Afro-Egyptian Journal of Infectious and Endemic Diseases
المجلة الأفريقية المصرية للأمراض المعدية والمتوطنة
ISSN (Online): 2090-7184
ISSN (Print): 2090-7613

An Official Publication of Endemic and Tropical Medicine Department, Faculty of Medicine, Zagazig University, Zagazig 44519, Egypt

Editor-in-Chief:
Mohamad El-Khashab
E-mail: ajied@zu.edu.eg
elkhashab2005@hotmail.com

Editorial Board:
Zagazig University, Egypt:
Hamed Suliman, Endemic and Tropical Medicine
Amr Murad, Endemic and Tropical Medicine
Faiza Elghohary, Endemic and Tropical Medicine
Salama Elghoniemy, Endemic and Tropical Medicine
Ahmad Mahmoud, Endemic and Tropical Medicine
Samy Eisa, Endemic and Tropical Medicine
Osman Elwerwary, Endemic and Tropical Medicine
Ibrahim Hegazy, Endemic and Tropical Medicine
Nahta Elgammal, Endemic and Tropical Medicine
Mohamad Abdel-Tawab, Endemic and Tropical Medicine
Rashed Hasan, Endemic and Tropical Medicine
Misaa Abdalla, Endemic and Tropical Medicine
Mohamad Imam, Endemic and Tropical Medicine
Nasr Bechit, Endemic and Tropical Medicine
Mostafa Elshamy, Endemic and Tropical Medicine
El-Said Elbadrawy, Endemic and Tropical Medicine
Amira Suliman, Endemic and Tropical Medicine
Eman Abdel-Aal, Endemic and Tropical Medicine
Maged Bahgat, Endemic and Tropical Medicine
Osama Rushdy, Endemic and Tropical Medicine
Walid Abdel-Dayem, Endemic and Tropical Medicine
Ahmad Sakr, Endemic and Tropical Medicine
Abeer Nafee, Endemic and Tropical Medicine
Tarak Zaher, Endemic and Tropical Medicine
Soha Esmat, Endemic and Tropical Medicine
Ghada Salem, Endemic and Tropical Medicine
Sahar Elsharif, Endemic and Tropical Medicine
Hala Ismail, Endemic and Tropical Medicine
Gehan Shawqy, Endemic and Tropical Medicine
Mohamad Refaey, Endemic and Tropical Medicine
Sherif Galal, Endemic and Tropical Medicine
Samah Telep, Endemic and Tropical Medicine
Tagrid Abдалlah, Endemic and Tropical Medicine
Nagla Abdel-Monem, Endemic and Tropical Medicine
Mohamad Saria, Endemic and Tropical Medicine
Noha Shaheen, Endemic and Tropical Medicine
Soha Elhawary, Endemic and Tropical Medicine
Mohamad Hassona, Endemic and Tropical Medicine
Talaat Fathy, Endemic and Tropical Medicine
Mohamad Magdy, Endemic and Tropical Medicine

Ihab Darwish, Endemic and Tropical Medicine
Ashraf Metwaly, Endemic and Tropical Medicine
Mohamad Emara, Endemic and Tropical Medicine
Ahmad Behiry, Endemic and Tropical Medicine
Reda Lami, Parasitology
Samia Etewa, Parasitology
Mohiddin Abdel-Fattah, Parasitology
Alaa Elgendy, Parasitology
Ahmad Shaheen, Microbiology
Ayman Marii, Microbiology
Shimaa Abdel-Azim, Microbiology
Marwa Abdel-Azim, Microbiology
Mahmoud Wahid, Pathology
Sahar Zagloul, Internal Medicine
Khaled Talaat, Internal Medicine
Amany Ibrahim, Internal Medicine
Ahmad Refaat, Medical Statistics
Mohamad Sand, Pediatrics
Mohamad Abdel-Raoof, Physiology
Shreen Elaraby, Physiology
Heba Pasha, Biochemistry and Molecular Biology
Randa Hussini, Biochemistry and Molecular Biology
Rasha Hussini, Biochemistry and Molecular Biology

Cairo University, Egypt:
Ahmad El-Garem, Endemic and Tropical Medicine
Shukry Hunter, Endemic and Tropical Medicine
Suhir Zakaria, Endemic and Tropical Medicine
Hosny Salama, Endemic and Tropical Medicine

Ain Shams University, Egypt:
Abdel-Rahman El-Ziady, Endemic and Tropical Medicine
Fawzy Montasir, Endemic and Tropical Medicine
Ramadan Baddar, Internal Medicine
Amr Fateen, Internal Medicine
Mahmoud Osman, Internal Medicine
Reda El-Wakil, Endemic and Tropical Medicine

Mansura University, Egypt:
Gamal Sheha, Internal Medicine
Magdy Hamed, Internal Medicine

Tanta University, Egypt:
Saber Ismail, Endemic and Tropical Medicine
Abdel-Raoof Abu-Elazm, Endemic and Tropical Medicine
Mohamad Sharaf, Endemic and Tropical Medicine
Assiut University, Egypt:
Ahmad Nasr, Endemic and Tropical Medicine

Benha University, Egypt:
Samir Qabil, Endemic and Tropical Medicine
Magdy Atta, Endemic and Tropical Medicine

Military Medical Academy, Egypt:
Mamdouh Elbahnasawy, Endemic and Tropical Medicine

Secretary:
Eman Abdel-Aal
E-mail: ajied@zu.edu.eg
                     emanelshamy2005@yahoo.com
Walid Abdel-Dayem
E-mail: ajied@zu.edu.eg
drwalid_dayem@yahoo.com
Abeer Nafee
E-mail: ajied@zu.edu.eg
        abeer-n2009@hotmail.com
Tarik Zaher
E-mail: ajied@zu.edu.eg
tarezqahter@zu.edu.eg
Soha Esmat
E-mail: ajied@zu.edu.eg
     sohaesmat@hotmail.com
Ghada Salem
E-mail: ajied@zu.edu.eg
       ghadasalem21@yahoo.com
Sahar Elnimr
E-mail: ajied@zu.edu.eg
          alnimrsahar@yahoo.com
Hala Ismail
E-mail: ajied@zu.edu.eg
         h_ao_am@yahoo.com
Mohamad Magdy
E-mail: ajied@zu.edu.eg
            mradwan@zu.edu.eg
Mohamad Emara
E-mail: ajied@zu.edu.eg

E-Archiving:
Hosam Dawood
Sherwet Sahlol
Mohamad Ibrahim
Amal Abdel-Fattah
Ahmad Farok
Abeer Hasan
Ibrahim Mohamad
Sameh Mahmoud
Said Saad
Besheer Helmy
Emad Abdel-Hamid
Ahmad Elgebaly
Nabila Hasan
Kamal Amer
Ahmad Abdel-Razik
Ahmad Attia
Ahmad Saaid
Ahmad Lotfy
Shereif Bahnasawy
Abdel-Monim Elshamy
Ahmad Abulkhair
Dena Mohamad
Sara Refae
Shimaa Abdel-Fattah
Ramy Elhendawy
Mona Amin
Marwa Attia
Mahmoud Khalil
Marwa Ayesh
Mona Abdelmaksoud

Published by: Communication and Information Technology Center (CITC), Zagazig University, Zagazig, Egypt
Atef Eraky
E-mail: atef_ eraky@yahoo.com
Wafaa Metwally
E-mail: wafaa@zu.edu.eg

Scope of the Journal
The Afro-Egyptian Journal of Infectious and Endemic Diseases (AJIED) is a peer-reviewed journal that publishes clinical, parasitological, microbiological, physiological, biochemical, immunological and pathological studies in the field of infectious, endemic and tropical diseases. The scope of the journal includes also articles of endemic gastroenterology and hepatology. The journal is published quarterly by Endemic and Tropical Medicine Department, Faculty of Medicine, Zagazig University, Zagazig, 44519, Egypt.

Submission Process
The Journal accepts online submissions only. Manuscripts can be submitted at http://mis.zu.edu.eg/ajied/home.aspx. Once the manuscript has been uploaded, our system automatically generates an electronic pdf, which is then used for reviewing. All correspondence, including notification of the Editor’s decision and requests for revisions, will be managed through this system. Authors can follow the progress of their paper using this system to final decision. For any problems please contact the Editorial Office at ajied@zu.edu.eg.

Authorship
All authors should have made substantial contributions to all of the following:
(1) the conception and design of the study, or acquisition of data, or analysis and interpretation of data
(2) drafting the article or revising it critically for important intellectual content
(3) final approval of the version to be submitted.

**Article types**

The following types of manuscripts are routinely accepted:

1. **Original Articles:** This should include an abstract, keywords, introduction, patients/material and methods, results, discussion and references. They should be no longer than 5000 words (word count excludes tables, figures and legends).

2. **Reviews:** An abstract and keywords are required. The text should be divided into sections by suitable headings. Tables and figures may be used as appropriate for the text. They should be no longer than 6000 words.

3. **Opinions, Commentaries and Letters to the editor:** These take the same form as a review.

4. **Short Communications:** These should be no more than 2,500 words, with up to 15 references and a maximum of 3 figures or tables.

5. **Case Reports:** Case reports should present only cases of exceptional interest including presentation, diagnosis and management of disease. They should contain short summaries, an introduction, the case report, discussion, a reference list, tables and figure legends.

6. **Images in Infectious and Endemic Diseases:** These consist of interesting cases with high quality images with a short text and no more than 10 references.

**Preparation of the manuscript**

Please ensure that the following are including in your submission: -One author designated as corresponding author: Their E-mail address , full postal address Telephone and fax numbers -Keywords -Cover letter addressed to the Editor, introducing the manuscript and confirming that it is not being submitted concurrently elsewhere -All figure captions -All tables (including title, description, footnotes) -All necessary files have been uploaded -Manuscript has been spell checked -All text pages have been numbered -References are in the correct format for this journal -All references mentioned in the Reference list are cited in the text and vice versa -Permission has been obtained for use of copyrighted material from other sources (including the Web) -Color figures are clearly marked as being intended for color reproduction or to be reproduced in black-and-white.-Manuscripts :Please type all pages with double spacing and wide margins on one side of the paper. Title page, abstract, tables, legends to figures and reference list should each be provided on separate pages of the manuscript. Use font such as Times New Roman or Arial. The text should be in single-column format. Number the pages. Keep the layout of the text as simple as possible. Most formatting codes will be removed and replaced on processing the article. In particular, do not use the options to justify text or to hyphenate words.

However, do use bold face, italics, subscripts, superscripts etc. Do not embed 'graphically designed' equations or tables, but prepare these using the facility in Word or as a separate file in Excel. When preparing tables, if you are using a table grid, use only one grid for each individual table and not a grid for each row. Do not prepare tables in PowerPoint. To avoid unnecessary errors you are strongly advised to use the spellchecker. The title page should include: the title, the name(s) and affiliation(s) of the author(s), an address for correspondence, and telephone/fax numbers for editorial queries. All articles should include an Abstract of no more than 300 words and 3-6 key words for abstracting and indexing purposes. Please write your text in good English. Use decimal points (not commas); use a space for thousands (10 000 and above).

Provide the following data in your submission (in the order given).

1. **Title page (separate page):** Title should be concise and informative. Avoid abbreviations and formulae where possible. Author names and affiliations. Where the family name may be ambiguous (e.g., a double name), please indicate this clearly. Present the authors’ affiliation addresses (where the actual work was done) below the names. Indicate all affiliations with an Arabic number immediately after the author’s name and in front of the appropriate address. Corresponding author: This should be indicated after authors affiliations. Clearly indicate who is willing to handle correspondence at all stages of refereeing and publication, also post-publication. Ensure that telephone and fax numbers (with country and area code) are provided in addition to the e-mail address and the complete postal address.

2. **Abstract:** (separate paper). A concise and informative abstract is required (maximum length 300 words). The abstract should state briefly the purpose of the research, the principal results and major conclusions. Do not cite references in the abstract. Non-standard or uncommon abbreviations should be avoided in the abstract, but if essential they must be defined at their first mention in the abstract itself. The abstract should be divided into: Background and study aims, patients/material and methods, results and conclusion. Keywords Immediately after the abstract, provide a maximum of 6 keywords.

3. **Abbreviations:** Define abbreviations that are not standard in this field at their first occurrence in the article (even if mentioned in the abstract). Ensure consistency of abbreviations throughout the article

4. **Introduction:** State the objectives of the work and provide an adequate background, avoiding a detailed literature survey or a summary of the results.

5. **Patients/Materials and methods:** Provide
sufficient detail to allow the work to be reproduced. Methods already published should be indicated by a reference. Only relevant modifications should be described. Include in figure legends and table texts, technical details of methods used, while describing the methods themselves in the main text.

6- Results: This should explore the significance of the results of the work, not repeat them. A combined Results and Discussion section is often appropriate in a Short Communication but not in an Original Article. Ensure that the chapter results stands by itself and explain all results of your work. Note that all tables and figures should be presented in separate papers.

7- Discussion: Discuss your results and avoid extensive citations and discussion of published literature.

8- Acknowledgement: Collate acknowledgements in a separate section at the end of the article and do not, therefore, include them on the title page, as a footnote to the title or otherwise. When the work included in a paper has been supported by a grant from any source, this must be indicated. A connection of any author with companies producing any substances or apparatus used in the work should be declared in this section. All contributors who do not meet the criteria for authorship as defined above should be listed in an acknowledgements section. Examples of those who might be acknowledged include a person who provided purely technical help, writing assistance, or a department chair who provided only general support. Authors should disclose whether they had any writing assistance and identify the entity that paid for this assistance.

9- References: References should be numbered consecutively (with parentheses) as they appear in the text e.g. [5]. Type the reference list with double spacing on a separate sheet. This includes family name and first name initial, up to 6 authors are required and more authors are marked with et al. Examples: 1- Abdel-Wahab M, Esmat G, El-Boraey Y, Ramzy I, Medhat E, Strickland G. The epidemiology of schistosomiasis in Egypt: methods, training, and quality control of clinical ultrasound examinations. Am J Trop Med Hyg 2000; 62 (suppl) :17-20. 2- Wright W. Geographical distribution of schistosomes and their intermediate hosts. Ansari N, ed. Epidemiology and control of schistosomiasis (bilharziasis). Baltimore :University Park Press 1973 :42-48. Do not include references to personal communications, unpublished data or manuscripts either 'in preparation' or 'submitted for publication'. If essential, such material may be incorporated into the appropriate place in the text. Recheck references in the text against reference list after your manuscript has been revised. All references listed in the text should be included in the reference list and all references in the reference list should be included in the text.

10- Illustrations: Photographs should be presented as high quality jpg. Illustrations will not be redrawn by the Publisher: line figures should be suitable for direct reproduction. They should be prepared with black on white background, or be black-and-white images; ; they should be completely and consistently lettered, the size of the lettering being appropriate to that of the illustration, taking into account the necessary reduction in size. Colour figures will be included

11- Tables: Number tables consecutively in accordance with their appearance in the text. Place footnotes to tables below the table body and indicate them with superscript lowercase letters. Avoid vertical rules. Be sparing in the use of tables and ensure that the data presented in tables do not duplicate results described elsewhere in the article.

Editorial Review
All manuscripts are subject to peer review. If changes are requested, revisions received later than 2 months after this request will be treated as new submissions. When changes are made, the corresponding author should go into resubmission under title of submission of revised manuscript, and a word document should be uploaded that indicates changes and modifications done.

Publication charges
After submission of original articles, reviews, case reports and short communications the editor will request 100 Egyptian pounds for editing process to be sent to:
Dr Sahr Abdel-Shafy Elnimr, Endemic and Tropical Medicine Department, Faculty of Medicine, Zagazig University, Zagazig 44519, Egypt, Mobile:+201007246450 via: Bank Misr, Egypt, Zagazig Islamic Branch, Swift code: BMISEGCX140, account number: 261/250/202064. After acceptance of the article; 200 Egyptian pounds will be requested by the editor for publication to be sent as above. 10 colored prints will be sent to the corresponding author inside Egypt. Additional postal charges will be needed for outside Egypt. Please send a copy of your bank transfer to: ajied@zu.edu.eg.

Off prints
The corresponding author, at no cost, will be provided with a PDF file of the article via e-mail. Authors can download the PDF from the journal web page and in the same way the journal cover image can be downloaded.

Policy and Ethics Declarations
Upon submission you will be required to complete this
form to declare funding, conflict of interest and to indicate whether ethical approval was sought. This information must also be inserted into your manuscript under the acknowledgements section. If you have no declaration to make please insert the following statements into your manuscript: Funding: None
Competing interests: None declared Ethical approval: Not required Ethics. Work on human beings that is submitted to AJIED should comply with the principles laid down in the Declaration of Helsinki; Recommendations guiding physicians in biomedical research involving human subjects. Adopted by the 18th World Medical Assembly, Helsinki, Finland, June 1964, amended by the 29th World Medical Assembly, Tokyo, Japan, October 1975, the 35th World Medical Assembly, Venice, Italy, October 1983, and the 41st World Medical Assembly, Hong Kong, September 1989. The manuscript should contain a statement that the work has been approved by the appropriate ethical committees related to the institution(s) in which it was performed and that subjects gave informed consent to the work. Studies involving experiments with animals must state that their care was in accordance with institution guidelines.

Competing interests
All authors must disclose any financial and personal relationships with other people or organizations that could inappropriately influence (bias) their work. Examples of potential conflicts of interest include employment, consultancies, stock ownership, honoraria, paid expert testimony, patent applications/registrations, and grants or other funding. Role of the funding source all sources of funding should be declared. Authors should declare the role of study sponsors, if any, in the study design, in the collection, analysis and interpretation of data; in the writing of the manuscript; and in the decision to submit the manuscript for publication. If the study sponsors had no such involvement, the authors should so state.
Contents

ORIGINAL ARTICLES

Assessment of C-Reactive Protein and Macrophage Migration Inhibitory Factor in Diabetic Foot Infection
Mohamed Rashad Ahmed, Gehan Abd-el-Kader Ibrahim, Hoda Gouda Bakr, Takwa E. Meawed 19

Percutaneous Local Injection of Ethanol and Mitoxantrone in Treatment of Hepatocellular Carcinoma
Ahmed S Bihery, Mostafa H El-Shamy, Talaat E El-Mokadem, Tarek I Zaher, Walid A Abd El Dayem, Talaat Fathy, Mohamad Emara 28

Interferon Therapy for Elderly Egyptian Patients with Chronic Hepatitis C of Genotype 4
Mohammed Emam, Waleed A. Abd El-Dayem, Soha AElhawari, Hayam Heeba, Amany Emara 37

Probiotics in Minimal Hepatic Encephalopathy
Soha A Elhawari, Emad F Hamed 45

The Pattern, Risk Factors, and Clinico-Aetioloical Correlate of Tinea Capitis among the Children in a Tropical community Setting of Osogbo, South-Western Nigeria
Adeolu O Akinboro, Olayinka A Olasode, Olaniyi Onayemi 53

VIDEO CASE

Video Case: Extraction of a Coin from the Stomach of a 6 Months Infant
Tarek I Zaher 65

IMAGE CASE

Image Case: Esophageal Polyp in Old man, Is it Common?
Mohamed Hassan Emara, Salem Yousef 66

CASE RECORD

Case 1-2011: A 60 Years Old Male with Coma and Fever with Recent Travel to South Sudan
Tarik Zaher, Nahla Elgammal, Dina Mohamed 68

CONFERENCE

9th Annual Conference of Hepatology, Gastroenterology and Infectious Diseases, November 17, 2011, Zagazig Faculty of Medicine, Zagazig, Egypt 72
Assessment of C-Reactive Protein and Macrophage Migration Inhibitory Factor in Diabetic Foot Infection

Mohamed R. Ahmed, Gehan A. Ibrahim, Hoda G. Bakr, Takwa E. Meawed

1Internal Medicine Department, Faculty of Medicine, Zagazig University, Egypt.
2Microbiology & Immunology Department, Faculty of Medicine, Zagazig University, Egypt.

Background and study aim: Diabetic foot ulcer is a universal health problem. Neuroischemic changes and infection are responsible for its occurrence and complications. Altered or complete loss of sensation and microvascular disease complicated by unchecked infection can precipitate tissue necrosis and gangrene. A threat for a rapid test predicting early infected foot ulcer emerges. C-reactive protein (CRP) and macrophage migration inhibitory factor (MIF) are involved in innate inflammatory response. We aimed at evaluation of the ability of C-reactive protein and macrophage migration inhibitory factor to differentiate between early infected and non infected diabetic foot ulcers and to detect risk factors of diabetic foot ulceration.

Patients and methods: 52 diabetic patients were selected, examined and classified into 3 groups: Group (I): Included 12 patients with non-infected diabetic foot ulcer (grade I), group (II): Included 30 patients with mildly infected diabetic foot ulcer (grade II) and group (III): Included 11 diabetic patients free from foot wounds used as a control group. In addition to routine laboratory investigations, serum CRP was measured using Enhanced Immuno-turbidimetric Assay. MIF level was detected by ELISA. Swabs from the diabetic foot ulcers were taken for aerobic and anaerobic cultures.

Results: Statistically significantly elevated Hb A1C%, MIF and CRP levels were detected in mild infected diabetic foot ulcer compared to studied groups (P <0.05). Dermatological changes were statistically significant risk factors for diabetic foot ulcers, accounted for 88.1% of ulcer cases. The most frequently isolated organism was E. coli. The most common site for ulcers was the toes representing 50% of the cases.

Conclusion: CRP and MIF can differentiate early infected from non-infected diabetic foot ulcers.

INTRODUCTION

Foot ulcers are common diabetic complications. About 15% of diabetics develop foot ulcers within their life time and up to 70% of all non-traumatic amputations in the world occur in diabetics. Many of these amputations are preventable as about 85% are preceded by a foot ulcer [1]. Although most foot infections stay superficial, they can spread to muscle, joints and bone. Unchecked infection can precipitate tissue necrosis and gangrene, especially in an ischemic limb [2].

Diagnosis of infection must be based not on microbiological findings only but also on clinical criteria to avoid unnecessary antimicrobial treatment and emergence of multidrug-resistant organisms [3]. Aerobic gram-positive cocci are the predominant pathogens, Staph aureus and the β-hemolytic Streptococci are the most commonly isolated pathogens [4].

Biochemical parameters such as sedimentation rate and leucocytosis are reputed to be of poor value for diagnosing diabetic foot infections [5]. C-reactive protein (CRP) is a highly conserved acute phase protein of innate inflammatory response synthesized by hepatocytes under cytokines stimulation originating at the site of pathology and leading to dramatic rise in CRP level within 48 h after stimulation [6,7]. An altered immune response in diabetic foot ulcer is a universal health problem. Neuroischemic changes and infection are responsible for its occurrence and complications. Altered or complete loss of sensation and microvascular disease complicated by unchecked infection can precipitate tissue necrosis and gangrene. A threat for a rapid test predicting early infected foot ulcer emerges. C-reactive protein (CRP) and macrophage migration inhibitory factor (MIF) are involved in innate inflammatory response. We aimed at evaluation of the ability of C-reactive protein and macrophage migration inhibitory factor to differentiate between early infected and non infected diabetic foot ulcers and to detect risk factors of diabetic foot ulceration.

