Nitroglycerine Ointment for Prevention of Osteoporosis in Postmenopausal North Indian Women

1Ruchika Garg, 2Soniya Dhiman, 3Prabhat Agrawal, 4Manish Bansal, 5Prashant Prakash

ABSTRACT

Introduction: Osteoporosis is a disease characterized by decreased bone strength and increased risk of fracture. A huge number of drugs are available for treatment of osteoporosis, but a number of factors limit their use. An inexpensive and widely available drug nitroglycerine (NG) applied topically might prove to be an effective. It increases bone formation and help in reduction of bone resorption by releasing the nitric oxide (NO), which improves bone cell function. In our study, we observed the effect of application of once-daily NG ointment to increase bone mineral density (BMD) at the lumbar spine and hip joint in a period of 24 months in postmenopausal women.

Aim and objectives: To determine the effect of topical NG on lumbar spine BMD and hip BMD.

Materials and methods: A single-center, double-blinded, placebo-controlled randomized trial conducted in Department of Obstetrics and Gynecology in collaboration with Department of Medicine, SN Medical College, Agra for 24 months. One hundred postmenopausal women (mean age 54.3 years) with lumbar spine T-scores of between 0 and –2 who completed a 1-week run-in period taking NG ointment were randomly assigned second group received the placebo ointment for 24 months. Bone mineral density was measured at the level of lumbar spine (L1–L4) and total hip using DXA.

Observations and results: Compared with the women in the placebo group, those in the NG group had significant increases in BMD at the lumbar spine (6.9%) and total hip (6.0%) at 24 months (p < 0.001 for all measures).

Conclusion: We have observed that by using NG, osteoporosis can be prevented and treated effectively and it leads to reduced risk of fractures. Headache was the only serious side effect which leads to drop outs in the study. But conflicting results of other studies and small sample size of our study suggest that efficacy of nitrates should be tested in larger RCTs before recommending its routine use in osteoporosis.

Keywords: Bone mineral density, Dual X-ray absorptiometry, Nitric oxide, Nitroglycerine.

INTRODUCTION

Osteoporosis, a disease where decreased bone strength increases the risk of a broken bone, increases worldwide with increasing age of population.1,2 In the developed world, depending upon the method of diagnosis, 2 to 8% of males and 9 to 38% of females were affected.3 Incidence of disease in the developing world is unclear.4 A huge number of drugs are available for treatment of osteoporosis but a number of factors limit their use, such as gastric problems and osteonecrosis of jaw are well known side effect of bisphosphonates. Hormonal treatment has its own limitations. An inexpensive and widely available drug is there which has shown its beneficial role in treatment of osteoporosis. Nitroglycerin (NG) applied topically might prove to be an effective and inexpensive agent for prevention of osteoporosis.5 It increases bone formation and also help in reduction of bone resorption.5 This phenomenon has not been observed with any of the current agents used to treat osteoporosis.5 Nitroglycerine act by releasing the nitric oxide (NO), which improves bone cell function.5 Many physiological processes in the body including bone remodeling are regulated by this short lived free radical NO.5,7 Organic nitrates (e.g. nitroglycerin, isosorbide dinitrate, ISMO) can act as NO donors.8

Nitric oxide can inhibit osteoclast activity and acts as a signaling molecule in osteoblasts and osteocytes.9-14 It has been observed in rodents that NO donors, such as NG, isosorbide mononitrate and isosorbide dinitrate, can prevent bone loss associated with estrogen deficiency and glucocorticoid administration.15,16 In vitro studies consistently demonstrate that NO has a biphasic effect on osteoclast activity and bone resorption.17,21
In our study, we observed the effect of application of once-daily ointment to increase bone mineral density (BMD) at the lumbar spine and hip joint in a period of 24 months.

AIM AND OBJECTIVES
To determine the effect of topical NG on lumber spine bone mineral density and to evaluate the changes in hip BMD.

MATERIALS AND METHODS
A single-center, double-blinded, placebo-controlled randomized trial conducted in Department of Obstetrics and Gynecology, SN Medical College, Agra in collaboration with Department of Medicine, SN Medical College, Agra. The study was conducted for 24 months started in July 2013 and completed in July 2015. One hundred twenty-six postmenopausal women (mean age 54.3 years) with lumbar spine T-scores of between 0 and –2 were taken into the study. Out of which 100 patients who completed a 1-week run-in period taking NG ointment were randomly assigned into two groups. The first group received NG ointment while the second group received the placebo ointment. 15 mg of 2% NG ointment rubbed to the skin of upper arm per day for 24 months, at bedtime instead of every 6 to 8 hours, as given in angina patients. Patients were advised to squeeze about 1 inch of the medicine. Calcium and vitamin D supplementation was given to every patient.

