Journal of
GANDAKI MEDICAL COLLEGE-NEPAL (J-GMC-N)

कर्मण्येवाधिकारस्ते
My right is to my work

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Swine flu (Pig flu)

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(Bird flu, Avian flu, Avian influenza, Chicken Ebola, Dog flu, Horse flu, human flu)

In March 2009, a new H1N1 Influenza (swine flu) virus strain was detected and it spread from person to person and caused a pandemic. The Ministry of Health and Population held a press conference on October 15, 2009 to confirm the first findings of community transmission of H1N1 influenza in Nepal. In 2014, thousands of Nepalese suffered from the pandemic flu. According to the Epidemiology and Disease Control Division (EDCD), its outbreak resulted in the death of 18 people. Again in February, 2015 there was an outbreak of swine flu epidemic in Nepal, particularly in Kathmandu, Chitwan, and Pokhara.

Swine flu is a respiratory disease commonly found in pigs, caused by highly contagious strain of Influenza virus type A. Around 1 – 4% of the pigs that develop the disease die from it.

Human influenza is an acute infectious disease of the respiratory tract which occurs in sporadic, epidemic and pandemic forms. The modern history of the disease may be considered to date from the pandemic of 1889-90.

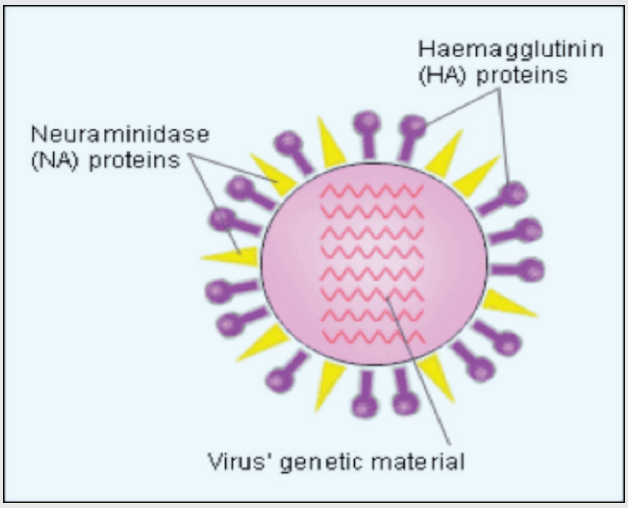
Influenza also occurs in animals and birds in nature. Indeed, the avian influenza virus was demonstrated as early as in 1901, but the association between the two remained unknown till 1955, when Schaefer demonstrated that the fowl plague virus was antigenically related to type A influenza virus. Not only did the swine influenza resemble human influenza clinically but there was also epidemiological association between the two. It was widely held that the virus spread to swine from humans at the time of the 1918 pandemic. Influenza viruses have also been isolated from horses, whales and seals.

The swine flu virus or influenza virus (**Orthomyxovirus**) is a spherical shaped, enveloped, RNA virus, measuring 80 – 120 nm in diameter. Nucleocapsid has helical symmetry. RNA genome is negative sense, single stranded, and segmented into 8 pieces. Envelope is composed of an inner protein layer called matrix (M) protein, and an outer lipid layer. Two types of glycoprotein spikes or peplomers project from the surface of the envelope: A triangular hemagglutinin (H/ HA), and mushroom shaped neuraminidase (N/ NA) (Fig 1).

When viruses are mixed with RBC, viruses are adsorbed on to mucoprotein receptors on RBC surface by HA spikes, known as **hemagglutination**. When RBC are added to a serial dilution of viral suspension, highest dilution that produces hemagglutination provides **hemagglutination titre**. Hemagglutination can be

used for detection and assay of influenza virus in culture fluids, and for titration and standardization of killed influenza vaccines.

Fig 1: Influenza virus structure (Source: myDr.com.au)



Hemagglutination can be inhibited by its specific antibody, known as **hemagglutination inhibition (HI)**. **HI test** is used for detection and quantitation of antibody to virus, and therefore used as a serological diagnostic test for influenza.

Hemagglutination is followed by detachment of virus from cell surface, a process reverse to hemagglutination, known as **Elution**. It is due to an enzyme neuraminidase (sialidase), which acts by splitting N-acetylneuraminic acid from receptors, hence known as Receptor Destroying Enzyme (RDE). Hemagglutination and elution can be used for purifying and concentrating influenza viruses.

The virus contains two types of antigens: Internal, and surface antigens. Internal antigens are type specific, and include ribonucleoprotein (RNP) antigen, and Matrix (M) protein antigen. Surface antigens are strain specific, and include hemagglutinin (H/ HA), and neuraminidase (N/ NA). Based on the nature of type specific RNP antigen, influenza viruses are classified into 3 serotypes: Influenza virus type A, influenza virus type B, and influenza virus type C.

All isolates from nonhuman (animal) hosts belong to influenza virus type A. All known viruses that cause influenza in birds belong to Influenza type A virus. Thus, Swine flu virus or Avian flu virus is the influenza type A virus. Influenza virus types B and C are exclusively human viruses and natural infection with them is not identified in animals or birds. Ordinarily, non-human influenza viruses do not cause human infection. However, they play an important role in the emergence of pandemic influenza.

Hemagglutinin (H/HA) is strain specific antigen, and exhibits great antigenic variation. It exists in 15 HA subtypes, named H1 to H15. Only 4 HA subtypes (H1 – H3, and H5) are found in humans. Antibodies to hemaggutinin (Anti-HA) are produced following infection and immunization, and are protective.

Neuraminidase (NA) or sialidase is also strain specific antigen, and exhibits antigenic variation. 9 NA subtypes (N1 – N9) have been identified. In Humans, 2 subtypes (N1, N2) are found. Antibodies to neuraminidase (Anti-NA) are produced following infection and immunization, but not effective in protection.

The unique feature of the influenza virus is its ability to undergo antigenic variation. This is of great importance in the epidemiology of the disease. Antigenic variability is highest in Influenza virus type A, and less in type B, while it has not been demonstrated in type C. Influenza virus type A undergoes antigenic variation/change in hemagglutinin (H/ HA), and neuraminidase (N/NA) from time to time. Type B virus also exhibits antigenic variation but changes are not marked. Type C virus does not undergo antigenic variation.

Antigenic variations may be: i) Complete (Major changes) — **antigenic shift** (Responsible for emergence of pandemics) ii) Partial (Minor changes) — **antigenic drift** (Responsible for emergence of Epidemics).

What makes influenza (flu) an important and challenging disease is its propensity for causing **pandemics**. It is for this reason that worldwide surveillance is maintained on influenza, under the auspices of the WHO. Influenza pandemics have been recorded at irregular intervals from 1173 AD. The most severe pandemic of modern times occurred in 1918-19 (**Spanish flu**), during which over 200 million people were affected and more than 20 million perished. India suffered the most, with some 10 million deaths. An unusual feature of this pandemic was the very high rate of mortality among young adults.

The next pandemic occurred in 1957 when the Asian strain H2N2 originated in China and spread throughout the world within a short period. However, the mortality rate was low, but caused widespread morbidity. The Hong Kong strain H3N2 appeared in 1968 also caused a pandemic but it was much less severe. In 1977, epidemic influenza appeared in China and then in Russia, was known as **Red flu**. The disease was mainly confined to the young people under 20 years age group. The isolate was identified as H1N1 virus, antigenically very close (similar) to the strains prevalent from 1946 to 1957. This H1N1 virus has spread through most of the world, and with the H3N2 virus, currently causes human influenza.

The reason the virus is able to cause epidemics and pandemics lies in its ability to undergo antigenic variation. Antigenic drift, resulting from mutation and selection, is responsible for periodical epidemics. Pandemics are caused by a virus strain that has undergone antigenic shift. **When a human or pig (swine) is coinfected by both avian and human influenza virus strains, genetic reassortment/recombination may occur between their genomes which may result in the emergence of a new virus strain that may cause a pandemic of influenza** (Fig 3). Such recombinant hybrids can be produced by growing human and nonhuman strains together in eggs or in experimental animals exposed to mixed infection. It has been shown by genetic studies that both the 1957 Asian virus and the 1968 Hong Kong virus were such recombinant hybrids.

The mere appearance of a new recombinant hybrid virus strain may not lead to a pandemic. For this the new virus strain should be capable of spreading rapidly among people. In fact there have been several instances when new recombinant hybrid virus strains have been detected, which failed to spread. The swine flu virus H1N1 caused a localized outbreak in a military camp in New Jersey, USA in 1976, leading to much anxiety and panic vaccination, but did not spread. Though a few similar incidents have occurred since then, what raised a real threat of a new pandemic was the outbreak in Hong Kong of chicken flu in 1997 with a new virus strain H5N1 influenza virus, which caused 18 confirmed human cases with 6 deaths. However, all human cases were shown to have spread directly from chickens, without any transmission from person to person. Immediate containment measures and the slaughter of all (over 1.6 million) chickens in Hong Kong stopped the danger before the strain developed person-to-person transmissibility, which could have initiated a pandemic. This incident indicated the value of influenza surveillance and the potential danger from avian strains.

Aquatic and migratory birds appear to be primary reservoirs and natural hosts of all 15 HA subtypes of influenza type A virus (Fig 2). Viruses do not cause any disease in them or undergo any mutational changes. In birds, influenza type A virus causes asymptomatic intestinal infection. Birds shed the viruses abundantly in feces, which contaminate lakes

and ponds. In cold climates, the viruses persist in such waters for long periods and can readily be isolated from them. Domestic birds like ducks can get infected from wild birds and carry the infection to pigs, which may be an important link in the chain, as they are susceptible to infection by both human and avian influenza strains. Recombination may take place in pigs and such hybrid virus strains may lead to human infection with a potential for the emergence of pandemics (Fig 3). The postulated role of ducks and pigs in the development of new hybrids explains why pandemic strains tend to originate in China where millions of birds, pigs and people live closely together. The reappearance of old strains like the H1N1 in 1977 may have been from an avian reservoir of strains. Similarly, it is possible that an old pandemic strain present in wild birds may suddenly reappear. If this hypothesis is true, it would be prudent to keep wild and domestic birds separate, and also to keep pigs away from them. The practice of keeping several species of birds along with chickens in live bird markets is potentially dangerous.

Fig 2: Aquatic and migratory birds: Primary reservoirs and natural hosts of all 15 H subtypes of influenza type A virus

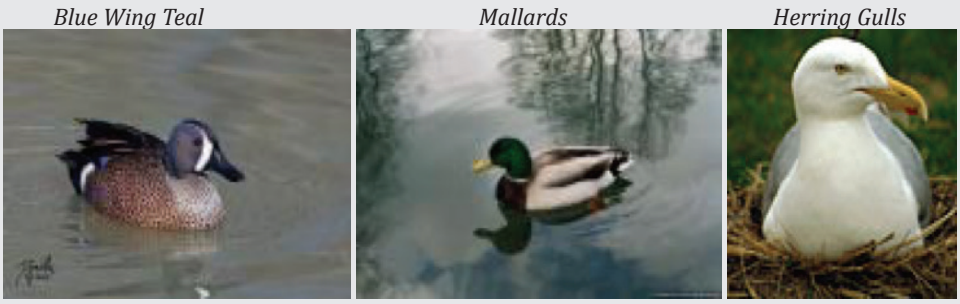
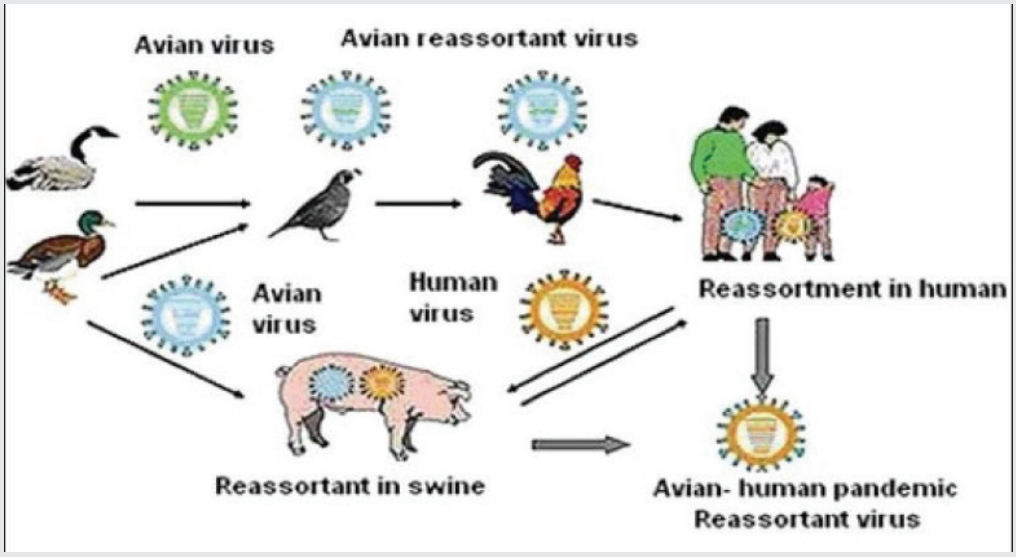


Fig 3: When a human or pig (swine) is coinfected by both avian and human influenza virus strains, genetic reassortment/recombination may occur between their genomes which may result in the emergence of a new virus strain that may cause a pandemic of influenza (Source: www.sysrevpharm.org)



A unique feature of influenza epidemiology was that once an antigenic variant emerged, it completely displaced the preexisting strain. The H1N1 strains were displaced by Asian H3N2 strains in 1968. However, this rule is not observed in recent years. Even after the reemergence and wide dissemination of the H1N1 strains in 1977, the Hong Kong H3N2 strains continue to be prevalent. The reason for this coexistence is not known.

There is considerable evidence to suggest that there occurs an orderly recycling of the virus subtypes at least with regard to their hemagglutinin (H) antigen. Sero-epidemiological studies indicate that the severe pandemic of 1889 was caused by a virus with the antigenic structure H2N8 and that this was followed in 1900 by the subtype H3N8 which led to a moderate pandemic. In 1918, came the most severe of all pandemics, caused by **'Swine type' H1N1 virus** (Formerly HSW N1). Mild epidemics occurred around 1933 and 1946 associated with minor variations in the hemagglutinin (H) antigen. The next severe pandemic came in 1957 with the H2N2 (Asian) subtype. This was followed in 1968 by the H3N2 (Hong Kong) virus strain leading to moderate pandemic. The year 1977 saw the reappearance of H1N1 virus strain. Thus the sequence of variation in the hemagglutinin (H) antigen has been H2⟶H3⟶H1⟶H2⟶H3⟶H1 from 1989 to present time. From 1977, both H3N2 and H1N1 viruses have been circulating together.

In March 2009, a new H1N1 virus strain was detected which was a reassortant between previously circulating swine virus and a Eurasian swine virus and was also called **swine origin influenza virus (S-OIV)**. It spread from person to person and caused a pandemic. As of 1 August 2010, worldwide more than 214 countries reported laboratory-confirmed cases of pandemic influenza H1N1 2009, including over 18449 deaths.

Evaluation of Postmenopausal Women and Men Aged above 50 for Risk Factors Associated with Osteoporosis

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Keywords
Bone Mineral Density, Men aged above 50, Osteoporosis, Postmenopausal women, Risk factors.

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ABSTRACT

Objectives: To study risk factors for osteoporosis in postmenopausal women and men aged above 50.

Methods: It is a cross sectional, descriptive, observational study. Demographic profile of participants and risk factors for osteoporosis were studied by direct interview with predefined questionnaire and with Bone Mineral Densitometry. Osteoporosis was defined as a T-score of - 2.5 or less and osteopenia, -1.5 to -2.5.

Results: Total number of participants were 200. Among them, postmenopausal women were 100 and men aged above 50 years were 100. Among postmenopausal women 15% were found to be with low T-score with diagnosis of osteoporosis. 21% of postmenopausal women and 16% of men were found to have osteopenia. All the diagnosed cases were attributed by minimum of two clinical risk factors. The most common risk factors were less BMI, and age above 55 years (50-60).

Conclusions: The risk factors of osteoporosis are not only associated with bone fractures but also for the cardiovascular disease and are modifiable (Body mass index, smoking, diet, life style). The health providers can take opportunity to advice risk factors of cardiovascular disease as well.

INTRODUCTION

Osteoporosis is a progressive and systemic skeletal disorder characterised by low bone mass and micro architectural deterioration of bone tissue, with a consequent increase in bone fragility and susceptibility to fracture[1].

Bone density or mass relates to the density of osteocytes in a particular area of skeletal bone. The main mineral found in osteocytes is calcium. It is the main contributor to bone strength. Bone density is measured by bone densitometry. The lower the bone density, the higher the risk of fracture even with low impact injury. Bone fractures secondary to osteoporosis cause significant disability and cost, both to the individual, their family and to the community health care costs. The spinal verterbrae, hip and forearm are the main areas affected by osteoporotic fractures. Osteoporotic fractures lead to significant morbidity and mortality, particularly in case of the hip fractures[2,3].

Osteoporosis does not produce symptoms unless associated with fracture or fractures. Osteoporosis has no warning signs, and fracture might be the presentation

of disease. Nearly all non vertebral fractures are caused by a fall; however, vertebral fractures occur without a fall, and need not necessarily be painful. Only one third of vertebral fractures are painful and rest two third are painless[4].

Marked height loss over years may be a sign of underlying vertebral compression fractures, even without significant associated back pain[5]. Wrist or other fractures may occur at a younger age than vertebral or hip fractures and may also be early clinical presentation of osteoporosis. Age is the most important factor associated with bone loss[1]. The kyphosis caused by thoracic vertebral wedge fractures (Dowager's hump) is common feature in the elderly and may also contributes to respiratory morbidity.

Bone density or mass is the result of a constant remodelling of bone and a balance between bone formation by osteoblasts and resorption by multinucleated giant cells called osteoclasts. This balance alters through out life and peak bone mass is reached at approximately 40 years of age. From then the bone mass declines at a slow steady rate of about 0.4% per year. This rate of loss accelerates in women during the climacteric, reaching 2 - 5% loss per annum for the first 5 – 10 years following menopause, slowing thereafter to 1% per year[4].

Skeletal bone structure consists of outer cortical bone and inner trabecular bone. The latter found in greater concentration in the central skeleton. Osteoporosis particularly affects the trabecular bone so predisposes to the increased risk of fracture in these areas[5,8].

A major problem of bone strength is determined by measurement of bone mineral density (BMD). BMD is measured by Bone Densitometry. Dual-energy x-ray absorptiometry (DEXA) is the most common method used in assessing BMD. Measurement of bone strength other than bone density at these sites may predict fracture risk but cannot be used to diagnose osteoporosis[5,6,7]. That is why BMD measurements are effective tools for identifying patients at high risk for fracture[10]. The use of serial Bone Densitometry start at age of 50 or postmenopause can screened out osteoporosis and preventive measures can be taken to prevent fracture in near future. However, bone strength and fracture risk are also affected by other qualities of bone such as rate of remodelling, size and geometry, microarchitecture, mineralization, damage accumulation and matrix quality[5,8].

Primary osteoporosis is usually due to bone loss that occurs with aging. Falling estrogen levels through the climactreric and in particular after the menopause are the leading causes of bone loss in women. A declining level of testosterone is the leading factor in men[6].

Bone loss is maintained by dominating activity of osteoblast in young age and before menopause.Both aging and hypoestogenaemia (Post menopausal state), activate osteoclasts, which results in demineralisation of bone and bone becomes thin and fragile.

Secondary osteoporosis is a result of medication (e.g. Glucocorticoids) or medical disease (e.g. Malabsorption), various cancers, smoking, alcohol consumption[2]. This adversely affects the skeletal health. The cause of osteoporosis is multi factorial. Primary factors are age, heredity, estrogen, androgen status and dietary calcium intake.

In addition, a decrease in calcium intake or impaired absorption of calcium from the gut lowers serum level of ionized calcium. This stimulates parathyroid hormone (PTH) secretion to mobilize calcium from bone by stimulation of osteoclastic activity. As a result bone becomes more thin and fragile[1]. With a minimal stress, the thin and fragile bones get fracture very easily.

METHODS

It was a cross sectional, descriptive, observational study. Demographic profiles of participants and risk factors for osteoporosis were studied by direct interview with predefined questionnaire and with measurement of Bone mineral density. The study was conducted in Gandaki Medical College Teaching Hospital (Pokhara) with collaboration of Orthopaedic Department, General Surgery department and Gynaecology Department on 22nd December 2013. This was an opportunistic study. Patients who presented to OPDs (Gynaecoloy, Orthopaedics and General Surgery) for their other problems were invited one month prior to study.

Inclusion criteria were

1. Postmenopausal women or postsurgical menopause (bilateral oophorectomy with or without hysterectomy).
2. Men aged above 50 years.

Exclusion criteria were

1. Known case of osteoporosis.
2. Women who had undergone hysterectomy with bilateral oophorectomy or post menopause on hormone replacement therapy or calcium or vitamin D preparation.
3. Men or women suffering from Rheumatoid arthritis and under treatment of glucocorticoids and regular intake of alcohol.

An idea for clinical risk factors and low BMD associated with osteoporosis are obtained from the medical literature and following risk factors were included.

1. Risk factor 1: Age
2. Risk factor 2: Body Mass Index
3. Risk factor 3: Current smoking last 6 months
4. Risk factor 4: Duration of menopause

Type of Equipment: (Peripheral) Radial Densitometry.

Bone Mineral Density based definitions of bone density (WHO):

1. Normal: T score above (i.e. greater) than or equal to – 1 to 0.
2. Low Bone Mass (Osteopenia): T score between – 1.5 to above - 2.5.
3. Osteoporosis: T score below or equal to - 2.5.

RESULTS

Total 200 participants were enrolled in this study. But among them 100 participants were men aged above 50, found to be healthy (none of them were having osteoporosis). Further analysis was not needed for them. The remaining 100 participants post menopause were analysed for risk factors.

