EVALUATION OF VITAMIN D LEVELS IN TERM PREGNANCY AND ITS OBSTETRIC OUTCOME IN INDIAN WOMEN

By

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IN

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I hereby declare that this dissertation entitled **"EVALUATION OF VITAMIN D LEVELS IN TERM PREGNANCY AND ITS OBSTETRIC OUTCOME IN INDIAN WOMEN"** is a bonafide and genuine research work carried out by me under the guidance of **Dr. MUNIKRISHNA. M,** Professor, Department of Obstetrics and Gynecology and **Dr. C.D. DAYANAND**, Professor, Department of Biochemistry, Sri Devaraj Urs Medical College, Tamaka, Kolar.

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Dr. SHILPA SURI CIRYAM

LIST OF ABBREVATIONS USED

Abbrevations	Description
1. 1, 25(OH) 2D	1, 25, DiHydroxy Vitamin D
2. 25(OH) D	25 Hydroxy Vitamin D
3. ACOG	American congress of obstetricians and gynecologists
4. BMI	Body Mass Index
5. ELISA	Enzyme linked immunosorbent assay
6. GDM	Gestational Diabetes Mellitus
7. IL	Interleukin
8. IUGR	Intrauterine growth retardation
9. LSCS	Lower segment caesarean section
10. NICE	National Institute for Health Care and Excellence
11. NICU	Neonatal Intensive Care Unit
12. PE	Preeclamsia
13. RCOG	Royal College of Obstetricians and Gynaecologists
14. SGA	Small For Gestational Age
15. TLR	Toll 2 Receptor

16. TNF	Tumor Necrosis Factor
17. VDBP	Vitamin D Binding Protein
18. VDR	Vitamin D Receptor
19. WHO	World health organization

ABSTRACT

EVALUATION OF VITAMIN D LEVELS IN TERM PREGNANCY AND ITS OBSTETRIC OUTCOME IN INDIAN WOMEN

INTRODUCTION:

Vitamin D deficiency is currently a global pandemic affecting all age groups in both developed and developing nations. Vitamin D is considered a fundamental hormone in calcium homeostasis and bone health. Its important role in cellular proliferation and differentiation, vascular function and immune regulation has been brought to light. Risk of vitamin D deficiency increases during pregnancy due to increased maternal and fetal demands and altered vitamin D metabolism. Recently, maternal vitamin D deficiency has been linked to adverse pregnancy outcomes, including preeclampsia, gestational diabetes, fetal growth restriction and preterm birth. Adequate vitamin D status appears to be relevant to health at all ages, and even in prenatal life.

OBJECTIVES:

1. To estimate serum vitamin D levels in term pregnancy.
2. To determine the association between serum vitamin D levels in term pregnancy and its obstetric outcome.

STUDY DESIGN:

This is a cross sectional, observational study conducted in the Department of Obstetrics and Gynecology at R.L. Jalappa Hospital and Research centre, Tamaka, Kolar, from December 2015- March 2017

MATERIALS AND METHODS:

A total number of 160 subjects were included and samples were collected after obtaining an informed consent. Under aseptic precautions 5ml of venous blood was collected from

median cubital vein of all subjects. The sample was centrifuged at 3000 rpm for separation of serum and stored at - 80°C till analysis. Analysis of vitamin D (25-hydroxy Vitamin D) was done using ELISA kit obtained from Calbiotech, Inc., California.

RESULTS:

Majority of the subjects were vitamin D deficient (81.87%) and 12.5% were vitamin D insufficient and only 5.63% were vitamin D sufficient. Majority of the cases were in the age group of 23-28 years (52.5%) with a normal BMI (18.5 to 24.99 kg/m2). The prevalence of vitamin D deficiency was more among the Primigravida (85.6%). High prevalence of vitamin D deficiency was seen in lower middle socioeconomic class (62.5%). Low vitamin D level during pregnancy was associated with higher rates of ceasarean section (92.4%). Maternal vitamin D deficiency was associated low birth weight of neonates (100%). Vitamin D deficiency was not associated with any adverse maternal outcome.

CONCLUSION:

In the present study, majority of women had deficient serum vitamin D levels. Most of these vitamin D deficient women belonged to the lower socioeconomic strata. A significant number of vitamin D deficient pregnant women developed fetal distress at term, which raised the cesaerean section rates in the vitamin D deficient group. Most of the neonates born to vitamin D deficient mothers were of low birth weight.

As there is a high prevalence of vitamin D deficiency worldwide and screening for maternal serum vitamin D levels is not proven to be cost effective, we conclude that, there is a need for vitamin D supplementation for all pregnant women as a routine antenatal practice.

Keywords: Vitamin D, Term gestation, Obstetric outcome.

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INTRODUCTION

Vitamin D is considered a fundamental hormone in calcium homeostasis and bone health. Its important role in cellular proliferation and differentiation, vascular function and immune regulation has been brought to light.[1] Recently, maternal vitamin D deficiency has been linked to adverse pregnancy outcomes, including preeclampsia, gestational diabetes, fetal growth restriction and preterm birth. Adequate vitamin D status appears to be relevant to health at all ages, and even in prenatal life.[2]

Vitamin D is called calciferol and has 2 physiologically active forms – D2 (ergocalciferol) which is synthesized from plants and D3 (cholecalciferol) which is subcutaneously produced upon exposure to ultraviolet B rays.[3]

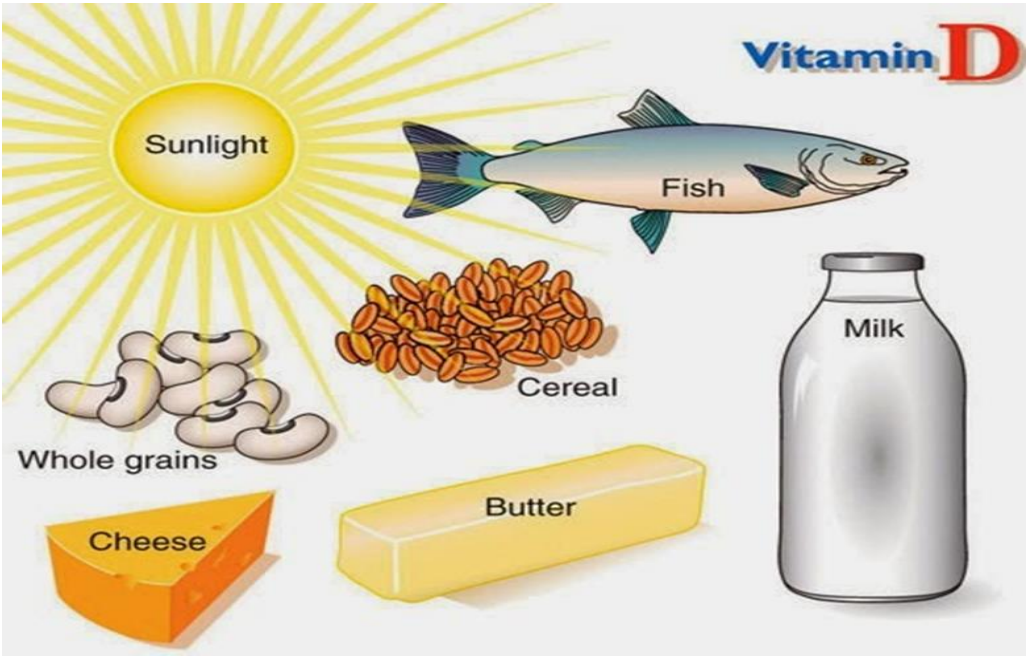

Figure 1 - Source of vitamin D

Vitamin D3 is three times more effective than vitamin D2 in raising serum concentrations of vitamin D and maintaining those levels for a longer time. Its metabolites have superior affinity for vitamin D-binding proteins in plasma.[3,4]

Vitamin D2 and D3 are first metabolised in the liver to 25 (OH)D – calcidiol D2 and then in the kidneys to 1,25(OH)2 D – calcitriol D3 which is the active form responsible for calcium homeostasis and bone mineralisation.

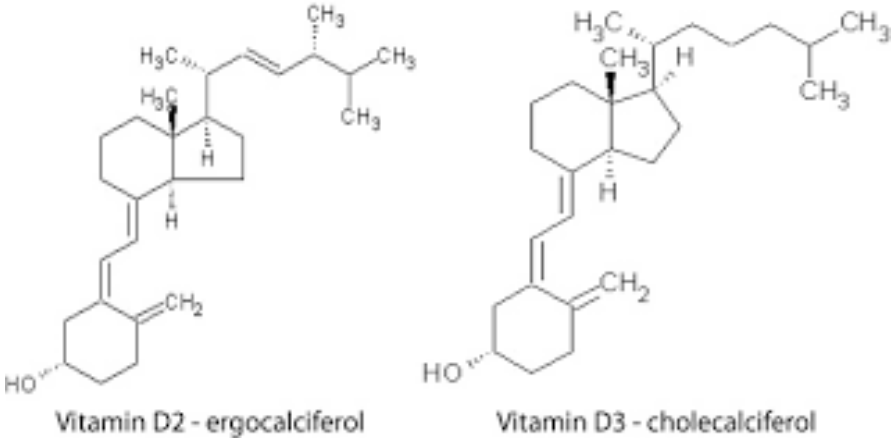

Figure 2 - Vitamin D chemical structure

Vitamin D has a short half-life, and thus adequate vitamin D intake is necessary in order to ensure sustained circulating levels.[3] Vitamin D status is affected by factors that regulate its production in the skin such as skin pigmentation, latitude, dressing codes, season, aging, sunscreen use, and air pollution and also by factors affecting its absorption or metabolism. Melanin acts as a filter for ultraviolet (UV) rays hence reducing the production of vitamin D by the skin. Differences in latitude have also been shown to influence the concentration of vitamin D. Seasonal variation in the concentration of vitamin D with higher levels during the summer compared with the winter months is also seen. Vitamin D metabolism is also affected in obese individuals, as vitamin D is deposited in body fat stores, making it less bioavailable.[5]

Assessment of vitamin D status is done by 25(OH)D, as it reflects the sum total of vitamin D produced cutaneously and that obtained by food and supplements.

NEED FOR THE STUDY

Vitamin D deficiency is currently a global pandemic affecting all age groups in both developed and developing nations. Risk of vitamin D deficiency increases during pregnancy due to increased maternal and fetal demands and altered vitamin D metabolism.

Immunomodulatory role of vitamin D during pregnancy enables successful implantation by attenuating decidual T cell function. Vitamin D is also important for maintaining pregnancy by regulating calcium metabolism in the myometrium. Vitamin D also contributes to the production of antimicrobial peptides that prevent infection in both mother and fetus during pregnancy and early childhood.[4]

Vitamin D deficiency is unexpected in a tropical country like India, where there is abundant overhead sun for almost all around the year. However it was observed that 96% of Indian pregnant women had vitamin D insufficiency and 60% were vitamin D deficient.[6] This paradox maybe partly explained by the many prevalent social and cultural practices in India that precludes adequate exposure of adolescent girls and young women to sunshine. Revealing clothing is frowned in traditional Indian households, both rural and urban. Newly married women are expected to cover themselves even more and are discouraged from outdoor activity. Increased urbanization that results in poor outdoor activity and greater pollution, coupled with skin pigmentation, may further compound this problem.[7]

In a population that already has a high prevalence of vitamin D deficiency, the problem is likely to worsen during pregnancy. Vitamin D deficiency during pregnancy may lead to life threatening complications both in the mother and the neonate. Recently, maternal vitamin D deficiency has been associated with elevated risk of gestational diabetes mellitus (GDM), preeclampsia, preterm birth and caesarean section in the mother and also

neonatal complications like fetal hypovitaminosis D, low birth weight, small for gestational age (SGA) and intrauterine growth retardation (IUGR).[8-16]

Taking into consideration the above mentioned risks with hypovitaminosis D, the necessity for diagnosing vitamin D deficiency in pregnant women visiting RLJ hospital was felt. This will help to find prevalence of hypovitaminosis D in pregnant women for better obstetric outcome and to create awareness amongst rural women in order to prevent above mentioned maternal and fetal complications.

OBJECTIVES

1. To estimate serum vitamin D levels in term pregnancy.
2. To determine the association between serum vitamin D levels in term pregnancy and its obstetric outcome.

REVIEW OF LITERATURE

Vitamin D was classified in the second half of the 20th century as a prohormone. Vitamin D is a unique vitamin because unlike other vitamins it can be obtained exogenously from food and synthesized in the skin from sunlight. It also contains hormone-like traits in its function and seco-steroid structure. Impaired vitamin D levels during pregnancy is associated with adverse outcomes such as preeclampsia, gestational diabetes, preterm birth and poor neonatal outcome. Maintenance of normal pregnancy requires an effective coordination of anti-inflammatory and antimicrobial responses within the fetoplacental unit. Vitamin D helps in maintaining normal pregnancy upto term by modulating both these anti-inflammatory and antimicrobial processes.[17]

Vitamin D metabolism:

The de novo synthesis of vitamin D3 in humans begins in the skin. 7-dehydrocholesterol is the precursor molecule, which is called provitamin D3. Following exposure to Ultraviolet B rays in the range of 280-320nm, 7-dehyrocholesterol (provitamin D3) is converted to previtamin D3. Through a subsequent thermal hydroxylation reaction in the skin, isomerization takes place converting previtamin D3 to vitamin D3. Thus it is seen that the prerequisites for de novo synthesis of vitamin D3 are 7-dehydrocholesterol and sunlight of a specific wavelength. Without these prerequisites, humans are completely dependent on dietary intake of vitamin D, which maybe in the form of either vitamin D2 or vitamin D3.[18,19]

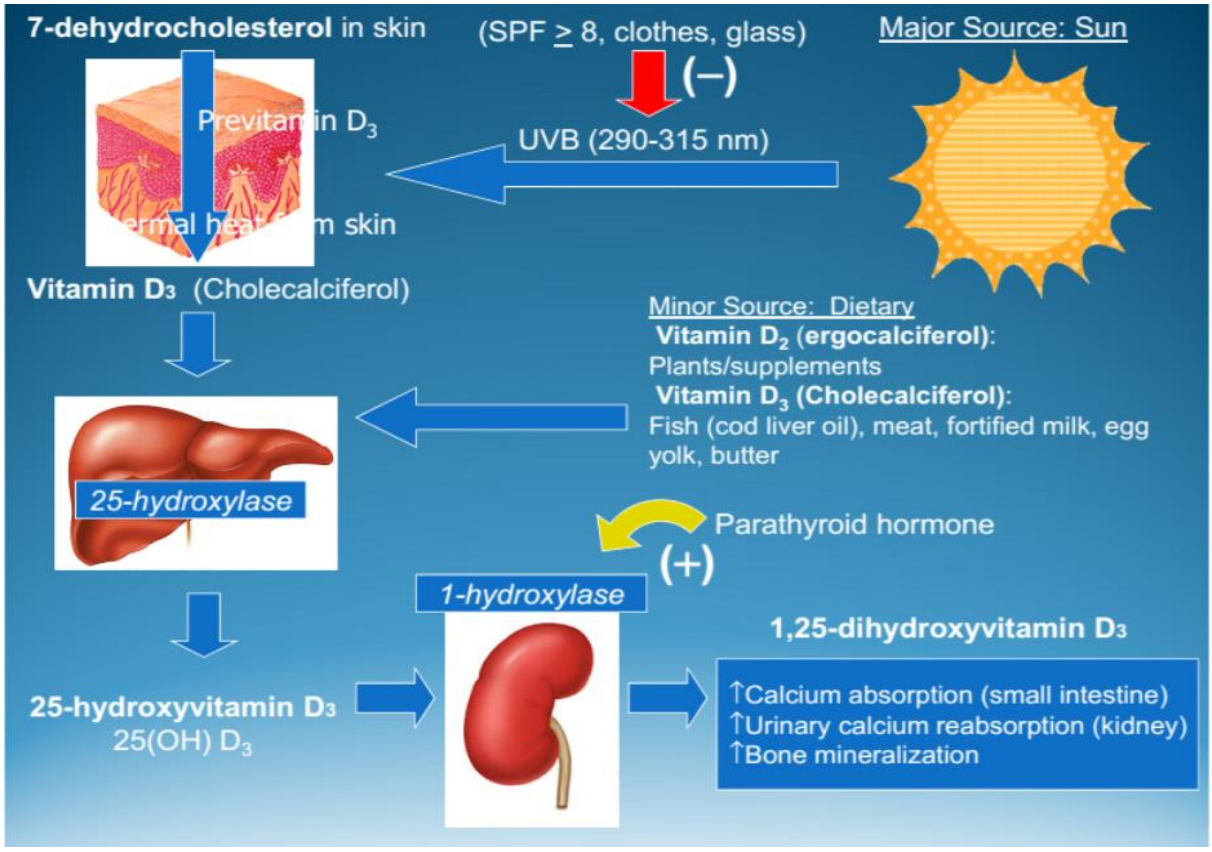

Figure 3 - Vitamin D metabolism

Following its synthesis, vitamin D binds to vitamin D binding protein (VDBP) and enters the circulation. The concentration of vitamin D in circulation is represented as international units (IU) or micrograms with a known conversion of 40IU = 1 microgram.

