like result for culture and sensitivity were absent, no data on clinical signs and symptoms were obtained. Also, a number of socio-demographic factors and clinical factors such as hemoglobin level were not obtained. This limited, comprehensiveness of our work. However, variables that we so far analyzed are potential of providing findings that can guide clinical approaches and can simultaneously provoke further research.

Additionally, we did not achieve sample size calculated due to limited study records. However, this sample size is moderately sufficient for the few variables (inherent with secondary data studies) and so internal validity is likely [22]. Our work, though does not consist culture and sensitivity, is the first of its kind to be done in South Western Uganda and is consistent with treatment approaches of fevers other than Malaria in our resources constrained health setting.

VI. CONCLUSIONS

The prevalence of invasive Bacterial and malaria co-infection among pregnant women attending Kampala International University Teaching Hospital of peri-urban Western Uganda is high. Education level, previous history of blood transfusion and being of Munyankole tribe were potential predictors of Invasive Bacteria-Malaria co-infection.

There is a need for Uganda ministry of health to train antenatal health care providers about awareness and management of IBI-Malaria among pregnant women in Uganda especially those from rural areas where there is possibility of malaria endemicity. Also there is need to equip peripheral health facilities with laboratories that can carry out culture and sensitivity tests for the diagnosis of Invasive bacterial infections.

There is need to sensitise communities about the issue of IBI-Malaria co-infection especially among the mothers with primary level education and lower concerning possible prevention strategies of IBI-Malaria co-infection.

Further studies are recommended using more specific and more sensitive techniques such as culture and sensitivity in identification of the IBI in abnormally raised neutrophils in pregnant women.

Data Availability

Data used to support these findings are available from the corresponding author upon request.

Conflicts of Interest

The authors declare that there is no conflict of interest regarding publication of this article.

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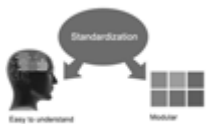
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AUXILIARY MEMBERSHIPS

Institutional Fellow of Open Association of Research Society (USA) - OARS (USA)

Global Journals Incorporation (USA) is accredited by Open Association of Research Society, U.S.A (OARS) and in turn, affiliates research institutions as “Institutional Fellow of Open Association of Research Society” (IFOARS).

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The IFOARS institution is entitled to form a Board comprised of one Chairperson and three to five board members preferably from different streams. The Board will be recognized as “Institutional Board of Open Association of Research Society”-(IBOARS).

The Institute will be entitled to following benefits:



The IBOARS can initially review research papers of their institute and recommend them to publish with respective journal of Global Journals. It can also review the papers of other institutions after obtaining our consent. The second review will be done by peer reviewer of Global Journals Incorporation (USA). The Board is at liberty to appoint a peer reviewer with the approval of chairperson after consulting us.

The author fees of such paper may be waived off up to 40%.

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The IBOARS can organize symposium/seminar/conference in their country on behalf of Global Journals Incorporation (USA)-OARS (USA). The terms and conditions can be discussed separately.

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We shall provide you intimation regarding launching of e-version of journal of your stream time to time. This may be utilized in your library for the enrichment of knowledge of your students as well as it can also be helpful for the concerned faculty members.



After nomination of your institution as “Institutional Fellow” and constantly functioning successfully for one year, we can consider giving recognition to your institute to function as Regional/Zonal office on our behalf.

The board can also take up the additional allied activities for betterment after our consultation.

The following entitlements are applicable to individual Fellows:

Open Association of Research Society, U.S.A (OARS) By-laws states that an individual Fellow may use the designations as applicable, or the corresponding initials. The Credentials of individual Fellow and Associate designations signify that the individual has gained knowledge of the fundamental concepts. One is magnanimous and proficient in an expertise course covering the professional code of conduct, and follows recognized standards of practice.



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We shall provide print version of 12 issues of any three journals [as per your requirement] out of our 38 journals worth \$ 2376 USD.

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- The professional accredited with Fellow honor, is entitled to various benefits viz. name, fame, honor, regular flow of income, secured bright future, social status etc.



- In addition to above, if one is single author, then entitled to 40% discount on publishing research paper and can get 10% discount if one is co-author or main author among group of authors.
- The Fellow can organize symposium/seminar/conference on behalf of Global Journals Incorporation (USA) and he/she can also attend the same organized by other institutes on behalf of Global Journals.
- The Fellow can become member of Editorial Board Member after completing 3yrs.
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- • This individual has learned the basic methods of applying those concepts and techniques to common challenging situations. This individual has further demonstrated an in-depth understanding of the application of suitable techniques to a particular area of research practice.

Note :

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- In future, if the board feels the necessity to change any board member, the same can be done with the consent of the chairperson along with anyone board member without our approval.
- In case, the chairperson needs to be replaced then consent of 2/3rd board members are required and they are also required to jointly pass the resolution copy of which should be sent to us. In such case, it will be compulsory to obtain our approval before replacement.
- In case of “Difference of Opinion [if any]” among the Board members, our decision will be final and binding to everyone.

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

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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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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.

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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.

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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.

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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.

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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.

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

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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.

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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.



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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.

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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.

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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.

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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.

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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 through 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.
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- 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.
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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.

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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.

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- 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:

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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.

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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."



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- 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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<i>Introduction</i>	Containing all background details with clear goal and appropriate details, flow specification, no grammar and spelling mistake, well organized sentence and paragraph, reference cited	Unclear and confusing data, appropriate format, grammar and spelling errors with unorganized matter	Out of place depth and content, hazy format
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<i>Result</i>	Well organized, Clear and specific, Correct units with precision, correct data, well structuring of paragraph, no grammar and spelling mistake	Complete and embarrassed text, difficult to comprehend	Irregular format with wrong facts and figures
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<i>References</i>	Complete and correct format, well organized	Beside the point, Incomplete	Wrong format and structuring



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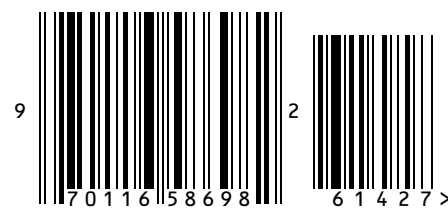
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