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## CONTENTS

### EDITORIAL

New Antibiotics: Dalbavancin (Zeven) and Tedizolid Phosphate (Sivextro)

*Reddy KR*

### ORIGINAL ARTICLES

1. MIPPO Vs ORIF with Plate in Distal Tibial Fractures 1  
*Shrestha RP, Ghale TM*
2. Clinico-microbiological Correlation of Vaginal Discharge in Reproductive Age 5  
*Pandit C, Dhungana PR, Dubey NR, Gurung RT, Gurung TK, Thapa E*
3. Use of Polypropylene Mesh for Treatment of Complete Rectal Prolapse: Should We Renounce? 9  
*Upadhyay PK, Neupane BR, Gurung NV, Pun PB*
4. Minimal Invasive Plate Osteosynthesis of Distal Tibia Fractures 14  
*Kandel IS, Shrestha B, Tripathi RB, Ghale TM*
5. Isoflurane Vs Halothane: A Comparative Study of Haemodynamic Response due to Carbondioxide Pneumoperitoneum During Laparoscopic Cholecystectomy 19  
*Ghale TM, Shrestha RP*
6. Incidence of Tuberculous Lymphadenitis 23  
*Gurung B, Nepali R*
7. Femoral Artery Variations in Nepalese Cadavers 27  
*Baral P, Jha AK*
8. Immunostimulatory and Anti-tumor Activity of Aqueous Extract Isolated from *Cnidium monnieri* Fruits 30  
*Malla B, Chang BY, Lee MK, Kim SY*
9. Retrospective study of Ureterorenoscopy +/- Procedure in Gandaki Medical College Teaching Hospital, Pokhara 34  
*Bhujju S*
10. Bacteriological Study of Postoperative Wound Infections and Their Antibigrams 37  
*Reddy KR, Reddy SR*

### Instructions to authors

# Editorial

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## **NEW ANTIBIOTICS: DALBAVANCIN (ZEVEN) AND TEDIZOLID PHOSPHATE (SIVEXTRO)**

The Food and Drug Administration (FDA), USA approved two new antibacterial agents under the FDA's new Generating Antibiotic Incentives Now (GAIN) Act of 2012, providing physicians and patients with a new treatment option for serious skin infections.

- 1) Dalbavancin on 23<sup>rd</sup> May, 2014 for the treatment of adults with Acute Bacterial Skin and Skin Structure Infections (ABSSSI) caused by certain susceptible bacteria such as *Staphylococcus aureus* including Methicillin-Susceptible and Methicillin-Resistant strains of *Staphylococcus aureus*.
- 2) Sivextro or Tedizolid phosphate on 20<sup>th</sup> June, 2014 to treat severe *Staphylococcus aureus* and other skin infections in adults, including those caused by Methicillin-Resistant *Staphylococcus aureus* (MRSA).

Dalbavancin and Sivextro are the first medicines to be designated as Qualified Infectious Disease Products (QIDP) by FDA. The drugs were granted QIDP designation because they are antibacterial human drugs intended to treat serious or life-threatening infections.

**DALBAVANCIN** (Trade name **ZEVEN**) is a novel second generation lipoglycopeptide antibiotic. It belongs to the same class as vancomycin, the most widely used and one of the few treatments available to patients infected with Methicillin-Resistant *Staphylococcus aureus* (MRSA).

Dalbavancin is a semisynthetic lipoglycopeptide that was designed to improve upon the natural glycopeptides currently available, vancomycin and teicoplanin.

It possesses *in vitro* activity against a variety of Gram positive bacteria, including Methicillin-Resistant *Staphylococcus epidermidis* (MRSE). It is a once-weekly, two-dose antibiotic, given intravenous dosage form.

The drug's safety and efficacy were evaluated in 2 clinical trials involving a total of 1289 adults with Acute Bacterial Skin and Skin Structure Infections (ABSSSI). Results showed that Dalbavancin was as effective as vancomycin for the treatment of ABSSSI.

The most common adverse effects identified in the participants in the clinical trials were nausea, headache, and diarrhea and elevations in one of their liver enzymes. The drug label for dalbavancin provides recommendations on dosage adjustment with renal impairment.

**TEDIZOLID** marketed as the phosphate salt (Trade name **SIVEXTRO**) is a novel oxazolidinone with *in vitro* activity against clinically significant susceptible Gram positive pathogens including MRSA and is now approved in both an intravenous and oral formulations. Administered once daily, Sivextro offers an effective, short six-day course of therapy.

Clinically tested in more than 1,300 adults with serious skin infections, Sivextro was found to be as effective as linezolid, an antibiotic presently in use that has a similar mechanism of action. The New Drug Application (NDA) for Sivextro was supported by two global phase-3 studies, which met primary and secondary end points defined by the FDA and the European Medicines Agency (EMA). These studies demonstrated that Sivextro 200 mg administered once daily for six days was statistically non-inferior to 600 mg linezolid taken twice a day for 10 days. In these studies, the adverse event rates were similar for patients treated with Sivextro and linezolid.

Sivextro is indicated for the treatment of Acute Bacterial Skin and Skin Structure Infections (ABSSSI) caused by susceptible strains/isolates of *Staphylococcus aureus* (including Methicillin Resistant and Methicillin Susceptible isolates), *Streptococcus pyogenes* (Group A streptococci), *Streptococcus agalactiae* (Group B streptococci), *Streptococcus anginosus* group (including *Streptococcus anginosus*, *Streptococcus intermedius*, and *Streptococcus constellatus*) and *Enterococcus faecalis*.

The most common adverse reactions for Sivextro are nausea, headache, diarrhea, vomiting, and dizziness. The following warnings and precautions must be taken during the administration of Sivextro:

- 1) The safety and efficacy of Sivextro in patients with neutropenia (neutrophil counts  $<1000$  cells/mm<sup>3</sup>) have been adequately evaluated. In an animal model of infection, the antibacterial activity of Sivextro was reduced in the absence of granulocytes. Alternative therapies should be considered when treating patients with neutropenia.
- 2) *Clostridium difficile* associated diarrhea (CDAD), ranging from mild diarrhea to fatal colitis, has been reported with nearly all systemic antibacterial agents, including Sivextro. Evaluate all patients who present with diarrhea following Sivextro use.
- 3) Prescribing Sivextro in the absence of a proven or strongly suspected bacterial infection or prophylactic indication is unlikely to provide benefit to the patient and increases the risk of the development of drug resistant bacteria.

# MIPPO Vs ORIF with Plate in Distal Tibial Fractures

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## ABSTRACT

### Key words:

Distal Tibial Fracture,  
MIPPO (Minimally  
invasive percutaneous plate  
osteosynthesis),  
ORIF with Plating,  
Open reduction and internal  
fixation.

**Objectives:** To evaluate healing time and complications of distal tibial fractures treated by MIPPO and ORIF with plate.

**Methods:** 30 patients aged 20 - 65 years of either sex with fracture of distal tibia admitted in Fewa City Hospital, Pokhara were randomly divided into two groups. Group A – MIPPO, and Group B – ORIF with plating.

**Results:** For type A and B fractures healing time in both groups were similar but in type C healing takes longer in ORIF group. Similarly delayed union, non-union and infection were seen in ORIF and malunion in MIPPO.

**Conclusions:** It can be concluded from the study that MIPPO is better than ORIF for distal tibial fractures.

## INTRODUCTION

Closed fractures of the tibial shaft traditionally have been treated with closed reduction and a cast. Since the late 1950s, open reduction and internal fixation (ORIF) was reserved for situations in which an adequate reduction could not be obtained or maintained by conservative means. ORIF often necessitates extensive dissection and tissue devitalisation, creating an environment less favorable for fracture union and more prone to bone infection. As a result, other less invasive methods were developed to treat diaphyseal fractures of the tibia. The most successful, closed intramedullary (IM) nailing, has been associated with shorter time to union and a shorter period of disability before working compared with closed reduction and fixation with a cast<sup>1,2,3</sup>. IM nails have been greatly improved in recent years and indications for their use have been extended to

fractures closer to the ankle joint<sup>4,5</sup>.

In recent years, with the development of biological fixation and locking plates, the application of minimally invasive percutaneous plate osteosynthesis (MIPPO) technique in treating extremity fractures has been widely accepted. MIPPO has several advantages compared with traditional treatments. It can avoid direct exposure of metaphyseal fracture lines and adopt indirect induction to preserve vascular per fusion, among others<sup>6-10</sup>. MIPPO has been successfully used in some fractures<sup>11-14</sup>. Several studies reported that MIPPO technique could reduce periosteum damage, provide favorable micro environment for fracture healing, improve indirect bone healing, and reduce fracture healing time<sup>15-19</sup>.

Therefore the present study aims to identify whether the clinical results of MIPPO for Distal Tibial Fractures are better compared with those of traditional open reduction and internal fixation (ORIF) and findings latter evaluated and discussed appropriately. For that purpose we compared the radiographic and clinical results of patients with fractures of the distal third of the tibial shaft, treated with ORIF with plate and those treated with MIPPO.

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## METHODS

The study was conducted at Fewa City Hospital Pvt. Limited, Pokhara, between January 2011 to July 2012. Patients aged 20 - 65, diagnosed with closed distal tibial fracture with or without fibular fracture were included in the study. Open fracture, pathological fracture, head injury were major exclusion criteria for the study. This is a prospective study based on convenient sampling. Operation was performed 1 to 5 days after the incidence. Before the operation, patient was treated with analgesic, antibiotic, serratopeptidase, elevation and ice compression.

### Operative procedure

For all the patients, pre-anaesthetic check-up was performed. Patients were in supine position. Spinal anaesthesia or general anaesthesia was given according to the condition of the patient in most of the cases. Tourniquet was used for open cases only. Semi-tubular plates were used when there was associated fibular Fracture. Antero-medial incision was given. Fracture site was exposed without periosteum stripping. One or two cortical screws used as a lag screw in spiral or oblique fractures. Fracture was completely fixed with a locking plate. In closed group, operation was performed under image intensifier. Plate was inserted percutaneously from distal to proximal in closed group. Alignment and length was maintained under image intensifier. Plate was fixed with locking screws. 6 - 9 screws were applied according to the extent of fracture type. Suction drain was used only in open cases.

### Post operative care

Antibiotic (Inj. Cefazolin) was used at least for seven post-operative days may increase up to 15 according to the wound condition. Drain was removed after 48 hours of operation. Active ankle and range of motion were allowed after 24 hours. Patient was allowed non-weight bearing crutch walking after swelling diminished. After radiographic evidence of callus formation appeared, patient was encouraged to weight bear after partial weight bearing exercise. X-ray was taken after 3 - 6 weeks and 3 months after operation. Other complications like sepsis, skin necrosis were recorded. Evaluations were performed based on the range of ankle motion, limb rotation, fracture healing and radiographic alignment. Healing was evaluated by radiological callus formation and pain-free full weight bearing walking. Healing time less than 6 months was considered normal, 6 - 9 months was considered delayed union and non-union after 9 months. Angulation or rotational deformity of 5 degree or more was considered as malalignment.

## RESULTS

During the study period of 1 year, a total of 35 patients were included. Out of those total patients 13 were in group A (ORIF) and 17 in group B (MIPPO). 5 patients were omitted as they did not come for follow up. In group A with open reduction and plating 8 were males and 5 were females whereas among 17 patients in group B (MIPPO) 13 were males and 4 females. Mean age in group A was 43.4±12.5 and in group B was 44.3±13.2.

Table 1: Sex Distribution

Group A			Group B		
Male	Female	Total	Male	Female	Total
8	5	13	13	4	17

Fractures were classified according to AO classification. Fractures were classified into 3 types: Type A 56.7% (17 patients), Type B 26.7% (8 Patients) and Type C 16.6 % (5 patients).

Table 2: Types of the Fractures

	No of Patients	%
Type A	17	56.7%
Type B	8	26.7%
Type C	5	16.6%

The average follow up time was 16.3 months (13 to 28 months) for Group A and 17.3 months (12 to 30 months) for Group B. In group A, 8 fractures healed within 6 months where delayed union, non-union and wound infection was seen in each fracture but there is no mal-union seen. Similarly, in group B, 12 fractures healed in 6 months where delayed union in 1 and mal-union in 2 fractures. Posterior angulation was seen in both the mal-union. No statistically significant difference seen in the healing time of types A and B of two groups. The healing time of type C in group A was longer than group B ( $p < 0.05$ ).

Table 3: Healing Time in months

	Group A	Group B
Type A	4.99 ± 1.25	5.32 ± 1.29
Type B	6.40 ± 1.72	5.58 ± 0.58
Type C	5.60 ± 1.58	10.32 ± 1.68

Table 4: Complications

	Group A	Group B
Delayed Union	1	1
Non Union	1	0
Malunion	0	2
Infection	1	0

## DISCUSSION

In recent years, various reports have argued that the MIPO technique is a safe method of managing distal tibial fractures, while avoiding some complications associated with conventional open plating methods. Some studies have also revealed defects of the MIPO technique. Hasenboehler *et al* reported that although MIPO seems more advantageous for soft tissue and bone biology, prolonged healing times were observed in simple fracture patterns<sup>20</sup>. Khoury *et al* pointed out that for the MIPO technique, reduction should be performed cautiously due to the tendency of sagittal plane malreduction<sup>21</sup>. Therefore, whether the virtues of the MIPO technique exceed ORIF is

not clear. The MIPO technique requires only realignment of tibial mechanical axis, and clear exposure of the fracture is not necessary. It is a typical method based on biological fixation which involves minimal soft tissue dissection with preservation of vascular integrity of the fracture. The intraoperative image intensifier helps with the closed reduction and avoids excessive disturbance of the fracture fragments. For comminuted fractures, the MIPO technique is particularly advantageous over ORIF. Based on the theoretical considerations, the MIPO technique claims an earlier union process, as well as lower risks of infection, non-union and other complications.

Cheng W *et al* didn't find any significant difference between the MIPO and ORIF for the treatment of distal tibia fractures. They also found out that irritation symptoms were more frequently encountered in the MIPO group<sup>22</sup>. Similar results were reported by Lau *et al* whereby 52% (25/48) of distal tibia fracture cases treated by MIPO had the implants removed due to skin impingement<sup>23</sup>. This could be related to the thin subcutaneous tissue and suboptimal premoulding of plates. Despite the advantages of closed reduction and slight disturbance of soft tissue, MIPO has the disadvantages of non-accurate reduction. The fragments may be not tightly compressed which could increase the risks of delayed union and non-union, especially for simple fractures. Several studies have reported the rate of delayed union or nonunion to be 5–17%<sup>20,24</sup>.

Cadaver research suggests that the MIPO technique may carry a higher risk of injury for saphenous nerve and long saphenous vein<sup>25</sup>. Zou J *et al* concluded that MIPPO was superior to ORIF for treating Type C tibia fractures<sup>26</sup>. The healing time of Type C in the open group was longer than that in the closed group ( $p < .05$ ). The first choice for Type C fractures is MIPPO, whereas that for Type A is open reduction<sup>27</sup>.

In our study we found no difference in healing time in group A and group B in type A and fracture type B. For these fractures both the methods of operation is preferable except there is a long scar in open method. But in case of type C fractures which was comminuted healing time was more in open group than closed one. Less soft tissue entrapping, eliminate many small fragments and periosteal stripping cause faster healing in closed group where as delayed union and non-union seen in open group.

This result was similar to the study done by Collinge and Protzman<sup>28</sup> and Zou and Shi<sup>29</sup>. In their study MIPPO technique showed few problems like delayed union and non-union due to intact periosteum, no anatomical reduction due to small incision, rotational and angular deformity. By this study MIPPO is the first choice in type C distal tibial fractures. For group A fractures MIPPO can try first if not perfect reduction achieved, then switch to open reduction.