Patients and methods: 52 diabetic patients were selected, examined and classified into 3 groups: Group (I): Included 12 patients with non-infected diabetic foot ulcer (grade I), group (II): Included 30 patients with mildly infected diabetic foot ulcer (grade II) and group (III): Included 11 diabetic patients free from foot wounds used as a control group. In addition to routine laboratory investigations, serum CRP was measured using Enhanced Immuno-turbidimetric Assay. MIF level was detected by ELISA. Swabs from the diabetic foot ulcers were taken for aerobic and anaerobic cultures.

Results: Statistically significantly elevated Hb A1C%, MIF and CRP levels were detected in mild infected diabetic foot ulcer compared to studied groups (P <0.05). Dermatological changes were statistically significant risk factors for diabetic foot ulcers, accounted for 88.1% of ulcer cases. The most frequently isolated organism was E. coli. The most common site for ulcers was the toes representing 50% of the cases.

Conclusion: CRP and MIF can differentiate early infected from non-infected diabetic foot ulcers.
ulcer patients is interesting as some markers of inflammation are upregulated (CRP, fibrinogen, IL-6, MIF, macrophage inhibitory protein (MIP-1α) and IP-10) while others are not (IL-8, IL-18, and macrophage chemo-attractant protein (MCP-1)) [8].

Macrophage migration inhibitory factor (MIF) is a proinflammatory cytokine of the innate immune system that plays a major role in the induction of immune-inflammatory reaction. MIF may play a causal role in the etiology of type 2 diabetes and elevated MIF levels confer a higher disease risk [9].

AIM OF THE WORK

Evaluation of the ability of CRP and MIF to differentiate between early infected and non-infected diabetic foot ulcers and to detect risk factors of diabetic foot ulceration.

PATIENTS AND METHODS

A prospective case control study was conducted at diabetic foot clinic of Internal Medicine and Microbiology and Immunology Departments, Faculty of Medicine-Zagazig University Hospitals during the period from Feb 2009 to Jan 2010.

Fifty-two diabetic patients were included, 42 of them were suffering from diabetic foot ulcer and 10 were control group. They were treated by oral antidiabetic drugs, insulin or combined treatment. Inclusion criteria: Cases to be included should have history of diabetes mellitus, fasting plasma glucose ≥126 mg/dl (7.0 mmol/l) or 2-h post prandial plasma glucose ≥200 mg/dl (11.1 mmol/l). They were classified into 3 groups according to the grade of diabetic foot ulcer (IDSA-IWGDF) (3) into :

**Group (I):** Included 12 patients with non-infected diabetic foot ulcer as there were no symptoms or signs of infection (Grade I), 7 patients were males, 5 were females with age range 35-64 years and mean of age 50.16 ± 8.5 years. Their mean duration of diabetes was 7.16 ± 3.51 years, 75% of them were using insulin and 25% were using oral anti-diabetics.

**Group (II):** Included 30 patients with mildly infected diabetic foot ulcer (Grade II), 15 patients were males, others were females with age range 26-76 years and mean 52.3 ± 11.4 years. Their mean duration of diabetes was 14.6±7.18 years, 70% of them were using insulin, 16.7% were using oral anti-diabetics and 13.3% on combined therapy. Grade II diabetic foot ulcer was diagnosed if there was infection involving skin and subcutaneous tissue only (without involvement of deeper tissues and without systemic signs). At least two of the following signs were fulfilled:
- Local swelling or induration
- Erythema > 0.5 - 2 cm around the ulcer
- Local tenderness or pain
- Local warmth
- Purulent discharge (thick, opaque to white secretion).

**Group (III):** Included 10 diabetic patients free from foot wounds used as a control group. 5 patients were males and 5 were females, their age range (35-75) years and mean age was 52.7±11.1 years, 80% of them using insulin, 10% using oral anti-diabetic agents and 10% were on combined therapy.

**Exclusion criteria :** Any patient who had any of the following was excluded: foot wound that was more than grade 2, other causes of inflammatory response of the skin (e.g. trauma, gout, acute Charcot neuro-osteoarthropathy, fracture, thrombosis and venous stasis), treatment with antibiotics and lastly peripheral arterial disease.

Any systemic illness that might elevate the inflammatory markers as: allergic complication of infection (rheumatic fever, and erythema nodosum).

Other inflammatory diseases as (rheumatoid arthritis, chronic arthritis, systemic vasculitis, Familial Mediterranean Fever and Chron's disease), necrosis, trauma and malignancy.

Informed consent was taken. Patients included in this study were subjected to full history taking, complete clinical examination and foot examination were performed to detect any of the following:
- Signs of neuropathy that included 10 gm monofilament detection and deep tendon reflex.
- Signs of vasculopathy included pulse examination for dorsalis pedis and posterior tibial arteries, presence of edema and ankle brachial index.

- Risk factors that promote ulcer development as presence of toes deformity, bunions, Charcot foot, drop foot, equinus, prominent metatarsal heads and amputation) or dermatological factors as (corns, callus, dry skin, hair loss, nail changes and changed color of skin).

www.mis.zu.edu.eg/ajied/home.aspx
**Methods:**
Consent was taken from all of patients included in the study. After fasting for at least 12 h, 10 ml of blood was aseptically collected. Complete blood count was done on automated cell counter (cell Dyn 1700). Liver and kidney functions were done on automated analyzer (cobas 6000). Hemoglobin A1C was done on automated analyzer (cobas 6000). Swabs from the diabetic foot ulcers were taken for aerobic and anaerobic cultures. Tissue specimens were obtained from the debrided base and sides (Levine technique) [10].

Serum CRP was done on automated analyzer (Integra 400), CRP latex is an in vitro test for quantitative determination of CRP in human serum and plasma on Roche/Hitachi Cobas systems using particles Enhanced Immunotubidimetric Assay [11].

MIF level detection was done by using RayBio® Human MIF ELISA.

**Statistical analysis:**
Data entry and analysis were performed using SPSS (statistical package for social science version 10) (SPSS, Inc., Chicago, IL, USA). Data were presented as number and percentage, mean and standard deviation. The chi-square ($\chi^2$), t-test and ANOVA were used. Mann Whitney-U test and Kruskal- Wallis H test are non parametric tests equivalent of the t-test and ANOVA. P value of <0.05 was considered significant.

**RESULTS**
Non-significant difference was found between cases and control group regarding age, sex, type of diabetes or treatment modalities of diabetes. Ulcer size and depth was not statistically different in mildly infected than non infected ulcers. But in mildly infected ulcer patients, there was significantly longer duration of diabetes 14.6±7.18 years compared to 7.16±3.51, 10.7±6.7, in non infected ulcer and control groups respectively (P<0.05) also hypertension was significantly found in group II (33.3%), compared to 8.3 % and 0.0 %, in (group I) and control group respectively) (P <0.05) Table (1).

Decreased Hb % and increased WBC were found in group II than group I but it was statistically insignificant (p>0.05). A significantly elevated HB A1C% and highly statistically significant elevated CRP level were detected in (group II) compared to the studied groups (p<0.05) and (p<0.001) respectively. Also highly significantly elevated MIF levels were detected in (group II) compared to the studied groups (p<0.001) Table (2). There was no statistical significant difference between group (III) and group (I) as regarding HB%, WBC count, A1C%, ALT, AST, serum creatinine and CRP level Table (2).

A statistical significant difference was found between ulcer cases and non ulcer (control group) as regards dermatological changes (corns, callus, dry skin, hair loss, nail changes and color of skin), 88.1% of diabetic ulcer cases had dermatological changes compared to 50 % of non ulcer cases (P <0.05). While, there was no statistical significant difference between cases and control group regarding neuropathy, 76.2% of the diabetic ulcer cases compared to 70 % of non ulcer ones (P <0.05). Also, no significant difference was found regarding deformity, 28.6% cases compared to 10% of non ulcer cases (P <0.05), Table (3).

The most frequently isolated organism from infected diabetic foot ulcer was *E-coli* 12 (43.3%) then *Staph aureus* 9 (30.0%) followed by *Proteus mirabilis* 3 (10.0%) then *Klebsiella* 2 (6.7%), *Candida* 1 (3.3%) and lastly sterile culture was found in 2 (6.7%) Table (4).

The most common site for diabetic foot ulcer was the toes representing 50% of cases then metatarsals heads, mid foot and heel (45.2%), then dorsum of foot (4.8%) (Figure 1).
Table (1): Comparison between the studied groups as regards demographic data and clinical criteria

<table>
<thead>
<tr>
<th></th>
<th>Group (І) Non infected ulcer (N = 12)</th>
<th>Group (ІІ) Mild infected ulcer (N = 30)</th>
<th>Group (ІІІ)</th>
<th>Test statistics</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age(years)</td>
<td>Mean ± SD 50.1± 8.5</td>
<td>52.3 ± 11.4</td>
<td>52.7± 11.1</td>
<td>F= 0.21</td>
<td>0.81 (NS)</td>
</tr>
<tr>
<td>Sex : no (%)</td>
<td>Male 7 (58.3%) Female 5(41.7%)</td>
<td>15(50%) Male 15(50%) Female 5 (50%)</td>
<td>χ2= 0.25</td>
<td>0.88 (NS)</td>
<td></td>
</tr>
<tr>
<td>Ulcer (mean±SD)</td>
<td>Size(cm): 1.7±0.7</td>
<td>1.7±1.1</td>
<td>-----</td>
<td>Z=0.22*</td>
<td>0.83 (NS)</td>
</tr>
<tr>
<td></td>
<td>Depth(cm): 0.8±0.4</td>
<td>0.7±0.5</td>
<td>Z=0.44*</td>
<td>0.69 (NS)</td>
<td></td>
</tr>
<tr>
<td>Diabetes (Type І)</td>
<td>1(8.3%)</td>
<td>7(23.3%)</td>
<td>2(20%)</td>
<td>χ2  1.246</td>
<td>0.536 (NS)</td>
</tr>
<tr>
<td></td>
<td>Diabetes (Type ІІ)</td>
<td>11(91.7%)</td>
<td>23(76.7%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Duration of DM mean ± SD (years)</td>
<td>7.16± 3.51</td>
<td>14.6±7.18</td>
<td>F 5.99</td>
<td>0.048 (S)</td>
</tr>
<tr>
<td>Treatment of DM</td>
<td>Insulin 9 (75%) Oral 3(25%) Oral combined 0(0.0%)</td>
<td>21(70%) Oral 5(16.7%) Oral 1(10%)</td>
<td>χ2  2.42</td>
<td>0.658 (NS)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ischemic heart diseases</td>
<td>0(0.0%)</td>
<td>4(13.3%)</td>
<td>χ2  3.17</td>
<td>0.2 (NS)</td>
</tr>
<tr>
<td></td>
<td>Hypertension</td>
<td>1(8.3%)</td>
<td>10(33.3%)</td>
<td>χ2  6.53</td>
<td>0.038 (S)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>(*)</td>
<td></td>
</tr>
</tbody>
</table>

(*) = Kruskal – Wallis test

Table (2): Serum levels of CRP, MIF and other laboratory criteria of the studied groups.

<table>
<thead>
<tr>
<th>Lab Data</th>
<th>Studied groups</th>
<th>Group (І) Non infected ulcer (N = 12)</th>
<th>Group (ІІ) Mild infected ulcer (N = 30)</th>
<th>Group (ІІІ) (N = 10)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb %</td>
<td></td>
<td>12.2±1.7</td>
<td>11.6±1.8</td>
<td>11.8±1.8</td>
<td>0.534 (NS)</td>
</tr>
<tr>
<td>WBCx10³</td>
<td></td>
<td>7.2±1.9</td>
<td>9.1±3.1</td>
<td>7.9±2.4</td>
<td>0.133 (NS)</td>
</tr>
<tr>
<td>HbA1C%</td>
<td></td>
<td>7.8 ± 1.1</td>
<td>8.9±1.8 ab</td>
<td>7.7±2.1</td>
<td>0.05 (S)</td>
</tr>
<tr>
<td>CRP mg/dl</td>
<td></td>
<td>13.4±27.1</td>
<td>17.4±7.5 ab</td>
<td>4.6±3.3</td>
<td>0.0001 (HS)</td>
</tr>
<tr>
<td>MIF ng/dl</td>
<td></td>
<td>7.7±1.3</td>
<td>11.9±2.2 ab</td>
<td>4.5±0.9</td>
<td>0.0001 (HS)</td>
</tr>
<tr>
<td>ALT U/L</td>
<td></td>
<td>24.6±11.3</td>
<td>27.1±18.9</td>
<td>25.5±12.33</td>
<td>0.997 (NS)</td>
</tr>
<tr>
<td>AST U/L</td>
<td></td>
<td>26.2±15.2</td>
<td>26.4±19.1</td>
<td>24.9±10.6</td>
<td>0.778 (NS)</td>
</tr>
<tr>
<td>Creatinine mg/dl</td>
<td></td>
<td>0.8±0.2</td>
<td>1.2±1.6</td>
<td>0.9±0.2</td>
<td>0.928 (NS)</td>
</tr>
</tbody>
</table>

a= Significant difference between group (ІІІ) and group (І).
b= Significant difference between group (ІІ) and group (ІІІ).
   No-significant statistical difference between group (ІІІ) and the group (І).

Table (3): Culture results of the diabetic foot ulcers.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Group (І) Non infected ulcer (N = 12)</th>
<th>(Group ІІ) Mild infected ulcer (N = 30)</th>
<th>χ2</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>E-coli</td>
<td>1(8.3%)</td>
<td>12(43.3%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staph aureus</td>
<td>0 (0.0%)</td>
<td>9(30.0%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proteus mirabilis</td>
<td>1(8.3%)</td>
<td>3(10.0%)</td>
<td>25.608</td>
<td>0.0001 (HS)</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>0(0.0%)</td>
<td>2(6.7%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Candida</td>
<td>0(0.0%)</td>
<td>1(3.3%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sterile Culture</td>
<td>10(83.3%)</td>
<td>2(6.7%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
www.mis.zu.edu.eg/ajied/home.aspx

Table (4): Risk factors of ulceration in diabetic foot patients.

<table>
<thead>
<tr>
<th>Risk factor</th>
<th>Ulcer cases (N = 42) (%)</th>
<th>Control group (N = 10) No (%)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neuropathy</td>
<td>32 (76.2%)</td>
<td>7 (70.0%)</td>
<td>0.697 (NS)</td>
</tr>
<tr>
<td>Deformity</td>
<td>12 (28.6%)</td>
<td>1 (10.0%)</td>
<td>0.419 (NS)</td>
</tr>
<tr>
<td>Dermatological changes</td>
<td>37 (88.1%)</td>
<td>5 (50.0%)</td>
<td>0.022 (S)</td>
</tr>
</tbody>
</table>

Table (5): MIF ELISA sensitivity, specificity and positive predictive value

<table>
<thead>
<tr>
<th></th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Positive Predictive Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>MIF ELISA Test</td>
<td>93.3%</td>
<td>83.3%</td>
<td>93.3%</td>
</tr>
</tbody>
</table>

DISCUSSION

There are 285 million people suffering from DM, corresponding to 6.4% of the world’s adult population, which is estimated to rise to 438 million by 2030. Estimated Egyptian diabetics among adults aged 20-79 years is 11.4% of diabetic world population for the year 2010 and to the national population is 10.4 % [13].

C-reactive protein (CRP) takes part in the systemic inflammatory response, acute injury, infection or other stimuli, CRP binds to specific molecular configurations typically present in case of cell death and additionally found on the surface of pathogens, and therefore CRP increases rapidly after tissue injury or infections and reflects the intensity of the inflammatory process [6,14].

Serum concentration of CRP represents a very useful non specific inflammatory biomarker and plays an important role in screening for organic diseases, monitoring the response to treatment and helps in detection of recurrent infection [10].

MIF is a proinflammatory cytokine of innate immune system, High levels of MIF were detected in chronic non healing diabetic ulcers then began to fall with successful healing [9].

In this study, there was no significant difference between studied groups regarding age and gender, the same as our results was reported by Kumar et al. [15], however, Frykberg [16] found that male sex was a risk factor for ulceration.

Data from the National Hospital Discharge Survey (NHDS) 1987–1990 in the US reported the highest percentage of hospital discharges for foot ulcers in patients aged 45–64 years and lowest in patients < 45 years. Elderly patients are
usually less mobile, have poor vision, live alone and have other medical problems [17].

Longer duration of diabetes and elevated percentage of hypertension (33.3%), were detected in mild infected ulcer patients than others. Reiber [17] showed a six-fold increased risk of foot ulcer in patients ≥20 years DM duration than patients ≤9 years. In another study, diabetic patients with foot ulcer had a longer duration of DM, 17 years versus 12 years [16].

In this study, HbA1c % was significantly increased in mild infected than non infected diabetic foot ulcers. The level of glycaemic control has been shown to play a role, with a 26% increased risk of peripheral vascular disease for every 1% increase in HbA1c [18]. Moss et al [19] found that increasing HbA1c % was associated with subsequent foot ulcer in their cohort study. Boyko et al. [20] found that, severe hyperglycaemia was associated with a higher risk for diabetic foot ulcer. There was a great evidence that leukocyte functions such as migration, phagocytosis, intracellular killing and chemotaxis were impaired in the presence of uncontrolled diabetes [21].

In the present study, there was a high significantly elevated CRP level in mild infected diabetic foot ulcer compared to non infected ulcer and control groups. Lee et al. [22] sharing us the same result and reported that CRP was more useful method in predicting and diagnosing infection than WBC, ESR in diabetic foot ulcer patients. Upchurch et al. [23] supported our results, they reported increased CRP and fibrinogen levels in diabetic patients with a foot ulcer compared with diabetic patients without foot ulcer.

Lin et al. [24] added that, reduced CRP levels (<50 mg/L), indicates a low infection severity and may serve as a major predictor of successful percutaneous transluminal angioplasty outcome in diabetic patients with infected foot ulcers.

Jeandrot et al.[25] studied value of CRP, procalcitonin and other usual biological inflammatory markers in differentiating early infected from uninfected diabetic foot ulcers. They found that CRP as a single marker had the highest sensitivity and specificity and the use of a high-sensitivity CRP assay brought no additional accuracy of diagnosis.

CRP values have been shown to significantly increase in response to local infection, while procalcitonin increases more in systemic infection [26].

Regarding MIF level, a high significantly elevated MIF level was detected in our mild infected diabetic foot ulcers compared to non-infected ulcer and control groups. Both sensitivity of MIF ELISA test and precision (positive predictive value) were relatively high (93.3%) indicating that MIF ELISA test can be used as a screening test for mass population to differentiate between early-infected and non-infected ulcer. It doesn't need high technical skills, but further tests should confirm diagnosis. Its specificity was (83.3%) indicating that MIF ELISA test is not highly able to identify negative results (Table 4).

Only few researchers studied MIF level changes in diabetic foot ulcer patients. However, our results regarding elevated levels of CRP and MIF agreed with the results reported by Weigelt and his colleagues [8] that Patients with an acute foot ulcer had significantly higher levels of CRP, fibrinogen, interleukin (IL-6), MIF, macrophage inflammatory protein-1α, and interferon-γ-inducible protein-10.

Other authors discussed MIF level in diabetic patients with complications including ulcers, reported that elevated levels of MIF or its cell surface receptor (CD74) were found in patients with diabetic complications including diabetic nephropathy [27], diabetic retinopathy [28], and diabetic foot syndrome.

In two different cohort studies done by Herder et al [9,27], there was a stronger association between MIF with impaired glucose tolerance (IGT) and type 2 diabetes much more than the associations of CRP and IL-6 with IGT and type 2 diabetes.

A consistent triangular relationship between MIF genotypes, serum levels and incident type 2 diabetes was found especially in women indicating that MIF may play a causal role in the etiology of type 2 diabetes and elevated MIF levels confer a higher disease risk [29].

Regarding risk factors for the development of diabetic foot ulcer, the statistically-significant factor (P<0.05) was dermatological changes (88.1%) versus (50%). Deformity and neuropathy were risk factors, but the difference was insignificant.

Chronic, repeated pressure and recurrent trauma from biomechanical changes can lead to
hyperkeratosis. Callus tissue is tough and increasing pressure leading to increasing the incidence of plantar ulcerations [30,31]. The same finding was reported by El-Nahas et al. [32] in their large-scale study that included 1200 Egyptian diabetic patients.

One of the commonest combinations causing ulceration is peripheral neuropathy, foot deformity and inappropriate footwear. Patients with deformed feet bones are at risk of skin damage and infection [33,34,35].

Kumar et al. [15] found that over 40% of type 2 DM patients had significant neuropathy. Bowering [36] reported that 60% of diabetic foot ulcers are the result of underlying neuropathy. Loss of sensation is one of the strongest risk factors for ulceration [37]. This was supported by Abbot et al. [38] in a cohort study included 6613 diabetic patients found that, neuropathy predictors as abnormal ankle reflex and 10 gm monofilament insensitivity predict new foot ulcers.

In this study E. coli was the most frequently isolated organism from ulcer cases (43.3%) followed by S. aureus (30.0%), then Proteus (10.0%), however, the picture was different in the study done by Gadepallli et al. [39] as S. aureus represented (13.7%). Proteus was isolated from (12.6%), followed by E. coli (12.0%) from of diabetic foot ulcer isolates. Richard et al. [40] reported S. aureus was the most frequent pathogen (36.5% of all isolates).

In this study, the most common site for the ulceration was the toes, representing 50% of the cases. The study done by Reiber et al. [41] supported our results, they reported that, lesion sites were in toes in 52%, while, metatarsal heads, mid foot & heel in 37%, dorsum of foot in 11%. Gefen [42] found nearby results as up about 60% of all diabetic ulcers, typically involve sites exposed to high pressure such as near the metatarsal heads and toes.

We can conclude that, CRP and MIF can differentiate early infected from non infected diabetic foot ulcers to avoid unnecessary antimicrobial treatment leading to emergence of multidrug-resistant pathogens. The statistically significant risk factor for development of diabetic foot ulcers was dermatological changes. However, Deformity and neuropathy were other non-significant risk factors. Toes were the most common site exposed to diabetic foot ulceration.

**Recommendations**

The role played by other immune-mediators should be investigated in diabetic patients with ulcers, to determine the outcome of this problem and try to minimize the adverse effects by modification of these mediators.

**Funding:** Non.

**Conflicts of interest:** Non.

**Ethical approval:** Approved by the Ethical Committee of Faculty of Medicine, Zagazig University. Informed consents were taken from all patients.