Table 1: Age wise distribution and trend of osteopenic and osteoporotic postmenopausal women

Age (in years)	Normal BMD (%)	Osteo-penia	Osteopo-rosis	Total
40 - 50	4	3	0	7
50 - 60	29	12	9	50
60 - 70	25	4	3	32
70 - 80	6	2	2	10
80 +	0	0	1	1
Total	**64**	**21**	**15**	**100**

Fig 1: Age wise distribution and trend of osteopenic and osteoporotic postmenopausal women

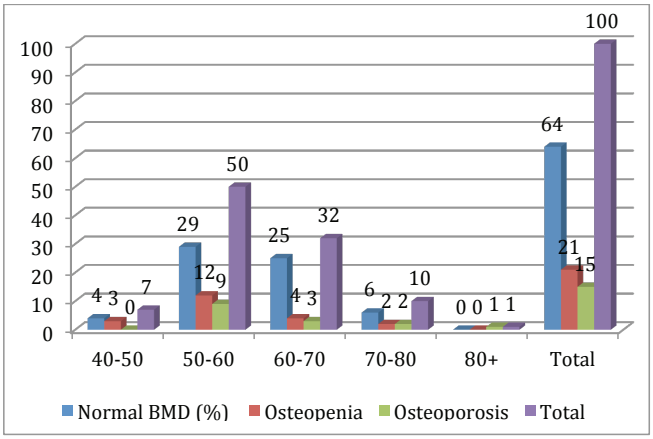


Table 2: Age wise distribution and trend of osteopenia and osteoporosis in men age above 50

Age (in years)	Normal BMD (%)	Osteope-nia	Osteopo-rosis	Total
40 - 50	6	3	0	9
50 - 60	55	10	0	65
60 - 70	6	5	0	11
70 - 80	11	1	0	12
80+	3	0	0	3
Total	**81**	**19**	**0**	**100**

Fig 2: Age wise distribution and trend of osteopenia and osteoporosis in men age above 50

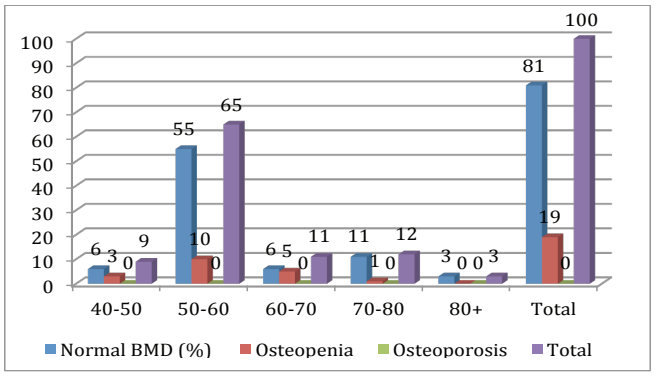


Table 3: Distribution of weight and trend of osteopenia and osteoporosis in postmenopausal women

Body Mass Index (BMI)	Normal BMD	Osteo-penia	Osteopo-rosis	Total
Under Weight (<18.5 Kg/m^2)	32	16	12	60
Normal Weight (18.5 Kg/m^2 - 24.9 Kg/m^2)	27	4	3	34
Over Weight (25.0 Kg/m^2 - 29.9 Kg/m^2)	5	1	0	6
Total	64	21	15	100

Fig 3: Distribution of weight and trend of osteopenia and osteoporosis in postmenopausal women

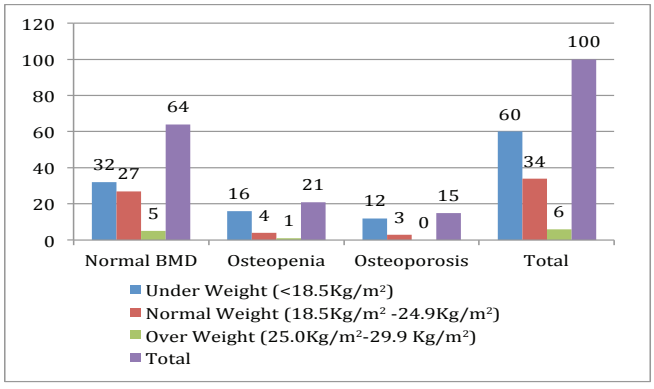


Table 4: Relation between current smoker and BMD in postmenopausal women

	Normal BMD	Osteo-penia	Osteopo-rosis	Total
Non Smoker	71	10	1	82
Currently Smoking	8	3	7	18
Total	79	13	8	100

Fig 4: Relation between current smoker and BMD in postmenopausal women

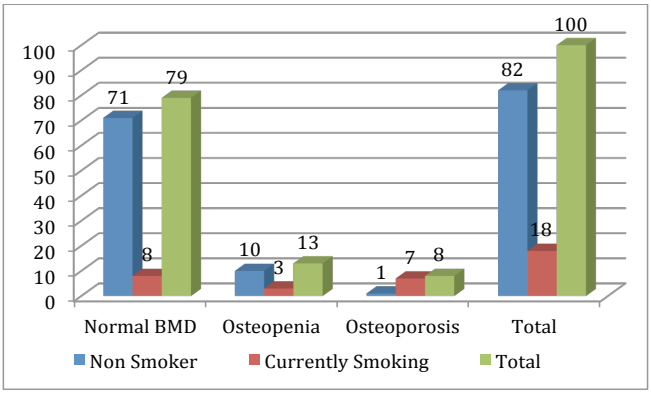
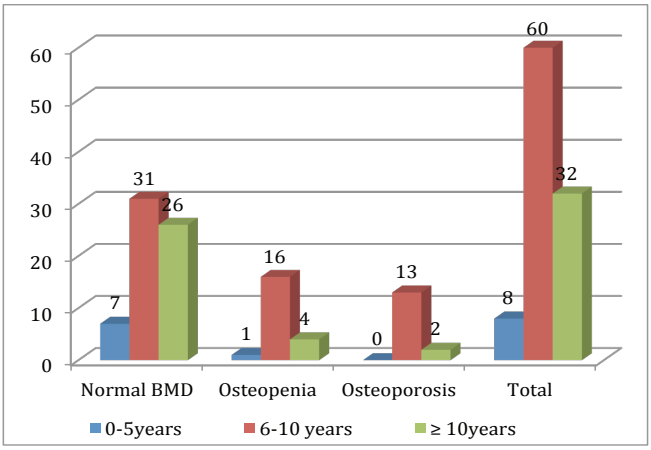


Table 5: Relation between duration of menopause and Bone Mineral Density

Duration of menopause	Normal BMD	Osteo-penia	Osteo-porosis	Total
0 - 5years	7	1	0	8
6 - 10 years	31	16	13	60
≥ 10 years	26	4	2	32
Total	64	21	15	100

Fig 5: Relation between duration of menopause and Bone Mineral Density



DISCUSSION

It is easy to say that prevention is better than cure, but very difficult to apply in practice in case of chronic health problem. Osteoporosis is degenerative, multifactorial problem occurring mostly in the women with gradual onset. Chronic health problem of women does not come into priority in the family because of lack of awareness, education and poor socioeconomic condition.

In this study we found that the osteoporosis is more common in women than men for the same age group[6]. Our results were comparable to the study carried out by Mardas AK *et al* where they found 25.6% of women had osteoporosis[8]. We had also observed that none of the men aged above 50, had osteoporosis. Since none of the men aged above 50 had osteoporosis, further analysis for men did not require. But it does not mean that men do not suffer from osteoporosis. We can say that age at 50 is not the appropriate time to evaluate osteoporosis for men[6].

The majority of women are having osteoporosis because the women are at greater risk, as they have smaller bone and lower total bone mass, less intake of vitamin D and calcium in their diet[4]. They loss bone more quickly after menopause[4,7]. The study carried out by Baddoura R *et al* found that 33% of women had osteoporosis with the use of DXA as measuring tool for BMD at total hip[9]. So women are more vulnerable to get osteoporosis whichever the measuring tool was used to measure BMD. This study showed that our women with post menopause having low body mass index (BMI) were found to be more osteoporotic (12%) as compared to women with normal weight and overweight.

Smoker women (current) were found to be suffering from osteoporosis more than nonsmokers. Similarly those women who had menopause for more than 5 years were more osteoporotic. This finding is comparable to study carried out by Najam R and Huda N, where duration of menopause was 6 to 10 years[10].

CONCLUSIONS

Peripheral Radial Densitometry is simple, inexpensive method for measuring Bone Density. Our study has some strength to identify risks factors for osteoporosis and it recommends for BMD only to those post menopausal women who had two or more than two risk factors.

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Effectiveness of Addition of Dexamethasone to Local Anesthetics in Supraclavicular Brachial Plexus Block

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ABSTRACT

Background: Supraclavicular brachial plexus block is a popular and widely employed regional nerve block technique for perioperative anesthesia and analgesia for upper extremity surgery. Several adjuvants like ketamine, epinephrine, opioids, alpha-2 agonists etc. can be added to local anesthetics to prolong the duration of regional blocks and also to intensify the quality of regional blocks.

Objectives: Our objective was to determine whether the addition of dexamethasone to bupivacaine would prolong the duration of analgesia after supraclavicular brachial plexus blockade for patients undergoing ambulatory upper-limb surgery.

Methods: A total of 60 physical status ASA I-II patients aged 20 - 70, weight >60 kg of either sex, undergoing upper limb surgery below the shoulder joint (both elective and emergency surgery) were enrolled in the study. The duration of study was February 2014 to February 2015. Patients were randomly allocated to one of the two groups, consisting 30 patients in each group.

Group I (Case): Patients in this group received 1.0% adrenalized xylocaine/ lignocaine (20 ml) and 0.5% bupivacaine (16 ml) plus dexamethasone 8 mg (2 ml) making a total volume of 38 ml.

Group II (Control): Patients in this group received 1.0% adrenalized xylocaine/ lignocaine (20 ml) and 0.5% bupivacaine (16 ml) plus 0.9% normal saline (2 ml) making a total volume of 38 ml.

Results: Duration of motor block and duration of analgesia were prolonged in dexamethasone group (Case) as compared to control group which is statistically very significant (P<0.0001). The mean duration of motor block in case group and control group were 415 ± 30.0 and 216 ± 32.0 respectively (P<0.0001) which is statistically highly significant. The mean duration of analgesia in case group was 925 ± 60.0 and in control group was 322 ± 40.2 which is statistically highly significant (P<0.0001).

Keywords
Dexamethasone, Local anesthetics, Supraclavicular brachial plexus block

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Conclusions: The addition of dexamethasone to 1.0% adrenalized xylocaine for axillary brachial plexus block significantly helps in prolonging the duration of analgesia and motor blockade in patients undergoing upper limb surgeries and is comparatively safe and cost-effective method of providing post-operative analgesia.

INTRODUCTION

Regional anesthesia is an excellent adjunct or alternative to general anesthesia for extremity surgery[1]. It provides an important role in facilitating ambulatory anesthesia and reducing immediate post-operative pain[2]. Uncontrolled pain, nausea and vomiting are the common causes for delayed discharge and unanticipated hospital admission[3,4]. Brachial plexus block is popular and widely employed regional nerve block technique for peri-operative anesthesia and analgesia for surgery of the upper extremity. It can significantly reduce pain and nausea allowing for faster discharge from hospital when compared with general anesthesia[5]. The duration of the sensory nerve blockade and therefore analgesia with single shot regional anesthesia is relatively short lived and the challenge remain to prolong the duration of analgesia while minimizing adverse effects. Another advantage is that it can be performed at the patients' arm in any position to provide excellent anesthesia for elbow, forearm and hand surgery[6]. Prolonging blockade time and thus analgesia could potentially benefit both patients and the healthcare system. Various perineural adjuvants have been used with local anesthetics in regional anesthesia in an attempt to optimize block characteristics and improve clinical outcomes[7,8]. Vasoconstrictors can be used to vasoconstrict the vessels, thereby reducing vascular absorption of the local anesthetic. Clonidine has also been used in combination with local anesthetics (lidocaine, bupivacaine, mepivacaine) in axillary brachial plexus block[9]. Corticosteroids are widely used in peripheral nerve blocks for acute pain control and are routinely injected into the epidural space for treating radicular pain with a reliably acceptable side effect profile[10,11].

Glucocorticoids have been shown to prolong nerve blockade in proportion to their rank-order anti-inflammatory potency, an effect that can be mitigated by the corticosteroid antagonist cortexolone[12]. Recent research has focused on the addition of the glucocorticoid dexamethasone as a local anesthetic adjuvant in regional anesthesia. Intravenous dexamethasone has been previously shown to be opioid-sparing in the early postoperative phase between 24 - 48 hours following its administration[13,14] and also serves to reduce postoperative nausea and vomiting (PONV)[15]. Several clinical studies have evaluated the effectiveness of dexamethasone applied perineurally with aocal anesthesia in regional nerve blocks including epidural[16,17] brachial plexus[18,19], femoral and sciatic[20], and facial and dental blocks[21,22]. Clinical studies investigating the analgesic efficacy of dexamethasone added to local anesthetic agents have been encouraging.

Our objective was to determine whether the addition of dexamethasone to bupivacaine would prolong the duration of analgesia after supraclavicular brachial plexus blockade for patients undergoing ambulatory upper-limb surgery. We hypothesized that the addition of dexamethasone to bupivacaine would significantly prolong the duration of analgesia after single shot supraclavicular brachial plexus block compared with bupivacaine and xylocaine.

METHODS

A randomized, prospective, double blinded and case control study was carried out at Department of Anesthesiology, Gandaki Medical College Teaching Hospital, Pokhara after getting institutional approval and written informed consent from the patients. A total of 60 physical status ASA I-II patients aged 20 - 70, weight >60 kg of either sex, undergoing upper limb surgery below the shoulder joint (both elective and emergency surgery) were enrolled in the study. The duration of study was February 2014 to February, 2015.

Patients unwilling to give consent and patients with a history of allergy to local anesthetic, a history of peptic ulcer disease, diabetes mellitus, hepatic or renal failure, severe respiratory disease, patients receiving any premedication (including opioids, benzodiazepines and clonidine), neurological patients with deficit in brachial plexus and pregnant women were excluded from the study.

Patients were randomly allocated to one of the two groups, consisting 30 patients in each group using computer-generated randomization list.

Group I (Case): Patients in this group received 1.0% adrenalized xylocaine/ lignocaine (20 ml) and 0.5%

bupivacaine (16 ml) plus dexamethasone 8 mg (2 ml) making a total volume of 38 ml.

Group II (Control): Patients in this group received 1.0% adrenalized xylocaine/ lignocaine (20 ml) and 0.5% bupivacaine (16 ml) plus 0.9% normal saline (2 ml) making a total volume of 38 ml.

Both local anesthetic solutions were prepared by an anesthesia assistant not involved in the performance of supraclavicular brachial plexus block, patient care, or data collection. No premedications were applied to the patients. Thorough history was elicited in each patient and clinical examination was investigated in detail. The brachial plexus block was performed using supraclavicular approach. The block was performed using a nerve locater in all cases after preanesthetic evaluation with monitoring instrument. During injection of drugs negative pressure aspiration was performed after every 5 - 6 ml to avoid intra-vascular injection. Sensory and motor block of all four nerve territories (radial, ulnar, median and musculocutaneous) were assessed. All nerve blocks were performed by a single experienced anesthesiologist blinded to group allocation. The onset and duration of analgesia and motor block and any complications were evaluated. The patients were routinely monitored with electrocardiogram (ECG), non-invasive blood pressure (NIBP) measurement, and pulse oximetry (SpO_2) during procedures. Surgery was conducted with patients awake, and a surgical tourniquet was used in all cases. Additive general anesthesia was at the discretion of the anesthesiologist and was based on sensory blockade at 30 minutes and the intended area of surgery (leading to exclude the patient).

Parameters Studied

After the end of the supraclavicular brachial plexus (SCBP) block, an anesthesiologist blinded to the solution type evaluated sensory block and motor block as follows.

A. Onset of sensory block

Onset of sensory block can be defined as the time interval between the end of local anesthetic administration and the loss of sensation to pinprick (sensory score = 1/2p) in nerve specific territory. Sensory blockade of each nerve was evaluated every five minutes until 30 minutes using a spirit soaked cotton swab using a three-point scale and compared with the control arm as a reference.

0 = Normal sensation (No block)

1 = Decreased sensation (Partial block)

2 = No sensation (Complete block)

Table 1: Sensory test sites and motor test

Major Peripheral Nerve Distribution	Motor test	Sensory test site
Median	Flexion of three fingers	Thenar eminence
Ulnar	Abduction of fingers	Hypothenar eminence
Radial	Extension of the elbow and wrist	Dorsum of hand
Musculocutaneous	Flexion at the elbow	Overbase first metacarpal
Medial cutaneous ante brachial		Median side of arm

B. Onset of motor block

The onset time of the motor blockade was determined at the time interval between the end of local anesthetic administration and absent movement (Bromage modified scale = 0) in nerve specific territory. The motor block was evaluated by Bromage modified scale at 0, 10, and 20 and at the end of the 30 minute period (Table 2).

Motor block was evaluated by thumb abduction (radial nerve), thumb adduction (ulnar nerve), flexion of the elbow in supination and pronation of the forearm (musculocutaneous), and thumb opposition (median).

Table 2: Modified Bromage scale

Score	Definition
4	Full power in relevant muscle group
3	Reduced power but ability to use muscle group against resistance
2	Ability to move relevant muscle group against gravity but inability to move against resistance
1	Flicker of movement in relevant muscle group
0	No movement in relevant group

C. Duration of analgesia

Duration of analgesia was considered as the time interval between the administration of local anesthetic and the first postoperative pain. During the procedure, anesthesia was considered satisfactory if the patient did not complain of any pain or discomfort and if no sedation was necessary.

Post operative follow up was carried out in the recovery and post operative ward. Duration of analgesia was noted according to 0 - 10 Visual Analog Scale (VAS) for pain at every half an hour for first 10 hours and then hourly till 24 hours. When patients began to experience worst pain (VAS = 8 - 10), it was considered that analgesic action of the drug was terminated and rescue analgesic (IM Diclofenac 1 - 1.5 mg/kg) was administered to the patients.

D. Duration of motor block

The duration of the motor block was defined as the time interval between the end of local anesthetic administration and recovery of complete motor function (Bromage modified scale = 4).

The duration of motor blockade post operatively was assessed every hourly by asking the patients to move their fingers and to see whether they are able to raise the hand or not. This time was recorded and taken as the cessation of motor block effect.

Duration of postoperative analgesia and proportion of patients requiring analgesic supplementation for the first 24 hrs were the primary outcome variables. Onsets of anesthesia, potency of analgesia were the secondary end points.

E. Incidence of side effects during block in two groups

Patients were monitored routinely and any untoward side effects like drowsiness, pruritus, nausea/ vomiting, voice changes, Horner's syndrome, and dyspnea or chest discomfort were looked for and noted if any.

Data Analysis

The collected data was reviewed, coded, verified and statistically analyzed using the Statistical Package for Social sciences (SPSS) version 16 and Microsoft Excel 2007. Descriptive statistics was used for all studied variables. For statistical analysis of demographic data and for comparison of groups, Chi-square, Mann-Whitney U-test, and independent Student's t-test analyses were performed. Results were considered statistically significant where p value was less than 0.05 ($p<0.05$).

RESULTS

A total of 60 physical status ASA I-II patients were included in the study. In both groups, patient's demographic profiles were comparable with regards to mean age, sex, weight, height and duration of surgery (Table 3).

Table 3: Patient Characteristics

Characteristics	Group I (Case)	Group II (Control)	P value
Age	40 ± 8.336	38 ± 8.324	>0.05 NS
Weight	72 ± 6.220	74 ± 5.824	>0.05 NS
Height (Cm)	170 ± 5.5	168 ± 6.0	>0.05 NS
Sex (M/F)	18/12	20/10	>0.05 NS
Duration of surgery (min)	122 ± 30	115 ± 28	>0.05 NS

Values are expressed as Mean Standard deviation, NS: Not significant

Table 4. Comparison of quality of block in two groups

Quality of block	Group I (Case)	Group II (Control)	P value
Onset of sensory block (min)	12 ± 2.8	13.5 ± 2.6	0.825 (>0.05)
Onset of motor block (min)	16.5 ± 2.0	17.8 ± 3.0	0.720 (>0.05)
Duration of analgesia (min)	925 ± 60.0	322 ± 40.2	0.0001 (<0.05)*
Duration of motor block (min)	415 ± 30.0	216 ± 32.0	0.0001 (<0.05)*

Values are expressed as Mean Standard deviation, *: Statistically highly significant

The mean time to onset of sensory block in minutes in case group and control group was 12 ± 2.8 and 13.5 ± 2.6 respectively which is statistically not significant as revealed by P value (P>0.05).

The mean time to onset of motor block in minutes in case group was 16.5 ± 2.0 and in control group was17.8 ± 3.0. This is statistically not significant (P>0.05).

Duration of motor block and duration of analgesia were prolonged in dexamethasone group (Case) as compared

to control group which is statistically very significant as revealed by P value (P<0.0001).

In our study the mean duration of motor block in case group and control group were 415 ± 30.0 and 216 ± 32.0 respectively (P<0.0001) which is statistically highly significant.

The mean duration of analgesia in case group was 925 ± 60.0 and in control group was 322 ± 40.2 which is statistically highly significant as revealed by P value (P<0.0001).

Table 5: Incidence of side effects during block in two groups

Side effects	Group I (Case)	Group II (Control)
Horner's Syndrome	1	1
Dysponoea or chest discomfort	2	1
Recurrent laryngeal nerve block	0	0
Inadequate block	1	2

Incidence of the side effects observed during our study was one case of Horner's Syndrome in both groups, two cases of dysponoea in case and one in control group which are a known complication of supraclavicular block and they subsided as the effect of block wore off.

DISCUSSION

Regional anesthesia is a simple, safe, effective technique of anesthesia having distinct advantages over general and IV regional anesthesia very particularly for day care surgeries. The main reason is they can be utilized for analgesia during post-operative period besides avoiding all the problems associated with general anesthesia[23]. Supraclavicular brachial plexus block is a popular and widely employed regional nerve block technique for perioperative anesthesia and analgesia for upper extremity surgery[5]. Several adjuvants like ketamine, epinephrine, opioids, alpha-2 agonists etc. can be added to local anesthetics to prolong the duration of regional blocks and also to intensify the quality of regional blocks[24]. In our study we evaluated the efficacy of dexamethasone added to bupivacaine for supraclavicular brachial plexus block.

Synthetic glucocorticoid dexamethasone is preferred in various studies because of its potential and lack of mineralocorticoid activity. Dexamethasone is also known for its anti-emetic property. Dexamethasone is the preferred anti-emetic agent in cases of refractory nausea and vomiting.

The present study indicates that the addition of 8 mg dexamethasone to 1.0% adrenalized xylocaine for axillary brachial plexus block results in a significant increase in duration of analgesia and motor block but that the onset time of sensory and motor blockade is similar. The mechanism of the analgesia induced by corticosteroids is not fully understood. This effect is suspected to be mediated by their anti-inflammatory or immune-suppressive effects[25,26].

Steroids are very potent anti-inflammatory and immunosuppressive agents. Perineural/ IV injection of steroids is reported to influence post-operative analgesia as well[27,28].

In our study, patient's demographic profiles were comparable with regard to mean age, sex, weight, height and duration of surgery in both groups. The mean time to onset of sensory block in minutes in case group and control group was 12 ± 2.8 and 13.5 ± 2.6 respectively, which is statistically not significant as revealed by P value (P>0.05). The mean time of onset of motor block in minutes in case group was 16.5 ± 2.0 and in control group was 17.8 ± 3.0. Both these data were not significant statistically as p>0.05. So our study showed that there was no significant difference in the onset time of sensory and motor block between two groups.

In one study by Shrestha BR *et al* onset of action was 10 - 30 minutes in local anesthetic group (mean 18.15 ± 4.25) and 10 - 20 minutes (mean 14.5 ± 2.10) in the local anesthetic plus steroid group. They found statistically significant difference between two groups[19]. However another study by Ali Movafegh *et al* found that the onset time of sensory and motor blockade was similar in both the groups[29]. Our study also showed that dexamethasone does not produce significant difference in the onset time of sensory and motor block. Similarly, Pathak *et al* showed that there was no significant difference in the onset time to sensory and motor blocks between two groups in their study, which correlated with the findings of our study[30].

In present study, duration of motor block and duration of analgesia were prolonged in dexamethasone group (Case) as compared to control group which is statistically very significant as revealed by P value (P<0.0001). In our study the mean duration of motor block in Case group and Control group were 415 ± 30.0 and 216 ± 32.0 respectively (P<0.0001) which is statistically highly significant. The mean duration of analgesia in case group

was 925 ± 60.0 and in control group was 322 ± 40.2 which is which is statistically highly significant as revealed by P value (P<0.0001).

Several studies have shown that addition 4 - 8 mg of dexamethasone to local anesthetics effectively and significantly prolongs the duration of analgesia. Kopacz and Holte *et al* found that addition of small amounts of dexamethasone to bupivacaine incorporated in microcapsules prolonged local analgesia compared with microcapsules with plain bupivacaine after subcutaneous administration in humans[31]. Estebe IP *et al* studied the effect of dexamethasone on motor brachial plexus block with bupivacaine and with bupivacaine-loaded microspheres in a sheep model and found that the incorporation of dexamethasone in bupivacaine-loaded microspheres dramatically increases the duration of action[32]. Kopacz DJ *et al* also reported that the intercostal injection of dexamethasone-containing bupivacaine microspheres produces a prolonged duration of anesthesia and analgesia[33].

Many authors believe that the block prolonging effect of dexamethasone is due to its local action and not a systemic one[34]. They found that steroids produce analgesia by blocking transmission in nociceptive c-fibres and suppressing ectopic neuronal discharge. Local application of methylprednisolone has been found to block transmission in c-fibres but not in α and β fibres[35]. The effect was reversible, suggesting a direct membrane action of steroids. Steroids might bring about this effect by altering the function of potassium channels in the excitable cells[36-38]. There are others who believe that analgesic properties of corticosteroids are the result of their systemic effect[39,40].