## CONCLUSIONS

In conclusion, MIPPO is one of the better choice of treatment of distal tibial fractures than ORIF.

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# Clinico-microbiological Correlation of Vaginal Discharge in Reproductive Age

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## Key words:

Foul smell,  
Gram stain,  
Microorganisms,  
Vaginal discharge.

## ABSTRACT

**Objectives:** The main objective was to study vaginal discharge and its management in relation to commonest microorganisms involved.

**Methods:** This is hospital based prospective, cross sectional study in 100 consecutive cases of reproductive age women (15 - 49 years) from October through December 2013, in Fewa City Hospital, Pokhara, Nepal.

**Results:** The most common age group is 30 - 35 years (33%). The most common finding on direct smear is polymicrobial (53%). The most common type of discharge is mucopurulent (53%).

**Conclusions:** In experienced hand, clinical diagnosis correlates with microbiological findings and patients improved with the medication of WHO regime for syndromic approach to vaginal discharge.

## INTRODUCTION

In health, the vagina has a mixed bacterial flora with unique bacterial community but dominated by Lactobacilli<sup>1</sup>. Many other microorganisms may be present in lower concentration including anaerobic, facultative anaerobic bacteria and *Candida* species<sup>2</sup>. The hormonal environment alters on a monthly basis with additional disturbances to the ecosystem produced by menstruation, hygiene practice and sexual activity. It can introduce a number of new species and pathogens as well as alter pH<sup>3,4</sup>.

A disturbance in the vaginal ecosystem is in which the lactobacilli are replaced by an overgrowth of vaginal commensals. It is recognized by the most common cause of abnormal vaginal discharge in women of reproductive age group. Abnormal vaginal discharge is the most common gynecologic disorder that manifests with an offensive, non bloody discharge per vagina with or without symptoms of itch<sup>5</sup>.

Vaginal discharge is the most common complaint in women of reproductive age<sup>6,7,8</sup>. Microbial agents of abnormal vaginal

discharge and cervicitis include *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, beta haemolytic streptococci, *Ureaplasma urealyticum*, *Candida albicans*, *Trichomonas vaginalis*<sup>9,10</sup> etc.

Although vaginitis or inflammation of the vagina generally is both treatable and mild; when left untreated, it is a possible risk for acquisition of human immunodeficiency viral infection (HIV) and acquired immunodeficiency syndrome (AIDS) as well as other complications<sup>11</sup>. Other complications of vaginal infection include pelvic inflammatory disease with subsequent infertility, ectopic pregnancy, pelvic abscess, menstrual disorders, spontaneous abortion, preterm birth and cancer of the cervix<sup>9,11</sup>.

It is very expensive and complex to carry out gold standard investigation in a country with low resources<sup>12</sup>. At least one microscopic examination of vaginal swab helps to guide for antibiotic treatment against Gram positive and Gram negative bacteria. This will help to avoid empirical treatment<sup>13,14</sup>. Therefore, over and above the desire to relieve the women of unpleasant symptom, there may be a role in prevention by early diagnosis and prompt treatment of this condition.

## METHODS

A hospital based cross sectional study in 100 consecutive cases of reproductive age women between 15 - 49 years with vaginal discharge visiting Gynecology Department from October to

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December 2013 in Fewa City Hospital, Kaski, Nepal.

A clinical inference was drawn on microbiological etiology of infective discharge based on history, nature of discharge and gynaecological examination. Nature of vaginal discharge was analyzed according to subjective assessment by the examining doctor and only mucopurulent, white, creamy, yellowish and greenish in color were enrolled. The posterior and lateral vaginal fornices and cervix were sampled by high vaginal and cervical swabs after exposing the cervix with a sterile, unlubricated cuscus speculum. Samples were processed immediately after collection. The characteristics of the discharge such as quantity, color and smell were recorded and the pH of discharge was determined directly with a pH indicator paper and an immediate fishy odor on addition of 1 - 2 drops of 10% KOH was considered as a positive amine test. Bacterial vaginosis is diagnosed if any three out of five possible criteria were present in the vaginal discharge:

- Vaginal pH > 4.7
- Positive amine test
- Presence of clue cells
- Thin homogenous vaginal discharge
- Presence of Gram positive or negative bacteria

High vaginal swab was obtained and sent for wet mount, whiff test and Gram stain.

Exclusion Criteria: Foreign body, urinary and or feculent discharge, bloody discharge, patients on oral antimicrobial therapy or any form of vaginal medication.

The approval from ethics review committee of the hospital has been obtained for this study.

## RESULTS

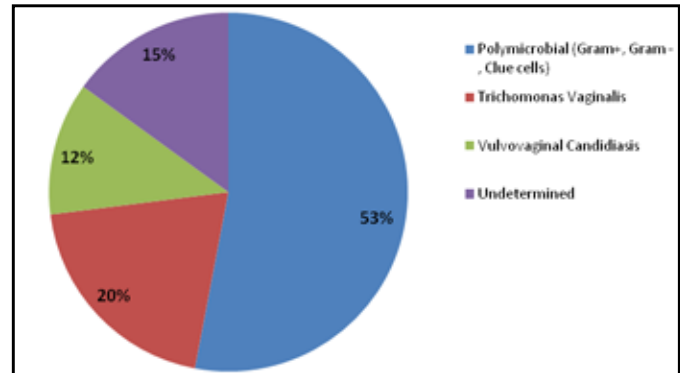
In three months of study period, there were 702 gynaecological cases in our gynaecological Out Patient Department. Among them 250 cases had abnormal vaginal discharge. First 100 cases which had fulfilled the inclusion criteria were enrolled in this study. And 35% had abnormal vaginal discharge as chief complaint.

Table 1: Age wise distribution of the cases (n = 100).

Age	No. of patients (in %)
15 - 20	11
20 - 25	17
25 - 30	21
30 - 35	33
35 - 40	10
40 - 45	6
>45	2
Total	100

The most common age group is 30 - 35 (33%).

Fig 1: Types of infection on direct smear evaluation



Polymicrobial infection was the commonest type of vaginitis (53%) followed by *Trichomonas vaginalis* (20%) and candidiasis being the least common (12%). Among them, two cases turned out to be human immunodeficiency viral infection (HIV) positive and three were diabetic. These patients were diagnosed upon request of additional diagnostic tests based on their histories and complaints at the time of consultation.

Table 2: Distribution of cases on the basis of nature of vaginal discharge (n = 100).

Clinical Diagnosis	Nature of Discharge	Percentage (%)
Mixed Infection	Mucopurulent	53
Bacterial Infection	Milky white	20
Trichomoniasis	Thin brown	15
Candidiasis	Curdy	12

## DISCUSSION

This study was conducted in a small group of women and only in one centre. But this study can be considered as one milestone in the future for other similar studies. This study supports that vaginal discharge is the commonest problem in gynaecological practice and one third of patients were with vaginal discharge in our gynaecological outpatient department.

Noble H<sup>15</sup> found similar type of incidence in his study. The results of this study are similar to the study done by Khan SA<sup>16</sup>. Our results showed predominance of polymicrobial infection as the most common cause of vaginal discharge with the same sample size as in the study by Khan SA<sup>16</sup>. Poor knowledge, lack of hygienic practice and low socioeconomic status were noted to be cause of polymicrobial vaginal infection. The use of non-culture laboratory methods in the initial assessment of abnormal vaginal discharge can be a useful adjunct in the syndromic cases of abnormal vaginal discharge<sup>17</sup>.

Undetermined agents contributed to 15% of cases in this study. Inability to overcome the undetermined causes in this study needed further evaluation by referring to higher centers due to lack of investigation tool in our hospital. It could be possible that a number of women might be suffering from *Chlamydia*, *Ureplasma urealyticum* and other organisms including viruses.

Laboratory diagnosis for *Chlamydia* and viruses are complex and very expensive<sup>14,18</sup>.

Other opinions for undetermined causes of abnormal vaginal discharge is associated with psychosocial problem of women. One of the study carried out in Goa, India found that the psychological problems are associated with abnormal vaginal discharge<sup>19</sup>. It invites further evaluation to establish relation between psychosocial problems and abnormal vaginal discharge or excessive normal discharge. Physical and chemical factors associated with abnormal vaginal discharge were not looked upon by our study tool.

A comparison of Gram stain versus clinical criteria reveals that both are effective for the diagnosis of symptomatic bacterial vaginosis. Gram stain is a common method of assessing bacterial vaginosis in research studies but it requires skilled personnel<sup>9</sup>. Bacterial vaginosis is considered as the most common cause of vaginal discharge. In clinical setting, bacterial vaginosis is diagnosed by the presence of the following criteria described by Amsel *et al*:

- Elevated pH of  $\geq 4.5$
- Thin homologous gray-white discharge.
- Amine odor upon addition of 10% potassium hydroxide to vaginal fluid is equally comparable to high vaginal swab analysis for diagnosis of bacterial vaginosis<sup>6,10</sup>.

Clinical findings and syndromic approach are very effective methods for managing genital tract infections and helps in reproductive enhancement in a country with low socioeconomic status where there is a lack of diagnostic tools and trained man power<sup>12,20</sup>. The syndromic approach does not require identification of one underlying etiology. Instead it is based on identification of syndrome that is a group of symptoms and easily recognized signs associated with a number of well defined etiologies. Treatment is provided for majority of the organisms locally responsible for this syndrome<sup>12,20</sup>. Syndromic management for abnormal vaginal discharge has been recommended by the World Health Organization<sup>11,21</sup>.

## CONCLUSIONS

The complaint of vaginal discharge is very common. An accurate diagnosis is based on the knowledge of epidemiology of lower genital tract infection, consistent application of laboratory tests where needed. Because of limited resources we have to rely on our clinical findings and guidelines for maximum benefits of our patients in the periphery.

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# Use of Polypropylene Mesh for Treatment of Complete Rectal Prolapse: Should We Renounce?

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## ABSTRACT

### Key words:

Rectopexy,  
Polypropylene mesh,  
Rectal prolapse.

**Background:** Controversy exists regarding the preferred surgical technique for treatment of adult complete rectal prolapse. Which surgery is appropriate? Open or laparoscopic, abdominal or perineal, to fix, to use mesh anteriorly or posteriorly, use of absorbable or non-absorbable mesh, to resect, to narrow, to suture or to use natural things e.g., omentum. About 130 operation modifications are available. Insertion of a foreign material during rectopexy is commonly performed with the assumption that this material evokes more fibrous tissue formation than ordinary suture rectopexy. The polypropylene mesh has been criticized for eliciting foreign body reaction resulting in sepsis, gradual erosion and subsequent fistula or other complications.

**Objectives:** This study was done to see demographic, clinical and functional outcome of open abdominal mesh rectopexy in our setup.

**Design:** Retrospective study from 17th July, 2006 to 16th July, 2013 (2063-4-1 to 2070-4-1).

**Setting:** Surgical department, Western regional hospital, Pokhara, Nepal.

**Methods:** The charts of 22 patients (excluding 4 patients which we could not follow up) of abdominal mesh rectopexy were reviewed retrospectively and the patients were followed up in surgical OPD/telephone questionnaire. All patients had undergone standard abdominal mesh rectopexy without the division of lateral ligaments with utmost care for prevention of bleeding and nerve damage. The excluding criteria were age under 20 years, perineal procedures, recurrent prolapse, ASA score 3 and above, high Wexner score for constipation (15 and above), D grade Park's Browning classification to grade fecal incontinence.

**Results:** Out of 22 cases of abdominal rectopexy 4 (18%) were males and 18 (82%) were females. 18 (82%) were within 61 - 80 yrs of age group. The most common symptoms were mass per anum (100%) followed by incontinence for liquid stool and flatus (55%). There was no operative mortality. The average hospital stay 7.8 days (5 - 11 days). The constipation ( $p = 0.019$ ) and incontinence ( $p = 0.0165$ ) were better post operatively. There was 1 (4.5%) partial recurrence of rectal prolapse in 91 year old lady in 2 years follow up.

**Conclusions:** The recurrence rate as well as clinical and functional outcome of open abdominal mesh rectopexy seems to be satisfactory, however long-term effect has to be seen.

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## INTRODUCTION

Rectal prolapse has been known since the Egypt and Greek civilization in 1500 BC<sup>1</sup>. The method of treatment described by Hippocratic Corpus included hanging the person by the heels and shaking the person until the gut returned to its normal

place. After the prolapse was reduced, a 'caustic potash' was applied to the rectal mucosa, and the thighs were bound together for 3 days<sup>1,2</sup>. Rectal prolapse, or procidentia, is defined as a protrusion of the rectum beyond the anus. An occult (internal) rectal prolapse or a rectal intussusception term is used when it does not protrude beyond the anal canal<sup>2,3</sup>. The complete or full thickness rectal prolapse (protrusion of the entire rectal wall) should be distinguished from mucosal prolapse in which there is protrusion of the rectal or anal mucosa only<sup>1,2,4</sup>.

It commonly occurs at the extremes of age. Its incidence in children with cystic fibrosis is about 20%. Females are usual sufferers and peak incidence after fifth decade representing 80% - 90%. More than half of the patients with rectal prolapse have coexisting incontinence<sup>1,4</sup>. In 15% - 65% patients, it is associated with constipation. The solitary ulcer results due to mucosal trauma by straining as it forces the anterior wall of rectum into anal canal<sup>2</sup>. The primary cause of rectal procidentia remains unclear. The abnormal deep pouch of Douglas, lax and atonic pelvic floor and anal canal muscles, internal and external anal sphincter weakness evidenced by pudendal nerve neuropathy<sup>14</sup>, lax lateral ligament and lack of normal fixation of rectum with mobile mesorectum- are considered prerequisites for development of rectal prolapse<sup>6,8</sup>. This is compounded by predisposing factors such as multiparity<sup>7</sup>, neurological illness or connective tissue disorders<sup>6</sup>.

The operative procedure that yields best results with minimum morbidity is argued though more than hundred surgical operation techniques are available to correct this disabling condition. The basic type of surgical approaches to rectal prolapse include<sup>9</sup>-

- Anal encirclement.
- Perineal mucosal sleeve off (Delorme's) or recto sigmoid resection (Altemeier's procedure) with or without pelvic floor repair.
- Laparoscopic (Robotic) or open trans abdominal rectopexy (suture or mesh) or resection or both (Well's, Ripstein, De-hoore, Orr-loygue, Frykman-Goldberg, etc).
- Making a reverse intussusception as Devadhar's concept<sup>10</sup>.

However doubt still remains on the following subjects—

- Operative or non-operative management? Considering the very elderly and infirm.
- Is abdominal or perineal approach better than other?
- Which method of rectopexy is superior?
- Is laparoscopic (robotic) or open method better?
- Is resection and pexy better than pexy alone?
- What are the long term consequences of these procedures<sup>4,9</sup>?
- Is new technique Stapled Trans Anal Rectal Resection (STARR) suitable for rectal intussusceptions (less than 3 cm)<sup>11</sup>?

Insertion of foreign material during rectopexy evokes more intense fibrous reaction than suture rectopexy which may be non-absorbable like nylon, polypropylene, marlex, polytef and absorbable e.g., polygalactin or polydioxanone<sup>12</sup>. Furthermore

its close association with bowel is blamed for sepsis, fistula formation and other complications<sup>4,9,12</sup>. This study does not compare results among various surgical operations but evaluates the recurrence rate, functional outcome of open non absorbable polypropylene mesh rectopexy in our setup.

## METHODS

The medical records of 26 patients operated for open abdominal mesh rectopexy from 17th July, 2006 to 16th July, 2013 (2063-4-1 to 2070-4-1) were reviewed retrospectively. The four patients who could not be followed up were excluded from the study. All patients had mechanical and bacteriological bowel preparation 24 hrs prior to operation. All patients had undergone standard abdominal mesh rectopexy without the division of lateral ligaments with utmost care in preventing bleeding and autonomic neuronal damage.