**REFERENCES**

11. Levine NS, Lindberg RB, Mason AD Jr, Pruitt BA Jr. The quantitative swab culture and smear: a quick, simple method for determining the number
of viable aerobic bacteria on open wounds. *J Trauma* 1976; 16(2):89-94


24. Lin CW, Hsu LA, Chen CC, Yeh JT, Sun JH, Lin CH et al., C-reactive protein as an outcome predictor for percutaneous transluminal angioplasty in diabetic patients with peripheral arterial disease and infected foot ulcers. *Diabetes Research and Clinical Practice* 2010; (2) 167-172


www.mis.zu.edu.eg/ajied/home.aspx


Percutaneous Local Injection of Ethanol and Mitoxantrone in Treatment of Hepatocellular Carcinoma

Ahmed S Bihery¹, Mostafa H El-Shamy¹, Talaat E El-Mokadem², Tarik I Zaher¹, Walid A Abd El Dayem¹, Talaat Fathy¹, Mohamad Emara¹
¹Tropical Medicine Department, Faculty of Medicine, Zagazig University, Egypt.
²Clinical Oncology Department, Faculty of Medicine, Zagazig University, Egypt.

Background and study aim: New therapeutic choices have been developed for hepatocellular carcinoma (HCC), including percutaneous ablation therapy, transarterial chemoembolization, radiation therapy and molecular target therapy. Ablation of liver tumors is currently the main alternative to formal liver resection. This work aimed at comparing percutaneous ethanol injection (PEI) with combined percutaneous ethanol and mitoxantrone injection (PIM) in treatment of HCC.

Patients and methods: This study included 125 patients with 131 HCC lesions which were randomly divided into two groups; group I composed of 68 lesions in 65 patients treated with PEI. Group II composed of 63 lesions in 60 patients treated with PEI and PIM. Clinical assessment, laboratory evaluation and CT studies were performed to all patients pre treatment and at 3, 6, and 12 months post treatment. Each focal lesion was considered as one subject.

Results: The percentage of ablation in both groups at 3, 6, 12 months were 60.3%, 48.5% and 39.7% in group I respectively versus 85.5%, 74.6% and 68% in group II respectively with a statistical significant difference between the two groups. There is an increased number of local recurrence in group I compared to group II. Side effects and complications are comparable in both groups.

Conclusion: Combination of PEI and PIM is better than PEI alone without additional complication and recurrence rate seemed to be better in combination therapy than PEI alone.

INTRODUCTION

Hepatocellular carcinoma (HCC) is a highly malignant cancer and it is the sixth most common cancer worldwide and the third most common cause of cancer related deaths with higher prevalence in Asia and sub-Saharan Africa [1].

Advancement in diagnostic radiology and nuclear medicine contributed to the accurate and early diagnosis of HCC. Ultrasound, CT, Triphasic CT and MRI are used in diagnosis of these tumors [2].

Surgical resection, liver transplantation and cryosurgery are considered the best curative options for HCC. Regional interventional therapies have led to a major breakthrough in the management of unresectable HCC[3].

Furthermore, experiences in interventional radiology, radiation oncology and surgery fields have grown, and new therapeutic choices have been developed including percutaneous ablation therapy, transarterial chemoembolization (TACE), radiation therapy and molecular target therapy[4]. Ablation of liver tumors is currently the main alternative to formal liver resection [5]. Percutaneous ethanol injection (PEI) is a procedure of easy execution, good tolerability and low cost, which can be applied in repeated sessions [2].
Mitoxantrone is a cycle specific anthracyclin which induces persistent intracellular DNA damage. It is used as an anticancer agent and has demonstrated clinical activity when administered via multiple routes: intravenous, intraperitoneal, intrapleural, intrapericardial, or intrathecal [6]. Mitoxantrone was selected for palliative local treatment of malignant liver lesions because of its low tissue toxicity, high intratumoral concentration after intratumoral instillation, since it has a tendency to remain at the application site [7].

This work aimed at comparing PEI with combined PEI and intralional mitoxantrone (PIM) in treatment of HCC.

PATIENTS AND METHODS

This prospective interventional study was conducted in Tropical medicine and Clinical Oncology Departments, Faculty of Medicine, Zagazig university, Egypt, during the period from February 2009 to May 2011 and included 125 patients presented with 131 focal hepatocellular carcinoma lesions, the lesions were randomly divided into 2 groups;

Group I: Comprised 68 focal lesions presented in 65 patients, which were injected intralesionally with ethanol in multiple sessions.

Group II: Comprised 63 focal lesions presented in 60 patients, which were injected intralesionally with ethanol in multiple sessions followed by intralional injection with Mitoxantrone.

Each focal lesion was considered as one subject.

The diagnosis of HCC was based on typical characters of focal lesion in triphasic CT: filling of the dye in arterial phase and rapid fade out in venous and delayed phases, CT and/or ultrasound focal lesions with a serum alpha-fetoprotein >200 IU/ml or by histological confirmation.

Inclusion criteria in both groups are:

1- Single lesion 2-5 cm or 2 lesions each<3 cm [8],
2- Child- Pugh class A and B,
3- Serum creatinine < 2mg/dl,
4- Performance status 0-2 [9] and

Pretreatment assessment

Pre-treatment assessment of all patients was done by full history taking, thorough clinical examination, laboratory investigations including CBC, liver function, kidney function, α fetoprotein, serological examination for HCV and HBV. Radiological examination including X ray chest, CT study, ultrasound and ultrasound guided biopsy when indicated.

Ethanol injection.

All lesions were injected by absolute alcohol; ultrasound guided in multiple sessions, twice weekly, under complete aseptic condition and 10 mg midazolam as a sedative agent.

The same operator used spinal needle (20 gauge) to inject ethanol intralesionally and leave the needle for 2 minutes in place, then injection of local anesthetic during withdrawal of the needle to minimize the irritant effect of refluxed ethanol to the capsule.

The total amount of ethanol can be calculated according to the following equation:

\[ V = \frac{4}{3} \pi (r+0.5)^3 \]

Where: \( V \) = Volume of ethanol, \( \pi = \frac{22}{7} \), \( r \) = radius of the tumor by cm plus 0.5 cm as safety margin [10].

The average amount per session was 6.8 cc, with average 5 sessions per lesion and average amount of 35 cc per lesion which was calculated according to the above mentioned equation used by Shiina et al [10].

Mitoxantrone injection.

This was done to patients of group II after complete sessions of ethanol.

Ultrasound guided injection of mitoxantrone mixed with lipiodol at the time of injection in a single session, the dose of mitoxantrone is 0.5 mg per cubic centimeter of the tumor size.

Re-evaluation of the patients was done by laboratory investigations, ultrasound and triphasic CT after treatment and every 3 months up to one year.

Statistical analysis

Data were checked, entered and analyzed using SPSS 15 for Windows. Data were expressed as mean ± SD for quantitative variable, number and percentage for qualitative one. Chi-squared (X²) or fisher exact, t test and paired t test were used.
when appropriate. P <0.05 was considered significant.

RESULTS
Clinical presentations of the studied patients is shown in table 1. Group I included 48 male patients and 17 females with a mean age of 61.1 years. Group II included 44 male patients and 16 females with a mean age of 60 years. There was no statistically significant difference as regard age and sex between the studied groups.

Chronic HCV infection was the predominant virus in our study, 112 patients were HCV antibodies positive, where 11 patients were HBsAg positive and two patients had co-infection of both viruses. Local ablation therapy for HCC is associated with a variety of complications as shown in table (2). The most frequent complication was tolerable pain while intolerable pain (needs analgesics) detected in 27.6% and 20% of patients in group I and group II respectively. The most serious complications were less frequent and tend to occur in subjects in group I who developed peritoneal collection, subcapsular hematoma and pleural effusion each one detected in 2 subjects (3.1%) , while portal vein thrombosis detected in 3 subjects(4.6%). All these complications were controlled by conservative management .Table (3) compares biochemical parameters among patients of groups I before and 3 months after injection, there was no statistically significant difference as regard all parameters except for serum alpha-feto protein(α FP) and AST which showed statistically significant improvement after treatment. Table (4) compares biochemical parameters among patients of groups II before and 3 months after injection, there was no statistically significant difference as regard αFP, AST, serum bilirubin(BIL), serum albumin (ALB) and serum creatinin (CRT), where ALT and prothrombin time (PT) show statistically significant improvement in these patients after injection. Table 5 and 6 compare the success of ablation and rate of recurrence at 3,6 and 12 months with good ablation recorded in group II than in group I.

Table (1): Clinical presentations of all patients

<table>
<thead>
<tr>
<th>Condition</th>
<th>Group I (n=65 patients)</th>
<th>%</th>
<th>Group II (n=60 patients)</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Right hypochondral pain</td>
<td>35</td>
<td>53.8%</td>
<td>34</td>
<td>56.7%</td>
</tr>
<tr>
<td>Anorexia</td>
<td>22</td>
<td>33.8%</td>
<td>15</td>
<td>25%</td>
</tr>
<tr>
<td>Loss of weight</td>
<td>14</td>
<td>21.5%</td>
<td>16</td>
<td>26.7%</td>
</tr>
<tr>
<td>Low grade fever</td>
<td>11</td>
<td>16.9%</td>
<td>9</td>
<td>15%</td>
</tr>
<tr>
<td>Splenomegally</td>
<td>38</td>
<td>58.5%</td>
<td>35</td>
<td>58.3%</td>
</tr>
<tr>
<td>Lower limb edema</td>
<td>23</td>
<td>35.4%</td>
<td>19</td>
<td>31.7%</td>
</tr>
<tr>
<td>History of jaundice</td>
<td>6</td>
<td>9.2%</td>
<td>5</td>
<td>8.3%</td>
</tr>
<tr>
<td>History of ascites</td>
<td>4</td>
<td>6.2%</td>
<td>5</td>
<td>8.3%</td>
</tr>
</tbody>
</table>

Fig. (1): Viral serology among the studied groups

www.mis.zu.edu.eg/ajied/home.aspx
Table (2): Frequency of complications after injection in group I and group II

<table>
<thead>
<tr>
<th>Complication</th>
<th>Group I (n=65 patients)</th>
<th>Group II (n=60 patients)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No</td>
<td>%</td>
</tr>
<tr>
<td>Pain</td>
<td></td>
<td></td>
</tr>
<tr>
<td>tolerable</td>
<td>47</td>
<td>72.3%</td>
</tr>
<tr>
<td>Intolerable</td>
<td>18</td>
<td>27.6%</td>
</tr>
<tr>
<td>Fever</td>
<td>8</td>
<td>12.3%</td>
</tr>
<tr>
<td>Vomiting</td>
<td>7</td>
<td>10.8%</td>
</tr>
<tr>
<td>Peritoneal collection</td>
<td>2</td>
<td>3.1%</td>
</tr>
<tr>
<td>Pleural effusion</td>
<td>2</td>
<td>3.1%</td>
</tr>
<tr>
<td>Subcapsular hematoma</td>
<td>2</td>
<td>3.1%</td>
</tr>
<tr>
<td>Portal vein thrombosis</td>
<td>3</td>
<td>4.6%</td>
</tr>
</tbody>
</table>

Table (3): Biochemical tests in group I before and 3 months after injection.

<table>
<thead>
<tr>
<th></th>
<th>Group I before</th>
<th>Group I after</th>
<th>t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>α FP (normal 10 u/dl)</td>
<td>247.9±477</td>
<td>227.2±421</td>
<td>2.38</td>
<td>0.019</td>
</tr>
<tr>
<td>AST (normal up to 40 u/dl)</td>
<td>1.8-2690</td>
<td>2.1950</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ALT (normal up to 40 u/dl)</td>
<td>69.6±32.3</td>
<td>61.7±19.3</td>
<td>2.19</td>
<td>0.03</td>
</tr>
<tr>
<td>BIL (normal 0.3-1.2 mg/dl)</td>
<td>19-150</td>
<td>29-117</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PT (normal 11-14 second)</td>
<td>51.3±20.7</td>
<td>46.9±11</td>
<td>1.7</td>
<td>0.07</td>
</tr>
<tr>
<td>ALB (normal 3.5-5.3 g/dl)</td>
<td>0.6-2.1</td>
<td>0.8-3.1</td>
<td>0.4</td>
<td>0.67</td>
</tr>
<tr>
<td>ALP (normal 75-250 u/dl)</td>
<td>3.5±0.4</td>
<td>3.4±0.47</td>
<td>1.31</td>
<td>0.19</td>
</tr>
<tr>
<td>(normal 0.3-1.2 mg/dl)</td>
<td>2.9-4.5</td>
<td>2.5-4.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(normal 75-250 u/dl)</td>
<td>238±40</td>
<td>245±45</td>
<td>1.6</td>
<td>0.12</td>
</tr>
<tr>
<td>(normal 0.3-1.2 mg/dl)</td>
<td>258.5±477</td>
<td>227.2±421</td>
<td>2.38</td>
<td>0.019</td>
</tr>
<tr>
<td>(normal 11-14 second)</td>
<td>115-350</td>
<td>76-590</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>14.4±0.9</td>
<td>14.6±1.3</td>
<td>0.6</td>
<td>0.75</td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>12.2-16</td>
<td>12.05-18.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CRT (normal 0.5-1.4 mg/dl)</td>
<td>0.98±0.16</td>
<td>0.99±0.2</td>
<td>1.0</td>
<td>0.3</td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>0.6-1.3</td>
<td>0.6-1.6</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table (4): Biochemical tests in group II before and 3 months after injection.

<table>
<thead>
<tr>
<th></th>
<th>Group II before</th>
<th>Group II after</th>
<th>t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>α FP (normal 10 u/dl)</td>
<td>330.6±580.4</td>
<td>185.5±320.4</td>
<td>1.009</td>
<td>0.31</td>
</tr>
<tr>
<td>AST (normal up to 40 u/dl)</td>
<td>4.8-1890</td>
<td>3-1250</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ALT (normal up to 40 u/dl)</td>
<td>72.5±37.2</td>
<td>66.8±48.3</td>
<td>0.752</td>
<td>0.45</td>
</tr>
<tr>
<td>BIL (normal 0.3-1.2 mg/dl)</td>
<td>13-143</td>
<td>32-270</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PT (normal 11-14 second)</td>
<td>53.7±29.1</td>
<td>46.8±15.2</td>
<td>2.52</td>
<td>0.014</td>
</tr>
<tr>
<td>(normal 0.3-1.2 mg/dl)</td>
<td>11-147</td>
<td>15-76</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(normal 0.3-1.2 mg/dl)</td>
<td>1.13±0.32</td>
<td>1.4±1.26</td>
<td>1.67</td>
<td>5.1</td>
</tr>
<tr>
<td>ALB (normal 3.5-5.3 g/dl)</td>
<td>0.6-1.8</td>
<td>0.8-6.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(normal 3.5-5.3 g/dl)</td>
<td>3.4±0.46</td>
<td>3.27±0.41</td>
<td>1.69</td>
<td>0.09</td>
</tr>
<tr>
<td>ALP (normal 75-250 u/dl)</td>
<td>2.8-4.3</td>
<td>2.5-4.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(normal 75-250 u/dl)</td>
<td>225±41</td>
<td>255±41</td>
<td>1.4</td>
<td>0.21</td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>107-355</td>
<td>71-650</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PT (normal 11-14 second)</td>
<td>14.1±1.3</td>
<td>13.8±0.9</td>
<td>2.95</td>
<td>0.004</td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>11.7-16.3</td>
<td>12.5-16.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>1.01±0.2</td>
<td>1.03±0.2</td>
<td>1.76</td>
<td>0.08</td>
</tr>
<tr>
<td>(normal 0.5-1.4 mg/dl)</td>
<td>0.6-1.5</td>
<td>0.8-1.4</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table (5): Follow up of complete ablation in both groups at 3, 6 and 12 months after injection.

<table>
<thead>
<tr>
<th></th>
<th>Group I (n=68 lesions)</th>
<th>Group II (n=63 lesions)</th>
<th>X2</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No</td>
<td>%</td>
<td>No</td>
<td>%</td>
</tr>
<tr>
<td>3 months</td>
<td>41</td>
<td>60.3</td>
<td>54</td>
<td>85.7</td>
</tr>
<tr>
<td>6 months</td>
<td>33</td>
<td>48.5</td>
<td>47</td>
<td>74.6</td>
</tr>
<tr>
<td>One year</td>
<td>27</td>
<td>39.7</td>
<td>43</td>
<td>68</td>
</tr>
</tbody>
</table>

Table (6): Follow up of complete ablation in both groups at 6 and 12 months after injection.

<table>
<thead>
<tr>
<th></th>
<th>Group I (n=41 lesions)</th>
<th>Group II (n=54 lesions)</th>
<th>X2</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No</td>
<td>%</td>
<td>No</td>
<td>%</td>
</tr>
<tr>
<td>After 6 months</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Still ablated</td>
<td>33</td>
<td>80.4</td>
<td>47</td>
<td>87</td>
</tr>
<tr>
<td>New lesions</td>
<td>4</td>
<td>9.7</td>
<td>4</td>
<td>7.4</td>
</tr>
<tr>
<td>Local Recurrence</td>
<td>4</td>
<td>9.7</td>
<td>3</td>
<td>5.5</td>
</tr>
<tr>
<td>Died</td>
<td>3</td>
<td>7.3</td>
<td>1</td>
<td>1.8</td>
</tr>
<tr>
<td>After one year</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Still ablated</td>
<td>27</td>
<td>65.8</td>
<td>43</td>
<td>79.6</td>
</tr>
<tr>
<td>New lesions</td>
<td>6</td>
<td>14.6</td>
<td>7</td>
<td>13</td>
</tr>
<tr>
<td>Local Recurrence</td>
<td>8</td>
<td>19.5</td>
<td>4</td>
<td>7.4</td>
</tr>
<tr>
<td>Died</td>
<td>8</td>
<td>19.5</td>
<td>7</td>
<td>13</td>
</tr>
<tr>
<td>One year survival</td>
<td>33</td>
<td>80.5</td>
<td>48</td>
<td>87</td>
</tr>
</tbody>
</table>

Fig. (2): (a) CT study shows HCC with rapid uptake in the arterial phase before treatment. (b) CT study of the same focal lesion shows no uptake (complete ablation) after ethanol injection.
DISCUSSION
Percutaneous ablation is the best treatment option for patients with early stage HCC who are not suitable for surgical resection or transplantation [8]. Ethanol induces immediate coagulative necrosis and injury then thrombosis of tumor cells and enable the complete ablation of small neoplastic lesions without adversely affecting liver function. PEI is a procedure of easy execution, good tolerability and low cost, which can be applied in repeated sessions [2] and that is why we used this maneuver for percutaneous ablation in our low resource community. PEI performed under ultrasonographic guidance achieves complete tumor necrosis in 70%–80% of solitary HCC ≤ 3cm [11] and in almost 100% in tumors less than 2 cm. Tumor necrosis is less likely to be achieved in large tumors; 70% necrosis is reported for tumors between 2 and 3 cm and 50% necrosis for HCC between 3 and 5 cm [12].

The median age of patients in this study is 60.7 years which is slightly higher than ages recorded in another Egyptian study and estimated to be 56 years [13], and this may be due to improvement of general health by preventive programs and introduction of antiviral agents in treatment of chronic HCV and HBV.

In our study the male (n=92) to female (n=33) ratio was 2.7:1, which is in agreement with both the local Egyptian ratio 3:1 stated by Gad El-Mawla et al.[13], and the international ratios of 2.6:1 in China and 3.1:1 in Italy [14].

HCC usually develops following chronic liver inflammation caused by hepatitis C or B virus [15]. This is also applied to our study where chronic HCV infection is the most common cause of liver disease in our series (89.6% of patients versus 8.8% chronic HBV and 1.6% cases of coinfection) and this reflects the situation in Egypt where HCV prevalence
reported among several population groups reaches up to 20% [16], while low prevalence of chronic HBV coincide with intermediate endemicity (2-8%) of HBsAg carrier rate in Egypt[17]. These results are not in agreement with Abdel-Wahab et al., [18] who found positive virology in only 82.5% of HCC patients (61% HCV, 14.5% HBV and 7% coinfection); this may be attributed to the advancement in the diagnostic methods.

Abdominal pain (right upper quadrant), anorexia and loss of weight were the most common presenting symptoms, while liver cirrhosis and splenomegaly were the predominant signs, this is in agreement with Kew [19].

As regard treatments for HCC, liver transplantation can eliminate tumors and cirrhosis at the same time, and is considered to be the most appropriate treatment for patients with early HCC [20]. But, the lack of liver donors is a major limitation [21]. In Egypt further obstacles include high prevalence of chronic liver diseases in particular HCV in addition to financial constraints for the high cost of transplantation and the lack of experience for living donor liver transplantation.

Till the time this study is planned for in February 2009, many studies had been published to evaluate effect of percutaneous injection of ethanol in treatment of HCC, but few studies evaluating percutaneous injection of mitoxantrone in treatment of HCC were published and none -to our knowledge- evaluated effect of percutaneous combined injection of ethanol and mitoxantrone in treatment of HCC.

In a study done by Sung et al;[22] between January 1995 and April 1999, 64 patients with HCC were treated by PEI as first-line treatment and therapeutic efficacy was assessed by US, CT and AFP. Overall survival rates at one year was 92% and the corresponding cancer free survival rates were 56%. The local tumor progression rates were 23% (9 of 39 HCCs) for tumors ≤ 2cm in diameter. The local tumor recurrence is comparable to our results (19.5%), while the survival rate is better in their study due to different etiology of their patients as HCC develop in chronic hepatitis B viral infection with good synthetic function of the liver.

In our study the percentage of complete ablation in group I at 3, 6, 12 months after the use of PEI alone is 60%, 48.5% and 39.7% respectively. Which is in agreement with Sung and his colleagues[22] who obtain the same percentage of ablation in large lesions 2-5 cm in diameter.

The use of combined PEI and PIM resulted in a significantly higher rates of complete ablation at 3, 6 and 12 months: 85.7%, 74.6% and 68%, respectively and indicate the efficacy of mitoxantrone addition to PEI. These results may attributed to effect of ethanol on blood vessels draining the tumor leading to their thrombosis that impair systemic absorption of mitoxantrone and maximize its local effect.

Lipidol has high affinity to malignant hepatocytes, when mixed with mitoxantrone leads to selective uptake by malignant hepatocytes leading to additional ablative effect and more prevention of local recurrence rate observed in group II compared to group I.

The percentage of ablation after treatment with combination of PEI and PIM is 85.7% after 3 months. These results are comparable with many studies reporting similar frequency of ablation after radiofrequency ablation (RF) in patients with similar criteria[23]. We are in need for further controlled studies to compare RF and combination of PEI and PIM.

Intratumoral instillation of mitoxantrone results in a 1000-fold higher concentration in the tumor compared with intravenous administration. We preferred to do percutaneous interventions to obtain a higher drug concentration within the tumor without systemic toxicity and to preserve the integrity of the healthy liver parenchyma, which is an advantage over (TACE) or repeated surgery. This factor is of paramount importance as survival is dependent on the integrity of the liver function [24].

