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MGM Journal of Medical Sciences

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From the Editor's Desk

World Hepatitis Day was celebrated all over the world on 28th July like every year. In 2015, viral hepatitis accounted for 1.34 million deaths worldwide. Around 257 million people have chronic hepatitis B virus (HBV) infection and 71 million have hepatitis C virus (HCV) infection. Causes of death are chronic liver disease (cirrhosis) and hepatocellular carcinoma. The World Health Organization (WHO) has resolved to eliminate viral hepatitis by 2030. Elimination has been defined as 65% reduction in mortality and 90% reduction in incidence. Hepatitis A virus (HAV) which is transmitted by contaminated food and water caused 11,000 deaths (0.8% of viral hepatitis mortality) worldwide in 2015. An effective and safe vaccine is available against HAV. About 3.5 percent of global population is infected with HBV, and one percent with HCV. These two infections account for 96% of hepatitis deaths. Unfortunately, 95% of patients infected with HBV and HCV do not know that they are infected. One-fifth of these patients develop end-stage liver disease. Alcohol abuse and HIV infection accelerate progression of liver failure. Both these viruses are transmitted by unsterile injections, unsafe blood transfusions, unprotected sex with infected partners and mother-to-child transmission. With the availability of the safe and effective vaccine, HBV is totally preventable. It needs to be supplemented with ensuring safety of blood transfusions, safe injection practices and diagnosis of asymptomatic carriers of the viruses in the population by robust testing programs. Now effective and safe antiviral drugs, approved by the WHO, are available to treat HBV- and HCV- infected individuals. By eliminating the viruses in the infected individuals with appropriate antiviral drug therapy, we can reduce the source of infections.

Hepatitis D virus (HDV) is also transmitted through parenteral route and usually coinfects those who already have HBV infection. It responds to older interferon-based therapies. Hepatitis E virus (HEV) infection is acquired mostly by contaminated water. About 20 million infections occur every year and estimated 44,000 patients sucuumb to it. China is believed to have developed a vaccine against HEV.

To summarize, WHO stresses on HBV vaccination, blood safety, injection safety and provision of clean water (in HAV & HEV) as the pillars of viral hepatitis elimination strategy, which can be achieved by 2030 if fully implemented. For this, all-out efforts need to be made by all nations individually and jointly with WHO.

We have pleasure in presenting before our esteemed readers the current issue of MGM Journal of Medical Sciences (MGMJMS) with original papers, review articles, and interesting case reports. We are delighted to note increase in the number of contributors from India and abroad to this scientific journal, submitting quality papers.

Shibban K Kaul MS MCh FIACS
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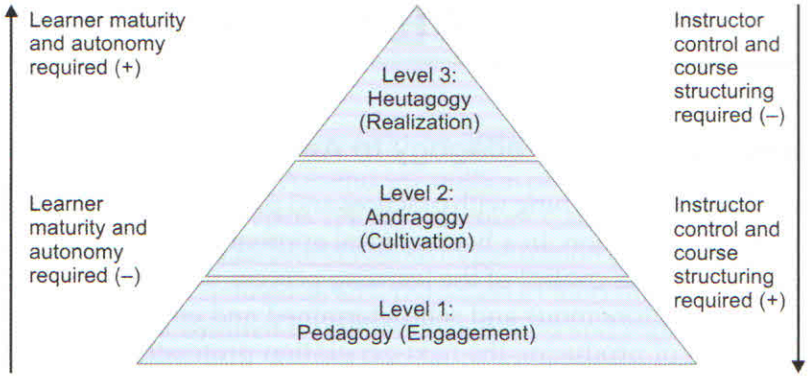

Fig. 1: Progression from pedagogy to andragogy to heutagogy[4]

Considering the development in educational technology, pedagogy and andragogy have become insufficient because they lack reflection of what is learned and how it is learned by students. Perhaps a heutagogical learning environment can make it possible to create capable students and can result in both the development of student competencies and capability and capacity to learn. Sometimes, heutagogy has been known as a net-centric theory serving as a framework for digital-age teaching and learning. Higher education educators should utilize the inherent quality of self-determination of students to help them to learn to accomplish their goals in life. This should be recognized and ample opportunity must be provided for students to nurture this behavior by providing learning opportunities that support learner autonomy. Educators should not view heutagogy with suspicion, as it crosses the boundaries of pedagogy and andragogy and places full control of all aspects of learning into the hands of the student.[5]

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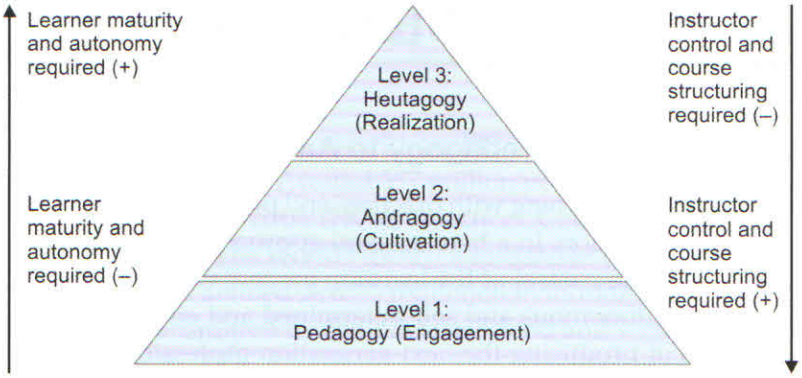

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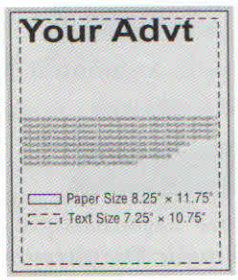

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ORIGINAL ARTICLE

TlyA Expression is Necessary for Mycobacterial Susceptibility to Capreomycin: Wild-type Strains are Naturally Resistant

[1]Shikha Nag, [2]Krishnasastry Musti

ABSTRACT

The human pathogen *Mycobacterium tuberculosis* has coevolved with humans and uses novel strategies for intracellular survival as well as for drug resistance. In this communication, we have compared the drug resistance of *Mycobacterium marinum* as well as the H37Ra, an avirulent form of the human pathogen *M. tuberculosis* against second-generation antibiotic capreomycin. Interestingly, *M. marinum* and its three mutants are naturally resistant to capreomycin, while the H37Ra showed susceptibility and also it showed surface expression for the TlyA protein since function of TlyA is necessary for susceptibility to capreomycin. It is postulated that the resistance to capreomycin can occur in wild-type strains through suppression of expression of TlyA, while the H37Ra is unable to do the same.

Keywords: Capreomycin, Drug resistance, Drug susceptibility, H37Ra, *Mycobacterium marinum*, *Mycobacterium tuberculosis*.

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Conflict of interest: Nil

INTRODUCTION

Emergence of drug-resistant strains of mycobacterial species is a serious problem, which has been highlighted by the World Health Organization.[1] Although many aspects have come to light regarding this problem, the major reasons attributed to drug resistance could be due to evolution of mutant forms and or other mechanisms. Considering the number of possibilities that can contribute to drug resistance, the following are distinct possibilities, viz.

- Drug is not experienced by the bacterium, i.e., its failure to penetrate the bacterium or effectively pumped out of the bacterium.
- The drug does not find its target.
- Processing of pre- or pro-form of the drug fails.
- Emergence of mutations that prevent the drug binding either in nucleic acid sequences or its product proteins.

Based on these broad categories, one can easily see many subcategories except for the first possibility which requires specialized efflux pumps which are yet to be delineated at the molecular level. It remains to be explored whether they can pump all drugs that are currently in use. However, the possibilities 2 to 4 have an underlying phenomenon, i.e., whether or not heterogeneity can contribute to drug resistance. For example, a given drug may not find its target if a single bacterium has evolved in such a way that it does not express or partition a particular protein during cell division as pictorially depicted in Figure 1. In such a scenario, the bacterium that retained the bulk of the protein will be susceptible while the bacterium that did not receive or make the same protein will be resistant. In this possibility, it is important to note that the genome of both bacteria is the same, while one is susceptible and the other resistant. Hence, the question, whether such bacteria in principle exist or not can be experimentally explored. In this present work, we are able to identify that *M. marinum*, a close relative of the human pathogen *M. tuberculosis*, and its three mutants appear to be resistant to the second-generation antibiotic, capreomycin, while the H37Ra, the avirulent form of the same human pathogen, is susceptible to the same drug.

MATERIALS AND METHODS

Bacterial Strains

The bacterial strains *M. marinum* and its transposon mutants M1, M2, and M3 were a generous gift from Dr Eric Brown, Genentech, USA, which were described in Gao et al.[2] The *M. tuberculosis* H37Ra was obtained from Microbial Type Culture Collection, Institute of Microbial Technology, Chandigarh, India. All the antibiotics used in this study were of analytical grade and were obtained from Sigma-Aldrich Co., USA.

Bacterial Growth

All the bacterial strains were cultured as per protocols suggested by American Type Culture Collection (ATCC) using complete 7H9 media glycerol (0.2% v/v), albumin,

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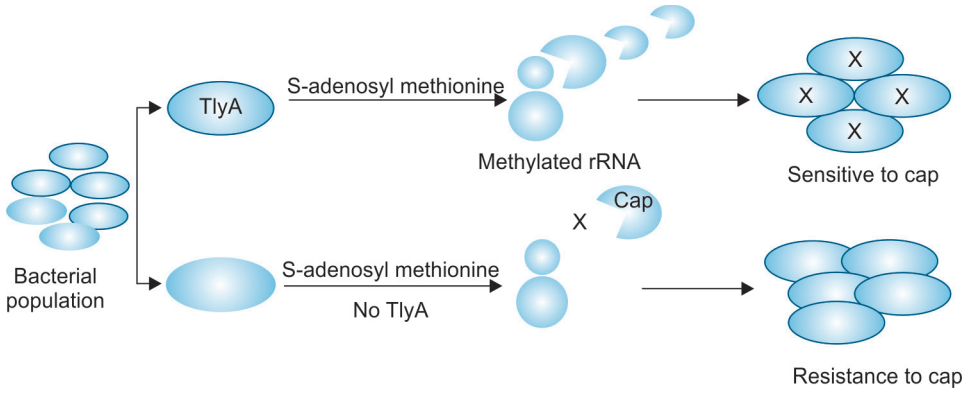


Fig. 1: Illustration of TlyA expression-dependent susceptibility. The bacteria that express the TlyA protein are susceptible to capreomycin due to methylation of 16S and 23S rRNA, while the bacteria that do not express the protein will be resistant in the absence of methylation. It is known in literature that the TlyA is not essential for *in vitro* growth of mycobacteria (based on transposon mutagenesis) and hence, the possibility of lack of its expression

dextrose and catalase (ADC), and Tween-80 (0.05% v/v). Each of the strains (WT, M1, M2 and M3) was enumerated (taken from a log phase culture) and inoculated in flasks with equal volume of medium with an inoculum ratio at 1:100 with or without antibiotics. The growth curve was monitored by taking A_{600} of each sample for every 6 hours for indicated time intervals.

Antibiotic Disk Susceptibility Assay

The bacterial culture described earlier was washed thrice washed in phosphate-buffered saline containing Tween-80 (0.05%) and passed through 26-gauge syringe to dissociate clumps. All cultures having 10^6 cells/mL were spread onto complete 7H10 agar plates containing glycerol (0.2% v/v), oleic acid, albumin, dextrose and catalase (OADC), and Tween-80 (0.05% v/v). The antibiotics were applied with Whatmann filter paper No.1 disc that were saturated with desired antibiotic concentration. The plates were incubated at 30°C or 37°C and observed after 21 days. A plain 7H10 agar plate was also kept as negative control. The antibiotics tested were ampicillin (100 and 50 µg/mL), hygromycin B (50 µg/mL), and capreomycin with concentrations of 100, 80, 50, 40, 10, 5, 1, and 0.1 µg/mL.

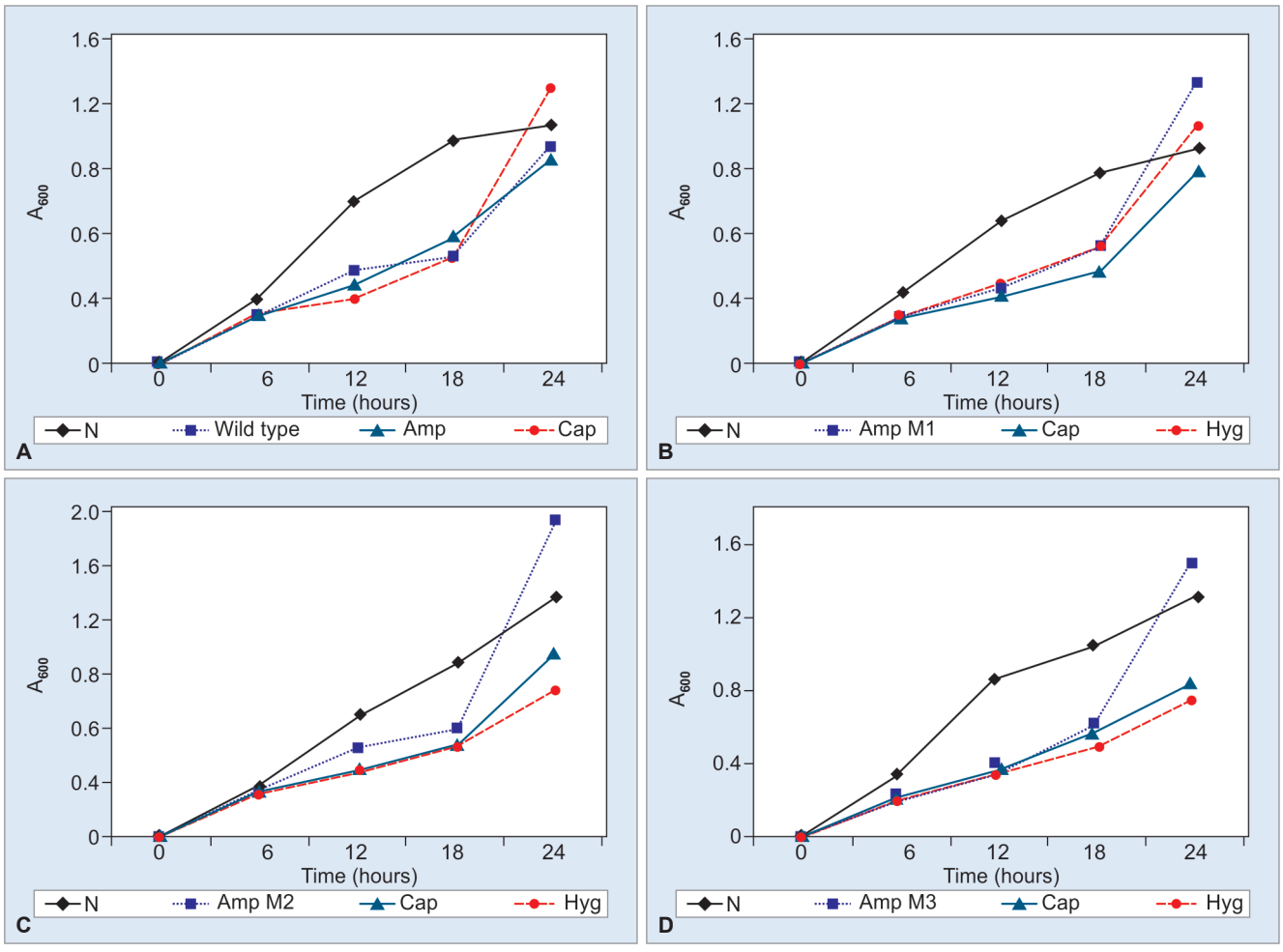
Confocal Staining

Surface staining of H37Ra was carried out as per earlier report using same reagents.[3]

RESULTS AND DISCUSSION

The genus *Mycobacterium* has many members that are pathogenic to variety of species and in humans, it is known by the tuberculosis disease. The *M. marinum* causes systemic tuberculosis like disease in fish and frogs and also localized skin suppurations in immunocompromised humans.[4] Both the human and animal infections are marked by the presence of a granulomatous host response, the hallmark of *M. tuberculosis*. *Mycobacterium marinum* is genetically closely related to *M. tuberculosis* and has been used increasingly as a model for understanding the pathogenesis of tuberculosis. *Mycobacterium tuberculosis* and *M. marinum* share >90% sequence homology based on 16S ribosomal ribonucleic acid (rRNA) sequences.[5] The virulence of nonpathogenic strains of mycobacterial species can be restored with the counterparts of the *M. tuberculosis* which is well demonstrated in the literature.[2,6,7] Detailed study of *M. marinum* can greatly help in developing screening methods for antimycobacterial agents since: (i) It has phylogenetically close relationship with *M. tuberculosis;* (ii) it has a relatively rapid doubling time; (iii) it shows similar drug susceptibilities to *M. tuberculosis;* and (iv) it is less expensive for biosafety level environment and its evolutionary aspects can be studied safely.

The strains designated with M1, M2, and M3 respectively, represent the transposon insertion strains of *M. marinum* that disrupt Mh3866, Mh3867, and Mh3868, which are homologous to Rv3866, Rv3867, and Rv3868 of *M. tuberculosis* respectively. Based on the literature, these genes are necessary for hemolysis exhibited by *M. marinum*.[2] We have earlier shown that the *M. marinum* expresses the *tlyA* gene product which has been independently shown to possess both the activities, i.e., hemolysis and S-adenosyl-L-methionine-dependent rRNA methylation activities.[3,8] Susceptibility to capreomycin by a bacterium is dependent upon the expression of *tlyA* gene product, which methylates the nucleotides C1409 and C1920 of 16S and 23S rRNA respectively. Methylation of rRNA results in reduced translational ability, as the methylation of the ribosomes facilitates the binding of capreomycin.[9] Hence, bacteria that do not carry the *tlyA* gene are naturally resistant to capreomycin, e.g., *Escherichia coli* has no natural homologue and is resistant to capreomycin which, upon expression of the *tlyA* gene, shows susceptibility.[10]



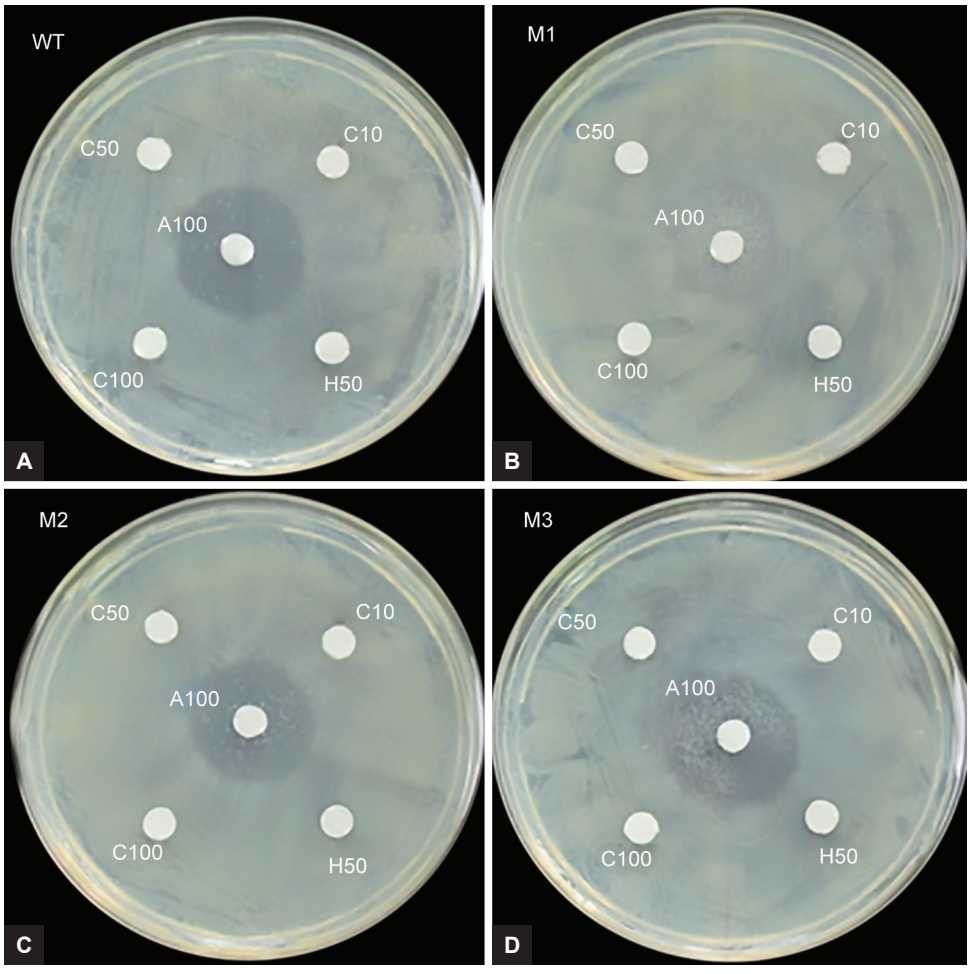
Graphs 1A to D: *In vitro* culture of *M. marinum* wild type, M1, M2, and M3 in the presence of indicated antibiotics: all the strains were cultured as described in Methods section in the presence of ampicillin (100 µg/mL), hygromycin B (50 µg/mL), and capreomycin (100, 50, and 10 µg/mL) and their growth was monitored for 24 hours

In view of the above observations, all the four strains were examined for growth in the presence of ampicillin, hygromycin B, and capreomycin as shown in Graph 1. The growth curves of all the four strains in the absence of the antibiotic show rapid growth, while in the presence of these three antibiotics, growth only retarded for about 18 hours, after which the growth has dominated that of the wild-type bacteria. Consistent with this observation, the antibiotic disc diffusion assay also showed no inhibition of growth as seen in Figure 2 in which we could see some inhibition only in ampicillin, but no significant inhibition in case of hygromycin B while we could not see any kind of inhibition in case of capreomycin.

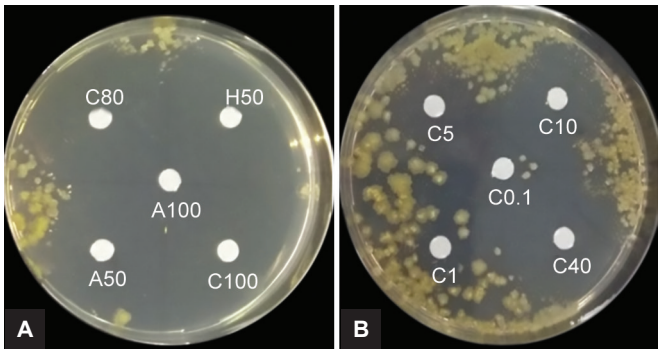
Attenuated Strain H37Ra is Susceptible to Capreomycin and exhibits Surface Expression of TlyA

Mycobacterium tuberculosis strain H37Ra, commonly used in studies with the virulent H37Rv, was originally derived from virulent strain H37.[11] Several studies have attempted to determine the genomic and proteomic differences leading to the basis of virulence attenuation of H37Ra, which appears to possess insertions, deletions, and mutations in some transcription factors.[12-14] However, detailed studies on H37Ra can have implications both in understanding the pathogenesis of virulent counterpart and the development of new vaccines and therapeutic agents. We, therefore, sought to examine H37Ra and its susceptibility to capreomycin in context of *tlyA*.