The exclusion criteria for operation were age below 20 years, perineal procedures, recurrent prolapse, high ASA score (3 and above), Wexner constipation score (15 and above), D grade Park's Browning classification for incontinence<sup>13,14</sup>. The patients were followed up in surgical OPD/ telephone questionnaire when possible. The data was analyzed statistically with the help of online SISA calculator and P value was calculated.

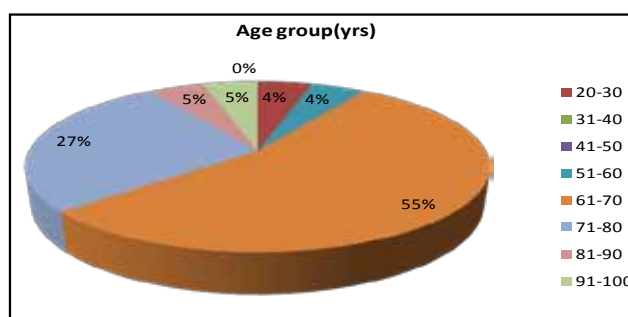
## RESULTS

There were 26 patients of open abdominal mesh rectopexy, in which 4 patients were excluded because of inadequate data and follow up. Out of 22 patients 4 (18%) were males while majority of patients were females 18 (82%). The age distribution of patients is shown in Table 1 and Fig 1.

Table 1: Age distribution

Age group (years)	No. of patients
20-30	1
31-40	-
41-50	-
51-60	1
61-70	12
71-80	6
81-90	1
91-100	1
<b>Total (n)</b>	<b>22</b>

Fig 1: Age distribution



18 (82%) patients were between 61 – 80 years age groups.

ASA scores of 22 patients who underwent open abdominal rectopexy is given in Table 2.

Table 3: ASA Scores

ASA Scores	Clinical status	No of patients
1	A normal healthy patient	17
2	A patient with mild systemic disease.	5
3	A patient with severe systemic disease.	-
4	A patient with severe systemic disease that is a constant threat to life.	-
5	A moribund patient who is not expected to survive without an operation.	-
6	A declared brain dead patient	-

All most all patients were fit and healthy or with minor systemic illness.

Table 4: Symptoms of complete rectal prolapse patients

S. No	Symptoms	No. of cases	%
1	Mass per anum	22	100%
2	Constipation	7	30%
3	Incontinence	15	68%
4	Bleeding	6	29%
5	Mucus discharge	20	95%
6	Solitary ulcer	1	4%

Almost all patients had mass per anum as chief complain making 100%, however the patients had more than one symptom. There was no mortality.

The immediate postoperative complications are shown in Table 5.

Table 5: Immediate postoperative complications

S.No	Complications	No. of patients
1	Retention of urine	4
2	Bleeding	-
3	Postoperative ileus	2
4	Wound dehiscence	1
5	Chest infection	1
6	Wound infection	1

All complications resolved during hospital stay. The hospital stay was 5 - 11 days (mean 8 days). Only one patient of 91 yrs female developed partial recurrence of rectal prolapse after 1 year otherwise there was no recurrence in short term follow up (3 - 24 months).

For the functional outcome the constipation and incontinence were used and 2x2 contingency table was made, the Pearson's Chi-squared test was used and p value calculated (confidence interval 95%) with SISA online statistical calculator.

Table 6: Constipation

S.N	Constipation	Pre-op.	Post-op.
1	Yes	7	1
2	No	15	21
	Total	22	22

P value <0.019.

Table 7. Incontinence

S.N	Incontinence	Pre-op.	Post-op.
1	Yes	15	4
2	No	7	18
	Total	22	22

P value <0.0165.

The recurrence rate was 4.5%.

Fig 2: Open posterior mesh rectopexy



## DISCUSSION

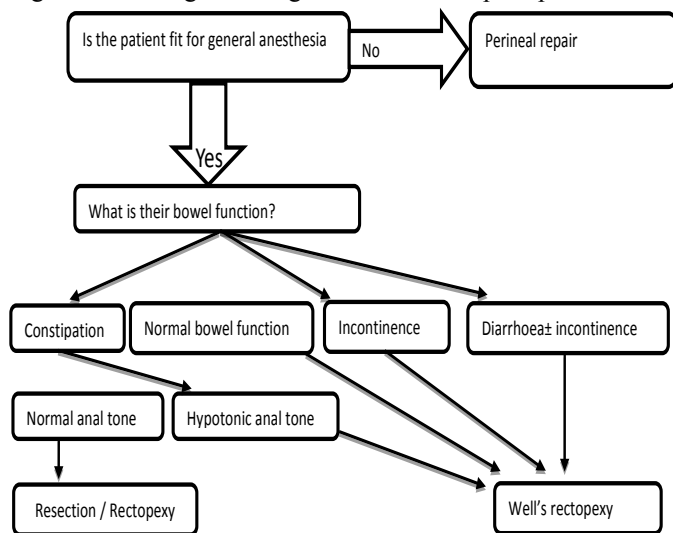
Rectal prolapse is a socially devastating disease, when patients seek for medical advice. Surgery remains the treatment of choice although severely ill or patients with high ASA scores cannot be offered proper treatment. Perineal approaches are reserved for elderly and unfit patients of high anesthetic risk<sup>4,12</sup>. Kaiwa and co-workers after treating 9 elderly patients laparoscopically concluded advanced age is not a contraindication to surgery as high ASA score<sup>15</sup>. The gangrenous or incarcerated rectal prolapse cases were not encountered in our series, demanding for Delorme's or Altemeier's operation, however 3 cases of Theirsch's operations were excluded from the study.

One case of 91 year old female had abdominal rectopexy but she developed recurrence (4.5%) after 2 years. These results are consistent with other reports on recurrence after anterior and posterior rectopexy. Yoshioka and co-workers reported a recurrence rate of 1.5% full-thickness rectal prolapse and 7% mucosal prolapse after posterior rectopexy with Marlex mesh

attached to the sacrum<sup>16</sup>. Marchal and co-workers reported a 4% recurrence rate 14 – 276 months after Orr–Loygue rectopexy<sup>17</sup>. In this series of patients, one of three patients experienced a recurrence 14 months after simple rectopexy, and one of 12 prolapses (8%) recurred 2 years after resection rectopexy. Molen *et al* reported 0% recurrence after performing posterior mesh rectopexy in 18 patients in 42 months of follow up<sup>12</sup>. Prospective studies of Galli and Rabbu reported 3% recurrence rate in 37 patients<sup>12</sup>. Similarly, Aitola reported 6% recurrence rate<sup>18</sup>.

Abdominal operation has lower recurrence and better functional outcome than perineal procedures. The use of polyvinyl alcohol has been absolute because of increased infection rate<sup>5,12</sup>. Similarly, Ripstein procedure was blamed for aggravation of constipation. In patients with redundant sigmoid colon along with history of constipation, resection rectopexy seems to be better option<sup>5,12</sup>.

Fig 3: The management algorithm for rectal prolapse<sup>20</sup>



As retrograde autonomic nerve damage occurs after the division of lateral ligaments, the problem of constipation is aggravated postoperatively. The choice of division or preservation of lateral ligaments depends upon surgeon's experience and further study is needed to access its efficacy<sup>5,12</sup>.

Laparoscopic surgery has the advantages of less pain, shorter hospital stay, early recovery, and early return to work as compared with laparotomy<sup>13</sup>. Apart from these advantages, the results are similar to those with the open procedures irrespective of the method used (suture, resection, or posterior mesh). Therefore, where expertise is available, this approach may be preferred<sup>4,12,13</sup>. Suture and mesh rectopexy are still popular with many surgeons, and the choice depends on the surgeon's experience and preference.

## CONCLUSIONS

Being a retrospective observational analysis, it limits the scientific conclusion. The recurrence rate as well as clinical and functional outcome of open abdominal mesh rectopexy seems to be satisfactory, however long-term effect has to be seen. This

may be due to avoidance of high score Wexner constipation and Park's incontinence score. Old is gold? Should we change to other method for the management of adult complete rectal prolapse (in terms of cost), the question remains to be answered.

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# Minimal Invasive Plate Osteosynthesis of Distal Tibia Fractures

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## Key words:

Distal tibia,  
Locking plate,  
Osteosynthesis,  
Ankle joint.

## ABSTRACT

**Background:** Fractures of the distal third of tibia and its plafond are complex injuries that present a challenge as to their best method of management. The involvement of the ankle joint and the vulnerability of the surrounding soft tissues further complicate these already complex injuries. Minimally invasive plate osteosynthesis (MIPO) is becoming a popular method of treating these injuries when there is no articular comminution and the soft tissue envelope is minimally disturbed.

**Methods:** Sixteen patients with tibial pilon fractures were treated by MIPO using an anatomical distal tibial locking plate, at Gandaki Medical College Teaching Hospital, Pokhara, Nepal from August 2009 to January 2011. Six patients had associated skeletal injuries as close lateral malleolar fracture. Two patients had a grade I open fracture.

**Results:** All fractures united at an average of 16 weeks. All patients had near to normal range of movement of contralateral ankle joint and had normal walking pattern with full weight bearing at 10 months of follow-up. Two patients had a superficial wound infection, 2 patients complained of prominence of the plate.

**Conclusions:** Minimally invasive plate osteosynthesis is a good and safe technique for treatment of distal tibial fractures without intra-articular comminution providing fracture healing, rapid functional recovery, and avoidance of major complications.

## INTRODUCTION

Fractures of the distal third of tibia and its plafond are complex injuries. The involvement of the ankle joint and the vulnerability of the surrounding soft tissues further complicate these already complex injuries. Most of these fractures are managed with an operative intervention such as closed reduction and intramedullary interlocking (IMIL) nailing or open reduction and internal fixation (ORIF) with plating<sup>12</sup> or closed reduction and per cutaneous plating or external fixators. Each of these techniques has their own merits and demerits. IMIL nailing has been reported with higher rate of malunion because it is difficult to achieve two distally locking screws<sup>1-3</sup>.

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Wound infection, skin breakdown and delayed union or non union requiring secondary procedures like bone grafting are some of the complications associated with conventional osteosynthesis with plates<sup>4-7</sup>. Similarly, pin tract infection, pin loosening, malunion and nonunion leading to osteomyelitis is potential complication of external fixators and hence not preferred as definitive fixation method<sup>8-11</sup>. Minimally invasive plate osteosynthesis (MIPO) with locking plate is becoming a popular method of treatment. Our aim of this study is to evaluate the radiological and functional outcome of fracture of distal third of tibia treated with MIPO in our setting.

## METHODS

This was a prospective, observational study carried out in eighteen months duration (August 2009 to January 2011) at Gandaki Medical College Teaching Hospital, Pokhara, Nepal. Total number of patients were 16. Six were females and 10 were males. Distal tibia including pilon fracture in 22 to 70 years of

patients presenting within two weeks of injury were included in this study. Compound Gustillo type II / type III fracture and grossly comminuted intraarticular fracture AO type C3 were excluded from the study. Clinical evaluation for the soft tissue condition, swelling and neurovascular status was done. Radiograph of affected leg including ankle anteroposterior, lateral and mortise views were taken. According to the AO-OTA classification, eight fractures were type A, and other eight fractures were type C (C1 = 3 and C2 = 5). Twelve patients had an associated fibular fracture. Ten patients were operated on within 48 hours and next six patients were postponed to allow their swelling to decrease.

**Surgical technique**

Prophylactic antibiotics Cefazoline 1 gm injection was given intravenously about half an hour before an incision was given. Lateral malleolus was fixed through a lateral approach in 10 cases. In the type I compound fractures, wound lavage and debridement was done before reduction and fixation. Provisional fixation of the articular fragments was done percutaneously with pointed reduction clamp and K wires. Lag screws used for definitive fixation. The appropriate length of the pre-contoured locking plate was determined. About 2 - 3 cm incision was made centering and just proximal to the medial malleolus. Subcutaneous extraperiosteal tunnel was created for the insertion of the plate.

Fig 1: Incision is made over medial malleolus region.



Fig 2: Tunnel is made for passage of plate



About 3-4 cm incision was made over the proximal end of the plate.

Fig 3: various sizes of locking plates and drill sleeves



Fig 4: Passage of plate through tunnel



Fully threaded 4 mm cancellous locking screws was passed through the distal end of the plate to fix the articular fragments.

Fig 5: Drilling done for proximal screws



Post-operatively, AP and lateral radiographs were made to check the fracture reduction and the congruency of articular surface of the ankle. The leg was elevated and gentle active/passive mobilization was started.

The patients were reviewed at 2 weeks, 6 weeks, 12 weeks, 18 weeks then every 3 months during the first year. During follow up visits, the patient was examined clinically and X rays anteroposterior and lateral views of the leg and ankle were obtained. Partial weight bearing was allowed when radiological union was evident on X rays, progressing to full weight bearing over one month or as patient tolerated. Clinical results were graded as Excellent (>92 points), Good (87 - 92 points), Fair (65 - 86 points) and Poor (<65 points) using Teeny and Wiss clinical assessment criteria<sup>12</sup> using 100 points score.

Fig 6: Pre-op radiograph of AO Type A distal tibia fracture with lateral malleolus



Fig 7: operated wound before skin closure



Fig 8: Postop radiograph and wound



## RESULTS

There were 16 patients including 10 males and 6 females. Age of patients varies from 22 years to 70 years with mean age being 44.12.

Table 1 : Demographic profile and outcome

S. No.	Characteristic		Values
1.	Age		44.12 (22-70) years
2.	Sex	Male	10
		Female	6
3.	Mode of trauma	RTA	8
		Fall from height	6
		Rotational injury	2
4.	Involved limb Side	Right	9
		Left	7
5.	AO Type	Type A	8
		Type C	8
6.	Healing time of fracture (in weeks)	Radiological union	16.5 (12-24)
		Clinical union	18.62 (12-30)
7.	ROM of Ankle (in degree)	Dorsiflexion	21.44 (13-28)
		Plantarflexion	29.69 (20-35)
8.	Full weight bearing walking (in Months) without crutches		5.31 (3-8)
9.	Results	Excellent	10
		Good	4
		Fair	2

All fractures were united at an average of 16 weeks. As regards of pain, 10 patients (62.5%) were pain-free, 4 patients (25%) had only occasional pain after stressful activities and not requiring medication, 2 patients (12.5%) had mild to moderate pain requiring analgesics. According to the clinical rating system by Teeny and Wiss, regarding range of movement 10 patients (62.5%) had Excellent, 4 patients (25%) had Good results, 2 patients (12.5%) had Fair results.

Few patients developed minor complications; two patients had a superficial wound infection that responded to medical treatment and dressing. Four patients had complained of prominence of the plate and one elderly female with thin skin had dehiscence of skin with exposure of two screws.

Fig 9: a) prominence of implant b) skin dehiscence over implant



## DISCUSSION

The results of operative treatment are dependent on the severity of the initial injury and the quality and stability of the reduction. The mechanism of injury, the status of the soft tissues and the degree of comminution affect the long term clinical result. The mean interval for radiological union was 16 weeks in our study is comparable to previous study done by Mahajan<sup>13</sup>. However, the most important factor is to achieve stable fixation and to allow early range of motion without unnecessary osseous and soft tissue devascularization<sup>14</sup>. Minimally invasive techniques are based on principles of limited exposure, indirect reduction methods and limited contact between bone and implant. As a result of these principles, this technique, as seen in present study, avoided major soft tissue complications and shortened the length of the patient's stay in the hospital<sup>15,16,17</sup>. In our study average time to full weight bearing walking without crutches was 5 months; it was almost similar to Hazarika *et al*<sup>18</sup>. The bone healing was excellent with this type of fixation because the stresses were distributed over a longer segment of bone. This technique can be used in fractures where locked nailing cannot be done like vertical slit and markedly comminuted fractures. There was rapid fracture consolidation due to preserved vascularity. There were fewer incidences of delayed union and non-union<sup>13</sup>. There was decreased need for bone grafting. There was less incidence of infection due to limited exposure. There were less chances of refracture. There was no chance of vascular complication by carefully inserting the plate submuscularly through limited incisions. There was no

need of any specialized instrumentation and the method was less time consuming and cost effective in the present study in a similar fashion as reported by previous authors<sup>19,20</sup>. With the introduction of Locking Compression Plates (LCP), minimally invasive techniques have become widely used. The plates act as internal fixators in a bridging manner, thus resulting in secondary bone healing<sup>9,18</sup>.