Limitations of our study are that we did not use ultrasound guided block as our experience was insufficient to use US based technique. Analgesics were only administered on request and a post-operative regimen with regular analgesic administration might have impacted our secondary outcomes. Such a regimen is often not easy to implement in an ambulatory setting. We did not follow up the patients for long periods >3 months for chronic neurological effects of dexamethasone. The dose we used in our study is a safe dose, which was proved in several clinical trials. No other significant side-effects were noted in the study group in our study except one case of Horner's Syndrome in both groups, two cases of dyspnea in case and one in control group which are a known complication of supraclavicular block and they subsided as the effect of block wore off.

CONCLUSIONS

The addition of dexamethasone to 1.0% adrenalized xylocaine for axillary brachial plexus block significantly helps in prolonging the duration of analgesia and motor blockade in patients undergoing upper limb surgeries and is comparatively safe and cost-effective method of providing post-operative analgesia.

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Changes in Anterior Chamber Depth and Angle Width After Phacoemulsification and IOL Implantation Using Anterior Segment OCT Visante

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Keywords
Phacoemulsification,
Optical coherence tomography,
Trabecular iris angle, IOL implantation.

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ABSTRACT

Background: Anterior segment optical coherence tomography is the recent advance technique to analyse anterior segment depth and width. Advanced cataract surgery (phacoemulsification) can change the anterior chamber depth and width and the optical coherence tomography is the useful tool to quantify it.

Objectives: To quantify anterior chamber depth and angle changes after phacoemulsification with posterior chamber intraocular implantation using Visante anterior segment optical coherence tomography.

Methods: In this prospective study done in Fatima University Ophthalmology Department, patients with significant cataract were enrolled. Of the 45 eyes of 35 patients, only 38 eyes of 30 patients were included in the study. Patients who had complicated surgery were excluded from the study. Anterior chamber depths and trabecular iris angles were measured using Visante AS-OCT. Examinations were done one day before and two weeks after surgery.

Results: Preoperatively, the mean central anterior chamber depth was 2.91 ±0.41 SD. Two weeks after phacoemulsification and intra ocular lens implantation the central anterior chamber depth increased to 3.54 ±0.31 SD. All angle parameters analyzed with Visante, showed a significant increase after cataract surgery for both the nasal and temporal angles. The mean preoperative trabecular iris angle (TIA) was 20.66° ±3.732 SD at the nasal angle and 21.64°± 3.613 SD at the temporal angle. After cataract surgery, TIA increased to 24.24° ±3.264 SD at the nasal angle and 25.45° ±3.343SD at the temporal angle with a mean difference of nasal anterior chamber angle width of 3.57 ±3.13 SD and the temporal anterior chamber angle width of 3.813 ±2.69 SD.

Conclusions: The angles changed significantly after cataract surgery, particularly in the temporal quadrant. Anterior segment optical coherence tomography can be used as a rapid and safe tool in addition to diagnostic gonioscopy in assessing anterior chamber angles.

INTRODUCTION

The crystalline lens, particularly the cataractous lens has an important role in the narrowing of the angle by pushing the peripheral iris anteriorly, as demonstrated by some studies, based on advanced imaging[1,2]. It is well known that cataract surgery and intraocular lens (IOL) implantation produce clinically evident changes in the anterior segment configuration. However, only a few studies have been made to quantify these changes[3-5]. Anterior segment cross-sectional imaging is essential in evaluating the anterior chamber configuration with quantitative data. Several methods have been studied and used for the diagnosis of glaucoma or other anterior segment pathology and quantification of angle configuration. Ultrasound biomicroscopy (UBM) provides high-resolution images of the anterior chamber and is able to penetrate opaque media. With UBM, detailed images of the iridotrabecular angle and ciliary body can be obtained. However, the UBM transducer requires the eye to be immersed in a water bath of saline with the patient supine; the procedure is uncomfortable for the patient and requires a skilled examiner. Scheimpflug photography is a non-invasive and repeatable technique that uses a slit beam and camera. However, because of its optical and acoustic characteristics, images must undergo processing and do not allow visualization of the actual angle recess.

Recently, anterior segment optical coherence tomography (AS-OCT) has emerged as a new imaging technique for the anterior segment that is non-invasive and non-contact. AS-OCT provides high resolution images by using a long wavelength (1,310 nm) of light; it offers rapid and easy quantitative analysis of various structures. AS-OCT has exhibited good repeatability and reproducibility with low intraobserver and interobserver variability. One limitation of AS-OCT is that it has incomplete penetration through the pigmented epithelium of the iris, thus making it difficult to obtain accurate images of the ciliary body, lens, and zonules behind the pigmented iris[6].

We conducted a prospective study to evaluate the effect of cataract extraction by phacoemulsification on the anatomy of the drainage angle in non glaucomatous eyes with open iridocorneal angles using Visante AS-OCT available in the OPD of Fatima Eye Center.

OBJECTIVES

To quantify anterior chamber depth and angle changes after phacoemulsification with posterior chamber intraocular lens implantation using anterior segment optical coherence tomography Visante (AS-OCT) in patients with cataract.

METHODS

Data collection

Patients who had cataract surgery with phacoemulsification and in-the-bag non foldable IOL implantation between January 2010 and October 2011 were enrolled in this prospective study. The study was done at the Department of Ophthalmology, Fatima University Medical Center, and Valenzuela City, Philippines. Written informed consent was obtained from all the participants. All the patients possessed visually significant cataracts and had best corrected visual acuity of less than 20/40 in the affected eye, and with angles of Shaffer grade 3 and 4 on gonioscopy.

Patients who had eye trauma, uveitis, glaucoma, previous intraocular surgery, corneal refractive surgery, complications related to cataract surgery (posterior capsule rupture, vitreous loss, peripheral anterior synechiae), eyes with substantial corneal abnormality (such as edema, dystrophy, abrasion, marginal degeneration, or pterygium), and those lost to follow up were excluded in the study. Patients with axial length of <18mm and >24mm and IOL power of less than +18D and greater than +23D were also excluded in the study.

Preoperative and postoperative evaluation

A complete ophthalmologic exam was done before and after cataract surgery. The uncorrected (UCVA) and best corrected (BCVA) visual acuities were measured with the Snellen chart. Under slit lamp biomicroscopy, cataract grading using LOCS III was done. Keratometric values were measured with a Shin Nippon keratometer. Axial length was measured with A-scan ultrasonography and IOL power calculation was by the SRK II formula. Funduscopy was done with a +90.0 diopter lens in eyes without dense cataract. When funduscopy was not possible, the posterior segment was evaluated with B-scan ultrasonography. An ICA examination was performed using a Goldmann 3-mirror lens 1 day before surgery. All 4 quadrants were graded, using the Spaeth Grading System. The IOP was measured with a Goldmann applanation tonometer. All preoperative examinations were done by the one examiner.

The ACD and ICA were measured by Visante AS OCT the day before surgery and 2 weeks postoperatively

by the same examiner. Follow-up time points included preoperative, a day after, 2 weeks, and one month after surgery. Seidel test was also performed by applying moistened sterile fluorescein dye to anesthesized eye and using cobalt blue light under slit lamp biomicroscopy to check if there is wound leakage.

AS-OCT findings

Images of the anterior segment were obtained using a commercially available AS-OCT device (Visante OCT; Carl Zeiss) by one experienced operator, who was masked to the results of the clinical ophthalmic examinations. Standard resolution scans captured the temporal and nasal quadrants (nasal- temporal 0° - 180°) in 1 image with participants looking straight ahead and having a good central corneal reflex. All the images were taken in the same dark conditions with patients in a sitting position. After several scans were acquired, the operator selected the best image with no motion artifacts or image artifacts from the eye lids. Assessment of the superior and inferior quadrants often requires manual manipulation of the eye lids, which may distort the angle. To prevent systematic bias in angle assessment of groups of patients who may require eyelid manipulation, only images of nasal and temporal quadrants were included in this study. ACD and ICA were measured using the caliper and angle tool respectively, by one examiner (Author).

Surgical technique

All the operations were performed by the author using conventional surgical procedures. Pupils were dilated with 0.5% tropicamide, and 10% phenyl ephrine hydrochloride. Topical anesthesia was used. 2 side ports were made with 15 degree stab incision. Intracameral injection of 1% lidocaine was done. Trypan blue dye was used to visualize the anterior capsule and viscoelastic was injected. A 2.75 mm clear corneal incision was done temporally. After the incision, the continuous curvilinear capsulorrhexis measuring approximately 5.5 mm in diameter was performed using a Utrata forceps. Hydrodissection/ hydrodelination, in the bag phacoemulsification using the divide and conquer technique, and cortical aspiration were done. The main wound was extended to 5 mm prior to insertion of a rigid 5 mm IOL in the capsular bag. After viscoelastics clean up, stromal hydration was done to close side ports and main wound. Intracameral injection of Moxifloxacin 1 cc was given. The incisions were not sutured. Postoperatively, patients were given topical antibiotics and topical steroids 4 times daily for 1 month. Patients were checked one day after operation. Seidel test was done to determine if there is a wound leak.

RESULTS

43 eyes of 35 patients were enrolled in this study. Three patients had complicated surgery and two patients were lost to follow up, hence, only 38 eyes of thirty patients were included in the study. There was no incidence of infection noted.

Table1: Average age of the subjects included in the study

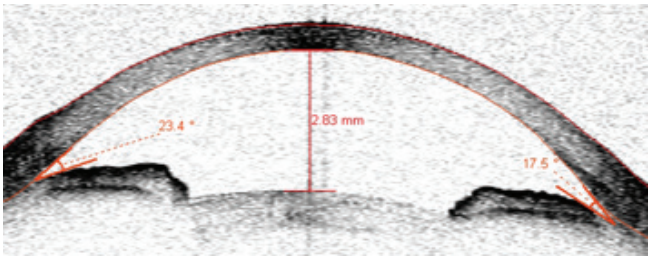
	Mean	Standard Deviation
Age	57.4	10.54

Table 2: parameters of anterior chamber depth before and after cataract surgery

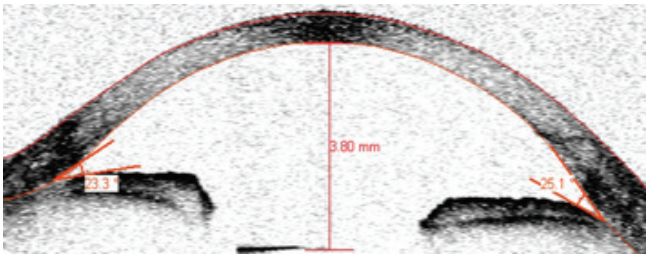
		Mean	N	Standard Deviation	Mean Difference	p-value
Pair 1	pre op	2.916579	38	.4108349	0.6247368	<0.001
	post op	3.5413	38	.31305		

Table 1 shows that the mean age of subjects was 57.4 years, with age range of 37 - 77 years old. The mean anterior chamber depth preoperatively was 2.91 ±0.41 SD (Table 2). Two weeks after phacoemulsification and IOL implantation the ACD increased to 3.54 ±0.31 SD or a 23% increase with a mean difference of 0.62 ±0.38 SD. The difference is statistically significant, p=<0.001.

Fig 1: Pre and post operative images using Visante OCT. Caliper and angle tool were used to measure the ACD and ICA



A. Pre operative image



B. Post operative image

Also, there was significant increase in Visante AS-OCT

parameters for both nasal and temporal angles after surgery (Fig 1). All angle parameters analyzed with Visante AS-OCT showed a significant increase after cataract surgery for both the nasal and temporal angles (Fig 1). The mean preoperative TIA was 20.66° ±3.732 SD at the nasal angle and 21.64° ±3.613 SD at the temporal angle. After cataract surgery, TIA increased to 24.24° ±3.264 SD or 19.76% increase at the nasal angle and 25.45° ±3.343 SD or 19.09% at the temporal angle with a mean difference of nasal anterior chamber angle width of 3.57 ±3.13 SD and the temporal anterior chamber angle width of 3.813±2.69 SD. The difference is statistically significant, p= <0.001 (Table 3).

Table 3: Anterior chamber angle (TIA) parameters before and after cataract surgery

	Mean	Standard Deviation
Percent change	23.1	16.63

		Mean	N	Standard Deviation	Mean Difference	p-value
Pair 1	nasal quadrant pre-op	20.668	38	3.7326	3.5737	<0.001
	nasal quadrant post op	24.242	38	3.2644		
Pair 2	temporal quad-pre-op	21.642	38	3.6131	3.8132	<0.001
	temporal quadrant post op	25.455	38	3.3434		

DISCUSSION

The area in the anterior chamber where the cornea and iris join is known as the angle. This is comprised of several structures that make up the eye's drainage system. The angle structures include: the outermost part of the angle, the front of the ciliary body, the trabecular meshwork, and the Canal of Schlemm. Aqueous is formed in the ciliary body behind the iris. It flows through the pupillary space into the anterior chamber. From there, the fluid travels into the angle structures and drains from the eye. As the aqueous fluid leaves the angle, it passes through a filter called the trabecular meshwork. After leaving the trabecular meshwork, the aqueous travels through a tiny channel in the sclera called the Canal of Schlemm. The aqueous fluid flows into other tiny channels and eventually into the eye's blood vessels. The production and drainage of aqueous fluid determines the eye's intraocular pressure (IOP).

Anterior segment -OCT is a light-based system that rapidly provides high-resolution images of very fine anatomical structures. Anterior segment OCT has all three elements: longer wavelength of 1310 nm, telecentric transverse scanning and very high speed axial scanning with a grating–based rapid scanning optical delay (RSOD) mechanism. Its non-contact nature ensures patient comfort and allows for rapid image acquisition in the sitting position, without risk of mechanical distortion of the angle. It also allows quantitative and dynamic data analysis with high reproducibility and repeatability[8]. This study demonstrates changes in anterior segment configuration after phacoemulsification and IOL implantation in normal eyes as measured quantitatively by AS-OCT.

The method we used to measure anterior chamber angle is measuring trabecular iris angle (TIA), which is regarded as a standardized parameter for measuring the trabecular meshwork opening. However, to obtain an exact value it is essential to localize the appropriate scleral spur. The other parameters which can be used to measure the anterior chamber angle are AOD500 and AOD750. Localization of the scleral spur is essential for these parameters and manipulation of the scale bar increases the frequency of errors.

Anterior chamber depth is the distance measured between the edge of the optic of intraocular lens and the endothelium. With the Visante OCT calipers, the safety distance between the implant and the endothelium can be precisely measured and one knows right away if the implant is in the right position. If the edge of the optic is measured <1.50 mm from the endothelium, corneal distortions that occur during eye rubbing can give rise to endothelial alteration by contact with the edge of the implant which is dangerous. If the implant is < 1mm from the endothelium, explanation may be required[8].

This study confirmed angle widening of up to 19% (at the nasal angle) and chamber deepening of up to 23% after cataract surgery, as other studies have previously demonstrated. Mean anterior chamber depth preoperatively was 2.91 ±0.41 SD. Two weeks after Phacoemulsification and IOL implantation, the ACD increased by 23% with a mean difference of 0.62 ±0.38 SD.

The mean preoperative TIA was 20.66° ±3.732 SD at the nasal angle and 21.64° ±3.613 SD at the temporal angle. After cataract surgery, TIA increased to 24.24° ±3.264 SD or 19.76% increase at the nasal angle and 25.45° ±3.343 SD or 19.09% at the temporal angle with a mean

difference of nasal anterior chamber angle width of 3.57 ±3.13 SD and the temporal anterior chamber angle width of 3.813 ±2.69 SD. The difference is statistically significant, p= <0.001.

These results are similar to results from other studies which has analyzed changes in ACD and ACA following cataract surgery[3]. The patients in our study were considered to represent the normal population without intraocular abnormalities. However, the patients enrolled in this study were primarily elderly patients with cataractous lenses; the mean age of the patients was 57 years. The method we used to measure anterior chamber angle by measuring TIA (Trabecular iris angle), is regarded as a standardized parameter for measuring the trabecular meshwork opening. However, to obtain an exact value it is essential to localize the appropriate scleral spur. The other parameters which can be used to measure the anterior chamber angle are AOD500 and AOD750. Localization of the scleral spur is essential for these parameters and manipulation of the scale bar increases the frequency of errors.

CONCLUSIONS

AS-OCT Visante measurement of the anterior chamber depth and trabecular iris angle in this study proves that phacoemulsification with IOL implantation can significantly deepen the ACD, and widen the anterior chamber drainage angle.

Summary

This prospective study done in Fatima Eye Laser Center was to quantify anterior chamber depth and angle changes after phacoemulsification with PCIOL implantation using Visante anterior segment optical coherence tomography. Patients with significant cataract were enrolled in the study. Of the 45 eyes of 35 patients, only 38 eyes of 30 patients were included in the study. Patients who had complicated surgery were excluded from the study. Anterior chamber depths and trabecular iris angles were measured using Visante AS-OCT. Examinations were done one day before and 2 weeks after surgery. After phacoemulsification and IOL implantation there was a significant increase in anterior chamber depth and angle width.

Recommendations

There is significant change in anterior chamber depth and angle width after the phacoemulsification and IOL implantation. So, it is recommended to do anterior segment OCT Visante before and after the phacoemulsification to note the changes. It may be a useful examination in patients with phacomorphic glaucoma where removal of cataract can open the trabecular iris angle. Also anterior segment OCT Visante is useful to assess the changes in TIA (trabecular iris angle) in patients with angle closure glaucoma.

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Comparison of Two Different Doses of Ketamine Used Along With Variable Doses of Propofol in Short Surgical Procedures

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TIVA, Ketamine, Propofol.

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ABSTRACT

Background: Total intravenous anesthesia (TIVA) has a definite advantage over gaseous induction on maintenance of anesthesia so, ketamine and propofol has been used as agents of choice since years.

Objectives: The study was done to compare two different doses of ketamine with variable doses of propofol.

Methods: Sixty patients were randomly selected and were divided into two groups. Induction, maintenance and recovery pattern were observed in short surgical procedures (procedures lasting <30minutes). Group K1 received 0.5mg/kg of ketamine whereas Group K2 received 1mg/kg of ketamine as induction along with variable doses of propofol to supplement induction and maintain anesthesia.

Results: Demographic pattern and intraoperative hemodynamics were observed which were relatively insignificant. Intraoperative complications like apnea, airway manipulation and hypotension were found to be significantly present in Group K1. Postoperative emergence delirium was seen in 13 out of 30 patients (43%) in Group K2 whereas it was seen in 2 patients only out of 30 patients in Group K1 (6%). Postoperative VAS score was better in Group K2 however awakening time, vital parameters postoperatively were all insignificant.

Conclusions: Thus, use of ketamine in lower dose produced better hemodynamics and preserved the advantages thus producing rapid, pleasant and safe anesthesia with fewer side effects

INTRODUCTION

The ideal anesthetic agent is the one that safely provides relief from pain, anxiety and unpleasant memories for wide variety of procedures[1]. The need for combination of agents arose as there is negligible number of drugs that can provide all the above advantages[2,3].

Propofol produces dose-dependent sedation, hypnosis, anxiolysis and amnesia with rapid onset and short duration of action. It also has anti-emetic properties however does not possess analgesic property and tends to depress the hemodynamic parameters[1,4-7]. Ketamine is a phencyclidine derivative which provides dissociative anesthesia and is also known to have analgesic and amnestic property[8,9]. It causes little or no respiratory or cardiovascular depression[4,10,11]. It is still not preferred as single agent for procedural sedation and analgesia especially in adults due to the occurrence of emergence phenomenon and concern of inducing vomiting and laryngospasm[4,8,12,13].

Ketamine and Propofol administered in combination have been used successfully in various surgeries. This combination has been favored because of the opposing hemodynamic and respiratory effects of each drug[10,11,14,15]. Use of ketamine with propofol has shown to achieve desired sedation[10]. The theoretical advantage of

combining these drugs has been tested and found to be true in various studies[1,4,16,17]. It has been associated with less nausea and vomiting, rapid recovery, fewer morbid events, greater patient satisfaction and faster hospital discharge[18].

This study was conducted to compare the effectiveness of two different doses of ketamine with variable doses of propofol. We wanted to know that the amount of ketamine required for balancing out the adverse events associated with propofol without hampering its advantages.

METHODS

This study was conducted in Gandaki Medical College Teaching Hospital in the year 2012. Sixty patients in between 10 - 60 years of age were taken into consideration. These patients belonged to American Society of Anesthesiologists states I and II, undergoing elective short surgical procedures (Procedures lasting <30 minutes). Patients who were hypersensitive to study drugs, patients with unstable hemodynamics, emergency surgeries, patients with liver and kidney diseases, any patient with chronic opiate use, pregnant females and patients with neuropsychiatric diseases were all excluded from the study.

In the pre-anesthetic visit, all patients were made familiar with the study plan and visual analogue scale to be used post-operatively. Intra-operatively, respiratory rate, non-invasive blood pressure, heart rate and oxygen saturation were monitored. Various complications were then monitored intra and post operatively. As the patient was taking into the operation theatre, intravenous cannula was inserted according to the age and availability in all patients and operation was carried out in face-mask with 3 - 4 litres/minute of oxygen. All patients received glycopyrrolate 8 mcg/kg and midazolam 50 mcg/kg as premedication.

These patients were randomly classified into two groups. In Group K1, patients received ketamine 0.5mg/kg and then anesthesia was maintained with variable doses of propofol to prevent immobilization and suitability for surgery whereas Group K2 patients received ketamine 1mg/kg instead and propofol was administered according to the requirement. Heart rate, systolic and diastolic blood pressure and saturation were measured from start to every 5 minutes till the surgery ended. Intraoperatively, patients were observed for laryngospasm, hypotension as defined by fall of systolic and diastolic blood pressure to >20% of baseline blood pressure. Nausea and vomiting during the procedure were observed and managed accordingly. Patients' whose airway manipulation had to be done during the surgery was also noted.

Postoperative monitoring included the need of airway manipulation, sedation scoring was done using Ramsey's sedation score and Kain's scoring was done for emergence delirium. VAS was measured in all patients till half an hour post-surgery. The overall satisfaction of the patient was assessed with 10 cm scale of VAS (No satisfaction at 0 cm and best satisfied at 10 cm)

Approval of this study was obtained from the ethics committee of the hospital and written informed consent was taken from each patient before the study. The collected data was analyzed using SPSS version 16. Analysis of variance (ANOVA) was used for comparison of mean values between groups and Chi-square test was used to compare discrete variables between groups. A p-value of <0.05 was considered significant.

RESULTS

The data of 60 patients were included for calculations. There was no statistically significant difference in demographic parameters like age, height, weight, American Society of Anesthesiologist grading, hemoglobin in between the two groups as shown in Table 1.

Table 1: Demographic profile of the patients

	Group K1 (Ketamine 0.5mg/kg)	Group K2 (Ketamine 1mg/kg)	P value
Age (Years)	30.6 ± 1.6	30.6 ± 1.76	0.944
Height (Meters)	1.42 ± 1.98	1.40 ± 2.1	0.780
Weight (Kg)	45 ± 9.7	46.3 ± 1.16	0.730
Hb (g/dl)	12.26 ± 1.38	12.20 ± 1.18	0.842

Duration of surgery in both the groups was insignificant with p value of 0.851. However, consumption of propofol was significantly higher in group K1 (p value <0.000). Group K1 had significant decrease in heart rate, systolic and diastolic blood pressure in first few minutes pertaining to increased use of propofol as shown in Figures 1 - 3.