In contrast to the observations described above, the H37Ra did not grow at all in the presence of various concentrations of the capreomycin in liquid media, while the agar plate showed dramatic loss of growth in the presence of capreomycin as seen in Figure 3 and Table 1. The colony morphology of H37Ra is consistent with literature pictures as well as the ATCC cultures examined in the laboratory. We have also examined for the presence of TlyA protein on the surface of H37Ra, which showed an unambiguous presence, whereas the expression of TlyA in M1, M2, and M3 is unobservable, while the wild type is noisy (unpublished observations). As seen in Figure 4, the confocal microscopic visualization



Figs 2A to D: Disc diffusion assay of *M. marinum* wild type, M1, M2, and M3: disc diffusion visualization was carried out as described in Methods section. The C100, C50, C10, A100, and H50, respectively, represent capreomycin 100, 50, and 10 µg/mL, ampicillin 100 µg/mL, and hygromycin B 50 µg/mL respectively



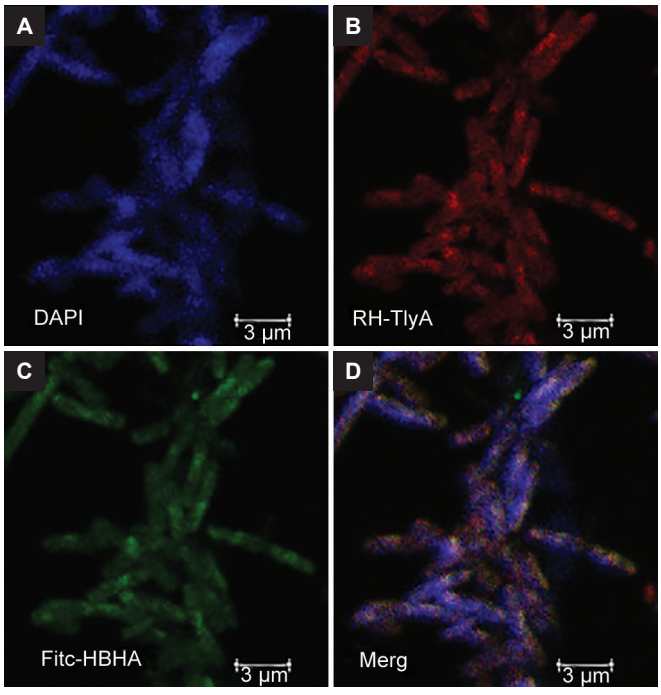
Figs 3A and B: Disc diffusion assay of H37Ra in the presence of capreomycin: Disc diffusion assay for H37Ra was carried out as described for *M. marinum* in the presence of capreomycin and other antibiotics. The C 0.01, C1, C5, C10, C40, C80, C100, A50, and H50, respectively, represent capreomycin, ampicillin, and hygromycin B with respective concentrations

Table 1: Antibiotic susceptibility of H37Ra and *M. marinum*

Antibiotic	*Conc, µg/mL*	*H37Ra*	*M. marinum*
Hygromycin B	40	s	r
	100	s	s-r
	50	s	r
Kanamycin	30	s	8
Capreomycin	100	s	r
	80	s	r
	50	s	r
	40	s	r
	10	s	r
	5	s	r
	1	s	r
	0.1	s	r

of H37Ra also showed positive surface staining for TlyA (Rh-Rhodamine channel) and heparin-binding hemagglutinin (HBHA), a well-known surface protein of H37Rv (FITC-HBHA). The observations shown in Graph 1, Fig. 2 and Table 1 are also consistent with a published literature in which many mycobacterial strains seem to be naturally resistant to capreomycin and isoniazid.[15]

It is interesting to note here that the wild-type and mutant strains of *M. marinum* are resistant to capreomycin for lowest to highest usable concentrations,



Figs 4A to D: Confocal visualization of H37Ra for TlyA expression: Surface staining of H37Ra was carried out as described earlier for *M. marinum* (3). The panels labeled with 4',6-diamidino-2-phenylindole (DAPI), rhodamine, fluorescein isothiocyanate (FITC) respectively, represent staining for deoxyribonucleic acid, TlyA staining with rhodamine-antirabbit-immunoglobulin G (IgG) and HBHA detected with FITC-antimouse-IgG

while the H37Ra is susceptible to it. Based on Figure 1, it is important to understand the expression profile of TlyA for evolution of possible heterogeneity or noise in mycobacterial species in *in vitro* culture conditions and its significance for intracellular survival.[16] In addition, the high prevalence of TlyA on the surface of H37Ra is very important for further studies, as H37Ra has the ability to survive in humans and mice, but does not cause the classical disease and also has not been shown to form granuloma. It is relevant to mention here that TlyA expression can aid in intracellular survival of even nonpathogenic versions upon expression of the same.[17] The H37Ra has been shown to contain mutations in PhoP regulon and it is necessary to focus the future studies on PhoP regulon and its role for evolution of susceptibility or resistance to second-generation antibiotic, such as capreomycin, which is not often used for treatment of tuberculosis, to enable its usage.[18] It is also not surprising that the possibility discussed in Figure 1 is found to be true, as many clinical isolates with wild-type *rrs* and *tlyA* genes have minimum inhibitory concentration values well above 0.5 to 2 mg/mL.[19] Hence, it is very important to study the evolution of this phenotype for combating the drug resistance problem posed by tuberculosis disease and such studies are currently underway.

ACKNOWLEDGMENT

Authors would like to thank Dr Shekhar Mande for encouragement and support.

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ORIGINAL ARTICLE

Actinomycosis Mimicking Gynecological Malignancy: Imaging Patterns in Seven Cases

[1]Nikhil Bhuskute, [2]Ranjeet Shinde

ABSTRACT

Pelvic actinomycosis is uncommon and believed to be related to the use of intrauterine devices (IUDs). It may present as a complex gynecological mass either uterine or adnexal with or without local pelvic spread or with peritoneal dissemination, all features which mimic gynecological malignancy. We describe seven women with proven actinomycosis who presented to a single cancer center gynecological cancer multidisciplinary team meeting (MDTM) to illustrate these imaging appearances and highlight discriminant features of actinomycosis. A minority of women had concurrent use of an IUD. Involvement of the pararectal space was a feature of pelvic disease extension. We describe the value of image-guided core biopsy (IGCB) in confirming the diagnosis.

Keywords: Arteriovenous fistula, Fistula, Hemodialysis, Infection, Patency, Primary failure, Rates, Steal syndrome, Thrombosis, Vascular access.

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INTRODUCTION

Otto Bollinger described bovine actinomycosis in 1877 and in 1879, James Israel described the first human involvement. Actinomyces israelii is a gram-positive, anaerobic, non-acid fast filamentous bacterium. It is a common commensal in the oral cavity, vagina, and large bowel.[1] Pelvic actinomycosis is an uncommon infection, most commonly described in association with use of IUDs[2,3] diverticular disease, cholecystitis, abdominal surgery, and penetrating trauma.[4,5] We describe our experience of seven women with pelvic actinomycosis mimicking gynecological malignancy at a tertiary oncology center which, to the best of our knowledge, is the largest such series yet reported.

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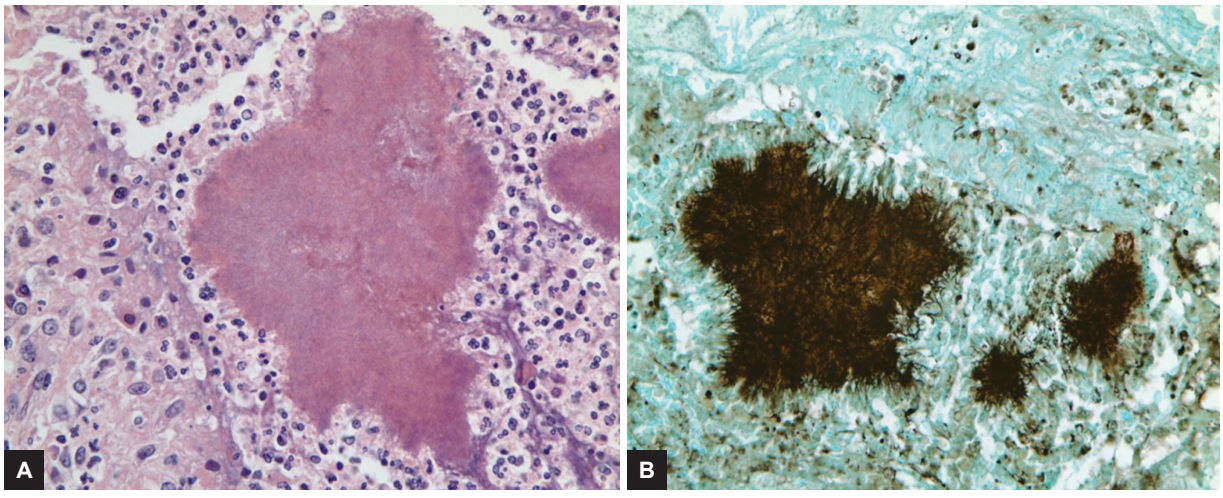
MATERIALS AND METHODS

Clinical

Over a 5-year period, seven cases of abdomino-pelvic actinomycosis were identified prospectively by one of the authors during attendance at the weekly MDTM. Five cases were histologically proven and two were assumed to be actinomycosis, based upon resolution of imaging abnormalities following specific antibiotic therapy. These two cases had no evidence of malignancy on multiple core biopsies which showed only a mixture of acute and chronic inflammation. No other organism was isolated from vaginal or uterine swabs. Because of a strong clinical and imaging suspicion of the diagnosis, the women were treated as actinomycosis with penicillin with excellent clinical recovery and complete or partial resolution of the imaging abnormalities. No other cases of proven pelvic urogenital actinomycosis were identified in a search of the pathology department database for this period.

The average age at presentation was 43 years (32–54 years). One woman had an IUD in place at presentation and two women had prior use of IUDs. All seven were referred to the regional MDTM with a provisional diagnosis of gynecological malignancy. Five women were suspected to have adnexal cancer and two to have suspected cervical cancer. One woman had prior grade II cervical carcinoma, treated by hysterectomy. Four women presented from our own local gynecology team and three were referral cases having already been discussed in other local cancer unit MDTM. No patient had a history of fever. The tumor marker CA-125 was raised in only two cases while the inflammatory marker C-reactive protein was raised in four of the seven cases. In five cases, the white cell count was raised (12.4–18.9 × 10^9/L) and in all cases, differential white cell count showed neutrophilia.

One woman was thought to have pelvic inflammatory disease (PID) after initial MDTM review and was treated in her local hospital as such. While undergoing antibiotic therapy, she required exploratory surgery for unremitting pain and for drainage of pelvic suppuration. Another woman with a complex pelvic mass suspected to represent ovarian cancer developed small bowel obstruction necessitating emergency surgery a few days after discussion in the MDTM. Both these patients were found to have actinomycosis on examination of the surgical specimens. Diagnosis in the remaining five patients was made by



Figs 1A and B: (A) The H&E stain shows sulfur granules, which are circumscribed masses of bacteria in branching filaments with a radial/palisading pattern at the periphery. (B) The Grocott stain shows the filaments to better effect. The granules are surrounded by a mixed inflammatory infiltrate, mainly neutrophil polymorphs with some histiocytes

image-guided biopsy or gynecological biopsy directed by imaging features. These women avoided surgery.

Confirmatory histology was thus obtained using IGCB in three women, from operative specimens for two women and image-guided biopsy for the other two women. They both had an abnormal cervix and uterus on imaging as well as on clinical examination, but with no clinical evidence of malignancy. After MDTM discussion, they underwent cervical and parametrial biopsy and/or uterine curettings, which showed inflammatory material only. Subsequent imaging in these two women showed resolution of abnormalities after antibiotic therapy.

Image-guided biopsies were taken following local anesthesia under either ultrasound (US) or computed tomography (CT) guidance; US-guided biopsy was diagnostic. For one woman, CT-guided biopsy was initially performed and showed nonspecific chronic inflammatory changes, but a repeat US-guided biopsy was diagnostic of actinomycosis. For IGCB, an 18-gauge cutting needle incorporating a spring-loaded device was used, producing a core of up to 1.8-cm-long specimen. Biopsies were taken from the infracolic omental cake (1), liver (1), and a pelvic mass.[1] The number of biopsy cores that were taken was at the discretion of the operator, but the aim was to provide material equivalent of two full biopsy cores. The IGCB was only performed after MDTM review and when there was imaging evidence of dissemination of the disease process.

The institutional review board granted a waiver to review the case notes in further detail in a retrospective fashion.

Pathology

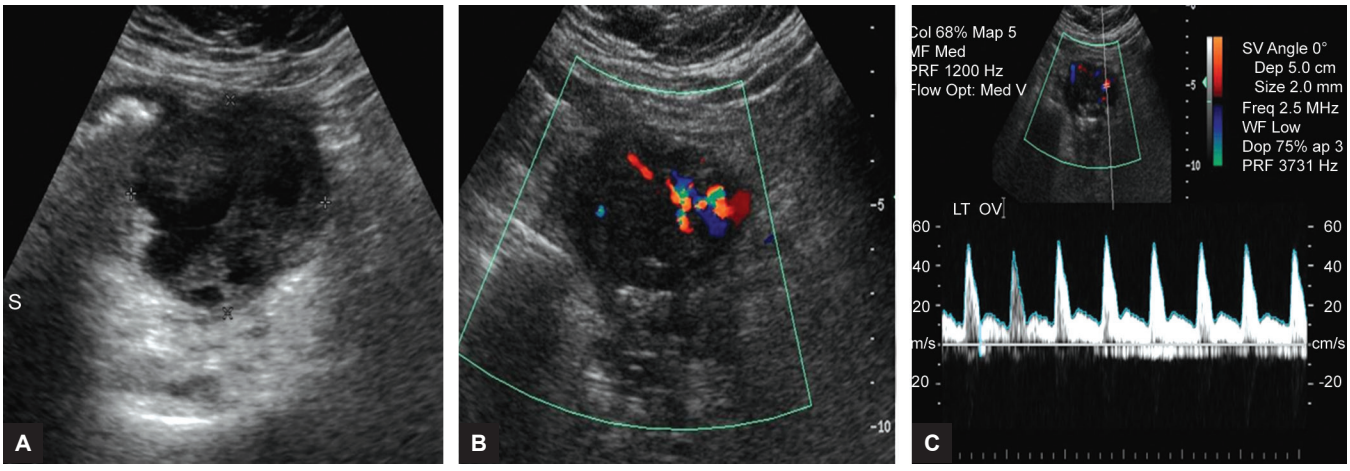
The sections were initially routinely processed in paraffin and stained with hematoxylin and eosin (H&E), followed by Gram, Grocott, Hexamine silver, and extended periodic acid–Schiff stains. Actinomyces colonies were readily identified as distinct "sulfur granules" on histological examination and these were typically surrounded by inflammation, granulation tissue, and fibrosis. This formation of an inflammatory mass may mimic a malignant tumor on macroscopic examination. The diagnosis of actinomycosis was made on microscopic examination of the specimen by the presence of sulfur granules and the absence of neoplastic cells. The sulfur granules comprised a central eosinophilic core surrounded by radiating gram-positive bacterial filaments (Fig. 1A). The diagnosis of actinomycosis was confirmed by the highlighted slender filaments of actinomyces on silver and Gram stains (Fig. 1B).

Imaging Features

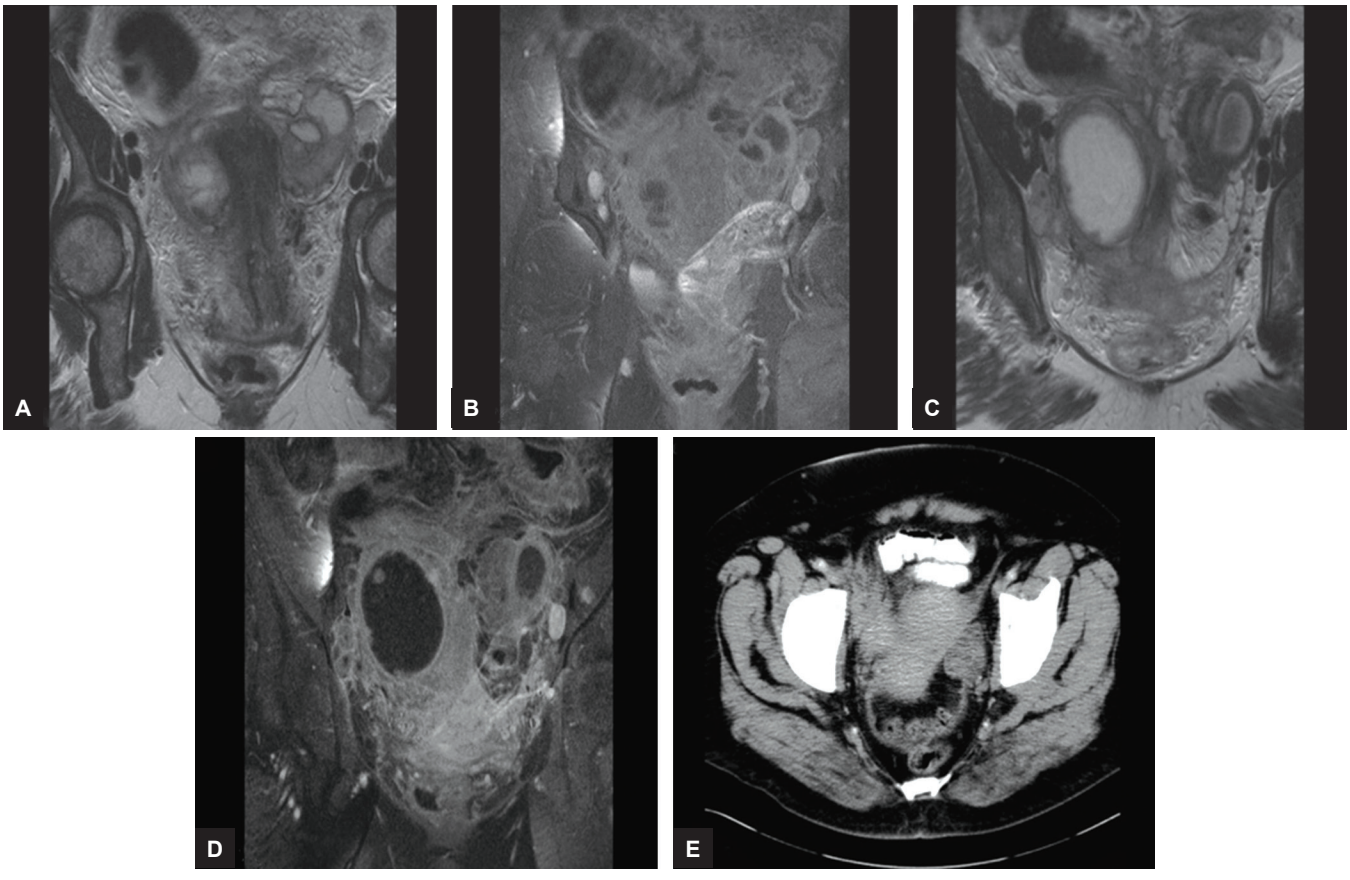
The presentations mimicking as gynecological malignancy are summarized in Table 1.

Table 1: Imaging features in seven cases of actinomycosis

Imaging feature	*Number of cases*
Features of primary cancer	
Adnexal mass	5
Uterine/cervical mass	2
Features of local extension	
Lymphadenopathy	2
Pelvic sidewall involvement	2
Pelvic fluid collections	2
Involvement of sigmoid mesentery	1
Involvement of pararectal spaces	3
Hydronephrosis	2
Features of dissemination	
Ascites: pelvic	2
Ascites: upper abdominal	0
Omental masses	2
Liver surface deposit	1

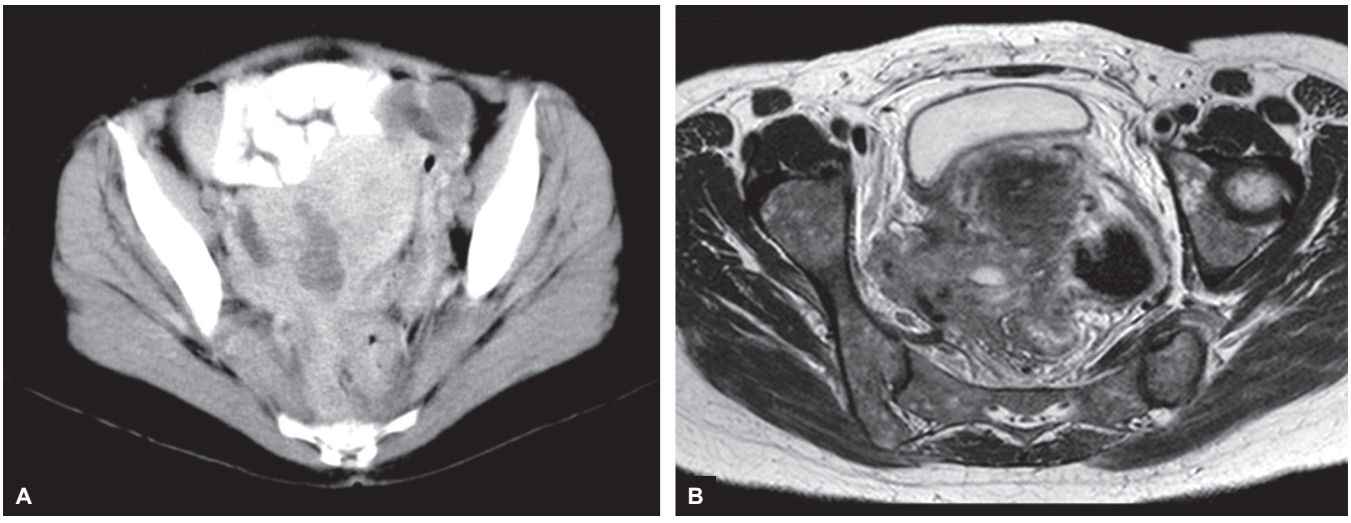


Figs 2A to C: (A, B) A sonographically complex solid-cystic mass in the left adnexa which shows abnormal color flow in the solid component and is thus suspicious for a malignant mass; (C) the spectral Doppler shows low-resistance velocity waveforms suggestive of malignancy