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# Isoflurane Vs Halothane: A Comparative Study of Haemodynamic Response due to Carbondioxide Pneumoperitoneum During Laparoscopic Cholecystectomy

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## ABSTRACT

### Key words:

Inhalational agent,  
CO<sub>2</sub> pneumoperitoneum,  
Isoflurane,  
Halothane.

**Objectives:** To evaluate Isoflurane Vs Halothane to the obtained haemodynamic response during laparoscopic cholecystectomy under balanced anaesthetic technique.

**Methods:** 30 patients aged 20-60 years of either sex belonging to ASA grade I and II scheduled for elective laparoscopic cholecystectomy admitted in Fewa City Hospital, Pokhara, were randomly divided into two groups.

Group I - O<sub>2</sub>: + Inhalational agent (Isoflurane 1.5 - 2 %)

Group II - O<sub>2</sub>: + Inhalational agent (Halothane 1 - 1.5 %)

**Results:** Hypertensive response due to CO<sub>2</sub> pneumoperitoneum was well suppressed by Isoflurane (1.5 - 2 %) {Group I} which maintained pulse rate at a relatively higher side than halothane, (1 - 1.5 %) {Group II} decreased mean arterial pressure more significantly than halothane without any difference in arterial oxygen saturation (SPO<sub>2</sub>).

**Conclusions:** It can be concluded from the study that Isoflurane (Group I) more effectively attenuated the haemodynamic response during laparoscopic cholecystectomy as compared to Halothane (Group II) under balanced anaesthetic technique.

## INTRODUCTION

Laparoscopic cholecystectomy is a relatively new surgical procedure which is enjoying ever increasing popularity and presenting new anaesthetic challenges. Though they are visually minimally invasive to the patient, the intraoperative requirement of laparoscopic surgery produces significant physiological changes, unique to the procedure. So far no anaesthetic agent has been used successfully to blunt these changes produced

due to CO<sub>2</sub> pneumoperitoneum. Volatile anaesthetics nowadays play an important role in the management of haemodynamic stability during a standardized balanced technique because of their ease of administration and predictable intraoperative characteristics. The extent of haemodynamics changes associated with laparoscopic cholecystectomy depends on the interaction of factors viz. exogenous gas insufflation, patient position and results of increased intra-abdominal pressure.

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- i) Patient Position- For initial trocar insertion patient is placed in trendelenburg position 10 - 20° which results in increase in central blood volume, decrease vital capacity. Then the patient is placed in supine reverse trendelenburg (20 - 30°) which improves diaphragmatic function, pressure, mean arterial pressure (MAP) and cardiac output (CO).

- ii) Neurohormonal effect of exogenous insufflating gas viz- CO<sub>2</sub>, N<sub>2</sub>O, helium. Absorption of CO<sub>2</sub> from the peritoneal cavity is the potential mechanism for hypercarbia and rise in end tidal carbon dioxide concentration (ETCO<sub>2</sub>). Haemodynamic alteration occurs only if PaCO<sub>2</sub> is increased by 30% above normal levels which causes rise in the pulse rate (PR), systemic vascular resistance (SVR), blood pressure (BP), central venous pressure (CVP).
- iii) Different mechanisms viz. pooling of blood in legs, inferior venacaval compression decrease venous resistance, decrease venous return, and cardiac output. On the other hand increased intra thoracic pressure, vascular resistance of intra-abdominal organs increases systemic vascular resistance (SVR) and hence mean arterial pressure (MAP). Release of neurohormonal factors viz. vasopressin, catechol amines also play a role in increasing mean arterial pressure (MAP).

## METHODS

30 patients aged 20 - 60 years of either sex belonging to ASA grade I and II scheduled for elective laparoscopic cholecystectomy under balanced general anaesthesia admitted in Fewa City hospital Pvt Ltd, Pokhara, from April 2007 to March 2010. After premedication with intravenous glycopyrrolate (0.2 mg) and I/V midazolam (0.3 mg/kg), I/V fentanyl (1 - 2 mg/kg), the patients were induced by I/V propofol (1.5 - 2 mg/kg). Volatile anaesthetic was then switched on before surgery. Intermittent positive pressure respiration was carried out to maintain normocarbia. The patients were randomly divided into two groups.

Group I: O<sub>2</sub> + Isoflurane (1.0 – 1.5% vaporizer concentration) + nondepolarizing muscle relaxant (vencurium 0.1 mg/kg)

Group II: O<sub>2</sub> + halothane (1.0 – 1.5% vaporizer concentration) + nondepolarizing muscle relaxant (vencurium 0.1 mg/kg).

For maintenance of anaesthesia, inhalation agent was delivered by Akoma vaporizer. Haemodynamic parameters like changes in mean pulse rate, mean arterial pressure, arterial oxygen saturation (SpO<sub>2</sub>) were recorded before induction (control value), at induction, at 5 min, 15 min, 30 min, 45 min after volatile anaesthetic is switched on after CO<sub>2</sub> pneumoperitoneum is created, after CO<sub>2</sub> removal (deflation), at the end of surgery and postoperatively at 15 min, and 30 min.

## RESULTS

Data was analysed and results were compared by student 't' test to determine the 'p' value. A 'p' value less than 0.05 was considered significant whereas 'p' value of <0.01 was taken as highly significantly.

Table 1: Demographic profile

	Group A (Isoflurane)	Group B (Halothane)
Age (yrs) Mean	48.21	49.32
Range	25 – 60	25 – 60
Sex ratio (M:F)	1.2 : 1	1.32 : 1
Body weight (kg)	61.92	64.2
Range	45 – 75	45 - 75

Above data shows group wise distribution of age, sex and weight. There was no significant difference between the two groups.

Table 2: Indications for laparoscopic cholecystectomy

Indications	Group A (Isoflurane)	Group B (Halothane)
Chronic cholecystitis with cholelithiasis	7	8
Chronic cholecystitis with cholelithiasis with jaundice	5	5
Acute cholecystitis with cholelithiasis	2	1
Mucocele of gall bladder	1	0
Empyema of gall bladder	0	1

Data shows 48% of patients in Group I and 56% in Group II had indication of chronic cholecystitis with cholelithiasis.

Table 3: Mean pulse rate (bpm)

Groups	Group I (Isoflurane)	Group II (Halothane)
Before induction (control value)	76.2 ± 3.23	74.92 ± 3.25
Induction	96.84 ± 8.92	94.52 ± 3.73
Volatile anaesthetic switched on (incision)		
5 min (CO <sub>2</sub> insufflated)	87.71 ± 2.37	78.9 ± 8.2
15 min	80.44 ± 3.66	67.64 ± 9.22
30 min	80.83 ± 3.89	62.6 ± 9.87
45 min	78.21 ± 3.82	58.5 ± 3.99
After CO <sub>2</sub> removal (Deflation)	76.70 ± 3.31	72.92 ± 1.95
End of Surgery	94.89 ± 8.21	96.89 ± 6.31
Post op 15 min	87.6 ± 1.11	88.39 ± 2.99
Post op 30 min	74.82 ± 2.79	72.11 ± 2.79

In Group I (Isoflurane) the mean pulse rate increased gradually from the control value. This rise in pulse rate was statistically insignificant (P is greater than 0.05) at 15 min, 30 min and at 45 min after the volatile anaesthetics is switched on. There was a gradual fall in mean pulse rate in Group II (Halothane) 5 min after volatile anaesthetics were switched on the pulse rate decreased from control value at 15 min, 30 min and at 45 min. This change was statistically significant (P is less than 0.05) at 15 min and highly significant (P is less than 0.001) at 30 min and 45 min intra operatively). Volatile anaesthetics were switched



off 5 - 7 minutes before the end of operation. Isoflurane (Group I) maintained the pulse rate towards higher side as compared to Halothane (Group II).

Table 4: Mean arterial pressure

Groups	Group I (Isoflurane)	Group II (Halothane)
Before induction (control value)	90.23 ± 4.16	92.33 ± 4.66
Induction	114.91 ± 5.33	112 ± 5.63
Volatile anaesthetic switched on (incision)		
5 min (CO <sub>2</sub> insufflated)	112.32 ± 3.66	130.23 ± 3.66
15 min	106.01 ± 4.44	128.64 ± 4.33
30 min	108.32 ± 3.92	126.33 ± 3.64
45 min	101.23 ± 4.23	124.62 ± 4.25
After CO <sub>2</sub> removal (Deflation)	112.78 ± 4.11	114.32 ± 2.12
End of Surgery	110.45 ± 2.44	124.22 ± 2.97
Post op 15 min	94.00 ± 2.33	112.66 ± 2.22
Post op 30 min	90.21 ± 3.38	96.22 ± 9.24

In this study, data shows change in mean arterial pressure at various time intervals in the two groups. In Isoflurane group, 5, 15, 30 and 45 minutes after the volatile anaesthetics was switched on, the Mean Arterial Pressure (MAP) increased. This change in MAP was statistically insignificant (P >0.05) when compared to control value. In Halothane group the MAP increased in 5, 15, 30 and 45 minutes. The increase in mean arterial pressure was statistically significant at 15 min (P <0.05) and highly significant (P <0.01) at 30 min and 45 min, when compared to control value. Thus, Isoflurane lowered the mean arterial pressure more as compared to Halothane.

Table 5: Arterial oxygen saturation

Groups	Group I (Isoflurane)	Group II (Halothane)
Before induction (control value)	97.2 ± 1.6	97.33 ± 1.7
Induction	98.21 ± 1.5	98.21 ± 1.2
Volatile anaesthetic switched on (incision)		
5 min (CO <sub>2</sub> insufflated)	98.3 ± 1.1	96.4 ± 1.2
15 min	97.4 ± 1.3	98.2 ± 1.1
30 min	97.5 ± 1.2	96.4 ± 1.2
45 min	97.2 ± 1.3	97.6 ± 1.1
After CO <sub>2</sub> removal (Deflation)	98.1 ± 1.2	97.7 ± 1.6
End of Surgery	97.8 ± 1.3	98.1 ± 1.1
Post op 15 min	98.00 ± 1.4	98.00 ± 1.8
Post op 30 min	97.6 ± 1.2	98.4 ± 1.6

Data shows the changes in arterial oxygen saturation (SPO<sub>2</sub>) in both groups. After the volatile anaesthetic was switched on there was significant change (p <0.05) in arterial oxygen saturation (SPO<sub>2</sub>) in both the groups at any time interval intra-operatively.

Table 6: ECG

	Group I (Isoflurane)		Group II (Halothane)	
	No. of patients	%	No. of patients	%
Sinus bradycardia	-	-	2	14%
Sinus tachycardia	1	7%	-	-
ST segment changes	-	-	1	7%
Ectopic changes	-	-	1	7%

Table 6 shows ECG change in lead II. Sinus bradycardia, ST segment changes and ectopic changes were observed in Group II (Halothane) which were transient and disappeared on withdrawal of inhalational agent.

Table 7: Complications of postoperative period

Complications	Group I (Isoflurane)	Group II (Halothane)
Nausea / Vomiting	2 patients	1 patient
Shoulder pain	1 patient	2 patients
Postoperative analgesic requirement I/V fentanyl (1 - 2 mg/kg)	10 patients	12 patients

Table 7 shows the complications of postoperative period. Nausea and vomiting was observed in 2 patients in Group I (Isoflurane) while 1 patient in Group II (Halothane). Shoulder pain was reported in 1 patient in Group I while in 2 patients in Group II. Injection fentanyl (1 - 2 µg/kg) was given in post-operative period. 10 patients in Group I whereas 12 patients in Group II required immediate postoperative analgesia.

Table 8: Recovery pattern

Recovery pattern	Time (min)	Time (min)
Opening eyes on command	4.42 ± 1.48	7.59 ± 1.50
Recall of time	5.10 ± 1.44	6.30 ± 3.01
Response to painful pinch	4.25 ± 1.97	5.25 ± 2.24
Handgrip	6.90 ± 1.52	8.20 ± 3.01
Spontaneous movement	3.60 ± 1.66	4.40 ± 2.16

Recovery from anaesthesia was judged after extubation by asking the patients to open eyes on command, recall on name, response to painful pinch, spontaneous movement and handgrip. This was done at intervals of every one minute after discontinuing the inhalational anaesthetic. It was seen that recovery times are factors with Isoflurane.

## DISCUSSION

For the past four decades halothane has been, by far the most commonly used volatile anaesthetic agent. However, as Smith<sup>1</sup> has pointed out, Halothane possesses some properties which fall short of the properties which an ideal anaesthetic agent should have. First it sensitizes the heart to endogenous and exogenous catecholamines and secondly, there is a rare but potentially lethal association with hepatitis. After about a quarter of a century, during which halothane remained virtually unchallenged as the safest and most versatile volatile anaesthetic, to agents which have a particularly wide role to play in contemporary hospital anaesthetic practice in attenuating response to CO<sub>2</sub> pneumoperitoneum during laparoscopic cholecystectomy. There is no correlation of age, sex and type of surgical procedure. Rise in pulse rate was observed in both the groups because of CO<sub>2</sub> pneumoperitoneum. Halothane lowered it significantly due to its negative chronotropic effect on SA node. Isoflurane maintained it towards a higher side due to its beta sympathomimetic activity but at clinically acceptable levels. Skovsted<sup>2</sup>, Seagard *et al*<sup>3</sup>, Kotrly *et al*<sup>4</sup> have reported that Isoflurane is more often associated with an increase in heart rate than Halothane. Two factors may be involved. Firstly, Isoflurane may have less effect on the rate than Halothane as changes in heart rate mediated by baroreceptors.

Kissin<sup>5</sup> suggested that Isoflurane provides a greater margin of haemodynamic safety than Halothane. The more favorable cardiac index of Isoflurane is likely to result from that, although Isoflurane decreases myocardial contractility, it does less than either Halothane or Enflurane (Beapure<sup>6</sup> *et al*). Hypertensive response was well suppressed with Isoflurane (1.5 – 2%) which decreased the mean arterial pressure (MAP) due to its vasodilating effect. Egers<sup>7</sup> speculated that both the volatile agents decrease systemic arterial pressure in a dose related fashion but by different underlying mechanism. Haemodynamic changes like cardiac performance were depressed by Halothane more than that by Isoflurane (McKinney MS<sup>8</sup>). Halothane and Isoflurane both produce a dose dependent decrease of systolic and diastolic function. In our study there is no significant difference in arterial oxygen saturation (SPO<sub>2</sub>) was found. There is change in specific ST segment in Halothane group. These findings are similar with Bornjak<sup>9</sup>. Atlee and Peterson<sup>10</sup> suggested that Halothane reduce conduction within the myocardium not with Isoflurane. Postoperatively Isoflurane group has better result like early recovery and faster wakeup from anaesthesia which was similar with Pandit *et al*<sup>11</sup>. Result of this study is almost similar with the study of Sethi C Jain A<sup>12</sup>.