Fig 1: Comparison of heart rate in between the two groups from the onset of surgery to the end of the procedure

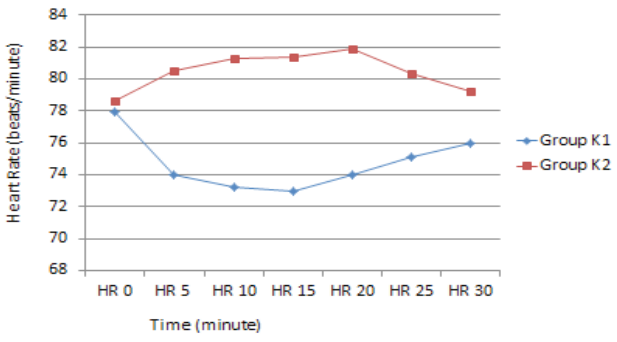


Fig 2: Comparison of systolic blood pressure between two groups

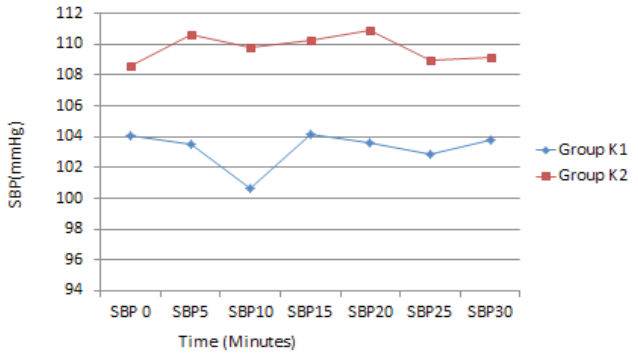
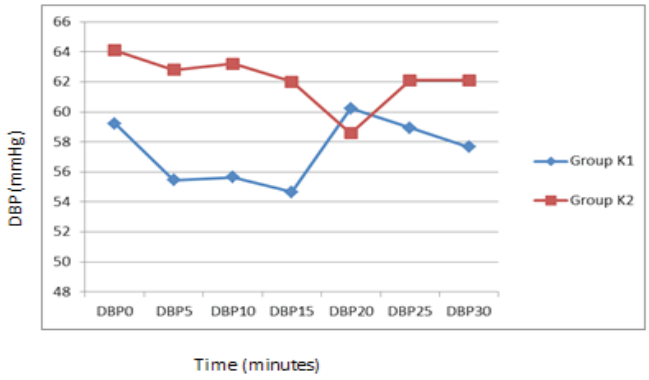
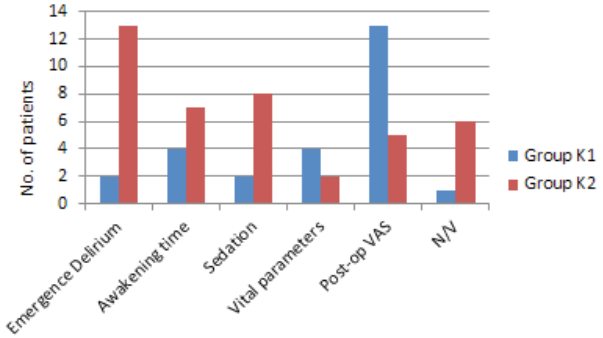


Fig 3: Comparison of diastolic blood pressure between two groups



There was also significant incidence of airway manipulation in Group K1 with p value less than 0.023 intra-operatively. However, rest of the expected complications in between groups, were insignificant. Post-operatively, Group K2 had significant incidence of emergence delirium and increased incidence of nausea/vomiting as shown in Figure 4. Mean VAS satisfaction was also significantly higher in Group K2. However, time for being awake from anesthesia as defined by Ramsey's sedation score was insignificant between the groups. Incidence of post-operative alterations in vital parameters was insignificant as well.

Fig 4: Comparison of Various features post-operatively



DISCUSSION

This study has demonstrated that even lower dose of ketamine upto 0.5 mg/kg at induction can be equally effective as higher doses. It has also shown that use of ketamine in addition to propofol did not alter the hemodynamics to much extent. However, there was requirement of airway manipulation if only analgesic dose of ketamine was used due to increased use of propofol. On the other hand, if higher dose of ketamine was used there was increased incidence of post-operative nausea and vomiting but with significantly improved VAS score.

Elaine VW *et al*[4] conducted a study to evaluate the effectiveness and safety of intravenous ketamine and propofol combination in Emergency Department. Here patients received 0.75 mg/kg of ketamine with 0.75 mg/kg of propofol in combination (Ketofol) for short surgical procedures. It was found to be safe and effective for painful procedures with minimal adverse effects. In a study done by Aouad MT[19], the combination of propofol with ketamine for invasive procedures in pediatric oncology resulted in reduced propofol and fentanyl consumption and preserved hemodynamics but agitation was a significant problem.

Bajwa *et al*[6] conducted a study in which the drug combination propofol-ketamine and propofol-fentanyl was tested. In this study, the 1st group received 1 mg/kg of propofol and 1 mg/kg of ketamine. It was noted that the hemodynamics were stable in this group whereas propofol-fentanyl group produced significant fall in pulse rate and blood pressure. However, wakefulness score, appearance of protective airway reflexes were better with propofol-fentanyl group. They concluded that both the combination provided a rapid, pleasant and safe anesthesia with few untoward effects.

Miner *et al*[20] conducted randomized controlled trial comparing ketamine 1 mg/kg with propofol 1 mg/kg and adding respective drugs as much required. They noticed an increased incidence of agitation which was nullified using midazolam.

Thus, we opted to use lesser dose of ketamine to avoid emergence delirium as seen with 1 mg/kg dose in most studies without blunting its advantage of providing analgesia. In our present study, apnea and laryngospasm was similar in both groups. Occurrence of nausea and vomiting was absent in both groups intra-operatively. However airway management had to be done in four patients in group K1 as the dose of propofol had to be increased for maintaining adequate surgical anesthesia.

There was no difference in the awakening time and patients were easily arousable post-operatively. There was

also no difference in sedation level. However, incidence of emergence delirium was found to be significant with 13 out of 30 patients in group K2 developing agitation, crying, restlessness which had to be treated with 0.05 mg/kg of midazolam. Patient had post-operative retching in group K2 though the post operative VAS score was better in the same group.

The limitation of this study was that it was done in a small setup with lesser number of patients. Patients were only observed for 30 minutes post-operatively. Thus, lower dose of ketamine 0.5 mg/kg combined to propofol can be better alternative to higher dose to prevent adverse reactions caused by increased use of ketamine though study has to be conducted in larger group before coming to a concrete conclusion.

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Analgesia for Newborns during BCG Vaccinations

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Keywords
Analgesia, Vaccination, Dextrose, Breast feeding.

Abbreviations

APGAR - Appearance, Pulse, Grimace, Activity, Respiration
BCG - Bacillus Calmette Guerin
NICU - Neonatal Intensive Care Unit
NIPS - Neonatal Infant Pain Scale
PAIN - Pain Assessment in Neonates

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ABSTRACT

Objectives: The aim of this study was to compare the analgesic effect of breast feeding and oral 10% dextrose during painful injections.

Methods: A convenient sampling was done among 400 healthy full-term newborns, at the United Mission Hospital, Okhaldhunga, Nepal. Two hundred each newborns were divided to receive either breast feeding or oral 10% dextrose ingestion while undergoing 0.05 ml intradermal BCG vaccination. An informed verbal consent was obtained from the guardian of eligible patient after aims and objectives were explained to them. To reduce the biasness, babies were alternately randomized to receive either breast feeding or oral 10% dextrose. The physiological and behavioral changes observed during the injection were recorded according to the NIPS (Neonatal Infant Pain Scale) pain scoring scale. The study groups were assessed according to the pain score and compared.

Results: The mean NIPS pain score out of 7score for breast feeding and oral 10% dextrose was found to be 1.74 $\pm$1.88 and 0.80 $\pm$1.24 respectively. About 60% of the babies in the oral dextrose group had a score of zero when compared with 28% in the breast feeding group. Overall, newborns in the oral 10% dextrose group showed excellent pain reduction (NIPS score <3) when compared with the breast feeding group (P <0.00029).

Conclusions: Oral 10% dextrose is found to be more effective than breast feeding alone as analgesia.

INTRODUCTION

Until now perception of minor pain of newborn has been under evaluated and has been ignored. Babies do have pain like children and adults but the way of expression is different. Some common practice done to reduce pain are holding the baby in the arm, breast feeding, feeding sweet solutions etc, and articles are now also showing similar good outcome concerning pain reduction.

It was generally assumed that newborn babies do not feel pain, or if they did, that it was a rather vague and easily forgotten experience that did not need painkillers. Because of this, little analgesia was used when newborns had operations or other medical procedures. The other reason analgesia was rarely used is because most strong painkillers can stop a baby from breathing unless carefully supervised. The belief that newborns felt little pain came from research that found that a newborn's pain sensing system was not yet fully developed, and that they had fewer pain-receptor cells and nerves than children and adults.

Recently it has been shown that single, very painful experiences result in the baby having exaggerated responses to later less painful experiences such as routine immunizations.

The International Association for the Study of Pain has defined as unpleasant sensory and emotional experience associated with actual or potential tissue damage or describe in terms of such damage[1]. The interpretation of pain is subjective, each person forms and internal construct of pain through encountered injury. Several experts[1] suggest that the neonates' expression of pain does not fit within the strict definition of the study of pain because of the requirement for self report. This lack of ability to report pain contributes to the failure of health care professionals to recognize and treat pain aggressively during infancy and early childhood. Because neonates cannot verbalize their pain, they depend on others to recognize access and manage their pain. Therefore, health care professionals can diagnose neonatal pain only by recognizing neonates associated behavior and physiological responses.

Studies indicate a lack of awareness among health care professionals a pain perception assessment and management in neonates. When analgesics were used in infants, they were often administered based only on the perception of health care professionals or family members. Fear of adverse reactions and toxic effects often contributed to the inadequate use of analgesics. In addition, health care professionals often focus on treatment of pain rather than the systemic approach to reduce or prevent pain.

Neonates are sensitive to pain and vulnerable to both short and long term effects. Recognition of the clinical importance of neonatal pain and stress has been delayed and hampered by the lack of awareness that newborns are capable of experiencing pain, insufficient knowledge about the development of the nervous system, difficulty in assessing neonatal pain, lack of evidence for the safety and efficacy of different modalities available for the treatment of pain, and fears about adverse effects associated with analgesic use.

Babies face painful injections from their first day of life as a part of routine national immunization programme. Simple measures are now available especially for the babies like breast feeding or oral glucose solutions during the injections. So far recent studies[3] have evaluated venepuncture or heel prick pain with different pain-scoring system found that active breast feeding or glucose solution during procedures reduces the pain. BCG vaccination is given by an intradermal injection. We consider that intradermal injection pain is more intense. But until now, there has been no study done to evaluate the pain perceived by newborn during intradermal injection. Newborn babies are routinely given BCG vaccination by intradermal route.

Therefore, this study will evaluate the pain perceived by neonates during intradermal injection and then will try to answer whether breast feeding or glucose solution feeding or both of these measures are effective in reducing the pain perception in neonates.

OBJECTIVES

1. To establish that simple measures are effective in reducing pain in neonates during painful injections.
2. To evaluate and compare the analgesic effect of oral 10% dextrose and breast feeding to reduce pain in the neonates during painful injections.

METHODS

Research Design

A Convenient sampling was conducted for a period of 6 months starting from October 2009 to May 2009 in Okhaldhunga Mission Hospital. A size of 200 cases in each group of breast feeding and oral 10% dextrose was collected. The first patient (Newborn baby) was selected for intervention with breast feeding, and the next consecutive one automatically went into intervention with oral 10% dextrose ingestion. So that by the end of the study there were equal number of newborns in each group when enrolled.

Sampling process

Inclusion criteria

All full term deliveries (Spontaneous vaginal, instrumental vaginal and cesarean deliveries) at the hospital and normal physical examination.

Exclusion criteria

- APGAR score less than 7 at both one and five minutes were excluded, premature babies/preterm babies or babies born before 37 weeks gestational age, weight less than 2000 gram, acute or chronic illness of the mother during late pregnancy, ill babies, babies on any medications.

Neonatal Pain Scoring Scale

Pain scoring tools developed by health care professionals show that the expression of pain is present and reliably observable. Neonates also remember pain, which is

apparent by observing their reaction to subsequent pain experience, such as immunization[2].

Neonatal Infant Pain Scale (NIPS) measures intensity of infant responses to pain or painful stress and the infant's ability to be consoled. NIPS scoring guidelines are shown in Table 1.

Table 1: Neonatal Infant Pain Scale

Pain assessment		Score
Facial Expression		
0 – Relaxed Muscles	Restful face, neutral expression	
1 – Grimace	Tight facial muscle: furrowed brow, chin, jaw (negative facial expression)	
Cry		
0 – No cry	Quiet, not crying	
1 – Whimper	Mild moaning, intermittent	
2 – Vigorous Cry	Loud scream: rising, shrill, continuous	
Breathing Patterns		
0 – Relaxed	Usual pattern for this infant	
1 – Change in Breathing	Indrawing, irregular, faster than usual: gagging; breath holding	
Arms		
0 – Relaxed/ Restrained	No muscular rigidity: occasional random movements of arms	
1 – Flexed/ Extended	Tense, straight legs: rigid and/or rapid extension, flexion	
Legs		
0 – Relaxed/ Restrained	No muscular rigidity: occasional; random leg movement	
1 – Flexed/ Extended	Tense, straight legs: rigid and/or rapid extension, flexion	
State of Arousal		
0 – Sleeping/ awake	Quiet, peaceful sleeping or alert random leg movement	
1 – Fussy	Alert, Restless, and thrashing	
	Score obtained	

Research Outcome Measure

The primary outcome measure was the NIPS (Neonatal Infants Pain Scale) score less than or equal to 3 was considered no pain or tolerable pain after the use of study agents in each group and the score more than 4 was considered pain. The score from both the study groups (breast feeding and oral 10% Dextrose) was recorded, and compared, as another outcome measure.

RESULTS

There were 200 newborns in each group (Breast feeding versus oral 10% dextrose) at the end of study. The cases were randomized to receive study agent on convenient alternate basis. Data were tabulated, and calculated using the SPSS (Statistical Program for Social Science) program. P value less than 0.05 was considered significant.

Results were analyzed in terms of difference in NIPS pain score observed with the study agent (Breast feeding versus oral 10% dextrose).

Table 2: Newborns characteristics, mean birth weight and gender distribution

Characteristics	Patients (n)		P value
	Oral 10% Dextrose (n=200)	Breast Feeding (n=200)	
Birth wight (gm)	2853± 421.72	2930± 372.19	0.05352
Gender (Male/Female)	99/101	108/92	0.38780

Table 2 shows no statistically significant difference in mean birth weight and sex between the groups.

Table 3: Showing the number of newborns showing different pain scores between the groups.

	Patients		P value
Pain scores	Dextrose 10% (n=200)	Breast Feeding (n=200)	
0	118	56	
1	33	55	
2	36	51	
3	05	09	
4	03	04	
5	01	03	
6	04	18	
7	00	04	
Mean±SD	0.80±1.24	1.74±1.88	0.0000

Table 3 shows that there was statistically significant difference in the mean NIPS pain score between the group (P value <0.0000).

Table 4: Showing the NIPS pain score above and below 3 among the newborns between the two groups

	Patients		P value
Pain scores	Dextrose 10% (n=200)	Breast Feeding (n=200)	
<3	192	171	
>4	08	29	0.00029

Table 4 reveals that there was clearly statistically significant difference noted in the oral dextrose group, large number of babies has NIPS pain score of less the 3, P value <0.00029.

DISCUSSION

Minor pain in newborn has always been under evaluated, especially when the procedures like venepuncture, vaccinations, blood sample collections etc were done for certain indications. Routine vaccinations like BCG which is given at the time of birth is an intradermal injection, the benefit of vaccination was always outweighed against the pain in newborns, and considered as unavoidable. This study came up with all these issues and tried to answer regarding pain perception by the neonates and the effect of simple basic measures to control pain.

Until now, breast feeding was considered for several advantages with immune protection, nutritional supplementation, to develop bonding between mother and baby etc, but at present literature is showing that, it also has additional another important advantage of pain reduction[6,7]. Whereas, the different sweet solutions especially sucrose has been used extensively for pain reduction in newborns with impressive outcomes[4,5,12,13]. Since the sucrose was not readily available, we used 10% dextrose solution to assess the antinociceptive effect for practical reason to compare with breast feeding.

This study was done at United Mission hospital, Okhaldhunga among the newborn healthy babies, who were randomized to receive either oral 10% dextrose or breast feeding during BCG vaccination. The BCG vaccination is routinely done on the day of discharge; about 0.05 ml of BCG is given intradermally over left upper arm or at deltoid region. The pain produced was unavoidable so there was full participation from the study group.

The mean birth weight and male to female ratio were not found to be significantly different among the two groups. Since the babies enrolled were clinically normal, all the babies received BCG vaccination within 24 to 48 hours after birth. This study has shown that both breast feeding and oral dextrose has efficient role in pain reduction with mean NIPS pain scores 1.74 ±1.88 versus 0.80 ±1.24 respectively.

This study has shown that oral 10% dextrose is more effective than breast feeding for pain reduction, about 60% from the dextrose group had NIPS pain score of zero compared with 28% from the breast feeding group. This was clearly statistically significant difference P value <0.0000. The overall mean pain score was also lower for the oral dextrose group with significant difference of P value <0.0000. We considered NIPS pain score less than 3 as no or very minimal pain. We found maximum number of babies in oral dextrose group had NIPS pain score less than 3 compared with the breast feeding, 96% versus 85.5% respectively. This difference in pain scores was also statistically significant with P value 0.00029. This study revealed that 10% dextrose had a statistically significant antinociceptive effect than breast feeding which reduced the immediate behavioral pain response and shortened crying time after BCG vaccination.

Similarly few other comparative studies have also shown that oral dextrose was more analgesic than breast feeding[8,15,16,20,21]. The analgesic effect elicited by sweet solutions is probably mediated by the release of endogenous opioids[4,5,12]. Although sucrose has been extensively used in clinical and animal studies, there are few studies that have investigated the anti-nociceptive effect of dextrose[12,13]. Several researchers[17] have demonstrated the effects of other sweet solutions including glucose, fructose and artificial sweeteners in reducing minor procedural distress in full-term infants. Some concern has been raised about the potential for increased risk of necrotizing enterocolitis as a result of repeated administration of hyperosmolar dextrose[4,18]. But we found no cases of such complications or other associated with oral dextrose ingestion in this study.

Breast feeding with proper positioning and contact has significant effect on pain reduction than just keeping the baby on the cot during painful procedure. But the analgesic effect of oral dextrose was more pronounced when compared with breast feeding. This was also well documented by Carbajel and group study[14] that, the analgesic effect of breast feeding was superimposed by dextrose solution with reduced crying time and minimal pain responses in the premature infants receiving venepuncture.

In this study we choose NIPS pain scoring chart as this was practically easy to do with no additional instrumental intervention. Moreover, several papers also showed that NIPS scoring system are more easy and valid, and associated with neonatal behavioral and pain response[9, 10,11,12,19]. We considered NIPS pain score less than or equal to 3 as minimal pain to compare the analgesic effect of breast feeding and oral 10% dextrose while BCG vaccination. We also found that most of the babies in the oral dextrose group had NIPS pain score of zero.

It seems that breast feeding is a simple method to reduce pain, but if available oral 10% dextrose should be used

during painful vaccinations, injections or venepunctures.

Further research is required for different preparations of oral dextrose solutions for its optimal effect, though the outcome of this study has clearly shown that oral 10% dextrose solution is efficient as analgesia in newborns during painful procedures.

CONCLUSIONS

This study has clearly demonstrated the analgesic efficacy of breast feeding and oral 10% dextrose ingestion during painful procedures like BCG vaccination. When compared individually oral 10% dextrose showed statistically significant analgesic effect over breast feeding during BCG vaccination.

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Dexmedetomidine as an Adjunct to Bupivacaine in Subarachnoid Block among the Patients undergoing Lower Extremity Orthopedic Surgery, a Randomized Prospective Study

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Keywords
Subarachnoid block, Dexmedetomidine, Bupivacaine.

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ABSTRACT

Background: Spinal anesthesia is commonly performed for lower extremity surgeries. When local anesthesia is used alone, high doses of opioids are given to relieve postoperative pain. Addition of vasoconstrictors and opioids enhances the quality of subarachnoid block. Dexmedetomidine (DXM) is one such drug. It is highly selective alpha-2 agonist and is approved as an intravenous sedative and co-analgesic drug. Its use is often associated with decrease in heart rate and blood pressure.

Objectives: The objective of the present study was to determine the various advantages of using DXM in combination with bupivacaine over bupivacaine alone in subarachnoid block among the patients undergoing lower extremity orthopedic surgeries.

Methods: It was a randomized, prospective study. A total of 40 patients were included with 20 in each group B and group D. All patients with American society of Anesthesiologists (ASA) grading I and II, in between age 18 - 65 years, scheduled for surgery under spinal anesthesia either elective or emergency was included in the study. Patients were randomly divided and assigned into two groups. In group B, patients received only bupivacaine 0.5% 12.5 mg for subarachnoid block (SAB) and in group D, patients received Dexmedetomidine 5 mcg in addition to Bupivacaine 0.5% 12.5 mg for SAB.

Results: The result showed that there was significantly faster motor and sensory onset and longer duration of action in the group which DXM was used. However there was no significant difference in the hemodyanamics.

Conclusions: To increase the duration of action (sensory and motor blockade) and fasten the onset of action, DXM can be added to Bupivacaine.

INTRODUCTION

Spinal anesthesia is commonly performed for lower extremity surgery[1]. The advantage of subarachnoid blockade (SAB) is its simplicity in technique, rapid onset and avoidance of many life threatening complications of general anesthesia[2,3]. Hyperbaric bupivacaine and tetracaine are the most commonly used agents for

SAB[4,5]. Both are relatively slow in onset (5 – 10 mins) and have prolonged duration (90 – 120 mins)[3]. When local anesthetic agents are used alone, high doses of opioids are given to provide adequate post-operative pain relief[6,7,8]. The addition of drugs that prolong the anesthesia can lessen the occurrence of side-effects like nausea, vomiting and respiratory depression[4,9].

Addition of vasoconstrictors (alpha-2 agonist) and opioids may enhance the quality and or prolong the duration of SAB[9,10]. Vasoconstrictors appear to decrease the uptake and clearance of local anesthesia from cerebrospinal fluid and may also have weak analgesic property[10]. Alpha-2 agonists produce diverse response including sedation, analgesia and anxiolysis each of which has been reported in treatment of surgical and chronic pain management[10,11]. Dexmedetomidine, a highly selective alpha-2 agonist is the most effective sedative and analgesic[12] which has been used in our study.

METHODS

It was a randomized, prospective study between use of bupivacaine alone or with addition of DXM for spinal anesthesia in lower limb orthopedic surgeries. A total of 40 patients were included with 20 in each group B and group D. All patients with American society of Anesthesiologists (ASA) grading I and II, in between age 18 - 65 years, scheduled for surgery under spinal anesthesia either elective or emergency was included in the study. Written and informed consent were taken from all the patients. Patients with unstable hemodynamics, ASA physical status >III, Patients with neuromuscular dysfunctions, history of chronic back pain or previous low back surgery or patients unable to consent for the trial were excluded from the study.

Patients were randomly divided and assigned into two groups. In group B, patients received only 0.5% bupivacaine 12.5 mg for subarachnoid block (SAB) and in group D, patients received Dexmedetomidine 5 mcg in addition to 0.5% bupivacaine 12.5 mg for SAB. Data was collected by the author with the help of an impartial assistant. Demographic profile of the patients along with the onset and duration of sensory and motor block, hemodynamic status (Blood pressure and heart rate) and postoperative analgesia were observed.

The onset of sensory blocked was defined as the time for loss of sensation to T10 dermatome level and achievement of Bromage 3 was taken as the time taken for onset of motor blockade. Similarly, reversal of sensory symptom was signified by regression of sensory block to S1 dermatome level and return of muscle power to Bromage 1 signified the time for reversal of motor blockade.

SPSS version 20 was used in processing the data to ensure accuracy and computations.

RESULTS

There were 20 patients in each groups B and D. Out of 40 patients, 70 % were males and rest were females. The mean age among the patients was 33+/-9 in group B, and 32+/-9 in group D. 18 patients in group B were ASA I and the remaining 2 belonged to ASA II, whereas 7 patients out of 20 in group D belonged to ASA II.