Vitamin D conversion rate is dependent on a healthy functional liver and the activity of an enzyme 25-hydroxylase that is synthesized in the liver. Thus those with impaired liver function will have a diminished 25(OH)D levels. Vitamin D is mostly bound to VDBP in circulation, and only a small quantity is unbound. The half-life of 25(OH)D is 2-3 weeks, which makes it a good indicator of body's vitamin D status.[20,21]

VDBP bound vitamin D is taken up from the circulation by the epithelial cells of the proximal tubules of the kidney. Here 25 (OH)D is converted to 1,25(OH)2D or calcitriol, which is the active form of vitamin D, by the action of the mitochondrial enzyme 1-α-hydroxylase.

Calcitriol's classic triad of action:

1. Increase in intestinal calcium absorption through calbindin.
2. Increase in urinary calcium reabsorption.
3. Regulation of parathyroid hormone (PTH) in a negative feedback loop that prevents demineralization of bone due to PTH and allows calcium to be absorbed from the gastrointestinal tract and reabsorbed from urine thus maintaining calcium homeostasis.

Adequate concentration of 25(OH)D (>30mg/ml) must be present in circulation to provide sufficient substrate to form 1,25(OH)2D. In individuals with vitamin D deficiency only trace amounts of 25(OH)D will be present in circulation as it will be converted to 1,25(OH)2D in order to maintain calcium homeostasis.

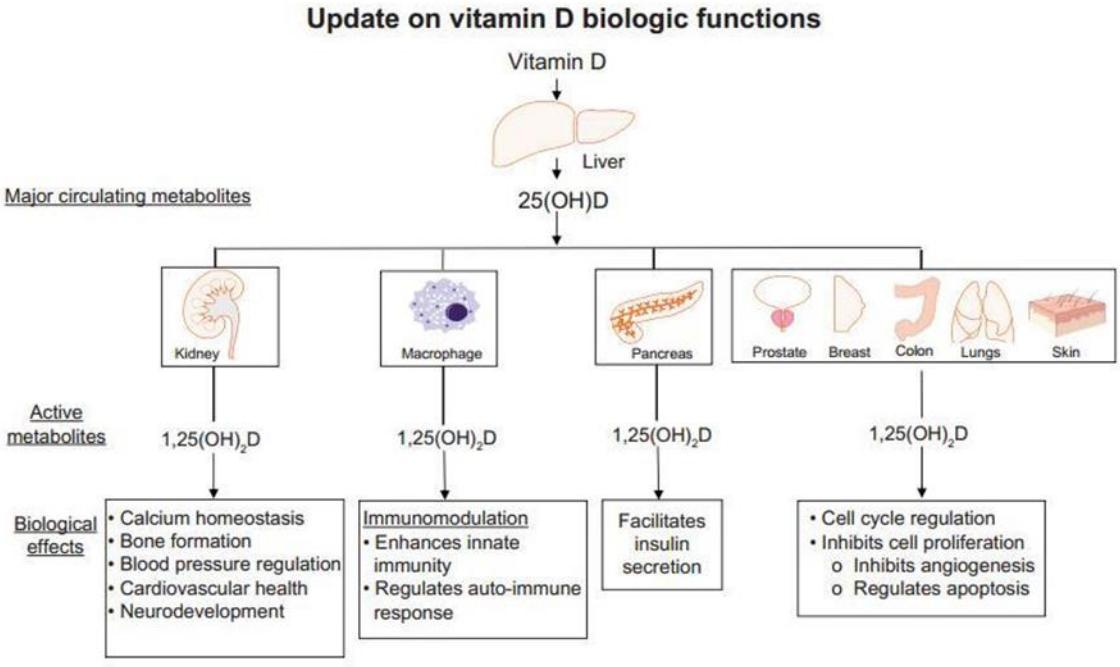

Figure 4 - Biologic functions of vitamin D

Metabolism of vitamin D during pregnancy:

Vitamin D metabolism is important for a range of key developmental events, including decidualization, modulation of maternal immune function and fetal bone formation.

To elicit biological effects, vitamin D, which is obtained through the diet or synthesized in the skin via UVB exposure must be converted to its active form 1,25(OH)2D. This is achieved by first converting circulating vitamin D to 25-hydroxyvitamin D (25(OH)D) by hepatic 25-hydroxylase, which is then hydroxylated to 1,25(OH)2D via the enzyme 1α-hydroxylase, encoded by the CYP27B1 gene in the kidney. Placenta also exhibits abundant expression and activity of 1α-hydroxylase as in the kidneys. This indicates functional importance of local placental vitamin D metabolism during gestation.[22]

Circulating 1,25(OH)2D binds to the vitamin D receptor (VDR) and induces subsequent gene transcription through vitamin D response element (VDRE) binding to target gene promoter regions to elicit genomic effects. VDR is also abundantly expressed in placental tissue, which allows for localized placental 1,25(OH)2D binding and subsequent gene transcription. The precursor 25-(OH)D readily crosses the placenta; however, 1,25(OH)2D cannot be transported across the placenta, and thus relatively low levels of 1,25(OH)2D is found in fetal circulation.[22]

Immune adaptations are vital for successful pregnancy outcome and vitamin D likely acts to promote implantation due to its anti inflammatory role in inflammatory pathways and its role in immune function.

Vitamin D deficiency

As the statistics keep surfacing at an alarming pace, currently vitamin D deficiency is recognized as the most un-treated nutritional deficiency worldwide. Vitamin D deficiency is a significant public health problem in all the developed and developing countries including India.[23-25]

Vitamin D deficiency is common, especially in women with pigmented skin. In a study conducted in London in the antenatal population, vitamin D level of less than 25 nmol/l

(vitamin D deficiency)was found in 47% of Indian Asian women, 64% of Middle Eastern women, 58% of black women and 13% of Caucasian women.[26]

In the general adult population, reduced vitamin D concentrations are found in obese subjects. Prepregnancy obesity has been associated with lower levels of vitamin D in both pregnant women and their neonates; 61% of women who were obese prior to pregnancy with a prepregnancy BMI of >30kg/m² were found to be vitamin D deficient, compared to 36% of women with a prepregnancy BMI of less than 25kg/m².[27]

A study conducted in northern India found a high prevalence of physiologically significant hypovitaminosis D among pregnant women and their newborns, the magnitude of which warrants public health intervention.[28]

Vitamin D in pregnancy

Pregnancy results in an increased risk of vitamin D insufficiency due to the increased demand for calcium to support fetal bone formation.[29] Current research suggests vitamin D deficiency is related to the development of gestational diabetes and pre-eclampsia. Assessment of vitamin D sufficiency in pregnancy is best done by measuring serum 25(OH)D levels in the mother as this is the form that crosses the placenta and thus adequately depict the fetal vitamin D status. Calcitriol is formed from 25(OH)D within the fetal kidney.[30-32]

Maternal complications in hypovitaminosis D:

1. Preeclampsia

Vitamin D plays a critical role in placental development and function. This is proven by maternal vitamin D deficiency being implicated in pregnancy

complications associated with placental dysfunction such as preeclampsia, preterm birth and IUGR and the presence of a local vitamin D metabolic pathway. The key mechanisms include effects on implantation, inflammation, immune function and angiogenesis.[22]

In three studies, it was seen that women with serum vitamin D levels less than 50nmol/l had 5 fold increased risk of severe preeclampsia. It was also seen that, women who developed pre-eclampsia had lower levels of vitamin D than women who did not.

In a study done in Sweden, an increase in 25(OH)D concentration during pregnancy of at least 30 nmol/L, regardless of vitamin D status at first trimester, was associated with a lower odds ratio for preeclampsia.[33]

A prospective cohort study of 697 pregnant women, found that a serum 25(OH)D levels less than 50nmol/L at 24-26 weeks gestation were significantly associated with an increased risk of developing pre-eclampsia.

However, many studies have shown a weak or no relationship between vitamin D and hypertensive disorders in pregnancy. A Canadian study showed that women with low circulating maternal vitamin D levels are more likely to have hypertension in pregnancy in the univariate analysis, but not the multivariate analysis.[34]

Two other studies also failed to show any association between vitamin D levels and the development of pre-eclampsia,gestational hypertension or preterm birth.[35,36]

However, in a meta-analysis including 31 studies, demonstrated that vitamin D insufficiency was associated with pre-eclampsia and small for gestational age (SGA) infants.[37,38]

2. Impaired glucose tolerance

Vitamin D deficiency is closely associated with Gestational Diabetes Mellitus (GDM). Vitamin D enhances insulin-dependent glucose transport by inducing insulin secretion and insulin receptor expression through Vitamin D receptor. Vitamin D is also a potential immunosuppressant, which down-regulates the expression of pro-inflammation markers, such as TNF-α and IL-2, among pregnant women with GDM.[39]

Zhang et al observed that the risk of gestational diabetes increased 2.66 fold in participants that had serum 25(OH)D levels <50nmol/L. Serum 25(OH)D and glucose levels after a glucose tolerance test were inversely correlated.[40]

In a study including 184 pregnant women with gestational diabetes, it was found that, maternal vitamin D deficiency was associated with an increased risk of adverse neonatal outcomes, such as neonatal hypoglycemia requiring ICU and SGA newborns.[41]

There are some data to suggest that the association between 25(OH)D levels and GDM risk is specific to ethnicity. In a majority non-Hispanic white population, 25(OH)D concentrations at 16 weeks of gestation were significantly lower in GDM subjects than in controls, whereas no association was found in Indian GDM mothers where 25(OH)D concentrations were measured at 30 weeks of gestation.[42]

3. Maternal obesity

Studies have consistently found that those who are obese have an increased risk of vitamin D deficiency. No definite explanation has been determined. However there are two theories that have been hypothesised, the first one is the sequestration of

vitamin D by adipose tissue and the second one is the regulation of adipose tissue by vitamin D.[43]

Wortsman et al did an interventional study where non-pregnant obese and lean participants had serum 25(OH)D drawn 24 hours after they were either exposed to UV radiation or given a 50,000 IU dose of Vitamin D3. The study found that BMI was inversely correlated with serum vitamin D3 levels after exposure to equal amounts of UV light or vitamin D3 supplementation. This was explained by the fact that subcutaneous fat stores vitamin D3 and it was thought that more vitamin D was sequestered in the obese than the non-obese subject due to increased fat metabolism.[44]

4. Preterm birth

Two studies examined relationships with blood levels of 25(OH)D during pregnancy and preterm birth. In Tanzania, no difference in risk of preterm birth (<37 weeks, RR=0.84[0.55–1.28]) or early preterm birth (<34 weeks, RR=0.77 [0.50, 1.18]) was observed among vitamin D deficient women using a cut-off of 80 nmol/L.[45]

Similarly, no difference in the third trimester 25(OH)D levels was found for adolescents in UK.[46]

Some observational studies suggested longer gestational duration associated with higher 25(OH)D levels.

In the Netherlands, women with serum levels >50 nmol/L had a slightly longer gestational length of 40.2 weeks vs. 40.0 weeks in women in with lower intake (P<0.001).[47]

An Australian study noted a significant 0.7-week shorter gestation among women with blood 25(OH)D levels <28 nmol/L compared with those with higher levels.[48]

5. Other complications

Vitamin D deficiency (< 37.5 nmol/l) has been associated with a four-fold increased risk of primary caesarean section (caesarean section performed for the first time), although this has not been demonstrated in all studies.[49]

Vitamin D deficiency is also associated with bacterial vaginosis in pregnant women.[50]

Neonatal complications:

Neonatal vitamin D levels are correlated with those of their mother, with maternal vitamin D deficiency increasing the risk of neonatal vitamin D deficiency.

1. Neonatal hypocalcaemic seizures

In an Australian study, hypovitaminosis D was found in 15% of pregnant women and 11% of neonates. Low maternal serum levels of 25(OH)D was associated with low levels of vitamin D and calcium in the neonate. Vitamin D deficiency was observed to be a major cause of hypocalcaemic seizures in neonates and infants.[51]

Hypocalcaemia is not uncommon in neonates and is a potentially severe problem.

In another study from Egypt, all mothers of babies with hypocalcaemic seizures had severe vitamin D deficiency.[52]

Maternal vitamin D deficiency is a common, and potentially preventable, cause of neonatal hypocalcaemia. This is especially common in South Asian women.

2. Skeletal development and growth

An association between small-for-gestational age babies and maternal vitamin D deficiency was seen in the first trimester.

Hossain et al found a positive correlation between maternal serum vitamin D status and fetal cord blood vitamin D levels. They also observed an inverse relationship between cord blood serum 25(OH)D and birth weight.[53]

Hypovitaminosis D is associated with impaired growth and bone development in the fetus. There is increasing evidence to show that more profound maternal 25(OH)D insufficiency may lead to suboptimal bone size and density after birth without overt rachitic change.[54] But this is likely to lead to an increased risk of osteoporotic fracture in later life.