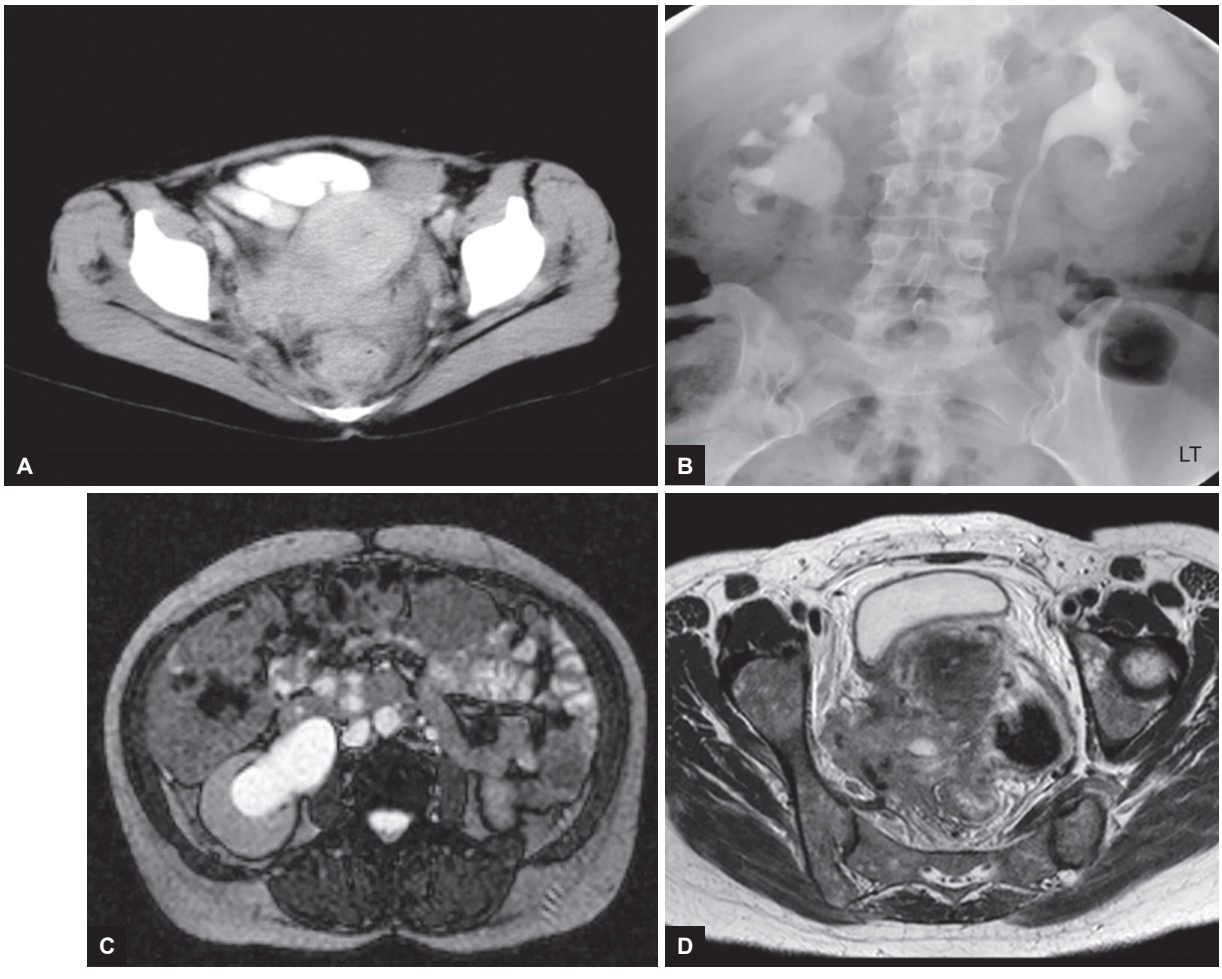


Figs 3A to E: (A) Coronal oblique T2-weighted MRI shows bilateral complex adnexal masses that have low signal intensity with some regions of high signal intensity and (B) the corresponding contrast-enhanced fat-suppressed T1-weighted MRI shows enhancement of the multilayered wall around cystic spaces. A 10 mm lymph node with enhancement is seen in the left iliac chain. Note the generalized "stranding" of the fat of the pelvic peritoneum in (A) which enhances on (B) suggestive of a diffuse infiltrative process. (C) Coronal T2-weighted MRI shows mural nodules in the cystic part of the right adnexal mass and (D) the corresponding contrast-enhanced fat-suppressed T1-weighted MRI which shows enhancement of the mural nodules along with the thick cyst walls on the right side. Note also right iliac lymphadenopathy showing small foci of necrosis. (E) Contrast-enhanced CT at an earlier time shows the infiltrative pattern of extension of disease from the right adnexal mass along the broad ligament and sigmoid mesentery. Simultaneous extension to the right pelvic sidewall and sigmoid mesentery is an odd pattern with primary ovarian malignancy

There were complex adnexal masses with solid components which showed abnormal Doppler flow on US and abnormal gadolinium enhancement at magnetic resonance imaging (MRI) (Figs 2 and 3). The solid components included mural nodules or mural thickening and irregularity exceeding 3 mm in thickness. Pelvic lymphadenopathy was seen in two women, one with solid lymphadenopathy and the



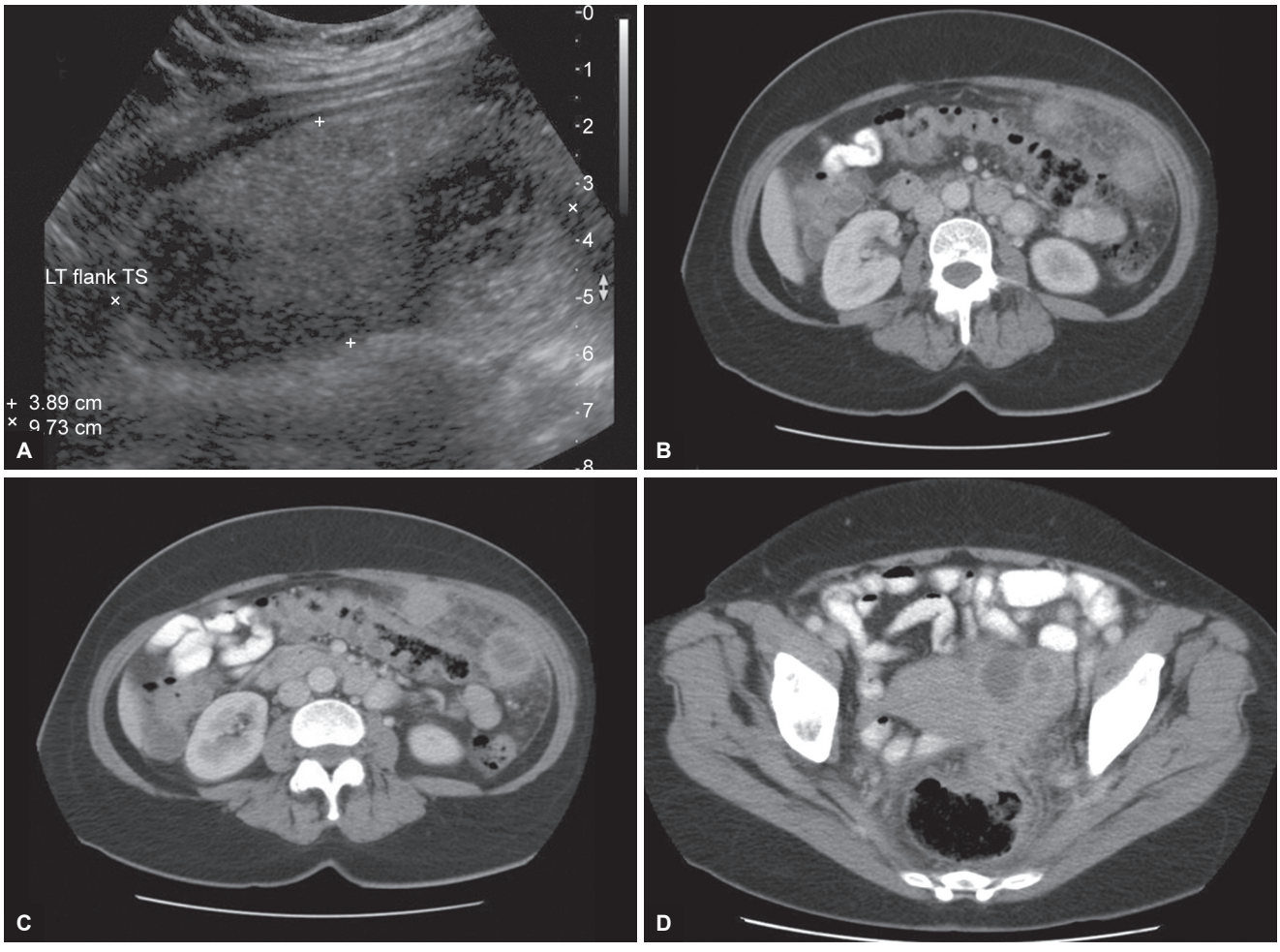
Figs 4A and B: (A) Contrast-enhanced CT scan shows a right adnexal mass with large areas of necrosis. Also note the right pelvic side wall involvement and a "tongue-like" direct extension though the right para-rectal space to the sacrum, again an odd feature for primary adnexal malignancy and (B) an axial T2-weighted MRI shows the same process. The adnexal mass has intermediate signal intensity with some regions of high signal intensity



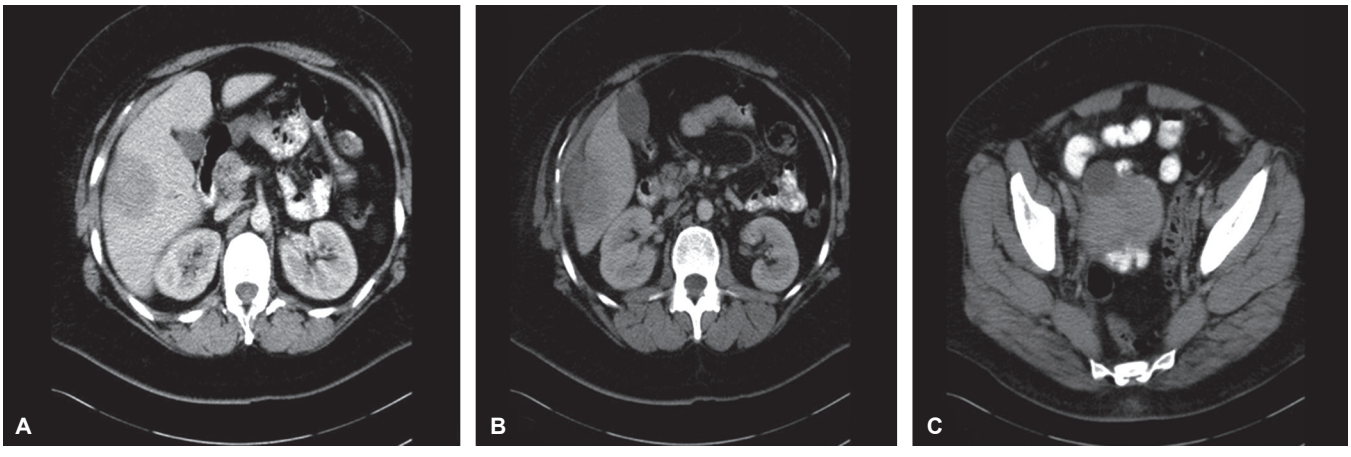
Figs 5A to D: The adnexal mass causing hydronephrosis on various modalities

other with multiple small areas of micro-necrosis in enlarged nodes (Fig. 3).

Local pelvic infiltration extended into sigmoid mesentery, pararectal and mesorectal spaces, or laterally to the pelvic sidewall (Fig. 4). The infiltrative process also involved the ureter causing hydronephrosis in two women (Fig. 5). When disease was predominant in the subperitoneal space of the pelvis, there was marked enhancement of the pelvic peritoneum and florid stranding of the fascial and fat spaces above and below this (Fig. 6).



Figs 6A to D: (A) Ultrasound of the left flank shows a low echo texture solid mass just below the anterior abdominal wall in keeping with an omental cake. (B) and (C) Contrast-enhanced CT which confirms an infiltrative mass in omentum with at least two large cavitating masses and small amount of free fluid hepatorenal pouch. (D) The CT of the pelvis shows a complex left adnexal mass with invasion of the mesorectal fat, an unusual feature of primary ovarian cancer



Figs 7A to C: (A) and (B) Contrast-enhanced CT shows a liver surface deposit extending as a round, solid mass into the parenchyma of the right lobe of the liver and which simulates a metastatic deposit and (C) CT at the level of pelvis of the same patient shows a predominantly solid adnexal mass. Note the absence of ascites on these images and indeed no intervening disease between adnexa and liver

Omental masses and liver surface deposits were seen in two women (Fig. 7). Notably on CT, the omental masses were nodular, more like "buns" than "cakes," and these showed central low attenuation (Fig. 6). This feature correlated histologically with central necrosis. Ascites were minimal or absent.

The MR signal characteristics were varied. There was bland T1-signal in the solid elements of pelvic

Table 2: Unusual imaging features in actinomycosis and comparison with PID and malignancy features

Features	*Acute PID*	*Actinomycosis*	*Primary ovarian malignancy*
Primary disease site	Uniform, thick-walled predominantly cystic. Occasional thickening of peritoneal folds	Ill-defined cystic or solid masses. No respect for fascial planes in spite of smaller primary mass	Usually well defined, solid, cystic, or mixed. Fascial planes offer longer resistance to local spread for smaller masses
Omental disease	Usually fine peritoneal thickening and enhancement	Masses like "buns" with or without cavitations	Large omental cakes or wispy disease. Rarely cavitate prior to treatment
Lymphadenopathy	Reactive	Micro-abscess within nodes or solid nodes	Usually solid with exception of cervical cancer
Liver surface disease	Rarely seen except in Fitz-Hugh-Curtis syndrome	Liver surface disease in absence of ascites	Liver surface or parenchymal disease almost always with ascites
Mesorectal invasion	Rare, thickening of uterosacral fold common with posterior extension	Mesorectal invasion out of proportion to size and site of primary mass	Unusual with adnexal masses, seen more commonly in cervical cancer
Pelvic sidewall involvement	In the form of enlarged reactive nodes	Involvement out of proportion to size and site of primary mass	Commoner with cervical cancer than other adnexal masses
Hemorrhage	Rare	Seen in solid masses	Unusual feature of solid ovarian mass

actinomycosis. T1 high signal suggestive of hemorrhage was not seen. T2-weighted images showed both low to intermediate signal in some of the solid areas, possibly suggesting a fibrotic process. High signal intensity was associated with cysts, fluid collections, or necrotic areas, and intermediate signal in the mural nodules of predominantly cystic masses. Involved pelvic floor and pelvic sidewall muscles also showed increased T2 signal. T1-weighted gadolinium-enhanced images showed intense enhancement of the solid elements or the solid components of lymphadenopathy and cystic masses and within the septae of complex cystic-solid masses. Enhancing infiltration of pelvic fat on fat-suppressed T1-weighted images was a prominent finding.

DISCUSSION

While 20% of IUD users have actinomyces-like organisms as part of their normal genital flora,[6] pelvic actinomycosis is a very rare, but serious infections may require long-term medical (antibiotic) therapy and may necessitate intervention and surgery to manage complications. Two of our seven cases required surgery for symptoms, one for small bowel obstruction and another for unremitting pain and pelvic sepsis which did not respond to standard antibiotic therapy for PID. There are thus similarities between actinomycosis and advanced gynecological malignancy in both clinicoradiological presentation and management. Pelvic actinomycosis is a chronic suppurative and granulomatous disease which does not respect anatomical barriers.[5] This property may result in condition being mistaken for a malignant "frozen pelvis." Thus, in some cases, unnecessary radical cancer surgery has been performed.[7,8]

The unifying feature of our seven cases is that they all presented to a gynecological oncology MDTM in a Cancer Unit with suspicion of new or recurrent cancer. None had features of a septic condition, none had fever, and only mild neutrophilia was present. One was suspected to be complex PID after initial MDTM discussion and one was suspected to have ovarian cancer, but there were uncertainties in diagnosis for the five women who proceeded to core biopsy. Three were diagnosed based on IGCB and two from core biopsies which were taken by a gynecologists from sites of concern identified on MRI at the MDTM review.

Some clinical and some imaging aspects of these cases did not fit with the typical presentations of gynecological cancer. Only two cases had a raised CA-125 level. This is rare with ovarian malignancy, especially when it has spread to the peritoneum and this further raised concerns.

Imaging features which were "out of character" for malignancy were: (i) Invasion of the pelvic side wall musculature or necrotic sidewall lymphadenopathy related to an adnexal mass, features more associated with an advanced primary cervical cancer (Fig. 4); (ii) a liver surface lesion without ascites and intervening omental cake that would be expected with typical spread of primary ovarian cancer (Fig. 7); (iii) invasion of the adnexal mass into the mesorectum or pararectal spaces, compartments usually respected by untreated ovarian cancer; and (v) cavitating omental masses in a patient who had not undergone treatment (Fig. 6). These discordant imaging features (Table 2) prompted the need for a firm histological diagnosis prior to treatment.

With a firm histological diagnosis, the primary treatment plan was medical/interventional and thus five of the seven women avoided unnecessary cancer surgery. One woman had percutaneous placement of a ureteric stent and another had CT-guided insertion of

a pelvic drain early in their treatment plan. However, two women later required surgery for associated complications, including bowel obstruction and unremitting pain with radiologically inaccessible and multiloculated abscess formation. Thus, actinomycosis, even when diagnosed quickly and accurately, further mimics advanced gynecological malignancy in its protracted and complex clinical course and its requirement for multidisciplinary care.

The diagnosis is highly likely when there are some of the key imaging features we have outlined and illustrated and when core biopsies show a mixture of acute and chronic inflammation and absence of malignant cells or granulomas which might suggest tuberculosis, a condition also recognized to mimic local and disseminated gynecological malignancy. There may be histological mimics of actinomycosis. Pseudo-actinomycotic radiate granules are a recognized mimic of sulfur granules[9]; these are non-infectious and lack a distinct central core with broad, club-like peripheral projections replacing the filamentous projections of actinomyces. Conversely, the histological diagnosis of actinomycosis is often difficult because many specimens contain only a few granules. In one series, only a single granule was identified in 25% of samples. Granules may not be detected in histology samples in cases of culture-proven actinomycosis.[10,11] For two of our seven women, we did not identify sulfur granules nor obtain positive cultures, but their clinical response to antibiotic therapy was dramatic.

A variety of imaging features of pelvic actinomycosis have been previously described and our findings largely confirm these.[12-15] For our series, it was the combination of clinical concern for the diagnosis combined with some unusual imaging features (Tables 1 and 2) which prompted core biopsy. We have previously shown that IGCB is a valuable tool in the investigation of peritoneal disease in women suspected to have gynecological malignancy.[16,17] A small minority have non-malignant disease including actinomycosis and tuberculosis and malignancies with more favorable prognosis like lymphoma.[17]

CONCLUSION

Pelvic actinomycosis has a variety of imaging patterns which can mimic locally advanced gynecological malignancy or its local, regional, and distant metastatic spread. Diagnosis can, however, be achieved prospectively after MDTM review and using IGCB, when clinical and imaging features are atypical for primary gynecological cancer (Tables 1 and 2).

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ORIGINAL ARTICLE

Scope and Feasibility of Student-friendly Environmental Sustainability Measures in Educational Institutes: Our Experience in MGM Institute of Health Sciences, Navi Mumbai Campus, Maharashtra, India

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ABSTRACT

Introduction: Environmental sustainability is an important component for transforming higher education for a sustainable tomorrow hence, there is a pressing need to step up initiatives for environmental sustainability. Institutes of higher education play a key role in imparting environmental awareness and in employing eco-friendly measures. While institutes have several academic and administrative audits, environmental audit is comparatively neglected. Environmental sustainability includes protecting and restoring ecological systems, optimal utilization of resources, enhancing the well-being of all people, and motivating students to take up environmental educational projects as part of their curriculum.

Objectives: To evaluate environmental initiatives currently being practiced for environmental sustainability in MGM Institute of Health Sciences (MGMIHS) and to assess the scope and feasibility of their implementation and evaluation in educational institutes in a developing country.

Materials and methods: The key components of eco-friendly initiatives include carbon footprint reduction, water management, waste recycling, sustainable buildings, energy conservation, and environmental hygiene. Methods to achieve environmental sustainability also include using academic resources, such as faculty and students for campus education initiatives in order to produce awareness, motivation, and behavioral change. These methods are studied and assessed with a view to implementation in our local setting. In addition, environmental indicators for auditing the beneficial effects of these methods are examined.

Results: A campus score measuring physical qualities of university campuses like Urbanism, Greenness, and On-Campus Living is suggested to be included in accreditation process like National Assessment and Accreditation Council (NAAC). Various student- and faculty-friendly environmental initiatives were taken by MGMIHS to make the campus eco-friendly. At policy level the Institute became committed to a plastic bag-free zone. Pooling of cars and vehicles was promoted. There was a felt need to test the feasibility of implementing a completely paperless system of functioning for university and constituent institutions. The recently introduced enterprise resource planning (ERP) software can be considered as a starting point for going paperless. Energy conservation by using renewable energy in order to reduce carbon footprints was promoted along with water conservation efforts. Increasing the green cover of MGMIHS university campuses was given highest priority. Advocacy, communication, and social mobilization for environmental awareness was achieved by arranging regular lectures for motivation of students and faculties to maintain eco-friendly campus. Constitution of a Campus Environmental and Sanitation Committee was a step in the right direction.

Conclusion: This article discusses the measures that can be easily and effectively adopted in educational institutes in a developing country for contributing toward environmental preservation. In addition the article discusses the importance of auditing environmental sustainability in educational institutes using various indicators like the campus index. United Nations Environmental Programme (UNEP) Greening Universities Toolkit in 2013 can be a starting point to make MGMIHS more environmental friendly.

Keywords: Environmental audit, Environmental indicators, Environmental sustainability, Student-friendly environmental initiatives.

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INTRODUCTION

Environmental sustainability is an important component for transforming higher education for a sustainable tomorrow. Beginning with the Stockholm Declaration of 1972, there has been a steady development of national and international sustainability declarations relevant to higher education.[1] There is, therefore, a pressing need to step up initiatives for environmental sustainability and include the same in the accreditation process. With the rise of nontraditional governance methods, the third

party audit has become a critical part of governing.[2] Institutes of higher education play a key role in imparting awareness about the problem and its solutions, and are in themselves centers of high population density and turnover which need to employ eco-friendly measures. Although institutes have several academic and administrative audits, environmental audit is comparatively neglected. Environmental sustainability includes protecting and restoring ecological systems, optimal utilization of resources, enhancing the well-being of all people, and motivating students to take up environmental educational projects as part of their curriculum.

OBJECTIVES

- To evaluate environmental initiatives currently being practiced for environmental sustainability in MGMIHS.
- To assess the scope and feasibility of environmental initiatives implementation and evaluation in educational institutes in a developing country.

MATERIALS AND METHODS

The key components of eco-friendly initiatives include carbon footprint reduction, water management, waste recycling, sustainable buildings, energy conservation, and environmental hygiene. The current initiatives undertaken at MGMIHS, Navi Mumbai Campus, are studied in our research.

Methods to achieve environmental sustainability also include using academic resources, such as faculty and students for campus education initiatives in order to produce awareness, motivation, and behavioral change. These methods are studied and assessed with a view to implementation in our local setting. In addition environmental indicators for auditing the beneficial effects of these methods are examined.

DISCUSSION

Environmental sustainability is an important component for transforming higher education for a sustainable tomorrow. Velazquez et al[3] define a sustainable university as "a higher educational institution, as a whole or as a part, that addresses, involves and promotes, on a regional or a global level, the minimization of negative environmental, economic, societal, and health effects generated in the use of their resources in order to fulfill its functions of teaching, research, outreach and partnership, and stewardship in ways to help society make the transition to sustainable lifestyles."

Various student- and faculty-friendly environmental initiatives were taken by MGMIHS university to make the campus eco-friendly. At policy level the Institute became committed to a plastic bag-free zone. It was also recommended that the ban should be strictly enforced and canteen/shop vendors should be sensitized regarding the same. Tobacco and alcohol-free zones were actively advocated. The university encouraged faculty and students to use pooling of cars and vehicles and to follow no vehicle days to reduce emission of toxic gases. Studies in certain international universities show that the most obvious barriers to environmental sustainability were financial and that leadership, incentive, and demand are required to move forward with improving sustainability at universities.[4]

An important environmental-friendly initiative is the move toward paperless functioning of Departments. A recently published study in IOSR Journal of Environmental Science, Toxicology and Food Technology[5] states that paperless campus is a new pedagogical approach where most of the pedagogic activities are dependent on electronic gadgets. Electronic books, an e-learning campus, digital libraries, computer-based learning, database management systems, video conferences, distance learning, smart card applications, web mail, teleconference, and web-based applications are common components of a paperless campus.

There was a felt need to test the feasibility of implementing a completely paperless system of functioning for MGMIHS university and constituent institutions in general and Internal Quality Assurance Cell in particular. Once achieved with the help of an electronic Management Information System (e-MIS), the initiative will go a long way for environmental conservation. The recently implemented ERP system at MGMIHS can be considered as a first step toward achieving a paperless system of functioning.