Table 1: Patients' Characteristics

	Group B (n=20)		**Group D** (n=20)		**P value**
Profiles					
Sex					
Females	8	40%	6	30%	
Males	12	60%	14	70%	0.741
Age (mean +/- SD)	33.45	9.72	32.10	9.36	0.657
ASA					
I	15	75%	11	55%	0.185
IE	3	15%	2	10%	0.633
II	1	5%	5	25%	0.077
IIE	1	5%	2	10%	0.548
Complications					
Shivering	8	40%	3	15%	0.077
None	12	60%	17	85%	0.077

Systolic blood pressure was significantly different at baseline and after two minutes of SAB however the values were insignificant thereafter and as for diastolic blood pressure, the fluctuations were significant from baseline till 30 minutes after SAB. There was no significant difference in heart rate of the patients between the groups from the start to 60 minutes of SAB. The onset of motor blockade (defined by Bromage 3) and sensory blockade (time to reach T10 dermatome) were relatively slower in group B as compared to group D. The mean time of motor onset was 4.29 minutes in group B and 3.31 minutes in group D whereas sensory onset was 2.86 minutes and 2.14 minutes respectively.

Similarly, reversal of motor symptoms as defined by return of muscle power to Bromage 1 and sensory symptoms by regression of sensory block to S1 dermatome level was relatively faster in group B as compared to group D. In group B, sensory reversal occurred at a mean time of 4.35 hours and motor reversal at 3.45 hours whereas in group D, the time increased to 7.48 hours and 5.58 hours respectively.

The result also showed that the duration of post-operative analgesia was significantly higher in group D which was 10 hours as compared to group B where analgesia lasted for an average of five hours. Eight patients out of 20, in group B and three patients in group D had developed shivering intraoperatively. The occurrence of shivering was insignificant among the groups with p value of 0.077.

Fig.1: Significant difference seen in onset of motor and sensory blockade (P <0.005)

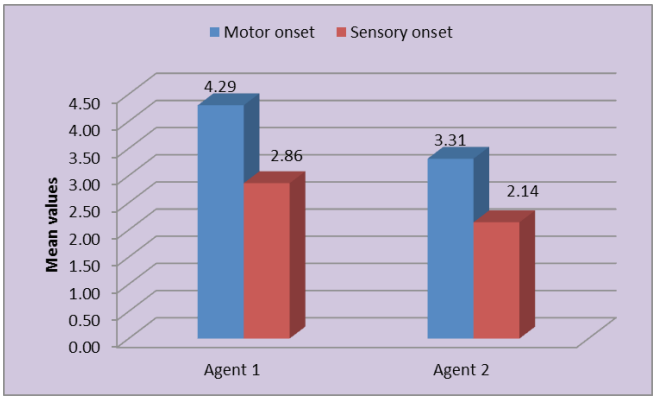


Fig 2: Significant differences seen in reversal of motor and sensory blockade (p <0.005)

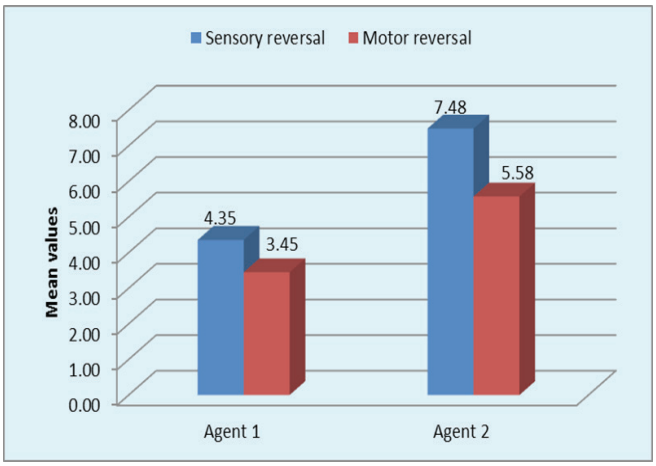
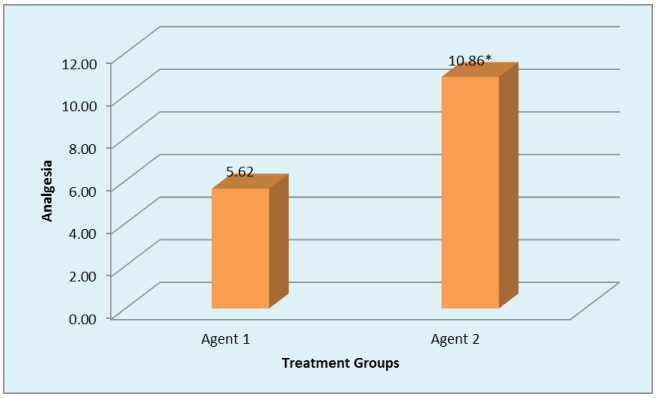


Fig 3: Postoperative analgesia seen to be significantly longer in agent 2, i.e. group D



DISCUSSION

Neuraxial blocks results in sympathetic blockade, sensory analgesia and motor blockade depending on dose, concentration of volume of anesthetic agent[1,2]. There are many choices of drugs for SAB[5]. Lidocaine is often chosen for shorter procedures however the clinical use has decreased because of incidence of back and leg pain (Transient neurological symptom)[3]. Agents like bupivacaine, tetracaine, ropivacaine and levobupivacaine for surgeries lasting >90minutes[4]. Adrenergic agonists such as epinephrine, phenylephrine, clonidine etc have been used in an effort to prolong the duration of spinal anesthesia[8,10].

Dexmedetomidine, a highly selective alpha-2 agonist, is an intravenous sedative and co-analgesic drug[11,12]. The improved specificity of DXM to alpha-2 receptor causes it to be more effective sedative and analgesic[12,13]. It is eight times more specific to alpha-2 receptor than clonidine. Besides its action in the locus cerulus of brain, it directly acts on alpha-2 receptors in spinal cord, thus inhibiting the firing of nociceptive neurons[12]. Its use is often associated with decrease in heart rate and blood pressure[11]. Thus it has been used intravenously as well as adjuvants in regional anesthesia. It has shown to reduce the requirements of opioids and inhalational anesthetic agents[13,14].

Kanazi*et al*[14] found that 3 mcg DXM and 30 mcg Clonidine are equipotent intrathecally when added to bupivacaine in patients undergoing urological procedures. It was found that there was significant reduction in onset time of sensory and motor blockade. Its use also significantly prolonged the duration of sensory and motor block without serious side-effects.

Shukla *et al*[15] conducted a comparative study of intrathecal DXM with intrathecal magnesium sulphate ($MgSO_4$) used as adjuvants to bupivacaine in which they found that the onset of anesthesia was rapid and of prolonged duration in DXM group. In $MgSO_4$ group, although the onset of block was delayed, the duration was significantly prolonged as compared to control group but to a lesser degree than DXM group. The groups were similar with respect to hemodynamic variables and there were no significant side-effects in either group. Subhi M *et al*[16] also did a study comparing the effects of adding DXM and fentanyl to intrathecal bupivacaine for gynecological procedures. The group receiving DXM with bupivacaine had longer sensory and motor block than those who received fentanyl though the onset of both motor and sensory block was not significant among groups.

Al-Mustafa *et al*[17] conducted a study to find out the

effect of DXM added to spinal bupivacaine for urological procedures in 2008. They divided patients into three groups: 1St group received bupivacaine alone, 2nd had 5 mcg DXM added and 3rd group had 10mcg DXM added to bupivacaine. They found that DXM had dose dependent effect on onset and regression of sensory and motor block when used as an adjunct to bupivacaine in spinal anesthesia.

In our study we noticed that there was no significant difference in the occurrence of shivering among the groups. There is a study which showed that there was significant reduction in shivering during spinal anesthesia without increased side-effects[18]. However, this study was conducted using intravenous infusion of dexmedetomidine.

CONCLUSIONS

Thus the addition of 5 mcg of dexmedetomidine in 0.5% bupivacaine increases the duration of sensory and motor blockade. It also significantly increases post-operative analgesia without much difference in the hemodynamic parameters.

Limitations of the study

Duration of hospital stay and amount of money spent by patients at the hospital for the trial was not estimated in this study. Small sample size, single centre study is other limitations. These things can be addressed in future well conducted comparative studies.

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Study of Anemia amongst Adolescents and the Effect of Information, Education and Communication (IEC) in Rural Area of Chagunarayan VDC of Bhaktapur District, Nepal

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ABSTRACT

Background: Adolescence is a period of rapid change and opportunities. It is an age in which children grow into young adults. It constitutes 10 - 19 years of age. In Nepal 23% of the population are adolescents.

Objectives: To find out the prevalence of anemia in school children in Chagunarayan, and to study the effect of information, education and communication (IEC) on anemia.

Methods: A cross-sectional community based study was carried out in three schools in Chagunarayan (Bhaktapur). Three hundred fifty school children were screened for hemoglobin estimation by simple random sampling method. Hemoglobin estimation was done by cynomethemoglobin method in their respective schools. Anemia was diagnosed according to WHO guidelines. Information, education and communication was given to all adolescent school children thrice in a month and again the effect of IEC was justified by estimating hemoglobin by cyanmethemoglobin method.

Results: The prevalence of anemia in Chagunarayan was 43.1% with males having more 54.1% than females 35.3%. Majority of the adolescents who were between the age group 13 - 15 years were anemic in both the sexes. The mean hemoglobin before IEC in males was 12.51 gm/dl and after IEC 12.97 gm/dl. Similiarly, in females mean hemoglobin was 12.26 gm/dl and after IEC 12.81 gm/dl. The mean increase in hemoglobin in males was 5.16% and in females 4.48% which shows statistically significant difference between the two hemoglobin concentrations.

Conclusions: This study shows that male adolescents can also be victims of anemia if proper nutrition and good healthy behavior is not practiced. The effect of IEC was justified.

Keywords
Anemia, Information, Education, Communication.

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INTRODUCTION

Adolescence constitutes 10 - 19 years of age. In Nepal 23% of the population are adolescents[1]. Adolescence is a period of rapid growth, weight gain, and blood volume expansion. Due to the rapid growth, adolescents are especially vulnerable to anemia. The overall nutrition requirement of the body increases

during this period and if not met adolescents will be anemic. Adolescence is not simply an extended childhood, but it is a non-homogenous group of growing individuals reaching adulthood. It is also the period of opportunities and change. This is a period of life when not only physical but psychological and behavioral changes take place in the individual. This is probably the most critical and vulnerable phase of human development to prepare generation of citizens to take over the responsibility.

METHODS

A cross-sectional community based study was conducted in Chagunarayan village in Bhaktapur District. Chagunarayan is located in North Eastern part of Kathmandu valley. It consists of rural and semi-urban areas. The study period was from July 2011 to August 2012. The study population was all school going adolescents in Chagunarayan and 350 adolescents participated in the study from three schools. Simple random sampling technique was applied.

Data collection and analysis

Blood sample for hemoglobin estimation was taken in respective schools and hemoglobin estimation was done in a room provided by the school authorities. The hemoglobin test was estimated by cyanamethemoglobin method. Written voluntary consent was taken from participants before drawing blood sample. The results were disclosed to the entire participants in their respective schools and appropriate IEC advice was given. WHO guidelines were used for diagnosing and grading anemia. A pre-tested, pre-coded questionnaire schedule was used for the study purpose. School children were examined and interviewed in their respective schools in Chagunarayan. Socio-demographic information of the adolescents and their families were collected in pretested proforma. All school children were given health information, education and communication before the procedure and at every three weeks thrice about anemia and dietary habit. The effect of IEC was observed after three months by estimation of hemoglobin concentration again by same cynamethemoglobin method using same standard calorimeter. Data thus generated was collected, compiled and analyzed by using SPSS 16.

RESULTS

Table 1: Sex wise distribution of anemia in adolescents

Sex	Anemia	No anemia	Total
Males	79 (54.1%)	67 (45.9%)	146 (100%)
Females	72 (35.3%)	132 (64.7%)	204 (100%)
Total	151 (43.1%)	199 (56.9%)	350 (100%)

Chi Square value=12.28, df=1, P value=0.000

Fig 1a: Age wise distribution of anemia in male adolescents

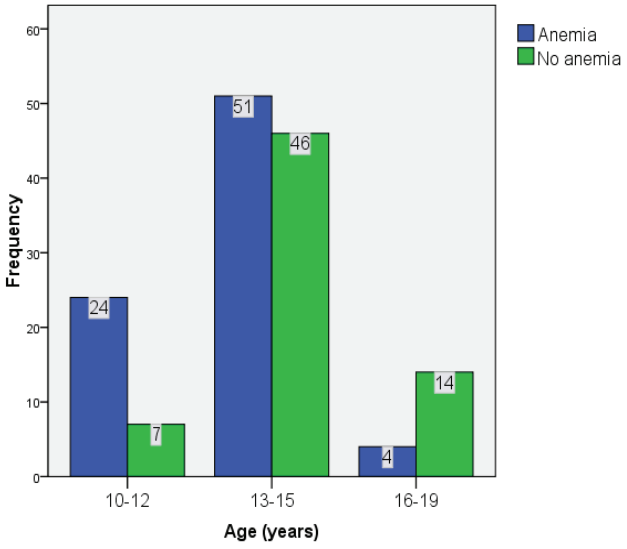


Fig 1b: Age wise distribution of anemia in female adolescents

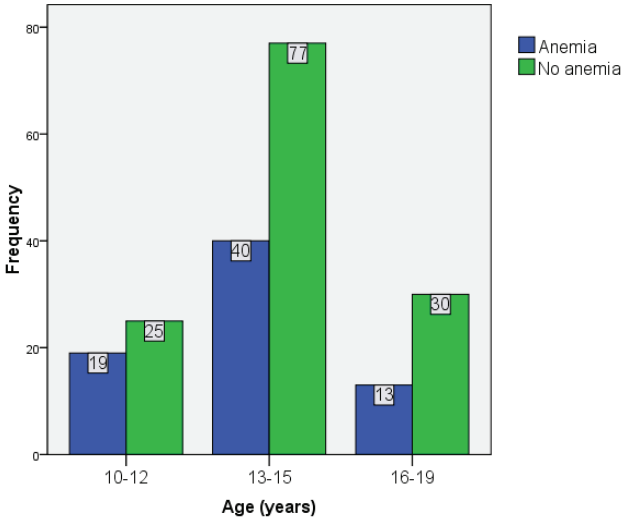


Table 2: Economic status of family and anemia in adolescents

Family monthly income (NPR)	Males (n=146)		Females (n=204)	
	Anemia	No anemia	Anemia	No anemia
< 8000	19 (61.3%)	12 (38.7%)	14 (34.1%)	27 (65.9%)
8000-12000	35 (56.5%)	27 (43.5%)	40 (34.2%)	77 (65.8%)
12000-16000	12 (42.9%)	16 (57.1%)	12 (44.4%)	15 (55.6%)
> 16000	13 (52.0%)	12 (48.0%)	6 (31.6%)	13 (68.4%)
Total	79 (54.1%)	67 (45.9%)	72 (35.3%)	132 (64.7%)

Males: Chi square value=2.253, df=3,
P value = 0.522;

Females: Chi square value=1.191, df=3,
P value = 0.755

Table 3: Hemoglobin (Hb) level before and after information, education and communication

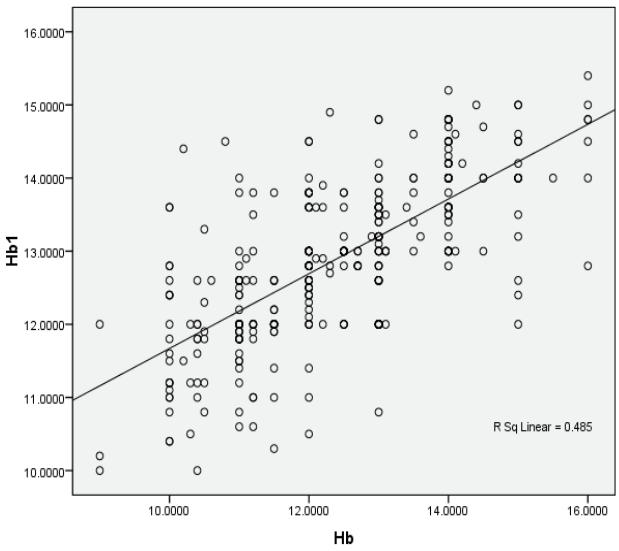
Sex	Hb	n	Min.	Max.	Mean	Standard Deviation
Males	Hb	146	10.00	16.00	12.51	±1.51
	Hb1	146	10.40	15.40	12.97	±1.10
Females	Hb	204	9.00	16.00	12.26	±1.43
	Hb1	204	10.00	15.00	12.81	±1.05

Hb: Before IEC; Hb1: After IEC

Table 4: Change in percentage of hemoglobin in average after IEC

Sex	Frequency (n)	Mini. (%)	Max. (%)	Mean Increase (%)	Std Deviation
Males	146	-20.00	36.00	5.1648	±8.64
Females	204	-20.00	41.18	4.4824	±9.68

Fig 2: Scatter diagram showing correlation between initial and after Hb in Adolescents after information, education and communication intervention



Hb- Pre IEC Hb level; Hb1-Post IEC Hb level

The prevalence of anemia in Chagunarayan was observed in 151 (43.1%) adolescents. Out of 146 males, 79 (54.1%) had anemia and out of 204 females, 72 (35.3%) had anemia (Table 1).

Fifty one males had anemia in the age group of 13 - 15 years followed by 24 males in 10 - 12 years age group and minimum number of participants with anemia (4) were detected among 16 - 19 years age group. Fourty females had anemia in the age group of 13 – 15 years, followed by 19 in age group 10 - 12 years and minimum number of participants with anemia were found in the age group 16 - 19 years. Anemia in the age group of 13 - 15 years was almost double the age group of 10 - 12 years (Fig 1 a & b).

Maximum anemic males were observed in those whose parental income is <8000 NPRs and contributes in males 19 (61.3%) and in females income between NPRs 12000 - 16000, 12 (44.4%) respectively. In male subjects having parental income >16000, 13 (52.0%) had anemia and in females 6 (31.6%) were anemic (Table 2). Both male and female groups exhibited increase in hemoglobin significantly P value 0.000. Mean hemoglobin in males before IEC was 12.51 SD ±1.51 and after IEC it was Mean 12.97 SD ±1.10. Similiarly in females before IEC Mean 12.26 SD ±1.43 and after IEC it was Mean 12.81 SD ±1.05 (Table 3).

The Mean increase in percentage after IEC was 5.16 SD ±8.64 in males and 4.48 SD ±9.68 in females. The range was -20 to 36% in males and -20 to 41.18%

in females (Table 4). The Coefficient of relationship (Karl Pearson coefficient of correlation) between two hemoglobin levels before and after giving IEC is 0.696, which is statistically significant with positive correlation (p=.000) and r^2=0.485 (Fig 2).

DISCUSSION

Adolescence (10 - 19 years of age) is a period of rapid transition in life from "childhood" to "adulthood". This phase of life is full of opportunities and healthy adolescents are a great asset for contributing to nation's development. However adolescents are also exposed to risk of infection and are vulnerable group at the same time. Adolescence is generally perceived to be a healthy period of life because mortality is relatively low in this age group. This is, however deceptive, since adolescents face many challenges in their life and several of these challenges relate to their health. Many nutritional surveys have identified that adolescent group is at increased risk for anemia all the time. The overall nutritional requirement increases during this period and if not met it will result in anemia. Anemia though global in occurrence is more of a concern in the developing countries because of its high prevalence in these regions. Inspite of its high prevalence in children, studies on prevalence in adolescents especially in boys are relatively less from developing countries. Adolescence is a period of rapid growth, weight gain and blood volume expansion and with inadequate and improper dietary habits, one is vulnerable to all kinds of nutritional morbidities. Malnutrition and worm infestation further aggravate the problem especially in rural area.

The WHO proposed a scheme for classification of public health severity of anemia and anemia was considered as: No public health problem if prevalence is below 4.9%, mild if prevalence is 5 - 19.9%, moderate if it is 20 - 39.9%, severe if it is more than 40%. Accordingly the present study shows that 43.1% anemia as severe among the subjects in study, and was considered a health problem in this area. The prevalence of anemia in the adolescents of Chagunarayan VDC, Bhaktapur was 43.1%. Studies done in other parts of Nepal showed higher prevalence than our study, Shah *et al*[2] 68.8%, Baral *et al*[3] 65.6%, Tiwari *et al*[4] 60.5%, Sinha *et al*[5] 56.3%.

Similiarly studies done in our neighboring country India also shows higher prevalence of anemia amongst adolescents. A study conducted by Chaturvedi *et al*[6] in rural Rajasthan, India among adolescents recorded 73.3% prevalence of anemia. Mehta[7] found out an anemia prevalence of 63.8% in urban slums of Bombay among 10 - 18 years adolescents. Raina *et al*[8] documented a prevalence of 85.3% in rural Haryana. Anemia was found to be more prevalent in adolescent males (54.1%) as compared to females (35.3%) in this study. Similar studies done by Baral *et al*[3] in Morang district of Nepal showed higher prevalence in females as compared to males. Studies done by Basu *et al*[9] in Chandigarh, India shows prevalence of anemia significantly higher amongst girls (23.9%) as compared to boys (7.7%).

In Nepal, intervention programmes in anemia are mostly female oriented because most of the data in our country shows female adolescents to be anemic. In Chagunarayan too supplementation of folic acid and iron tablets are done every year to female school children and males not having the advantage of it. So this may be the reason as the neglected male adolescents to have a higher prevalence rate. Tiwari *et al*[4] reported the prevalence of anemia in females to be 60.5% which was done in urban Kathmandu and was much higher than this study. In this study anemia was detected more in the age group of 13 and 15 years in males and females respectively. However it was followed by less in the age group of 16 - 19 years. Similar study done by Baral *et al*[3] shows higher prevalence in age group 10 - 14 years and improvement in the prevalence of anemia among adolescents with the increase in age in both males and females respectively.

In this study high prevalence of anemia was found more amongst rural adolescents as compared to semi-urban. These findings are similar to those described by Basu *et al*[9] and Vasanthi *et al*[10] but the findings reported by Baral *et al*[3] showed higher prevalence of anemia in urban adolescents.

Hemoglobin was examined before and after giving IEC for 3 times in each group at intervals of 1 month. The Mean Hb in males before IEC was 12.51 gm/dl and after IEC was 12.97 gm/dl. In females before IEC, it was 12.26 gm/dl and after IEC, Hb 12.81 gm/dl. Both male and female groups exhibited increase in hemoglobin significantly. The increase in range was -20 to 36% in males and -20 to 41.18% in females. The Mean increase in percentage was 5.16 in males and 4.48 in females. Fig 2 shows statistical significant positive correlation in all males and females. Both combined after IEC intervention in the adolescents also shows correlation. IEC was relatively more

appreciated by male students as their concentration towards food is more as compared to that of females in social life. The females are concentrating more for scholastic career. This needs further detail and long term follow up.

Recommendations

IEC regarding dark, green leafy vegetables and iron rich foods should be given to all school children every 6 months. Integrated community and school based approach would be the most appropriate strategy to reach adolescent population for prevention and control of anemia in Nepal and especially in mountainous area.

Teachers should be trained to identify the anemia in adolescent age groups so that early referral can be made to pediatrician in any Hospital or health sector which exists nearby.

Time to time IEC session should be held for the children of primary and secondary schools in view of present existing situation of anemia in adolescent age group in this developing country.

Acknowledgements

We would like to thank all the school children of Chagunarayan who took part in this study

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Level of Knowledge on Breast Feeding Among Primi Mothers in Western Regional Hospital, Kaski, With a View to Develop Information Booklet

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Keywords
Breast feeding, Primi mother, Information booklet.

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ABSTRACT

Background: Breastfeeding has been accepted as the most vital intervention for reducing infant mortality and ensuring optimal growth and development of children. It is the ideal form of feeding in the neonate. Artificial feeding exposes the infant to infection and resulting in over a million deaths annually worldwide due to its ill effects.