In a UK mother–offspring cohort study, 31% of the mothers had circulating concentrations of 25(OH)D in late pregnancy of 27–50 nmol/dl. There was a positive association found between maternal 25(OH)D concentration measured in late pregnancy and whole body bone mineral content and density assessed using dual energy X-ray absorptiometry (DEXA) in the offspring at 9 years of age.[55]

Additionally, maternal UVB exposure during pregnancy was positively associated with whole body bone mineral content in the offspring at the 9 years of age in the Avon Longitudinal Study of Parents and Children.[54]

3. Fetal lung development

Low maternal vitamin D intake in pregnancy is associated with wheeze and asthma in the offspring.[56]

Low cord blood 25(OH)D concentrations have been associated with respiratory syncytial virus bronchiolitis and respiratory infections in neonates.[57]

4. Childhood immune disorders

There are plausible physiological mechanisms for explaining the association between prenatal vitamin D status and immune development in childhood. The metabolite 1,25(OH)2D has been shown in animal and in vitro models to have an immune-modulatory role and low levels of neonatal vitamin D have been linked to childhood asthma. In this study it was seen that a higher maternal intake of vitamin D during pregnancy decreases the risk of recurrent wheeze in early childhood.[58]

Maternal vitamin D supplementation is associated with cord blood gene expression of tolerogenic immunoglobulin such as immunoglobulin-like transcripts 3 and 4 (ILT3 and ILT4).Cord blood 25(OH)D is correlated with mononuclear cell release of IFN-γ and hence Th1 cell development. Cord-blood levels of 25(OH)D had inverse associations with risk of respiratory infection and childhood wheezing but no association with incident asthma.[59]

Screening for vitamin D deficiency in pregnancy

There are no data to support routine screening for vitamin D deficiency in pregnancy in terms of health benefits or cost effectiveness. Pregnant women benefitting by screening test are: darker skin colour or coverage, obesity, risk of pre-eclampsia, or gastroenterological conditions limiting fat absorption.[60]

As the test is expensive, offering it to all at-risk women also may not be cost effective as compared to offering universal supplementation, as treatment with vitamin D supplements is regarded as being very safe.

At present, there are no data to support a strategy of measurement of vitamin D levels followed by treatment in the general female population. Measurement of vitamin D in a

hypocalcaemic or symptomatic pregnant woman as part of their management is applicable. This includes women with a low calcium concentration, bone pain, gastrointestinal disease, alcohol abuse, a previous child with rickets and those receiving drugs which reduce vitamin D.

Supplementation and treatment in pregnancy

Daily vitamin D supplementation with oral cholecalciferol or ergocalciferol is safe in pregnancy. The 2012 recommendation from UK Chief Medical Officers and NICE guidelines state that all pregnant and breastfeeding women should take 10 micrograms of vitamin D supplements daily after being informed about the importance of vitamin D.[61]

Three categories of vitamin D supplementation are recommended.

1. In general, vitamin D 10 micrograms (400 units) a day is recommended for all pregnant women in accord with the national guidance.[61]
2. High-risk women are advised to take at least 1000 units a day (women with increased skin pigmentation, reduced exposure to sunlight, or those who are socially excluded or obese).[62]

   The RCOG has highlighted the importance of addressing suitable advice to these women. Women at high risk of pre-eclampsia are advised to take at least 800 units a day combined with calcium as treatment.[63]
3. For the majority of women who are deficient in vitamin D, treatment for 4– 6 weeks, either with cholecalciferol 20,000 IU a week or ergocalciferol 10,000 IU twice a week, followed by standard supplementation, is appropriate.[64]

For women who require short-term repletion, 20,000 IU weekly appears to be an effective and safe treatment of vitamin D deficiency. A daily dose is likely to be appropriate to maintain subsequent repletion (1000 IU daily).[65]

In adults, very high doses of vitamin D (3,00,000–5,00,000 IU intramuscular bolus) may be associated with an increased risk of fractures and such high doses are not recommended in pregnancy.

A study by Hollis et al demonstrated that supplemental doses of 4000 IU cholecalciferol a day were safe in pregnant women when compared to higher doses and most effective compared to the lower doses.[65]

Safety of vitamin D

In a normal pregnancy there is enhanced intestinal calcium absorption. If vitamin D toxicity occurs it is manifested as hypercalcaemia and hypercalciuria. Therefore, there is a hypothetical concern that when secondary hyperparathyroidism (increases serum calcium levels) follows vitamin D deficiency, calcium given with vitamin D may be associated with temporary hypercalcaemia. However, this is self-limiting due to the associated hungry bone syndrome and has not been demonstrated to represent a clinical problem.[60]

MATERIAL AND METHODS

Study Design:

This is a cross sectional, observational study.

Source of Data:

Study was conducted in Department of Obstetrics and Gynaecology, R L Jalappa Hospital and Research Center, Tamaka, Kolar attached to Sri Devaraj Urs Medical College.

Duration of study:

December 2015 to March 2017

Sample size:

Estimated sample size is 160.

This is based on vitamin D levels in pregnant women found in a study in Lucknow - Vitamin D Deficiency in Rural Girls and Pregnant Women Despite Abundant Sunshine in Northern India. Reported standard deviation is 7.92, with an average vitamin D level of 15.12, with 10% absolute error in mean and 95% confidence.

Formula used: $n = \frac{Z\alpha^2 S^2}{d^2}$

Where: n = sample size S = 63

d = 10% of mean Z = 1.96

Inclusion Criteria :

All Singleton pregnant women between 37 to 40 weeks of gestation who came to Sri R L Jalappa Hospital and Research Centre during the study period.

Exclusion Criteria :

Pregnant women with-

1.Multiple gestation

2. Premature rupture of membranes

3. Preterm labor

4.Uterine anomalies

5.Previous caesarean section

6.Preexisting medical illness like hypertension, diabetes mellitus, cardiac disorders, renal diseases.

7.Chronic granuloma forming conditions like sarcoidoses.

8.On treatment with anticonvuslant, antifungal and anti retroviral drugs.

Method of collection of data:

Subjects: A total number of 160 samples was collected. All pregnant women fulfilling the inclusion criteria were registered for the study.

Detailed history regarding age, parity, duration of gestation, menstrual history, obstetric history and any complications in present pregnancy was taken. General clinical examination and complete obstetric examination was done. Necessary investigations along with Non stress test was done.

Sample collection:

After obtaining an informed consent, under aseptic precautions 5ml of venous blood of median cubital vein of subjects was collected in a plain tube. The sample was centrifuged at 3000rpm for separation of serum and stored at -80^0 C till analysis.

Ethical clearance:

Ethical clearance was obtained by the instituitional ethical committee on 26/12/2015

Analysis of vitamin D:

Vitamin D kit was procured from Calbiotech company and assay procedure was carried out as per kit. The Calbiotech, Inc. 25-hydroxy (25-OH) Vitamin D ELISA is designed for the quantitation of total 25-OH Vitamin D in human serum and plasma.

Figure 5 - Vitamin D ELISA kit

Procedure:

All reagents and specimens were allowed to come to room temperature before use.

10µl of 25(OH)D Standards, controls and samples along with 200µl working solution of biotinylated 25(OH) D reagent were dispensed into each well. Contents in the wells was mixed for 20 seconds using a plate shaker at 200-400 RPM (or equivalent motion). First incubation of the sealed plate was done for 90 minutes at room temperature. First wash was given by dispensing 300µl of 1X Wash Buffer into each well, and then briskly shaking out the 1X Wash Buffer into a waste reservoir. This was repeated 2 more times for a total of 3 washes. Following the wash 200µl of enzyme conjugate (Streptavidin-

HRP) was dispensed into each well. Second incubation was done for 30 minutes, at room temperature. Wash was repeated for the second time with 300µl of 1X Wash Buffer, similar to the first wash. After which 200 µl of TMB Substrate was dispensed into each well. Third incubation was done for 30 minutes at room temperature. The assay was stopped by adding 50 µl of Stop Solution into each well to stop the enzymatic reaction. Absorbance on ELISA Reader was read at 450 nm within 10 minutes of adding the Stop Solution.

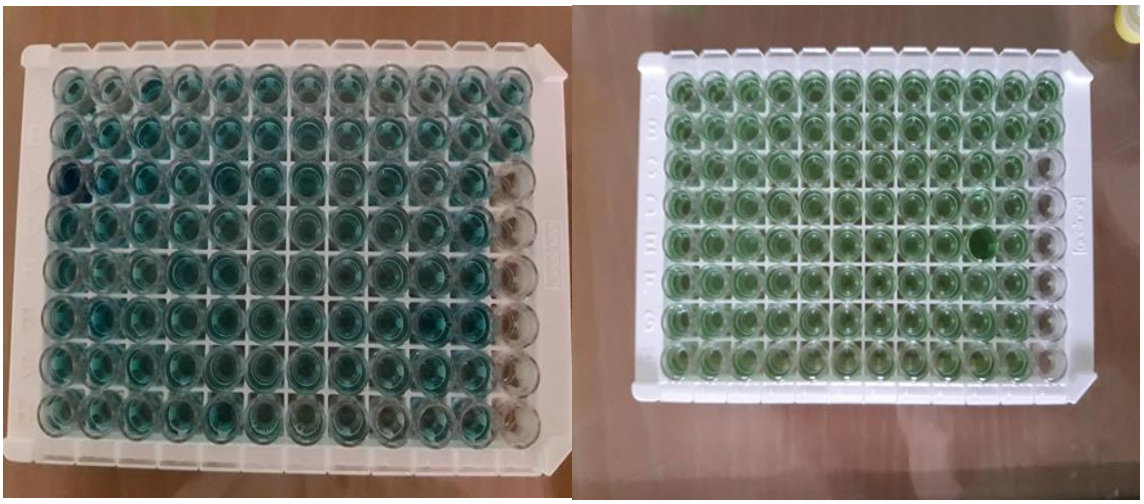

Figure 6 - Steps of vitamin D analysis

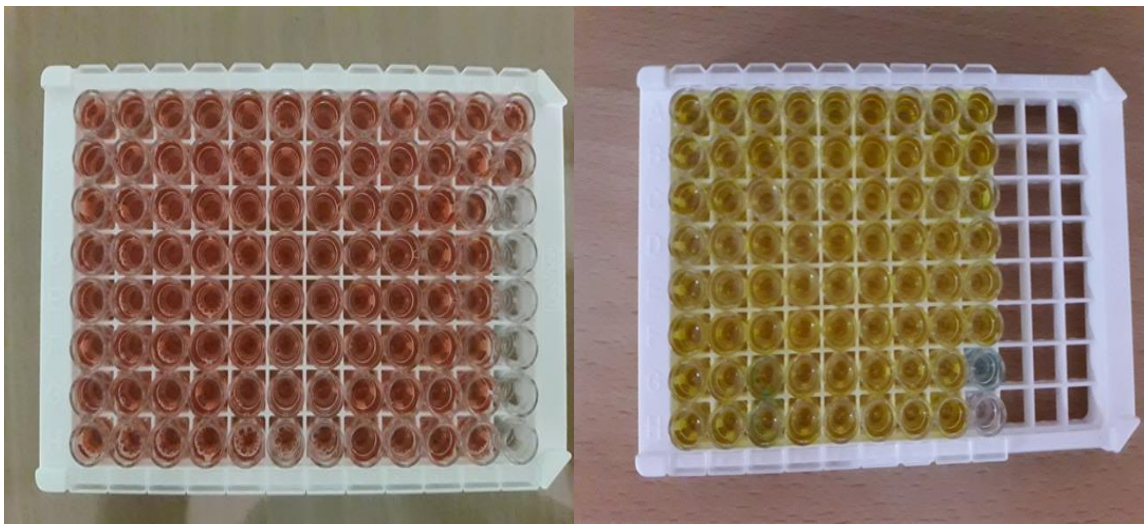

Figure 7 - Steps of vitamin D analysis

Figure 8 - Washer

Figure 9 - ELISA microplate analiser

Figure 10 – Centrifuge

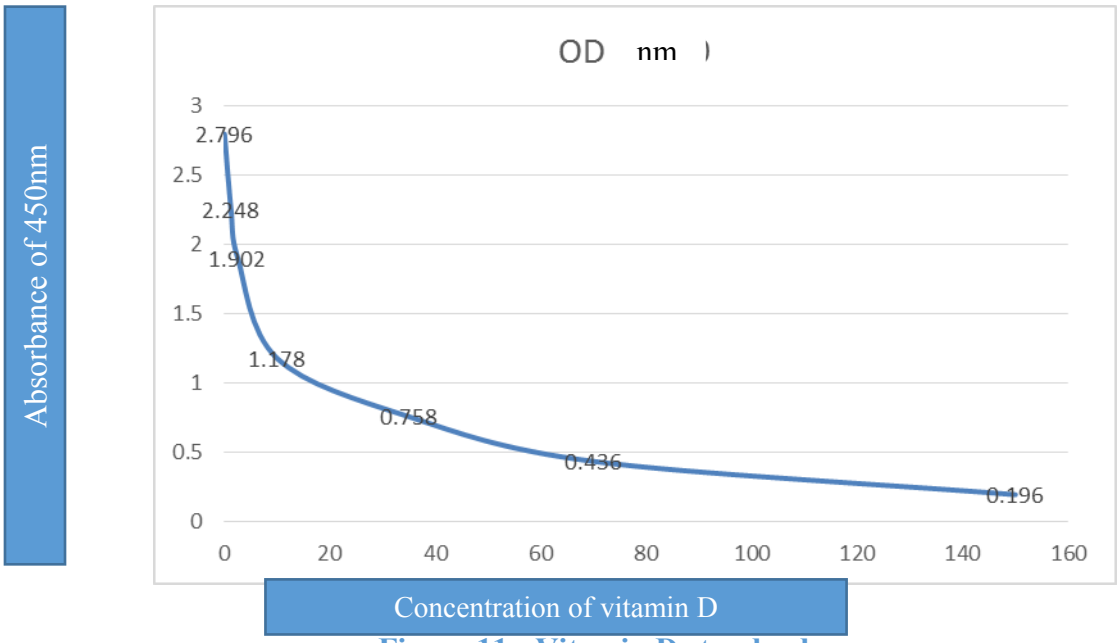

Figure 11 - Vitamin D standard curve

Results were calculated using the formula:

y = mx + c

Where, m = -0.0141 and c = 1.9001

y = -0.0141x + 1.9001

Vitamin D values was categorised according to the reference range given below:[66]

Table no. 1:- Serum vitamin D reference range – Endocrine society 2011

Vitamin D status	Concentration in ng/ml
Deficiency	<20
Insufficiency	21-29
Sufficiency	>30
Intoxication	>150

Statistical analysis

Various maternal and fatal outcomes like mode of delivery, APGAR score of the child, NICU admission etc. were considered as outcome variables.

Presence or absence of vitamin D deficiency was considered as Primary explanatory variable.

Descriptive analysis: Descriptive analysis was carried out by mean and standard deviation for quantitative variables, frequency and proportion for categorical variables. Data was also represented using appropriate diagrams like bar diagram, pie diagram and box plots.

Vitamin D deficiency and outcomes was assessed by cross tabulation and comparison of percentages. Chi square test was used to test statistical significance.

P value < 0.05 was considered statistically significant.

RESULTS

This prospective study was conducted in the Department of Obstetrics and Gynecology, R.L. Jalapa Hospital, attached to Sri Devraj Urs Medical College, Tamaka, Kolar, Karnataka.

A total number of 160 samples were collected and serum vitamin D levels were analysed.

Table 2:- Serum vitamin D levels in term pregnancy

Vitamin D status	Number of subjects N = 160	Percentage
Deficiency (<20ng/ml)	131	81.87
Insufficiency (21-29ng/ml)	20	12.50
Sufficiency (>30ng/ml)	9	5.63
Intoxication (>150ng/ml)	0	0

Out of the 160 subjects studied, 131 (81.98%) were having serum vitamin D level less than 20ng/ml and none of them had serum vitamin D level above 150ng/ml. Mean vitamin D level was 11.22ng/ml at 95% Confidence interval for Vitamin D (11.23±1.89).