Farres et al. [25] evaluated PIM in hepatic focal lesions. They used 10 mg of mitoxantrone for lesions < 3 cm and 20 mg for lesions >3 cm. Three treatment sessions were performed 1 month apart for each of the 11 lesions. Follow-up by CT examination one month after therapy showed very low attenuation of the lesion indicating tumor necrosis. All injections were painless except four and patients never required analgesics during or after the procedure. Doses of 10-20 mg PIM produces no side effects and no hematologic toxicity was observed [25]. When we compare these results with our results, there were higher rate of intolerable pain, fever and vomiting and other local aggressive complications, ethanol injection would be accused as the cause of these complications.

www.mis.zu.edu.eg/ajied/home.aspx
The histologic effects of locoregional mitoxantrone treatment are characterized by complete tumor necrosis in which dead tumor cells are surrounded by an inflammatory infiltrate and a fibrotic organization of liver tissue around the tumor. This structure tends to isolate the lesion, preventing their expansion and promoting the persistence of the drug at the injected site. This fibrous rim reaction could also explain why lesion remain stable and do not shrink after PIM treatment. Furthermore this rim could prevent proliferation of residual cancer cells and this explain absence of aggressive local complications in group II of our study e.g. peritoneal reaction.

The drawback of PIM as with all localized treatments is that it does not preclude the emergence of other tumor foci or the progression of untreated tumors.

In this study the rate of local recurrence is higher in group I in comparison to group II and this additional benefit may be attributed to PIM and its induced perilesional fibrous reaction.

In this study the synthetic liver functions and consequently Child class seems to be improved in group II than group I and hence combined use of PEI and PIM seems to be superior to PEI alone.

In conclusion, PEI followed by PIM seems to be better than PEI alone and this is reflected by higher rates of complete ablation of focal hepatic lesions at 3,6 and 12 months. Improvement of synthetic liver functions and consequently Child classes is better in group II than group I.

Funding: Non.

Conflicts of interest: Non.

Ethical approval: The protocol of the study was approved by the ethical committee of Faculty of Medicine, Zagazig University. Informed consents were obtained from all patients.

REFERENCES

Interferon Therapy For Elderly Egyptian Patients With Chronic Hepatitis C of Genotype 4

Mohammed Emam¹, Waleed A. Abd El-Dayem¹, Soha AEIhawari¹, Hayam Heeba², Amany Emara³

¹ Tropical Medicine Department, Faculty of Medicine, Zagazig University, Egypt.
² Internal Medicine Department, Faculty of Medicine, Ain Shams University, Egypt.
³ Clinical Pathology Department, Faculty of Medicine, Ain Shams University, Egypt.

Background and study aim: Hepatitis C (HCV) affects nearly one in every 5 Egyptians which is the highest incidence all over the world. Most of the Egyptian Chronic Hepatitis (CHC) patients are of genotype 4 where it represents 90% of all Egyptian HCV cases. The clinical utility of antiviral therapy in elderly patients in our locality is not clear, also little information is available in literatures all over the world on treatment of such group of patients with genotype 4. The present study aimed at evaluating the efficacy and safety of combination therapy (pegylated interferon alpha 2a (PegIFN-alpha2a) and ribavirin) in treatment of elderly Egyptian patients with HCV genotype 4.

Patients and Methods: 60 elderly Egyptian patients (more than 55 years) with chronic HCV (group 1) and another group of 72 younger (less than 55 years) age patients (group 2) were enrolled in the present study. Both group of patients were compensated and all of genotype 4. Both groups received 180 mcg PegIFN-alpha2a subcutaneously once weekly and ribavirin (1000-1200mg/daily) for 48 weeks. Patients were followed for 48 week and sustained virological response and safety were assessed in both group.

Results: A significant improvement in both end of treatment response (ETR) and sustained virologic response (SVR) was noted in both group, where ETR was achieved in 32 (53.3%) and 41 patients (56.9%) in both groups respectively, and 27 patients in group 1 (45.0%) and 38 (52.8%) in group 2 could retain negative viraemia SVR by the end of follow up period. SVR showed a non-significant negative correlation with age. Viral clearance after 4 weeks of therapy was associated with high incidence of ETR and SVR (P <0.001), but without significant difference between both group. Rate of discontinuation and periods of discontinuation and side effect and safety was not significantly different in both groups.

Conclusion: Despite these challenges, the present study showed that HCV treatment was generally well tolerated by the elderly Egyptian patients (55-68 years) with a little or no significant difference in SVR as well as therapy discontinuation rates secondary to adverse effects compared to younger age groups. Therefore, we recommend that chronic HCV Egyptian patients of age 55 years and more should be included in trials of chronic hepatitis C treatment and old age is no more contraindication for interferon/ribavirin therapy and the risk-benefit of antiviral therapy should be assessed on an individual basis.

INTRODUCTION

The World Health Organization has declared hepatitis C a global health problem, with approximately 3% of the world’s population (roughly 170-200 million people) infected with HCV. In the US, approximately 3 million people are chronically infected, many of whom are still undiagnosed[1]. In Egypt the situation is quite worse. Egypt has a population of 80 million and contains the highest prevalence of hepatitis C in the world. The national prevalence rate of HCV antibody positivity has been estimated to be between 10-13%[2]. Hepatitis C
affects nearly one in every 5 Egyptians which is highest prevalence all over the world. Most of Egyptian HCV patients are of genotype 4 where it represents 90% of all Egyptian HCV cases [3]. Genotype 4 is one of the neglected hepatitis C virus allow the world[4]. In our locality elderly patients with chronic HCV infection have been an understudied population due to several factors. These factors include exclusion of subjects older than 55 years of age from governmental programs of treatment, shortage of financial support, reluctance to treat HCV infection in the elderly due to fear of dealing with more HCV therapy related adverse effects, co- morbidities and risk factors of aging.

In Egypt anti-schistosomal injection therapy was the main cause of contamination, followed by procedures performed by informal providers and traditional healers such as dental care, wound treatment, circumcision, and deliveries. CHC is also highly prevalent in sub-Saharan Africa and in the Middle East. In Europe, its prevalence has recently increased particularly among intravenous drug users and in immigrants[4]. Chronic HCV is the main cause of liver cirrhosis and liver cancer in Egypt and, indeed, one of the top five leading causes of death[5]. Preliminary evidence suggests that genotype 4 hepatitis C infection may place the patient at greater risk for hepatocellular carcinoma more than other HCV genotypes. Despite the decrease in the incidence of acute hepatitis C, the prevalence of long-standing chronic hepatitis C infection is increasing among older adults[6]. Prevalence of HCV infection in elderly patients are variable all over the world, in a study that was done in Italy among 496 elderly with a mean age of 79.31 years old, the prevalence of positive HCV antibody was found to be around 11%[7]. A study from Japan revealed 8.8% and 13.1% of HCV antibody seropositivity in hospital and autopsy cases older than 60 years of age, respectively[8]. However no much data are available for the prevalence of CHC in elderly patients in Egypt. However Sayed et al.[9] reported that the highest prevalence rates in Egypt were seen in patients between 55-65 years old. The age at the time of infection was significantly associated with the development of cirrhosis[7]. The median time from the age of infection to the onset of cirrhosis that was 33 years in patients who acquired the HCV infection at the age of 21 to 30 was reported to decrease to 16 years in patients who had the infection after the age of 40[10]. The mean time from the age of blood transfusion to the development of cirrhosis was reported to be 9.8 years in patients who had blood transfusion at the age of 50 or more. Also investigators from Japan reported a significantly shorter interval between the time of blood transfusion and the diagnosis of HCV-associated hepatocellular carcinoma if the blood transfusion was received at an older age[8].

The hepatitis C virus (HCV) genotype is one of the main predictors of response to interferon (IFN)-based therapies. Information about treatment response of HCV genotype 4 patients is scarce and conflicting results have been reported[13]. Although the safety and efficacy of hepatitis C therapies have been extensively studied in patients between ages of 18 and 60, patients who are over 65 still remain understudied and difficult to treat population. Some past research indicates that older individuals do not respond as well to interferon-based therapy, but data are inconsistent. Adverse effects of antiviral therapy are more prevalent in elderly patients (age ≥60 years) with chronic hepatitis C (CHC) than in their younger counterparts, and often interrupt or halt their treatment. Interferon and ribavirin treatment is, therefore, considered to have reduced efficacy in this group[14]. Elderly patients with CHC, however, have an increased risk of developing hepatocellular carcinoma, and antiviral therapy is an effective way of reducing this risk and improving survival. However, the clinical utility of antiviral therapy in elderly patients is not clear and little information is available on their prognosis, with or without such therapy. Canadian consensus guidelines recommended that old age more than 65 years is no more contraindication for interferon/ribavirin therapy[15]. In Egypt nearly no data are available about combination (interferon/ribavirin) therapy for this group of patients with HCV of genotype 4 as elderly patients are out of treating governmental programs due to shortage of financial support, reluctance to treat HCV infection in the elderly due to fear of dealing with more HCV therapy related adverse effects, co morbidities and risk factors of aging. The purpose of the present study was to study the effectiveness and safety of IFN-α2a (Pegasys, 180mcg) once weekly in combination with ribavirin, in elderly Egyptian compensated HCV patients of genotype 4.
PATIENTS AND METHODS

**Detailed Description**: A prospective, multi-center, case control study conducted in Tropical Medicine Department, Zagazig University, Egypt and Riyadh National Hospital (RNH) (KSA) enrolling 60 Egyptian patients with chronic hepatitis C of genotype 4 who are 55 to 68 years of age (group 1) and other sex- and HCV genotype-matched 72 chronic hepatitis C Egyptian patients who are 22 to 54 years of age (group 2) conducted for comparison. Most of patients in RNH group were of old age those patients entered KSA before the obligatory pre-employment HCV examination starting since 15 years. All patients received pegylated interferon-alpha 2a (PEGASYS®) 180 mcg/week and Ribavirin 1000-1200 mg/day, the combination therapy given for 48 weeks of treatment. The primary outcome measurement was sustained virological response and safety of treatment in both studied groups. Secondary Outcome Measures, rapid virological response (RVR), defined as HCV RNA <15 IU/mL at week 4 treatment.

**Inclusion Criteria**: Patients have never been treated with traditional interferon plus ribavirin or peginterferon plus ribavirin. the patient were subjected to the following:

- Serologic evidence of chronic hepatitis C infection by an anti-HCV antibody test (performed using a third-generation enzyme immunoassay (MEIA; Abbott Laboratories, Abbott Park, IL, USA).
- Detectable serum HCV-RNA was assessed both qualitatively (COBAS AMPLICOR-HCV Test, v2.0; Roche Diagnostics; lower limit of detection 50 IU/mL) and quantitatively (COBAS AMPLICOR-HCV MONITOR Test, v2.0, Roche Diagnostics, lower limit of quantitation 15 IU/mL).
- HCV genotype was determined in all patients by using hybridisation techniques (Innolipa HCV, Bayer).
- Liver biopsy findings consistent with the diagnosis of chronic hepatitis C infection with compensated cirrhosis (Exception: patients with bleeding tendency in whom biopsy is medically contra-indicated and patients more than 60 years do not require biopsy).
- Compensated liver disease (Child-Pugh Grade A classification) with no history of esophageal Varices, Ascites and /encephalopathy.
- Measuring of serum TSH, AFP and ANA.
- Negative urine or blood pregnancy test (for women of childbearing potential) documented within the 24-hour period prior to the first dose of study drug.
- All fertile males and females receiving ribavirin must be using two forms of effective contraception during treatment and for six months after treatment end.
- Patient consent was obtained.

**Exclusion Criteria**: Patients were excluded from the treatment protocol if they had any of the following well-known contra-indications to antiviral therapy: If the patient is classified as child B or C chronic liver disease, haemoglobin <12 g/dL, white blood cells <4000 cell/mm3 (neutrophil count <2000/mm3), platelet count <100,000 cells/mm3, concomitant antibodies (ANA, AMA and ASMA positive), psychiatric disorders, infection with the human immunodeficiency virus, HBV positive patients, alcohol and/or drug abuse, severe cardiac or pulmonary disease. Also patients who are not responding at week 12 should stop treatment. All patients had normal thyroid function prior to the study. Patients with a creatinine clearance of <50 mL/minwere excluded form the study as ribuverin should not given in this cases.

**Follow up**: all patients were followed up for 48 weeks. Hematological parameters were assessed every 2 week for the first 8 weeks of treatment and patients underwent a complete blood count on a monthly basis. HCV RNA assessments (both qualitative and quantitative) were performed at weeks 4, 12 and 24 weeks and at the end of treatment. The dose modifications and the cause of treatment tailoring or stopping, where appropriate. The study endpoint in virological response includes : (1) 48 weeks negativity defined as undetectable serum HCV RNA after a 48 week of treatment, (2) SVR, defined as undetectable serum HCV RNA after a six months untreated follow-up period.

**Statistical analysis**:
Data were expressed as mean ± standard deviation or number and percentage. Data were compared using Chi-square test or Fisher’s exact, t- test when appropriate. P-value less than 0.05 were considered statistically significant.

**RESULTS**
This study was conducted in Department of Tropical Medicine, Zagazig University in Egypt and RNH hospital in KSA. The conducted study involving 132 chronic HCV Egyptian patients...
between December 2007 and May 2011. The patients were classified into two groups: 60 patients older than 55 years old (group 1), and 72 patients younger than 55 years of age (group 2). All scheduled for 48 weeks of combination therapy with interferon and ribavirin. Baseline demographic (apart from age) data and disease characteristics were similar in both groups (Table 1).

The age of acquisition of infection and mode of infection can not be defined exactly in both studied groups, however, the suspected duration of infection was significantly higher in old age group. Also history of blood transfusion, surgical, dental sable anti bilharzial treatment were reported more in old age patients.

One hundred twenty-four patients out of 132 completed the study and follow up periods. Five patients (8.3%) Of the elderly patients, had to discontinue therapy due to adverse effects such as severe thrombocytopenia and low white blood cell retinal hemorrhage. In the same group, 8 patients (13.3%) had to adjust or tailor treatment due to laboratory abnormalities, while in the younger age group only 4 (5.6%) and 9 (12.5%) of patient had to discontinue and adjust the treatment due to side effect and abnormal laboratory finding respectively. The discontinuation more than 10 weeks occurred in 10 (16.7%) and 8 (11.1%) patient in both groups respectively.

The viral load was significantly higher in old age grouping comparison to younger age group. In intent-to-treat analysis, the sustained virological response (SVR) rate was "substantially lower" for the older compared with the middle-aged group (45% vs. 52.8%, respectively), but the difference did not reach statistical significance (P = 0.88) The prevalence of adverse effects due to IFN therapy, especially lethargy, confusion, and changes in behavior, was higher for older patients.

Among older patients who experienced a rapid virological response at week 4 of treatment and received treatment for >80% of the duration of the treatment course, the SVR was responding at week 4 higher in those patients who r similar to those of younger age group (45% vs 52.8%, respectively) (p=0.37). The older group had a higher rate of treatment discontinuation but not significant compared with the middle-aged group (8.3% vs 5.6%, respectively; P = 0.77). The hematologic adverse events were the most common encountered adverse effects. There are three major problems encountered: neutropenia, thrombocytopenia, and anemia.

**Neutropenia**: Patients of both group showed significant neutropenia approximately 3-6 times during monthly follow up visits. Dose reductions occurred in 16 of individuals less than 55 years. Older patients were noted to have dose modifications for neutropenia in 12 patients'. The severity of neutropenia was higher in old age group than the younger group. It was the cause of stopping treatment in 3 patients in old age group and two patients in younger age group. Currently, dose reduction was the only management for neutropenia, no one in both groups has used GCSF.

**Thrombocytopenia**: 14 and 11 of patients in both group respectively needed reductions of treatment for thrombocytopenia during study. Dose reduction was recommended when platelet counts fall below 50,000. Two patients in old age group discontinued treatment due to severe thrombocytopenia. Discontinuation of therapy was recommended if platelet counts fell below 30,000.

**Anemia**: 23 and 34 patients in both group respectively experienced anemia (mild to moderate). Management of anemia was to reduce ribavirin dose for hemoglobin less than 10 g/dl, and obtain blood count levels every two weeks or more frequently. Ribavirin dosing was recommended to be discontinued in 4 patients in elderly group and only one patient in younger age group when hemoglobin falls below 8.5 g/dl. One patient in both groups were given erythropoietin [epoetin-alfa (Eprex, Janssen Cilag S.p.A)] at 40,000 international units weekly for few weeks (6-9 weeks) which significantly improves hemoglobin.

A regression analysis of sustained virologic response rate and ribavirin dosage shows a clear direct relationship between SVR and dose, that full dose scheduling of ribavirin permits virologic responders to have an ultimate SVR. Dose reduction results in a fall to 52.8% of younger age group compared to 45.09% in older age group individuals.

Intent to treat (ITT) analysis showed significant improvement in both of ETR and SVR with peg-IFN therapy, where ETR was 53.3% and 56.9%, respectively, while SVR was 45.09% and 52.8% (P < 0.37).

Regarding viraemia, there was a significant difference between responders and non-
responders in both groups and within the same group. Viral clearance after 4 weeks of therapy was associated with high incidence of ETR and SVR (P<0.6), but also without significant difference between both groups. Six patients in old age group and two in the other group could not continue the study due to sever side effects, which was of significant difference.

27 out of 34 old age patients who showed rapid virologic response maintained sustained virologic response, while 38 patients out of 44 showed the same response in younger age patients (Table 2).

### DISCUSSION

During the past decade, our knowledge of the pathogenesis, clinical course, and treatment of chronic hepatitis C virus (HCV) infection has increased tremendously. Chronic infection is prevalent and may be more severe in the elderly population. It is estimated that physicians will be encountering increasing numbers of elderly persons with liver diseases due to chronic HCV infection. However, there are hardly any data on the various aspects of pathogenesis and treatment of the disease in old age [17]. The aim of this article was evaluation the of efficacy and safety of the standard combination therapy in chronic HCV Egyptian elderly patient (age, more than 55 years) and to suggest an approach to management of the infection in this population.

The mechanisms underlying the relatively rapid progression of liver disease in older patient are not known. Most of the older adults with chronic hepatitis C virus infection acquired the disease earlier in life. These patients often present with complications of liver disease, mainly cirrhosis and hepatocellular carcinoma. The burden of chronic hepatitis C virus infection in elderly persons is expected to increase significantly in our locality in Egypt during the next 2 decades.

There are few clinical studies conducted in HCV-positive elderly patients especially in HCV

### Table (1): Demographic, clinical and biochemical characteristics in both groups

<table>
<thead>
<tr>
<th></th>
<th>Group 1 (n = 60)</th>
<th>Group 2 (n = 72)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>58.4 ± 7.5</td>
<td>43.2 ± 7.6</td>
<td>&lt;0.001**</td>
</tr>
<tr>
<td>Sex : Male</td>
<td>54 (90.0%)</td>
<td>57 (79.2%)</td>
<td>0.09</td>
</tr>
<tr>
<td></td>
<td>6 (10.0%)</td>
<td>15(20.8%)</td>
<td></td>
</tr>
<tr>
<td>HCV RNA IU/ml</td>
<td>387.262 ± 167.854</td>
<td>316,923 ± 154.849</td>
<td>&lt;0.05*</td>
</tr>
<tr>
<td>ALT &lt; 2 upper limit of normal n (%)</td>
<td>51(85.0%)</td>
<td>58 (80.5%)</td>
<td>0.05*</td>
</tr>
<tr>
<td>Suspected Duration of HCV infection (years)</td>
<td>26.4 ± 6.5</td>
<td>12.8 ± 4.3</td>
<td>&lt;0.001*</td>
</tr>
<tr>
<td>Platelet count(1000/µl)</td>
<td>135.28 ± 49.7</td>
<td>146.2 ± 52.8</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Hemoglobin (g/dL)</td>
<td>11.87 ± 2.5</td>
<td>13.20 ± 1.5</td>
<td>&lt;0.001*</td>
</tr>
<tr>
<td>Absolute neutrophil count (cells/µl)</td>
<td>1584±189</td>
<td>1615±164</td>
<td>&gt;0.05 NS</td>
</tr>
<tr>
<td>Discontinuation of treatment owing to adverse effects</td>
<td>5 (8.3%)</td>
<td>4 (5.6%)</td>
<td>0.77 NS</td>
</tr>
<tr>
<td>Modification of treatment owing to laboratory abnormalities</td>
<td>8 (13.3%)</td>
<td>9 (12.5%)</td>
<td>0.88 NS</td>
</tr>
<tr>
<td>Weeks of discontinuations</td>
<td>10 (16.7%)</td>
<td>8 (11.1%)</td>
<td>0.35 NS</td>
</tr>
<tr>
<td>1. Less than 10 weeks</td>
<td>2 (3.3%)</td>
<td>1 (1.4%)</td>
<td>0.87 NS</td>
</tr>
<tr>
<td>2. More than 10 weeks</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dose reduction due to :</td>
<td>12 (20.0%)</td>
<td>16 (22.2%)</td>
<td>0.75 NS</td>
</tr>
<tr>
<td>Neutropenia</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thrombocytopenia</td>
<td>14 (23.3%)</td>
<td>11 (15.3%)</td>
<td>0.23 NS</td>
</tr>
<tr>
<td>Anemia</td>
<td>23 (38.3%)</td>
<td>34 (47.2%)</td>
<td>0.3 NS</td>
</tr>
<tr>
<td>Number of patients could not continue the study</td>
<td>6 (9.9%)</td>
<td>2 (2.8%)</td>
<td>&lt;0.001*</td>
</tr>
</tbody>
</table>

*P<0.05 (significant)  
NS : non significant

### Table (2): Virological response in both group.

<table>
<thead>
<tr>
<th></th>
<th>Group 1 (n = 60)</th>
<th>Group 2 (n = 72)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 week response (RVR)</td>
<td>34/60 (56.7%)</td>
<td>44/72 (61.1%)</td>
<td>0.6 NS</td>
</tr>
<tr>
<td>48 week response (ETR)</td>
<td>32/60(53.3%)</td>
<td>41/72 (56.9%)</td>
<td>0.67 NS</td>
</tr>
<tr>
<td>Sustained virological response (SVR)</td>
<td>27/60(45.0%)</td>
<td>38/72 (52.8%)</td>
<td>0.37 NS</td>
</tr>
</tbody>
</table>

www.mis.zu.edu.eg/ajied/home.aspx
patients genotype 4. One of the major reasons for this could be the exclusion of subjects who were 55 years or older in HCV related clinical studies[16-18]. In several randomized trials, the mean age was reported to be around 40 years[19-21]. Other reasons for exclusion of older participants could be the increased co-morbid conditions at older age, the fear of facing more adverse effects during the HCV treatment, or reluctance to perform liver biopsies in the elderly. In Egypt, patients more than 55 years old are out of governmental programs for HCV treatment, and nearly no studies were conducted on these a group in our country.