Objectives: This study was conducted to assess the level of knowledge on breast feeding and find out the association between level of knowledge and demographic variables among primi mothers in Western Regional Hospital, Kaski.

Methods: A descriptive study was conducted among 60 primi mothers admitted in postnatal ward of selected hospital in Kaski. Participants were selected through purposive sampling. Data was collected through interview by using socio-demographic proforma and structured breast feeding related questionnaire. After data collection, information booklet was distributed to each participant.

Results: The overall analysis of knowledge was mean and mean percentage [12.3(58.9%) with SD±2.15]. There was no significant association between level of knowledge among primi mothers and selected variables.

Conclusions: The study concluded that postnatal primi mothers had inadequate level of knowledge regarding breast feeding. There is a need to impart the knowledge and provide proper guidance on breastfeeding to prevent infant mortality.

INTRODUCTION

Breast milk is a complete food and it provides all the nutrients a baby needs during first 6 months of life. It contains a number of anti-infective substances and antibodies. Breast fed babies are less likely to suffer from allergic disorders like asthma and eczema. Breast feeding provides immunological benefits to the baby for the life time. This also provides emotional security. During breast feeding there is a release of oxytocin to eject the milk, which helps to contract the uterus. Breast feeding delays ovulation and onset of menstruation. Breast feeding is convenient and aids to maintain and regain the pre

pregnancy body weight. Mothers who breast feed their babies have a reduced risk of development of breast and ovarian cancer[1].

In Nepal, a report given by National Health Demographic Survey 2006 showed that the prevalence of the infants ever breastfed was 98.4% and exclusive breast feeding 53%. The newborns receiving breast feeding within one hour was 31.1% and feeding within 24 hours was 85%. The pre-lacteal feeding practice was 36%[2].

Nowadays, the women literacy rate has been increased and more women have started to work outside their home. This may reduce the prevalence of exclusive breast feeding practice. WHO recommends exclusive breast feeding for 6 months means that the infant receives only breast milk from her mother or expressed breast milk. There are 340,500 births per day in the world. Globally 450 neonates die in every hour[3].

According to the report of National Health Demographic Survey in 2006, Infants Mortality Rate (IMR) of Nepal was 48/1000 live births and 56/1000 live birth in the Western development region which is much higher than that of the developed world which stands at 8/1000 live births[2].

The second long term health plan of Government of Nepal (2017) had set a target to decrease neonatal mortality to 15/1000 live birth and infant mortality to 34.4/1000 live birth. As the breast feeding aid in the prevention of diseases like diarrhoea and respiratory diseases, it indirectly aid in the reduction of neonatal, infant and childhood mortality and improve health and development of infants and young children[4].

There are 340,500 births per day in the world. Globally 450 neonates die in an hour. India had the Infant Mortality Rate of 57/1000 live births (2005/2006) whereas Bangladesh had 65/1000 live births in the year 2004. According to the report of National Health Demographic Survey in 2006, Infants Mortality Rate (IMR) of Nepal was 48/1000 live births and 56/1000 live births in the western development region. The report of National Family Health Survey division (NFHS) shows the declining rate of IMR from 79 to 48 deaths per 1000 live births from the year of 1996 to 2006. The report also depicts that the infant mortality rate in Eastern development region was 45/1000 live birth, Central development region 52/1000 live births, Mid-Western region 97/1000 live birth and Far-Western region 74/1000 live births in the year 2006[2].

OBJECTIVES

- To assess the knowledge of primi mothers regarding breast feeding.
- To find out the association between knowledge of primi mothers with demographic variables.
- To prepare an information booklet regarding the enhancement of breast feeding.

METHODS

This was a descriptive study, using a quantitative approach performed at a selected hospital of Kaski, Pokhara. A non probability purposive sample of 60 primi postnatal mothers, immediate after delivery to 15 days of postnatal period and admitted in postnatal ward were selected. Two part structured questionnaire was developed to cover entire aspect of the study.

Section I: The study sample was assessed using the socio-demographic schedule includes age, religion, educational status of participants, educational status of participant's husband, occupation, type of family, place of residence and source of information questionnaire. Written consent was taken from every participant before data collection.

Section II: It is a 20 item questionnaire which enquires about knowledge on different aspects of breast feeding. It was intended to be a short, simple instrument. It was designed with the intention of capturing major dimension of the concept of anatomy and physiology, initiation, advantages, duration, diet, hygiene and contraindication of breast feeding. Each right response carries 1 mark and each wrong response carries zero mark (Total score 20).

Based on the percentage gained by the primi mothers, the knowledge of the respondents was arbitrarily categorized in the following groups :

- Inadequate : below 50%
- Moderately adequate : 50-75%
- Adequate : above 75%

Data pertaining to the study was collected through face to face interview with primi mothers admitted in postnatal ward with the help of trained enumerators. After data collection information booklet to enhance breast feeding was distributed to each participant. Descriptive and inferential statistics were used in order to analyze the data using SPSS.

RESULTS

Section I: Description of demographic characteristics of primi mothers

Fig 1: Distribution of respondents based on their age

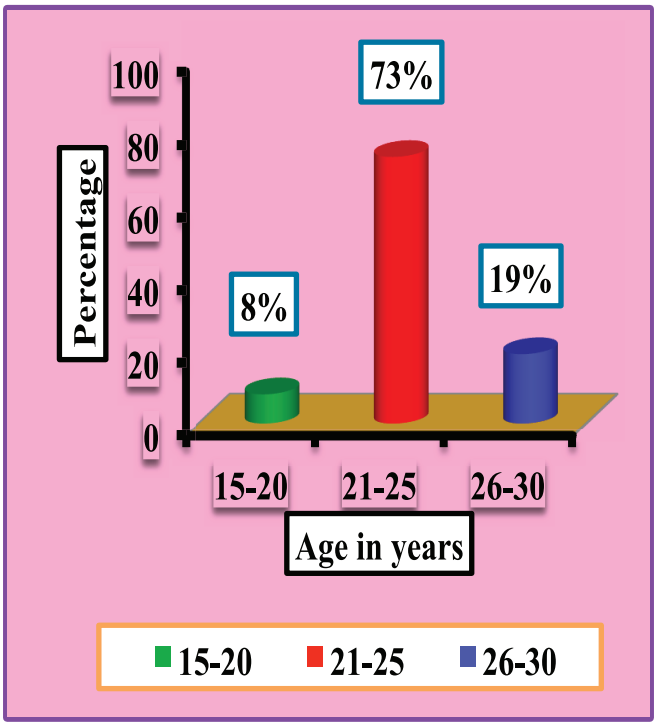

Figure 1 showed that majority of participants (73%) were belonged to age group of 21 - 25 years, 19% were belonged to age group 26 - 30 years, and remaining (8%) were belonged to age group of 15 - 20 years.

Fig 2: Distribution of respondents based on their religion

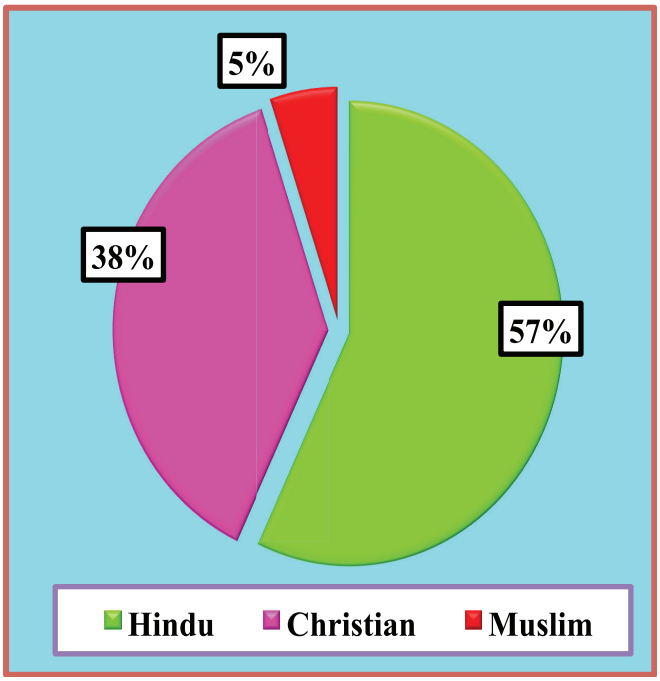

Figure 2 showed that majority of the participants (57%) were Hindus, 38% were Christians and remaining 5% were related to Muslim religion.

Fig 3: Distribution of respondents based on their educational status

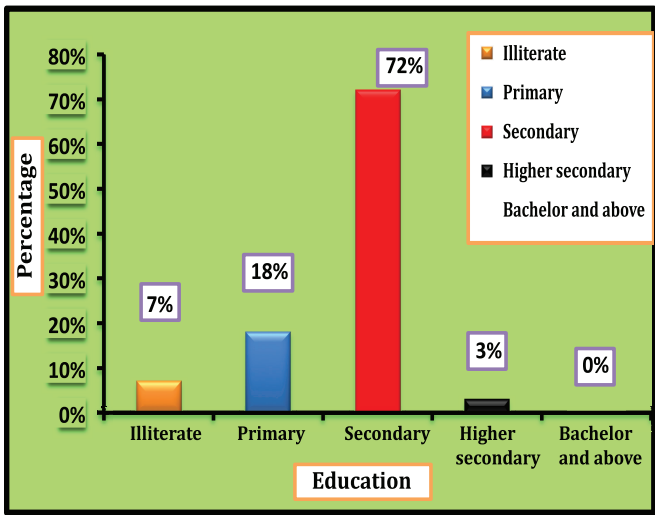

Figure 3 showed that Most of the participants (73%) had secondary level education, 18% had primary level education, only 3% had higher secondary level education, while7% participants were illiterate.

Table 1: Frequency and percentage distribution of socio-demographic characteristics **(**n=60)

S. No.	Variables	Frequency (f)	Percentage (%)
1	**Educational status of participant's husband**		
	a. Illiterate	2	3
	b. Primary	7	12
	c. secondary	49	82
	d. Higher secondary	2	3
	e. Bachelor and above	0	0
2	**Occupation**		
	a. Unemployment	58	97
	b. Government service	0	0
	c. Private	0	0
	d. Self employment	2	3
3	**Family income per month**		
	a. <10,000	7	12
	b. 10,001-20,000	53	88
	c. >20,001	0	00
4	**Family type**		
	a. Nuclear	48	80
	b. Joint	12	20
	c. Extended	0	0
5	**Place of residence**		
	a. Urban	8	13
	b. Rural	52	87
6	**Advice regarding ANC/ PNC**		
	a. Mother –in-law	0	0
	b. Friends	3	5
	c. Husband	0	0
	d. Health personnel	57	95

Table 1 showed that most of the participant's husbands

(82%) had secondary level education, 12% had primary level education, and only 3% had higher secondary level education, while 3% participants were illiterate. Majority of the participants (97%) were unemployed, and only 3% were self employed. The monthly income of maximum participants (88%) was Rs 10,001 - 20,000, and only 12% participants had less than 10,000. Most of the participants (80%) were belonged to nuclear family and only 12% joint family; among them majority (87%) were from the rural area and remaining (13%) were from the urban area.

Majority of the participants (95%) had received information from health personnel regarding antenatal care and postnatal care and only 5% had received information from their friends.

Section II: Distribution of knowledge level on breast feeding among primi mothers

Table 2: Distributions of knowledge level on breast feeding among primi mothers based on mean, median, mean percentage, and standard deviation (area wise) (n=60)

S. No	Area	No of items	Max. score	Mean	SD	Mean %
1	Anatomy & physiology	5	5	3.5	1.4	49.9
2	Initiation	2	2	1.0	0.7	75.8
3	Advantages	2	2	1.0	0.7	53.3
4	Duration	4	4	2.2	0.7	63.3
5	Diet	2	2	1.0	0.7	50.0
6	Hygiene	2	2	1.0	0.7	72.0
7	Contraindication	3	3	1.0	1.4	42.0
	Over all knowledge score	**20**	**15**	**12.3**	**2.25**	**58.9**

Table 2 shows that primi mother's level of knowledge on breast feeding based on various aspects. With regard to anatomy and physiology, mean 3.5 (34.50%) with SD±1.4, which was the highest score, where as initiation of feeding mean 1.0 (75.8%) with SD±0.7, advantages mean 1.0 (53.3%) with SD±0.7, diet for lactating mothers mean 1.0 (50.0%) with SD±0.7, importannce of hygiene mean 1.0 (72.0%) with SD±0.7, were similar lowest score. The duration of feeding mean 2.2 (63.3%) with SD±0.7, and with regard to contraindication of feeding mean 1.0 (42.0%) with SD±1.4.

The combined mean score of samples regarding the level of knowledge were 12.3 (58.9%) with SD±2.15, which indicates that primi mothers having moderately adequate knowledge regarding breast feeding.

Section III: Description of association between knowledge of primi mothers with demographic variables

Table 3: Association between knowledge of mother with demographic variables (n=60)

S. No	Demographic variables	Degree of freedom	Chi-square value χ^2	Table value	Level of significance	Inferences
1	Age of participants	1	2.3	3.84	0.05	NS
2	Religion	1	0.0075	3.84	0.05	NS
3	Education of the participants	1	2.97	3.84	0.05	NS
4	Education of the participant's husband	1	0.33	3.84	0.05	NS
5	Occupation of the participants	1	0.35	3.84	0.05	NS
6	Income per month	1	0.15	3.84	0.05	NS
7	Family type	1	0.01	3.84	0.05	NS
8	Place of residence	1	0.26	3.84	0.05	NS
9	Source of information	1	0.07	3.84	0.05	NS

NS: not significance at p 0.05 level

Table 3 shows that there is no significant association between level of knowledge among primi mothers and selected variables like; age (χ^2=2.3, p>0.05), religion (χ^2=0.0075, p>0.05), educational status of participants (χ^2=2.97, p>0.05), educational status of participant's husband (χ^2=0.33, p>0.05), occupation (χ^2=0.35, p>0.05), family per month income (χ^2=0.15, p>0.05), types of family (χ^2=0.01, p>0.05), place of residence (χ^2=0.26, p>0.05), sources of information (χ^2=2.3, p>0.05). Here χ^2 value is lesser than table value at 0.05 level of significance, so there is no association between demographic variables of participants with the knowledge level of participants.

DISCUSSION

Section I: Demographic characteristics of primi mothers

Percentage distribution of samples in reference to the age showed that 73% were in the age group 21 - 25 years, 19% were in the age group of 26 - 30 years and 8% were in the age group of 15 - 20 years. With reference to religion, the

percentage distribution of respondents showed that 57% were Hindus, 38% were Christians and 5% were Muslims. The findings were supported by Nandhini Subbach[5] study (85% were Hindus). Distribution of the respondents according to the educational status reveals that 93% of the respondents were literates and 7% were illiterates. Distribution of samples with regard to their husband educational status reveals that 97% were literates and 3% were illiterates. With regard to occupational status of samples showed that 97% were unemployed and 3% were self employed. Percentage distribution of participants with regard to family income per month reveals that 88% have income between Rs.10,001 - 20,000 and 12% of them have below 10,000. With reference to type of family reveals that 80% were from nuclear family and 20% were from joint family. Distribution of respondents according to their place of residence showed that 87% were from rural area and 13% were from urban area. With reference to source of information, percentage distribution of respondents reveals that 95% gained information from health personnel and 5% gained information from their friends.

Section II: Knowledge level of participants on breast feeding

Area wise analysis denotes that maximum (75.8%) mean percentage was in the area of initiation of breast feeding and the minimum (42%) was in the area of contraindication of breast feeding. The similar finding was supported by the findings of the study by Kronborg H, Vaeth M (2004), reveals that 98.7% had the knowledge on initiation of breast feeding[6].

The similar findings were supported by the findings of the study by Ramakrishna MN (1998), reveals that 97% of mothers had knowledge regarding the importance of breast milk[7].

The mean knowledge score obtained by the primi mothers was 12.3 (58.9) with standard deviation 2.25 and the knowledge score were in the range of 9-15.

Section III: Association between knowledge of mothers with demographic variables

No significant association was found between knowledge of primi mothers regarding breast feeding with demographic variables like age, religion, educational status of participant, educational status with participant's husband, occupation, family income per month, type of family, place of residence and source of information.

CONCLUSIONS

Assessment of the level of knowledge of primi mothers revealed that over all knowledge score is 12.3 (58.9%). This finding shows that primi mothers have inadequate knowledge regarding breast feeding. Area wise analysis of knowledge score was more (75.8%) in the area of initiation of breast feeding and least (42%) in the area of contraindication of breast feeding.

No significant association was found between knowledge of primi mother regarding breast feeding with demographic variables like age, religion, educational status of participants, educational status with participant's husband, occupation, family income per month, type of family, place of residence and source of information.

The study concluded that postnatal primi mothers had inadequate level of knowledge regarding breast feeding. There was a need to impart the knowledge and provide proper guidance on breast feeding to prevent infant mortality. Therefore researcher prepared the information booklet related to enhance breast feeding and distributed to each participants after data collection.

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Study of Factors Sustaining Tobacco Consuming Practices and Diseases in Residents of Dhankuta Municipality

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ABSTRACT

Background: Every year 15,000 deaths in Nepal are attributable to tobacco smoking and using other products of tobacco.

Objectives: The main objective of the study is to find out the association between factors initiating tobacco use among smokers and diseases in the last one year.

Methods: The cross-sectional study was conducted among residents of Dhankuta Municipality where 205 households were taken as subjects. Semi-structured questionnaire was administered to the participants and face to face interview was conducted. Chi-square test was applied to find out the association between factors initiating tobacco use among smokers and diseases in the last one year.

Results: The prevalence of diseases was significantly higher among those using drugs with tobacco (77.3%) and where the adult smokers (40.9%) were present in the family. The prevalence of diseases was found to be high among those consuming both smoking and chewing (30%), and taking cannabis with smoking (27.9%). The prevalence of diseases was significantly higher among those who were consuming tobacco inside the house (25.6%) than those not allowed to smoke inside the house (8.6%) ($P<0.05$).

Conclusions: Some social factors like presence of adult smoker and no restrictions of smoking in the house, and personal factors like taking illicit drug with smoking was significantly associated with diseases.

INTRODUCTION

Annually 4.9 million people in worldwide lose their lives as a result of tobacco consumption[1]. In the 20th century, 100 million people across the globe lost their lives due to consumption of tobacco[2]. Mathers and Loncar[3] estimated that deaths due to tobacco consumption are on the rise, from 5.4 million in 2005 to 6.4 million in 2015 and projected to be 8.3 million in 2030. Annually, tobacco is responsible for 1.4 million cancer deaths. Lung, oral, and nasopharyngeal cancers are some of the major cancers caused by tobacco consumption[1,4]. Chronic diseases due to cigarette smoking, elevated risk of cardiovascular disease, diabetes and respiratory diseases are also the consequences of tobacco use[5].

The World Health Organization (WHO) projected that there is an increasing trend of tobacco use in developing countries ranging from 4.9 million in 2000 to more than 10 million by 2020[1]. The South East Asia region of the WHO alone shares the burden of 90% of global smokeless tobacco (SLT) consumers[1,6].

Every year 15,000 deaths in Nepal are attributable to tobacco smoking and using other products of tobacco[7]. Based on the Nepal Demographic and Health Survey (NDHS, 2006) dataset, Sreeramareddy *et al*[8] reported that the prevalence of 'any tobacco use', 'tobacco smoking' and 'tobacco chewing' was 30.3%, 20.7% and 14.6%, respectively.

Therefore this study was designed to find out the association between factors initiating tobacco use among smokers and diseases in the last one year.

METHODS

The cross-sectional study was conducted from 1st July, 2014 to 30th April, 2015 among the residents of Dhankuta municipality of Nepal. Dhankuta is located in the Eastern geographical region of Nepal. This research was based on random selection of the study area, Dhankuta municipality. A National survey revealed that the prevalence of tobacco use was 33% (Khan S *et al* in India in 2013)[9], more than that 45% (Karki YB *et al* in Nepal in 2002)[10] and highest 52.07% (Zahiruddin QS *et al* in India in 2011)[11]. So taking lower value 33% of prevalence of tobacco use, sample size was calculated at 95% CI and 80% powers then it became 205 persons aged above 17 years. There are 9 wards in Dhankuta Municipality. Among 9 wards, 5 wards were randomly selected. The list of households of five selected wards was prepared and equal number of households (41) from each ward was selected on the basis of simple random sampling.

Ethical clearance was taken by Institutional Ethical Review Board of B P Koirala Institute of Health Sciences, Dharan, Nepal. Participants were first explained the purpose of study, its implications and assurance about the confidentiality of the information provided was given to the participants. Name of the individuals or participating group was not disclosed after the study.

Written permission was taken from concerned authority (Head of house) and the participants of the study. Those individuals who were available after three visits and willing to give written consents were included in the study. Pretested semi-structured questionnaire was administered to the study subjects in the presence of investigator and face to face interview was conducted.

The collected data was entered in MS Excel 2000. The quantitative data was analyzed using Statistical Package for the Social Sciences (SPSS) software package. The prevalence was calculated, Chi-square test was applied to find out the association between factors initiating tobacco use among smokers and diseases in the last one year. The probability of occurrence by chance is significant if P< 0.05 with 95% Confidence Interval.

RESULTS

Table 1: Association between personal factors initiating tobacco use among smokers and diseases in the last one year (n=117)

Characteristics	Suffered from diseases in last one year		Total	P-value
	Yes	No		
Age of start consuming tobacco (n=117)				
<15 years	14 (25.9)	40 (74.1)	54 (46.2)	
15-25 years	5 (11.6)	38 (88.4)	43 (36.8)	0.192
>25 years	5 (25.0)	15 (75.0)	20 (17.1)	
What form you started consuming tobacco at first (n=117)				
Smoking	11 (18.6)	48 (81.4)	59 (50.4)	
Chewing	7 (18.4)	31 (81.6)	38 (32.5)	0.514
Both	6 (30.0)	14 (70.0)	20 (17.1)	
Taking other substance with smoking (n=117)				
Alcohol	9 (19.6)	37 (80.4)	46 (39.3)	
Pan parag	3 (10.7)	25 (89.3)	28 (23.9)	0.211
Cannabis	12 (27.9)	31 (72.1)	43 (36.8)	
Illicit drug use (n=117)				
Yes	17 (77.3)	5 (22.7)	22 (18.8)	<0.001
No	7 (7.4)	88 (92.6)	95 (81.2)	
Total	**24 (20.5)**	**93 (79.5)**	**117**	

Among 205 study population, almost 117 (57.1) of respondents were found to be consuming tobacco. The prevalence of diseases was higher among those who were consuming both smoking and chewing (30%) than smoking (18.6%) and chewing (18.4%) but the difference was not significant. The prevalence of diseases was significantly higher among those who were using drugs with tobacco than those not using (P<<0.001) (Table1).

Table 2: Association between social factors initiating tobacco use among smokers and diseases in the last one year (n=117)

Characteristics	Suffered from diseases in last one year		Total	P-value
	Yes	No		
Why you started consume tobacco (n=117)				
Peer pressure	7 (10.6)	59 (89.4)	66 (56.4)	
Recreation	8 (27.6)	21 (72.4)	29 (24.8)	0.005
Adult smoker in family	9 (40.9)	13 (59.1)	22 (18.8)	
Parents smoke (n=117)				
Yes	24 (22.9)	81 (77.1)	105 (89.7)	0.063
No	0 (0.0)	12 (100.0)	12 (10.3)	
Friends smoking (n=117)				
Yes	22 (22.7)	75 (77.3)	97 (82.9)	0.329
No	2 (10.0)	18 (90.0)	20 (17.1)	
Household restrictions (n=117)				
No restrictions	21 (25.6)	61 (74.4)	82 (70.1)	
No smoking allowed inside the house	3 (8.6)	32 (91.4)	35 (29.9)	0.03
Total	**24 (20.5)**	**93 (79.5)**	**117**	

The prevalence of diseases was significantly higher where adult smokers were present in the family (P <0.05). The prevalence of diseases was significantly higher among those who were consuming tobacco inside the house than those not allowed to smoke inside the house (P <0.05). This is due to passive smoke effect on the health (Table 2).