## CONCLUSIONS

It can be concluded from the study that Isoflurane more effectively attenuated the haemodynamic response during laparoscopic cholecystectomy as compared to Halothane under balanced anaesthetic technique.

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# Incidence of Tuberculous Lymphadenitis

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## ABSTRACT

### Key words:

Tuberculosis,  
Lymphadenitis,  
FNAC,  
Ziehl-Neelsen's staining.

**Background:** Nepal is a high-burden country for tuberculosis (TB). About 45% of the total population is infected with TB and an estimated 20,000 new infectious cases of TB are reported each year.

**Objectives :** The purpose of this study is to determine the incidence of tuberculous lymphadenitis in comparison with other diagnosis of enlarged head and neck nodes by fine needle aspiration cytology (FNAC).

**Methods :** This is a retrospective study of 126 patients with neck swellings in Gandaki Medical College, Pokhara. FNAC was performed. The slides were then stained and evaluated under the microscope.

**Results :** This study showed that lymphadenitis of tuberculous nature (necrotizing granulomatous lymphadenitis consistent with tuberculosis and granulomatous lymphadenitis suggestive of tuberculosis) were found in 35 patients out of 126 (27.78%) which was the second most common diagnosis and other non tuberculous lesions (reactive lymphadenitis, non Hodgkin's lymphoma, metastatic carcinoma to lymph nodes and lymphangioma) were found in 91 patients (72.22%).

**Conclusions :** Our study showed that incidence of tuberculous lymphadenitis in this region of Nepal is not very rare and is easy to be diagnosed due to access to very easy procedure like FNAC. We believe this information would be useful in many clinical settings and facilitate pathological reporting and focused clinical investigation under the current Nepalese perspective especially in the rural areas. Considering the frequency of tuberculous lymphadenitis of the head and neck region in our part of Nepal we also believe that in the future, this data will help further to diagnose and treat the patients timely and accurately.

## INTRODUCTION

Nepal is a high-burden country for tuberculosis (TB). About 45% of the total population is infected with TB and an estimated 20,000 new infectious cases of TB are reported each year<sup>1</sup>. Tubercular lymphadenopathy is the most common extra pulmonary form of tuberculosis and cervical lymphnodes are the most commonly affected group of nodes<sup>2</sup>. The purpose of this study was to determine the incidence of tuberculous lymphadenitis in enlarged head and neck nodes by FNAC.

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Clinical examination and often diagnostic aids like radiology and blood tests fail to resolve the vexing problem regarding the nature of mass. Fine Needle Aspiration Cytology (FNAC) is a procedure where by small amount of tissue or cells is aspirated from a pathological lesion with the help of fine disposable syringe. Lymph node aspiration is of great value for the diagnosis of lymphadenitis, lymphomas and metastatic carcinoma<sup>3,4</sup>. Sensitivity and specificity of FNAC have been documented by several studies in the past<sup>5,6</sup>. This procedure was first done by Griey and Gray in 1904, in patients with sleeping sickness<sup>7</sup>. The experiment of fine needle aspiration (FNA) developed gradually, until 1921, when Guthrie tried to correlate FNA results with various disease process<sup>8</sup>. The main benefit of FNAC is to avoid the need for surgical biopsy, which requires local or general anesthesia, increased hospital stay and costs<sup>9</sup>.

## OBJECTIVES

To determine the incidence of tuberculosis affecting the neck nodes.

**Design:** Retrospective study

**Place and duration of study :** This study was carried out in Gandaki Medical College, Prithivi Chowk, Pokhara, Nepal. It started from January 2008 till 2010 January, lasting two years.

## METHODS

All the patients presenting with palpable cervical lymph nodes (CL) in the Hospital were recruited in the study regardless of age and sex. The patients with enlarged CL but having contraindication of FNAC (Bleeding disorder, cardio respiratory failure) were excluded from the study. All patients were asked detail history pertaining to neck swelling and relevant questions to the etiological factors and past family history of tuberculosis. Apparatus used included 10 ml disposable plastic syringe 22–25 gauge, 0.6–1.0 mm external diameter disposable needle 3.98 cm and 8.8 cm long with or without stylet, antiseptic sponges, sterile gauze pads, microscopic glass slides.

All FNAC were carried out by pathologists. A 23-gauge needle was connected to a 10 ml syringe mounted on a syringe holder. The palpable cervical node was fixed with one hand and needle was inserted into the lymph node and a full suction pressure was applied. The tip of the needle was moved around at varying angles and depths and with constant negative pressure (never emerging outside the skin). Before final withdrawal, the negative pressure was released prior to the needle emerging from the skin. The cytological material was transferred on to glass slides. The aspirated material was then smeared on 2 - 4 slides, fixed in 95% ethanol and air dried to be stained with papanicoloau and May-Grunwald Giemsa stains respectively. The slides were then evaluated by the pathologist. Suspicious cases for tuberculosis in microscopic findings were subjected to Zeihl Neelsen stain for AFB (TB). The data analysis was performed using SPSS version 17.

## RESULTS

In this study 126 patients were involved. Among them males were 67 (53.17%) and females 59 (46.83%). Mean age was 24.93 years (SD±20.0). Age ranged from 1 year to 86 years. The age distribution of the study population is shown in Figure 1.

Patients with age group 0 - 20, 21 - 40, 41 - 60 and 61 - 80 and above 80 years were 53.97%, 29.37%, 7.14%, 7.94% and 1.59% respectively (Fig 1). Most of the patients were between the age 0 - 20 years. Males were higher in frequency (n=67 ; 53%) than females (n=59; 47%).

FNAC was done from various sites. From neck lymph nodes it was 93 (74%), supraclavicular lymph nodes 9 (7%), submandibular lymph nodes 9 (7%), submental lymph nodes 6 (5%), preauricular lymph nodes 5 (4%) and postauricular lymph nodes 4 (3%) as shown in the figure 2.

This study showed that lymphadenitis of tuberculous origin were found in 35 patients out of 126 (27.78%) and other

nontuberculous lesions were found in 91 patients (72.22%), as shown in figure 3.

Among the lymphadenitis of tuberculous nature, 19 patients were diagnosed with necrotizing granulomatous lymphadenitis consistent of tuberculosis (15.1%) and granulomatous lymphadenitis suggestive of tuberculosis was 16 (12.7%) of all the 126 patients. Reactive lymphadenitis 78 (61.9%), non Hodgkin's lymphoma 5 (4.0%), metastatic carcinoma to lymph nodes 7 (5.6%) and lymphangioma was 1 (0.8%) were other diagnosis made in the cytopathological study among these patients (Table 1).

Cases in microscopy showing caseous necrosis, epithelioid cell granulomas, Langerhans giant cells in a lymphoid background were diagnosed as Necrotising Granulomatous Lymphadenitis (NGL) whereas cases with all the above findings but devoid of caseous necrosis were diagnosed as granulomatous lymphadenitis. Few of the NGL cases showed positivity for acid fast bacilli in Ziehl Neelsen Stain (Fig 4 and 5).

Fig 1: Age group of patients

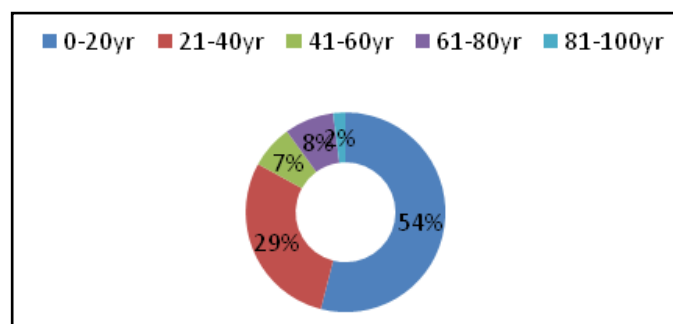


Fig 2: Sites of FNAC of cervical lymphnodes (n=126)

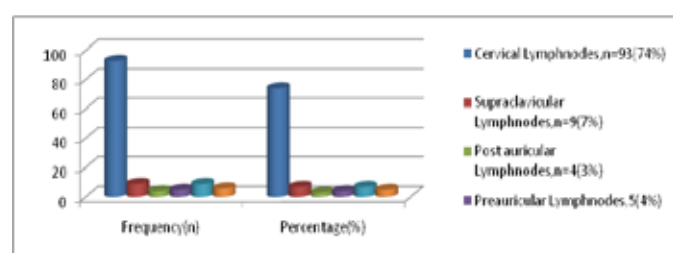


Fig 3: Incidence of Tuberculous and Nontuberculous diseases

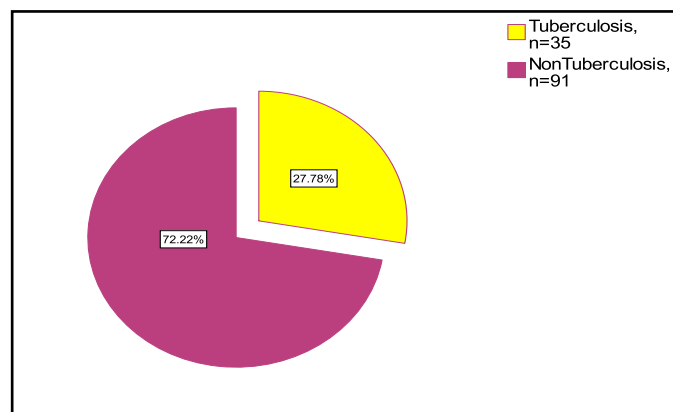


Fig 4: A. Epithelioid cell Granuloma; B. Caseous necrosis

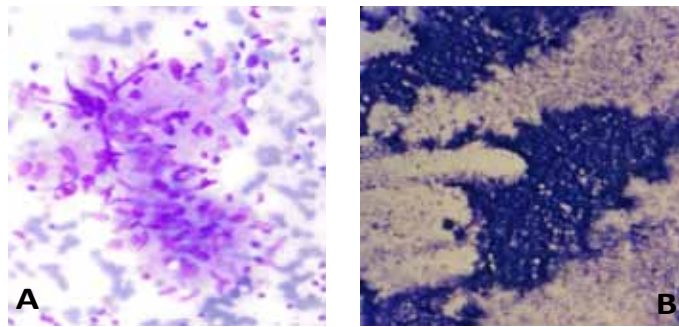


Fig 5: C. Langerhans giant cell; D. Acid fast bacilli seen in Ziehl Neelsen stain

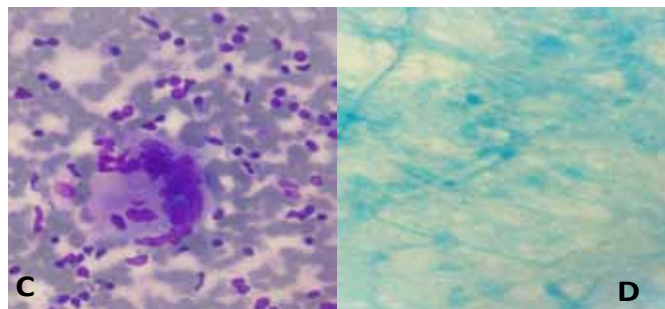


Table 1: Result of FNAC of cervical lymph nodes

Diagnosis	Number of patients	Percentage (%)
<b>Tuberculous nature</b>		
Necrotising Granulomatous Lymphadenitis, consistent with Tuberculosis	19	15.1
Granulomatous Lymphadenitis, suggestive of Tuberculosis	16	12.7
<b>Nontuberculous nature</b>		
Reactive Lymphadenitis	78	61.9
Non Hodgkin's Lymphoma	5	4.0
Metastatic Carcinoma to Lymphnodes	7	5.6
Lymphangioma	1	.8
Total	126	100.0

**DISCUSSION**

This study was carried out to find out the relative frequencies of tuberculosis in the lymphnodes in the neck in our region. Diagnostic FNAC was performed. This technique is safe simple and quick with a low complication rate and helps to select people preoperatively for surgery<sup>10</sup>. The FNA for cytological evaluation of neck masses was first reported by KUM in 1947<sup>11</sup>.

In present study, the age of patients ranged from 1 to 86 years with mean of 24.93 years and SD±20.0 years. Cervical lymph

nodes were most commonly affected (74%). Males (53.17%) were more affected than females (46.83%) and the male to female ratio was 1 : 0.88. We found that majority of patients in our study (53.97%) were from 0 - 20 years age while in similar study by Shakya G showed that age group 21 - 40 were mostly affected<sup>12</sup>. Necrotising granulomatous lymphadenitis, consistent with tuberculosis along with granulomatous lymphadenitis suggestive of tuberculosis was found to be the second most common pathology in our study (27.8%). Similarly, the studies by Tariq *et al*<sup>13</sup>, ElHag *et al*<sup>14</sup>, Kamal F *et al*<sup>15</sup> and Muhammad J *et al*<sup>16</sup> showed that the incidence of tuberculosis of neck nodes were 36%, 21%, 13% and 26.1% respectively. A comparison between these studies are illustrated in the Table 2.

Table 2 : Comparison of results with other studies

Authors	Current study	Tariq <i>et al</i> <sup>13</sup>	El Hag <i>et al</i> <sup>14</sup>	Kamal F <i>et al</i> <sup>15</sup>	Muhammad J <i>et al</i> <sup>16</sup>
Year of publication		2008	2003	1996	2006
No. of patients	126	50	225	847	42
Duration of study (years)	1.5	1	5	1	1
TB Lymphadenitis (%)	27.7	36	21	13	26.1
Reactive Lymphadenitis (%)	69.1	18	33		16.6
Malignant neoplasms (%)	4	14	13	11	4.6

In most studies an accuracy rate of FNAC in diagnosing lymph node lesions of 85% to 94.4% has been reported<sup>9,17</sup> while sensitivity of FNAC in the detection of tubercular lymphadenopathy was reported to have 77%<sup>18</sup>. Sensitivity, specificity and diagnostic accuracy was reported to be 97%, 97.5% and 97.4% respectively in yet another study in which, cytomorphological features of epithelioid and giant cells with caseation necrosis was associated with higher percentage of AFB positivity<sup>19</sup>. Early diagnosis is particularly important in tubercular lymphadenopathy due to its curability.

Our study also showed that more than half the cases (61.9%) were reactive lymphadenitis which is similar with the study by Mohammad R, Azadeh R<sup>20</sup>, in which 58.9% of the lesions were reactive lymphadenitis. AlAlwan *et al*<sup>21</sup> and Narang *et al*<sup>22</sup> also showed reactive lymphadenitis in 55.3% and 61.6% of the lymph nodes, respectively<sup>21,22</sup>.

Non Hodgkin's Lymphoma was seen in 7 (5.6%) in our study. Similar finding was shown in study by Haque MA 6 (5.6%)<sup>23</sup>. Metastatic carcinoma to lymph nodes was found in 5.6% cases. In another large study, nasopharyngeal carcinoma was reported as most frequent primary site in cervical metastatic lymphadenopathy<sup>6</sup>. In the diagnosis of metastatic malignancy, the lymph node FNAC is as rewarding as the surgical biopsy. Lymphangioma was seen only in 1 case.

**CONCLUSIONS**

Our study showed that incidence of tuberculous lymphadenitis in this region of Nepal is not very rare and is easy to be diagnosed due to access to very easy procedure like FNAC. It has reported the frequency of tuberculous lymphadenitis in one of the Western part of Nepal. It also showed different patterns of cytological diagnosis on FNAC of enlarged cervical lymph

nodes among Nepalese population in this region. Despite the limitations, FNAC provides a reliable and convenient method for the initial management of cervical lymphadenopathy specially in case of tuberculosis as it is medically treatable disease. One can always avoid surgical procedures and come to the correct and timely decision of treating medically treatable diseases. We believe this information would be useful in many clinical settings and facilitate pathological reporting and focused clinical investigation under the current Nepalese perspective. Looking at the results of frequency of tuberculosis in the neck region in our part of Nepal we also believe that in the future this data will help further to diagnose and treat the patients timely.