Outcome Measured
Bone mineral density was measured at the level of lumbar spine (L1–L4) and total hip using dual X-ray absorptiometry (DXA).

Ethical approval was given concerned by authority. Study medication was stopped if a clinical spine, proximal forearm, or hip fracture occurred; or if she began medical treatment with a nitrate. All participants gave written informed consent.

Inclusion Criteria
All women of 45 years or older who were at least 1 year postmenopausal attending the OPD in the Department of Obstetrics and Gynecology and Department of Medicine having BMD T scores between 0 and –2.0 at the lumbar spine and higher than –2.0 at the total hip were included in the study.

Exclusion Criteria
Women were excluded if they had:
- Medical conditions that influenced bone metabolism including hyperthyroidism, renal disease, malignancy.
- Used androgen, calcitonin, estrogen, progesterone, methotrexate or other antimetabolite drug, fluoride in a tablet form, raloxifene, tamoxifen, etidronate, prednisone, or equivalent, anticoagulants.
- Used lithium or anticonvulsants for 6 months before study entry.
- Women who were prescribed nitrates for cardiac conditions.
- Had a systolic blood pressure of 100 mm Hg or less or a diastolic blood pressure of 110 mm Hg or more at the baseline screening examination.
- An abnormal electrocardiogram at the baseline screening examination.
- A history of myocardial infarction, angina, valvular, or congenital heart disease.
- Had migraine headaches.
- Reported a hypersensitivity to nitrates.
- Smoking.
- Who did not give consent.

At initial examination, proper history taking and physical examination was done. Complete blood count, liver function test, kidney function test, blood sugar screening, electrocardiography and BMD of lumbar spine and hip was done for baseline. Patients were routinely followed-up up to 2 years. Bone mineral density of lumbar spine and hip was again measured at 1st and at 2nd years.

Assignment
Treatment was assigned by simple randomization using computer generated codes. Each tube (placebo or active nitroglycerin) identical packets and labeled with a nonrepeating allocation number that could be revealed only for safety concerns.

End Points
Areal BMD was measured by dual-energy X-ray absorptiometry at the lumbar spine and hip at baseline, 12 and 24 months. Measurements were performed by International Society of Clinical Densitometry certified technicians blinded to treatment assignment using a densitometer (GE Lunar Prodigy, Madison, Wisconsin). The reproducibility measurements in our study were 1.2% at the spine, 1.5% at the femoral neck, and 0.9% at the total hip.

At baseline, 12 and 24 months, volumetric BMD were calculated using the manufacturer’s software (version 6.00). All scans were performed by a technician and a phantom was scanned each day before performing scans on study participants.

Blinding
Collection and review of data were blinded to treatment assignment. Results of bone densitometry during follow-up
were not available to study participants. Treatment assignments were kept in a locked file by the pharmacist who generated the random allocation sequence.

**OBSERVATIONS AND RESULT**

Out of 126 postmenopausal women, who entered the run-in trial, 100 were randomized (n = 50 in nitroglycerin ointment group and n = 50 in placebo group). Out of the 26 women who were not randomized, 18 discontinued the study due to headaches, six lost interest, and two had an allergic reaction. The mean (SD) age of the participants was 54.3 (3.7) years, none of the participants had osteoporosis based on T-scores at the spine or hip, and there were no statistical differences in the distribution of baseline characteristics by treatment assignment (Table 1). During the 24 months, none of the participants initiated medications that are known to influence bone metabolism.

At 2 years, we observed that women randomized to the NG group had significant increases in areal BMD at the lumbar spine [1.07–1.16 gm/cm² vs placebo from 1.08–1.09 gm/cm²; percentage change, 6.9%; 95% confidence interval (CI), 5.2 to 8.2%; p < 0.001]; and total hip (0.84–0.94 gm/cm² vs placebo from 0.80–0.84 gm/cm²; 6.0%; 95% CI, 5.6–7.0%; p < 0.001) (Table 2).

Incidence of serious adverse events did not differ between NG [2 (4.0%)] and placebo [2 (4.0%)] groups.