DISCUSSION

Smoking and the use of tobacco is the single major cause of non-communicable diseases. Resulting in four million deaths a year, smoking has a great risk, socially and economically[12]. The study was reported that the proportion of people consuming any form of tobacco in Nepal was almost 29.8%[13].

This study shows that most of the smokers had started smoking before the age of 15 years. A study revealed that majority of the smokers (68.8%) had started smoking at the age of 11 to 20 years[14]. Pandey *et al* in Nepal reported that 93% of all daily smokers had started smoking before the age of 20 years. Among them 27% of smokers had started before the age of 15 years[15]. The fact that the effect of tobacco increased with age corroborates the findings of one of the earlier studies in India[16]. Another study showed that Nepal has high prevalence rate of health problems from 20 - 40% in persons above the age of 20 years; this was found to be significantly associated with tobacco smoking[14], but our study did not show any association between the age of starting tobacco use and health effect.

In our analysis it was found that cigarette smoking constituted more than half of the total tobacco consumption. It is greater than India where cigarette smoking constituted only 14% of the total tobacco use[17]. The Southern region of Nepal has close cultural relations with the Indians. Likewise, many Nepalese go to India in search of jobs for their livelihood. The situation of tobacco consumption in India and Nepal are, therefore, comparable and the use of tobacco products in India may influence Nepal[17].

This study showed that most of the people smoking with alcohol (39.3%) and cannabis (36.8%). Studies conducted by Orak *et al* indicated that the people were using alcohol with smoking (15%)[18], and Turkey (16.5%)[19] which are lower than our study. Tot *et al* carried out a study in Mersin, Turkey revealed that 5% of the people use cannabis[20], and lhan *et al* in Turkey, 5.9%[21] which are lower than our study. These substances have a high addiction potential and that they are illegal; we think that being exposed to these subtances even once still creates an important risk that should not be neglected. Another major point to be considered is that there may be an underestimation of the prevalence of substance use, because it is illegal in Nepal.

Most of the respondents started consuming tobacco due to peer pressure (56.4%) and recreation (24.8%). Other studies also showed most of the people started consuming tobacco because of peer pressure, and for entertainment[22,23]. Wang *et al* reported that peer influence proved to be the most significant and consistent predictor of smoking[24]. Having a smoker as the best friend increases significantly the probability of smoking[25].

This study showed that almost 89.7% of respondents' parents were smokers. Study conducted by Suryawanshi *et al* in India also reported that 4.7% of the parents were smoking[26], which is lower than our study. Another study showed that parental smoking is the main reason for the high tobacco users[27]. Similar study conducted by Uprety S *et al* showed family members of the smokers, smoke about 38%[28], Paudel S *et al* in Dharan, Nepal, 21%[14], which are lower than our study findings. This was also observed by

Leather dale ST *et al* where smoking is influenced when there is smoker in their family and surroundings.

CONCLUSIONS

Some social factors like presence of adult smoker in family and no restrictions of smoking in the house were significantly associated with health problems. The personal factor like taking illicit drug with smoking was also significantly associated with diseases. The prevalence of disease in some personal factors like respondents consuming both smoking and chewing, taking cannabis with smoking was found to be high but our study couldn't significantly associated with diseases. So, further strong study probably a case control study can make picture clearer.

Limitation of the study

Limitation of this study includes the cross sectional nature of data which precludes from drawing causal inferences. Tobacco use, specially smoking, is sometimes associated with social stigma. Therefore, some of the individuals may under-report their smoking habits.

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Conflicts of interest

The authors declare that they have no competing interests.

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Drug Resistant Tuberculosis : Molecular perspectives

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ABSTRACT

Drug-resistant tuberculosis is a growing threat to global public health. As a major factor, improper use of anti-TB drugs, either due to prescription errors or low patient compliance led to the widespread emergence of multi drug resistant tuberculosis (MDR-TB). More recently, severe forms of drug resistance called extensively drug-resistant (XDR-TB) have been described. Drug resistance in *Mycobacterium tuberculosis* arises from mutational changes in the bacterial chromosome at low frequency. A better knowledge of the molecular mechanisms of drug resistance of *M. tuberculosis* will improve the available techniques for rapid drug resistance detection and will help to explore new targets of drug activity and development.

The purpose of this review is to summarize our current understanding of the mechanisms of action anti-TB drugs and to explore the molecular basis of drug resistance in *M. tuberculosis*, highlighting the emerging areas of research.

INTRODUCTION

Tuberculosis (TB) remains as an important infectious disease and public health concern worldwide. According to WHO report, there were an estimated 9.0 million cases of TB in 2013 and 1.5 million deaths were attributed to this disease. More than half a million cases occurred in children and 360,000 deaths were reported among HIV-infected persons[1]. However, even more disturbing is the emergence of drug resistance. In 2013, there was an estimated 48,000 incident cases of multidrug resistant (MDR)-TB and approximately 210,000 deaths were due to it.

MDR-TB is caused by strains of *Mycobacterium tuberculosis* that are resistant to at least two first-line drugs, rifampicin and isoniazid. Since 2006, it has been recognized the presence of even more resistant strains of *Mycobacterium tuberculosis* known as extensively drug resistant (XDR)-TB. These strains in addition to being MDR are also resistant to any fluoroquinolone and to at least one of the three injectable second-line drugs: kanamycin, capreomycin or amikacin. More recently, a more worrying situation has emerged with the description of *Mycobacterium tuberculosis* strains that have been found resistant to all antibiotics that were available for testing, known as totally drug resistant: TDR-TB[2,3].

Early detection of drug resistance in TB is very essential to reduce and contain the spread of these resistant

strains. A better knowledge of the mechanisms of action of anti-TB drugs and the development of drug resistance will allow identifying new drug targets and better ways to detect drug resistance. We review the mode of action and resistance mechanisms of the main anti-TB drugs as well as newly approved drug in following sections.

First-Line anti-TB Drugs

Rifampicin

Rifampicin together with isoniazid has long been considered as the back bone of multidrug treatment regimen for TB. Rifampicin is active against growing and non-growing (slow metabolizing) bacilli[4]. Rifampicin is bactericidal antibiotic that acts by inhibiting the bacterial DNA-dependent RNA polymerase and targets the β-subunit of the RNA polymerase, physically by blocking the elongation of mRNA[5]. The majority of isolates of *Mycobacterium tuberculosis* resistant to rifampicin, harbor mutations in 81-bp spanning codons 507 – 533 of the *rpoB* gene, that codes for the synthesis of β-subunit of the RNA polymerase. As a result of this, conformational changes occur and decrease the affinity for the drug and results in the development of resistance[6]. This region is also known as the "hot-spot region" or "rifampicin resistance-determining region (RRDR)"[7]. Mutations in codons 516, 526 and 531 are the most commonly associated with rifampicin resistance. Other codons include 511, 518, 519, 522, 529 and codon 533[8,9]. Although less frequent, some reports have also noted the occurrence of mutations outside the RRDR of *rpoB*[10]. Cross-resistance with other rifamycins can occur. Mutations in some codons (e.g., 518 or 529) have been associated with low level resistance to rifampicin but still susceptible to other rifamycins, such as rifabutin or rifalazil[11]. This is important for TB patients that need to receive antiretroviral therapy since rifabutin is less effective inducer of the cytochrome P450 CYP3A oxidative enzyme[12]. On the other hand, monoresistance to rifampicin is quite rare and almost all rifampicin-resistant strains are also resistant to isoniazid. This is the reason why rifampicin resistance is considered as a surrogate marker for MDR-TB[13]. Recent genome sequencing studies have uncovered the acquisition of compensatory mutations in *rpoA* and *rpoC*, encoding the α and β subunits of RNA polymerase, in rifampicin resistant strains with mutations in rpoB[14], responsible for restoring the fitness of these strains *in vivo* and have also been associated with higher transmissibility in some settings[15,16].

Isoniazid

Isoniazid is another first-line very important drug for the treatment of TB. Unlike rifampicin, isoniazid is only active against metabolically-active replicating bacilli. Also known as isonicotinic acid hydrazide, isoniazid is a pro-drug that requires activation *in vivo* by the catalase/peroxidase enzyme KatG, encoded by *KatG* gene, to exert its effect[17]. Mutations in two genes *KatG* and *inhA* or its promoter region are most commonly associated with isoniazid resistance[18]. The most prevalent gene mutation has been identified as S315T in *KatG* resulting in an isoniazid product deficient in forming the isoniazid-NAD adduct needed to exert its antimicrobial activity[19]. This mutation has been consistently associated with high-level resistance (MIC >1 μg/mL) to isoniazid[20] and occurs more frequently in MDR strains[18]. The second most common mutation occurs in the promoter region of *inhA* causing an overexpression of *inhA* or less frequently, a mutation in its active site, which decreases its affinity for the isoniazid-NAD adduct[21]. The most prevalent mutation found is at position -15C/T and is more commonly associated with low level resistance to isoniazid (MIC <1 μg/mL). Mutations in *inhA* also cause resistance to the structurally related drug ethionamide, which shares the same target[22,23]. A recent study found that a mutation in the *inhA* regulatory region together with a mutation in the *inhA* coding region produced high-level isoniazid resistance and also cross-resistance to ethionamide[24]. One recent interesting finding showed that the 4R isomer of the isoniazid-NADP adduct causes inhibition of the dihydrofolate reductase (DfrA) in *M. tuberculosis*, suggesting that mutations in *dfrA* could possibly play a role in resistance to isoniazid[25]. Moreover, an analysis of the proteome of isoniazid targets in *M. tuberculosis* identified 16 other proteins, in addition to *inhA* and *dfrA*, that were bound by these adducts with high affinity, which could signal other not yet clearly defined actions of isoniazid on the bacteria[26]. Several studies have found single nucleotide polymorphism in other genes in isoniazid resistant clinical isolates of *M. tuberculosis*, including *kasA* and the *oxyR-ahpC* and *furA-katG* intergenic regions[18,27,28]. However, their direct role as a cause of isoniazid resistance has not been fully demonstrated. On the other hand, co-resistance to isoniazid and ethionamide has been clearly demonstrated to be caused by mutations in *ndh* in *M. smegmatis* and

M. bovis BCG, by altering the NADH/NAD ratios inside the cell, leading to a competitive inhibition of the INH-NAD adduct[29]. A recent study also found that a silent mutation in *mabA* conferred isoniazid resistance through upregulation of *inhA* in *M. tuberculosis*[30].

Pyrazinamide

Pyrazinamide constitutes now part of the standard first-line regimen to treat the disease. Pyrazinamide is an analog of nicotinamide and its introduction allowed reducing the length of the treatment to 6 months. It has the characteristic of inhibiting semi-dormant bacilli residing in acidic environments such as found in the TB lesions[39]. The pyrazinamide is similar to isoniazid in that it is a prodrug that requires activation to pyrazinoic acid, by the enzyme pyrazinamidase/nicotinamidase, a bacterial enzyme coded by 561 nucleotide *pncA* gene[40] before exerting its effects. The proposed mechanism of action of pyrazinamide involves conversion of pyrazinamide to pyrazinoic acid, which disrupts the bacterial membrane energetics inhibiting membrane transport. Pyrazinamide would enter the bacterial cell by passive diffusion and after conversion to pyrazinoic acid it is excreted by a weak efflux pump. Under acid conditions, the protonated pyrazinoic acid would be reabsorbed into the cell and accumulated inside, due to an inefficient efflux pump, resulting in cellular damage[41]. One study reported that pyrazinoic acid and its n-propyl ester can inhibit the fatty acid synthase type I in replicating *M. tuberculosis* bacilli[42]. Mutations in the gene *pncA* are the most common finding in pyrazinamide resistant strains. These mutations are scattered throughout the gene but most occur in 561-bp region in the open reading frame or in an 82-bp region of its putative promoter[43]. A recent survey of *pncA* mutations reported that single nucleotide substitutions were far by the most common, followed by multiple mutations and insertions/deletions. Some studies have reported the occurrence of pyrazinamide resistant strains without any mutations in *pncA* stating that the resistance could be due to mutations in another not yet identified regulatory gene[44]. Based on the current evidence, the contribution of mutations in *rpsA* to pyrazinamide resistance remains limited[45].

Ethambutol

Ethambutol is also a common first-line agent in the treatment of TB. Ethambutol is bacteriostatic against multiplying bacilli interfering with the biosynthesis of arabinogalactan in the cell wall by inhibiting arabinosyl transferase[31]. The inhibition of arabinosyl transferase prevents the formation of the mycolyl-arabinogalactan-peptidoglycan complex, which functions to increase cell wall permeability. In *M. tuberculosis*, the genes *embCAB*, organized as an operon, code for arabinosyl transferase, which is involved in the synthesis of arabinogalactan, producing the accumulation of the intermediate D-arabinofuranosyl-P-decaprenol[32]. The mutations in the gene *embCAB*, at position *embB306* is most prevalent according to most of the studies[33,34]. Some studies, however, have also found mutations in *embB306* in ethambutol susceptible isolates[35]. Moreover, a study with a large number of *M. tuberculosis* isolates found that mutations in *embB306* were not necessarily associated with resistance to ethambutol but with a predisposition to develop resistance to increasing number of drugs to be transmitted[36]. In fact, allelic exchange studies have shown that individual mutations causing certain amino acid substitutions produced ethambutol resistance, while other amino acid substitutions had little or no effect on ethambutol resistance[37]. More recently it has been reported that mutations in the decaprenylphosphoryl-β-D-arabinose (DPA) biosynthetic and utilization pathway genes, *Rv3806c* and *Rv3792,* together with mutations in *embB* and *embC* accumulate, giving rise to a range of MICs of ethambutol depending on mutation type and number[38]. There remain about 30% ethambutol resistant strains that do not present any mutation in *embB* stressing the need to identify other possible mechanisms of drug resistance to this drug.

Streptomycin

Streptomycin was the first antibiotic to be successfully used against TB. Unfortunately, resistance to it emerged, a result of being administered as monotherapy[46]. Streptomycin is an aminocyclitol glycoside active against actively growing bacilli and its mode of action is by inhibiting the initiation of the translation in the protein synthesis[47]. Streptomycin specifically acts at the level of the 30S subunit of the ribosome at the ribosomal protein S12 and 16S-rRNA coded by the genes *rpsL* and *rrs*, respectively[48]. Consequently, mutations in *rpsL* and *rrs* are the major mechanisms of resistance to streptomycin but account for 60% - 70% of the resistance found[49]. Among the mutations reported in *rpsL,* a substitution in codon 43 from lysine to arginine has been the most commonly reported, which produces high-level resistance to streptomycin. In *rrs* the most common mutations occur around nucleotides 530 and

915. There remain an important percentage of strains resistant to streptomycin that lack mutations in either of these two genes, suggesting additional mechanisms of resistance. It has also been reported that mutations in *gidB*, a gene encoding a convserved 7-methylguanosine methyltransferase specific for the 16S rRNA, confers low-level resistance to streptomycin[50].

Second-Line anti-TB Drugs

Ethionamide

Ethionamide is a derivative of isonicotinic acid structurally similar to isoniazid. It is also a pro-drug requiring activation by a monooxygenase encoded by the *ethA* gene. It interferes with the mycolic acid synthesis by forming an adduct with NAD that inhibits enoyl-ACP reductase enzyme. *EthA* is regulated by the transcriptional repressor *EthR*[66]. Resistance to ethionamide occurs by mutations in etaA/ethA, ethR and also mutations in inhA, which cause resistance to both isoniazid and ethionamide[67]. Studies with spontaneous isoniazid- and ethionamide-resistant mutants of *M.tuberculosis* found that an enzyme encoded by *mshA*is essential for mycothiol biosynthesis[68].

Fluoroquinolones

Fluoroquinolones are currently in use as second-line drugs in the treatment of MDR-TB. Both ciprofloxacin and ofloxacin are synthetic derivatives of the parent compound nalidixic acid, discovered as a by-product of the antimalarial chloroquine. Newer-generation quinolones such as moxifloxacin and gatifloxacin are being evaluated in clinical trials and proposed as first-line antibiotics with the purpose shortening the length of treatment in TB[51,52]. The mode of action of fluoroquinolones is by inhibiting the topoisomerase II (DNA gyrase) and topoisomerase IV, encoded by the genes *gyrA, gyrB, parC* and *parE*, respectively. In *M. tuberculosis*, only type II topoisomerase (DNA gyrase) is present and, thus, is the only target of fluoroquinolone activity. Type II topoisomerase is a tetramer formed by two α and β subunits, coded by *gyrA* and *gyrB*, respectively, which catalyzes the supercoiling of DNA[53]. The development of fluoroquinolone resistance in *M. tuberculosis*is by chromosomal mutations in the quinolone resistance-determining region of *gyrA* or *gyrB*. Most frequently mutations are found at position 90 and 94 *gyrA* but mutations at position 74, 88 and 91 have also been reported[54,55]. Cross resistance is assumed to occur between fluoroquinolones although isolated reports have acknowledged the presence of strains resistant to gatifloxacin and moxifloxacin that were still susceptible to ofloxacin[56]. Also, the involvement of efflux mechanisms has been suggested as a possible cause for fluoroquinolone resistance in *M. tuberculosis*[57].

Kanamycin, Capreomycin, Amikacin, Viomycin

All these four antibiotics have the same mechanism of action by inhibiting the protein synthesis but, while kanamycin and amikacin are aminoglycosides, capreomycin and viomycin are cyclic peptide antibiotics, used in the management of MDR-TB as second-line drugs. Kanamycin and amikacin inhibit protein synthesis by alteration at the level of 16S rRNA. The most common mutations found in kanamycin-resistant strains are at position 1400 and 1401 of the *rrs* gene, conferring a high-level resistance to kanamycin and amikacin. However, mutations at position 1483 have also been reported[58, 59]. Full cross-resistance between kanamycin and amikacin is not complete. Some studies have shown variable levels and patterns of resistance suggesting that other mechanisms of reistance might be possible[60]. In concordance with this, a low-level resistance to kanamycin has been associated with mutations in the promoter region of the *eis* gene, encoding an aminoglycoside acetyl transferase[61]. Mutations at position -10 and -35 of the *eis* promoter led to an overexpression of the protein and low-level resistance to kanamycin but not to amikacin. These mutations were found in up to 80% of clinical isolates showing low-level resistance to kanamycin[61]. Capreomycin and viomycin, on the other hand, have a similar structure and bind at the same site in the ribosome, at the interface of the small and large subunits[62]. They show full cross-resistance[63]. Mutations in the *tlyA* gene have also been associated with resistance to capreomycin and viomycin. *TlyA* is an rRNA methyltransferase specific for 2'-O-methylation of ribose in rRNA. Mutations in *tlyA* determine the absence of methylation activity[64]. Although some studies did not find this association, a recent meta-analysis, evaluating the association of genetic mutations and resistance to second-line drugs, has confirmed the presence of *tlyA* mutations in addition to mutations in *rrs* and *eis*[65].

Para-Amino Salicylic Acid (PAS)

Although it was one of the first anti-TB drugs used in the treatment of the disease, together with isoniazid and streptomycin, PAS is now considered as a second-line drug part of the treatment regimen for MDR-TB. Until recently, its mechanism of action was not completely defined. It has been proposed that being an analog of para-amino

benzoic acid, it must compete with it for dihydropteroate synthase, interfering in the process of folate synthesis. A study using transposon mutagenesis identified mutations in the *thyA* gene associtated with resistance to PAS that were also present in clinical isolates resistant to PAS[69]. A recent study has also identified various missense mutations in *folC* encoding dihydrofolatesynthase that conferred resistance to PAS in laboratory isolates of *M. tuberculosis*[70] in a panel of 85 clinical MDR-TB isolates, mutations in *folC* were identified in 5 isolates resistant to PAS. Nevertheless, just less than 40% of PAS-resistant strains had mutations in *thyA* indicating that still other mechanisms of resistance to the drug might exist[71].

Cycloserine

Cycloserine is an oral bacteriostatic second-line anti-TB drug used in MDR-TB treatment regimens. It is an analog of D-alanine that by blocking the activity of D-alanine: D-alanine ligase inhibits the synthesis of peptidoglycan. It can also inhibit D-alanine racemase (*AlrA*) needed for the conversion of L-alanine to D-alanine[72]. Although the actual target of cycloserine in *M. tuberculosis* is not completely elucidated, in previous studies in *M. smegmatis* it was shown that overexpression of *alrA* led to resistance to cycloserine in recombinant mutants[73]. More recently, it has also been shown that a point mutation in *cycA*, which encodes a D-alanine transporter, was partially responsible for resistance to cycloserine in *M. bovis* BCG[74].

Thioacetazone

Thioacetazone is an old drug that was used in the treatment of TB due to its favorable *in vitro* activity against *M. tuberculosis* and its very low cost. It has toxicity problems, however, especially in patients co-infected with HIV. It belongs to the group 5 drugs of the WHO and acts by inhibiting mycolic acid synthesis[75]. A recent study reported mutations at codon 101 of *mmaA4*, at codon 61 of *hadA* and at codon 85, 157 and 123 of *hadC* genes associated with resistance to thioacetazone. Moreover, the study showed that overexpression of *hadAB, hadBC or hadABC* in *M. tuberculosis* led to high level resistance to thioacetazone[75].

Macrolides

Macrolides are more frequently recommended for the treatment of other mycobacterial infections due to their limited activity against *M. tuberculosis*. Among them clarithromycin is considered as part of the group 5 drugs of the WHO. Intrinsic resistance to macrolides has been attributed to low cell wall permeability and the expression of *emr37,* a gene that codifies for a methylase at a specific site in the 23s rRNA, blocking the binding of the antibiotic. In studies performed with *M. tuberculosis* and *Mycobacterium microti* it was found that this intrinsic resistance was inducible with sub-inhibitory concentrations of clarithromycin, leading to four- to eight-fold increase in MIC values[76]. Moreover, in studies performed with clinical isolates of *M. tuberculosis,* subinhibitory concentrations of ethambutol reversed resistance to clarithromycin, signaling a permeability barrier as the cause of the intrinsic resistance to the macrolide[77].

Clofazimine

Clofazimine is a riminophenazine compound reported long ago as having anti-TB activity[78]. Due to the availability of other effective anti-TB drugs at the time and some undesirable side-effects, such as pigmentation of the skin, its use was more limited to the treatment of leprosy[79]. It is now considered in the group 5 drugs of the WHO for the management of MDR-TB. Recent studies, however, have signaled the outer membrane as the possible target of clofazimine[80]. Another study found that in *M. tuberculosis*clofazimine is reduced by NADH dehydrogenase and subsequently after spontaneous reoxidation liberates bactericidal levels of reactive oxygen species[81]. Resistance to clofazimine has not yet been fully characterized; however, a recent study has found that in spontaneous mutants of the reference strain H37Rv, mutations in the transcriptional regulator Rv0678 caused an upregulation of MmpL5, a multisubstrate efflux pump, which not only caused resistance to clofazimine but also to bedaquiline[82].

Linezolid

Linezolid is an oxazolidinone originally approved for clinical use in the treatment of skin infections and nosocomial pneumonia caused by Gram positive bactera[83], also part of the category 5 drugs of second-line anti-TB drugs. The mode of action of linezolid is by inhibition of an early step in the synthesis of proteins, binding to the 50S ribosomal subunit[72]. Resistance to linezolid in *M. tuberculosis* is still unusual, but a study analyzing 210 MDR strains found 1.9% of strains being resistant to linezolid[84]. Further analysis of *in vitro* selected linezolid-resistant mutants found that strains with mutations in the 23S-rRNA had MICs of 16-32 μg/mL, while strains

with MICs of 4 - 8 µg/ml or susceptible strains showed no mutations[85]. A more recent study using next-generation sequencing has also found the mutation T460C in *rplC*, encoding the 50S ribosomal L3 protein, in *in vitro*-selected mutants and clinical isolates of *M. tuberculosis* resistant to linezolid[86]. Previous studies have also found evidence of the possible involvement of efflux pumps in the resistance of *M. tuberculosis*to linezolid[57].