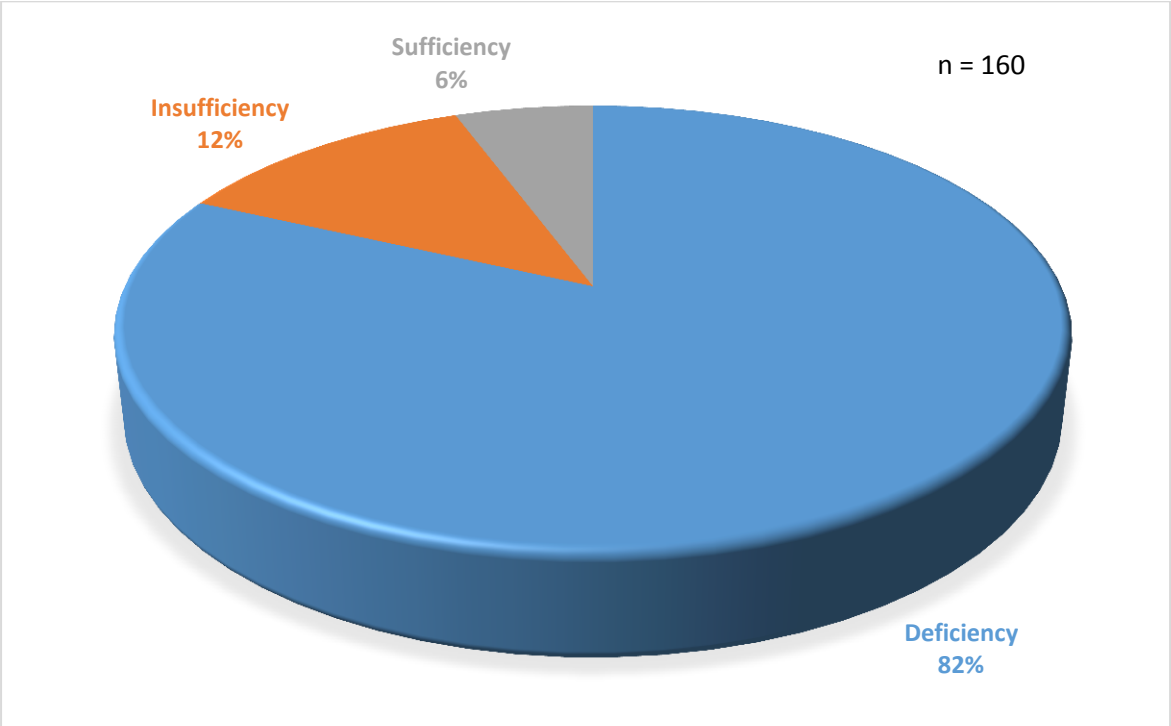

Figure 12:- Graph showing distribution of subjects according to serum vitamin D level.

Table 3:- Distribution of subjects according to age group and Vitamin D level

Age Group	Vitamin D								P Value
	Deficiency n = 131		Insufficiency n = 20		Sufficiency n = 9		Total n = 160		
	No.	%	No.	%	No.	%	No.	%	
18-22yrs	26	19.8	6	30	1	11.1	33	20.6	0.430
23-28yrs	71	54.2	8	40	5	55.6	84	52.5	
29-33yrs	21	16	6	30	2	22.2	29	18.1	
34-40yrs	13	9.9	0	-	1	11.1	14	8.8	

Out of 160 subjects, 52.5% of subjects were in 23-28yrs age group, 20.6% of subjects were in 18-22yrs age group, 18.1% were in 29-33yrs age group, 8.8% were in 34-40yrs age group.

There was no statistical significant association found between age group and vitamin D level.

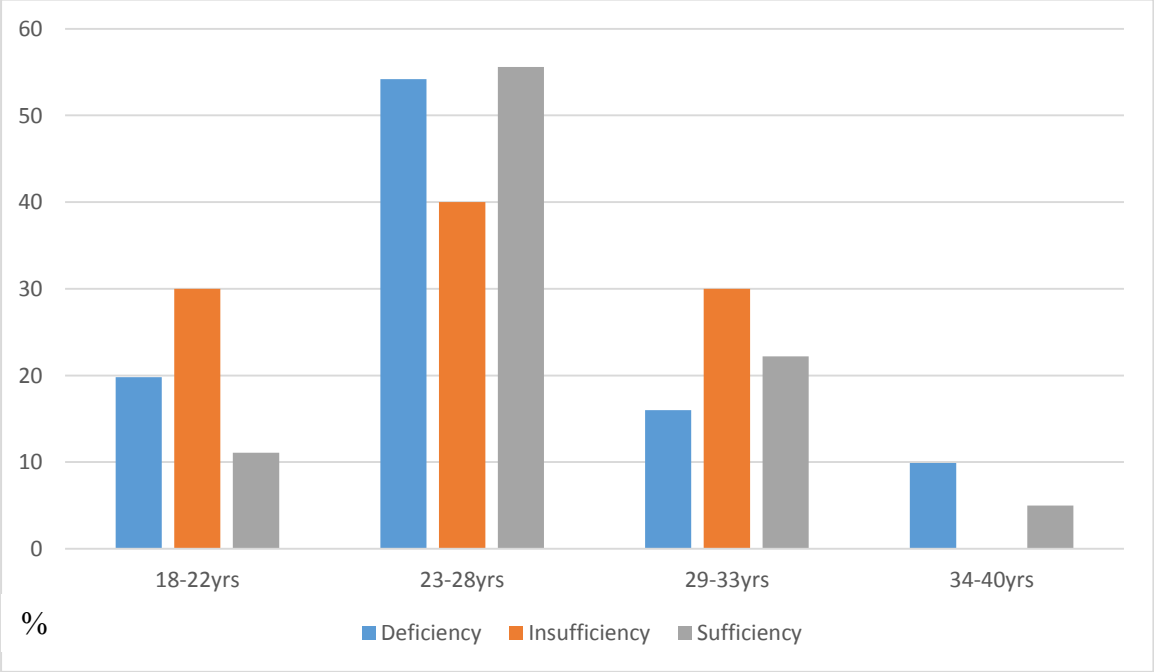

Figure 13:- Graph showing distribution of subject according to Age group and Vitamin D level.

Table 4:- Distribution of subjects according to parity and vitamin D level

Parity	Vitamin D								P Value
	Deficiency n = 131		Insufficiency n = 20		Sufficiency n = 9		Total n = 160		
	No.	%	No.	%	No.	%	No.	%	
Primigravida	112	85.5	17	85	8	88.9	137	85.6	0.982
Gravida 2	15	11.5	3	15	1	11.1	19	11.9	
Gravida 3	3	2.3	0	-	0	-	3	1.9	
Gravida 4	1	0.8	0	-	0	-	1	0.6	

Out of 160 subjects, 85.6% of subjects were primigravida, 11.6% of subjects were gravida 2, 1.9% were gravida 3, and only 0.6% were gravida 4.

There was no statistical significant association found between parity and vitamin D level.

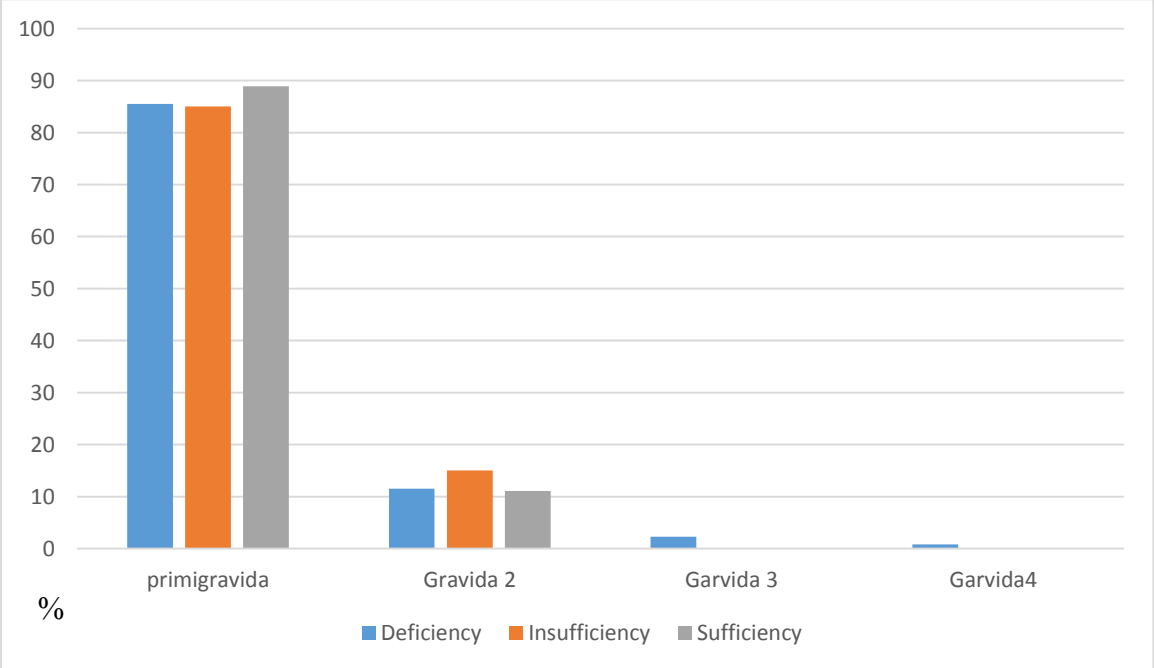

Figure 14:- Graph showing distribution of subjects according to parity and vitamin D level.

Table 5:- Distribution of subjects according to period of gestation and vitamin D level

POG	Vitamin D								P Value
	Deficiency N = 131		Insufficiency N = 20		Sufficiency N = 9		Total N = 160		
	No.	%	No.	%	No.	%	No.	%	
37 – 37+6 weeks	18	13.7	3	15	2	22.2	23	14.4	0.952
38 – 38+6 weeks	49	37.4	8	40	3	33.3	60	37.5	
39 – 39+6 weeks	59	45	9	45	4	44.4	72	45	
40 – 40+6 weeks	5	3.8	0	-	0	-	5	3.1	

There was no statistical significant difference found between period of gestation and vitamin D level.

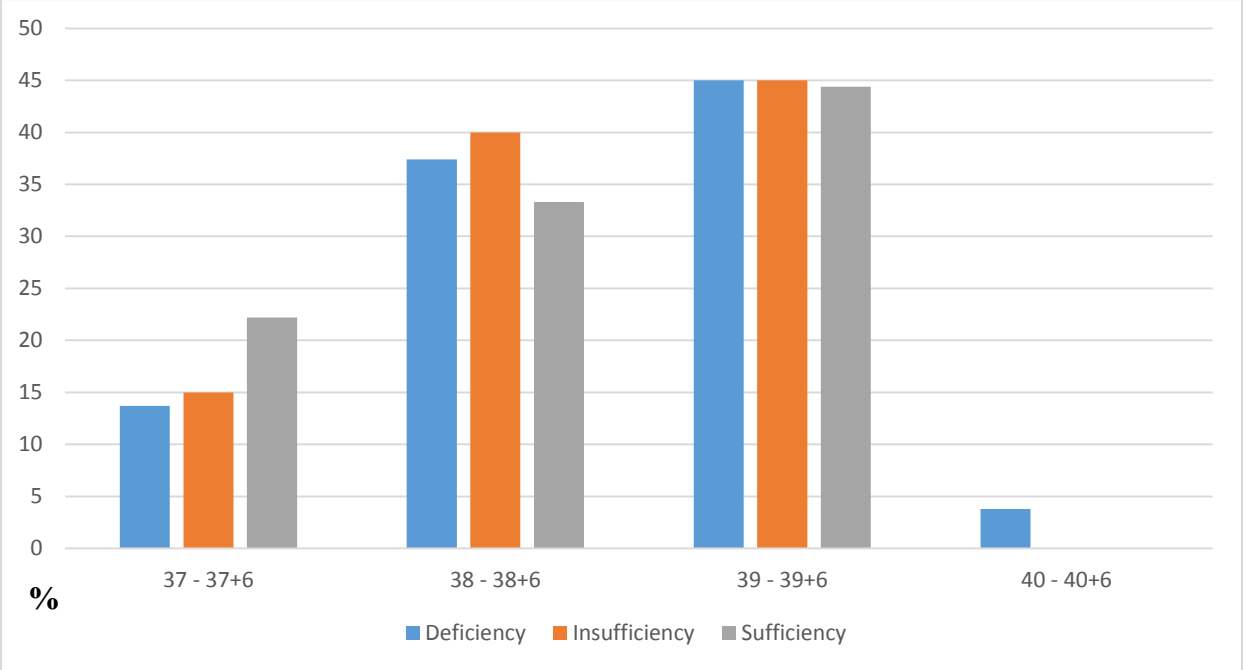

Figure 15: - Graph showing distribution of subjects according to period of gestation and vitamin D level.

Table 6:- Distribution of subjects according to BMI and vitamin D level

BMI	Vitamin D								P Value
	Deficiency N = 131		Insufficiency N = 20		Sufficiency N = 9		Total N = 160		
	No.	%	No.	%	No.	%	No.	%	
Underweight <18.5	7	5.3	2	10	0	-	9	5.6	0.747
Normal 18.5 – 24.99	121	92.4	18	90	9	100	148	92.5	
Overweight >25	3	2.3	0	-	0	-	3	1.9	

Out of 160 subjects, 92.5% of subjects had BMI in normal range, 5.6% of subjects were underweight and 1.9% of subjects were overweight. There was no statistical significant difference found between BMI and vitamin D level.

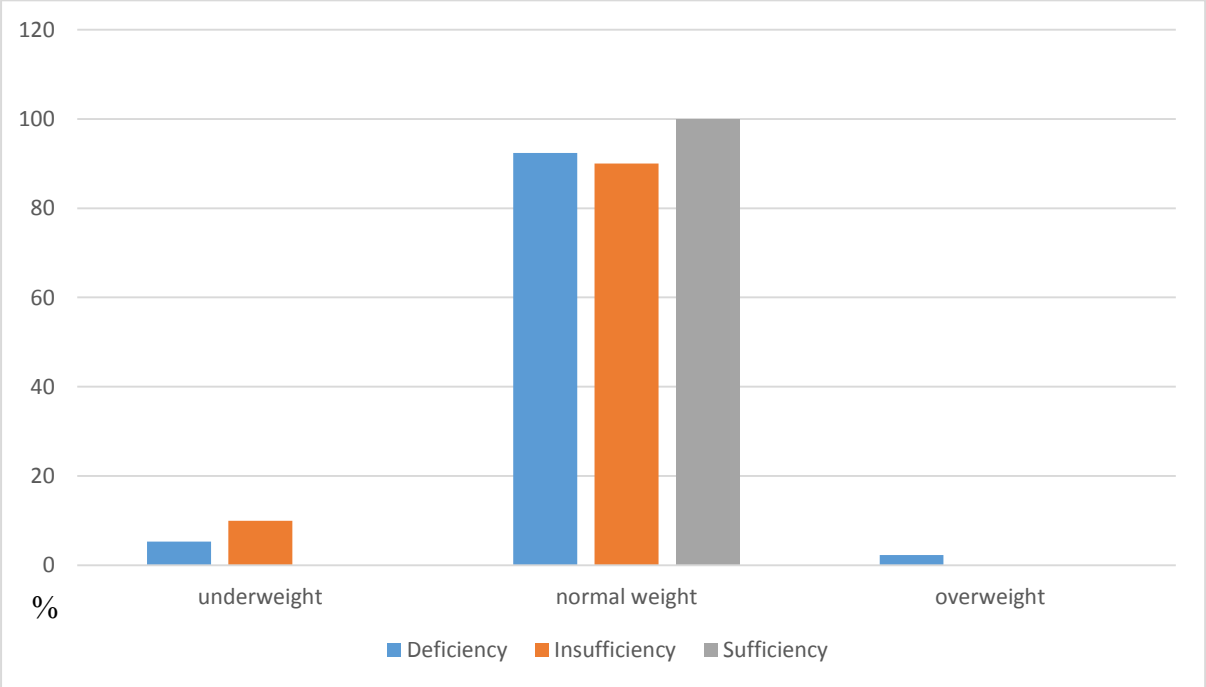

Figure 16: - Graph showing distribution of subjects according to BMI and vitamin D level.