#### Conflicts of interest

The authors declare that they have no competing interests.

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# Femoral Artery Variations in Nepalese Cadavers

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## ABSTRACT

### Key words:

Femoral artery,  
Anatomic variation,  
Medial and lateral circumflex  
arteries.

**Background:** Femoral artery is a continuation of external iliac artery below inguinal ligament. It is an important artery with clinical importance.

**Objectives:** The objective of the study is to find out the prevalence of various anatomic variations of femoral artery in Nepalese.

**Methods:** Around 106 lower limbs of both sides were dissected in Anatomy dissection hall and any variations of femoral arteries were observed and noted down.

**Results:** It was found that lateral and medial circumflex artery directly arising from femoral artery in 30% and 36% cases and from profunda femoris artery in 70% and 64% respectively.

**Conclusions:** The lateral circumflex artery had directly arisen from femoral artery in 30% and from profunda femoris artery in 70% cases. The medial circumflex artery had directly arisen from femoral artery in 36% cases and from profunda femoris artery in 64% cases.

## INTRODUCTION

The femoral artery is the primary artery of lower limb. It is a continuation of external iliac artery below inguinal ligament. The branch of femoral artery which supply thigh is profunda femoris artery which gives off medial circumflex artery, lateral circumflex artery and perforating arteries. The femoral artery continues as popliteal artery in adductor hiatus and run into popliteal fossa where it divides into tibial and peroneal artery. The tibial artery supplies back of leg and terminates by dividing into medial and lateral plantar arteries which supplies sole of foot. The peroneal artery supplies front and lateral part of leg and dorsum of foot.

The anatomical knowledge of variations of femoral artery and its branches is important as it is frequently accessed by surgeons and radiologists. The femoral artery is generally

preferred and easily accessible for catheterization in number of investigations and angiographies. The awareness of variation in height of origin of profunda femoris artery and the distribution of its branches are of great importance. Knowledge of variation of lateral circumflex femoral artery is important when undertaking clinical procedures in the femoral region and hip joint replacement. It can be used as an anterolateral thigh flap in coronary artery bypass grafting. The lateral circumflex femoral artery flap is used for the reconstruction of large defects in the face secondary to gunshot wounds.

The present study was undertaken since there was no study of femoral artery variations in Nepalese, so far. The findings of study will be useful for general surgeons, radiologists, plastic surgeons, anatomists etc.

## METHODS

Around 106 dissected lower limbs of both sides of Nepalese cadavers were studied from 2003 to 2013 in dissection halls of B. P. Koirala Institute of Health Sciences, Manipal College of Medical Sciences and Gandaki Medical College. The anatomic variations in lower limbs were noted down.

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### Correspondence:

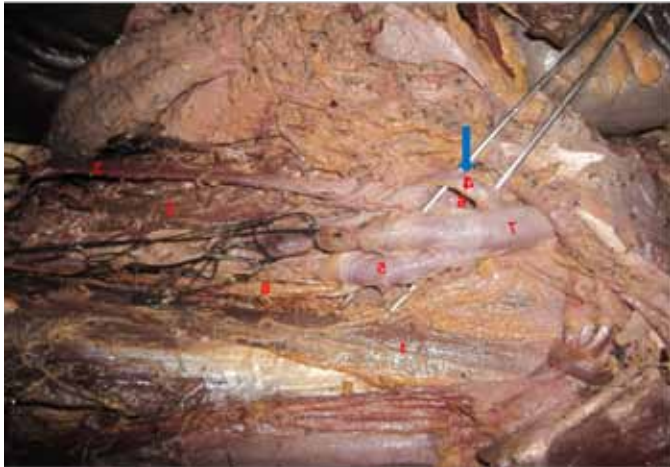
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## RESULTS

Around 106 lower limbs of both sides were observed. It was found that lateral and medial circumflex artery directly arising from femoral artery in 30% and 36% cases respectively (Fig 1 and 2). Other types of variations could not be noted in Nepalese cadavers.

Fig 1: Variation of femoral artery- medial circumflex artery directly originating from femoral artery



- 1) Sartorius 2) Adductor longus 3) Great saphenous vein 4) Medial circumflex femoral artery 5) Profunda femoris artery 6) Femoral vein 7) Femoral artery 8) Femoral nerve

Fig 2: Variation of femoral artery- lateral circumflex artery directly originating from femoral artery



- Sartorius 2) Adductor longus 3) Lateral circumflex femoral artery 4) Medial circumflex femoral artery 5) Profunda femoris artery 6) Femoral vein 7) Femoral artery 8) Femoral nerve

## DISCUSSION

Prakash *et al* observed that the medial femoral circumflex artery in 43 out of 64 (67.2%) extremities originated from the profunda femoris artery; whereas in 21 out of 64 (32.8%) extremities it originated from the femoral artery. On the other hand, the lateral femoral circumflex artery in 52 out of 64 (81.25%) extremities originated from the profunda femoris artery, whereas 12 out of 64 (18.75%) extremities, it originated from the femoral artery<sup>1</sup>.

The present study observed that lateral and medial circumflex artery directly arising from femoral artery in 30% and 36% cases and from profunda femoris artery in 70% and 64% respectively.

However Siddhartha P *et al* found the medial circumflex artery originated from the deep femoral in 63% of the specimens. It arose separately from the common femoral and more proximally in 26% of the specimens<sup>2</sup>.

Siani A *et al* observed an unexpected anatomical variant of the femoral artery in a patient with acute lower limb ischemia. In this anomaly, the deep femoral artery arises from the external iliac artery, 2 cm above the inguinal ligament, runs with a parallel course with the superficial femoral artery, and placed between the branches of femoral nerve<sup>3</sup>.

Satheesh Nayak B *et al* reported unusual branches of femoral artery in the femoral triangle. One of the rare but clinically important variations was the origin of deep circumflex iliac and inferior epigastric arteries from femoral artery bilaterally instead of from external iliac artery<sup>4</sup>.

Atulya *et al* found a variation in origin and branching pattern of lateral circumflex femoral artery. Double origin of lateral circumflex artery from lateral and anterior aspects of profunda femoris artery was observed respectively in the left lower extremity of a 50 year old male cadaver<sup>5</sup>.

## CONCLUSIONS

The present study of 106 lower limbs of Nepali cadavers showed variations of femoral artery branching system. The lateral circumflex artery had directly arisen from femoral artery in 30% and from profunda femoris artery in 70% cases. The medial circumflex artery had directly arisen from femoral artery in 36% cases and from profunda femoris artery in 64% cases.



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# Immunostimulatory and Anti-tumor Activity of Aqueous Extract Isolated from *Cnidium monnieri* Fruits

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## ABSTRACT

### Key words:

*C. monnieri* fruits extract,  
Immunostimulation,  
Macrophages.

**Background:** Immunostimulants are the biological response modifiers which are designed as alternative drugs to amplify the host's immune system against the infections and cancer cells.

**Objectives:** The objective of the research is to investigate the immune modulating activity of *Cnidium monnieri* fruits extract *in vitro*.

**Methods:** RAW 264.7 mouse macrophages were used. Macrophages were treated with either LPS or various concentrations of *C. monnieri* fruits extract. Nitric oxide (NO) was detected from the cell supernatant. Macrophages phagocytosis and tumoricidal activities were investigated.

**Results:** The extract significantly enhances the production of nitric oxide (NO), phagocytosis and tumoricidal activities of RAW 264.7 mouse macrophages.

**Conclusions:** *C. monnieri* fruits extract significantly enhances the activity of RAW 264.7 mouse macrophages *in vitro*. The augmented tumoricidal effect of *C. monnieri* fruits extract treated macrophages was believed due to the production of nitric oxide. Therefore, *C. monnieri* fruits extract is estimated as a novel immunostimulating herbal extract which can be developed as an alternative drug to recruit the host's immune system against various pathogens as well as cancer cells.

## INTRODUCTION

Immunostimulants, the biological response modifiers (BMR), are designed as an alternative drug to amplify immune system of the host against infectious pathogens and tumor cells. Many aqueous extracts isolated from plants are relatively nontoxic in nature. Therefore, extensive research has been conducted to explore their ability to modulate immune function both *in vitro* and *in vivo*<sup>1</sup>. In particular, these extracts have been shown to enhance macrophage cytotoxicity against microorganisms and cancer cells, stimulate phagocytosis activity, enhances secretion of NO, TNF- $\alpha$ , IL-6, IL-12 INF- $\gamma$  and so on. Thus, plant extracts

are ideal candidates for therapeutics with immunomodulatory and antitumor effects<sup>1,2</sup>.

Macrophages are multifunctional cells that play a major role in host defense against infection and cancer by connecting innate immune system with adaptive immune system. Pathogenic stimuli, injury and cancer cells activate the macrophages to release numerous pro-inflammatory cytokines (like TNF- $\alpha$ , IL-1), NO, chemokines, inflammatory molecules and prostaglandins<sup>3,4</sup>. Once they are activated, they act as effectors cells and involved in the reorganization and eradication of foreign pathogens including tumor cells directly via phagocytosis<sup>5</sup>. Thus, the purpose of this study was to investigate the immunomodulatory effect of *C. monnieri* fruits extract on macrophage RAW 264.7 cells and to present *C. monnieri* fruits extract as a novel Biological Response Modifier (BRM).

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## METHODS

The chemicals were obtained from the following sources: Dulbecco's Modified Eagle's Medium (DMEM), RPMI 1640 and fetal bovine serum (FBS) were obtained from GIBCO (Grand Island, NY, USA), penicillin, streptomycin and amphotericin B from Sigma-Aldrich, USA. 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazoliumbromide (MTT), lipopolysaccharide (LPS), were purchased from Sigma Chemical Company. NO detection kit was obtained from INTON biotechnology, INC. Phosphate buffer saline 10× was obtained from Bioplus Fine Research Chemicals and Neutral red was obtained from Showa Chemicals INC, Japan.

### Plant materials

*C.monnieri* fruits extract used in this study was supplied by the college of Pharmacy, Chungbuk National University, South Korea.

### Cells and treatment

The RAW 264.7 mouse macrophages and CT-26 colon cancer cells were obtained from the American Type Culture Collection (ATCC) and grown in DMEM and RPMI medium respectively, supplemented with 10% heat inactivated fetal bovine serum (FBS) and antibiotic solution (containing 10,000 units/ml of penicillin, 10 mg/ml streptomycin and 25 µg/ml of amphotericin B) at 37°C in an air containing 5% CO<sub>2</sub>/95% air atmosphere.

*C. monnieri* fruits extract was dissolved in the medium. 100 µl of macrophages (1×10<sup>6</sup> cells/ml) were seeded in 96 well plate for 24 hrs and *C. monnieri* fruits extract (10, 30 and 100 µg/ml) was added while, negative control was treated with medium only and incubated for further 24 hrs. 100 µl of MTT (1 mg/ml) was added and further incubated for 3 hrs, the supernatant was aspirated, and 100 µl of dimethyl sulphoxide was added to each well to dissolve the remaining MTT-formazan crystals. Absorbance was measured at 540 nm by ELISA reader<sup>6</sup>. Data are expressed as percentage of control.

### Determination of NO production

The macrophages were seeded as above. After 0, 6, 12, 24, 48 and 72 hrs, the nitrite (the stable reaction product of NO with molecular oxygen) accumulated in cell supernatant was measured by Griess reagent and used as an indicator of nitric oxide (NO) production in medium. 50 µl of culture supernatants were mixed with equal volume of Griess reagent (1 : 1 ratio of 0.1% N-[1-naphthyl]-ethylenediamine dihydrochloride and 1 % sulfanilamide in 5% phosphoric acid) in 96-well plate and incubated at room temperature for 10 min. The absorbance was measured at 545 nm by ELISA reader. Nitrite concentrations in culture supernatants were measured to assess NO production in macrophages. NaNO<sub>2</sub> was used as standard to calculate nitrite concentration<sup>2</sup>. The concentration of nitrite (µM) was calculated from a standard curve drawn with known concentration of sodium nitrite dissolved in water.

### Phagocytosis assay

Phagocytosis assay of the *C. monnieri* fruits extract activated macrophages were analyzed by neutral red uptake method. Macrophages were seeded as above. Cells were stimulated with LPS (0.1 µg/ml) or various concentration of the extract (10, 30, and 100 µg/ml) for 24 hrs. To evaluate the neutral red phagocytosis by macrophages, 100 µl of 0.075% aseptic neutral red solution was added and incubated for 1 hr. The supernatant was discarded and the cells were washed twice with PBS. Then 100 µl of cell lysate solution (ethanol 50% and 1% acetic acid at the ratio of 1 : 1) was added into 96-well plate to lyse cells at room temperature for 2 hrs. The optical density at 540 nm was measured by ELISA reader.

### Tumoricidal activity of macrophages

Tumoricidal activity of macrophages was based on an assay described elsewhere<sup>7</sup>. The cells were seeded as above and were washed twice with PBS. They were then co-incubated with the CT 26 colon cancer cells (1×10<sup>4</sup> cells/well); an initial effector/target cell ratio of 10 : 1). The cells were treated with various concentration of *C. monnieri* fruits extract (10, 30, and 100 µg/ml) for 24 h. The cell density was evaluated by MTT assay. The cytolytic activity is expressed as the percentage tumor cytotoxicity as follows:

$$\% \text{ Cytotoxicity} = \left\{ \frac{\text{O.D. of [(target+macrophages) - macrophages]} - \text{O.D. of target (nontreated)}}{\text{O.D. of target (nontreated)}} \right\} \times 100$$

### STATISTICAL ANALYSIS

Experimental results were expressed as means ± SD. Statistical significance was calculated by two tailed student's *t*-test. *P*-values of less than 0.05 were considered to be statistically significant.

## RESULTS

### 1. Effects of *C.monnieri* fruits extract on macrophage RAW 264.7 cells

The cytotoxic effect of *C. monnieri* fruits extract in macrophages was measured by MTT assay. The extract showed no cell toxicity at the tested doses. Thus, we treated cells with the extract ranging from 10 - 100 µg/ml during subsequent experiments (Fig 1).

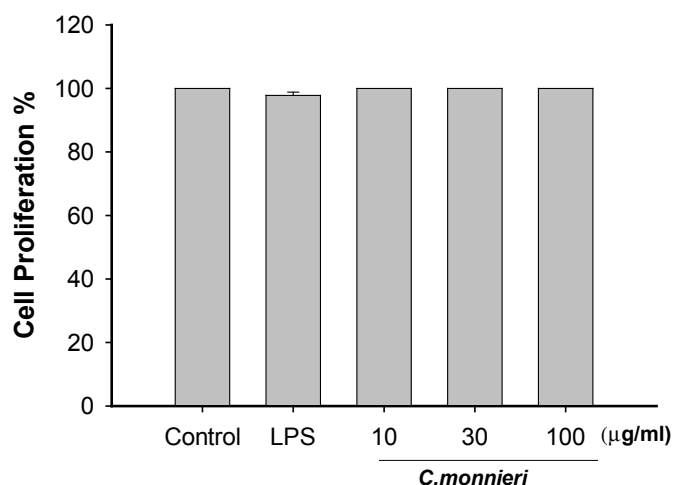


Fig 1: Effect of *C. monniieri* fruits extract on the cells viability. Cells were incubated with various concentrations of *C. monniieri* fruits extract (10- 100 µg/ml). Each bar represented as means  $\pm$  S.D.

## 2. Immune stimulating effects of *C.monniieri* fruits extract

To investigate whether the production of NO by *C. monniieri* fruits extract was time-dependent, macrophages were cultured for 6 - 72 hr at a concentration of 100 µg/ml of the extract. As shown in Fig 2, at time as early as 12 hr, the level of NO induced by the extract was higher ( $p < 0.001$ ) than untreated control and continued to increase sharply up to 48 hrs. After 48 hrs saturation state was achieved till 72 hrs of incubation.