The mechanisms underlying the relatively rapid progression of liver disease in older adults are not known. Possible mechanisms for the role of aging in fibrosis progression are higher vulnerability to environmental factors (especially oxidative stress)[18], reduction in the rate of hepatic blood flow, and reduced mitochondrial capacity[19,20], as well as impaired immunity, may explain the significantly higher viremic load in older patients[16].

The current standard of care for HCV infection is pegylated IFN-α and oral ribavirin[16,30]. The goal of antiviral treatment is to prevent complications of the disease, mainly cirrhosis and HCC. We propose that, for all older patients, treatment decisions should be individualized on the basis of the severity of the liver disease, potential for serious adverse effects, likelihood of treatment response, and presence of comorbid conditions. Therapy is contraindicated for patients with decreased life expectancy due to severe hypertension, heart failure, or coronary artery disease; poorly controlled diabetes; or obstructive lung disease[16]. Chest radiography and electro-cardiography are prudent to exclude significant pulmonary and cardiac disease that may be exacerbated by ribavirin-associated anemia[36]. We believe that, owing to the higher risk of adverse effects from antiviral treatment for elderly persons, the degree of liver fibrosis should be assessed before consideration of therapy. There are only a few, nonrandomized studies on treatment of HCV infection in elderly patients. Early reports did not assess the rate of sustained virologic response[19-22].

A study in France comparing chronic HCV infection in patients more than 65 years of age with that in younger patients demonstrated that the older group had a significantly longer duration of infection (26 vs. 20 years), a higher age at infection (50 vs. 24 years) and increased likelihood of a history of transfusion (51% [449 of 881] vs. 29% [957 of 3301]) [23], this also was in agreement with the present study where the suspected duration of infection was higher in our old age group. Among persons who underwent liver biopsy, the fibrosis stage was higher for those aged 65 years, regardless of infection duration.

A different message derives from a population-based study in Italy of adults aged 60 years[24]. Although 4.1% of the participants (44 of 1063) had HCV antibodies, only 54.3% (19 of 35 anti-HCV-positive persons studied) had HCV viremia, all of whom were either asymptomatic or had mild liver disease. The discrepancy between these studies may be due to the differing characteristics of the study populations. The first population consisted only of patients referred for treatment, whereas the second was community based and included all individuals with HCV antibodies. It may be that many older patients had either recovered from the disease or were asymptomatic carriers.

The efficacy of therapy with IFN (or pegylated IFN) and ribavirin in older adults was reported in 2 small case series. Among 20 patients >65 years of age, the rate of sustained virologic response was 45% (9 patients)[29], and among 30 patients with a mean age of 65 years, 30% (9 patients) achieved a sustained response[28]. The rate of sustained virologic response reported for younger populations treated with pegylated IFN and ribavirin is, on average, 55%[29-30], this results were also in accordences with our results for both old age and younger age group.

It seems important that, for elderly patients with chronic hepatitis C, the risk-benefit of combination antiviral therapy consisting of pegylated interferon and ribavirin should be assessed on an individual basis. Assessment should be performed in all cases before considering treatment, and it should include evaluation of the degree of liver fibrosis by means of liver biopsy or, possibly, by means of noninvasive methods. However, there is a need for prospective randomized controlled trials to be conducted in HCV Egyptian patients older than 68 years of age for better evaluation of the safety and efficacy of HCV treatment in this age group[25]. In addition, more epidemiologic studies are needed for better assessment of the prevalence as well as the risk factors of chronic HCV infection in these elderly subjects. Therefore, we recommend that patients of age 55 years and more should be included in trials of
chronic hepatitis C treatment. For those groups of patients, risk-benefit of antiviral therapy should be assessed on an individual basis[26]. In the present study the age more than 55 years was an independent predictor of poor response (OR for sustained response of those aged more than 55 years). It is not known whether the rate of response in persons more than 68 years of age is the same as or worse than that for persons 55–68 years of age. Nevertheless, the AASLD guideline does not stipulate an upper age limit for antiviral therapy[16], although, in practice, elderly patients are less considered and referred for treatment. We believe that therapy should be considered for patients up to the age of 68 years.

**Funding:** Non.

**Conflicts of interest:** Non.

**Ethical approval:** The protocol of the study was approved by the ethical committee of Faculty of Medicine, Zagazig University. Informed consents were obtained from all patients.

**REFERENCES**


Probiotics in Early grades of Hepatic Encephalopathy

Soha A Elhawari¹, Emad F Hamed²
¹Tropical Medicine Department, Faculty of Medicine, Zagazig University, Egypt
²Internal Medicine Department, Faculty of Medicine, Zagazig University, Egypt.

Background and study aim: Modification of intestinal flora, through different mechanisms is regarded as a therapeutic option in management of hepatic encephalopathy. We conducted this study to test the ability of probiotics in treatment of early grades of hepatic encephalopathy in Egypt.

Methods: One-hundred educated cirrhotic patients without overt hepatic encephalopathy were included. Diagnosis of cirrhosis based on clinical, ultrasonographic and laboratory findings and patients were screened for grade-I and minimal hepatic encephalopathy by psychometric tests. Hepatic encephalopathy was diagnosed if at least one of psychometric tests was abnormal. Thirty patients with abnormal psychometric tests were included in the final analysis. Acidophilus capsules and banana were given to all patients 30 minutes before meals 3 times daily for 2 weeks.

Results: Minimal and grade-I hepatic encephalopathy were found in thirty out of the one hundred screened patients (30%). Minimal and grade-I hepatic encephalopathy were more common in Child class C, than in class B and A. Two weeks after Lactobacillus acidophilus intake, patients showed significant improvement in the performance of psychometric tests (P<0.001) while liver function tests and Child classes did not improve (P>0.05). Probiotics improved the clinical grades of hepatic encephalopathy; 96.7% (n=29) of patients showed improvement in clinical grade of hepatic encephalopathy and only one patient (3.3%) did not improve (P<0.05).

Conclusion: Probiotics could be used to treat early grades of hepatic encephalopathy. Child classes and liver functions did not improve significantly after probiotic therapy.

INTRODUCTION

Probiotics have been defined as viable microorganisms that when ingested have a beneficial effect in the prevention and treatment of specific pathologic conditions [1]. Currently, the best studied probiotics are lactic acid producing bacteria, particularly Lactobacillus and Bifidobacterium species [2]. Probiotics action is mediated through different mechanisms including: modulation of the intestinal microflora composition, increasing numbers of health – promoting genera (Lactobacillus and Bifidobacterium) and decreasing numbers of potentially harmful ones (several strains of Clostridium and Enterococcus); blocking the adhesion sites of pathogens; degradation of toxin receptors; reduction of luminal pH; competition for nutrients with gut microbes; production of inhibitory substances e.g. lactic acid; production of protective substances e.g. butyric acid; enhancement of the gut flora metabolic activity; modification of the mucus layer; enhancement of epithelial barrier integrity and finally stimulation of the immune system [3,4]. It thus could be used as alternative to or as adjuvant with antibiotics for gut sterilization [5]. Consequently; probiotics were used in treatment of different GIT diseases including lactose intolerance, inflammatory bowel disease, and various types of diarrhea [6].

The general term of hepatic encephalopathy (HE) covers all the neurological and psychological symptoms in patients with liver disease that cannot be explained by the presence of other pathologies.
It is a clinical syndrome with a wide range of variability extending from minimal impairment of intellectual function only detectable by psychometric testing- called minimal hepatic encephalopathy (MHE) to profound coma with signs of decerebration. The pathogenesis of hepatic encephalopathy is unknown. Many theories have been proposed; the most frequently discussed is the production of endogenous neurotoxins in particular ammonia. Since the pathogenesis of HE is mostly due to endogenous neurotoxins produced in the intestine that lack detoxification by the diseased liver, so modification of intestinal flora has been examined as a possible therapeutic option in treatment of HE [7], through different mechanisms, including suppression of harmful micro-organisms or stimulation of beneficial organisms [8,9]. And this was reflected in some studies that used probiotics to treat minimal [10,11], episodic [12,13] and grade II HE [14], but none to our knowledge conducted in Egypt. We conducted this study to test the ability of probiotics to treat early grades (minimal and grade I) hepatic encephalopathy.

**PATIENTS AND METHODS**

This study was conducted in the Tropical Medicine Department, Zagazig University Hospitals, Egypt. It comprised 100 educated (all finished secondary school education) cirrhotic patients without overt HE. The diagnosis of cirrhosis based on clinical and ultrasonographic examination and laboratory assessment and they were screened for grade-I HE and MHE by psychometric tests. Forty eight educated apparently healthy subjects were also included as controls to determine cut off values of psychometric tests; because psychometric tests are influenced by level of education and culture. Hepatic encephalopathy was diagnosed if at least one of the psychometric tests was abnormal. Thirty patients had abnormal psychometric tests and were included in the final analysis.

**Exclusion criteria**

Patients who were illiterate, of grade II and more HE, patients who were exposed to factors that might affect gut flora such as treatment with antibiotics, lactulose, enemas or oral antimicrobials and patients recently receiving antacids, H2-blockers or proton pump inhibitors were excluded from the study. Also patients with chronic diseases other than cirrhosis e.g. renal failure, uncontrolled diabetes…..etc. were also excluded.

**Patient assessment**

All the studied individuals were subjected to:

1. Detailed history taking, with special attention to history of any criterion of the West Haven criteria of altered mental state

2. History of fermented dairy products consumption was taken and subjects who were taking fermented milk were advised to discontinue the intake at least 2 weeks before the study

3. Thorough clinical examination for signs of chronic liver disease.

4. Routine investigations which include liver function tests, prothrombin time (PT), kidney functions, complete blood picture and viral markers.

5. Child-Turcotte-Pugh classing was evaluated for each patient.

6. Abdominal ultrasonographic examination was done to diagnose liver cirrhosis and to detect the presence of ascites.

7. Psychometric tests were done to diagnose the presence of HE and were repeated after two weeks of *Lactobacillus* intake.

**Probiotic supplementation**

Acidophilus capsule (10 mg *Lactobacillus acidophilus*, which contains 100 million active organism, its naturally occurring metabolic product and mixture of rice flour, gelatin and magnesium stearate) was given to the patients 30 minutes before meals 3 times daily (Acidophilus, Walgreens, USA). Banana, as a source of fructooligosacharides was given (in equal amounts to each patient) with each acidophilus capsule 30 minutes before meals. Morelli et al., [16] found that noticeable increase of probiotic count in stools was achieved at the 5th day of oral intake which suggests that potential probiotic benefits can be obtained after only few days of intake, therefore all patients were given probiotic preparation containing *Lactobacillus acidophilus* for two weeks. Patients who showed no major improvement at two weeks of *Lactobacillus* intake were advised to take conventional therapy for HE.

**Psychometric tests**

The following tests were used for all patients and their results were compared to that of our control subjects.
1. Circle connection test (CCT)

The standardized test comprises ten circles of variable sizes distributed on a sheet of paper, and there are five variants of distributions of equal degree of difficulty. The subject is asked to connect the circles according to their sizes by drawing line starting from the smallest circle proceeding to the next larger in size until the largest circle is reached. The test score was the time taken to complete connection of the circles measured in seconds, including the time needed to correct any errors [17]. In this work, CCT was considered abnormal when the time taken by the patients was greater than the mean+2SD from that of the healthy controls (>26 seconds).

2. Number connection test A (NCT)

In the NCT, which measures cognitive motor abilities [18], patients must connect numbers from 1 to 25 (written in Arabic, with identification of both the starting and finishing point), printed on a paper as quickly as possible. The test was explained to the subject and a demonstration was shown for him, after he understood the test he was asked to perform it quickly. The test score was the time in seconds required to complete the test, including the time needed to correct any errors. Errors were not enumerated, but patients were instructed to return to the preceding correct number and then carry on. In this work, NCT was considered abnormal when the time taken by the patients was greater than the mean+2SD from that of the healthy controls (>47 seconds).

3. Digit symbol test (DST)

It is a subtest of the Wechsler Adult Intelligence Scale-Revised (WAIS) [19]. The patient was given a list of digits from 1 to 19 associated with symbols and was asked to fill in blanks with symbols that correspond to each number. The test score was the total number of correct sequential matching of symbols to numbers in a 90 second interval. After explanation of the test, an abbreviated demonstration was administered to ensure that the patient understood the test correctly. The test was in the Arabic version of the Wechsler Adult Intelligence Scale revised by Meleka [20]. In this work, DST was considered abnormal when the score gained by the patients was less than the mean-2SD from that of the healthy controls (<29).

STATISTICAL ANALYSIS

Data were checked, entered and analyzed using Epi-Info 2000 for data processing and statistics. Data were expressed as mean ± SD for quantitative variable, number and percentage for qualitative one. Chi-squared (X²) or fisher exact, t test and paired t test were used when appropriate. P <0.05 was considered significant.

RESULTS

In the present work out of 100 educated patients with cirrhosis, 30 patients had HE by psychometric tests. According to West Haven criteria of altered mental state [15], 25 patients (83.3%) had MHE and 5 patients (16.7%) had grade-I HE; 2 cases were females (6.7%) and 28 cases were males (93.3%). All patients in this study were of viral etiology, most of them were due to hepatitis C virus (table 1).

CCT, NCT and DST could diagnose 25, 28 and 24 cases respectively; all of them had improved performance after probiotic therapy except one who consumed big amounts of red meat (table 2). The number of patients who had normal scores of psychometric tests after the 2 weeks of probiotic therapy was 22, 21 and 10 for CCT, NCT and DST indicating a reversal rate of 88%, 75% and 42% respectively (table 2).

Concerning liver function tests there was a non-significant reduction in the serum bilirubin level, ALT level, and PT. Also, there was a non-significant increase in serum albumin level after probiotic therapy (table 3).

In this study Child-Turcotte-Pugh class improved in 2 cases (6.7%) who improved from class C to class B, while one case (3.3%) worsened from class B to class C with no change from class B or C to class A (table 4).

Although CCT was the most simple test used, it had low sensitivity, specificity, predictive value and diagnostic accuracy (table 6) and it was the least agreed upon screening test for HE in this study (kappa coefficient = 0.75). By the end of the two weeks therapy with Lactobacillus acidophilus, 29 patients (96.7%) were MHE and only one patient (3.3%) remained in grade-I HE, this patient is the same patient who consumed big amounts of red meat as mentioned. No side effects related to Lactobacillus acidophilus intake was reported during the study period.
### Table (1): Characteristics of studied groups.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>All cirrhotic patients (n= 100)</th>
<th>Patients with HE (n= 30)</th>
<th>Controls (n= 48)</th>
<th>Test of significant</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td>$\bar{X} \pm SD$</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>50±12 (16-55)</td>
<td>46.7±9.2 (16-54)</td>
<td>43.5±9.6 (15-50)</td>
<td>t =1.7</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>85</td>
<td>28</td>
<td>37</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>15</td>
<td>2</td>
<td>11</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Etiology of cirrhosis</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis B virus</td>
<td>20</td>
<td>6</td>
<td>20</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis C virus</td>
<td>60</td>
<td>17</td>
<td>56.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatitis B &amp; C</td>
<td>20</td>
<td>7</td>
<td>23.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Clinical grade of HE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grade-0</td>
<td>25</td>
<td>83.3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grade-1</td>
<td>5</td>
<td>16.7</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Child-Turcotte-Pugh class</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Class A</td>
<td>31</td>
<td>5</td>
<td>16.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Class B</td>
<td>39</td>
<td>12</td>
<td>40</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Class C</td>
<td>30</td>
<td>13</td>
<td>43.3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Table (2): Psychometric test scores among patients with HE before and after therapy.

<table>
<thead>
<tr>
<th>Psychometric test</th>
<th>Before therapy</th>
<th>After therapy</th>
<th>Mean difference</th>
<th>t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>CCT</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>33.6±7.4 (20-58)</td>
<td>19.9±4.4 (12-28)</td>
<td>-13.7±8 (-32:7)</td>
<td>9.34</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>NCT</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>64.5±15.2 (40-99)</td>
<td>41.7±12.3 (28-85)</td>
<td>-22.8±16.4 (-52-2)</td>
<td>7.62</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>DST</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>21.7±8.1 (8-45)</td>
<td>29.9±8.1 (19-51)</td>
<td>8.1±4.9 (-5:17)</td>
<td>9.05</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

### Table (3): Liver function tests before and after therapy among studied patients with HE.

<table>
<thead>
<tr>
<th>Liver function test</th>
<th>Before therapy</th>
<th>After therapy</th>
<th>Difference</th>
<th>t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total bilirubin (mg/dl)</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>2.58±1.96 (0.48-8.84)</td>
<td>2.44±1.67 (0.57-7.8)</td>
<td>-0.13</td>
<td>0.53</td>
<td>0.60</td>
</tr>
<tr>
<td>Albumin (gm/dl)</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>2.9±0.7 (2.1-4.5)</td>
<td>3.1±0.7 (2.2-4.7)</td>
<td>0.15</td>
<td>1.36</td>
<td>0.18</td>
</tr>
<tr>
<td>PT (in seconds)</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>18.5±4.2 (11-31)</td>
<td>18±2.99 (13.1-27)</td>
<td>-0.4</td>
<td>0.95</td>
<td>0.64</td>
</tr>
<tr>
<td>ALT (IU/L)</td>
<td>$\bar{X} \pm SD$</td>
<td>(Range)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>52.3±18.2 (24-109)</td>
<td>49.4±14.8 (20-82)</td>
<td>-2.9</td>
<td>1.39</td>
<td>0.17</td>
</tr>
</tbody>
</table>
Table (4): Child-Turcotte-Pugh classes before and after therapy among patients with HE.

<table>
<thead>
<tr>
<th>Child-Turcotte-Pugh class</th>
<th>Before therapy</th>
<th>After therapy</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>A</td>
<td>5</td>
<td>16.7</td>
<td>5</td>
</tr>
<tr>
<td>B</td>
<td>12</td>
<td>40.0</td>
<td>13</td>
</tr>
<tr>
<td>C</td>
<td>13</td>
<td>43.3</td>
<td>12</td>
</tr>
</tbody>
</table>

*Mc Nemar’s chi-square test

Table (5): Relationship between psychometric tests and Child-Turcotte-Pugh classes.

<table>
<thead>
<tr>
<th></th>
<th>Class A n = 5</th>
<th>Class B n = 12</th>
<th>Class C n = 13</th>
<th>Total n=30</th>
<th>X²</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients who were diagnosed by CCT</td>
<td>5</td>
<td>8</td>
<td>12</td>
<td>25</td>
<td>4.15</td>
<td>0.12</td>
</tr>
<tr>
<td>Patients who were diagnosed by NCT</td>
<td>4</td>
<td>12</td>
<td>12</td>
<td>28</td>
<td>2.31</td>
<td>0.31</td>
</tr>
<tr>
<td>Patients who were diagnosed by DST</td>
<td>3</td>
<td>9</td>
<td>12</td>
<td>24</td>
<td>2.67</td>
<td>0.26</td>
</tr>
<tr>
<td>Patients who were abnormal in all test</td>
<td>3</td>
<td>8</td>
<td>11</td>
<td>22</td>
<td>1.57</td>
<td>0.45</td>
</tr>
</tbody>
</table>

Table (6): Validity of psychometric tests.

<table>
<thead>
<tr>
<th></th>
<th>Sensitivity %</th>
<th>Specificity %</th>
<th>Predictive value %</th>
<th>Kappa of agreement</th>
<th>Diagnostic accuracy %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cases</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CCT</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;26 seconds</td>
<td>25</td>
<td>83.3</td>
<td>91.7</td>
<td>86.3</td>
<td>89.8</td>
</tr>
<tr>
<td>≤26 seconds</td>
<td>5</td>
<td>83.3</td>
<td>91.7</td>
<td>86.3</td>
<td>89.8</td>
</tr>
<tr>
<td>NCT</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;47 seconds</td>
<td>28</td>
<td>93.3</td>
<td>97.9</td>
<td>96.6</td>
<td>95.9</td>
</tr>
<tr>
<td>≤47 seconds</td>
<td>2</td>
<td>93.3</td>
<td>97.9</td>
<td>96.6</td>
<td>95.9</td>
</tr>
<tr>
<td>DST</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;29</td>
<td>24</td>
<td>80.0</td>
<td>100</td>
<td>100</td>
<td>88.9</td>
</tr>
<tr>
<td>≥29</td>
<td>6</td>
<td>80.0</td>
<td>100</td>
<td>100</td>
<td>88.9</td>
</tr>
</tbody>
</table>

- CCT was considered abnormal if the patient took more than 26 seconds.
- NCT was considered abnormal if the patient took more than 47 seconds.
- DST was considered abnormal if the patient scored less than 29.

DISCUSSION

Minimal HE was reported, world wide, to be present in 10% to 84% of cirrhotic patients without overt HE, depending on the diagnostic techniques used and patients selected for the studies [21]. In contrast to patients with symptomatic HE, patients with MHE have no recognizable symptoms of brain dysfunction [22]. MHE was assumed to have a negative effect on patients' daily functioning because psychomotor rather than verbal abilities tend to be affected and therefore treatment is recommended [3,23] and is regarded as a prophylactic measure against development of overt HE [24]. Patients with MHE are in need for close monitoring because of the fear to develop overt HE; more than 50% of them develop overt HE within a short period of time [24].

Clinicians may have difficulty in distinguishing patients with grade-I HE from patients with minimal HE [25]. That is why patients of grade 0 (MHE) and grade I were analyzed together in this study. In our study it occurs at a frequency of 30% in cirrhotic patients.

No study, to our knowledge, evaluated the effect of probiotics on early grades of HE in Egyptian cirrhotic patients. But Liu et al. [10] evaluated symbiotic modulation of gut flora on MHE in Chinese patients. They used symbiotic preparation consisting of 4 freeze-dried, non-urease producing bacteria, namely *Pediococcus pentosaceus* 5-33:3, *Leuconostoc mesenteroides* 32-77:1, *Lactobacillus paracasei* 19, *Lactobacillus plantarum* 2592, along with a fermentable fiber preparation consisting of glucan, inulin, pectin and resistant starch. They evaluated the effect of symbiotics on 58 cirrhotic patients with MHE. Patients were given symbiotic preparation for one month. Evaluation of therapy was done with the aid of NCT, brainstem evoked potentials and Child-Turcotte-Pugh classification. They found improvement of Child-Turcotte-Pugh class, improvement of MHE, improvement of liver functions, improvement of evoked potentials and NCT with
a positive correlation between NCT and Child-Turcotte-Pugh class. The discrepancies in the results between our study and that of Liu et al. may be due to: (1) all our cases are of viral etiology compared to 74% in their study, (2) longer duration of therapy one month in their study and two weeks in our study, (3) multiple probiotic strains they used compared to one strain in this study and (4) many fermentable prebiotics fibers they used. They reported that none of their patients developed overt hepatic encephalopathy during their study. Although the same applies to the present study yet no meaningful conclusion can be drawn from those observations as the duration of the study was 4 weeks in the study of Liu et al. and 2 weeks in the present study.