New anti-TB Drugs

There are several anti-TB drugs in the process of development and some of them are already being evaluated in clinical trials and in new combinations with the purpose of reducing the length of TB treatment. The new anti-TB agents bedaquiline, delamanid, and PA-824 have been shown to have potential to enhance our capacity to cure XDR-TB. Recently, bedaquiline is approved by the USA FDA for treatment of MDR-TB after 40 years of the last approval of rifampicin.

Bedaquiline

Formerly known as TMC207 or R207910, bedaquiline is a diarylquinolone with specific activity against *M. tuberculosis*, which has also shown *in vitro* activity against other non-tuberculous mycobacteria[87]. Bedaquiline was discovered after a high-throughput evaluation of thousands of compounds using *Mycobacterium smegmatis* in a whole-cell assay[88]. The drug showed *in vitro* and *in vivo* activity against *M. tuberculosis* and then entered into clinical evaluation for drug susceptible and MDR-TB[52,89,90]. Based on the results of two phase II clinical trials, bedaquiline has recently received conditional approval for the treatment of MDR-TB under the trade name Sirturo. A "black box' warning is, however, accompanying this authorization due to the reported unexplained deaths and QT interval prolongation. Recent reviews and evaluation of this new drug have been published[91,92]. Bedaquiline is also being evaluated in new combination regimens with the purpose of reducing the length of treatment[93]. The mode of action of bedaquiline is by inhibiting the ATP synthase of *M. tuberculosis,* which wasa completely new target of action for an anitimycobacterial drug, discovered by analyzing *M. tuberculosis* and *M. smegmatis* mutants resistant to bedaquiline. By sequencing the genome of these mutants and comparing to that of the susceptible strains, the only mutation found was in the *atpE* gene, which encodes the c part of the F0 subunit of the ATP synthase[88]. This is a complex structure that generates the ATP needed by the mycobacterial cell[94] for which bedaquiline has a favored specificity compared to mitochondrial ATP synthase[95]. The most prevalent mutation in the *atpE* gene found in bedaquiline resistant mutants is A63P but also I66M has been found. The latter introduces a modification that interferes the proper binding of bedaquiline to its target[96,97].Nevertheless, in a study to further assess the mechanisms of resistance to bedaquiline in *M. tuberculosis,* it was found that only 15 out of 53 resistant mutants had mutations in *atpE*. The other 38 strains lacked mutations in *atpE*or even in the F0 or F1 operons, which suggests that other mechanisms of resistance are still possible[98]. Bedaquiline resistance in clinical isolates has not been reported till date.

CONCLUSIONS

Drug resistance in *M. tuberculosis* emerges as a result of spontaneous chromosomal mutations that render the bacteria resistant to the most commonly used anti-TB drugs. Among the reasons for this, the non-compliance with the treatment regimens is signaled as the first cause.The standard treatment of TB calls for a six-month regimen of four drugs that in the case of MDR-TB is extended to 18-24 months involving second-line drugs. This makes compliance with the treatment regimens very challenging and the rates of nonadherence could be high, resulting in poor outcomes and further dissemination of MDR strains. Thus drug resistance in TB remains a man-made phenomenon.

Though mutations in several genes are clearly associated with drug resistance in *M. tuberculosis,* there are still many cases where resistant strains do not harbor any known mutation. A recent study using whole-genome sequencing identified new genes and intergenic regions that were associated with drug resistance and its evolution, showing that TB drug resistance is a phenomenon more complex than previously assumed[99]. More clarification is needed on the role of specific gene mutations and the development of MDR- or XDR-TB or the relation between drug resistance and fitness of the bacteria. A better knowledge is also required on the role of efflux pump mechanisms and the development of clinical drug resistance, or the role of porins, if any, on the intrinsic resistance to certain antibiotics. It is thus, quite important to further our knowledge of additional mechanisms of drug resistance to the available anti-TB drugs. This could have a major impact on the dynamics of TB transmission and for the discovery and development

of new anti-TB drugs. As new treatment regimens containing new drugs are implemented, we will have to establish the spectrum of epidemiologically relevant mutations as soon as possible.

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Does Practical Demonstration Class Help Medical Students to Develop Their Skill Domain?

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ABSTRACT

Background: Practical demonstration by teachers has been practiced in the Institute because of difficulty in handling 50 students in a particular lab exercise (Labex) which has been questioned by many teachers and students.

Objectives: To evaluate the efficiency of practical demonstration class at BP Koirala Institute of Health Sciences (BPKIHS), Dharan, Nepal.

Methods: It was a questionnaire based study administered to MBBS students of first and second year (n=63) and basic medical science (BMS) teachers (n=24).

Results: Most of the students (57/63) and all the teachers (24/24) were satisfied with the present method of practical class (i.e. demonstration by the teachers). They believed that practical demonstration was very helpful to students for developing their practical skills. However, most of the students (53/63) and teachers (17/24) felt and suggested that practical class should be of demonstration by teachers followed by the practical work done by the students, others (students= 8/63 and teachers= 3/24) suggested that it should be briefed by teachers and done by the students followed by the feedback from the teachers. Almost all the teachers (21/24) responded that they clarify queries raised by the students in the labex. However, students mention that queries were clarified in few practical classes of Basic Medical Science disciplines like in anatomy, biochemistry and microbiology. Both the students (63/63) and the teachers (22/24) accepted that question repetition at BPKIHS is a common phenomenon. They have encountered mistakes in the previous questions. Almost all the teachers (20/24) and students (51/63) agreed to undergo training about the questions and exam pattern at BPKIHS.

Conclusions: Students' skill domain could be improved by demonstration of practical by the teachers followed by the maximum participation of the students in the practical procedures. To improve and to gain students' excellence, it seems to improve, review, update and minimize the maximum repetition of old questions in the practical examinations.

INTRODUCTION

Many attempts are made by the teachers and the administration to enrich the students learning environment. It is most accepted and believed that the combination of didactic lecture, practical demonstration by faculty and performing the experiments by each student will enrich the students learning environment. As well as, practical demonstration and performance in experiments by the students will help them in developing and understanding the concepts. It also builds their attitude towards learning.

Across the globe many teaching learning methods are adopted. However, by time to time it is necessary to monitor that the applied method is working well or not. The ways to improve any adopted method helps to minimize the flaws of the method and assist the students learning environment too[1]. Written examinations (essays and multiple choices) test cognitive knowledge, which is only one aspect of the competency[2]. Practical examination tests the skill domain of the students. Students develop skills in the practical class. On observing lab exercise (Labex) at BPKIHS, we found many students have least interest on practical performance or in the technical aspect of the practical. They had a list of old objective structured practical examination (OSPE) questions that they were willing to ask at the end of the lab demonstration. They do attend the labex but for solving the old questions, and for the sake of performing well in the exam. It was good that they were willing to solve the queries but it seemed that they were not interested on the skills and technical aspect of the practical. In addition, while performing duties in the examination and during correction of answer sheets, we encountered many questions that lack desirable characteristics of the evaluation tools. Therefore, this study was aimed to solve some of the problems faced by the students and teachers during the lab demonstration. In addition, we performed this study to search perception of students and teachers on repetition of OSPE questions in the practical examination. As well as, the other objective of the study was to document ways to improve the practical class in the Institute.

METHODS

It was a cross sectional descriptive study. The pre-designed and pretested structured questionnaires on practical demonstration classes were administered to MBBS students (n=63) and basic medical science teachers (n=24). Basic medical science disciplines included were anatomy, biochemistry, microbiology, pathology, pharmacology and physiology. Ethical clearance was taken from the Institute ethical review board at BPKIHS. Data were expressed in proportions or frequencies. The questionnaires designed for the teachers and students included both the open and closed ended questions**.** We also studied students and teachers perception on OSPE questions repetition in the practical examination via the pretested questionnaire. They were also asked to provide suggestion for improving the present form of the practical examination i.e. objective structured practical examination (OSPE).

Some sample questions present in the questionnaire were as follows:

1. A practical class should be (Encircle one)
   a. demonstration by teachers only
   b. demonstration followed by practical work by students
   c. practical work by students followed by assessment
   d. briefing followed by practical work by students and feedback from the teachers
   e. Other any, specify….
2. How were practical demonstrations? (Please encircle one)
   Very helpful Helpful somewhat
   Helpful Not Helpful
3. Do you have collection of old questions asked to your senior students?
4. Is question repetition a common phenomenon at BPKIHS?
5. Is repeating old questions justifiable for a future medical graduate?
6. Have you ever encountered mistakes in the previous questions?
7. What sorts of mistakes have you encountered? (Please tick one)
   - Entire concept/theme ☐
   - Main head line ☐
   - Marks distribution ☐
   - Focused on certain elements ☐
   - Repetition of questions ☐
   - Any other, specify…….
8. How do you rate the BPKIHS exam pattern to student achievement?

Small group discussion for ways to improve the practical class in the institute

After finding disagreement between students and teachers about a few questions in the questionnaire, we attempted to find the ways to improve the practical class in the Institute. For this, we again created the environment for the discussion among teachers and students. Small group discussion was organized to arrive at an agreement between them. Finally, the suggested outcomes were

included in the study.

Statistical analysis

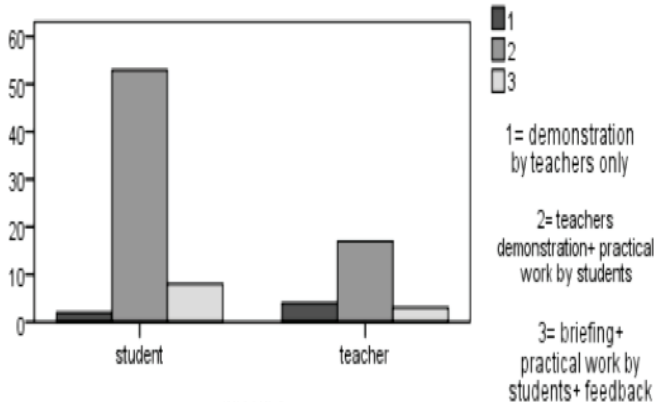
In dichotomized data, e.g. the response of teacher and student, chi-square tests was used. Whereas, for the open-ended responses (textual data): the responses were classified under theme- and concept-wise and qualitative weight was given consulting concerned experts. Based on the majority of teachers and students suggestions, we compared responses from the students with the teachers to get the conclusions.

RESULTS

A. Practical class scenario at BPKIHS

We analysed answers from the teachers and students and divided answers into theme- and concept-wise and presented them as follows. On the present scenario, both teachers (24/24) and students (57/63) believed that practical demonstrations were very helpful to students for developing their practical skills. Although most of the students (57/63) and all the teachers (24/24) were satisfied with the lab exercise (Labex) but both gave mixed response on the practical class. Most of the students (53/63) and teachers (17/24) felt and suggested that practical should be demonstration by teachers' followed by practical work by the students, and some of them (students= 8/63 and teachers= 3/24) suggested practical work (Labex) should be briefed and done by the students followed by a feedback (Fig 1).

Fig 1: Students and teachers response on type of practical class



Almost all the teachers (21/24) responded that they clarify queries raised by the students in the Labex. However, students mention that queries were clarified in a few practical classes of basic medical science disciplines (BMSD) like in anatomy, biochemistry, and microbiology. However, in other departments like in physiology, pathology and pharmacology disciplines the clarification of queries perceived by the students was less (Table 1).

Table 1: Students perception on queries clarification in BMSD practical classes

Subjects (BMS)	Perception of students on clarification of queries during practical classes (n= 63)		
	Yes	Partial clarification	No
Anatomy	38	25	0
Biochemistry	33	27	2
Physiology	19	41	3
Pharmacology	26	26	11
Pathology	24	35	4
Microbiology	31	29	3

Regarding training on the questions and exam pattern, overwhelming majority of teachers (20/24) agreed to provide training and majority of students (51/63) wanted to undergo the training.

B. Perception of students and teachers on repetition of old questions in the practical examination

1. Repetition of the questions in the examination by basic science disciplines

All the students (63/63) and almost all the teachers (22/24) accept that question repetition at BPKIHS is a common phenomenon. Both (students=32/63 and teachers=16/24) of them have encountered mistakes in the previous questions. According to students' experience, among basic sciences more repetition of old questions were in Pathology (42/63), Biochemistry (19/63), Anatomy (16/63) and Pharmacology (15/63) whereas in Microbiology (8/63) and Physiology (3/63) it was less.

Interestingly, it was found that students experience those subjects easy to study and pass the examination which had more repetition of old questions in exams like in anatomy 15/63, biochemistry 16/63, and pathology 10/63.

2. Benefits to students on repetition of the questions

Majority of students (58/63) responded that they were benefited by reviewing the previous questions and appearing in the examination. They mentioned that they were benefited by reviewing old questions because it helped them to secure high marks easily as maximum questions were repeated. They also mentioned that it helped them to obtain pass marks at least by selective reading from important topic in a limited time.

It helped them to cover whole syllabus by revising past questions too. In addition they mentioned that by knowing and observing the old questions helped them to know about the questions and exam and question marking pattern. It helped them to build confidence, cover must known areas, study well. Moreover, some students mentioned that viewing old questions inspired them to know many new things by seeing questions that are tough and it stimulated them to study in depth. However, some students mentioned that it did not benefit them because it forced them to go for the selective study. Which led them to miss out lots of basic concepts, limiting not only their knowledge but also their competence and caliber were diminished. They also mentioned that it decreased their confidence to face difficult situations or questions because of the selective study.

C. Small group discussion for ways to improve the practical class in the Institute

In small group discussion, students and teachers suggested that practical should be demonstration by teachers followed by practical work by the students and feedback by the teachers at the end. There should be time to time revision class for the practical skill development, monitored by the teachers. There should be training about the questions and exam pattern to both the teachers and the students. To minimize the mistakes in the questions they suggested timely reviewing of the old questions. Teachers agreed to improve, review, update and minimize the maximum repetition of old questions in the OSPE exam.

The practical examination in the Institute (BPKIHS) is based on the objective structured practical examination (OSPE). Each OSPE station is of 5 min duration. Sets of practical questions were displayed in each OSPE station. Hence students have to answer within 5 min in each OSPE station and they have to shift from one station to another and complete their task by writing answers to the questions asked in the station. Therefore, teachers mentioned that practical class should be designed or conducted in such a way that it will help students to perform the OSPE station well. In addition to this, there should be improvement in the practical examination system too. To test the skill domain, students should show the practical procedure by doing themselves. Therefore, doing the procedure will actually test the skill domain of the students. Hence, teachers suggested adding some procedural stations along with the present style of OSPE stations will improve the present practical examination system. Teachers also mentioned that they have to improve the practical class by involving students in the practical procedures. Teachers were also agreed for organizing revision class at the end of the particular organ system or block.

DISCUSSION

Examination is considered a good tool to assess students' performance on their expression of knowledge, skills or abilities. A single examination does not fulfill all the functions of assessment[3]. Among different types of examination tools, practical examination helps to judge the skill and techniques of the students and practical class is the way to train and help them to enrich their knowledge.

Therefore, this study was undertaken to observe some of the problems faced by the students and the teachers during the labex. In addition, this study was done to find ways to improve the practical class, ultimately helping students to improve their skill or psychomotor domain.

We found that there are many ways to improve the practical class at BPKIHS. Both teachers and students have suggested that there should be maximum participation by students during the laboratory procedures. There are reports that students working together in the lab helped them to develop their concept of team work, encouraged students' capabilities when seeking, acquiring, and processing the knowledge[4]. Acquiring skills help student to be a better communicator, analyst, professional and citizen[5]. Therefore, by making students to perform in group during the practical class will help them to develop team work quality, skills as well as enhance their learning ability too.

In our study, both the study groups after small group discussion suggested that doing the practical procedure will actually test the skill domain of the students. It has been mentioned that OSPE has scope for being structured in such a way that all the objectives of laboratory teaching can be tested and each aspect can be assigned the desired weightage[6]. Although there are reports showing OSPE as a better method for evaluation of practical skills and gain more students' satisfaction as compared to traditional examination[7]**.** Teachers at BPKIHS suggested adding some procedural station in between the OSPE station. In a study done in 42 Malaysian pharmacy students, OSCE (objective structured clinical examination) was perceived to be more stressful than other methods of assessment (32%), yet only 10% thought it was unfair[8]**,** whereas few countries have considered it as a gold standard tool for assessing the student skills[9]**.** In this context, adopting OSPE and adding few procedural stations will help to assess the skill domain of the students very well. Along with the improvement in

the practical class demonstration and doing practical by students will help to meet the requirement in performing well in the practical examination. Finally, it will also help to minimize the students' stress and will be most satisfying tool to assess students' psychomotor and attitude domains too.

Our study found that question repetition has to be sorted out at BPKIHS. Interestingly, it was found that students experience those subjects easy to study and pass the examination which had more repetition of old questions in the examination. However, they have stressed that practicing old questions made them deficit in knowledge due to selective study habits. It has been found that both learning outcome and perceived course quality were enhanced by the increased frequency of examinations, possibly by promoting consistent student study habits[10]. As frequency of examination is high at BPKIHS, it is promoting students learning. However, if practical class is taken effectively and question reviewing, updating is done regularly, then it will certainly improve the learning environment of the students. Therefore, it seemed that reviewing old questions will be helpful to create a rich question bank that will help in emergency situations i.e. when there is lack of time for preparing questions. But, it should be regularly updated by the teachers. In addition, reviewing and updating old questions is necessary for the improvement of student performance and to provide justice to students stress tolerated by them during examination time. Preparing standard questions consumes lots of time and preparing them in hurry leads to lots of mistakes that leads to poor performance. Some of the oversights we encountered were like highly ambiguous questions, inappropriate marks distribution, focused on certain elements, repetition of questions, questions not within the time format of the examination etc. Therefore, it seems that it is the right time to start the review and update of the old questions. This study probably disclosed the hidden flaws and helps teachers to take an effective labex and make students to participate actively in the practical classes. In addition, in future the chance of having repetition and mistakes in the questions will be least.

CONCLUSIONS

For the effective improvement on students' skill domain, practical class should not be limited on demonstration by teachers only but it should be practiced by making students maximum participation on practical procedures. To improve and to gain students' excellence, it seems to improve, review, update and minimize the maximum repetition of old questions in the OSPE. Repetition of old questions helped students to pass the examination but it forced them to go for the selective study.

Limitations of the study

The study would have been more conclusive, if it had focused and included responses from students who were in clinical years and from interns too.

Conflict of Interest

Nil

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An Unusual Case of Sexual Assault on an Infant: A Perineal Laceration in a Seven Month Old Girl

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Keywords
Anogenital injuries, Child maltreatment syndrome, Child sexual abuse.

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ABSTRACT

Child sexual abuse is a very common form of sexual offence in younger age groups. It also falls under child maltreatment syndrome. Here, we report a case of a seven month-old baby girl with gross anogenital injuries. On anogenital examination of the infant baby girl, a skin deep laceration measuring 0.6 x 0.2 cm was present over the posterior fourchette. Along with that, redness was noted over inner aspects of bilateral labia minorae and majorae. Hymen was intact.

INTRODUCTION

Sexual abuse occurs when a child is engaged in sexual activities that the child cannot comprehend, for which the child is developmentally unprepared and cannot give consent, and/or that violate the law or social taboos of society. The sexual activities may include all forms of oral-genital, genital, or anal contact by or to the child, or non-touching abuses, such as exhibitionism, voyeurism, or using the child in the production of pornography[1].

Amongst all the victims of sexual abuse, children are the commonest. In such cases where children are the victims, usually there are few or no signs of general violence as the child usually is unable to comprehend the act and is also unable to offer resistance. Narrowing down the child sexual abuse victim to girls, hymen is deeply seated and vagina is small in such group. It is quite impossible for the penetration of adult organ to take place. Usually penis is placed either within the vulva or between thighs. As such hymen is usually intact and there may be little redness and tenderness of the vulva. Bruises may present over the labia minora and majora depending upon the force applied by the perpetrator. The organ may be forcibly introduced causing rupture of the vaginal vault and associated visceral injuries in rare cases. Vaginal discharges due to gonorrhea and inflammation of body parts lead to suspicion in many cases[2].

Case presentation

This sexual assault case was referred to Forensic Medicine Department, Teaching Hospital, Maharajgunj from Metropolitan Police Circle, Balaju on 2070-2-3 (May 17, 2013). She was previously taken to the Kanti Children's Hospital`s emergency at around 12.25 PM and later on was brought to our department at 3.15 PM on 2070-2-3 (May 17, 2013). The victim was a seven months old female baby accompanied by her mother and a female constable. A valid written consent was taken from the mother. According to the examinee`s mother, she had been working as a waiter in a hotel nearby for seven months. She used to leave her seven months old daughter at Mrs. X`s house since 2070-1-29 (May 12, 2013) on payment basis. She often used to drop two to three packets of milk in the morning for her daughter. She used to daily check her baby after bringing her to home. That day, on 2070-2-3 (May 17, 2013), she had not noticed anything bad till 8 am in the morning. At around 9 AM, Mrs X came to the hotel where she worked and handed her baby to her telling that she would be leaving for Dhading and would return after two to three days. When the baby woke up at around 10 am to urinate, she saw blood in her undergarment and her external genitalia. She along with the hotel owner Ms. Y noticed white foamy secretions noted over her external genitalia. The baby cried and did not stop even after she was fed

milk. The victim`s mother further gave the history of washing the genital region and the underwear of the baby girl. On examination, the averagely built baby girl weighed 7 kg and was 60 cm in height. Her vitals were within normal limit. No any evidence of secondary sexual characteristics development was noted. Her tooth had not erupted yet. There was no any fresh injury noted on general body examination from head to toe. On anogenital examination, redness was noted over inner aspects of bilateral labia minorae and majorae. Hymen was intact. A skin deep laceration measuring 0.6 x 0.2 cm was present over the posterior fourchette. Clitoris, fossa navicularis, anal and perianal regions were free from fresh injury. A swab was collected from the exterior of the genitalia for laboratory investigation. Tests for HIV, HBV and other venereal diseases were recommended and she was further referred to pediatrician for further management.

DISCUSSION

Anogenital injuries caused by sexual abuse are uncommon. Coexisting allegations and lesions in the anogenital area lend support to the hypothesis of sexual abuse. The differential diagnosis of genital trauma also includes accidental injury (e.g. fall) and physical child abuse. This differentiation may be difficult and may require a careful history and multidisciplinary approach. Because many congenital malformations like hymenal notch, bump and infections e.g. lichen sclerosis, bacterial, viral or fungal skin manifestations or other causes of ano-genital abnormalities like diaper/napkin rash may be confused with abuse, familiarity with these other causes is important[3,4]. Although physical examination is important, the diagnosis of child sexual abuse is generally based on the affected child's statements, which should be obtained according to the proper procedure. All physicians should know that the physical findings are normal in more than 90% of cases and understand why this is so[5].

In very young children, due to deeper placement of the hymen and less capacity of the vagina, full penetration with rupture of hymen may not occur. Instead, there may be congestion, bruise or even tear laceration of the structures of posterior wall of introitus and vagina which may extend up to perineum[6]. Typical findings described in the virgin victim may not be elicited in a child victim, due to anatomical disproportion in genitals of the victim and accused[7]. Price J concluded his article with a debate on normal findings that are reported to occur in up to 99% of children referred for examination[8]. Assault on children mostly involve only fondling, simulated intercourse such as intercrural connection (i.e. penile friction between the inner thighs and external genitalia), or oral, or anal penetration. Hymen, therefore, is usually found intact and there may be redness and tenderness of vulva[9]. Bays and Jenny stated that a "sudden, accidental violent abduction of the legs may cause splitting injuries of the midline genital structures," although until then the only reported case was caused by sudden violent abduction during sexual abuse[3]. Lynch *et al* described 22 girls with "blunt urogenital trauma" without signs of sexual abuse[10].

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Journal of Gandaki Medical College-Nepal (J-GMC-N)

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