Sustainability issues in higher educational institutions have attracted increasing levels of attention from both the public and policy makers in recent decades.[6] For the purpose of energy conservation, use of renewable energy in order to reduce carbon footprints was promoted at MGMIHS. Solar energy utilization is being actively promoted in the campuses. Solar heaters were installed for water heating at hospital, college, and hostel. The campus buildings were constructed in such a way that ample natural light and ventilation is made available. This resulted in the conservation of electricity. Special awareness programs were carried out for the stakeholders to conserve energy. For achieving efficiency in energy consumption, process has been initiated to replace fluorescent tube lights and compact fluorescent lamp (CFL) bulbs with light-emitting diode (LED) lights only. Use of energy-efficient appliances is actively advocated. Use of four-star rating low energy consumption equipments/machinery including star-rated refrigerators and air

conditioners are promoted. Unit settings for all air conditioners are usually kept at 22 to 24°C to conserve power. Staff and students are inspired and encouraged to conserve energy, by placing reminders placards (switch off artificial lighting and unplug electrical and electronic devices) in various departments, lecture halls, and corridors.

An interesting research by Hajrasouliha[7] proposes an index, called Campus Score, which measures the main physical qualities of university campuses. Campus Score is composed of three latent variables representing Urbanism, Greenness, and On-Campus Living, with 10 indicators. This index has been calculated for 103 research-intensive universities in the United States. The study states that physical campus characteristics can impact student satisfaction, academic performance, and graduation rates. A similar score can be thought of for purpose of conducting environmental audit and can be included in accreditation process like NAAC.

Rational use of water can be a powerful tool to promote sustainability on university campuses. Other than resource and financial savings, it aims to support technological and behavior innovation toward a more balanced relationship between human activities and nature.[8] In order to promote water conservation, an artificial pond with check dam is constructed in the MGMIHS campus and water from the same is used for gardening purpose. Sewage treatment plant is installed for treatment of waste water and this treated water is reused for gardening and plantation purpose at the university campus.

Carbon neutrality is promoted at MGMIHS by sending laboratory waste to an outsourced authorized biological waste disposal agency. The biodegradable wastes are used for compost making and for in-house gardening purpose. Recycling of waste paper is promoted. The MGMIHS can take ideas from universities in Malaysia which have been working toward implementing Green Initiatives as outlined in the UNEP Greening Universities Toolkit in 2013 to become sustainable Green Campuses.[9]

Increasing the green cover of MGMIHS university campuses is given highest priority. For the purpose of promoting this MGMIHS conducts a green audit of its campuses. Tree plantation drives on a periodic basis are conducted by the university to keep the campus green and to provide fresh oxygen; this also helps to sequester carbon dioxide emitted in the atmosphere. Garden, lawn, and plant nurseries in the campus are well maintained. Census of plants and trees in campus is maintained.

Advocacy, Communication, and Social Mobilization (ACSM) for environmental awareness was achieved at MGMIHS by arranging regular lectures for motivation of students and faculties to maintain eco-friendly campus. Awareness talks regarding Swachata Abhiyan and Swachata Mobile Application (developed by the Ministry of Urban Development) was conducted for stakeholders like Dean, Medical Superintendent, and Students.

Inspection and supervision of campus and facilities was done within the institution by Head of Institution and site office in-charge on a regular basis. A special Health Inspector was appointed under Department of Community Medicine, Navi Mumbai who with the guidance of Dean, Medical Superintendent, and Professor and Head of the Department (Community Medicine) contributed to environmental protection steps. Regular water surveillance and testing for bacteriological examination is done to promote water quality. Biomedical waste management rules are strictly followed as part of environmental sustainability measures. Supervision over disposal of condemned materials for all building and structures in order to prevent fire hazard and checking for mosquito breeding sites and taking appropriate preventive measures are given priority. The constitution of a Campus Environmental and Sanitation Committee headed by the Medical Superintendent of the hospital was a move toward significantly improving the environmental sustainability of MGM teaching hospital. As a future initiative, MGMIHS can go for environmental audit and implement ISO 14001 environmental standards.

RECOMMENDATIONS

Environmental sustainability measures like plastic-free zones, car pooling, tobacco/alcohol-free zones, paperless administration, energy conservation, water conservation, measures for ensuring carbon neutrality, increasing green cover with tree plantation, biomedical waste management, e-waste management, formation of campus environmental and sanitation committee, and ACSM activities for promoting environmental sustainability can be easily and effectively adopted in educational institutes in a developing country for contributing toward environmental preservation. Further auditing of environmental sustainability is of vital importance in educational institutes using various indicators like number of departments implementing e-MIS and promoting paperless functioning. For energy conservation, indicators, such as percentage of CFL lights/bulbs replaced by energy-efficient LED lights and percentage of equipments having either four star or five star energy ratings can be implemented. In the area of water conservation, one can use indicators like percentage of buildings having rain water harvesting systems in place and facilities for reusing Sewage Treatment Plant treated water for nondrinking purposes. For the purpose of increasing green cover, indicators like percentage of land area which is designated as open green places and periodicity of conducting tree audit can be developed. Policy-level indicators which could be developed can be Adoption of Policy level initiatives like polythene-free

zones, tobacco/alcohol-free zone, and car pools. The ACSM indicators for environmental sustainability could include a number of sensitization meetings, lectures, and training sessions done for promoting environmental awareness. Once these environmental indicators are developed, the institution needs to formulate a data collection and surveillance system to know the progress made in these areas. Periodic environmental audit can then be conducted. Campus Score, which measures the main physical qualities of universities like Urbanism, Greenness, and On-Campus Living, can be implemented at MGMIHS. The time is also right for implementing ISO 14001 environmental standards so that MGMIHS can show the way to other universities of developing countries that Environmental Sustainability can become a reality and not just remain an utopian dream.

CONCLUSION

Environmental sustainability measures can be easily and effectively adopted in educational institutes in a developing country for contributing toward environmental preservation. Further auditing of environmental sustainability is of vital importance in educational institutes using various environmental indicators. Once these environmental indicators are developed, the institution needs to formulate a data collection and surveillance system to know the progress made in these areas. Periodic environmental audit can then be conducted based on these parameters. Once found effective, environmental indicators and environmental audit as a process can be suggested to NAAC for inclusion as a criteria for assessment, appraisal, and as part of their Annual Quality Assurance Reporting System.

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ORIGINAL ARTICLE

Intralesional Platelet-rich Plasma Therapy *vs* Intralesional Triamcinolone Acetonide for the Treatment of Alopecia Areata

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ABSTRACT

Introduction: Alopecia areata (AA) is a common autoimmune condition causing hair loss. Platelet-rich plasma (PRP) has emerged as a new treatment modality in dermatology, and preliminary evidence has suggested that it might have a beneficial role in hair growth.

Aims and objectives: To compare the efficacy of intralesional PRP therapy *vs* intralesional triamcinolone acetonide (TrA) for the treatment of AA in a randomized, single-blinded, placebo- and active-controlled parallel study.

Materials and methods: Thirty AA patients were randomized into two groups to receive intralesional injections of PRP or TrA. Half the number of patches in each patient were treated and other half were injected with placebo (normal saline). A total of three treatments were given for each patient at one month intervals. The evaluation was done by severity of alopecia tool (SALT) scoring, dermoscopic evaluation of dystrophic hair, and patient satisfaction. Patients were followed up for 6 months.

Observations and results: Intralesional TrA was found to give better results as compared with intralesional PRP.

Conclusion: Intralesional TrA gives superior results in AA as compared with intralesional PRP.

Keywords: Alopecia areata, Intralesional triamcinolone acetonide, Platelet-rich plasma therapy.

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INTRODUCTION

Alopecia areata (AA) does not destroy hair follicles, and therefore, the potential for regrowth of hair is retained for many years, and is possibly lifelong.[1] A variety of therapeutic options are available. Search for new modalities continues as there is a high relapse rate.

Intralesional corticosteroids are the most popular drugs available for the treatment of AA.[2] But localized atrophy is a common complication, particularly if TrA is used.[1] Other therapies like topical minoxidil, anthralin, immunotherapy, topical/systemic corticosteroids, cyclosporine, and Psoralen and Ultra Violet-A light therapy are commonly used with varying success.

Platelet-rich plasma (PRP) is an autologous concentration of platelets in concentrated plasma.[3] Recently, there have been a few studies investigating the clinical results of PRP applications as treatment for AA. It is hypothesized that growth factors released from platelets may act on stem cells in the bulge area of the follicles, stimulating the development of new follicles and promoting neovascularization in cases of AA.[4]

So, PRP is a potential useful therapeutic tool for alopecias, without major adverse effects. A study has been carried out to evaluate the effectiveness of intralesional PRP in cases of AA and compare its efficacy with the most popular treatment option, i.e., intralesional TrA.

AIMS AND OBJECTIVES

- To compare the efficacy and safety of intralesional PRP *vs* intralesional TrA for the treatment of stable scalp AA.
- To compare the overall patient satisfaction in the two treatment groups.

MATERIALS AND METHODS

Ethical Considerations

The study design was approved by the Institutional Ethics Review Committee. Written informed consent was obtained from each subject prior to enrollment in the study.

Study Area

The study was conducted in the outpatient department (OPD) of a tertiary care center situated in Navi Mumbai, in the periphery of Mumbai, the capital of Maharashtra state in India.

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Duration

The study was conducted over a period of 22 months from February 2014 to November 2015.

Inclusion Criteria

- Patients with patches of AA over scalp
- Patients with stable AA—patient denying history of increase in patch size or of development of new patches and a negative hair pull test
- Patients with a minimum of two patches
- Patients with a normal platelet count (>1.5–4.5 lakh/cu.mm)
- Patients willing to be part of the study after informed consent

Exclusion Criteria

- Patients below 15 years of age
- Patients with active AA
- Patients with alopecia totalis/universalis
- Patients who have been treated with topical agents in the past 15 days or intralesional/systemic agents in the past 1 month
- Pregnant and lactating women
- Unwilling patients

Patients

- A sample size of 30 patients fulfilling the inclusion and exclusion criteria was taken in the study.
- Patients were selected from Dermatology OPD, MGM Hospital, Kamothe, Navi Mumbai, Maharashtra, India.
- A written informed consent was taken from all selected patients.
- Patients were explained the procedure, possible side effects, and postprocedure care.
- Information sheet was given to all patients. It was explained to them that they can drop out of the study at any given point of time for any reason. It will not affect their further management in any way.
- All patients were photographed prior to starting the study and at last follow-up visit.

Preparation of PRP

- All patients were sent to the blood bank located within MGM Hospital for preparation of PRP.
- After taking informed consent, patients' 20 cc blood was obtained by venipuncture in two tubes containing 0.9 mL of anticoagulant citrate dextrose A.
- After blood collection, the tubes were made to stand vertically for half an hour.
- The citrated blood was then centrifuged in Heraeus Cryofuge 5500i centrifuge at 1000 rpm for 12 minutes (soft spin) at 22°C.
- After the first step, each tube gave 3 mL of plasma (containing buffy coat with platelets and leukocytes).
- Subsequently, this 6 mL plasma was taken up from both the tubes using a sterile micropipette and was transferred to another sterile tube.
- A second round of centrifugation was performed at 2000 rpm for 10 minutes (hard spin) at 22°C.
- The pellet containing platelets accumulated at the bottom and the platelet-poor plasma surfaced to the top. The upper 2 mL was discarded and the lower 4 mL was used to resuspend the platelets.
- The suspended pellet in 4 mL of plasma was used as PRP.
- The platelet count was then checked to ensure adequate amount of platelets in PRP.

Procedure

- Randomization of the patients was done by using random number table and it was known only to the one doing the procedure.
- Patients were divided into two groups of 15 patients each.
- One group received intralesional PRP and the other group received intralesional TrA.
- Half of the patches in each patient were treated, whereas the other half were given placebo (normal saline). In case of odd number of patches, more number of patches were treated and less number were given placebo.
- The alopecia patches in each patient were randomized to receive intralesional PRP or intralesional TrA.
- Three monthly therapies were given.
- New patches developing during the course of the study were treated, but not included in the study evaluation.

Procedure for Injecting

- Area was cleaned by betadine solution and normal saline.
- 0.1ml of PRP/intralesional TrA/normal saline was injected into the dermis of affected patches at multiple points 1 cm apart.
- Treated site was again cleaned and left open.
- Patients were followed up for a total of 6 months.
- Evaluation was done at:
  - T0—1st visit (1st injection)
  - T1—2nd visit (2nd injection)
  - T2—3rd visit (3rd injection)
  - T3—4th visit (6 months after 1st injection)

Evaluation

Evaluation was done by a non-treating dermatologist at every visit.

Table 1: Age and sex distribution

	PRP				ILS				
	Male		Female		Male		Female		
Age	n	%	n	%	n	%	n	%	Total
Below 20	2	13	1	7	3	20	0	0	6
21–30	3	20	2	13	4	27	2	13	11
31–40	3	20	2	13	3	20	1	7	9
41–50	0	0	1	7	0	0	2	13	3
51–60	0	0	1	7	0	0	0	0	1
Total	8	53	7	47	10	67	5	33	30

Severity of Alopecia Tool Scoring

The proportion of scalp involvement is determined by dividing the scalp into five regions and estimating the percentage of the scalp surface that all the alopecic area would occupy if placed together. These five were named as regions:

1. S1: <25% hair loss
2. S2: 25 to 49% hair loss
3. S3: 50 to 74% hair loss
4. S4: 75 to 99% hair loss
   a. A: 75 to 95% hair loss
   b. B: 96 to 99% hair loss
5. S5: 100% hair loss

Severity of alopecia tool scoring was done at every visit.

Body Hair Loss

- B0: No body hair loss
- B1: Some body hair loss
- B2: 100% body hair loss (excluding scalp)

This involved a complete physical examination and included facial, axillary, truncal, genital, and extremity hair evaluation.

Nail Involvement

- No: No nail involvement
- N1: Some nail involvement
- N2: Twenty nail dystrophy/trachyonychia (i.e., all 20 nails)

Nail assessment was done at every visit to look for any improvement, worsening, or no change.

Dermoscopic Evaluation

It was done by a non-treating dermatologist. Evaluation was intended to determine the number of dystrophic hair in the patch. Markers for dystrophic hair included exclamation mark hair, black dots, yellow dots, coudability hair. The percentage of dystrophic hair was calculated on a 4-point scale:

1. 3: >50%
2. 2: 30 to 49%
3. 1: 1 to 29%
4. 0: no dystrophic hair

Patient Satisfaction

Based on the improvement perceived by the patients, they were asked to rate the improvement in hair growth on a 10-point scale:

- 7 and above: excellent response
- Between 4 and 6: good response
- 3 or less than 4: poor response

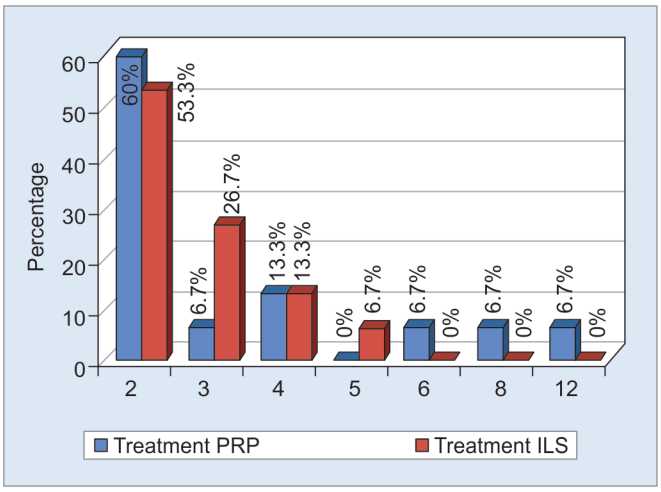
Data analysis was done by using statistical tests, such as unpaired t-test, paired t-test and chi-square test. Level of significance was set at 0.05. All p-values <0.05 were treated as significant. Statistical Package for the Social Sciences version 17.0 was used for data analysis.

OBSERVATION AND RESULTS

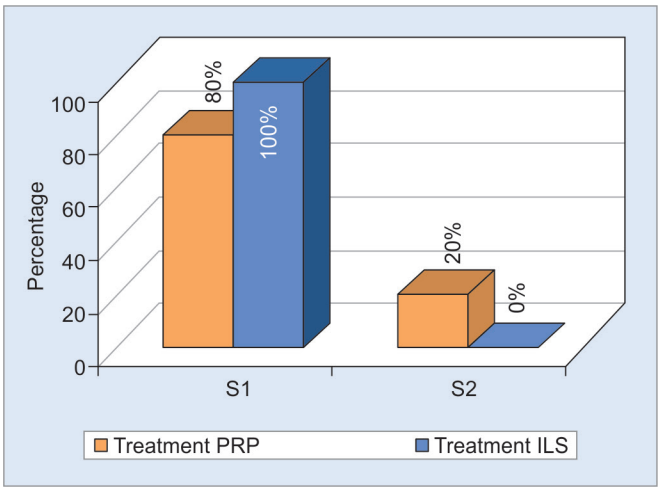
Out of the total 30 patients, 27 patients completed the study and 3 were dropouts. Assessment for hair regrowth was done by a blinded observer using SALT score and by another blinded observer for dermoscopic improvement. Also, subjective evaluation was done by the patient. Analysis of data was done by statistical tests mentioned above. Age and sex distribution has been depicted in Table 1. The disease duration of the patients enrolled in the study varied from less than 1 month to more than 6 months, depicted in Table 2. The number of patches of alopecia areata in the patients enrolled in the study varied from two to twelve, depicted in Graph 1. The extent of hair loss in patients, calculated by SALT score is depicted in Graph 2. Some patients gave history of similar episodes of patchy hair loss in the past, depicted in Table 3.

Table 2: Total disease duration of patients enrolled in the study

Duration	PRP (%)		ILS (%)		Total
≤1 month	2	13.3	4	26.7	6
1–6 months	10	66.7	11	73.3	21
>6 months	3	20.0	0	0.0	3
Total	15	100.0	15	100.0	30



Graph 1: Number of patches in all patients enrolled in the study



Graph 2: Extent of hair loss calculated using the SALT score

Table 3: Total number of prior episodes in each group of patients

		PRP		*ILS*		
		n	*%*	*n*	*%*	*Total*
Number of prior episodes	None	14	93.3	11	73.3	25
	One	1	6.7	2	13.3	3
	Two	0	0.0	1	6.7	1
	Multiple	0	0.0	1	6.7	1
	Total	15	100.0	15	100.0	30

Association of Infection with Onset of AA

No such association was seen in the study as none of the patients gave any history of, or symptoms suggestive of, any infection prior to onset of hair loss.

Family History

Only one patient enrolled in the study, from the ILS group, gave family history of AA.

Nail Involvement

Three (10%) patients had nail involvement which was in the form of pitting of nail plate. Of these, 1 was from the ILS group and two were from the PRP group.

Body Hair Involvement

Two (6.7%) patients had body hair involvement, one patient in each group.

Evaluation of Hair Regrowth by SALT Scoring

Thirteen (43.33%) patients had complete hair regrowth, of which 3 (20%) were from the PRP group and 10 (66.67%) were from the ILS group.

The association between treatment given and response was tested using chi-square test for association. The result indicates that there is significant difference in the proportion of responses between intralesional TrA and intralesional PRP therapy

Table 4: Dermoscopic improvement (active)

	PRP				*ILS*			
	Before		*After*		*Before*		*After*	
	n	*%*	*n*	*%*	*n*	*%*	*n*	*%*
G0	2	15.4	7	53.8	1	7.1	11	78.6
G1	6	46.2	6	46.2	7	50.0	1	7.1
G2	1	7.7	0	0.0	5	35.7	2	14.3
G3	4	30.8	0	0.0	1	7.1	0	0.0
Total	13	100.0	13	100.0	14	100.0	14	100.0

Table 5: Dermoscopic improvement (placebo)

	PRP				*ILS*			
	Before		*After*		*Before*		*After*	
	n	*%*	*n*	*%*	*n*	*%*	*n*	*%*
G0	3	23.1	9	69.2	3	21.4	13	92.9
G1	7	53.8	4	30.8	8	57.1	1	7.1
G2	1	7.7	0	0.0	1	7.1	0	0.0
G3	2	15.4	0	0.0	2	14.3	0	0.0
Total	13	100.0	13	100.0	14	100.0	14	100.0

Table 6: Patient satisfaction score

		Patient satisfaction score					
		0%	*10%*	*30%*	*60%*	*100%*	*Total*
Treatment	PRP	8	2	0	1	2	13
	ILS	1	1	1	0	11	14
Total	9	3	1	1	13	27	

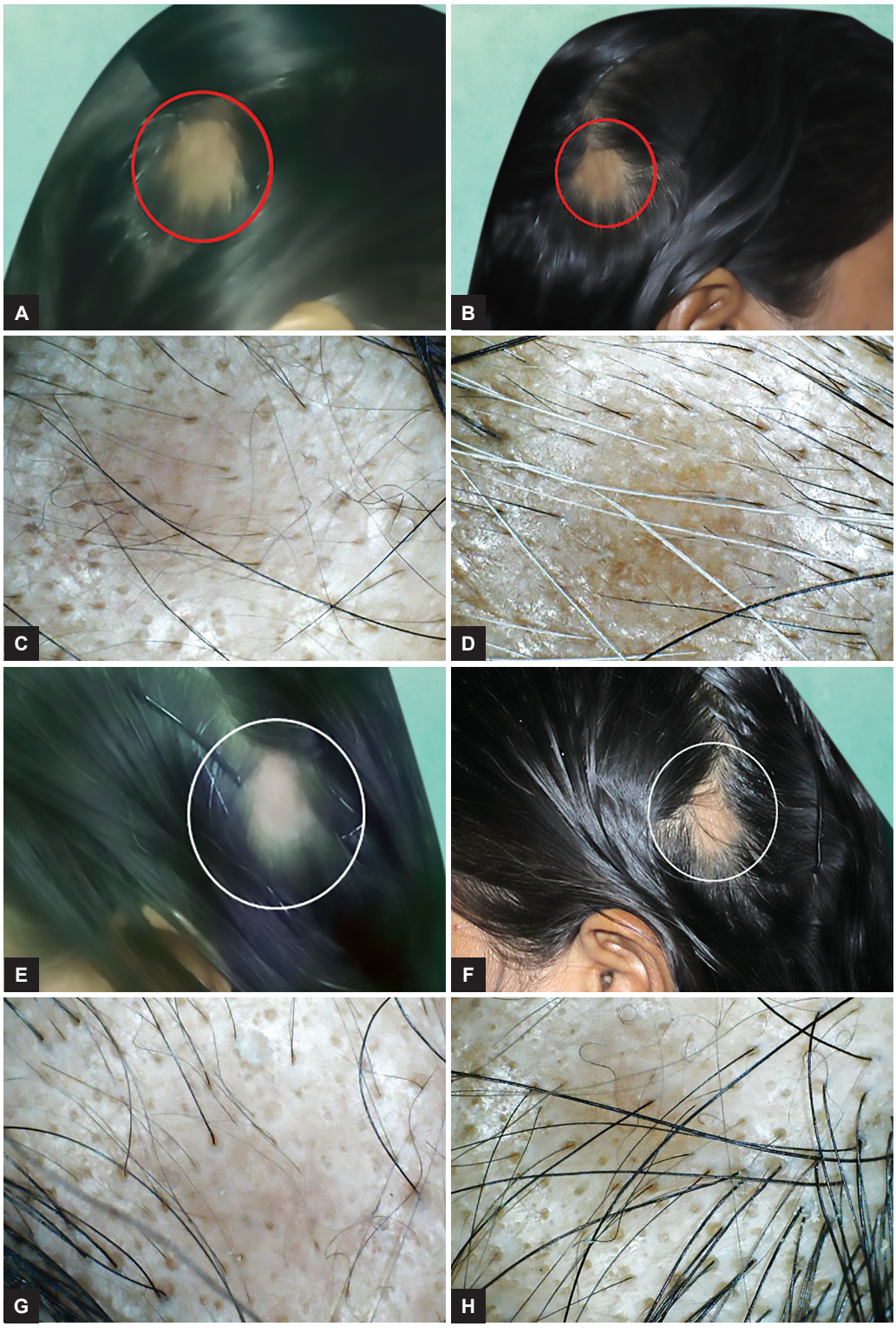
Chi-square = 13.897, $p < 0.05$, significant

Dermoscopic improvement in active and placebo patches are depicted in Tables 4 and 5 respectively.