Table 7:- Distribution of subjects according to socioeconomic status and vitamin D level

SE Status	Vitamin D								P Value
	Deficiency N = 131		Insufficiency N = 20		Sufficiency N = 9		Total N = 160		
	No.	%	No.	%	No.	%	No.	%	
Upper middle	0	-	1	5	7	77.8	8	5	<0.001
Middle class	15	11.5	13	65	2	22.2	30	18.8	
Lower middle	95	72.5	5	25	0	-	100	62.5	
Lower class	21	16	1	5	0	-	22	13.8	

In the total study population, 62.5% of subjects were in lower middle class followed by middle class consisting of 18.8% of subjects followed by lower class and upper middle class, which is 13.8% & 5% respectively.

Of the vitamin D deficient subjects (n=131), 72.5% were in lower middle class. 65.0 % of Vitamin D insufficiency subject were in middle class. 77.8% of vitamin D sufficiency subject were in upper middle. There was a statistical significant association (<0.001) found between socioeconomic status and vitamin D level.

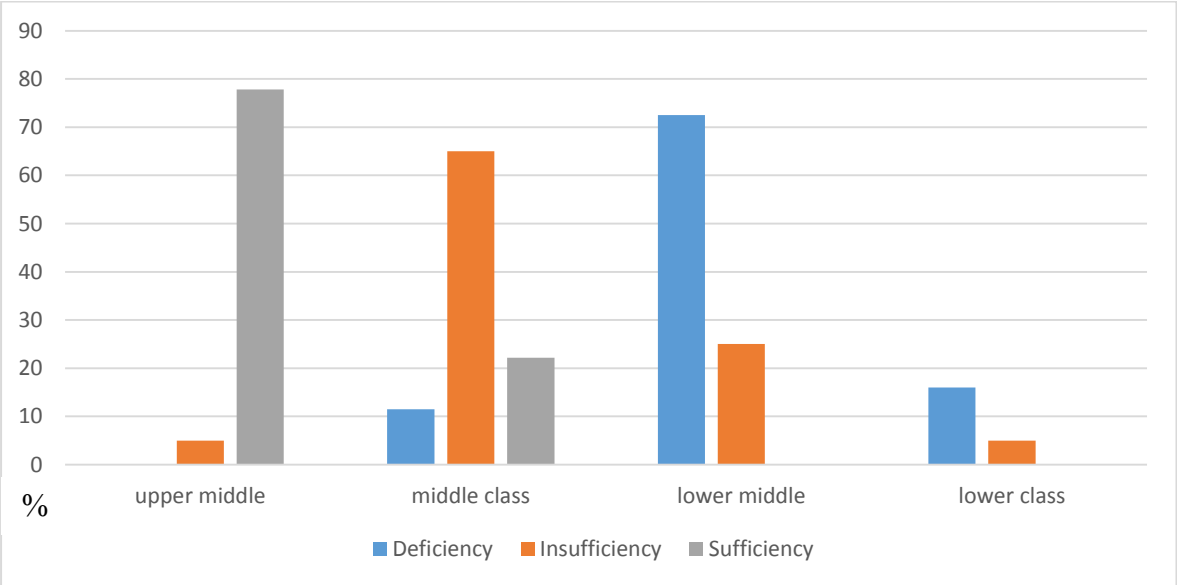

Figure 17:- Graph showing distribution of subjects according to socioeconomic status and vitamin D level.

Table 8:- Distribution of subjects according to education status and vitamin D level

Education Status	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
Uneducated	7	5.3	2	10	1	11.1	10	6.3	0.28
Primary school	63	48.1	11	55	2	22.2	76	47.5	
High school	58	44.3	7	35	4	44.4	69	43.1	
Graduated	3	2.3	0	-	2	22.2	5	3.1	

Out of 160 subjects, 47.5% of subjects had studied up to primary school, 43.1% of subjects had studied up to high school, 6.3% of subjects were uneducated and only 3.1% of subjects were graduates.

There was no statistical significant difference found between education status and vitamin D level.

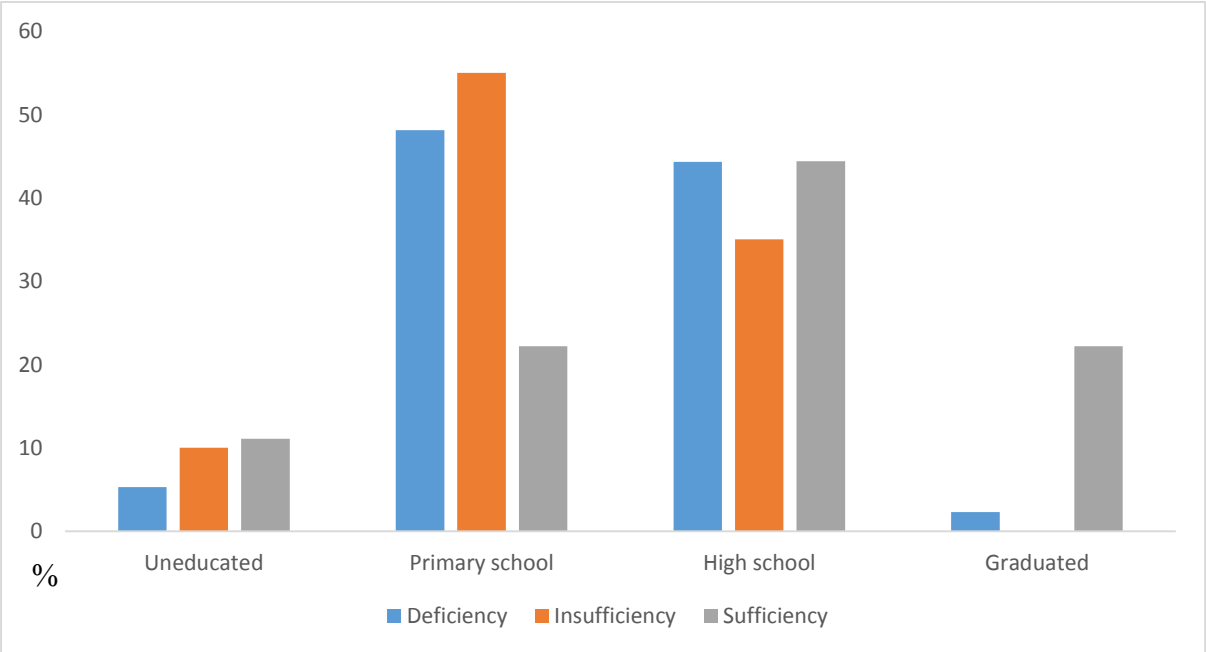

Figure 18:- Graph showing distribution of subjects according to education status and vitamin D level.

Table 9:- Distribution of subjects according to mode of delivery and vitamin D level

Mode of delivery	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
Vaginal	10	7.6	14	70	8	88.9	32	20	<0.001
LSCS	121	92.4	6	30	1	11.1	128	80	

Of the total study population, 80 % of subjects underwent LSCS and 20% of subjects had vaginal delivery.

70% of vitamin D insufficiency subjects had vaginal delivery, 88.9 % of vitamin D sufficiency subjects had vaginal delivery and only 7.6% of Vitamin D deficiency subjects had vaginal delivery rest 92.4% of Vitamin D deficiency subjects underwent LSCS . There was a statistical significant association (<0.001) found between mode of delivery and vitamin D level.

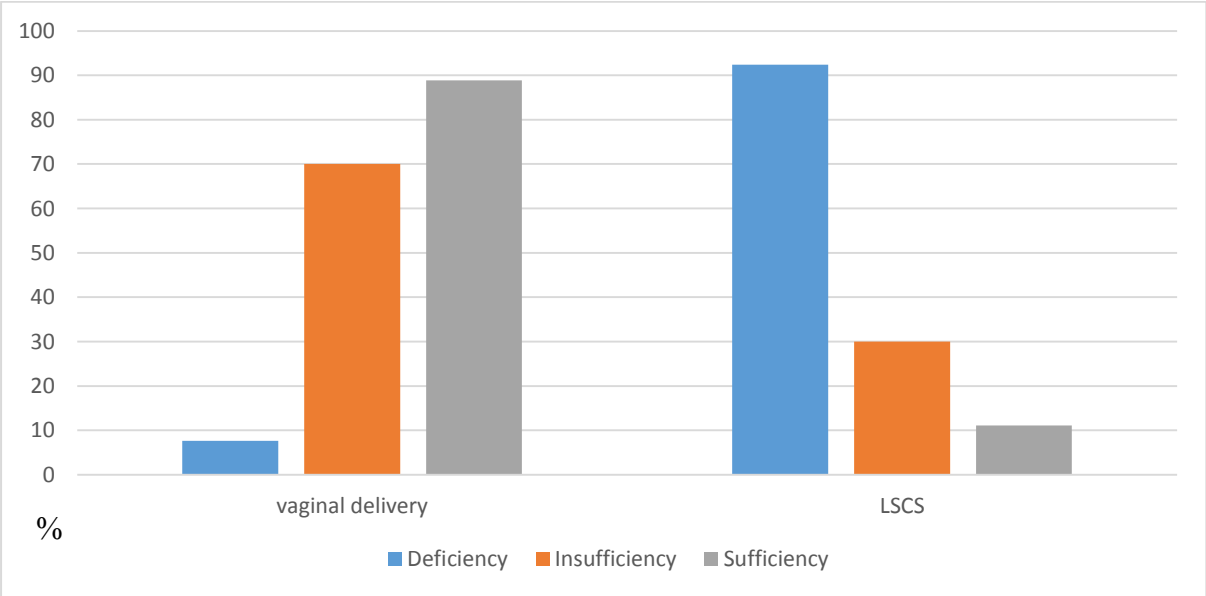

Figure 19:- Graph showing distribution of subjects according to mode of delivery and vitamin D level**.**

Table 10:- Distribution of subjects according to indication for LSCS and vitamin D level

Indication for LSCS	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
Fetal distress	81	61.8	12	6	4	44.4	97	60.6	<0.001
Nonprogression of labour	18	13.7	4	2	1	11.1	23	14.3	
Placenta previa	5	3.8	1	0.5	3	33.3	9	5.6	
Malpresentation	5	3.8	3	2.2	1	11.1	9	5.6	
Maternal medical disease	22	16.7	0	-	0	-	22	13.7	

Of the vitamin D deficient subjects who underwent LSCS, 61.8% had fetal distress, 13.7% had non progression of labour, 3.8% had placenta previa and another 3.8% had malpresentation and 16.7% had maternal medical disease. There was a statistically significant association between indication for LSCS and vitamin D levels (p=0.001).

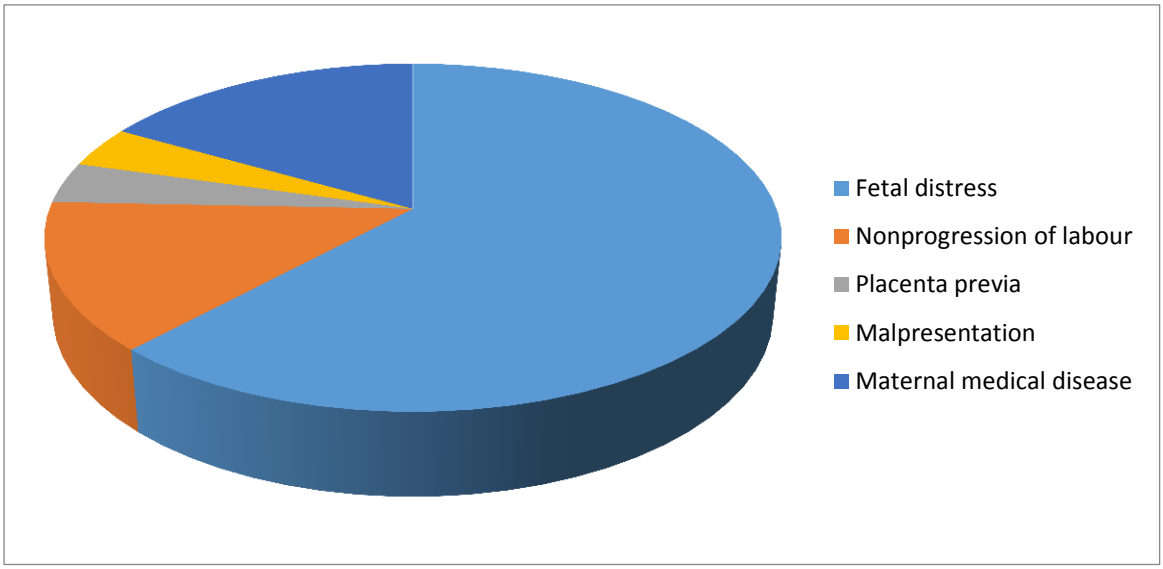

Figure 20:- Graph showing distribution of vitamin D deficient subjects according indication for LSCS.

Table 11:- Distribution of subjects according to birth weight and vitamin D level in mothers

Birth weight	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
1.5-2.5kg	25	19.1	0	-	0	-	25	15.6	<0.001
2.6-3.5kg	105	80.2	17	85	5	55.6	127	79.4	
>3.5kg	1	0.8	3	15	4	44.4	8	5	

Among 160 newborn babies, 25 newborn babies were LBW (1.5-2.5kg), 127 babies were normal birth weight (2.5-3.5kg) and eight babies were > 3.5kg. All the mothers of 25 LBW babies had Vitamin D deficiency(100%). There was a statistical significant association (<0.001) found between birth weight and vitamin D level in mothers.