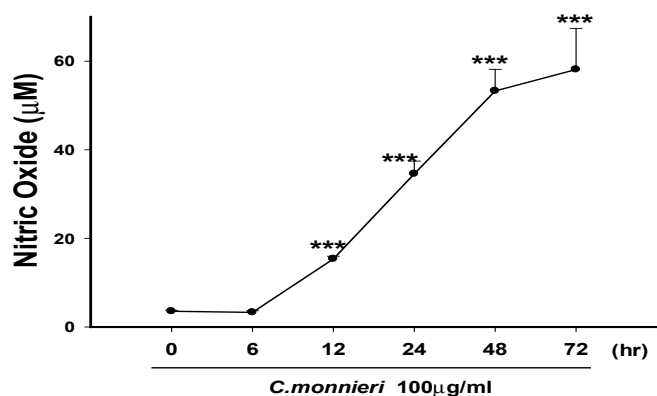


Fig 2: Effect of *C.monniieri* fruits extract on the time dependent production of NO from macrophages. The data are represented as means  $\pm$  S.D., where \*\*\* $p < 0.001$ , as compared to control.

## 3. Effect of *C.monniieri* fruits extract on phagocytosis activity

The phagocytic activity of macrophages was monitored by measuring the amount of neutral red internalized by macrophages. Results showed that 100 µg/ml of the extract significantly increased the phagocytosis of macrophages as compared to untreated control ( $p < 0.05$ ). Moreover, the

phagocytosis percentage of the extract 100 µg/ml was slightly higher than that of positive control, which was treated with 0.1 µg/ml of LPS (Fig 3).

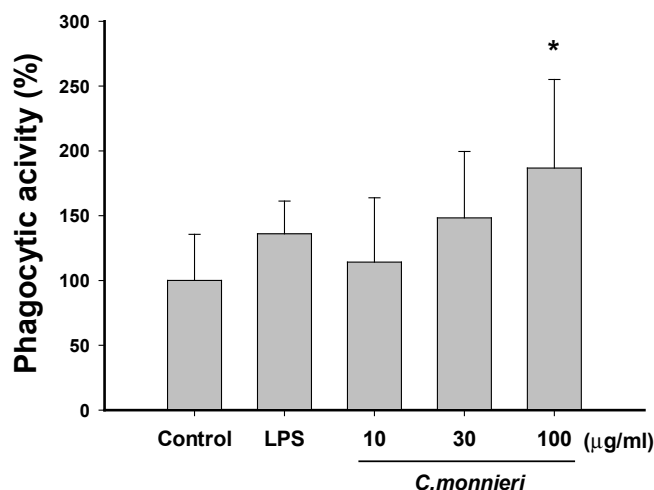


Fig 3: Effect of *C. monniieri* fruits extract on the phagocytosis of neutral red by macrophages. LPS 0.1 µg/ml was used as positive control. Each error bar represented as means  $\pm$  S.D., where \* $p < 0.05$ , as compared to control.

## 4. Effect of *C. monniieri* fruits extract on tumoricidal activity of macrophages

Here macrophages were considering as effectors and CT 26 colon cancer cells were used as the targets because they are either sensitive to TNF- $\alpha$  or NO. *C. monniieri* fruits extract significantly increased the cytostatic activity of the macrophages against the CT 26 colon cancer cells at the tested concentrations by enhancing the synthesis of tumoricidal molecules from the macrophages (Fig 4).

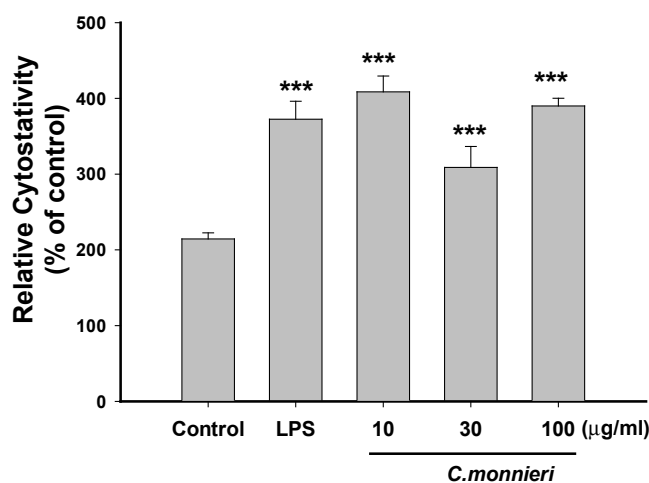


Fig 4: Effect of *C. monniieri* fruits extract on cytostatic activity of macrophages. Each error bar represented as means  $\pm$  S.D., LPS 0.1 µg/ml was used as positive control, where\*\*\* $p < 0.001$  as compared to control.

## DISCUSSION

Our experimental results suggested that *C. monnieri* fruits extract significantly and time dependently stimulated the production of NO, and enhanced phagocytotic activities of macrophages. The enhancements in the phagocytosis activity indicated the final and most indispensable step of the host's immunological defense system against the tumor cells<sup>8</sup>. Therefore, the activated macrophages are considered as one of the important components of the host defense system<sup>9</sup>.

NO is a gaseous molecule synthesized from L-arginine by nitric oxide synthase. It is a highly reactive free radical and it can form a number of oxidation products such as NO<sup>2</sup>, NO<sup>2-</sup>, N<sub>2</sub>O<sub>3</sub>, and S-nitrosothiols. It is an important mediator of the non-specific host defense against invading microbes and tumors<sup>10</sup>. Thus, NO can be used as a quantitative index of macrophage activation<sup>2,11</sup>. *C. monnieri* fruits extract (100 µg/ml) stimulated macrophages to produce NO in time dependent manner. Our results are in agreement with Cheng *et al*<sup>12</sup> where glycyrrhiza polysaccharide (100µg/ml) released the NO from peritoneal macrophages in time dependent manner till 0 to 72 hrs of incubation. Moreover, *C. monnieri* fruits extract was not toxic to the RAW 264.7 macrophages.

*C. monnieri* fruits extract significantly enhanced the cytolysis activity of the RAW 264.7 macrophages against those cancer cells but the cytolysis activity of *C. monnieri* fruits extract was not in a dose dependent manner. On the basis of these *in vitro* results we estimated that *C. monnieri* fruits extract is a novel immunostimulant.

## CONCLUSIONS

On the basis of our *in vitro* investigation we concluded that *C. monnieri* fruits extract can activate the macrophages. Thus, *C. monnieri* fruits extract can be developed as a novel Biological Response Modifier (BRM).

## Acknowledgment

We thank Professor Sung Yeon Kim and Mi Kyeong Lee for their support in conducting the entire research.

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# Retrospective study of Ureterorenoscopy +/- Procedure in Gandaki Medical College Teaching Hospital, Pokhara

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**Key words:**

Ureteric calculus,  
ureterorenoscopy,  
Lithotripsy.

## ABSTRACT

**Back ground:** Urinary calculus is one of the most common diseases in urology as the development of small caliber scopes and lithocast, the treatment of ureteric calculus changed from open surgery to minimally invasive surgery as ureterorenoscopic lithotripsy. Since September 2010, Gandaki Medical College Teaching Hospital starts ureterorenoscopic (URS), ureterorenoscopic lithotripsy (URSL) and other procedure.

**Objectives:** Objective of our study is to find out the result of URS/URSL of our hospital and compare it with the study of other parts of the world.

**Methods:** From September 2010 to August 2011, were treated 122 patients with URS 126 times by a single urologist. The result of URS was analyzed and compared with different studies of the world.

**Results:** Most of our patients were aged between 21 to 40 years with single stone in lower or mid ureter. We had success of insertion of URS without any dilation method is 99.2%. For ureteric calculus post URSL, we got stone free rate of 96.3% with stone migration rate of 3.7% and complication rate of 8.8% as perforation rate of 4%, conversion rate of 3.7%.

**Conclusions:** URS/L is safe and choice of management for lower and mid ureteric calculus.

## INTRODUCTION

Urinary calculus is one of the most common diseases in urology. As the awareness to health in general population and the development of new diagnostic technique increasing, the incidence of urinary calculus also increased. Till German's Chaussy in 1980 used first lithotripsy for urinary calculus, open surgery was only one procedure for urinary calculus. As the development of small caliber scopes as ureterorenoscope (URS) and strong lithocast, minimally invasive treatment has replaced open surgery for the treatment of urinary calculus. These days more than 90% of urinary calculus managed without open surgery. Ureterorenoscopic lithotripsy (URSL)

is very popular for the treatment of ureteric calculus as it has good success rate with less morbidity, less hospital stay and less anesthesia requirement. Similarly, with the help of URS it become quite easy to diagnose ureteric disease and carried out the procedure according to its disease.

In Gandaki Medical College Teaching Hospital, open surgery or DJ stenting was common procedure for ureteric calculus due to lack of trained man power till August 2010. From September 2010, we started URS and procedure for urological problem.

From September 2010 to August 2011 in Gandaki Medical College Teaching Hospital, about 31% of total general surgery surgeries were of urological surgeries of which about 34% were urinary calculus related surgeries. Among these urinary calculus related surgery, about 87% were done without open surgery which is comparative to other parts of the world.

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## OBJECTIVES

Objective of our study is to find out the result of URS/URSL of our hospital and compare it with the study of other parts of the world.

## METHODS

It is a prospective observational study done by single urologist in Gandaki Medical College & Teaching Hospital from September 2010 to August 2011. All cases who underwent URS/URSL for different disease was included in the study. Both sexes with different ages, procedure, site-size-side-number of stone, stone clearance and complications were analyzed. For the insertion of URS no any dilatation method was used, DJ stenting was done only in those patients with inflamed ureter, ureteric stricture, stone >10mm in size and stone migration during procedure. For URSL patients KUB X-ray was done 2 weekly post operations till patient was free from stone up to 6 weeks, if stone is still present after 6 weeks then another procedure was carried out.

## RESULTS

From September 2010 to August 2011, altogether 122 patients were treated 126 times with URS/URSL by single urologist. The age of our patients varies from 14 years to 75 years and most common age group of 21-30 years one. Average age was 32.7 yrs. Detail results can be seen in Table 1, 2 and 3 as below.

Table 1: Disease to perform URS/URSL

Diseases		No	Total
Calculus	Upper	14	108 + 2 (1 with stricture)
	Mid	36	
	Lower	60	
Stricture		2	2
Inflammation		2	2
Migrated DJ		1	1
NAD	Pre op suspected of stone	2	10
	1		
	2		
Suspected stricture		5	
Hydroureteronephrosis			
Ureteric colic with HDN			
			Total 125*

(\*all together 122 patients of which 126 procedures were done; successful only in 125 procedures (1 failed to insert) as some we did bilaterally. Each side consider as single patient)

Table 2: List of procedures done

Procedures	
URS +ICPL	107 + 2 (B/L)
Diagnosotic URS	13 + 2 (B/L)
Migrated DJ stent removal under URS (failed, refer for removal by PCN)	1
Failure of insertion (conservative Rx, stone passed spontaneously after 3 weeks)	1

Table 3: Complications and management during procedure

Complications:
<ul style="list-style-type: none"> <li>• <u>Perforation</u>: 5 (Upper 1, mid 1, lower 3)               <ul style="list-style-type: none"> <li>- converted to open ureterolithotomy 2 (lower)</li> <li>- DJ stenting 3 (1 Urinoma formation detected post op 7 days)</li> </ul> </li> <li>• <u>Stone migration</u>: 4 (Upper 3, mid 1)               <ul style="list-style-type: none"> <li>- open Pyelolithotomy 1</li> <li>- Ureterolithotomy 1</li> <li>- DJ stenting 2 (stone passed during removal)</li> </ul> </li> <li>• <u>Retained stone</u>: 4 (Upper 1, mid 2, lower 1)               <ul style="list-style-type: none"> <li>- stones passed after 4 wks 2 ( DJ stenting 1)</li> <li>- Passed after 6 wks 2</li> </ul> </li> <li>• <u>Re-admission for Pain</u>: 3 (upper 1, lower 2)               <ul style="list-style-type: none"> <li>- conservative Rx 2</li> <li>- Urinoma formation, DJ stenting 1</li> </ul> </li> </ul>

Stone diseases were commonest cause to perform URS with most of patients having single stone of size between 5 - 10mm. We got success of insertion of URS without any dilatation method of 125/126 (99.2%). We did DJ stenting in 32 patients for different causes. Our stone free rate post of 2, 4 and 6 weeks were 101/109 (92.7%), 103/109 (94.5%) and 105/109 (96.3%) respectively. We got complications rate of 11/125 (8.8%) with perforation rate of 5/125 (4%), stone migration rate of 4/109 (3.7%), conversion rate of 4/109 (3.7%) and re-admission rate of 3/125 (2.4%).

## DISCUSSION

URS is one of the most important diagnostic and procedure tool for the disease of ureter. Before the introduction of small caliber URS, different types of dilatation methods were used, even these days some urologists use those methods too. But in our study, we did not use any dilatation method for insertion of URS in ureter, still we got success rate for insertion of URS of 99.2% which is much better than Fasihuddin Q *et al* result of having 8% of failure to insertion rate<sup>1</sup>. As in other centers, stone diseases are main cause for doing URS. Similarly, in our hospital also stone disease was the commonest cause to perform URS.

These days URSL is the first line therapy for most of ureteric stones with stone free rate more than 90%. In China, Chen QS *et al* analyzed 515 cases undergone URSL and they got

97.2% of success rate<sup>2</sup> while in USA Baglay DS got >90% of success rate<sup>3</sup>, Rasool M *et al*, Pakistan got success rate of 94.2%<sup>4</sup>, Fasihuddin Q *et al*, in 125 patients underwent URSL with stone clearance rate 93.8%<sup>1</sup>, which is comparable to our success rate of 96.3%. They concluded that the therapeutic effects of pneumatic lithotripsy through URS were reliable and safe in the treatment of ureteral stones, with rapid postoperative recovery<sup>2</sup>. Similarly Zhong W *et al* used pneumatic lithotripter for 180 patients during URSL and got stone free rate of 93.3%<sup>5</sup> and in Pakistan, Ather MH *et al* got stone free rate of 85%. He concluded that with recent advances in endourology the indication for open surgery have decreased more than one third during these 10 years<sup>6</sup>.

Another big study done by Elashry OM *et al* in Egypt for 15 years in single center of 4512 patients who underwent 5133 URS for treatment of distal ureteric calculi, got stone free rate of 94.6 % with intraoperative complication rate of 6.67% and postoperative complication rate of 9.9% and concluded that URSL have high success rate with minor complications for distal ureteric calculi<sup>7</sup>.

During our study we got complication rate of 8.8% which includes perforation rate as 4%, stone migration rate of 3.7%, conversion rate of 3.7% and re-admission due to complication is 2.4%. Similarly, Zhong W *et al* used similar procedure as our and got perforation rate of 3.3% and retreatment rate of 2.2%<sup>5</sup> and Rasool M *et al* in Pakistan got perforation rate of 5.7% and stone migration rate of 5.7%<sup>4</sup>. Ko YH *et al* in Korea during URSL for upper ureteric calculus got stone free rate of 68.8% with complication rate of 21.9%, stone migration rate 15.6%, perforation rate 9.4% and conversion rate of 6.2%<sup>8</sup>. So our study complications rate also comparable to the result of URSL in different part of world.

## CONCLUSIONS

We got after URSL for ureteric calculus, stone free rate of 96.3% with complication rate of 8.8% and conversion rate of 3.7%. So, in our experience URS/URSL is safe and choice of management to diagnose ureteric disease and especially for mid and lower ureteric calculus.