Also Saji et al. [11] evaluated probiotics on Indian cirrhotic patients with MHE. They received a probiotic preparation in a dose of one gram sachet containing not less than 1.25 billion spores of Lactobacillus acidophilus, Lactobacillus rhamnosus, Bifidobacterium longum and Sacharomyces boulardi, three times daily after meals. Diagnosis of MHE based on NCT-A, evoked potentials and arterial ammonia level and they were treated for 4 weeks before re-assessed by the same tests. By the end of treatment duration they failed to demonstrate any benefit for probiotics over placebo. Furthermore, liver functions were not assessed in this study. When compared to our study; most of their patients were of alcohol etiology; and alcoholics are known to relapse and deny that, also our patients were all inpatient with close monitoring.

Improvement of liver functions in this study and other studies concerned with the use of probiotics in liver diseases may be due to decreased exposure of the liver to inflammatory mediators, oxidative stresses and endotoxins released from the GIT [26-28].

None of our patients developed overt HE or adverse effects related to the probiotics during the study period and this is probably due to: (1) all patients in the present work were hospitalized, (2) none of them was alcoholic, (3) all of them were closely monitored, (4) all clearly instructed to avoid exposure to precipitants of HE and (5) safety of the probiotic strain we used, none of our cases developed adverse effects due to intake of Lactobacillus acidophilus.

Our results cope with the doubt in the ability of probiotics to improve viral induced liver damage. Although in this study Child-Turcotte-Pugh score showed improvement at the end of the two weeks period of therapy when compared to pretreatment period this improvement was non-significant, this is secondary to the non-significant improvement in liver functions.

Psychometric tests were used in detection and follow up of HE. Circle connection test can be done quickly and easily more than NCT and it also can be used by illiterate subjects [29]. NCT and DST were used in this work because they are simple, inexpensive and sensitive and have been evaluated in several studies for their value in the diagnosis, prognosis and treatment of HE [30,31]. In addition of its simplicity, NCT is relatively unaffected by patient's level of learning [32].

In Egyptian cirrhotic patients screened with psychometric tests, NCT had positive predictive value of 96.6% and negative predictive value of 95.9% and greater diagnostic accuracy (96.6%) compared to other psychometric tests used. NCT is the most sensitive test used to diagnose HE, and it had high significance agreement among the screening tests (CCT, NCT and DST) used for diagnosis of HE (kappa coefficient = 0.92). These results agree with Amodio et al. [33] who confirmed the efficacy of NCT as a simple tool in assessing cognitive alterations in cirrhosis. DST was the highest specific test in this study; it had specificity of 100%, sensitivity of 80%, positive predictive value of 100%, and negative predictive value of 88.9% and diagnostic accuracy of 92.3%. These results are in agreement with Li et al. (2004) [21] who concluded that DST and NCT test battery was sensitive and specific for detection of MHE.

CONCLUSIONS

Probiotics could be used to treat early grades of HE. Child-Turcotte-Pugh class of cirrhotic patients and liver functions did not improve significantly after probiotic therapy. Psychometric tests (CCT, NCT and DST) are valid as screening tests for HE in Egyptian patients.

Funding: Non.

Conflicts of interest: Non.

Ethical approval: The protocol of the study was approved by the ethical committee of Faculty of Medicine, Zagazig University. Informed consents were obtained from all patients.
REFERENCES


Elhawari and Hamed Afro-Egypt J Infect Endem Dis 2011; 1 (2):45-52

www.mis.zu.edu.eg/ajied/home.aspx


The Pattern, Risk Factors and Clinico-Aetiological Correlate of Tinea Capitis Among the Children in a Tropical Community Setting of Osogbo, South-Western Nigeria

Adeolu O. Akinboro¹, Olayinka A. Olasode², Olaniyi Onayemi³

¹Dermatology Unit, Department of Internal Medicine, LAUTECH Teaching Hospital, and College of Health Sciences, Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria
²Department of Dermatology and Venereology, Obafemi Awolowo University Teaching Hospital Complex, and College of Health Sciences, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria
³Department of Dermatology and Venereology, Obafemi Awolowo University Teaching Hospital Complex, and College of Health Sciences, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria

Background and study aim: Tinea capitis is an important infective dermatologic disease of worldwide distribution among children. Its frequency is increasing especially in the developing world, and has become an endemic disease in some places. To determine the prevalence, the risk factors, pattern and clinico-aetiological correlate of Tinea capitis among the children in Ilie community.

Patients and Methods: A total of 370 children aged 5 – 16 years; 185 with Tinea capitis as subjects and 185 relatively healthy children as controls. Multistage sampling method was employed, and house to house survey for Tinea capitis was conducted. The diagnosis of Tinea capitis was made and scrapings were obtained for microscopy and culture.

Results: The mean age was 7.31 ± 2.52 years for the subjects and 7.40 ± 2.43 years for the controls. The mean age of onset of T. capitis was 5.2 ± 2.039 years. The prevalence of T. capitis in Ilie Community was 43.5%. Contact with animals, soil and individuals with T. capitis were the prevalent risk factors for T. capitis. Large family size did contribute but not significantly to the spread of Tinea. Also, history of atopy did correlate but not significantly with disease chronicity. A total of 120 isolate representing 7 different dermatophytes including; T. metangrophytes (67.5%) as the leading organism were isolated. There was no case of mixed infection. Other isolates include T. tonsuran (13.3%), T. rubrum (10.8%), M. audouini (2.5%), M. gypseum (2.5%), T. violaceum (1.7%), T. soudanense (1.7%). Trichophyton metangrophytes was the most frequent organism causing the predominant non-inflammatory type of T. capitis (Gray patch and Black dot type) though not exclusively among children age group 5-8 years and 9-12 years, followed by T. tonsurans. A bold step must be taken to effectively reduce contact with the sources of infection.

Conclusion: Tinea capitis remains an endemic disease reaching variable epidemic proportion in some populations as seen in Ilie among the children. The non-inflammatory type (GPTC, BDTC) were the prevalent clinical types of T. capitis in Ilie.

INTRODUCTION

Ringworm which can also be simply referred to as Tinea, is an infectious fungal disease of the skin and its integuments.[1] It is one of the superficial infections of worldwide distribution but of extreme prevalence in the humid regions of the world. [1] Tinea capitis (T. capitis) is a public health problem in Nigeria though not a reportable disease. Like other infectious diseases, it has its heavy toll on children between ages of 4 and 14 years because of frequent body contact that occur among children. [2,3] Gugnani and Njoku-obi [3] had earlier attributed the public health problems caused by dermatophytosis in Nigeria to the warm humid climate, crowded living, poor
sanitary conditions of the majority of the populace, and same has been enhancing the spread of the disease. In the past decades, the disease remained endemic in Nigeria, largely because of lack of information on its prevalence and the absence of necessary control measures in place [4]. The ringworm of the head is a public place ill: in the schools and other public places where children are found and have continue to be an issue of concern among parents and teachers that cares. The epidemiology of T. capitis varies from place to place [5]. These variations range from changing prevalence, shifting agents, to seasonal escalations. Fathi et al attributed this variation to people's habits, lifestyle, and standards of hygiene, climate conditions and levels of education. [5]

Previous works on T. capitis in our environment placed emphasis on prevalence and implicated aetiological agents, clinical types were not properly defined. Therefore, findings from a community based study of T. capitis was aimed at bridging existing gaps, proffer solution to lingering wildfire and continuous threat of T. capitis among children and may serve as an incentive to a more comprehensive approach to the management of T. capitis and generate data for further studies.

PATIENTS AND METHODS

This study was conducted in Ilie community, a village in Olorunda Local government headquarters in Osogbo, the capital of Osun State, Nigeria. Ilie is located in the tropical rain forest belt of South-Western part of Nigeria and is about 500 kilometers from Abuja the capital city of Nigeria. Geographically Ilie lies approximately on latitude 40°N of equator and longitude 7.34°E of Greenwich meridian. Ilie is an organized community outreach centre of Ladoke Akintola University Teaching Hospital, where medical students undergo their community medicine posting. Vegetation in Ilie is a mixture of Savannah and semi-tropical forest. There are two distinct seasons; the wet and the dry seasons. The former occurs between April and October, while the later takes place from November to March.

Largely the community is an agrarian and fishing community, other occupations of the people include trading, cloth dying and wood carving. The community is an example of typical African setting and settlement; the dwellers have large and extended family. Children and adults are living together in compounds with household numbers varying from two to five per compound. Houses are built mostly with mud. Only very few houses were cemented and were built years past and presently were dilapidated and in a state of disrepute. Also, domestic animals and pets are kept in very close association with humans.

The people are mainly Yorubas, but other ethnic groups including Ibos, Hausas, and some minority group that work as manual labourers, serving the need of the community.

From population projection of 2006 national census, about 5500 people are currently living in Ilie. Population growth rate was put at 2.9% per annum. The estimated under 15 years (44% of Nigeria population) population was projected at 2,420 by year 2009.

The social amenities in the community include post office, police post, electricity, pipe borne water and a community health centre. Community dermatology services are grossly lacking. Only the parents that filled informed consent form had their children recruited into the study. A systematic community based epidemiological study of Tinea capitis infection has not been conducted in the area.

Study design: Cross-sectional study.

STUDY POPULATION: Three hundred and seventy children were recruited. One hundred and eighty five children with T. capitis and 185 sex matched children without any chronic or severe disease as control. The subjects and control group were recruited between August and December 2010.

Selection criteria for patients and controls: Children with symptoms or signs suggestive of T. capitis, resident of Ilie, whose parents filled the consent form and aged 5 to 16years with voluntary intention to participate in the study, were recruited. The inclusion criteria for contols were the same except that, they were without T. capitis.

Exclusion criteria for patients and controls: All non - Ilie’s residents and those whose parent’s refused to sign the informed consent .

Clinical Survey for Tinea capitis: The purpose and benefits of the study were explained to the parents and their children in the local Yoruba language. Survey for T. capitis was done in the evening when parents and children had returned back to their homes from school and farm.
Multistage sampling method, comprising of successive random sampling was employed. Sixteen communities were randomly selected from the thirty two communities in Ilie village. From the randomly selected community, compounds were randomly recruited into the study. Then from the selected compounds houses were recruited. Compound with two houses had one house randomly selected; two out of three, three out of four, four out of five houses, and five out of six houses were also selected randomly for inclusion into the study.

Children from selected houses had the survey questionnaire self administered by the investigators. The questionnaire focused on socio-demographic characteristics like age, sex, and child’s education level, parent’s occupation, number of children in the family and average monthly income, race and religion. Clinical history such as age at onset of *T. capitis*, duration of *T. capitis*, history of contact with animal, soil, individuals with *T. capitis*, and place of barbing were recorded. All the children in the selected houses had all areas of their scalp thoroughly examined for clinical types of *T. capitis* and lymphadenopathy. Diagnosis of clinical types was also made as seen among the children include; Scaly annular patch, inflammatory black dot pattern, Inflammatory *T. capitis* (kerion), Inflammatory *T. capitis* (Favus), and inflammatory pustular type.

**Sample Collection and laboratory processing:** The affected area of the head was cleaned with alcohol, hairs and scales were collected into dry, clean envelope for mycological examination using the technique described by Fathi et al. [3]. The hair scrapping was transported from the field in a dry clean envelope. Identification of all specimens taken from the scalp was done by direct microscopy with 10% KOH, The scrapings and the pieces of hair were plated out immediately as soon as investigator arrived from the field on daily basis separately on culture media. Slide culture technique was also used. The dermatophyte specific Potatoe agar was used. Each of the culture plates were incubated at 27°C for 4 weeks and then macro and micro morphological studies of cultured colonies was done for the presence of dermatophytes.

**STATISTICAL ANALYSIS:** Data was analyzed using Statistical Programme for Service Solution 16.0 (SPSS Chicago Inc., IL, and U.S.A.,) The socio-demographic variables of the patients were summarized using the Student’s t-test for numeric variables and Chi square tests for categorical variables. Clinical variables such as age at onset, duration of *T. capitis*, family and atopy history, close contact with soil, animals, and individual with *T. capitis*. Other clinical variables like alopecia, scaling, pruritus, and lymphadenopathy were also summarized using the Student’s t-test for numeric variables and Chi square tests for categorical variable as applicable. The pattern of animal contact was represented with bar chart.

**RESULTS**
The mean age was 7.31 ± 2.52 years for the children with *T. capitis* and 7.40 ± 2.43 years for the apparently normal children recruited into the study as controls. There was no significant difference between the age of both group statistically (t test = 0.74 df = 278, p = 0.67). The range and mode were 5-16 and 5 years respectively for both *T. capitis* group and control. (Table 1)

A total of 425 children were randomly recruited and examined before the sample size was completed. Overall prevalence of *T. capitis* in this population was 43.53%. Prevalence was highest among the age group 5-8 years ,139 (57.43%) followed by age group 9-12 years, 35 (42.12%) and least among age group 13-16 years, 89 (20.94%). *Tinea Capitis* was prevalent among the boys than the girls. The overall prevalence among the boys was 131 (30.82%), and among the girls was 54 (12.70%). The highest prevalence was recorded among boys 5-8 years, 91(37.6%), the least was among girls 9-12 years, 6 (6.38%). *T capitis* was not recorded among girls 13-16 years(Table 1).

Table (1) also shows the distribution of children per families’ of *T. capitis* and the control groups. Most of the subjects (T. capitis group) came from a larger family with many children than the control. The mean number of children per family for the subjects and controls were, 4.81 ± 2.012 and 4.63 ± 2.210 respectively. The most frequent number of children encountered in the study per family was 5 for the subjects, and was 4 for the controls. The number of children per family for the subjects ranged from 1 – 15, for the controls it ranged from 1-12. However, this difference was not statistically significant, (t = 0.787, df = 368, p = 0.155).

*T. capitis* was rare before the age of one year, only 4 (0.9%) of the subjects had the infection before one year. Most of the subjects had *T.*
capitis by the age of 6 year. The mean age of onset of T. capitis was 5.2 ± 2.039 years and the most frequent age of onset was 5 years. The duration of T capitis among children was 0.6±1.031 with a range of 0.1 – 13years. , 99(49.1%) children have had T. capitis for about one year. Also, 52 children (28.1%) had T.capitis for at least 2 years, while 14 (7.6%), 9 (4.9%), 11 (5.9%) and 4 (2.2%) had T. capitis for at least 3, 4, 5 and 6 years respectively. Only 4 (2.2%) of T. capitis population had the infection recurrently for more than 8 years.

There was no statistically significant difference between children with T. capitis and the control group in terms of educational attainment (Table 2). Majority of the pupils, T. capitis and control group were in the primary school, 105 (56.8%) vs 94 (50.8%) respectively. This was closely followed by children in pre-school age, 68 (36.8%) vs 74 (40.0%) respectively for both T. capitis and control group. Only a few of the children randomly recruited were in the secondary school at the time of survey, 11 (5.9%) vs 17 (9.2%) for T. capitis group and control group respectively( X² =75.474, df= 2, p = 0.000 ). Whereas, the entire pupils in the control group were enrolled in one form of formal education or the other, one child, (0.5%) of the Tinea group had no form of education. Statistically, there was no significant difference in the educational attainment of both groups. (X² = 3.147 df = 3, p = 0.344)( Table 2).

The two groups however differ significantly in terms of occupation and religion. While majority of the parents of children with T. capitis group were predominantly farmers and fisher men, the control group were largely civil servants and men that were self employed in trades such as road side mechanics, drivers, patent medicine store operators and the likes. Statistically this difference was found to be significant. (X² = 99.219, df= 2, p = 0.000).

From Table (2) , children with T. capitis were likely to have farmers and fishermen as parent, while the parents of the control population were likely to be civil servants and self employed individuals.

The Average income of the families of the subject and controls were evaluated. Most families in the T capitis group, 140 (76.1%) earn less than N5 000; 00($33.00) per month which is much below the Nigerian minimum wage, while income among the parents of the children in the control group spread across the entire income bracket. The average family income per month for the subjects was N8 260 ($32.00) ± 11 749($74.36), and controls was N16 880:00($106.83) ± 14 513:00($91.85). The range of income per month for the subjects and control group was N1, 000 – N 50 000 ($6.300 - 316.00) and N3 000 - N 60 000 ($18.98 - 379.74) respectively. The difference in the family incomes was found to be statistically significant. (t test = -6.269, df = 368, p = 0.000) (Table 2).

Assessment was made for the possible risk factors for Tinea among the children. Majority of the children with T. capitis had significant animal contact, 166 (89.7%) than the children in the control group, 105 (56.8%). The existed difference was statistically significant. (X² = 51.316 df =1, p = 0.000). Meanwhile, figure( 1) shows the pattern of animal contact among the children with T. capitis and controls. The predominant animal contacted was goat among both group; 125/185 and 74/185 respectively. Frequent contact with goat and other animals being kept for commercial purposes was highest among children with T. capitis than control group (Goat and sheep: 26/185 and 2/185; sheep alone 5/185 and 2/185 respectively).

Contact with pets like dog and cat were frequent among controls than the children with T. capitis (14/185 and 8/185 respectively).

Contact with soil was also more common among children with T. capitis than control 156 (84.3%) vs 14 (7.6%) respectively. The difference was statistically significant, (X² = 2.194, df =1, p = 0.000). Also frequent among children with T. capitis than the control was the positive history of previous contact with individual that had ringworm, 146 (78.9%) vs 38 (20.5%) respectively. The observation was also found to be significant, (X² = 1.261, df =1, p = 0.000). Household contact was significantly higher than contact in the classroom. (X² = 72.339, df=1, p = 0.000). One hundred and thirty five (73.0%) of the children with T. capitis use various available village barbers while 30 (16.2%), either weaved or barb their head at home. The difference found between self barbing and public barbing was statistically significant, (X² = 8.096, df =1, p = 0.017). (Table 3)

History suggestive of various atopic diathesis was also commoner among children with T. capitis than controls, 54 (29.45%) vs 4 (2.2%), statistically the difference was also significant. (X² = 51.116, df =1, p = 0.000). (Table 3) Rhinitis was the prevalent atopy suggestive
symptom 45/54 (83.3%), this was followed by Vernal conjunctivitis 4/54 (7.45%), Atopic dermatitis 3 (5.5%), and Asthma 2 (3.7%) (Table 3).

Family history of T. capitis in the parents of children with T. capitis infection and the control was present in 45/185 (24.3%) and 4/185 (2.2%) of the control. Statistically, the difference between the two groups was significant ($X^2 = 39.543$, df = 1, $p = 0.000$). However, there was an non-significant positive correlation between history of atopy and the duration of T. capitis in this study. ($r = 0.024$, $p > 0.05$) (Table 3)

The study examined the pattern of symptoms found among the recruited children, scalp scaling was the predominant symptom found among 179/185 (96.8%) more than as found in the controls, 12/185 (6.3%). The difference was found to be statistically significant ($X^2 = 346.754$, df = 1, $p = 0.000$) (Table 3). Scalp pruritus followed by scaling as the second most frequently encountered symptom in this study among children with T. capitis than among the controls and the existed difference in this pattern of presentation was found to be statistically significant, 161/185 (87.0%) vs 27/185 (14.6%), ($X^2 = 3.018$, df = 1, $p = 0.000$) (Table 3).

However, hair loss (alopecia) was present both as symptom and examination finding significantly among children with T. capitis than the controls, 139/185 (75.1%) vs 0 (0.0) ($X^2 = 222.641$, df = 1, $p = 0.000$). Alopecia was predominantly patchy and non - scarring 134 (96.4%) than patchy scarring form which was present in 5 (3.6%) (Table 3).

Adenopathy was also significantly present among children with T. capitis than control 47 /185 (25.4%) vs 5/185 (2.7%), ($X^2 = 41.978$, df = 1, $p = 0.000$). Fourty four (23.78%) children had lymphadenopathy at the posterior cervical area and 3/47 (6.38%) had it in the post auricular area (Table 3).

The various clinical types of T. capitis were examined for; the non-inflammatory form of T. capitis was prevalent: “Gray patch” Tinea capitis (GPTC) 86/185 (46.5%), “Black dot” Tinea capitis (BDTC) 78(42.2), Seborrheic dermatitis type Tinea capitis (SDTC) 18 (9.7%), and the pustular inflammatory type 3 (2.2%). No case of Kerion or Favus was recorded (Fig 2,3,4&5).

Attempt was made to correlate clinical type of T. capitis with species of organism responsible. T. metangrophyte was responsible for non-inflammatory T. capitis: BDTC and GPTC and SDTC. There was no clear cut correlation between dermatophytes and clinical types ($r = 0.025$, $p = 0.567$).