**Table 1: Baseline characteristics of study participants**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Nitroglycerine (n = 50)</th>
<th>Placebo (n = 50)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>54.2 ± 3.6</td>
<td>54.4 ± 3.8</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>56.4 ± 4.4</td>
<td>55.6 ± 4.8</td>
</tr>
<tr>
<td>Years since menopause</td>
<td>5.2 ± 2.4</td>
<td>5.6 ± 2.8</td>
</tr>
<tr>
<td>T-score</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lumbar spine</td>
<td>−1.1 ± 0.5</td>
<td>−1.0 ± 0.6</td>
</tr>
<tr>
<td>Total hip</td>
<td>−0.8 ± 0.2</td>
<td>−0.7 ± 0.3</td>
</tr>
</tbody>
</table>

**Table 2: Absolute changes in value of BMD**

<table>
<thead>
<tr>
<th>Site and group</th>
<th>Baseline</th>
<th>12 months</th>
<th>24 months</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Lumbar spine</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>1.08</td>
<td>(1.06–1.10)</td>
<td>(1.07–1.11)</td>
</tr>
<tr>
<td>Nitroglycerine</td>
<td>1.07</td>
<td>(1.05–1.09)</td>
<td>(1.12–1.19)</td>
</tr>
<tr>
<td><strong>Total hip</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>0.80</td>
<td>(0.72–0.98)</td>
<td>(0.82–0.86)</td>
</tr>
<tr>
<td>Nitroglycerine</td>
<td>0.84</td>
<td>(0.82–0.96)</td>
<td>(0.92–0.96)</td>
</tr>
</tbody>
</table>

Osteoporosis, a progressive bone disease with decreasing BMD leading to an increase risk of fracture. It is characterized by distorted bone microarchitecture, and an altered amount and variety of protein inside the bone along with decreased BMD. There are limited options for treatment and prevention of osteoporosis. Calcium, vitamin D, bisphosphonates, calcitonin, SERMs, hormonal therapy and teriparatide, a lot of treatment options are available. All these have advantages and disadvantages but none of the above is a good management option for treatment of osteoporosis.

Topical NG is a cost effective, widely available, well known drug which act by releasing a free radicle NO. Adding NO to bone cultures decreases osteoclast maturation and bone resorbing activity and also enhances the differentiation and proliferation of osteoblasts. Continuous administration of nitrates induces tachyphylaxis to the effects. It was observed that older women taking nitrates intermittently for angina have higher BMD at the hip compared with nonusers and women taking it continuously.

Effects of nitrate was shown by a cross sectional study done in 1989 on 450 cardiac patients who were taking various doses of nitrates, this study demonstrated a dose dependent effect of nitrates on BMD. Rejnmark et al also found that use of nitrates were associated with an 11% reduced risk of any fracture and a 15% reduced risk of hip fracture over a period of 5 years for both men and women.

Jamal et al conducted a double blind, placebo controlled, randomized trial on postmenopausal women and found that treatment with 15 mg/day of NG ointment for 24 months increases bone formation and decreases bone resorption, resulting in an increased areal BMD at the lumbar spine and proximal femur.

An observational study suggested that women taking nitrates have a lower risk of all fractures, including hip fractures. A short-term randomized controlled trial (RCT) showed that isosorbide mononitrate taken once at bedtime decreased a marker of bone resorption and increased a marker of bone formation. An RCT of NG ointment (Nitro-Bid 22.5 mg/d) did not find increased BMD at the lumbar spine, femoral neck, or total hip; however, adherence to treatment in the study was poor.

In our study, we also found a significant increase in areal BMD at lumbar spine (6.9%) and total hip (6.0%) in postmenopausal females after topical application of 15 mg/day of NG ointment for 24 months.
Nitroglycerine has not been approved for this use. As NG is very well known drug and has no known serious and long-term side effects except for bothering headache. Nitroglycerine is very inexpensive, widely available in gel, tablet and cream form.

Our study has limitations. Headache was common, accounting for more than half of the dropouts during study. Sample size was small. Daily application of 15 mg/day NG ointment increases bone formation and decreases bone resorption; thereby, substantially improving BMD and bone structure. Furthermore, nitrates have a potential advantage of easy administration as an ointment, patch, or pill and wide availability of generic preparations. The efficacy of nitrates for reducing risk of fracture should be tested in a larger RCT.

REFERENCES