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# Bacteriological Study of Post-operative Wound Infections and Their Antibiograms

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## Key words:

Postoperative Wound infections,  
*Pseudomonas aeruginosa*,  
*Staphylococcus aureus*,  
Antibiotic.

## ABSTRACT

**Objectives:** To isolate and identify and to determine the distribution of pathogens in postoperative wound infections and to evaluate *in vitro* antibiotic susceptibility of isolated bacterial pathogens.

**Methods:** The study was carried out on 100 postoperatively wounded infected cases, during July 2008 - January 2009. Samples were collected by swabs and pus was aspirated with a sterile needle and syringe and transported to laboratory immediately. Samples were processed using standard bacteriological techniques by Direct Microscopic examination using Gram's stained smears and cultured on to MacConkey agar, blood agar, chocolate agar and isolates were identified. Antibiotic sensitivity test was conducted by using Mueller-Hinton agar medium, by disc-diffusion (Kirby-Bauer) method.

**Results:** Postoperative wound infections caused by Gram negative bacilli were more common. 82 (82%) were culture positive. *E.coli* was the commonest etiological agent 22 (53.6%) followed by *Staphylococcus aureus* 18 (43.8%), *Pseudomonas aeruginosa* 18 (43.8%), *Klebsiella pneumoniae* 16 (39%), *Proteus vulgaris* 6 (14.6 %), and *Staphylococcus epidermidis* 2 (4.8%). All isolates were sensitive to Imipenem and Chloramphenicol. *E.coli* was sensitive to all antibiotics tested except Ciprofloxacin, Ofloxacin, Co-trimoxazole, Penicillin/Tazobactam, Ampicillin and Linezolid. *Pseudomonas aeruginosa* was resistant to all antibiotics tested except Imipenem, Cefepime, Chloramphenicol, Penicillin/Tazobactam, Amoxicillin/Clavulanic acid and Clindamycin. *Staphylococcus aureus* were sensitive to all antibiotics tested except Gentamycin, Co-trimoxazole, Ampicillin/Sulbactam, Amoxicillin/Clavulanic acid, Polymyxin-B and Clindamycin.

**Conclusions:** Postoperative wound infections caused more commonly by Gram negative bacilli and predominant pathogens involved were *E.coli*, followed by *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Proteus vulgaris* and *Staphylococcus epidermidis*. All isolates were sensitive to Imipenem and Chloramphenicol. A high degree of multidrug resistance was observed with *Pseudomonas aeruginosa* and *Proteus vulgaris*.

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## INTRODUCTION

Surgical wound infections are the most frequent nosocomial infection in most hospitals<sup>1</sup> and are important cause of morbidity, mortality, and excess hospital costs<sup>3</sup>. Postoperative wound infections<sup>2</sup> delays recovery and often increases length of stay and may produce lasting sequelae and require extra resources for investigations, management and nursing care. Therefore, its prevention or reduction is relevant to quality patient care<sup>4</sup>.

## OBJECTIVES

Present investigations were undertaken with the following aims and objectives. i) To isolate and identify and to determine the distribution of aerobic bacterial pathogens in postoperative wound infections ii) To evaluate *in vitro* antibiotic susceptibility of bacterial isolates.

## METHODS

The study was carried out on 100 postoperatively wounded infected cases, during July 2008 - January 2009, at Prathima hospital, Karimnagar, Telangana, India. Specimens were collected by sterile cotton swabs and pus was aspirated with a sterile needle and syringe and transported to laboratory immediately. Samples were processed using standard bacteriological techniques by Direct Microscopic examination using Gram's stained smears and cultured on to MacConkey agar, blood agar, chocolate agar and isolates were identified by biochemical tests. Simultaneously antibiotic sensitivity test was conducted by using Mueller-Hinton agar medium by disc-diffusion (Kirby-Bauer) method for the antibiotics listed in Table 1.

Table 1: List of antibiotics tested

S. No	Antibiotic	Disc concentration
1	Imipenem	10 mcg
2	Cefepime	30 mcg
3	Gentamycin	10 mcg
4	Ciprofloxacin	5 mcg
5	Chloramphenicol	30 mcg
6	Ofloxacin	5 mcg
7	Co-trimoxazole (Trimethoprim/ sulphamethoxazole)	1.25/23.75 mcg = 0.052 mcg
8	Penicillin/Tazobactam	100/10 mcg
9	Vancomycin	30 mcg
10	Ampicillin/Sulbactam	10/10 mcg
11	Cefoperazone	75 mcg
12	Amoxycillin/Clavulanic acid	20/10 mcg (30 mcg)
13	Oxacillin	1 mcg
14	Polymyxin B	300 units
15	Erythromycin	15 mcg
16	Novobiocin	30 mcg
17	Gatifloxacin	5 mcg
18	Ampicillin	10 mcg
19	Clindamycin	2 mcg
20	Levofloxacin	5 mcg
21	Linezolid	30 mcg

## RESULTS

Table 2: Types of bacteria by Direct Microscopy (Gram's staining)

Bacteria seen	Number ( % )
Gram negative bacilli	62 (62%)
Gram positive cocci	20 (20%)
Total	82 (82%)

Table 3: Pathogens isolated from postoperative wound infections

S. No	Bacteria isolated	No. of isolates	Percentage (%)
1	<i>E.coli</i>	22	53.6
2	<i>Staphylococcus aureus</i>	18	43.8
3	<i>Pseudomonas aeruginosa</i>	18	43.8
4	<i>Klebsiella pneumoniae</i>	16	39
5	<i>Proteus vulgaris</i>	6	14.6
6	<i>Staphylococcus epidermidis</i>	2	4.8
	Total	82	82

Table 4: Antibiotic sensitivity pattern of isolated strains (Nil = All are Resistant; - = Not tested)

S. No	Antimicrobial agent	<i>E.coli</i> 22 (%)	<i>Pseudo aeruginosa</i> 18 (%)	<i>Staph aureus</i> 18 (%)	<i>Klebsiella pneumoniae</i> 16 (%)	<i>Proteus vulgaris</i> 6 (%)	<i>Staph epidermidis</i> 2 (%)
1	Imipenem	18 (81.8)	6 (33.3)	8 (44.4)	16 (100)	6 (100)	2 (100)
2	Cefepime	6 (27.2)	14 (77.7)	16 (88.8)	Nil	4 (66.6)	2 (100)
3	Gentamycin	2 (9)	Nil	Nil	Nil	-	2 (100)
4	Ciprofloxacin	Nil	Nil	6 (33.3)	12 (75)	6 (100)	-
5	Chloramphenicol	8 (36.3)	16 (88.8)	14 (77.7)	10 (62.5)	4 (66.6)	2 (100)
6	Ofloxacin	Nil	Nil	4 (22.2)	Nil	Nil	-
7	Co-trimoxazole	Nil	-	Nil	2 (12.5)	Nil	Nil
8	Penicillin/ Tazobactam	Nil	6 (33.3)	Nil	6 (37.5)	2 (33.3)	6 (33.3)
9	Vancomycin	2 (9)	Nil	16 (88.8)	Nil	Nil	Nil
10	Ampicillin/ Sulbactam	-	Nil	Nil	-	-	-
11	Cefoperazone	-	Nil	-	-	-	-
12	Amoxycillin/Clavulanic acid	-	4 (22.2)	Nil	-	-	2 (100)
13	Oxacillin	-	-	18 (100)	-	-	2 (100)
14	Polymyxin-B	14 (63.3)	-	Nil	Nil	Nil	-
15	Erythromycin	-	-	10 (55.5)	-	-	Nil
16	Novobiocin	-	-	4 (22.2)	-	-	2 (100)
17	Gatifloxacin	-	-	12 (66.6)	-	-	2 (100)
18	Ampicillin	Nil	-	2 (11.1)	10 (62.5)	Nil	Nil
19	Clindamycin	6 (27.2)	4 (22.2)	Nil	8 (50)	6 (100)	2 (100)
20	Levofloxacin	16 (72.7)	Nil	10 (55.5)	16 (100)	Nil	-
21	Linezolid	Nil	Nil	6 (33.3)	4 (25)	6 (100)	-

Table 2 shows the results of Gram's staining. Majority (88%) of the Gram's stained smears revealed the presence of pus cells.

Out of 100 cases of postoperative wound infections, 82 (82%) were culture positive (Table 3). *E.coli* was the commonest etiological agent 22 (53.6%) followed by *Staphylococcus aureus* 18 (43.8%), *Pseudomonas aeruginosa* 18 (43.8%), *Klebsiella pneumoniae* 16 (39%), *Proteus vulgaris* 6 (14.6%), and *Staphylococcus epidermidis* 2 (4.8%).

Among 22 isolates of *E.coli*, 18 (81.8%) were sensitive to Imipenem, 16 (72.7%) sensitive to Levofloxacin, 14 (63.3%) sensitive to Polymyxin-B, 8 (36.3%) sensitive to Chloramphenicol, 6 (27.2%) sensitive to Cefepime and Clindamycin and 2 (9%) sensitive to Gentamycin and Vancomycin (Table 4).

Among 18 isolates of *Pseudomonas aeruginosa*, 16 (88.8%) were sensitive to Chloramphenicol, 14 (77.7%) to Cefepime, 6 (33.3%) to Imipenem and Penicillin/Tazobactam, 4 (22.2%) sensitive to Amoxicillin/Clavulanic acid and Clindamycin. All 18 (100%) isolates were resistant to Gentamycin, Ciprofloxacin, Ofloxacin, Vancomycin, Ampicillin/Sulbactam, Cefoperazone, Levofloxacin and Linezolid (Table 4).

Among 18 isolates of *Staphylococcus aureus*, all (100%) were sensitive to Oxacillin (Methicillin sensitive), 16 (88.8%) to Cefepime and Vancomycin, 14 (77.7%) to Chloramphenicol, 12 (55.5%) to Erythromycin and Levofloxacin, 8 (44.4%) to Imipenem, 6 (33.3%) to Ciprofloxacin and Linezolid, 4 (22.2%) to Ofloxacin and Novobiocin, 2 (11.1%) to Ampicillin. All 18 (100%) were resistant to Gentamycin, Co-trimoxazole, Penicillin/Clavulanic acid, Polymyxin-B and Clindamycin (Table 4).

Among 16 isolates of *Klebsiella pneumoniae*, all (100%) were sensitive to Imipenem and Levofloxacin, 12 (75%) to Ciprofloxacin, 10 (62.5%) to Chloramphenicol and Ampicillin, 8 (50%) to Clindamycin, 6 (37.5%) to Penicillin/Tazobactam, 4 (25%) to Linezolid, 2 (12.5%) to Co-trimoxazole. All 16 (100%) isolates were resistant to Cefepime, Gentamycin, Ofloxacin, Vancomycin and Polymyxin-B (Table 4).

Among 6 isolates of *Proteus vulgaris*, all (100%) were sensitive to Imipenem, Ciprofloxacin, Clindamycin and Linezolid, 4 (66.6%) to Cefepime and Chloramphenicol, 2 (33.3%) to Penicillin/Tazobactam. All 6 (100%) isolates were resistant to Ofloxacin, Co-trimoxazole, Vancomycin, Polymyxin-B, Ampicillin and Levofloxacin (Table 4).

2 (100%) isolates of *Staphylococcus epidermidis* were sensitive to Imipenem, Cefepime, Gentamycin, Chloramphenicol, Penicillin/Tazobactam, Amoxicillin/Clavulanic acid, Oxacillin, Novobiocin, Gatifloxacin and Clindamycin; Resistant to Co-trimoxazole, Vancomycin, Erythromycin and Ampicillin (Table 4).

## DISCUSSION

Our findings were comparable with many other studies. Manyahi<sup>10</sup> suggested that absence of pus cells in Gram stained smears does not exclude the possible presence of bacteria and reported that both Gram positive and Gram negative bacteria showed high resistance rate to Ampicillin and Amoxicillin/

Clavulanic acid. All Gram negative bacteria showed high level of resistance to majority of antibiotics tested but were all sensitive to Imipenem.

Amatya<sup>11</sup> made a bacteriological study of 388 postoperative wound samples at B & B Hospital, Gwarko, Lalitpur, Nepal. 235 samples (182 single isolates and 53 multiple isolates) were culture positive. High infection rate was found in males (62.5%) than females (53.1%) and in the age group 30 - 40 years (25.5%). The majority of the organisms were isolated from surgical sites after orthopaedic surgery (63.01%), and from orthopaedic ward (25.7%). 292 isolates of 10 different bacterial species were obtained. The predominant isolates were *Pseudomonas* spp (33.9%), followed by *Escherichia coli* and *Staphylococcus aureus*. Other isolates were *Acinetobacter* spp, *Klebsiella* spp, *Enterobacter* spp, coagulase negative staphylococci, *Proteus mirabilis*, nonhemolytic streptococci and *Citrobacter* spp. The most effective antibiotic for Gram positive isolates was Oxacillin (96.1%) followed by Chloramphenicol (85.2%), and Ceftriaxone and Cefoperazone Sulbactam (66.7% each). Similarly, against Gram negative bacteria, Imipenem was most effective (74.8%), followed by Amikacin (58.8%). A total of 2.2% of *S. aureus* was Methicillin Resistant *S. aureus* (MRSA).

Our findings were similar to the study done by Lilani *et al*<sup>5</sup> and Rogers BA<sup>8</sup>. However, in our study the commonest isolate was *E.coli* followed by *Staphylococcus aureus*, whereas in a study by Lilani *et al*<sup>5</sup> and Olson<sup>9</sup> *Staphylococcus aureus* was the most commonest isolate followed by *Pseudomonas aeruginosa*. Murthy<sup>12</sup> investigated 406 postoperative clean wounds for the presence of sepsis and established antibiogram of organisms isolated. The over all postoperative sepsis rate was 13% (clinical) and 12% (bacteriological). *Staphylococcus aureus* (32%) and *Pseudomonas* species (21%) were the commonest organisms recovered and Netilmycin, Cephaloridine and Norfloxacin were the most effective antibiotics against both Gram positive and Gram negative bacterial infections.

## CONCLUSIONS

The study reveals that postoperative wound infections<sup>5,6,7</sup> caused by Gram negative bacilli were more frequent and predominant pathogens involved were *E.coli*, followed by *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Proteus vulgaris* and *Staphylococcus epidermidis*.

All isolates were sensitive to Imipenem and Chloramphenicol. *E.coli* was sensitive to all antibiotics tested except Ciprofloxacin, Ofloxacin, Co-trimoxazole, Penicillin/Tazobactam, Ampicillin and Linezolid.

A high degree of multidrug resistance was observed with *Pseudomonas aeruginosa*, being resistant to all antibiotics tested except Imipenem, Cefepime, Chloramphenicol, Penicillin/ Tazobactam, Amoxicillin/ Clavulanic acid and Clindamycin.

*Staphylococcus aureus* strains were sensitive to all antibiotics tested except Gentamycin, Co-trimoxazole, Ampicillin/ Sulbactam, Amoxicillin/Clavulanic acid, Polymyxin-B and Clindamycin. All isolates of *Staphylococcus aureus* were identified as Methicillin sensitive (MSSA).

All isolates of *Klebsiella pneumoniae* were sensitive to all the antibiotics tested except Cefepime, Gentamycin, Ofloxacin, Vancomycin and Polymyxin-B.

All isolates of *Proteus vulgaris* were sensitive to all antibiotics tested except Ofloxacin, Co-trimoxazole, Vancomycin, Polymyxin-B, Ampicillin and Levofloxacin thus exhibiting multidrug resistance.

All isolates of *Staphylococcus epidermidis* were sensitive to all antibiotics tested except Co-trimoxazole, Erythromycin and Ampicillin.

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