The scaling and hairs from children’s scalp were obtained for microscopy and culture. Scrapping was only possible and obtainable from 179 children. One hundred and sixty two samples were positive for fungal element microscopically while 17 samples showed no growth. Culture confirmed growth of dermatophytes only in 120 samples. Isolated dermatophytes represented 2 genera; Trichophyton and Microsporon, and 8 different dermatophytes including Trichophyton metangrophytes as the leading organism isolated. There was no case of mixed infection. Other isolates of this study include T. tonsuran, T. rubrum, T. violaceum, T. soudanence, Microsporon audounii Microsporum gypseum. Trichophyton metangrophytes was the most frequent organism causing infection in children, followed by T. tonsuran as shown in Table (4).
Table (1): General Characteristics of Children with *Tinea capitis* and the Controls

<table>
<thead>
<tr>
<th>Variables</th>
<th>T. capitis group</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age of participants(years)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>7.31 ± 2.52</td>
<td>7.40 ± 2.43</td>
</tr>
<tr>
<td>Mode</td>
<td>5</td>
<td>5</td>
</tr>
<tr>
<td>Range</td>
<td>5-16</td>
<td>5-16</td>
</tr>
<tr>
<td>Number of children per family</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>4.81 ± 2.012</td>
<td>4.63 ± 2.210</td>
</tr>
<tr>
<td>Mode</td>
<td>5</td>
<td>4</td>
</tr>
<tr>
<td>Range</td>
<td>1-15</td>
<td>1-12</td>
</tr>
<tr>
<td>Mean age at onset of <em>T. capitis</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>5.2 ± 2.039</td>
<td></td>
</tr>
<tr>
<td>Mode</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>1 month – 11 years</td>
<td></td>
</tr>
<tr>
<td>Duration of <em>Tinea capitis</em></td>
<td>0.6 ±1.031</td>
<td>1</td>
</tr>
<tr>
<td>Mode</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>1 month - 13 years</td>
<td></td>
</tr>
</tbody>
</table>

Age and Sex Prevalence of *T. capitis*

<table>
<thead>
<tr>
<th>Age in years</th>
<th>Number examined</th>
<th>Number infected(prevalence)</th>
<th>Total(prevalence)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td>Total (%)</td>
</tr>
<tr>
<td>5 – 8</td>
<td>242</td>
<td>91(37.60)</td>
<td>139 (57.43)</td>
</tr>
<tr>
<td>9 – 12</td>
<td>94</td>
<td>29(30.85)</td>
<td>35 (37.23)</td>
</tr>
<tr>
<td>13 – 16</td>
<td>89</td>
<td>11(12.35)</td>
<td>11 (12.35)</td>
</tr>
<tr>
<td>Total</td>
<td>425</td>
<td>131(30.82)</td>
<td>185 (43.53)</td>
</tr>
</tbody>
</table>

Table (2): Educational attainment, occupation and religion, ethnicity, and average family income per month distribution of patients and controls

<table>
<thead>
<tr>
<th>Educational attainment*</th>
<th><em>Tinea capitis</em> group (N, %)</th>
<th>Control group (N, %)</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>1 (0.5)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Pre-school</td>
<td>68 (36.8)</td>
<td>74 (40.0)</td>
</tr>
<tr>
<td>Primary</td>
<td>105 (56.8)</td>
<td>94 (50.8)</td>
</tr>
<tr>
<td>Secondary</td>
<td>11 (5.9)</td>
<td>17 (9.2)</td>
</tr>
<tr>
<td>Occupation**</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Predominantly farming</td>
<td>130 (70.3)</td>
<td>7(3.8)</td>
</tr>
<tr>
<td>Predominantly fishing</td>
<td>14 (7.6)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Other forms of self employment</td>
<td>19 (10.3)</td>
<td>104 (56.2)</td>
</tr>
<tr>
<td>Unemployed</td>
<td>0 (0.0)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Civil servants</td>
<td>22 (11.9)</td>
<td>74 (40.0)</td>
</tr>
<tr>
<td>Religion***</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Christianity</td>
<td>28 (15.1)</td>
<td>102 (55.1)</td>
</tr>
<tr>
<td>Islam</td>
<td>157 (84.9)</td>
<td>77 (41.6)</td>
</tr>
<tr>
<td>Traditional</td>
<td>0 (0.0)</td>
<td>6 (3.2)</td>
</tr>
<tr>
<td>Ethnicity****</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yoruba</td>
<td>183(98.9)</td>
<td>176 (95.1)</td>
</tr>
<tr>
<td>Hausa</td>
<td>0 (0.0)</td>
<td>6 (3.2)</td>
</tr>
<tr>
<td>Igbo</td>
<td>0 (0.0)</td>
<td>3 (1.6)</td>
</tr>
<tr>
<td>Others</td>
<td>2 (1.1)</td>
<td>0 (0.0)</td>
</tr>
<tr>
<td>Average income per month(Naira)*****</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 – 5000</td>
<td>140 (76.1)</td>
<td>56 (30.3)</td>
</tr>
<tr>
<td>5,001 - 10,000</td>
<td>17 (9.2)</td>
<td>31 (16.8)</td>
</tr>
<tr>
<td>10,001 - 20,000</td>
<td>13 (7.1)</td>
<td>52 (28.1)</td>
</tr>
<tr>
<td>20,001 - 50,000</td>
<td>14 (7.6)</td>
<td>43(23.2)</td>
</tr>
<tr>
<td>&gt; 50,000</td>
<td>0(0.0)</td>
<td>3 (1.6)</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>8,260 ± 11,749</td>
<td>16,880 ± 14,513</td>
</tr>
<tr>
<td>Range</td>
<td>1,000,000 - 50,000</td>
<td>3,000 – 60,000</td>
</tr>
</tbody>
</table>

*X²=3.14*X²=3.147, df= 3 p = 0.344 (N = 185),  **X²=3.147, df= 3, p = 0.369 (N = 185),
***X²=75.474, df= 2, p = 0.000 (N = 185), **** X² = 5.136 df =2, p = 0.048,
***** t- test = -6.269, df = 368, p = 0.000

Akinboro et al., Afro-Egypt J Infect Endem Dis 2011; 1 (2):53-64
www.mis.zu.edu.eg/ajied/home.aspx
Table (3): Comparison of some risk factors, clinical history and examination findings: children with Tinea. capitis and controls.

<table>
<thead>
<tr>
<th>Clinical variables</th>
<th>Tinea capitis group N (%)</th>
<th>Control group (N %)</th>
<th>X²</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td>Animal contact</td>
<td>166 (89.7)</td>
<td>19 (10.3)</td>
<td></td>
</tr>
<tr>
<td>Soil contact</td>
<td>156 (84.3)</td>
<td>29 (15.7)</td>
<td></td>
</tr>
<tr>
<td>Human contact</td>
<td>146 (78.9)</td>
<td>39 (21.1)</td>
<td></td>
</tr>
<tr>
<td>Atopy History</td>
<td>54 (29.8)</td>
<td>131 (68.5)</td>
<td></td>
</tr>
<tr>
<td>Family history</td>
<td>45 (24.3)</td>
<td>140 (75.7)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>51.316*</td>
</tr>
<tr>
<td>Clinical History</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Scalp pruritus</td>
<td>161 (87.0)</td>
<td>24 (13.0)</td>
<td></td>
</tr>
<tr>
<td>Scalp scaling</td>
<td>179 (96.8)</td>
<td>6 (3.2)</td>
<td></td>
</tr>
<tr>
<td>Adenopathy</td>
<td>47 (25.4)</td>
<td>138 (74.6)</td>
<td></td>
</tr>
<tr>
<td>Alopecia</td>
<td>139 (75.1)</td>
<td>46 (24.9)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1.942*</td>
</tr>
<tr>
<td>Type of Atopy</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rhinitis</td>
<td>45(83.3)</td>
<td>2(3.7)</td>
<td></td>
</tr>
<tr>
<td>Asthma</td>
<td>2(3.7)</td>
<td>0(0.0)</td>
<td></td>
</tr>
<tr>
<td>Dermatitis</td>
<td>3(5.5)</td>
<td>0(0.0)</td>
<td></td>
</tr>
<tr>
<td>Conjunctivity</td>
<td>4(7.4)</td>
<td>0(0.0)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2.331</td>
</tr>
<tr>
<td>Contacts</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Family member</td>
<td>135(73.0)</td>
<td>14(7.6)</td>
<td></td>
</tr>
<tr>
<td>Class member</td>
<td>14(16.7)</td>
<td>27(14.6)</td>
<td></td>
</tr>
<tr>
<td>Barbing saloon</td>
<td>155 (83.3)</td>
<td>30 (16.2)</td>
<td></td>
</tr>
<tr>
<td>Home Barbing</td>
<td>134 (16.7)</td>
<td>49 (26.5)</td>
<td></td>
</tr>
<tr>
<td>Weaving</td>
<td>0 (0)</td>
<td>2 (0.1)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>72.339*</td>
</tr>
</tbody>
</table>

*df =1, p < 0.001 N = number

Table (4): Distribution of dermatophytes isolated according to sex among the children in Ilie Community

<table>
<thead>
<tr>
<th>Species</th>
<th>Total (%)</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trichophyton mentagrophytes</td>
<td>81(67.5)</td>
<td>67</td>
<td>14</td>
</tr>
<tr>
<td>Trichophyton tonsuran</td>
<td>16(13.3)</td>
<td>10</td>
<td>6</td>
</tr>
<tr>
<td>Trichophyton rubrum</td>
<td>13(10.8)</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>Microsporum audouinii</td>
<td>3 (2.5)</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Microsporum gypseum</td>
<td>3 (2.5)</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Trichophyton violaceum</td>
<td>2(1.7)</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Trichophyton soudanense</td>
<td>2 (1.7)</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>120 (100)</td>
<td>95</td>
<td>25 (20.83)</td>
</tr>
</tbody>
</table>

Figure (1): shows the pattern of animal contact: patients and controls

X² = 51.316, df= 1, p < 0.001
Figure (2): Annular Patch *Tinea capitis*

Figure (3): Infected Annular Patch *Tinea capitis*

Figure (4): A child with *Tinea capitis* and posterior cervical lymphadenopathy.

Figure (5): Black dot type *Tinea capitis*

Figure (6): Dilapidated building, uncemented floor and dermatophyte infected animals living in close contact with humans
DISCUSSION

The overall prevalence of *T. capitis* in this study was 43.5%. This prevalence rate is widely in excess of previously recorded prevalence of 4% to 30% described among school children in the Western and Southern part of Africa. [6, 7] Soyinka [8] recorded a higher prevalence of 55% in a population based study and they thought there was an epidemic of *T. capitis* among school pupils. *Tinea Capitis* has been known to reach epidemic proportions among school children. The observed prevalence in this study is higher than 14.05% recorded by Ajao and Akintunde in Ile-Ife [9] and similarly prevalence was highest among boys (30.82%) compared to girls (12.7%).

In the past decades, several studies have concluded that *T. capitis* is an important dermatologic condition widely distributed throughout the world, more importantly among children. Its frequency is increasing, and aetiological agents vary from one geographic location to another. Several other Nigerian [3, 4, 8, 10, 11, 12, 13] and international studies [14, 15] had documented differences in occupation, parent income as a significant risk factors for *T. capitis* as shown in this study. Parents of children with *T. capitis* in this study were more likely to be predominantly farmers and fishermen, while those of controls were predominantly office workers or self employed in other forms of trade. This is in line with epidemiological reasoning that, low income earning or poverty, malnutrition and general poor social conditions experienced by subsistent farmers, coupled with unlimited exposure to potential sources of infections such as contaminated soil and animals highly predisposed children in Ilie to *T. capitis*. The strong link between animals and dermatophyte had been previously documented by Abdulkadir [16], Ameh and Okolo [17] in the Northern states of Nigeria.

The study also found that children in Ilie community live in close contact with animals (predominantly goat, sheep and dogs) which were being kept for business, family food supply and hunting purposes. Abdulkadir [16] had earlier confirmed the enzootic ringworm of horses, dogs, and livestock as common source of sporadic infection among owners or their care takers which might include the owner’s children. Animal type ringworm was also viewed by Macura as an occupational hazard for farmers and pet keepers [18]. Study from Sokoto state of Nigeria where livestock and pet domestication was found as a common practice in households, had suggested domestic animals as important reservoir of *tinea*. Direct or indirect contact with fungus contaminated objects of livestock rearing like dung, fencing, halters, rope, harness and grooming brushes were found as extremely important in the natural dissemination of the disease.[17]

Other authors in the region had observed that *Tinea* transmission was encouraged by poor living unsanitary condition with overcrowding [3]. Soil in the homes of livestock keepers and playground has been viewed by several authors as containing fungal element that dropped from the body of infected animals or primarily a geophillic agent [19]. The high frequency of *T. capitis* among children had therefore been linked to intense close contact among children especially at play grounds and at home. This study showed that contact were more intense at home amongst family members and neighbors than in the classroom, this is understandable because pupils spend few hours in the school, and most hours are spent at home and neighborhood play grounds.

In addition, most children with *T. capitis* came from large families (> 4 children), this seems not significant enough as a sole factor that could risk the children and sustain the infection in the community. This observation is similar to the former findings of Ajao et al. in Ile – Ife. [9]

Like other previous studies [3,4,8,10,11,12,13], boys were predominantly infected than girls, with a ratio of 2.4:1 which was greater than 1.8:1 recorded by Kalla et al.[20] in India but lower than a high ratio 5:1 recorded by Gugnani and Njoku-Obi in South-eastern Nigeria.[3] The mean age of infection with dermatophyte was 7.31±2.52 years, which is closer to the observation of other workers [8,21]. The most frequent age group affected was 5–8 years with most likely age of infection being 5 years. Gugnani and Njoku-Obi recorded age group 3-7 years in their study [3]. Ayambimpe et al [21] also documented the highest rate of infection in the age bracket 10–14 years; this was closely followed by similar age grouped 5–8 years which was observed in this study. *Tinea capitis* was not seen among girls older than 13 year. Soyinka and others had adduced this high prevalence of *T. capitis* among the boys to continuous and sustained exposure to infective agents and close body contact at the play ground, and the fact that most boys visit the same set of barbers that
harbor infective agents on their barbing equipment [8,12,21]. Furthermore, most of the girls older than 13 years may prefer to weave their hair rather than visiting barber’s shop, they practice better hair and general hygiene and may carry their own weaving equipment and thus reduce contact with infective agents on the hairdresser’s hand [9]. Encouragement of personal hygiene has therefore become important in the prevention and control of this endemic disease.

Many of the children infected with *T. capitis* barb in public saloon in this study. Barbing saloon in Ille are untidy and the practice of equipment sterilization was foreign to the operators of the four barbing saloon serving the village and the environment. David et al. [12] also demonstrated evidence of fungal element in barbing equipment in a recent study in Mubi, Adamawa state of Nigeria and suggested sensitization of public health workers and saloon customers on the need for sterilization of barbing equipment in all saloon. Ayanbimpe et al.[21] also attributed high incidence of *T. capitis* to continuous contact in barber’s shop. With continuous and intense transmission of *T. capitis*, unless a bold step is taken to control the infection at the barber’s shop, complete eradication of the condition might be a mirage.

This study demonstrated a non-significant positive correlation between duration of *T. capitis* and history of atopy. The commonest documented atopy condition was rhinitis. Hay and Shennan [22] and other workers [23] had previously reported association between the presence of atopy and chronic dermatophytosis, asthma and hay fever was the commonest atopic disease reported in their study. Other associations of chronic dermatophytosis include immediate- type hypersensitivity and elevated IgE levels which were not examined in this study.

In respect to the spectrum of isolated species of dermatophyte, it has been established that organism varies from one geographical location to another, even within a country, state, or local government area the patterns of isolates have varied overtime. In this study, the isolated dermatophytes were zoophylic, anthropophilic or geophilic. The organisms belonged to two general *Trichophyton* and *Microsporon*, and seven species which include *T. metagrophyte*, *T.tonsurans*, *T. rubrum*, *M. audouinii*, *M. gypseum*, *T. violaceum*, *T. soudanens* were isolated. The high prevalence of *T. metagrophyte* (67.5%) in this study was similar to a recent finding by Nweke [13] in Anambra among nomadic herdsman living in camps. Jain et al. in Rajasthan district of India also found *T. metagrophyte* as the second leading agent in their study[24]. Jha et al. in eastern Nepal also found similar pattern [25]. This finding is not surprising because 166 (89.7%) and 156 (84.3%) of the children recruited into the study had history of unlimited contact with animals mainly goats and dogs. These animals, mainly live stocks have been kept for commercial purposes and almost all the households have their share in the livestock rearing. The animals live in close contact with humans, direct contact is possible because animals are not been kept in special pen, and the children also participate in the rearing of the animals. Animals have been variously implicated in *Tinea* transmission. This profile of isolate in this study was similar to findings of Ajao and Akintunde [9] but Microsporum audouinii was the leading agent in their study.

The finding of *T. tonsurans*, *T. rubrum*, *T. soudanense*, *T. violaceus*, *M gypseum* is not strange in this environment. They have been isolated in various studies in Nigeria in the past as aetiological agent of *T. capitis*. [3, 4, 8, 10, 11, 12, 13]

It is worthy of note also, to comment on the high burden 42/179 (23.5%) of other non-dermatophyte fungi infection of the children’s scalp. Isolated in the study include: Penicillium spp, Blastomyces dermatidis, Candida albicans, and Gliocladium spp. This is also not unusual because the children live in close contact with soil both at home and in the school. Oyeka and Ugwu had also noted that this non – dermatophyte spores are ubiquitous and may transiently colonized human skin [26]. The finding of this organism should be taken serious because isolates of non – dermatophyte was not found mixed with other dermatophyte organism, which will portend their isolation as been contaminants. More so, dermatophyte specific culture media (Potatoe agar) was used.

In this current study, the non- inflammatory *T. capitis* (GPTC; 46.5%, BDTC; 42.2%, and SDTC; 9.7%), were more common than the inflammatory type (Pustular; 2.2%). Previous findings also corroborated the fact that, gray patch *T. capitis* (GPTC) or black dot (BDTC) type are likely to be the leading clinical type in any epidemiologic survey [20, 27]. The incidence of inflammatory type (2.27%) is lower when compared with the findings of other authors [27,28]. Mixed morphology of *T.capitis*
was rarely reported in literature [28]. There was no case of mixed morphology in this study. Several other studies confirmed the non inflammatory (GPTC or BDTC) to be the leading clinical types, and in some studies both almost occurred at the same proportion or frequency [20, 25, 27, 29]. The inflammatory types of T. capitis are uncommon in the present study. Nnoruka et al. [30] in Enugu state of Nigeria found a low incidence of kerion (9.3.1%) in their study, while Grover et al. [27] in India found a higher incidence of inflammatory T. capitis at 32%.

According to Gugnani and Najoku-Obi [3] the clinical appearance of T.capitis is most variable, and depends on the type of hair invasion, the level of host resistance and the degree of inflammatory host response. The ectothrix agent (T. metagrophyte) was prevalent in this study as a cause of the predominant non – inflammatory type, though not mutually exclusive for these clinical types alone in the study. The isolates of this study were mainly Trichophyton species that characteristically invaded the hair while producing large-spored ectothrix in chains as seen in T. mentagrophytes or the endothrix type as documented for T. tonsurans, T. soudanense, T. violaceum, T. yaoundei, T. gourvilii and rarely same may be demonstrated by T. rubrum. However, previous studies have shown that clinical presentation is not correctly indicative of the type of fungus or vice versa, as it also depends on other unknown factors [25,29], but Grover et al. in their study found endothrix agents to be responsible for BDTC and ectothrix agent to be responsible for GPTC, but the finding was not mutually exclusive also in their study. [27]

CONCLUSIONS

Tinea capitis remains an endemic disease reaching variable epidemic proportion in some populations as seen in Ilie among the children. The non – inflammatory type (GPTC, BDTC) were the prevalent clinical types of T. capitis in Ilie. Close animal and soil contact, poor sanitary condition at home, extreme of poverty were the most potent risk factors for contracting and sustaining the epidemic in this population. The cheapest means of prevention and controlling this infection depends on education of parent and sibling on reducing or preventing undue contact with the source of contagion.

RECOMMENDATIONS

The following are recommended:

1) Health education about infectious disease such T. capitis: including modes of transmission to the parents and children, and the importance of personal hygiene.

2) There is need for the establishment of community dermatological services to cater for the teeming needs of the children.

3) Since livestock keeping is a lifestyle of the Ilie people, I therefore suggest the need for construction of animal’s pen by each household to reduce in-house contact with animals. This can be shouldered by the local health authority.

4) There is need for effective community veterinary services for prompt treatment of infected animals.

5) Study of dermatophyte infections among animals in Ilie and its correlation with human dermatophytes is suggested as a future study.

Funding: This work was not funded by any agency.

Conflicts of interest: There are no conflicts of interest in the course of this research work.

Ethical approval: Ethical clearance was obtained from the ethical committee of Ladoke Akintola University Teaching hospital, Osogbo. Informed consents were obtained from parents of the children.

REFERENCES


Video Case : Extraction of a Coin from the Stomach of a 6 Months Infant

Zaher T.
Endemic and Tropical Medicine Department, Faculty of Medicine, Zagazig University, Egypt.
tareqzaher@zu.edu.eg

Comment
An Egyptian coin was accidentally ingested by a 6 months female infant. Repeated X rays over 2 weeks revealed the coin in the upper abdomen. Upper gastrointestinal endoscopy using Olympus GIF Q160 endoscope was performed. 1mg midazolam was administered intramuscularly. Endoscopy revealed impacted coin in the antrum of the stomach. Repeated attempts using shark tooth forceps were done. Finally the coin was successfully extracted.
An esophageal polyp is a type of abnormal growth that protrudes from the lining of the esophagus. Esophageal polyps are rather rare in the general population, and in a broad-spectrum autopsy study were identified with a frequency of only 0.5%. Additionally, these growths are more common in men than women, and generally do not occur until middle age [1]. When esophageal polyps do result, they are most often asymptomatic and benign. However, it may present with bleeding, dysphagia, chest pain and sometimes respiratory manifestations including aspirations and even asphyxia especially with fibrovascular polyps [2]. Different types of polyps were described in the esophagus; these includes, hyperplastic polyps, inflammatory fibroid polyps, squamous cell carcinoma, adenocarcinoma, gastrointestinal stromal tumors (GIST), fibrovascular polyps [2] and hamartomas e.g. Cowden's disease [3].

Squamous cell papilloma is usually considered benign, and does not usually correlate with the development of squamous cell carcinoma. Squamous cell carcinoma usually develop at several areas in the esophagus simultaneously, and form “frond-like” protrusions within the esophageal lumen [4].

Adenocarcinoma of the esophagus may develop following long history of Barrett’s metaplasia of the lower esophagus following long history of GERD, or may develop in ectopic gastric mucosa [4].

The fibrovascular polyp grows into a large stalk form, up to 50 cm long, and is constructed of loose fibrous connective tissue, fat, and blood vessels, covered by a layer of epithelium cells. These polyps usually develop in the upper third of the esophagus. In one study, 87% of patients with fibrovascular polyps reported dysphagia, 25% had respiratory problems, and 12% had experienced partial regurgitation of the top of the stalk into their mouth or throat. This type of esophageal polyps is also benign, but is very, very rare. [5]

Diagnosis of esophageal polyps usually occurs via barium swallow, endoscopy, or CT. Usually small polyps are removed endoscopically while those causing any sort of interference with a patient’s health are surgically removed. [1]

An esophageal polyp is a mucosal growth, when compared with esophageal ploypoidal masses that includes mucosal and submucosal tissues and sometimes muscle layer, it is much more benign and small in size and may not necessitates treatment, while masses are usually large, symptomatic and usually needs treatment.

We reported a 66 years old male patient with history of coronary artery bypass surgery one year ago, he recently developed progressive dysphagia with unsatisfactory response to multiple courses of proton pump inhibitors and prokinetics. When a diagnostic upper endoscopy was performed, large polyoid polyps mass was seen protruding within the esophageal lumen just above the cardia and a large ulcerated necrotic friable mass also seen in the fundus of the stomach both masses were biopsied and histopathological analysis revealed gastric adenocarcinoma in both lesions.
REFERENCES


Case records of Endemic and Tropical Medicine Department, Zagazig University Hospitals, Zagazig, Egypt

Case 1-2011: A 60 years Old Male with Coma and Fever with Recent Travel to South Sudan

Tarik Zaher, Nahla Elgammal, Dina Mohamed
tareqzaher@zu.edu.eg

Presentation of the case:

A 60 years –old business man admitted to the intensive care unit of the Endemic and Tropical Medicine Department, Faculty of Medicine, Zagazig University, Egypt because of deep coma, fever and tachypnea.