Side effects: None of the patients enrolled in the study showed any adverse effect to therapy.

Patient satisfaction: As depicted in Table 6, 2 (13.33%) patients from PRP group were 100% satisfied, whereas 11 (73.33%) patients from the ILS group were 100% satisfied with the therapy.

The association between treatment given and patient satisfaction was tested using Pearson's chi-square test.

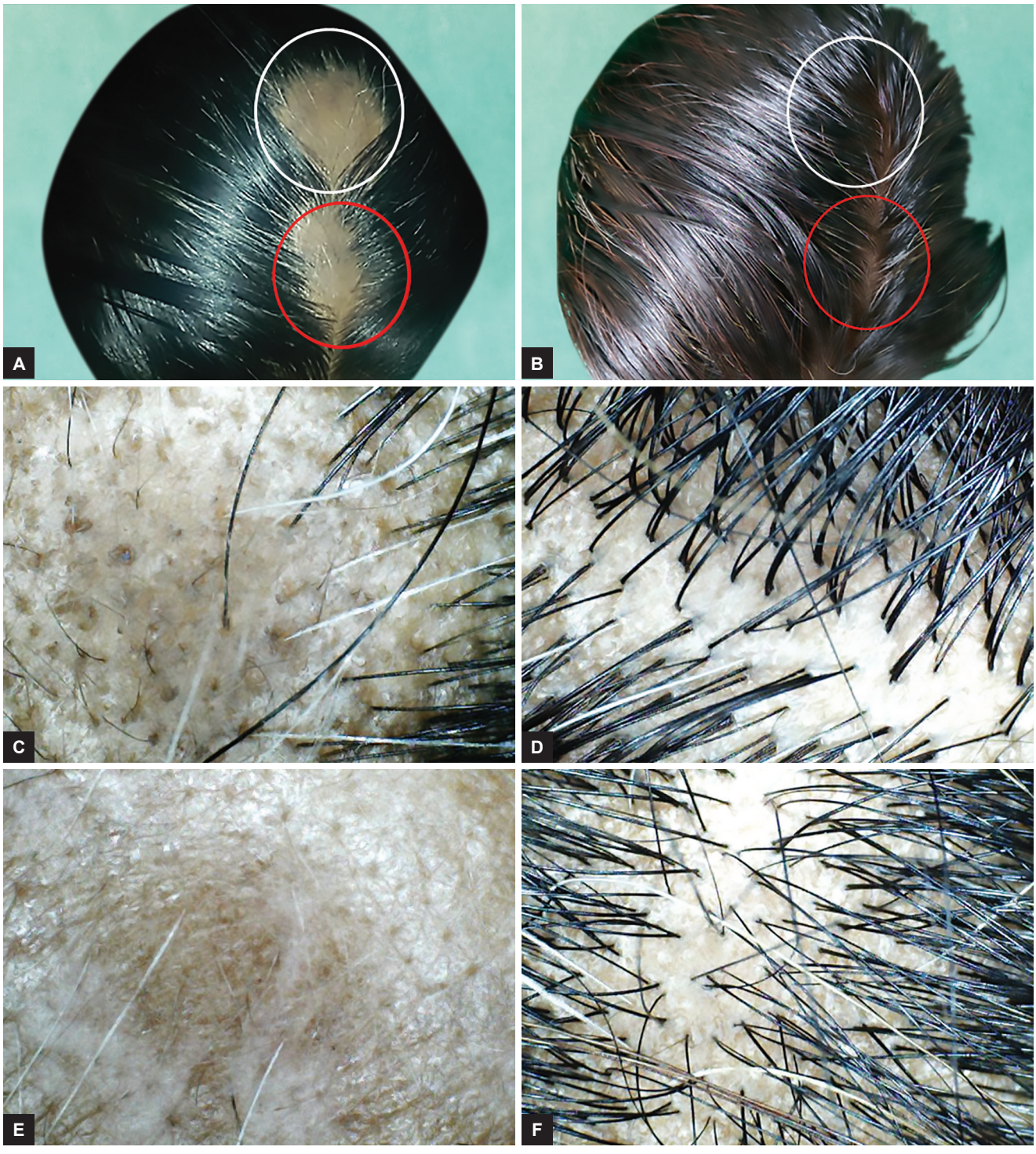


Figs 1A to H: Patient on PRP. 51 years/F, presented with two bald patches on the scalp since 15 months. No similar complaints in past. No body hair involvement. No nail involvement. No treatment taken in the past. No similar complaints in family members. H/o asthma in sister. Ix: WNL. Evaluation:

- SALT score: 0%
- Dermoscopic grading
  - Active patch G3 to G0
  - Placebo patch G3 to G1
- Patient satisfaction 60%

The result indicates that there is a significant difference in patient satisfaction between intralesional triamcinolone and intralesional PRP therapy.

Figures 1 and 2 compare two patients, one from each treatment group, highlighting the difference in treatment response.



Figs 2A to F: Patient on ILS. 42 years/F, presented with two bald patches on the scalp since 2 months. No similar complaints in past. No body hair involvement. No nail involvement. Topical herbal treatment taken in the past with partial improvement. No similar complaints in family members. H/o atopic dermatitis and vitiligo in daughter. Ix: Hb: 10.9; rest WNL. Evaluation:

- SALT score: 100%
- Dermoscopic grading
  - Active patch G1 to G0
  - Placebo patch G0 to G0
- Patient satisfaction 100%

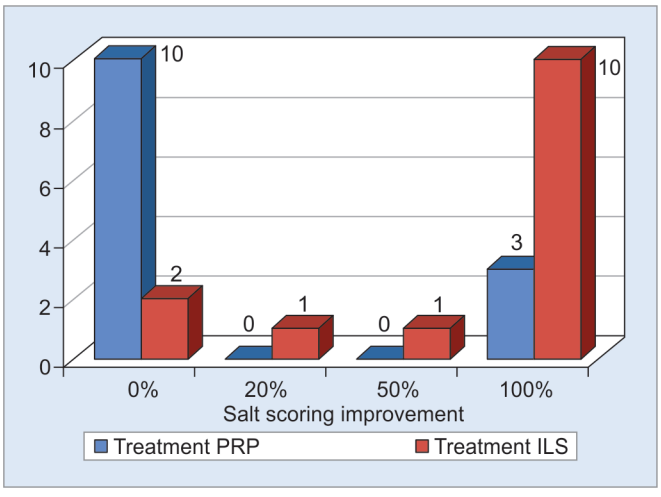
DISCUSSION

The practice of using PRP in dermatology is new and there are not many reports in the dermatology literature ascribing the benefits of this practice. It is therefore, subject to both overuse and over expectations, as well as over concerns and misunderstandings.[5] In 2013, Trink et al[6] performed a randomized, double-blind, placebo- and active-controlled, half-head study on 45 patients and

evaluated the efficacy of PRP in patients with AA. Both triamcinolone and PRP led to increased hair regrowth compared with the untreated side of the scalp. Additionally, patients treated with PRP had significantly increased hair regrowth compared with those treated with triamcinolone.

Taking this study into consideration, we formulated our study to compare the safety and efficacy of intralesional TrA vs intralesional PRP for the treatment of scalp AA. Very few similar studies have been published in literature so far. According to the assessment criteria used by us, i.e., SALT scoring, dermoscopic evaluation and patient satisfaction, significant proportion of difference (p-value <0.05) has been found between intralesional PRP therapy and intralesional TrA 5 mg/mL groups. The response was better in patients treated with intralesional TrA 5 mg/mL, with 10 out of 15 (66.67%) patients showing 100% hair regrowth by the end of 6 months in contrast to only 3 out of 15 (20%) patients showing complete hair regrowth in the intralesional PRP therapy group; 10 out of 15 (66.67%) patients showed no response with PRP therapy in contrast to only 2 (13.33%) patients showing no response with Intralesional TrA. This association between treatment given and improvement in SALT score is depicted in Graph 3.

On dermoscopic examination also, better result was seen in the intralesional TrA group with more number of patients showing reduced dystrophic hair at the end of the therapy; 11 out of 15 (78.6%) patients in the ILS group achieved grade 0, i.e., no dystrophic hair, by the end of therapy; 11 out of 15 (78.6%) patients in the intralesional TrA group achieving the same result. Comparable results were seen even in the placebo patches with 13 (92.9%) patients of intralesional TrA group achieving grade 0 by the end of therapy in contrast to only 9 (69.2%) patients achieving similar results in the PRP group.



Graph 3: Association between treatment given and improvement SALT score

The results of our study are in contrast to the study conducted by Trink et al[6] in which better results were seen in patients treated with intralesional PRP therapy. In another similar study done by Shumez et al,[7] better results were seen with intralesional PRP therapy but the results were statistically insignificant.

Surprisingly, it was also noticed that when treatment given led to hair regrowth in the active patch, placebo patches in which normal saline was injected also showed similar hair regrowth. This finding was consistent with both the therapies.

The overall patient satisfaction was better in the intralesional TrA group with 11 (73.3%) patients being 100% satisfied in contrast to only 2 (13.33%) patients of PRP group.

The PRP preparations are being extensively used in wound healing and tissue repair despite insufficient evidence. Blinded multicenter, randomized controlled studies are the need of the hour. There are no universally established standards for collection, quality control and administration of the product.

Transforming growth factor beta (TGF-β) and platelet-derived growth factor (PDGF) have been purported to be the most active contents in PRP for hair growth. So far, their biological actions are not fully known.[5] Mode of growth factor release and dynamics of the hair follicle environment may be important determinants of their outcome.

The TGF-β, in particular, is released in a latent form and requires activation for it to exert any biologic effects.[8] Whether latent TGF-β can be activated and, if activated, whether it is quantitatively adequate to produce a desired biologic effect, is still unknown. The PDGF can promote several biologic effects. The mitogenic actions of PDGF, as well as its expression by malignant cells, suggest that it may have a role in malignant transformation.[9]

CONCLUSION

The assessment criteria used in this study have shown significant proportion of difference (p-value < 0.05) between intralesional PRP and intralesional TrA 5 mg/mL for the treatment of scalp AA. This study showed that:

- Intralesional PRP therapy was not found to be efficacious standalone therapy for the treatment of scalp AA.
- Response of intralesional TrA 5 mg/mL for the treatment of scalp AA was found to be better as compared with that of intralesional PRP therapy, with 10 out of 15 (66.67%) patients showing 100% hair regrowth by

the end of 6 months in contrast to only 3 out of 15 (20%) patients showing complete hair regrowth in the intralesional PRP therapy group.

- Overall patient satisfaction was better in patients of intralesional TrA 5 mg/mL group as compared with intralesional PRP therapy.

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ORIGINAL ARTICLE

A Study of Factors affecting the Knowledge and Awareness about Effective Breastfeeding

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ABSTRACT

Aim: To study the knowledge and awareness about breastfeeding in women attending a tertiary care medical center, and correlate it with sociodemographic factors.

Materials and methods: A total of 331 participants were selected and interviewed with a prevalidated questionnaire. Their answers about essentiality and advantages of breastfeeding were evaluated as "wrong," "don't know," and "correct." Spearman's correlation coefficient graph was used to analyze the association of their answers with different sociodemographic factors.

Results: Over 80% participants had correct knowledge about early initiation of breastfeeding within 1 hour, exclusive breastfeeding for 6 months, and importance of colostrums in normally delivered babies; 58% showed lack of knowledge about hazards of prelacteals and initiation of breastfeeding after cesarean section. Advantages of breastfeeding for mothers were known to only 60 to 65% of participants. Our study did not establish any positive or negative correlation between age, parity, education, address, place of delivery, and information providers with knowledge about breastfeeding.

Conclusion: In this study, urban living, higher education, multiparity (i/v/o breastfeeding experience), and hospital delivery of women did not show positive correlation with higher degree of knowledge and awareness about breastfeeding, thus highlighting the need for specifically designed and focused training for the antenatal and postnatal women as well as the society.

Clinical significance: There is an urgent need for prompt and active action to be taken at health care system, community, and policy levels for strategic planning of different programs to improve effective breastfeeding rates. These plans should focus on clearing social myths and taboos and in accordance with local needs of the women.

Keywords: Awareness, Breastfeeding knowledge, Education relationship, Information provide.

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INTRODUCTION

Breastfeeding is a natural gift to every mammalian and it starts at the birth and continues for a species for a specified time. In 19th century, a higher infant mortality rate due to artificial feeding was a public health problem and it was addressed by encouraging breastfeeding and pasteurization of cow's milk. In early 20th century, commercial companies found a market for artificial formula feeds. As a result, by mid-20th century, breastfeeding trend declined and so entire generation of doctors and women grew without knowing or experiencing breastfeeding as a natural way to feed babies.

The World Health Assembly, in 2012, in its resolution 65.6 endorsed a Comprehensive Implementation Plan on maternal, infant, and young children nutrition which specified six global nutrition targets for 2025. The fifth target among them is: Increase rate of exclusive breastfeeding in the first 6 months of life up to 50%.[5] India's statistics in 2015, from the United Nations International Children's Emergency Fund[1] data, are showing early initiation of breastfeeding within 1 hour in 45% neonates and exclusive breastfeeding rate as 65%. The neonatal mortality rates are 28 per 1,000 live births and under 5 mortality rates between 45 and 50 per 1,000 live births which are significantly high. The World Health Organization (WHO)[2] has guided specifically the countries that are at or near 50% exclusive breastfeeding targets, to continue to strive for improvement, because of considerably high health and economic benefits of breastfeeding.

Inadequate rates of breastfeeding are due to poor knowledge about proper breastfeeding techniques and advantages of breastfeeding among women, their partners, families, health care providers, and policymakers.[2] We, as obstetricians, are important part of the WHO mission 2025. Because of counseling of mothers during pregnancy, immediately after child birth and during the postnatal visits, it has shown significant positive effects on improving the breastfeeding rates. So, we decided to undertake this research study to identify the basic knowledge level and barriers and facilitators for successful breastfeeding and thus we can use these data to

design contextualized intervention policies to improve breastfeeding rates among our society.

MATERIALS AND METHODS

Study type: Survey based correlational cross-sectional study.

Study period: 2015 to January 2016. Ethical committee approval was taken before conducting the study.

Study participants: Women attending the outpatient department (OPD) clinic of MGM Medical College and Hospital, Aurangabad, for various reasons.

Sample size: 331 women.

Participant selection: Participants were selected by convenient sampling after introducing the study to them and explaining to them the benefits of participating in this study.

Data collection: The participants, after written consent, were interviewed in a private room in the OPD campus of obstetrics/gynecology department for about 10 to 15 minutes with a prevalidated questionnaire. Participant's confidentiality was respected.

Questionnaire design: The questionnaire consisted of multiple-choice questions to gather data about sociodemographic variables like age, parity, educational status, residence, place of delivery, and information provider. The other part of the questionnaire included questions to test the essential knowledge about breastfeeding and its advantages. The answers were analyzed as "wrong," "don't know," and "correct." The themes of the questions were as follows:

1. Initiation of breastfeeding within ½ to 1 hour after child birth
2. Initiation of breastfeeding within 2 hours of cesarean section
3. Use of different prelacteals and its hazards
4. Health benefits of colostrums
5. Exclusive breastfeeding for first 6 months of life
6. Breastfeeding advantage in infants from recurrent infections
7. Breastfeeding health benefits for the mother
8. Breastfeeding in HIV-positive mothers
9. Contraception and breastfeeding
10. Breastfeeding benefits in protection from breast cancers

As the information gathered in question number 8 was of a specialized condition, it was omitted from the statistical analysis in this study. Though it was not a part of the study, researchers made an attempt to educate the participants about the correct knowledge regarding breastfeeding by counseling and providing them a list of correct answers for their reference.

Statistical Analysis

Spearman correlation coefficient is used to assess the strength and direction of association between two ranked variables. A perfect, monotonically increasing relationship between two variables would result in Spearman correlation coefficient of 1 while a perfect monotonically decreasing relationship would result in Spearman correlation coefficient of −1 (for no ties). In this article, the demographic parameters for subjects are divided into two levels (0 and 1), where 0 corresponds to a parameter value which would correspond to underprivileged subjects which would potentially lead to lack of knowledge about breastfeeding while 1 corresponds to a parameter value which would correspond to privileged subjects which would potentially lead to knowledge about breastfeeding. For example, education has two levels, where 0 corresponds to subjects having high school or lower education while 1 corresponds to subjects having at least college education. A new parameter called overall score is defined as sum of individual parameter values for every subject. Higher the overall score, more privileged the subject is. Once the overall score is calculated for each subject, the subjects are ranked according to the overall score. Their responses are assigned scores in the following manner: Wrong: 1, Don't know: 2, Correct: 3. Spearman correlation is used to assess if higher overall score or higher individual parameter values result in higher scores for the responses and *vice versa*. In other words, the goal is to see if more "privileged" subjects according to our definition are likely to give more correct answers on breastfeeding related questions. A higher correlation coefficient (>0.7) would indicate strong correlation between the privileged status and knowledge about breastfeeding, while a correlation coefficient close to 0 would indicate no relationship between the privileged status and knowledge about breastfeeding. The correlations with response score are analyzed for individual parameter scores as well as overall score.

RESULTS

The sociodemographic profile like age, education, parity, information provider, place of delivery, and address were considered; 18% of the participants were nulliparous and 13% were of age less than 20 years. Higher level of education was received by 64.9% of women participating in this study. Our hospital being a tertiary care center catering to a large area including rural and urban, along with 68.9% of urban women, 31% women were from rural areas. Majority of women (82%) had hospital deliveries, but the information providers were the nonmedical persons in 67.6% women (Table 1).

Table 1: Sociodemographic profile of participants

Name	*Category*	*Number*	*Percentage*
Age	<20	46	13.8
	>20	285	86.2
Parity	Nullipara	61	18.4
	Multipara	270	81.5
Address	Rural	103	31.1
	Urban	228	68.8
Education	Up to secondary	215	64.9
	Higher	116	35.0
Delivery place	Home	57	17.2
	Hospital	274	82.7
Information provider	Nonmedical	214	64.6
	Medical	117	35.34

Being a tertiary care center, the participants were belonging to rural areas and with less education also

Table 2: Essential knowledge about breastfeeding

Parameters analyzed	*Correct*	*Wrong*	*Don't know*
Initiation of breastfeeding after vaginal delivery	266 (80.4%)	42 (12.7%)	23 (6.9%)
Initiation of breastfeeding after LSCS	81 (24.5%)	182 (55%)	68 (20.5%)
Prelacteals (hazards)	124 (37.5%)	13 (3.9%)	194 (58.6%)
Importance of colostrums	301 (90.9%)	6 (1.8%)	24 (7.3%)
Exclusive breastfeeding for 6 months	295 (89.1%)	17 (5.1%)	18 (5.4%)

Awareness regarding essential knowledge is satisfactory, but far less than the goal of 100%. Wrong concepts are prevailing about initiation of breastfeeding after cesarean section; LSCS: Lower segment cesarean section

Table 3: Advantages of breastfeeding

Parameters analyzed	*Correct*	*Wrong*	*Don't know*
Protection of baby from illness	256 (77.3%)	33 (10%)	42 (12.7%)
Maintenance of figure	213 (64.4%)	44 (13.3%)	74 (22.4%)
Protection from breast cancer	190 (57.4%)	38 (11.5%)	103 (31.1%)
Contraception and breastfeeding	217 (65.6%)	51 (15.4%)	63 (19%)

Maternal advantages of breastfeeding are less known to women as compared with advantages for baby

Fairly correct knowledge (80–90%) was observed about the facts like early initiation of breastfeeding, exclusive breastfeeding, and importance of colostrums. Around 55 to 65% women were unaware about the maternal benefits of breastfeeding and 67.9% even do not have knowledge that breastfeeding protects the baby from infections through active and passive immunity. There is lot of confusion whether to give prelacteals or not and 58% agreed that they do not know the facts about hazards of giving prelacteals. Wrong concepts were predominant that is around 54%, regarding initiation of breastfeeding after caesarean section which is one of the leading causes of breastfeeding failures in patients delivered by cesarean sections (Tables 2 and 3).

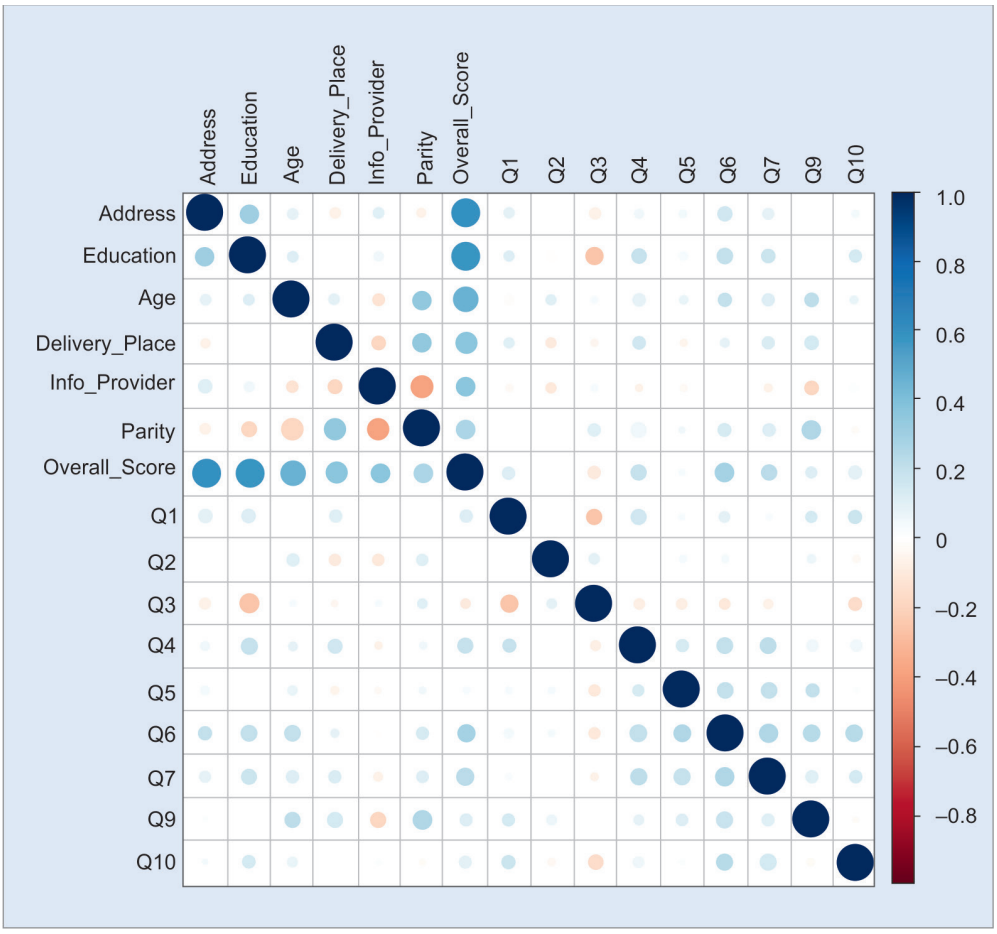
Spearman's correlation coefficient (Graph 1) is almost 0 with variables like address and information provider, thus denying any association between them and knowledge about breastfeeding. In case of variables like parity and contraceptive benefits, the correlation coefficient is shifted toward positive association but still not significant and commentable showing poor association. Education and use of prelacteals are negatively related to the knowledge as shown by Spearman's coefficient of –0.04. Even if we consider the overall combined scores, there is no strong association between knowledge of breastfeeding with all the six variables.