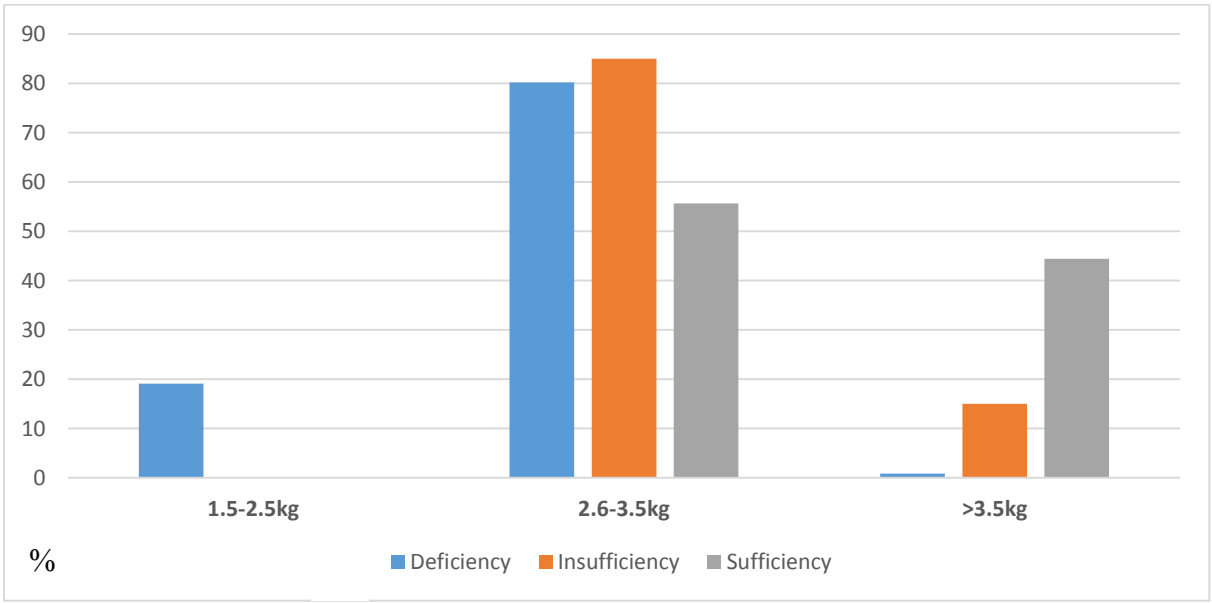

Figure 21:- Graph showing distribution of subjects according to birth weight and vitamin D level in mothers.

Table 12:- Distribution of subjects according to NICU admission of baby and vitamin D level in mothers

NICU Admission	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
Yes	14	10.7	1	5	1	11.1	16	10	0.727
No	117	89.3	19	95	8	88.9	144	90	

Out of 131 babies born to vitamin D deficient mothers, only 10.7% were admitted in NICU and the rest 89.3% were mother's side.

There was no statistical significant difference found between NICU admission of baby and vitamin D level in mothers.

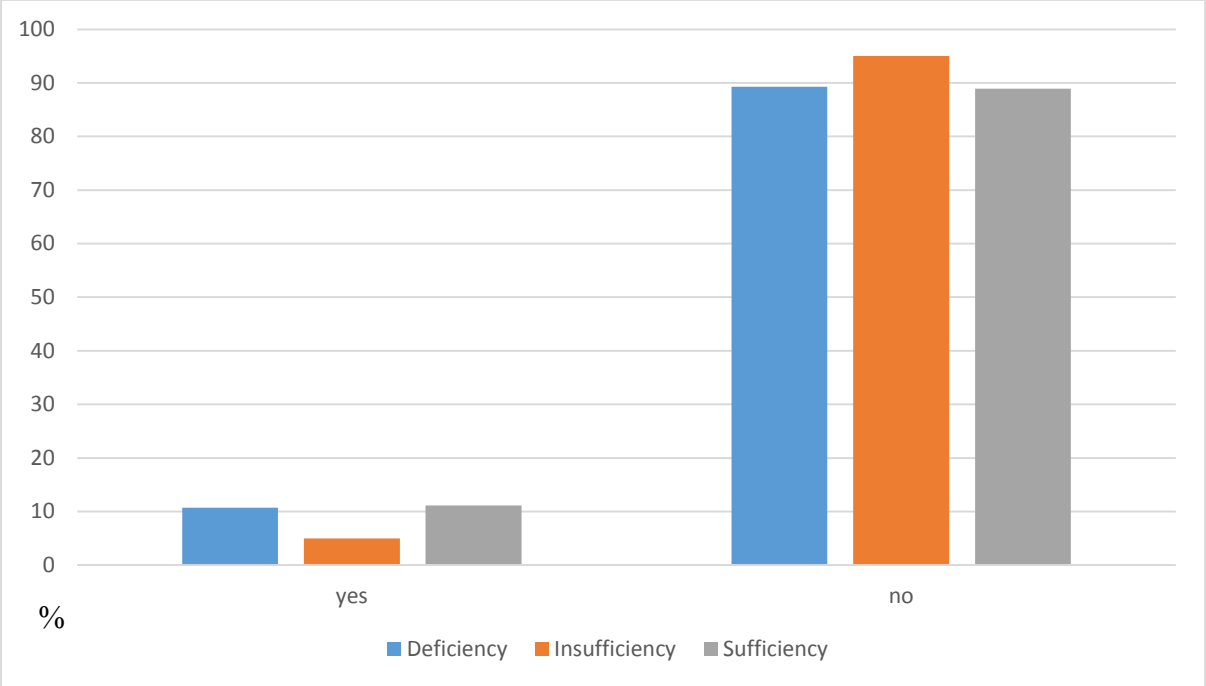

Figure 22:- Graph showing distribution of subjects according to NICU admission of baby and vitamin D level in mothers.

Table 13:- Distribution of subjects according to neonatal complication and vitamin D level in mothers

Neonatal complication	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
NIL	116	88.5	19	95	8	88.9	143	89.4	0.959
SGA	2	1.5	0	-	0	-	2	1.3	
IUGR	3	2.3	0	-	0	-	3	1.9	
Respiratory Distress	10	7.6	1	5	1	11.1	12	7.5	

89.4% of babies didn't have any complication, 7.5% of babies had respiratory distress, 1.9% of babies had IUGR and 1.3% of babies had SGA.

There was no statistical significant difference found between neonatal complication and vitamin D level in mothers.

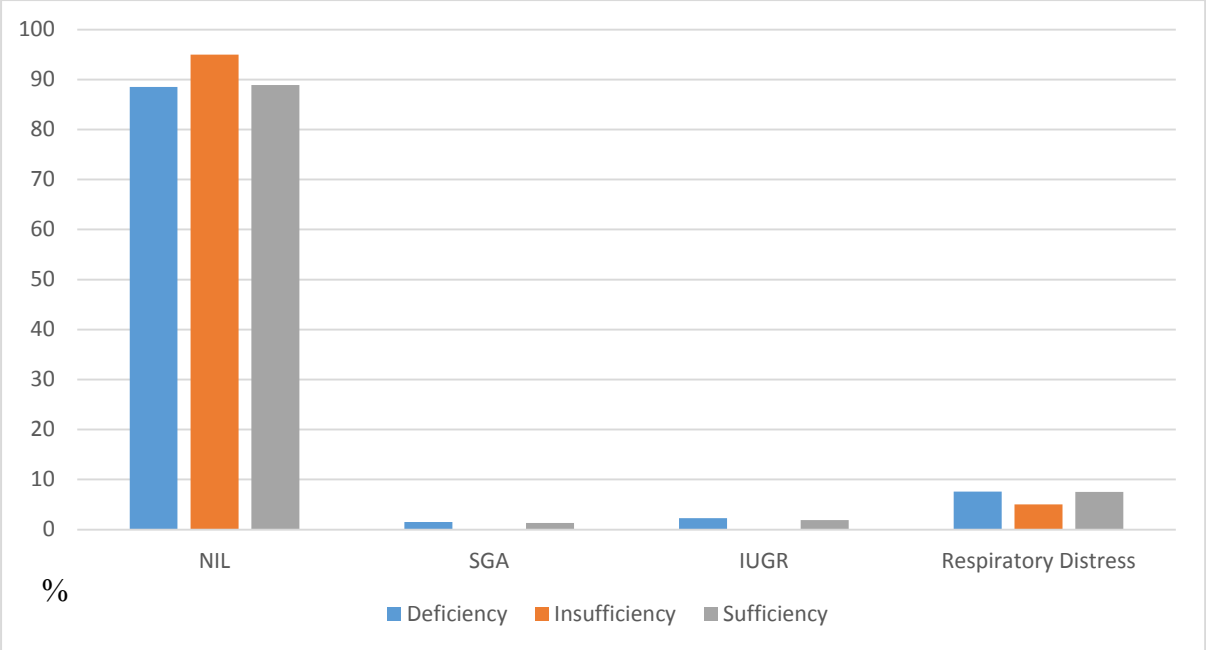

Figure 23:- Graph showing distribution of subjects according to neonatal complication and vitamin D level in mothers.

Table 14:- Distribution of subjects according to maternal complication and vitamin D level.

Maternal complication	Vitamin D								P Value
	Deficiency N=131		Insufficiency N=20		Sufficiency N=9		Total N=160		
	No.	%	No.	%	No.	%	No.	%	
NIL	103	78.6	17	85	9	100	129	80.6	0.908
PE	22	16.8	3	15	0	-	25	15.6	
Eclampsia	3	2.3	0	-	0	-	3	1.9	
GDM	2	1.5	0	-	0	-	2	1.3	
Anemia	1	0.8	0	-	0	-	1	0.6	

Out of 160 subjects, 80.6% did not have any complication, 15.6% of subjects had PE, 1.9% of subjects had eclampsia, 1.3% of subjects had GDM and 0.6% of subjects had anemia.

There was no statistical significant association found between maternal complication and vitamin D level.

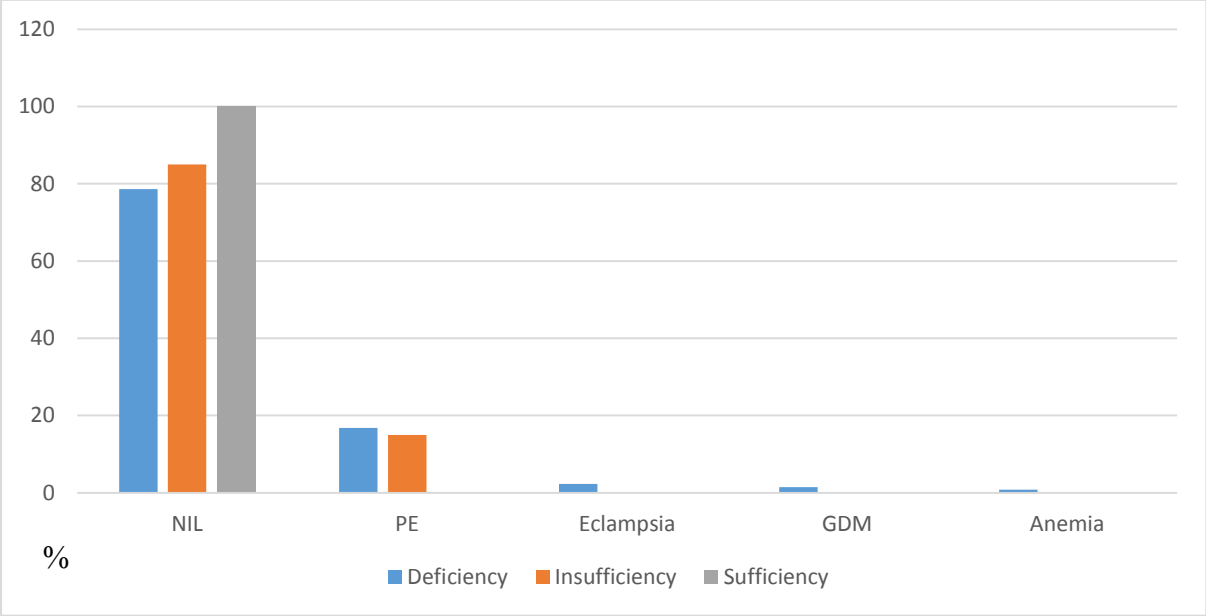

FIgure 24:- Graph showing distribution of subjects according to maternal complication and vitamin D level

DISCUSSION

Vitamin D is a prohormone which affects established physiological pathways including inflammation, immunomodulation, and transcription of genes involved in placental function.[67]

As the majority of female population residing in rural area of Kolar which comprises of women who are mostly housewives and spend most of their time indoors and are unaware of minimal adequate antenatal care, hence risk of hypovitaminosis D is high, which was reported in this study.

In this present study, majority (52.5%) of the study population belonged to the age group of 23 – 28 years and 85.6% were primigravida with 92.5% of the cases having a normal BMI of 18.5 to 24.99 Kg/m^2.

Serum vitamin D levels in term pregnancy

Studies	Place	Mean serum vitamin D levels	Percentage of vitamin D deficient population
1. Domaracki P et al – 2016[68]	Poland	18.20ng/dl	45.6%
2. Dawodu A et al – 2016[69]	Qatar	19.1ng/dl	78%
3. Pratumvinit B et al – 2015[70]	Bangkok	12.2ng/dl	75.5%
4. Liu Y et al – 2017[43]	Beijing	11.61ng/dl	20.4%
5. Lindqvist P et al – 2016[73]	Sweden	13.6ng/dl	54.9%
6. Amegah A et al, – 2017[4]	Ghana	17.2ng/dl	83%
7. Current study		11.22ng/dl	81.7%

In the present study, majority (81.87%) of cases had vitamin D deficiency (serum vitamin D concentration of < 20ng/dl) and 12.50% of cases had vitamin D insufficiency (20-29ng/dl) and only 5.63% of cases had sufficient vitamin D levels of >30ng/dl.

In a similar study done by Domaracki et al in Polish women showed only 10.8% of cases had sufficient serum concentrations of 25(OH)D, while 43.7% had vitamin D deficiency and 45.6% had insufficient serum vitamin D levels.[68]

In another study done by Dawodu et al in Qatar, a total of 47 (78%) mothers were vitamin D-deficient.[69]

In yet another study conducted on pregnant women in Thailand, the prevalence of hypovitaminosis D at delivery was 75.5%. Of these, 41.5% were vitamin D insufficienct and vitamin D deficiency was found in 34.0% of women.[70]

In different study done in Beijing, China by Liu Y et al in 2016, they found that, of the total ninety-eight patient, there were twenty patients whose serum levels of total 25-OH Vitamin D was lower than 50 nmol/L which was considered as Vitamin D deficiency.[41]

In another study done in Sweden by Lindqvist et al, the mean 25-OH vitamin D levels in women who underwent caesarean delivery due to suspected asphyxia was 43.6±18 nmol/L, which was significantly lower than in controls (p=0.04).[73]

Amegah et al observed in his study done in Ghana, that 83% of the study population had serum 25(OH)D levels <75 nmol/l which was considered as vitamin D deficiency.[4]

These are a few studies done in different parts of the world and all these studies showed results comparable with our study. From this observation it is evident that population of both, tropical and non tropical countries were largely vitamin D deficient

Parity and vitamin D deficiency

In our study 85% of the study population were primigravida, but there was no statistically significant difference in serum vitamin D concentrations and parity. In a study by Sachan et al also the study population had lower parity.[5]

Socioeconomic status

The study subjects were categorized into different socioeconomic class based upon modified BG Prasad classification. In this study, 62.5% of subject were in lower middle class followed by middle class consisting of 18.8% of subject followed by 13.8% & 5% in lower class and upper middle class, respectively. It was observed that 72.5% of Vitamin D deficienct subjects were in lower middle class. 65.0 % of Vitamin D insufficiency subject were in middle class. 77.8% of vitamin D sufficiency subject were in upper middle .There was a statistical significant difference (p=<0.001) found between socioeconomic status and vitamin D level.