The patient had history of recent travel to Juba, south of Sudan 2 weeks before admission. 5 days after return from Sudan, he noticed fatigue and mild fever. He was given non-specific treatment. 5 days later jaundice appeared on his skin, he was admitted to private hospital in Zagazig. The investigation showed total bilirubin :10 mg/dl , direct 7 mg/dl , ALT:150 , AST :120,Hg :10 gm/dl ,platelets :110000 / dl .The patient was managed as having acute liver disease. Later on malaria parasite test (MP test) was done and revealed P. falciparum in thick and thin blood films. The patient was referred to Zagazig Fever Hospital with deterioration of conscious level, quinine was given intravenously without improvement. In the next day, the patient was referred to Tropical Medicine Intensive Care Unit. The patient was deeply comatose, deeply jaundiced and pale, splenomegaly was found, bubbling chest crepitations were auscultated. The urine was black and the skin showed echymosis. The investigations showed total bilirubin :32 mg /dl, direct bilirubin 22 mg/dl, Hg :6 gm /dl ,platelets : 15000/dl, creatinin: 6 mg/dl , INR :7 ,PH: 7.31 , bicarbonate :12 mmol/l and glucose : 350 mg/dl. Hemoglobin was found in urine. Quinine was given by intravenous infusion in the dose of 20mg /kg loading dose then 10mg/ kg every 8 hours. Doxycyclin 100 mg /12 hours was given through the Ryle. Intravenous frusemide was given as well as oxygen inhalation as a measure against pulmonary edema, also chest consultation for the possibility of mechanical ventilation was requested. Transfusion of platelets, fresh frozen plasma, and backed red blood cells were given. Intravenous fluids as glucose 10% with 15 unit regular insulin and Ringer lactate solution were given according to the CVP. Regular insulin according to blood glucose level was given every 6 hours subcutaneously. The patient showed no response after one day of extensive care and death was the end due to multiple organ failure.

Differential diagnosis:

Febrile coma: Febrile coma occurs in cerebral malaria, meningitis, encephalitis, heat stroke, cerebral and pontine hemorrhage, hepatic coma, diabetic coma with infection and atropine poisoning[1].

Fever with jaundice: Fever accompanied by jaundice is caused by viral hepatitis, falciparum malaria, paratyphoid B, infectious mononucleosis, Weil’s disease, hemolytic crises, septic cholangitis, acute leukaemia, yellow fever and other viral hemorrhagic fevers as rift valley fever [1].

Discussion:

The above case is a case of severe malaria according to WHO definition of severe malaria[2] due to presence of coma, renal failure, pulmonary edema, high INR, hemoglobinuria and acidemia.
Table 1 -- 1990 WHO Definition of severe malaria[2]

1. Cerebral malaria – unrousable coma not attributable to any other cause in a patient with falciparum malaria. The coma should persist for at least 30 min (1 h in the 2000 definition) after a generalized convulsion to make the distinction from transient postictal coma. Coma should be assessed using the Blantyre coma scale in children or the Glasgow coma scale in adults.

2. Severe anaemia – normocytic anaemia with haematocrit <15% or haemoglobin <5 g/dL in the presence of parasitaemia more than 10 000/μL. Note that finger prick samples may underestimate the haemoglobin concentration by up to 1 g if the finger is squeezed. If anaemia is hypochromic and/or microcytic, iron deficiency and thalassaemia/haemoglobinopathy must be excluded. (These criteria are rather generous; and would include many children in high transmission areas. A parasitaemia of >100 000/μL might be a more appropriate threshold.)

3. Renal failure – defined as a urine output of <400 mL in 24 h in adults, or 12 mL/kg in 24 h in children, failing to improve after rehydration, and a serum creatinine of more than 265 μmol/L (>3.0 mg/dL). (In practice for initial assessment, the serum creatinine alone is used.)

4. Pulmonary oedema or adult respiratory distress syndrome.

5. Hypoglycaemia – defined as a whole blood glucose concentration of less than 2.2 mmol/L (40 mg/dL).

6. Circulatory collapse or shock – hypotension (systolic blood pressure <50 mmHg in children aged 1–5 years or <70 mmHg in adults), with cold clammy skin or core-skin temperature difference >10°C. (The more recent review declined to give precise definitions, but noted the lack of sensitivity or specificity of core-peripheral measurements.) Capillary refill time is not mentioned but recent studies indicate this simple test provides a good assessment of severity.

7. Spontaneous bleeding from gums, nose, gastrointestinal tract, etc. and/or substantial laboratory evidence of DIC. (This is relatively unusual.)

8. Repeated generalized convulsions – more than two observed within 24 h despite cooling. (In young children, these may be febrile convulsions, and the other clinical and parasitological features need to be taken into account.)

9. Acidaemia – defined as an arterial or capillary pH <7.35 (note temperature corrections are needed as most patients are hotter than 37°C; add 0.0147 pH unit per degree Celsius (°C) over 37°C), or acidosis defined as a plasma bicarbonate concentration <15 mmol/L or a base excess >10. (Operationally, the clinical presentation of ‘respiratory distress’ or ‘acidotic breathing’ is focused upon in the 2000 recommendations. Abnormal breathing patterns are a sign of severity indicating severe acidosis, pulmonary oedema or pneumonia.)

10. Macroscopic hemoglobinuria – if definitely associated with acute malaria infection and not merely the result of oxidant antimalarial drugs in patients with erythrocyte enzyme defects such as G6PD deficiency. (This is difficult to ascertain in practice: if the G6PD status is checked following massive haemolysis, the value in the remaining

11. Postmortem confirmation of diagnosis. In fatal cases a diagnosis of severe falciparum malaria can be confirmed by histological examination of a postmortem needle necroscopy of the brain. The characteristic features, found especially in cerebral grey matter, are venules/capillaries packed with erythrocytes containing mature trophozoites and schizonts of P. falciparum. (These features may not be present in patients who die several days after the start of treatment, although there is usually some residual pigment in the cerebral vessels.)

The 2000 recommendations also include the following:

12. Impairment of consciousness less marked than unrousable coma. (Any impairment of consciousness must be treated seriously). (Assessment using the Glasgow Coma Scale is straightforward, but the Blantyre Scale needs careful local standardization particularly in younger children.)

13. Prostration: Inability to sit unassisted in a child who is normally able to do so. In a child not old enough to sit, this is defined as an inability to feed. This definition is based on examination not history.

14. Hyperparasitaemia – the relation of parasitaemia to severity of illness is different in different populations and age groups, but in general very high parasite densities are associated with increased risk of severe disease, e.g. >4% parasitaemia is dangerous in non-immunes, but may be well tolerated in semi-immune children. In non-immune children studied in Thailand a parasitaemia ≥4% carried a 3% mortality (30 times higher than in all uncomplicated malaria) but in areas of high transmission values much higher may be tolerated well. Many use a threshold definition of 10% parasitaemia in higher transmission settings.

The followings were not considered criteria of severe malaria:

1. Jaundice – detected clinically or defined by a serum bilirubin concentration >50 μmol/L (3.0 mg/dL). This is only a marker of severe malaria when combined with evidence of other vital organ dysfunction such as coma or renal failure.

2. Hyperpyrexia – a rectal temperature above 40°C in adults and children is no longer considered a sign of severity.

The above case was treated by quinine infusion with doxycyclin by the Ryle as well as by supportive measures for severe malaria.

Treatment of Severe P. falciparum Malaria[3]:

Specific antimalarial treatment:

Regimen 1:

1st drug

Artesunate 2.4 mg/kg iv or im on admission; then at 12 h and 24 h, then once a day for at least
24 hours, followed by full course of ACT (artemisinin combined therapy), and

2nd drug

Doxycycline 100mgs BID (2.2mg/kg BID for <45kgs) for 7 days OR Clindamycin 20mg base/kg/day divided in three doses for 7 days in pregnancy OR Malaron 4 tab daily for 3 days OR Mefloquine 4 tab in 1st day, 2 tab in 2nd day.

Regimen 2:

1st drug

Artemether 3.2 mg/kg i.m. given on admission then 1.6 mg/kg per day for at least 24 hours, followed by full course of ACT, and

2nd drug

As above.

Regimen 3:

1st drug

Quinine 20 mg salt/kg on admission (iv infusion or divided im injection), then 10 mg/kg every 8 h; infusion rate should not exceed 5 mg salt/kg per hour; course for 3 days for malaria acquired in Africa and South America, 7 days for malaria acquired in South east (SE) Asia, and

2nd drug

Doxycycline and clindamycin as above. Do not use mefloquine in combination with quinine.

Regimen 4:

1st drug

Quinidine gluconate 10 mg salt/kg (equivalent to 6.2 mg base/kg) iv infused over 12 hours, followed immediately by 0.02 mg/kg/min salt (equivalent to 0.0125 mg/kg/min base) continuous iv infusion; course for 3 days for malaria acquired in Africa and South America, 7 days for malaria acquired in SE Asia, and

2nd drug

Doxycycline or clindamycin, do not use mefloquine.

ALWAYS AVOID THE FOLLOWING COMBINATIONS: QUININE, MEFLOQUINE, PRIMAQUINE, CHLOROQUINE WITH EACH OTHER.

Adjunctant Treatment in Severe P. falciparum:

Table -2 Adjunctant treatment [3]:

<table>
<thead>
<tr>
<th>Manifestation/complication</th>
<th>Immediate management (in addition to antimalarial treatment)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperpyrexia</td>
<td>Administer tepid sponging, fanning, cooling blanket and antipyretic drugs</td>
</tr>
<tr>
<td>Coma (cerebral malaria)</td>
<td>Maintain airway, place patient on his or her side, exclude other treatable causes of coma (e.g. hypoglycaemia, bacterial meningitis); avoid harmful ancillary treatment such as corticosteroids, heparin and adrenaline; intubate if necessary</td>
</tr>
<tr>
<td>Convulsions</td>
<td>Maintain airways; treat promptly with intravenous or rectal diazepam or intramuscular paraldehyde</td>
</tr>
<tr>
<td>Hypoglycaemia (blood glucose concentration of &lt;2.2 mmol/l; &lt;40 mg/100ml)</td>
<td>Check blood glucose, correct hypoglycaemia and maintain with glucose-containing infusion</td>
</tr>
<tr>
<td>Severe anaemia (haemoglobin &lt;5 g/100ml or packed cell volume &lt;15%)</td>
<td>Transfuse with screened fresh whole blood</td>
</tr>
<tr>
<td>Acute pulmonary oedema</td>
<td>Over-enthusiastic rehydration should be avoided so as to prevent pulmonary oedema. Prop patient up at an angle of 45°, give oxygen, give a diuretic, stop intravenous fluids, intubate and add positive end-expiratory pressure/continuous positive airway pressure in life-threatening hypoxaemia</td>
</tr>
<tr>
<td>Acute renal failure</td>
<td>Exclude pre-renal causes, check fluid balance and urinary sodium; if in established renal failure add haemofiltration or haemodialysis, or if unavailable, peritoneal dialysis. The benefits of diuretics/dopamine in acute renal failure are not proven</td>
</tr>
<tr>
<td>Spontaneous bleeding and coagulopathy</td>
<td>Fresh frozen plasma, platelets transfusions, vit K.</td>
</tr>
<tr>
<td>Metabolic acidosis</td>
<td>Exclude or treat hypoglycaemia, hypovolaemia and sepsis. If severe add haemofiltration or haemodialysis</td>
</tr>
<tr>
<td>Shock</td>
<td>Suspect sepsis, take blood for cultures; give parenteral antimicrobials, correct haemodynamic disturbances</td>
</tr>
</tbody>
</table>
The above case is an imported malaria because malaria is eradicated from Egypt except small focus in Elfayoum Governorate. In 2007, Zaher et al., reported a case of imported malaria died by cerebral malaria due to delayed diagnosis before admission to Almaza Military Fever Hospital, Cairo.[4]. Also Birnbaumr concluded that death of imported malaria cases was due to miss or delay diagnosis[5].

**Conclusion:**

Malaria in travelers typically manifests days or weeks after patients left the endemic area. Malaria symptoms are non specific and rapid diagnosis and treatment are needed. Specific chemoprophylaxis for travelers to chloroquine resistant areas should be given.

**References:**

1. Saif El Din S and Abdel Wahab MF. A Guide Book of Tropical and Infectious Diseases. Tropical Medicine Department, Ain Shams University, Cairo 1995; 244-246.
5. Birnbaumr D. Malaria diagnosis missed in nearly half of patients at risk; Imported malaria prospective analysis of problems in diagnosis and management. *Clin Infect Dis* 1998; 27; 142-149
Threat of Dengue Fever and Dengue Haemorrhagic Fever to Egypt from Travelers

El-Bahnasawy MM¹, KhaliL HH², Morsy AT³, Morsy TA²
Departments of Tropical Medicine¹, Internal Medicine² and Parasitology³, Faculty of Medicine², Ain Shams University, Cairo 11566, Egypt, Military Medical Academy¹, and The Ministry of Interior Hospitals³, Cairo, Egypt.
Mamdouh25@hotmail.com

ABSTRACT
Dengue (DF) and dengue hemorrhagic fevers (DHF) are present in urban and suburban areas in the Americas, South-East Asia, the Eastern Mediterranean and the Western Pacific, but dengue fever is present mainly in the rural areas of Africa. Several factors have combined to produce epidemiological conditions in developing countries in the tropics and subtropics that favor viral transmission by the main mosquito vector, Aedes aegypti as the rapid population growth, rural-urban migration, inadequate basic urban infrastructure (e.g. the unreliable water supply leading householders to store water in containers close to homes) and the increase in volume of solid waste, such as discarded plastic containers and other abandoned items which provide larval habitats in urban areas. Geographical expansion of the mosquito has been aided by the international commercial trade particularly in used car-tires which easily accumulate rainwater. Increased air travel and the breakdown of vector control measures have also contributed greatly to the global burden of DF and DH fevers. The presence of Aedes aegypti and endemic DF and DHF in the neighboring regional countries must be born in mind of the Public Health Authorities.

Intrafamilial Transmission of HCV
El-Bendary M
Tropical Medicine Department, Mansoura University

ABSTRACT
Egypt has high prevalence rate of HCV (10.8%) with many routes of transmission. However, in 40-45% of patients the route of transmission is not definite (community acquired). There are three routes of intrafamilial transmission of HCV: 1) Households of relatives due to reused scissors and needles with 4.7% chance of transmission per year, 2) Sexual with increase prevalence for more than 15 years of marriage and 1/2 – 2% chance of transmission per year, and 3) Vertical with 5% chance of transmission per year.

Afro-Egypt J Infect Endem Dis 2011 Dec;1(2):A3

Arthropod Vectors in Toshka, Egypt
Morsy T
Parasitology Department, Ain Shams University, Egypt

ABSTRACT
Toshka is a newly developed area in the south of Egypt near Sudan. Many tropical diseases could be transmitted from Sudan to Egypt. *Simulium* (vector of oncocerciasis), *Chrysops* (vector of Loaiasis), *Culicoides* (vector of filariasis and horse sickness), *Anopheles sergenti* (vector of malaria), *Aedes aegypti* (vector of Dengue), *Tabanus taeniola* (vector of anthrax), *Phlebotomus papatasi* (vector of cutaneous leishmaniasis) and *Phlebotomus langeroni* (vector of visceral leishmaniasis) all are found in Toshka.

Afro-Egypt J Infect Endem Dis 2011 Dec;1(2):A4

Recent Guidelines for Treatment of HCV
Heikal O
Military Medical Academy, Egypt

ABSTRACT
The prevalence of HCV in Egypt is 9% with 70000-140000 new infections each year and the mortality from liver diseases is 40000 deaths per year. The goal of treatment of chronic HCV is sustained virological response (SVR) which means PCR negative 6 months after the end of combined treatment by interferon and ribavirin. Patients with negative PCR at 4, 12, 24, and 48 weeks of therapy and SVR have 98% cure rate from the virus. Pre-treatment assessment includes: HCV RNA by PCR, liver biopsy elastography or Actitests and Fibrotest (when validated to substitute liver biopsy), genotype assessment, abdominal ultrasonography, alpha fetoprotein, history of alcohol intake and detection of auto-antibodies.

Afro-Egypt J Infect Endem Dis 2011 Dec;1(2):A5

HCV Update
Baddar R
Ain Shams University, Egypt

ABSTRACT
HCV is a major public health problem in Egypt. HCV is responsible for 20% of acute hepatitis, 70% of chronic liver diseases, 60% of HCC and 30% of liver transplantation. The predictors of response to combined treatment by interferon and ribavirin are: 1) genotype; 2 and 3 have better response, 2.5 age; younger patients have better response, 3. liver injury; minimal liver damage seen histologically has good response, 4. sex; females are more responders, 5. body mass index; thin patients are good responders, 6. viral load; HCV RNA less than 2000000 IU/ml has better response, 7. smokers have bad response, 8. low level of GGT has good response, 9. alcohols have bad response, 10. co-infections with HIV and HBV have bad response, 11. Interleukin 28B polymorphism predicts response to interferon, 12. patients with insulin resistant have poor response and 13. blacks with genotype 1 have poor response. HCV causes both hypo and hyper thyroidism and 7.3-15% of patients treated by interferon have thyroid dysfunction.

Afro-Egypt J Infect Endem Dis 2011 Dec;1(2):A6

Clostridium Difficile Colitis
Hunter S
Cairo University, Egypt

ABSTRACT
*Clostridium difficile* is a gram positive anaerobic spore forming bacteria. It secretes 2 potent exotoxins; enterotoxin A and cytotoxin B. It is called antibiotic associated diarrhea. The risk factors for *C. difficile* infection are antibiotic intake in the last 2 months, old age and hospitalization. Antibiotics as cephalosporins, penicillin, clindamycin, macrolides lead to disturbance of gut flora and colonization by *C. difficile*. The clinical spectrum ranges from asymptomatic, self limited diarrhea up to fulminant bloody diarrhea with perforation of the colon. Laboratory investigations revealed leucocytosis, ...
Ideal sedation has the following characters: easy administration, large safety profile, rapid onset, induces amnesia, air way safer, and keeps the patient cooperative. Side effects of sedation are: agitation, deep sleep, loss of protective air way reflexes, hypoxia and hypotension. The patient should be monitored during sedation by pulse oximetry, ECG or capnography. Supplemental O2 may be needed during sedation. Midazolam is commonly used during endoscopy in a dose of 1-2 mg I.V. up to 5mg, it has minimal side effects in the form of respiratory and cardiovascular instability. The antidote of midazolam is flumazenil. Propofol could be used due to its rapid onset and offset. Propofol depresses the respiratory center so, facilities for endotracheal intubation should be available during its use.

Afro-Egypt J Infect Endem Dis 2011 Dec ;1(2):A7

Spontaneous Bacterial Peritonitis in Cirrhotic Patients

El Sehly A
Cairo University, Egypt

ABSTRACT
Cirrhotic patients with poor liver functions are prone to bacterial infections as spontaneous bacterial peritonitis (SBP), urinary tract infections, bacteraemia and respiratory tract infections. SBP should be suspected in any deterioration of the cirrhotic patients. 10-27% of admitted ascetic patients have SBP. The recurrence rate and mortality of SBP are 70% and 31% respectively. One third of cases of hepato-renal syndrome have SBP. Clinically the patient of SBP has fever, abdominal pain, tender abdomen, encephalopathy and hypotension. The long term daily prophylaxis by oral norfloxacin carries the risk of resistance to quinolones and should be restricted to high risk patients. The diagnosis is established clinically, by findings of > 250 polymorphonuclear leucocytes (PMN)/dl in ascetic fluid and ascetic fluid culture. Treatment should be started before the result of the culture. Secondary bacterial peritonitis should be suspected if there is a lack of response to antibiotics. 2 or more organisms detected in ascetic fluid culture, ascetic fluid glucose < 50 mg/dl, ascetic fluid protien > 10 gm/dl and ascetic fluid LDH > serum LDH. Bacterascites means culture positive ascetic fluid with PMN < 250/dl in the ascetic fluid. Treatment of SBP is by empiric cefotaxime 2 gm/12 hrs for 5 days. Ceftriaxon, amoxicillin- clavulenic acid and quinolones (if no prophylaxis with norfloxacin) could be used.

Afro-Egypt J Infect Endem Dis 2011 Dec ;1(2):A8

Sedation for Endoscopy

Nouh A
Menofia University, Egypt

ABSTRACT
Ideal sedation has the following characters: easy administration, large safety profile, rapid onset, induces amnesia, air way safer, and keeps the patient cooperative. Side effects of sedation are: agitation, deep sleep, loss of protective air way reflexes, hypoxia and hypotension. The patient should be monitored during sedation by pulse oximetry, ECG or capnography. Supplemental O2 may be needed during sedation. Midazolam is commonly used during endoscopy in a dose of 1-2 mg I.V. up to 5mg, it has minimal side effects in the form of respiratory and cardiovascular instability. The antidote of midazolam is flumazenil. Propofol could be used due to its rapid onset and offset. Propofol depresses the respiratory center so, facilities for endotracheal intubation should be available during its use.

Afro-Egypt J Infect Endem Dis 2011 Dec ;1(2):A9

Anti Fibrotic Treatment: the Present Status and Future

Emam M
Zagazig University, Egypt

ABSTRACT
There is an increasing evidence that fibrosis is a dynamic and reversible process. Clarification of the mechanism of fibro genesis with particular action of stellate cell biology, has generated a great hope that novel therapies will evolve. Until now, however, no drug has been approved as an anti-fibrotic. In reality, there may already be many existing drugs with well-established safety profiles, whose mechanism of action will be also anti-fibrotic even though they have been developed for other indications. There are several points of attack in developing anti-fibrotic agents: 1- Eliminate the cause(s) of injury and their mediators, reduce inflammation and the immune response, 2- Target specific signaling: receptor ligand interaction, intracellular signaling, 3- Reduce fibro genesis by inhibiting matrix synthesis and 4- Resolve fibrosis by: increasing scar matrix degradation; stimulating apoptosis of stellate cells; and BM or cell transplantation. Emergence of effective drugs that ameliorate fibrosis approaching, and will transform the outlook for patients with chronic liver disease. To date with the available drugs, we can say that liver fibrosis is regressive but not completely reversible disease.

Afro-Egypt J Infect Endem Dis 2011 Dec ;1(2):A10

Fibroscan as a Tool in Diagnosis of Liver Fibrosis and Cirrhosis

Conference, Afro-Egypt J Infect Endem Dis 2011; 1 (2):72-75
www.mis.zu.edu.eg/ajied/home.aspx
El-Shamy M
Zagazig University, Egypt

ABSTRACT
The gold standard for diagnosis of liver fibrosis is liver biopsy, but it is an invasive technique needing relatively large size of liver tissue. An alternative to liver biopsy is the non-invasive elastography which depends on measuring the speed (velocity) of propagation of shear waves (vibration) through the liver tissues and it depends on the density of tissues. Fibroscan is a non-invasive, painless, outpatient, operator independent procedure. It has limitations as in obese patients with thick chest wall (use XL probe), in children with thin chest wall (use S probe) and in patients with narrow intercostal spaces. The normal elastography is 5.5±1.6 kilopascals (kpa). Fibroscan is useful in chronic HCV, HBV, alcoholic liver diseases, NASH, NAFLD and cirrhosis. Elastography according to meta-analysis study of 17 kpa is associated with cirrhosis, 49 kpa with ascites, 54 kpa with HCC and 63 kpa with bleeding varices.