DISCUSSION

Previously published world data show that there is wide variation (50–80%) in essential knowledge about breastfeeding in the public. Early initiation of breastfeeding within 1 hour of birth which is the first important step toward successful breastfeeding is known to 53% in a study by Noor et al,[3] as compared with 80.3% participants in this study. This difference may be because the study conducted by Dr Sofia Noor had participants from slum areas which are unserved and underserved pockets in urban areas. Similar variation is found in knowledge about duration of exclusive breastfeeding for 6 months: 16.9% in a study by Syed et al[4] from UAE, 28% by Noor et al,[3] and 88.8% in our study.

There is good awareness about importance of colostrum in the society, i.e., up to 80 to 90% (82%,[5] 99%,[6] 83.8%,[7] and 88% in our study). The knowledge about whether to give prelacteals or not and hazards of giving prelacteals are not known to 55% women from our study. Similar study carried out by Jha et al[7] in Kakatia Medical College, Warangal Telangana, India, found that there is positive association between level of formal education and not giving prelacteals. Breastfeeding protecting the baby from infection is known to 67% women from our study. Also 60 to 65% are aware about its protective cover in preventing breast cancer and its action as a contraceptive. Similar results are observed in a few other studies across the world.[4,8,9]

Family members and friends happen to be first information providers about breastfeeding. Younger women usually consult the senior female members in the family and thus the information providers are mainly nonmedical persons. This fact is observed by Mogre et al[10] from Africa, Syed et al[4] from UAE, and even in our study, i.e., 67.6% women agreed that their mothers, grandmothers, and other relatives have been the information providers



Graph 1: Correlation between tested variables. No strong positive or negative correlation is seen between any of the variables tested

for them. Mogre et al[10] have therefore, suggested that there should be active teaching learning programs conducted by the medical and paramedical workers for this important group of first information providers who are consulted by prospective young mothers about breastfeeding.

To dissipate correct knowledge in the society, the elder generations also need to be included. This fact has been strengthened by a study done by Pandey et al,[9] when they studied awareness about breastfeeding in two generations of Indian women. When we considered age and parity as variables to correlate with the correct answers, the Spearman's coefficient was very weak. In contrast to this, Obilade et al,[11] who conducted a study in two semi-urban areas of Lagos state of Nigeria, found that the number of children and educational levels were statistically significant among the study populations who feel that breastfeeding is essential for the baby.

It is expected that previous experience with breastfeeding in multiparas should improve their knowledge and attitude. Mogre et al[10] have found that the likelihood of breastfeeding the child improves with increase in parity, but the Spearman's correlation coefficient in our study is showing neither any positive nor any negative association.

Formal education always improves the knowledge levels and the women are exposed to a wide range of communication systems, information access, and literature. This fact is revealed in many studies.[5,12] On the contrary, attitude toward breastfeeding may worsen with literacy as stated by Tan KL[8] in the study, "Knowledge, Attitude and Practice of Breastfeeding in Klang, Malaysia" as women then search for different alternatives and options. Acharya et al[12] from Nepal studied the effect of educational status on early initiation of breastfeeding by analysis of three consecutive (2001, 2006, and 2011) demographic and health surveys, and found a significant correlation between them. But the authors have suggested that improving educational levels is a long-term goal and we should not wait till then. We should start searching for alternative educational and supportive initiatives like prenatal education. In our study, we could not find such association between formal education and knowledge.

Haroon et al,[13] in their systematic review, have concluded that special breastfeeding education and/or support increase(s) exclusive breastfeeding rates and decreases no breastfeeding rates at birth, less than 1 year, and 1 to 5 months. This indicates that only formal educa-

tion is not going to improve the knowledge and attitude toward breastfeeding.

A wide range of variation is observed when we compare different variables like age, parity, education, place of delivery, and information provider with breastfeeding knowledge across the world, because in different communities, different myths exist around breastfeeding. The status of women and their roles in different fields are also varied across the world and so are their needs. The social taboos attached with breastfeeding need to be addressed individually and also education and awareness must be imparted to community as a whole. The literacy rates will definitely affect the results of our efforts to achieve the WHO global goals, but simultaneous efforts will be needed at personal levels for the women, their partners, and other family members.

LIMITATIONS OF THE STUDY

The cross-sectional nature of this study makes it difficult to establish causality and can interpret only the association. As the study was health center-based mainly including the antenatal and postnatal women, it may not be representative of the situation in the entire community. Thus, the nature of the study is just a snapshot. It is therefore, imperative to conduct a longitudinal and community-based study to explore the knowledge about breastfeeding practices.

CONCLUSION

This study indicates that so-called "privileged" women according to our definition do not necessarily have higher degree of awareness and knowledge about breastfeeding. In other words, mere urban living, higher education, multiparity, hospital delivery, etc., are not sufficient to educate women about breastfeeding and there is need for a separate specific training focused on this subject.

There has to be a community-based strategic plan to impart knowledge about advantages and essentials of breastfeeding. The health care providers and the community workers together should identify the information needs and different skill sets required for successful breastfeeding practices in the society, so that training programs can be arranged and dissipation of information can be done through communication channels.

CLINICAL SIGNIFICANCE

Our study gives an insight into search for factors other than age, parity, education, and social background that affect practice of breastfeeding. It will be of help to strategy planners to formulate policies and programs at community and health care levels. After completing this study, we have started concentrating on one-to-one counseling for antenatal and postnatal patients along with their accompanying relatives.

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ORIGINAL ARTICLE

Effects of Diet Pattern on Nutritional Status of Schoolchildren: A Study in One Township of Navi Mumbai, Maharashtra, India

Rita Abbi

ABSTRACT

Diet pattern plays a key role in determining the nutritional status of children. The study was conducted to find the impact of diet pattern on the nutritional status of 105 school-going children in Navi Mumbai. Data were collected using survey questionnaire including observation and interview technique.

Food frequency questionnaires were administered to assess dietary intake. To assess growth status of children, z-scores [weight-for-height (WHZ), height-for-age (HAZ), and weight-for-age (WAZ)] were used. The objectives of the study were to determine diet variety and frequency of food intake, to find the nutritional status using food mean score for anthropometric parameters. The article further reveals the relationship between diet pattern and WAZ, WHZ, and HAZ of primary schoolchildren.

Results from the study revealed that there is a variation in the nutritional status of children. The children who eat less dietary variety may be related to malnutrition, while the large variety is related to heavyweight or obesity. The food variety should include all kinds of foodstuff from each category of major group.

Keywords: Diet pattern, Nutritional Status, Primary schools, z-score.

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INTRODUCTION

Malnutrition is one of most important public health problems in growing children, predominantly in low socioeconomic strata. Proper nutrition is therefore, an important prerequisite for better health. The dietary diversity helps to ensure nominal requirement of necessary nutrients. Nutritional status of children is influenced by the eating pattern.

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Adequate nutrition, healthy eating, and physical activities in childhood are the base for good health in adulthood.[1] Anthropometric parameters are the standardized methods for assessing the growth of children.

According to the United Nations International Children's Emergency Fund, "conceptual framework, household food insecurity, inadequate childcare, and the lack of access to health, safe water, and sanitation services are the key underlying causes of under nutrition of children."[2] Some research studies revealed mixed results of the relationship between food consumption and nutritional status of children.[3-7] More research is required to investigate the relationship between child nutrition status and food habits in a developing country like ours.

OBJECTIVE

To determine the impact of dietary patterns and the frequency of food intake using food mean score for anthropometric parameters on the nutritional status of schoolchildren in Navi Mumbai.

MATERIALS AND METHODS

Out of 14 nodes (township) in Navi Mumbai, one node, namely, Central Business District, Belapur, was selected randomly for the present study.

The list of private primary schools for the study area was prepared. From this list of primary schools, one school with children studying in 3rd and 4th standard was selected. All children (n = 105) studying in 3rd and 4th standard attending school at the time of the survey were included.

A pretested questionnaire was filled by the researcher with the help of class teachers and medical doctors. An informal atmosphere was created so that children should feel comfortable to respond to variety of food intake. An attempt was made to help children in recollecting various types of food items by showcasing the items.

To accomplish the objectives, a quantitative method of research was employed. For quantitative analysis, methods of descriptive statistics, inferential statistics, and multivariate statistics were used. Data entry and data analysis were done using Statistical Package for the Social Sciences, PC + version 10 package. To calculate the

growth status, the anthropometric measurements, HAZ, WAZ, and WHZ z-scores, were used.

The z-scores of anthropometric measurements were considered as dependent variables for data analysis. The description of the same is given as follows:

- Moderate stunting: HAZ is between −2.00 and −2.99;
- Severely stunting: HAZ is < −3.00;
- Moderate underweight: WAZ is between −2.00 and −2.99;
- Severely underweight: WAZ is < −3.00;
- Moderate wasting: WHZ is between −2.99 and −2.00;
- Severe wasting: WHZ is < −3.00;

Dietary diversity score was derived by adding food categories or food items for each food group separately for vegetables, fruits, and pulses.

The list of maximum possible varieties of edible oil, grains, pulses, vegetables, fruits, dairy product, nonvegetarian items, eggs, mixed food, beverages, sweets, fats available in this region of India was prepared in the form of a questionnaire. The objective of preparing the list was to enable the children to provide the correct information. The scores for each item were calculated and thereafter in addition, the overall scores were calculated. Table 1 shows the assignment of scores.

DISCUSSION

Nutritional Status of Children: Anthropometric Parameters

A poor z-score of WAZ is assumed to be a sign of malnourishment or, say, underweight and absence of significant wasting.

A short HAZ revealed the stunting and exhibited a slow development and proved that thinness means height is not increased in proportion to weight. A poor z-score for WHZ describes wasting and stunting, which explains thinness and low weight. This condition is related to acute hunger or some serious illness.

Table 2 presents the analysis of mean score of WAZ, HAZ, and WHZ, and body mass index (BMI) for primary schoolchildren. The mean score of dietary variety was calculated to assess the nutritional status of the children.

Weight for Age

It can be seen from Table 2 the total dietary variety scores for −2SD to +2SD, food mean score = 146.8 for primary schoolchildren who have standard weight, or z-score is >+2SD for overweight children whose food mean score = 213.4.

Table 1: Assignment of scores

Frequency of consumption	*Score assigned*
Daily	7
Weekly/2–3 times	3
Sometimes/monthly	1
Occasionally/never	0.5

Table 2: Mean z-score for WAZ, HAZ, and WHZ

	Total edible item score		
	Weight/age	*Height/age*	*Weight/height*
Z-scores	*Mean ± SD*	*Mean ± SD*	*Mean ± SD*
< −3SD	121.6 ± 19.0	0.0 ± 0.0	127.0 ± 26.2
−3SD to −2SD	119.4 ± 31.5	131.0 ± 38.3	116.2 ± 24.5
−2SD to +2SD	146.8 ± 40.6	146.5 ± 41.1	147.0 ± 40.6
>+2SD	213.4 ± 30.9	191.4 ± 16.6	207.0 ± 37.4

SD: Standard deviation

The z-score for WAZ was significantly higher with F = 32.197 at $p < 0.001$ level of significance who have normal weight or overweight. The overall mean score of all the food items were lowest for acute low weight children, and there is no significant difference between the means from the scores of moderately underweight respondents. The mean scores were more for children who are fat or overweight.

Height for Age

It can be seen from the column HAZ for primary schoolchildren (Table 2) that total dietary variety scores for −2SD to +2SD, food mean score = 131.0, and those who have normal height and those who have z-score >+2SD were taller (food mean = 191.4). The z-score for HAZ was significantly higher with F = 10.76 at, $p < 0.001$ level of significance.

Weight for Height

Total overall mean score of food groups was enhanced with better stunting children. Similar picture was observed in case of WHZ for food mean score. Table 2 reveals the overall mean score of food groups that moderately and severely stunted children have lower scores as compared with normal children and above-average height.

Total overall mean score of food was lower in moderately wasted children as compared with normal or overweight children (F = 40.831, $p < 0.001$). These data suggested that malnutrition is related to less dietary variety, while large variety was correlated with obese or fat children.

To determine which food group is associated with obesity of children, individual food group variety scores were analyzed. Tables 3 to 5 display the mean scores of food groups in terms of z-score for WAZ, HAZ, and WHZ respectively.

It can be seen from Table 3 that mean score for grains (wheat, rice, etc.) intake is high with rise in child's weight in primary schoolchildren. The data show that the mean score for grains is 19.8 for severely underweight (<3SD) as compared with children whose mean score for grains was 36.3 (>+2SD). This suggests that the mean score for

Table 3: Mean variety score of individual group in relation to WAZ z-scores

	WAZ			
Edible oil	*< −3SD*	*−3SD to −2SD*	*−2SD to +2SD*	*>+2SD*
Grains	19.8 ± 0.1	21.9 ± 5.8	25.7 ± 7.1	36.3 ± 5.2
Pulses	12.7 ± 0.0	13.5 ± 4.4	14.0 ± 4.4	17.8 ± 4.9
Vegetables	24.7 ± 4.9	25.9 ± 12.2	30.6 ± 10.8	39.7 ± 10.0
Fruits	6.9 ± 2.3	7.5 ± 7.9	10.4 ± 6.0	19.1 ± 10.4
Dairy product	10.3 ± 1.1	9.5 ± 2.9	12.3 ± 5.7	19.4 ± 6.4
Nonveg items	0.0 ± 0.0	0.8 ± 2.2	0.4 ± 1.1	0.8 ± 2.3
Eggs	0.2 ± 0.3	0.9 ± 1.6	1.6 ± 1.8	3.5 ± 2.0
Mixed food	14.2 ± 5.0	9.2 ± 5.0	14.2 ± 6.9	25.2 ± 6.4
Beverages	5.8 ± 3.7	6.4 ± 2.8	9.3 ± 3.4	13.4 ± 3.4
Sweets	16.3 ± 0.7	14.3 ± 6.0	17.8 ± 6.7	23.6 ± 3.4
Fats	9.4 ± 1.9	9.2 ± 2.0	10.5 ± 3.0	14.4 ± 3.1

SD: Standard deviation

Table 4: Mean variety score of individual group in relation to HAZ z-scores

	HAZ			
Edible oil	*< −3SD*	*−3SD to −2SD*	*−2SD to +2SD*	*>+2SD*
Grains	0.0 ± 0.0	23.1 ± 6.0	25.7 ± 7.2	28.9 ± 6.6
Pulses	0.0 ± 0.0	12.8 ± 5.0	14.0 ± 4.4	15.1 ± 1.9
Vegetables	0.0 ± 0.0	30.6 ± 15.4	30.5 ± 10.8	38.4 ± 9.1
Fruits	0.0 ± 0.0	8.9 ± 7.6	10.4 ± 6.1	15.7 ± 3.3
Dairy product	0.0 ± 0.0	11.5 ± 4.0	12.2 ± 5.7	18.1 ± 4.6
Nonveg items	0.0 ± 0.0	0.0 ± 0.0	0.4 ± 1.2	0.82 ± 1.6
Eggs	0.0 ± 0.0	1.3 ± 1.0	1.5 ± 1.8	3.2 ± 1.0
Mixed food	0.0 ± 0.0	12.2 ± 7.0	14.1 ± 7.0	19.4 ± 6.7
Beverages	0.0 ± 0.0	7.0 ± 2.4	9.2 ± 3.5	14.7 ± 2.3
Sweets	0.0 ± 0.0	14.9 ± 5.7	17.8 ± 6.7	24.4 ± 5.5
Fats	0.0 ± 0.0	9.6 ± 1.2	10.3 ± 3.0	12.4 ± 2.1

SD: Standard deviation

Table 5: Mean variety score of individual group in relation to WHZ z-scores

	WHZ			
Edible oil	*< −3SD*	*−3SD to −2SD*	*−2SD to +2SD*	*>+2SD*
Grains	22.3 ± 5.8	22.2 ± 5.6	25.7 ± 7.1	33.9 ± 6.4
Pulses	15.4 ± 5.1	12.3 ± 3.1	14.0 ± 4.4	17.2 ± 5.8
Vegetables	22.9 ± 6.6	23.4 ± 8.4	30.7 ± 10.8	40.4 ± 8.6
Fruits	8.9 ± 6.1	6.7 ± 5.0	10.4 ± 6.1	17.8 ± 6.1
Dairy product	10.0 ± 3.3	10.1 ± 3.2	12.3 ± 5.7	19.0 ± 7.4
Nonveg items	0.4 ± 1.2	0.8 ± 2.1	0.4 ± 1.1	1.0 ± 2.2
Eggs	1.8 ± 2.2	0.6 ± 0.8	1.6 ± 1.8	3.2 ± 2.0
Mixed food	10.2 ± 2.0	9.9 ± 5.0	14.2 ± 7.0	23.6 ± 7.0
Beverages	8.9 ± 3.6	6.7 ± 3.0	9.3 ± 3.4	13.2 ± 3.5
Sweets	15.9 ± 6.3	14.3 ± 5.4	17.8 ± 6.7	23.7 ± 6.9
Fats	10.1 ± 2.4	9.1 ± 1.8	10.5 ± 2.9	13.9 ± 3.7

SD: Standard deviation

grains intake is higher for heavier or higher weight children, the score being approximately twice the scores for children who were severely underweight.

According to study conducted by Liu et al,[8] "The weight gain was inversely associated with the intake of high-fiber, whole-grain foods but positively related to the intake of refined-grain foods in women, which indicated the importance of distinguishing whole-grain products from refined-grain products to aid in weight control."

The average scores for intake of pulses, vegetables, fruits, eggs, beverages, and fat is higher with more gain in weight of children; there was significant difference in mean scores among different WAZ categories.

According to Gopalan et al,[9] "The findings may be generally applicable to poor communities in several Asian countries where dietaries are based largely on cereals and to a smaller extent on legumes and pulses. The current home diets in the concerned poor communities were not satisfactory and need improvement. These diets were deficient in a number of nutrients, particularly vitamin A, riboflavin, iron, and possibly calcium. It should, however, be possible to overcome some of these deficiencies by improving the existing dietaries through the inclusion of relatively inexpensive foods that are locally available and well within the reach of the poor."

In this article, the moderately low weight children are associated with the poor score of dairy products and the higher score was found in obese children.

Many research studies made an attempt to find the relationship between measure of dietary variety and child's nutritional status.[10] The outcome of the study conducted by Sung et al[11] is in line with present findings that there was effect of food habits and physical training on the nutritional status of children. Table 4 exhibits the mean food scores for grain, vegetables, fruits, eggs, and combination of numerous types, and sweet intake is high by increasing child's stature.

There is increase in height of primary schoolchildren if mean food score namely beverages, pulses, dairy manufactured goods and fat intake is higher. A study in Mali also documented a significant relationship between diet variety and nutritional status, for 6- to 59-month-old kids. He has conducted the study in Mexico on 18- to 30-month-old children and concluded that there exists positive correlation between percentage of energy from animal groups (eggs and meat) and HAZ.[12] He further found that there was strong correlation between percentages of energy from dairy product.

A study from Ethiopia in 2000 reported that was a highly statistically significant relation between food-group variety for 1-day and/or 7-day recall and children's HAZ z-scores. Another study was conducted by Taren and Chen[13] in China. They have divided the food group scale (0–7) among rice, egg, vegetables, fruits, soya beans, meat, and other. The study was conducted on a sample of 12- to 47-year-old children.

The results revealed that there was a significant difference between z-scores of height for age for children whose intake is less than three groups of food as compared with others. Increases in this type of variety were associated with greater increases in growth for respondent with lower *vs* higher numbers of foods.

Table 5 reveals increase in food diversity with more intake of seasonal and other fruits and veggies in majority groups associated with lower energy consumption and thus a lower BMI, and perhaps also reduces the risk of obesity.[14]

McCrory et al[15] stated that "dietary variety within sweets, snacks, condiments and carbohydrate food was positively associated and dietary variety within the vegetable food group was negatively with energy intake and body fatness."

Also it was demonstrated that poor BMI was related with more variety of diet in girls as compared with their counterparts. The present findings are similar to Azadbakht and Esmaillzadeh.[16] Azadbakht and Esmaillzadeh[16] reported that "In Iran among female 18-28 years, demonstrated that a direct association between energy intake and dietary variety. Increase energy intake was related to increasing intake of fruit, vegetables and whole grain. This study also showed that there were inverse association among dietary variety score, obesity and abdominal adiposity."

CONCLUSION

The present study for 105 schoolchildren in Navi Mumbai township showed that "nutritional status" of schoolchildren is directly proportional to dietary diversity score, which is calculated by summing the frequency food items for each food group separately.

Children taking less dietary variety are prone to undernutrition.

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REVIEW ARTICLE

Diabetic Autonomic Neuropathy

Alaka Deshpande

ABSTRACT

Neuropathy is a debilitating complication of diabetes. Neuropathy can be motor, sensory, and autonomic. There is a wide variation in the prevalence of diabetic autonomic neuropathy (DAN), depending upon the definition and criteria used for diagnosis. The sympathetic and parasympathetic nerves supply various organ systems modulating their functions. Therefore, DAN has wide spectrum of clinical presentation.

The clinical manifestations differ depending upon the systemic involvement. The management consists of lifestyle modifications, control of hyperglycemia, and simple physical measures along with supportive care.

Keywords: Diabetes mellitus, Diabetic neuropathies, Hyperglycemia.

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INTRODUCTION

Neuropathy is a debilitating complication of diabetes. Neuropathy can be motor, sensory, and autonomic. It can be classified as:

- Bilaterally symmetrical polyneuropathy.
- Focal or multifocal neuropathy.

Diabetic autonomic neuropathy falls in the first category. It can be further classified as[1]

- Subclinical: Detected by physiological tests.
- Clinical: Diabetic patient presenting with symptoms and signs of neuropathy after excluding other causes.

Medical literature reports wide variation in the prevalence of DAN, depending upon the definition and criteria used for diagnosis. The other confounding factor is age of the patient. The sympathetic and parasympathetic nerves supply various organ systems modulating their functions. Therefore, DAN has wide spectrum of clinical presentation. The DAN can affect cardiovascular, gastrointestinal, genitourinary, pupillary, sudomotor, and neuroendocrine systems.

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PATHOGENESIS

As in the case of most of the complications of diabetes, multiple factors contribute to the pathogenesis of damage to the nerves both peripheral and autonomic nerves. The whole cascade starts with uncontrolled hyperglycemia. As nicely depicted by Verrotti et al[2] in Fig. 1, hyperglycemia induces various alternating metabolic pathways, resulting in production of oxidative stress and chronic inflammation causing irreparable damage to tissues. The hyperglycemia is responsible for mitochondrial overproduction of reactive oxygen and nitrogen species (RONS). RONS induce deoxyribonucleic acid (DNA) damage which overstimulates polyadenosine diphosphateribose polymerase (PARP) with endothelial dysregulation.

Chronic hyperglycemia leads to irreversible advanced glycosylation end-products (AGEs). Experimentally, it has been shown that AGEs have collagen linking activity which may be responsible for vascular complications.

The AGEs bind to Receptor for Advanced Glycation End products (RAGE) and activate these. The RAGE activation is pro-inflammatory. In addition, vascular complications like occlusion result into ischemic damage of the nerves. Peripheral nerve repair is impaired in diabetes.