In a similar study conducted by Rodriquez et al in a South European population, higher risk of vitamin D deficiency was related to lower social class (RR = 1.94, 95% CI 1.19, 3.16) which was comparable to our study.[71]

Whereas in 2 other studies conducted by Pratumvinit et al and Veena et al, it was observed that maternal serum vitamin D levels were negatively associated with socioeconomic status and education level, which are not in agreement with the present study.[70,72]

Mode of deliery

In our study, 80 % of subject underwent LSCS and 20% of subject had vaginal delivery. Only 7.6% of Vitamin D deficiency subject had vaginal delivery rest 92.4% of Vitamin D

deficiency subject underwent LSCS . Thus there was a statistical significant association (p=<0.001) found between LSCS and vitamin D deficiency.

In a study by Lindqvist et al, it was seen that women who underwent emergency caesarean section due to suspected fetal asphyxia had lower vitamin D levels in pregnancy which was similar to our study results.[73]

Baby birth weight and maternal vitamin D

In our study group of 160 newborn babies, 25 newborn babies were LBW (1.5-2.5kg), 127 babies were 2.5-3.5kg and eight babies were > 3.5kg. All the mothers of 25 LBW babies had Vitamin D deficiency which was statistically significant (p=<0.001).

In 2 similar studies conducted by Bowyer et al and Leffelaar et al, infants of mothers with 25-OH D deficiency during pregnancy had lower birth weight. These results were comparable with our study.[74,75]

In a study by Domaracki et al, serum levels of 25(OH)D did not correlate significantly with neonatal birth weight, as opposed to the results of the present study.[68]

Neonatal complications

In the present study, there was no statistical significant association found between neonatal complication such as small for gestational age, low birth weight and respiratory destress with vitamin D level in mothers.

In a study by Aghajafari et al showed a significant association between small for gestational age infants and 25-OHD insufficiency compared with the comparison group, which is not according to our study results.[16]

In another study conducted by Ataseven et al, respiratory distress syndrome was more common with severe (28 %) compared to mild-moderate 25(OH)D deficiency (14 %) ($p < 0.05$).[76]

Maternal complications

In the present study 80.6% of subjects did not have any maternal complications and there was no significant association seen between maternal complications like preeclampsia, eclampsia, GDM and anemia and serum vitamin D levels.

In the study of Domaracki et al, evident statistically significant differences ($p = 0.0021$) in serum concentrations of 25(OH)D in women with PE and healthy controls were noted. Women with PE had vitamin D deficiency. This was not consistent with our study. But 25(OH)D was not identified as a significant predictor of GDM in this study, which was consistent with our study.[68]

In Mirzakhani et al study, women who had sufficient vitamin D levels (≥30 ng/ml) in early and late pregnancy had a significantly lower rate of preeclampsia compared with women with insufficient vitamin D levels ($P = 0.04$). This was not comparable to our results.[77]

In a study by Nicholas et al, they found no association between hypovitaminosis D (plasma levels of <30 ng/ml) and preeclampsia, intrauterine growth restriction or gestational diabetes, which was consistent with our study results.[78]

Strengths:

- Maternal vitamin D was analyzed by serum 25 (OH) D which accounts for vitamin D that is synthesized in the skin and obtained from the diet.

- In this study we have excluded all the absolute indications for ceasarean section and preexisting maternal medical conditions, thus removing most of the confounding bias.

Limitations:

- Maternal vitamin D status was assessed late in pregnancy with an assumption that they were a representative of the vitamin D status early in pregnancy.
- Due to time constraint, seasonal variations in vitamin D and long term complications of hypovitaminosis could not be evaluated.
- Controls were not included in the study.

SUMMARY

The present study entitled –EVALUATION OF VITAMIN D LEVELS IN TERM PREGNANCY AND ITS OBSTETRIC OUTCOME IN INDIAN WOMEN.” was conducted at department of Obstetrics and Gynaecology, R.L. Jalappa Hospital and Research Centre attached to Sri Devaraj Urs Medical College, Kolar from March 2014 to August 2015.

A total number of 160 samples were collected, data was analysed and summarized as:

- Majority of the subjects were vitamin D deficient (81.87%) and 12.5% were vitamin D insufficient and only 5.63% were vitamin D sufficient.
- Majority of the cases were in the age group of 23-28 years (52.5%) with a normal BMI (18.5 to 24.99 kg/m^2)
- High prevalence of vitamin D deficiency was seen in lower middle socioeconomic class (62.5%).
- Low vitamin D level during pregnancy was associated with higher rates of ceasarean section (92.4%).
- There was a significant association between indication for ceasarean section (fetal distress) and serum vitamin D levels (61.8%).
- Maternal vitamin D deficiency was associated low birth weight of neonates (100%).
- Vitamin D deficiency was not associated with any adverse maternal outcome.

CONCLUSION

In the present study, majority of women had deficient serum vitamin D levels. Most of these vitamin D deficient women belonged to the lower socioeconomic strata. A significant number of vitamin D deficient pregnant women developed fetal distress at term, which raised the cesaerean section rates in the vitamin D deficient group. Most of the neonates born to vitamin D deficient mothers were of low birth weight.

As there is a high prevalence of vitamin D deficiency worldwide and screening for maternal serum vitamin D levels is not proven to be cost effective, we conclude that, there is a need for vitamin D supplementation for all pregnant women as a routine antenatal practice.

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CASE PROFORMA

NAME: IP NO:
AGE: DOA:
OCCUPATION: DOD:
ADDRESS:
EDUCATION:
HUSBANDS OCCUPATION:
SOCIOECONOMIC STATUS:

CHIEF COMPLAINTS:

HISTORY OF PRESENT ILLNESS:

OBSTETRIC HISTORY:
Marital life: Consanguinity:
Gravida: Para: living: Abortion: Dead:
Details of previous pregnancy:
Details of present pregnancy:

MENSTRUAL HISTORY:
Last menstrual period: Age of menarche:
Expected delivery date:
Period of gestation:
Period of gestation according to early scan:
Past menstrual cycles:

PAST HISTORY:
HTN/DM/BA/TB/blood dyscrasias/epilepsy/thyroid disorder/cardiac disease
H/O blood transfusions:
H/O Surgeries or hospitalization:

PERSONAL HISTORY:
Sleep and appetite:
Diet:
Bowel and bladder:

FAMILY HISTORY:

GENERAL EXAMINATION:
General condition: Fair/ moderate/ Poor
Built: Nourishment:
Ht: cms Wt: kgs BMI:
Pallor: Icterus:
Cyanosis: Clubbing:
Lymphadenopathy: Edema:

VITALS:
Pulse rate:
Respiratory rate:
Blood pressure
Temperature:

SYSTEMIC EXAMINATION:
Cardiovascular system:
Respiratory system:
Central nervous system:

Per abdomen: Uterus size:
Relaxed / Irritable / Acting
Presentation: cephalic/ Breech/ other
FHS:
Per vaginum: Effacement:
Dilatation:
Station:
Membranes:
Pelvis:

PROVISIONAL DIAGNOSIS:

INVESTIGATIONS:
Blood group and Rh typing:
CBC: HB: HIV:
PCV: HbsAG:
RBC: VDRL:

WBC:
PLT:
RBS:
Urine analysis: Albumin-
Sugar-
Microscopy-
Serum VITAMIN D level
OBSTETRICS SCAN:

DELIVERY DETAILS:
Mode of delivery: Vaginal delivery/ Caesarean section

VAGINAL-
Spontaneous /induced :
CAESAREAN-
Indication:

DETAILS OF NEONATE:
Sex: Date: Time:
Birth weight:
APGAR : 1'- 5'-
Admission to NICU:

MATERNAL COMPLICATIONS:
Hypertension
Convulsions
Premature rupture of membranes
Antepartum haemorrhage
Postpartum haemorrhage

FETAL COMPLICATIONS:
Low birth weight
Small for gestational age
Respiratory distress
Admission to NICU

CONDITION AT DISCHARGE:

SRI DEVARAJ URS MEDICAL COLLEGE & RESEARCH CENTRE, TAMAKA, KOLAR

PATIENT CONSENT FORM

Case no:

I have read the foregoing information, or it has been read to me. I have had the opportunity to ask questions about it and any questions that I have asked have been answered to my satisfaction. I have understood that I have the right to refuse consent or withdraw it at any time during the study and this will not affect my treatment in any way. I consent voluntarily to participate in this study –**EVALUATION OF VITAMIN D LEVELS IN TERM PREGNANCY AND ITS OBSTETRIC OUTCOME IN INDIAN WOMEN."**

Name of Participant_____

Signature/ thumb print of Participant _____

Date _____

Statement by the researcher/person taking consent:

I have accurately read out the information sheet to the potential participant, and to the best of my ability made sure that the participant understands that the following will be done:

1. Blood sample will be taken from the patient for serum vitamin D analysis.

I confirm that the participant was given an opportunity to ask questions about the study, and all the questions asked by the participant have been answered correctly and to the best of my ability. I confirm that the individual has not been coerced into giving consent, and the consent has been given freely and voluntarily.

Name of Researcher/person taking the consent_____

Signature of Researcher /person taking the consent_____

Date _____

Name and Address of Principal Investigator: Dr. Shilpa Suri Ciryam
R.L Jalappa Hospital
Tamaka, Kolar.

ತಿಳುವಳಿಕೆಯ ಒಪ್ಪಿಗೆ ಪತ್ರ

ಅಧ್ಯಯನ ಶೀರ್ಷಿಕೆ:- **"ಇಗಿಂಐಗಖಪಿಖಔಓ ಔಈ ಗಿಖಪಿಂಒಖಓ ಆ ಐಇಗಿಇಐಖ ಖಓ ಪಿಇಖಒ ಕಖಇಗಓಂಓಅಜ ಂಓಆ ಖಪಿಖ ಔಃಖಖಿಇಪಿಖಖಅ ಔಗಖಿಅಔಒಇ ಖಓ ಖಓಆಖಂಓ ಷಔಒಇಓ."**

ಶ್ರೀ/ಶ್ರೀಮತಿ_____ಆದ ನಾನು ಈ ಮೇಲಿನ ಸಂಶೋಧನ ವಿಷಯದ ಬಗ್ಗೆ ನನಗೆ ಅರ್ಥವಾಗುವ ರೀತಿಯಲ್ಲಿ ನನ್ನದೇ ಭಾಷೆಯಲ್ಲಿ ತಿಳಿಸಿರುತ್ತಾರೆ. ಈ ಸಂಶೋಧನಾ ವಿಷಯದಲ್ಲಿ ನಾನು ಒಬ್ಬ ವಿಷಯಿಯಾಗಿ ಭಾಗವಹಿಸಲು ನನ್ನ ಸಂಪೂರ್ಣವಾಗಿ ಒಪ್ಪಿಗೆ ಇರುತ್ತದೆ. ಈ ಸಂಶೋಧನಾ ಉದ್ದೇಶವನ್ನು ಪೂರ್ಣವಾಗಿ ಅರಿತಿರುತ್ತೇನೆ. ಮತ್ತು ಈ ಸಂಶೋದನೆಗೆ ಯಾವುದೇ ಪ್ರಯೋಗಾಲಯದ ಪರೀಕ್ಷೆಗಳು, ಔಷಧಿಗಳನ್ನು ಒಳಗೊಂಡಿರುವುದಿಲ್ಲ. ಈ ಸಂಶೋಧನೆಗೆ ನನ್ನಿಂದ ಯಾವುದೇ ಆರ್ಥಿಕತೆಯ ಅವಶ್ಯಕತೆ ಇರುವುದಿಲ್ಲ. ನಾನು ಯಾವುದೇ ಸಮಯದಲ್ಲಿ ನನ್ನ ಸಹಕಾರವನ್ನು ಹಿಂಪಡೆದು ಈ ಸಂಶೋಧನೆಯಿಂದ ಹೊರಹೋಗುವ ಹಕ್ಕನ್ನು ಹೊಂದಿರುತ್ತೇನೆ. ಇದರಿಂದ ನನ್ನ ಚಿಕಿತ್ಸೆಗೆ ಯಾವುದೇ ರೀತಿಯ ತೊಂದರೆಯಾಗುವುದಿಲ್ಲ. ಮುಖ್ಯವಾಗಿ ನನ್ನಿಂದ ಪಡೆದ ಈ ಮಾಹಿತಿಯು ಸಂಶೋಧನೆಗೆ ಮಾತ್ರ ಸೀಮಿತವಾಗಿರುತ್ತದೆ. ಮತ್ತು ಈ ಮಾಹಿತಿಯು ಎಲ್ಲೂ ಸೋರಿಕೆಯಾಗದಂತೆ ಎಚ್ಚರಿಕೆ ವಹಿಸುವುದಾಗಿ ತಿಳಿವಳಿಕೆ ನೀಡಿರುತ್ತಾರೆಂದು ನಾನು ದೃಢಪಡಿಸಿಕೊಂಡು ಒಪ್ಪಿಗೆ ನೀಡಿರುತ್ತೇನೆ.

ರೋಗಿಯ ಸಹಿ/ ಸಾಕ್ಷಿ ಸಹಿ. ಸಂಶೋದಕನ ಸಹಿ

ಬೆರಳಚ್ಚು.

KEY TO MASTERCHART:

A. Ip no - IP number

B. Age

C. Parity –
   1. primigravida
   2. gravida 2
   3. gravida 3
   4. gravida 4

D. POG - Period of gestation

E. BMI – body mass index
   1.underweight- <18.5
   2. normal- 18.5-24.99
   3. overweight - >25
   4. obese - >30

F. SE status – socio economic status
   1. class 1 upper class
   2. class 2upper middle
   3. class 3 middle class
   4. class 4 lower middle
   5. class 5 lower class

G. Edu status – education status
   1. uneducated
   2. primary school
   3. high school
   4. graduate

H. MOD – mode of delivery
   1. vaginal delivery
   2. lscs

I. Indication – indication for LSCS
   1. fetal distress
   2. non progression
   3. placenta previa
   4. malpresentation
   5. maternal medical disease