CLINICAL FEATURES

Sympathetic and parasympathetic nerves supply all the visceral organs; therefore, the signs and symptoms of DAN depend on the organ system that is involved. The

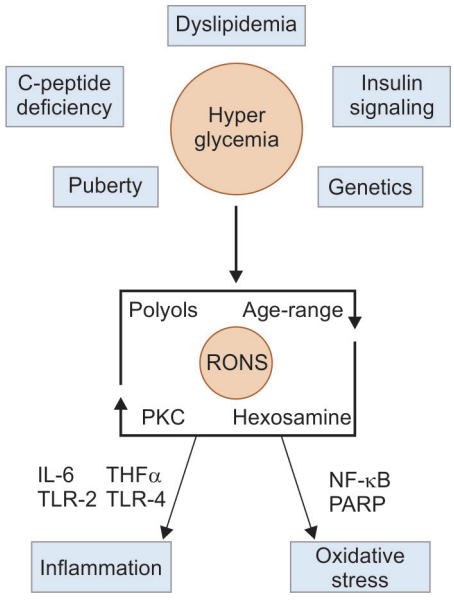

Fig. 1: Pathogenesis
Courtesy: Verrotti et al[2]

prevalence of this neuropathy is difficult to determine. It depends on the definition, the diagnostic criteria used, and the population studied.

Common manifestations due to DAN are:

- Cardiovascular autonomic neuropathy (CAN)
- Gastrointestinal
- Genitourinary
- Sudomotor
- Renal
- Arthropathy
- Pupillary
- Neuroendocrine
- Central nervous system

Cardiovascular Autonomic Neuropathy

Sympathetic and parasympathetic nerves control multiple cardiovascular functions including blood pressure.

Reduced heart rate variability is noted early in DAN. However, there are multiple risk factors contributing to the same, e.g., age, smoking, duration of diabetes, and control of hyperglycemia. Later on, as the vagal impairment sets in resting tachycardia and a fixed heart rate are common manifestations.

A fixed heart rate unresponsive to stress, moderate exercise or sleep indicates complete cardiac denervation. The CAN impairs exercise tolerance by blunting the heart rate and blood pressure response. Diabetics with multiple risk factors and preclinical coronary artery disease may be subjected to stress imaging testing so that hazardous exercise program can be avoided.[3]

Intra- and Perioperative Cardiovascular Instability

Patients with CAN may more often require vasopressor support than those without CAN. The normal autonomic response of vasoconstriction and tachycardia do not compensate for vasodilatory effect of anesthesia. There is an association between CAN and severe intraoperative hypothermia which impairs healing. The anesthesiologist and the surgeon should be alerted to CAN complications, particularly reduction in hypoxia-induced ventilatory drive.

Orthostatic Hypotension

It is defined as a fall in systolic blood pressure of >30 mm Hg or > 10 mm Hg in diastolic blood pressure in response to change in posture from supine to standing accompanied by symptoms of dizziness, faintness, even syncope. These symptoms may be misdiagnosed as due to hypoglycemia. It is therefore necessary that clinicians should pay more attention to DAN.

Silent Myocardial Ischemia

Cardiovascular autonomic neuropathy impairs appreciation of ischemic pain, thereby delaying the recognition and treatment. The electrocardiograph of a diabetic patient with exertional chest pain has shown prolongation of time of perception of chest pain from the onset of ST depression (angina perceptual threshold). Due to this unawareness, a diabetic patient with CAN may continue to exercise despite increasing ischemia. The studies have shown 28% cases with CAN had silent ischemia as against 10% cases without CAN.

Autonomic Cardiopathy

Cardiovascular autonomic neuropathy may be associated with diastolic and systolic dysfunctions in the absence of cardiac disease. Cardiovascular autonomic neuropathy can be evaluated more accurately by cardiac radionuclide imaging. Metaiodobenzylguanidine (MIBG) (the nonmetabolized norepinephrine analog) is actively taken up by postganglionic sympathetic nerve terminals of myocardium. Several studies have shown reduced myocardial MIBG uptake in patients with CAN.

Treatment

It aims at primarily prevention of disease progression. Hence, modifiable risk factors, such as hyperglycemia, hyperlipidemia, hypertension, and smoking need to be controlled. The ACCORD trial links intensive glycemic control to increased mortality. Therefore, target glycated hemoglobin level should be achieved gradually.

- Nonpharmacological measures
  - Increased consumption of water
  - Slow postural changes
  - Elevation of head end by up to 30 cm
  - Dorsiflexion of feet or hand grip exercise should be performed before standing
- Pharmacotherapy
  - Mineralocorticoid Fludrocortisone 0.1 to 0.4 mg/day
  - Alpha adrenoceptor agonist Midrodin used for postural hypotension, but it may cause severe supine hypertension so patients should not use it within 6 hours of bedtime. Norepinephrine prodrug Droxidopa is orally given, and it has recently received US Food and Drug Administration approval.

Gastrointestinal System

Gastroesophageal reflux disease (GERD), gastroparesis, and diabetic diarrhea are the commonest gastrointestinal (GI) manifestations of DAN. The autonomic neuropathy

decreases the pressure of the lower esophageal sphincter (LES), impaired clearance function of the tubular esophagus and hyperglycemia increases transient relaxations of LES. As a result, patient complains of heartburn and regurgitation. The GERD may also lead to laryngitis, chronic cough, and even bronchospasm. Control of hyperglycemia and use of prokinetic agents are advocated.

Gastric emptying time is delayed in up to 50% of cases. Gastroparesis diabeticorum is the term used to indicate altered GI motility. Food residue is retained in the stomach as a result of dysmotility. It may manifest as nausea, pain in abdomen, loss of appetite, early satiety, postprandial vomiting, and bloating. A gastric splash may be elicitable on clinical examination. It may also be associated with bezoar and bacterial overgrowth in the stomach and small intestines. It is prudent to rule out the obstructing lesion in stomach or intestine by endoscopy. The delayed gastric emptying time may be studied by scintigraphy.

Gastroparesis may impair glycemic control by mismatching plasma glucose and insulin levels. Diarrhea manifest as profuse, watery stools, usually nocturnal, lasting for a few days alternating with constipation. Reduced motility, reduced receptor-mediated fluid absorption, pancreatic insufficiency, and bacterial overgrowth contribute to diabetic diarrhea.

Fecal incontinence due to incompetence of anal sphincter is another manifestation of diabetic neuropathy. Chronic severe constipation is also common. Studies have implicated hyperglycemia as a cause of GI dysmotility. Control of hyperglycemia, use of prokinetic agents, some simple physical measures like small frequent meals and avoiding supine position immediately after food are advocated.

Genitourinary System

The urinary bladder is innervated by autonomic and somatic nerves. Diabetic autonomic neuropathy causes bladder dysfunction, retrograde ejaculation, erectile dysfunction, and dyspareunia due to lack of lubrication. Erectile dysfunction is associated with cardiovascular disease.

Bladder dysfunction may manifest initially as decrease in feeling the sensation of full bladder due to loss of autonomic afferent innervations. It results in frequent urination, while involvement of efferent fibers results in incomplete emptying. It leads to frequent urinary tract infections, dribbling due to overflow—incontinence. About 7 to 10% of cases may develop atonic bladder, necessitating regular catheterization.[4]

Management

The patient may initially be subjected to urine analysis and sonography to see the postvoidal residual volume. It is necessary to carry out complete urodynamic testing. The medications that impair detrusor activity like anticholinergic agents, tricyclic antidepressants, and calcium-channel blockers may be changed. Bladder training with strict voluntary micturition schedule, coupled with Crede maneuver, is advised. Intermittent catheterization may be needed.

Such problems are further contributed by smoking, alcohol, anxiety, and depression which need attention. Sexual dysfunctions in males include erectile dysfunction, retrograde ejaculation, and impotence. Sexual functions are affected in diabetic females too. Anxiety and depression play a role in sexual functions. Management should be tailored to individual needs. Phosphodiesterase inhibitors are used as first-line therapy in erectile dysfunction. However, they are contraindicated in the presence of heart disease which is being treated with nitrates.

Sudomotor System

Thermoregulation and sweat function are under the control of peripheral sympathetic cholinergic system. There is a loss of sweat function in glove and stocking area with compensatory proximal hyperhidrosis. Peripheral autonomic dysfunction manifests as sweating abnormalities, change in skin texture, loss of nails, itching, callous formation, edema, and foot ulceration. Quantitative sudomotor axon reflex testing may be used to detect denervation.

Other Changes

Pupillary abnormalities may cause failure in dark adaptation and difficulty in night driving. Peripheral neuropathy along with autonomic neuropathy is the main cause of diabetic arthropathy.

A prospective study in type I diabetes mellitus (T1 DM) with a 14-year follow-up has shown increasing hypoglycemic episodes due to autonomic neuropathy, resulting in decreased glucagon and epinephrine response to falling sugar levels. It is also termed as hypoglycemia-associated autonomic failure. Hypoglycemia unawareness is a major risk factor for strokes in diabetics independent of CAN. Surgeon and anesthesiologist have to be aware of DAN in diabetic patients undergoing any surgery. The American Diabetic Association recommends screening for DAN in T2 DM patients at the time of diagnosis and 5 years after the diagnosis of T1 DM.

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CASE REPORT

Hybrid Laparoscopic and Video-assisted Thoracoscopic Surgery Repair of Recurrent Traumatic Diaphragmatic Hernia: A Case Report and Literature Review

[1]Anith N Riduan, [2]Narasimman Sathiamurthy

ABSTRACT

Introduction: A case of recurrent traumatic diaphragmatic hernia (TDH) is presented in whom hybrid laparoscopic and video-assisted thoracoscopic surgery (VATS) was carried out.

Case report: A 38-year-old male with a history of emergency laparotomy for repair of traumatic diaphragmatic injury and left thoracotomy and repair of dissected descending thoracic aorta with plication and interposition graft repair presented to us 1-year postsurgery with chronic vomiting, electrolyte imbalance, and malnourishment. Diagnosis was established with contrast-enhanced computed tomography (CECT) of thorax and abdomen which showed a left hemi-diaphragm mid-part defect, measuring 5.1 cm in diameter, with stomach herniation. He underwent laparoscopic and left VATS repair of diaphragmatic hernia. Herniated stomach was adherent to the lower lobe of left lung and descending thoracic aorta.

Conclusion: In the age of minimally invasive surgery, hybrid laparoscopic and thoracoscopic surgery is an innovative, safe, and feasible option in managing recurrent diaphragmatic hernia.

Keywords: Recurrent diaphragmatic hernia, Thoracic surgery, Traumatic diaphragmatic hernia, Video-assisted thoracoscopic surgery.

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INTRODUCTION

Traumatic diaphragmatic hernia occurs in approximately 1 to 6% of major thoracic injuries, most commonly after high-velocity blunt trauma.[1] It is a rare entity that presents a diagnostic and therapeutic challenge which may lead to significant mortality and morbidity in general. Around 80 to 90%[2-4] will have major associated thoracoabdominal injuries involving liver, spleen, small bowl, and mesentery as well as lung contusions and rib fractures. Thoracic aorta injury occurs in up to 7.7% of patients with TDH.[2] Late presentation and recurrences can present with gastrointestinal and respiratory complications. Misdiagnosis can occur, resulting in delay in treatment and with consequent poor outcome.[5,6] Furthermore, in dealing with chronic and recurrent diaphragmatic hernia, in anticipation of significant adhesions to intrathoracic structures, selection of surgical approach for repair is of paramount importance. This report describes a case of recurrent TDH in which repair was performed using hybrid laparoscopic and video-assisted thoracoscopic surgery.

CASE REPORT

A 38-year-old Malay male presented with chronic vomiting and epigastric pain for a 6-month duration. He had a history of trauma after a fall from height in which he sustained a blunt left diaphragmatic injury, dissection of descending thoracic aorta, and fractures of left iliac crest, proximal third femur, and superior pubic rami. He underwent emergency laparotomy for repair of traumatic diaphragmatic injury, left thoracotomy, and repair of dissected descending thoracic aorta with plication and interposition graft repair and external fixation of the pelvic fracture. He recovered well after the surgeries and rehabilitations. After 1 year, he came back with chronic upper gastrointestinal symptoms, electrolyte imbalance, and malnourishment with body mass index of 14.7 kg/m^2.

Initial chest X-ray done showed an elevated left hemidiaphragm, with no apparent herniation (Figs 1A and B). Upper GI endoscopy showed a deformed elongated stomach and retracted pylorus with no evidence of gastric outlet obstruction. Barium meal study revealed abnormal stomach configuration with partial obstruction at level of pylorus (Figs 1C and D). Diagnosis was finally established with CECT of thorax and abdomen which showed a left hemi-diaphragm mid-part defect, measuring around 5.1 cm in diameter, with stomach herniation.

He underwent a laparoscopic and left VATS repair of diaphragmatic hernia. Operation was done using four laparoscopic ports (10, 10, 5, and 5 mm) with one

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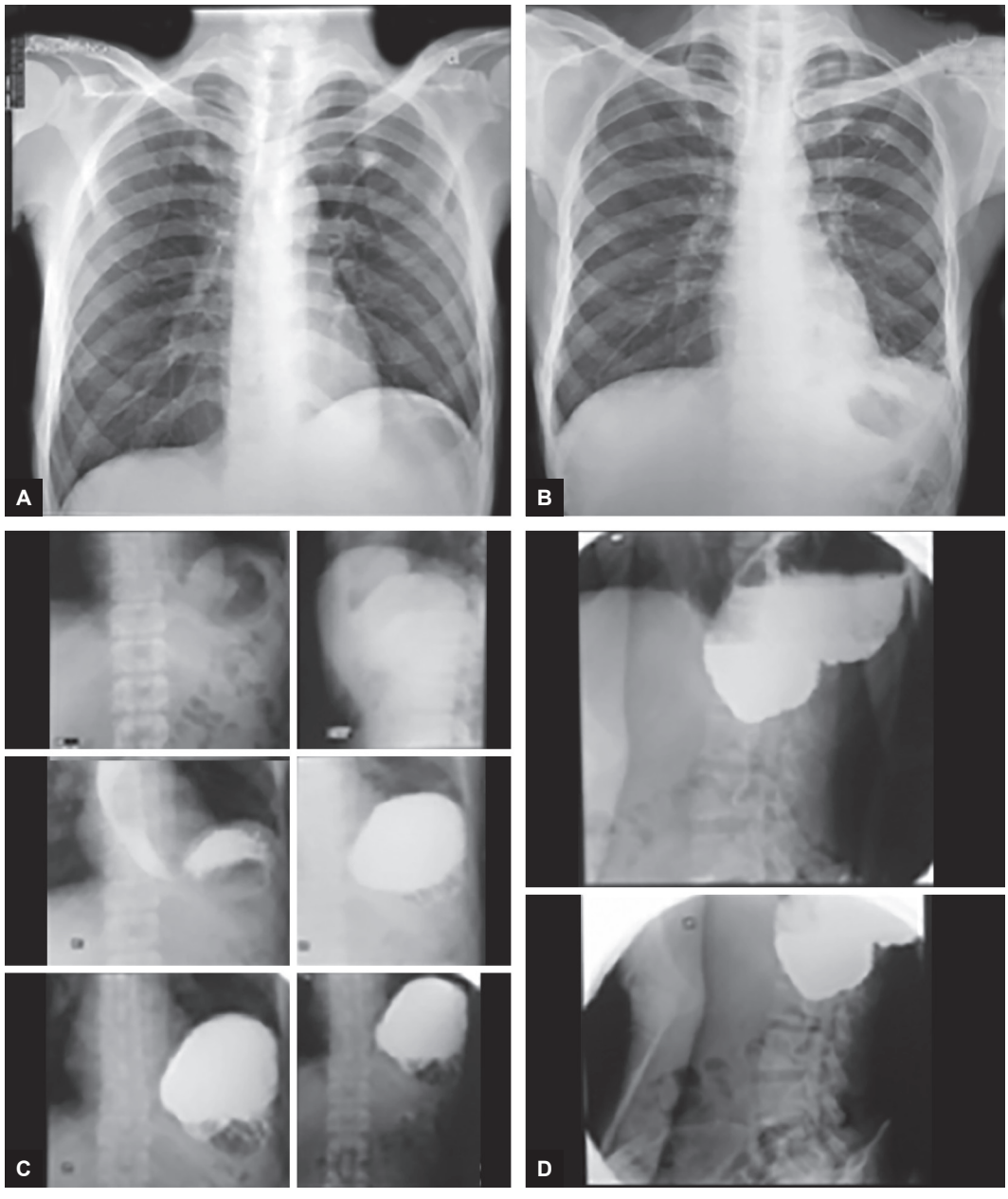
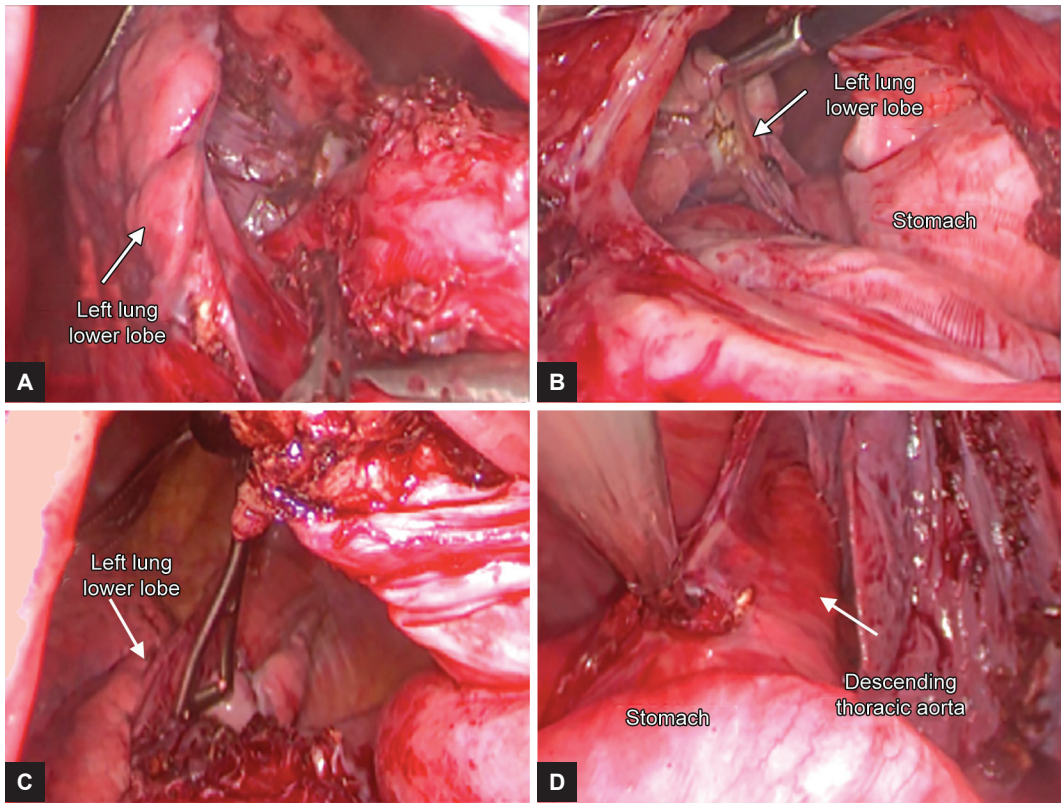


Figs 1A to D: (A) Chest X-ray showing elevated left hemi-diaphragm, (B) postoperative chest X-ray, (C and D) fluoro-barium meal study showing obstruction at pylorus level

thoracoscopic port (10 mm). Intraoperatively, the herniated stomach was adherent to the left lower lobe of left lung and descending thoracic aorta. Adhesions were released successfully by VATS (Fig. 2). In view of dense adhesions around the cardio-esophageal junction to the diaphragm, a mini upper midline laparotomy was done to aid adhesiolysis and diaphragmatic hernia repair with prolene mesh. Postoperatively, he was nursed in intensive care unit for 1 week and because of hypercapnic respiratory failure, weaning off ventilator was slow. Being malnourished preoperatively, his nutrition was carefully managed to avoid refeeding syndrome. He recovered well subsequently and was discharged to home. At our follow-up after 2 weeks, he had gained 6 kg in weight and was symptom-free.

DISCUSSION

Hybrid laparoscopic and VATS repair represents a minimally invasive technique sparing patients from morbidity and invasiveness of open thoracoabdominal surgery which requires a large and extensive incision and long postoperative recovery period. This patient had an eventful postoperative period in which ventilation weaning was difficult in view of hypercapnic respiratory failure. His arterial blood gases showed persistent respiratory acidosis with CO_2 retention. Effect of CO_2 insufflation in thoracoscopic surgery has not been widely documented in the current literature; however, a concern has been raised in a few publications that it can cause hypercarbia and acidosis[7] which can lead to prolonged ventilation in the postoperative period. Refeeding syndrome should also always be considered in



Figs 2A to D: Intraoperative finding; herniated stomach densely adhering to the left lower lobe and descending thoracic aorta

at-risk patients with chronic symptoms and malnutrition. Proper nutrition support plan and care are quintessential to avoid its catastrophic complications and morbidity.

CONCLUSION

Recurrent diaphragmatic hernia repair is an arduous task and the right approach in repair is fundamental to ensure the best outcome in surgery. In the age of minimally invasive surgery, hybrid laparoscopic and thoracoscopic surgery is an innovative, safe, and feasible option in managing recurrent diaphragmatic hernia.

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CASE REPORT

Acute Total Hip Arthroplasty in Unreconstructable Acetabular Fractures: Three Case Reports

[1]Kuldip Salgotra, [2]Sarabjeet Kohli, [3]Nilesh Vishwakarma, [4]Shaival Chauhan

ABSTRACT

The indications of acute total hip arthroplasty (THA) in acetabular fractures are limited and well defined. The reinforcement antiprotrusio cage offers an opportunity to conduct the THA in select unreconstructable acetabular fractures. The need to open-reduce the posterior column and dome fracture is not necessary and the fracture need not be aligned as the Bursch–Schneider's cage with flange can compensate for significant bone loss. Three patients in this study with unreconstructable acetabular fractures underwent acute THA. They were followed up for 3 years. The mean Harris hip score was 72.7. Two patients returned to their preexisting activity level. The short-term result in terms of Harris hip score is good and preinjury level can be achieved. There were no cases with complications of cage subsidence or osteolysis during the 3-year follow-up.

Keywords: Acetabulum fractures, Acute total hip arthroplasty, Bursch–Schneider's cage.

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INTRODUCTION

Traditionally, THA is indicated for primary hip osteoarthritis and hip arthritis secondary to multiple pathologies. Elderly patients with extensively comminuted acetabular fractures, acetabular fracture along with neck femur fracture, and impacted dome fracture with coexisting osteoporosis can be effectively treated with acute THA.[1,2] Acetabular fractures which cannot be reconstructed present a unique challenge. In selected elderly patients with acetabular fractures, acute THA helps to make them ambulatory, obviating the complications associated with prolonged bed recumbency.[3]

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CASE REPORTS

This article evaluates three cases of unreconstructable fractures of acetabulum who presented at our facility. Mean age was 57 years and mean duration since index injury was 9 days. Mechanism of injury was road traffic accident in all three cases. Two cases received first-aid treatment in the form of traction at their native places before they sought treatment. One patient had concomitant bladder injury and uncontrolled diabetes which delayed primary fixation. On radiological assessment with x-rays and three-dimensional computed tomography (3DCT), the acetabular fracture was deemed unreconstructable as per criteria enumerated by Mears and Velyvis[2] as well as Chana–Rodríguez et al.[4]

The fracture was classified as posterior wall posterior column fracture type of Letournel and Judet classification with involvement of superior dome of acetabulum. On X-ray of posterior wall, there was bone loss with comminution along the posterior wall (Fig. 1). On 3DCT of fractures, the defect in posterior wall and column and involvement of superior dome of acetabulum were delineated.

We relied on the landmark studies by Tidermark et al[5] and Sarkar et al[6] to decide regarding the use of reinforcement ring. The planning was done pre-op for the need of grafting and the method to fix the graft and the need of Bursch–Schneider reinforcement cage for the defective posterior wall as well as the weight-bearing dome of acetabulum were decided. With the help of template of the cage, the size of the graft was also determined. The bone stock of head was noted as the need of graft. A 3D model was made which helped in the delineation of the bone loss and the need of graft. Planning aimed to reconstitute the center of rotation of the hip with stability.

All the cases were operated by Kocher–Langenbeck approach with posterior dislocation of the hip. In all cases, the incision was extended distally enough to allow for femoral preparation. Necrotic tissues and devitalized tissues were excised to reduce the incidence of heterotopic ossification. The lesser sciatic notch was approached with retraction of obturator internus while protecting the sciatic nerve. Dissection was done in all cases between the hip capsule and short external rotators to expose the posterior column and sciatic notches. The part of hip posterior capsule and comminuted fragments were excised during exposure. A provisional femoral neck cut

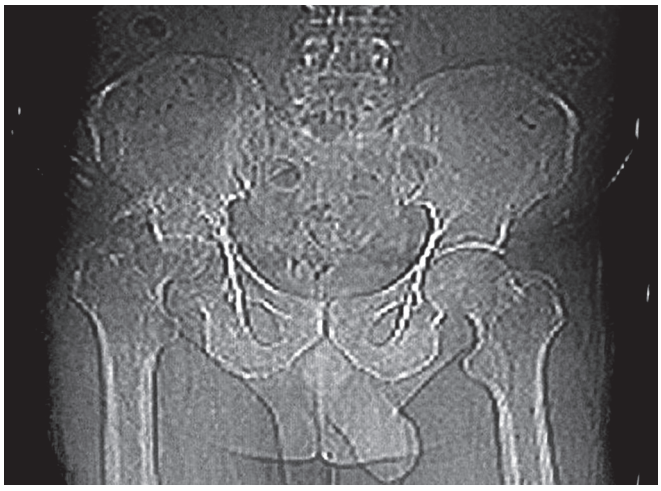


Fig. 1: Acetabular fracture dislocation

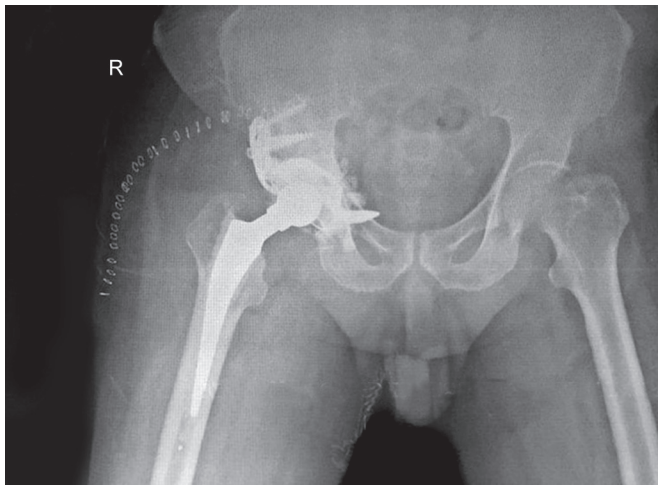


Fig. 2: Postoperative total hip replacement using Bursch-Schneider's cage

was taken. A 360° exposure of acetabulum was done. The bone defect along the acetabulum superior dome and posterior wall was demarcated and edges were freshened. The gluteus medius and minimus were partially elevated above the acetabulum with chisel so that the flange of cage can be attached there. No attempt was made to reduce the fracture. The acetabulum preparation was done with reamers as in primary THR, after the cartilage of remaining acetabulum was removed and there was punctuate bleeding; the reaming was stopped and the bone defect was marked.

The head of femur was cleaned of all cartilage and the graft of appropriate size was placed in the defect after reshaping it. Graft was provisionally fixed with K-wire and then the final reamer used previously was used to finally shape the graft to fit the Burch–Schneider's reinforcement cage trial. Appropriate size cage was selected after trial. The adaptation of the flange and/or nose was performed. Entry point made with chisel, the nose of cage was tapped into ischium until implant lay in the acetabulum floor. The cage was secured to Ilium with 3 to 4 screws; the graft was fixed to the cage with screws. The surrounding area of the cage was impacted with graft. The cemented cup was positioned and cemented stem inserted in femur after standard preparation and reduction. Postoperatively, patients were made to walk weight bearing on postoperative day 2, which patients tolerated well without much pain (Fig. 2).

All patients were followed up for 3 years. The mean Harris hip score was 72.7. Two patients returned to their preexisting activity level. There was one postoperative dislocation in immediate postoperative period which was managed with closed reduction and abduction pillow (Fig. 3). The leg discrepancy was found in all three patients but was well tolerated, as it was less than 1.5 cm in each. The follow-up was done at 6 weeks, 6 months,

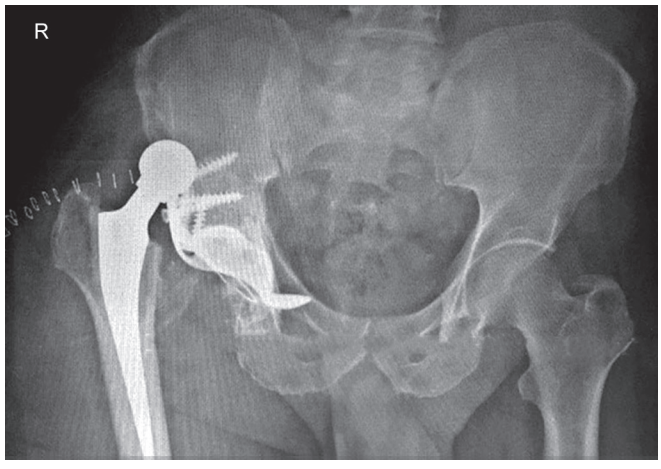


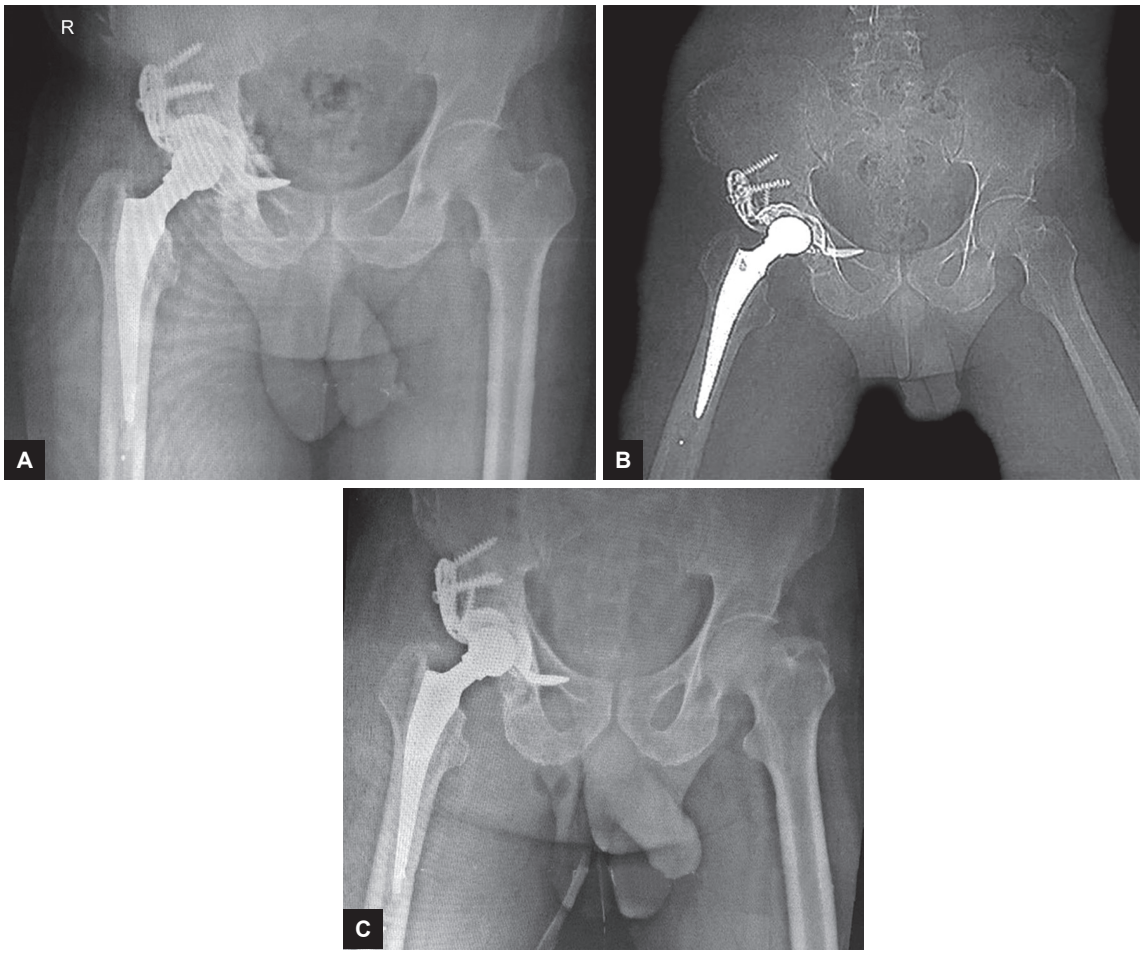
Fig. 3: Dislocation in immediate postoperative period

and then yearly. Radiological examination was done at each follow-up (Fig. 4). No case of cup subsidence or heterotopic ossification was noted in any of the patients and they retained their general level of satisfaction. No osteolysis was observed at the end of 3 years.

DISCUSSION

Acute THR serves as a salvage surgery in elderly patients with unreconstructable acetabular fractures. It enables immediate weight bearing, thereby saving the patient from complications of recumbency. Poor outcomes in acetabular fractures have been clearly enumerated by Mears and Velyvis[2] as severe intraarticular comminution with 10 fragments or more, impacted femoral head in acetabulum or femoral head fracture more than 40% involvement of weight-bearing arc including dome and preexisting osteoarthritis.[4]

Osteoporosis played an important factor in decision-making for acute THR. Marked osteoporosis was evident in all three patients as per Singh et al[7] index.



Figs 4A to C: Follow-up after 6 months of Bursch–Schneider's cage

Comminuted weight-bearing dome besides severe osteoporosis in all these patients were strong criteria to go for acute THA with reinforcement cage and autologous bone graft.[5]

Tidermark et al[5] advised using a reinforcement cage and this was supported by Sarkar et al.[6] They observed that reinforcement rings provide higher initial stability as compared with THR along with plates for acetabular reconstruction. Thus, we preferred Bursch-Schneider's rough blasted titanium cage with flanges over conventional THR. Tidermark et al[5] reported 10 patients, mean age 73 years (57–87), treated acutely with a THA supported by a reinforcement ring (Burch–Schneider antiprotrusion cage) and autologous bone grafting of the acetabulum. At a mean follow-up of 38 months (11–84), good-to-excellent outcomes were achieved in 60% (6/10), bone graft was completely incorporated in all cases, and no evidence of loosening was encountered. Early dislocations occurred in 30% of the subjects (3/10).

We had good results in all three cases (HHS–72.7). Two patients achieved preinjury level activity. One postoperative dislocation in immediate postoperative period was encountered. No cup subsidence or heterotopic ossification was seen in any patient and they retained their general level of satisfaction. No osteolysis was observed at the end of 3 years. Literature reports varied dislocation rates (7–30%).[5,8]

CONCLUSION

Acute THA with reinforcement ring along with autologous bone grafting gives promising results in a selected group of elderly patients with nonconstructable acetabular fractures. The short-term results are good with a high chance of patients returning to their preinjury level.

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SHORT COMMUNICATION

Accelerated Atherosclerosis: A Clinical Report

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ABSTRACT

Aim: The present study was conducted to determine the major risk factors for accelerated atherosclerosis.

Materials and methods: Patients between 50 and 70 years who had documented evidence of atherosclerotic vascular disease at three or more than three arterial sites in a short span of time were included in this study.

Results: Majority (83%) had diabetes for over 15 years mostly with poor control and most common dyslipidemia was low high-density lipoprotein (HDL). Evidence of coronary artery disease (CAD) was noted in all cases. Next common site for atherosclerosis was observed in carotid and lower limb arteries. Three amputation were done which showed evidence of inflammatory arthritis.

Conclusion: Accelerated atherosclerosis is an important complication of long-standing uncontrolled diabetes. Dyslipidemia characterized by low HDL and other inflammatory arthritic conditions also contribute to accelerated atherosclerosis.

Clinical significance: Intermittent claudication or pain in leg in long-standing diabetic cases particularly associated with CAD should arouse suspicion of peripheral arterial diseases. Early detection and management of peripheral arterial disease can save the involved limb from amputation.

Keywords: Accelerated atherosclerosis, Amputation, Cerebrovascular disease, Coronary artery disease, Diabetes mellitus, Intermittent claudication, Peripheral artery disease.

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INTRODUCTION

Coronary artery disease has assumed epidemic proportion in India and has become more aggressive and premature in its occurrence.[1] Its clinical presentation is rapidly changing and we see more often cases with accelerated atherosclerosis involving coronaries, cerebrovascular and peripheral arteries in a short span of time.[2] Recently, we came across number of such cases at a relatively early age where three or more than three vascular beds were involved. The term "accelerated atherosclerosis" has been used in experimental reports[3,4] and sometimes in clinical situations.[5,6] As this has far-reaching clinical and prognostic implication, we decided to share this short clinical experience for wider dissemination.

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MATERIALS AND METHODS

This is a prospective observational study of cases who presented with accelerated atherosclerosis with clinical evidence of atherosclerotic vascular lesions at three or more than three sites. The study comprises of six cases between age group 55 and 70 years. Subjects more than 70 years of age have not been included in this study. Data regarding age, sex, family history, known disease of subjects, tobacco details, body mass index, blood pressure, glycosylated hemoglobin (HbA1c), lipid profile, chest X-ray, carotid Doppler, peripheral Doppler, and computed tomography (CT) brain of each subject were recorded. Obesity was defined as per criteria described in Nature review.[7] Hypertension as per Eighth Joint National Committee criteria,[8] control of diabetes (HbA1c 8%) as per American Diabetes Association,[9] and carotid and peripheral Doppler were performed using a 7 MHz transducer as per American Society of Echocardiography guidelines,[10] and CT brain based on American College of Radiography guidelines.[11]

Statistical methods: Collected data were presented as mean, median, and standard derivation using MS Office Excel.

RESULTS

Mean age of study cases is 65.5 ± 5.25 years. Half of our cases were used to oral tobacco (Table 1). Lone female case though 68 years old had diabetes for as long as almost 28 years.

Table 1: Demographic profile in accelerated atherosclerosis

Total number of subjects	*6*
Male:female	5:1
Mean age (years)	65.5 ± 5.25
Current smoker	Nil
Oral tobacco	2 (M-2) (33.3)
Both (smoking + SLT)	1 (M-1) (16.6)

M: Male; SLT: Smokeless tobacco; All values are mean ± standard deviation or actual value with percentage in parenthesis

Table 2: Clinicobiochemical profile in accelerated atherosclerosis

Mean BMI (kg/m^2)	24.78 ± 3.59
Obesity	4 (66.6)
HTN	5 (83.3)
HTN duration (years)	11.2 ± 7.9
T2DM	5 (83.3)
T2DM duration (years)	18.8 ± 5.77
HTN + T2DM	4 (66.6)
Uncontrolled diabetes	4 (80)
Mean HDL (mg/dL)	34.7 ± 4.11

BMI: Body mass index; HTN: Hypertension; All values are mean ± standard deviation or actual value with percentage in parenthesis

Table 3: Imaging profile in accelerated atherosclerosis

Aortic calcification (CXR)	1 (16.6)
Plaque in carotid (Doppler)	
Unilateral	Nil
Bilateral	4 (66.6)
Plaque in peripheries (Doppler)	
Upper limb	Nil
Lower limb	3 (50)
Both upper and lower limb	1 (16.6)
Number of amputations	3 (50)
CAD (CAG)	6 (100)
Evidence of cerebrovascular disease (CT)	2 (33.3)
CKD (US abdomen)	3 (50)
Rheumatoid arthritis	1 (16.6)
Psoriasis	1 (16.6)
Ankylosing spondylitis	1 (16.6)

CKD: Chronic kidney disease; CXR: Chest x-ray; CAG: Coronary angiography; US: Ultrasound; All values are mean ± standard deviation or actual value with percentage in parenthesis

Five patients (83.3%) had hypertension and 4 patients (66.6%) had both diabetes and hypertension. Four patients (66.6%) had obesity. Almost all diabetic cases had been suffering with diabetes for over 15 years; 80% of diabetics had poor control of diabetes (HbA1c > 8%). The mean HDL was 34.7 ± 4.11 mg/dL (Table 2).

Documented coronary atherosclerosis was seen in all six cases; next common site for atherosclerosis was observed in carotid and peripheral arteries in lower limbs (Fig. 1). Evidence of aortic atherosclerosis in the form of aortic calcification was observed in one case (Table 3). Two cases presented with symptoms of vascular ischemia. Of these, one underwent lower limb amputation, whereas another needed amputation of left index finger and later left 5th toe amputation. Three cases had chronic kidney disease and three showed evidence of inflammatory arthritis.

DISCUSSION

Our study addresses the clinical concept of "accelerated atherosclerosis," a term used currently not unoften. It is worthwhile to note that cases of accelerated atherosclerosis involving coronary arteries, cerebrovascular tree, and peripheral arteries are more commonly seen these days. As evident in our small study, type II diabetes mellitus appears to be the key factor behind accelerated atherosclerosis. It is well known that atherosclerotic involvement of coronary, cerebrovascular, and large diameter peripheral arteries, such as carotids and/or arteries of extremities is due to macrovascular complication of diabetes.[12,13] It would be prudent to mention here that clinicians more often than not are preoccupied with microvascular complications of diabetes at the expense of macrovascular involvement of peripheral vessels.

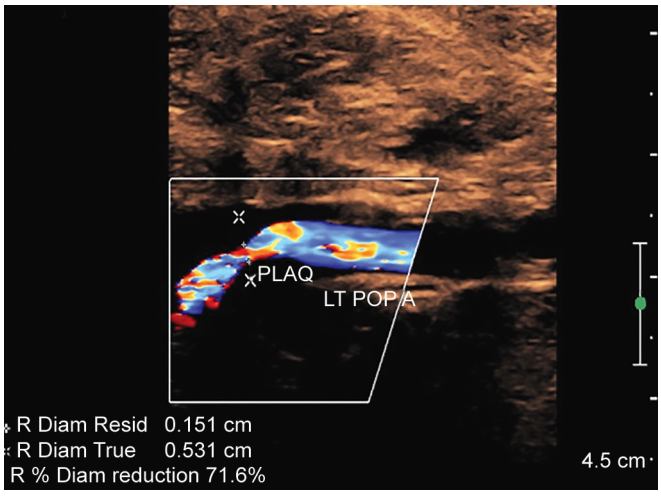

Fig. 1: Doppler study of a 68-year-old male postcoronary artery bypass grafting, known hypertension, and known diabetes of 17 years' duration. Left popliteal artery showing plaque with 71.6% luminal reduction

Moreover, the causes of accelerated atherosclerosis need to be worked out in each case. In our study, uncontrolled long-standing type II diabetes mellitus (T2DM) has been associated with accelerated atherosclerosis. The second important cause was dyslipidemia characterized by low HDL.[14] Diabetes along with hypertension increases the speed and extent of inflammation. Despite this, there are other clinical conditions associated with high inflammatory state like rheumatoid arthritis,[6] systemic lupus erythematosus[15], ankylosing spondylitis,[16] psoriasis,[17] human immunodeficiency virus,[18] and Tangier's disease.[19] Rapid involvement of limb vessels leads to dreaded complication of peripheral gangrene, which may result in amputation as happened in three of our subjects. Surprisingly, only 2 (33.3%) of our patients were tobacco users. One was smoker and the second was smoker as well as oral tobacco user. This low incidence could be due to small sample size in this study.

CONCLUSION

Diabetics are particularly prone to accelerated atherosclerosis. Besides coronary arteries, other arteries, especially

those of limbs, may get involved at an early stage and progress rapidly. Therefore, clinicians must look for signs of limb ischemia in these patients as a routine. Early detection and treatment of limb ischemia may help in reducing incidence of amputations.

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LETTER TO THE EDITOR

Hemolytic Disease of the Newborn due to Anti-C Antibodies

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Sir,

Our blood bank received samples of a newborn male child and his mother for blood group and cross match. The neonate developed jaundice on the 3rd day after birth (bilirubin = 27.5 mg/dL, indirect bilirubin = 24 mg/dL), which progressed further on days 4 and 5. Double volume exchange transfusion was planned and the demand was of 520 mL of fresh whole blood. The blood group of both the mother and child was B positive, so cross match with fresh B positive blood was performed. The first bag picked up randomly for cross match was found to be compatible with the baby's sample; however, it showed 4+ reaction with the mother's sample. Cross match with c negative, e negative, and K negative bag was performed next and was found to be compatible with both the mother's and baby's sample. This blood bag was issued. Subsequently, further workup with the pretransfusion samples was done. Direct Coombs test of the baby's sample was positive (4+ reaction) and indirect Coombs test of the mother's sample was positive (4+). The 3-cell and 11-cell panels of the mother's sample showed that there was presence of anti-c antibody in the mother.

The birth history of the baby was insignificant (full-term normal vaginal delivery). He had two older siblings in whom there was no history of jaundice at or after birth. The presence of anti-C antibodies in the mother could be due to exposure to C-positive fetal blood in the previous pregnancy, since there was no history of blood transfusion in the past. These antibodies were responsible for hemolytic disease of the newborn in the current pregnancy.

The most common cause of hemolytic disease of the newborn is the presence of anti-D antibodies in the mother. However, cases due to anti-c have been reported previously. Appelman et al[1] reported two cases of severe hemolytic disease of the newborn. In both these cases, the mothers were multiparous and had history of blood transfusion. Another case was reported in a neonate whose mother had one healthy child from the first pregnancy and two still births following that. She had also received blood transfusion.[2] Maternal alloimmunization to anti-D is low due to better detection and effective preventive options; other Rh antigens remain a significant albeit uncommon cause of hemolytic disease of the newborn. Prophylactic immunoglobulins are not available yet to prevent the formation of these antibodies.

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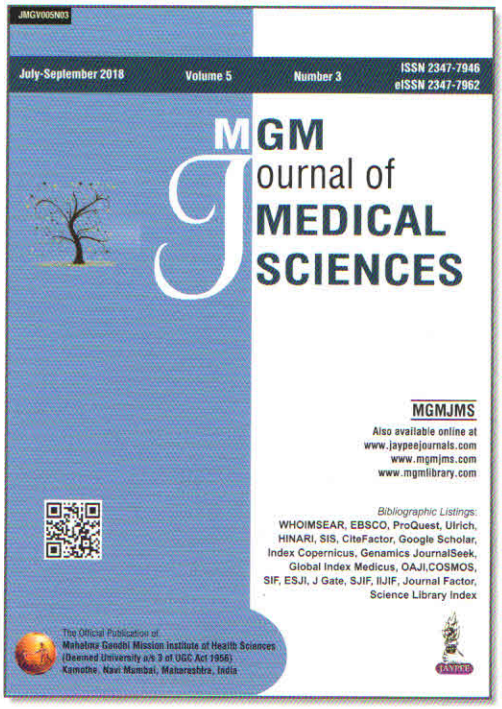

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