J. Bt wt – birth weight
   1. 1.5 to 2.5kg - LBW
   2. 2.5 to 3.5kg
   3. >3.5kg

K. Baby sex
   1. male
   2. female

L. Apgar 1 – apgar score at 1 min
   1. 5/10
   2. 6/10
   3. 7/10

M. Apgar 5 – apgar score at 5 min
   1. 7/10
   2. 8/10
   3. 9/10

N. NICU – neonatal ICU admission
   1. yes
   2. no

O. Neon comp – neonatal complications
   1. SGA
   2. IUGR
   3. resp distress
   4. MSAF
   5. mortality

P. Mat comp – maternal complications
   1. PE
   2. eclampsia
   3. GDM
   4. anemia

Q. Vit D – vitamin D
   1. Defficiency - <20
   2. Insuffiency - 21 - 29
   3. Sufficiency - >30
   4. Intoxication - >150

ip no	age	parity	POG	BMI	SE status	edu stat	MOD	indication	bt wt	baby sex	apgar 1	apgar 5	nicu	neon comp	mat comp	vit D
295943	20	1	39+1	2	4	3	2	1	2.25	2	3	3	2	0	0	2.51
263679	23	1	39+3	2	4	3	2	1	2.51	2	3	3	2	0	0	6.28
300548	25	1	39	2	4	3	2	1	2.3	2	3	3	2	0	0	2.09
211856	29	1	38+4	2	4	2	2	2	2.56	1	3	3	2	0	0	16.25
263587	23	1	39+2	2	4	3	2	1	2.5	2	3	3	2	0	0	4.42
301052	19	2	38	2	4	2	2	5	2	1	3	3	2	0	1	0.5
348106	24	1	38+2	2	5	3	2	1	3.15	2	3	3	2	0	0	1.65
312412	23	1	39+3	2	4	2	1	1	3	1	3	3	2	0	0	2.56
301476	27	1	37+6	1	4	2	1	5	1.98	2	1	1	1	3	1	1
293237	28	1	38+5	2	4	1	2	2	2.8	2	3	3	1	3	0	6.28
219183	21	1	38+2	2	3	3	1	5	2.76	2	3	3	2	0	1	1.65
268932	34	1	39+3	2	5	3	2	1	3.25	2	3	3	2	0	0	9.07
302182	25	2	37+5	2	4	3	1	5	3.5	1	3	3	2	0	2	9.07
302736	25	2	39+5	2	4	2	2	5	2.5	1	3	3	2	0	1	0.05
302365	24	2	39+4	2	4	3	2	1	2.6	1	3	3	2	0	0	7.21
345656	22	1	39+5	3	5	2	2	1	3.2	1	3	3	2	0	0	6.28
347129	23	1	40	2	3	2	2	1	3.4	1	2	3	2	0	0	7.49
347223	35	1	38+6	2	5	3	2	5	2.2	2	3	3	2	0	1	0.25
347128	19	1	37+5	2	4	2	2	2	2.8	2	3	3	2	0	0	22.5
347003	23	1	38+3	2	4	3	2	5	3.1	2	3	3	2	0	3	7.49
340950	23	1	39+3	2	4	3	2	1	3.24	2	3	3	2	0	0	2.09
338062	29	1	39+5	2	4	3	2	5	2.67	1	3	3	2	0	1	1.25
345820	24	1	39+5	2	5	2	1	1	2.54	1	3	3	2	0	0	2.5
348655	23	1	38+5	2	3	3	2	1	3.15	2	3	3	1	3	0	10.4
348699	34	1	39+1	2	4	3	2	1	3.5	2	3	3	2	0	0	7.49
348555	24	1	39+2	2	4	3	2	5	3.3	2	3	3	2	0	0	5.35
347107	18	1	39+2	2	4	2	2	1	3.24	1	3	3	2	0	0	10.21
348704	23	3	38+4	2	4	1	1	1	2.88	2	3	3	2	0	0	10.34
345278	25	1	38+4	1	5	3	1	1	2.9	1	3	3	2	0	0	9.07

346717	30	1	41	2	4	3	2	5	2.16	2	3	3	2	0	1	0.75
347986	25	1	37+6	2	5	3	2	2	2.6	2	3	3	2	0	0	6.28
350135	24	1	38+4	1	4	3	1	2	2.25	1	3	3	2	0	0	1.65
347117	25	1	37+5	2	5	2	2	1	2.62	2	3	3	2	0	0	7.49
350578	34	1	38+5	2	5	3	2	5	1.99	2	3	3	2	0	1	1.65
348469	26	1	38	2	4	3	2	5	2.34	2	3	3	2	0	1	7.21
349169	20	2	38+5	2	4	2	2	5	2.7	2	3	3	2	0	1	1.65
341569	26	1	39+4	2	4	3	2	1	3	1	3	3	2	0	0	9.07
348705	27	1	38+2	2	5	3	2	2	2.9	1	3	3	2	0	0	2.09
351493	21	3	39+5	2	5	3	2	4	2.83	1	3	3	2	0	0	5.35
349698	24	1	38+5	2	3	2	2	1	2.58	1	3	3	1	1	0	7.21
349774	31	2	38+3	2	4	3	2	5	2.15	2	1	1	1	3	2	0.07
350193	26	2	38+2	2	4	2	2	5	2.1	2	3	3	2	0	1	0.25
348624	25	2	39+4	2	4	3	2	1	2.8	2	3	3	2	0	0	5.35
343227	29	1	39+4	3	4	3	2	1	3.15	1	3	3	2	0	0	8.41
352407	22	1	39	2	5	1	2	1	2.77	1	3	3	2	0	0	6.28
348019	25	1	39+5	2	5	3	2	2	2.8	1	3	3	2	0	0	7.21
345278	26	1	37+4	2	4	3	2	2	3	1	3	3	2	0	0	5.35
337364	23	1	39+4	2	3	4	2	5	3.12	2	3	3	2	1	0	4.42
348203	24	1	39+3	2	4	3	2	1	2.22	2	3	3	2	0	3	1.25
351332	19	1	39+1	2	4	3	2	1	3.2	2	3	3	2	0	0	7.49
350994	30	1	39+3	2	4	2	2	1	2.8	2	3	3	2	0	0	4.42
352478	25	1	39+2	2	3	3	2	1	2.98	1	3	3	1	2	0	7.21
352302	20	1	38+2	2	4	2	2	1	2.75	2	3	3	2	0	0	7.21
329760	24	1	39	2	4	2	2	4	2.75	1	3	3	1	2	0	8.41
340985	23	2	38+3	2	4	2	2	1	3.25	1	3	3	2	0	0	9.07
351355	20	4	38	2	5	2	2	1	3.15	1	3	3	2	0	0	5.35
352978	30	1	37+5	1	4	2	2	1	3.32	1	3	3	2	0	0	4.42
341256	34	1	38+5	2	4	2	2	5	1.9	1	3	3	2	0	1	0.56
352495	21	1	38+4	2	4	3	2	1	2.6	2	3	3	2	0	4	9.07
352404	25	1	38+5	2	3	2	2	4	2.77	2	3	3	2	0	0	6.28
352996	34	1	38+5	2	4	2	2	1	2.8	2	3	3	2	0	0	10.36
353015	25	1	38+1	2	4	2	2	1	2.76	1	3	3	2	0	0	7.21

332977	24	1	39+4	2	4	2	2	1	2.8	2	3	3	2	0	0	9.07
339837	35	1	39+5	2	4	3	2	3	1.9	1	3	3	2	0	0	1.65
388726	24	2	39+1	2	3	2	2	1	2	2	3	3	2	0	1	1.65
393566	25	1	38+3	2	4	3	2	1	2.3	1	3	3	2	0	1	0.25
375766	23	1	37+6	2	4	2	2	1	2.45	1	3	3	2	0	0	8.41
395050	22	1	37+6	2	4	3	1	1	2.56	1	3	3	1	3	0	6.28
390399	26	1	39	2	4	3	2	3	2.6	1	3	3	2	0	0	9.07
394767	20	1	39+4	1	5	3	1	4	3.51	1	2	2	1	3	0	28.75
394250	18	1	38+4	2	3	2	2	5	3.5	2	3	3	2	0	1	10.24
385760	31	3	39+5	2	4	2	2	1	2.92	2	3	3	2	0	1	9.07
376388	25	1	38+2	2	4	2	2	1	2.9	2	3	3	1	3	0	6.28
311169	32	1	39+6	2	5	1	2	1	2.1	2	3	3	2	0	1	0.05
394622	32	1	39+4	2	3	2	1	4	2.8	1	3	3	2	0	0	22.5
399855	24	1	39+3	2	4	2	2	1	3.2	2	3	3	2	0	0	4.42
393268	34	1	39+6	2	4	2	2	1	3.1	2	3	3	2	0	0	1.65
394697	23	1	38+5	2	4	3	2	2	2.76	1	3	3	2	0	0	6.28
390423	25	1	38+4	2	3	3	1	1	2.9	1	3	3	2	0	1	22.5
391595	20	1	38+5	2	4	3	2	5	2.1	2	3	3	2	0	2	0.75
395250	26	1	38+6	2	4	3	2	1	2.8	2	3	3	2	0	0	7.21
345586	36	1	39+5	2	4	3	2	1	2.91	1	3	3	2	0	0	10.11
394410	23	1	39+5	2	5	4	2	1	2.9	1	3	3	2	0	0	7.21
395097	25	1	39+2	2	4	3	2	4	2.96	1	3	3	2	0	0	2.51
395170	26	1	39+3	2	4	2	2	2	3	1	3	3	2	0	0	2.09
393573	19	1	37+5	2	5	2	2	1	2.9	2	3	3	2	0	0	9.07
393685	26	1	38+5	1	3	2	1	2	3.1	1	3	3	2	0	0	22.5
323376	30	1	37+6	2	4	2	2	1	3.35	2	3	3	2	0	0	10.07
393795	25	1	38+6	2	4	2	2	1	2.8	1	3	3	2	0	0	6.28
393731	26	2	39+4	2	4	2	2	1	3.4	2	3	3	2	0	1	9.07
352996	20	1	39+6	2	2	2	2	1	3.12	1	3	3	2	0	1	22.5
390891	25	2	39+1	2	3	3	2	1	3.1	2	3	3	2	0	0	9.07
391457	24	1	38+1	3	4	1	2	4	3.22	2	3	3	2	0	0	7.21
390041	24	1	38+2	2	4	2	2	2	2.9	2	3	3	2	0	0	9.07
388221	21	1	39+2	2	4	2	2	1	2.1	2	3	3	2	0	0	6.28

391481	31	1	39+4	2	4	2	2	1	3.46	2	3	3	2	0	0	10.51
391469	21	1	37+5	2	4	3	2	1	3.14	2	3	3	2	0	1	8.41
391432	32	1	39+3	2	5	3	2	1	2.94	1	3	3	2	0	0	8.41
303732	23	2	37+6	2	3	3	1	2	3.5	2	3	3	2	0	0	22.5
386164	23	1	39+3	2	4	1	2	1	2.6	1	3	3	2	0	0	7.21
417456	30	1	38+3	2	4	2	2	5	2.9	1	3	3	2	0	1	2.58
410743	21	1	38+5	2	4	2	2	1	2.7	1	3	3	2	0	0	16.25
417412	25	1	39	2	3	2	2	1	3.24	1	3	3	2	0	0	6.28
416227	24	1	39+5	2	4	3	2	1	2.8	2	3	3	2	0	0	5.35
409329	25	1	40+1	2	4	3	2	1	3.51	2	3	3	1	3	0	10.15
414894	20	1	38+5	2	4	2	2	3	2.56	1	3	3	2	0	0	7.49
416289	31	2	39+4	2	4	2	2	2	2.9	1	3	3	2	0	0	8.41
341540	23	1	39+6	2	4	2	2	1	3.3	1	3	3	2	0	0	7.21
411763	25	1	38+5	2	4	3	2	1	2.78	1	3	3	2	0	0	5.35
414549	31	1	38+6	2	3	3	2	1	3.52	2	3	3	2	0	0	22.5
416944	26	1	39+2	2	4	3	1	2	2.8	2	3	3	2	0	0	22.5
415517	19	1	37+6	2	4	2	2	1	2.99	2	3	3	2	0	0	9.07
415713	33	1	37+3	2	4	2	2	1	3.3	2	3	3	1	3	0	10.34
414844	25	1	37+5	1	4	3	2	5	3.47	2	3	3	2	0	0	16.25
415833	36	1	38+6	2	2	4	1	1	3.55	2	2	2	1	3	0	35
416852	26	1	39+3	2	4	2	1	2	2.97	2	3	3	2	0	0	16.25
415832	33	1	39+5	2	4	2	2	1	2.45	1	3	3	1	3	0	6.28
416870	25	1	39+4	2	3	2	1	1	2.55	2	3	3	2	0	0	22.5
378808	25	1	38+4	2	3	3	2	1	2.67	1	3	3	2	0	0	22.5
416726	20	1	38+4	2	4	3	2	1	2.9	2	3	3	2	0	0	9.07
365129	32	2	37+5	2	4	3	2	1	3.2	1	3	3	2	0	0	10
415878	30	1	37+6	2	2	3	1	4	3.5	2	3	3	2	0	0	46.68
339669	22	1	38+4	2	4	1	1	3	3.6	1	3	3	2	0	0	28.75
342780	31	2	37+5	2	3	2	1	1	2.8	2	3	3	2	0	1	22.5
426336	23	1	39	2	3	2	2	1	3.5	1	3	3	2	0	0	22.5
415382	20	1	37+3	2	4	2	2	2	2.96	1	3	3	2	0	0	10.22
415385	33	1	38+4	2	4	2	1	1	2.7	2	3	3	2	0	0	22.5
416543	26	1	38+4	2	3	2	2	1	2.44	2	3	3	2	0	0	9.07

414904	35	1	39+5	2	4	2	2	2	3.24	2	3	3	2	0	0	9.07
416827	25	1	38+5	2	3	2	1	1	3.3	2	3	3	2	0	0	28.75
404546	26	1	39+2	2	2	2	1	1	3.3	1	3	3	2	0	0	58.34
416279	20	2	39+6	2	3	1	2	1	3.46	1	3	3	2	0	0	22.5
404781	31	1	38+2	1	5	2	2	2	3.11	2	3	3	2	0	0	8.41
417783	22	1	38+2	2	4	2	1	1	3.2	1	3	3	2	0	0	22.5
417767	34	1	41	2	4	2	2	1	2.9	2	3	3	2	0	0	16.25
383730	23	1	39+5	2	4	2	2	1	2.9	2	3	3	2	0	0	10.22
384017	30	1	39+5	2	3	3	1	4	3.24	1	3	3	2	0	0	22.5
347558	21	1	37+5	2	2	1	1	1	3.54	2	3	3	2	0	0	70
384233	26	1	39+4	2	3	2	1	1	3.5	1	3	3	2	0	0	46.68
384259	18	1	37+5	2	4	3	2	2	3.2	1	3	3	2	0	0	10.5
380901	25	2	38+2	2	2	3	1	2	3.24	2	3	3	2	0	0	35
380848	30	1	37+6	2	5	2	2	1	3.3	2	3	3	2	0	0	16.25
371792	26	1	38	2	4	3	2	1	3	1	3	3	2	0	0	9.07
381220	25	1	38+1	2	4	2	2	1	2.25	1	2	2	1	3	1	1.65
381145	34	1	39+1	2	4	2	2	1	2.8	2	3	3	2	0	0	7.21
371818	26	1	39+5	2	3	3	2	3	3.2	1	3	3	2	0	0	16.25
301084	26	1	38+2	2	4	2	2	2	3.33	1	3	3	1	2	0	16.25
370867	20	1	39+5	2	4	3	2	1	3.2	2	3	3	2	0	0	10
380267	25	1	38+2	2	4	2	2	1	2.9	2	3	3	2	0	0	8.41
381763	31	1	38+3	2	4	4	2	1	2.89	1	3	3	2	0	0	7.49
377852	25	1	39	1	5	3	2	1	2.6	1	3	3	2	0	0	4.42
381877	30	1	39+4	2	3	2	2	5	2.9	1	3	3	2	0	1	1.25
381847	34	2	40+2	2	4	3	2	3	3.2	2	3	3	2	0	0	7.21
369412	23	1	38+4	2	4	1	2	1	3	2	3	3	2	0	0	8.41
381890	24	1	38+3	2	4	2	2	1	3.24	2	3	3	2	0	0	8.41
377316	21	1	39+2	2	3	2	2	2	2.96	2	3	3	2	0	0	10.4
347671	31	1	39+3	2	3	2	1	1	2.98	2	3	3	2	0	0	22.5
376464	25	1	39+3	2	2	4	2	1	3.13	2	3	3	2	0	0	70
382243	32	1	39+2	2	2	3	1	1	3.51	2	3	3	2	0	0	46.68
382275	23	1	38+5	2	3	3	1	1	3.55	2	3	3	2	0	0	35

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