European Review for Medical and Pharmacological Sciences 2022; 26: 5926-5931

Vitamin D supplementation during pregnancy inhibits the activation of fetal membrane NF-κB pathway

N. GURKAN

Department of Obstetrics and Gynecology, Medical Park Hospital, Samsu[illegible]urkey

Abstract. – **OBJECTIVE:** Nuclear Factor-κB (NF-κB) is an important member of the basic cellular inflammatory pathway that regulates inflammation and apoptosis in fetal membranes. Vitamin D (VD) exerts its anti-inflammatory and immunomodulatory effects via the NF-κB pathway. This study was designed to investigate amniotic fluid NF-κB levels in pregnant women undergoing VD replacement therapy.

PATIENTS AND METHODS: Sixty patients who received antenatal vitamin D supplementation from the 14th week of pregnancy until delivery were included in the study. Participants were selected among those whose serum vitamin [illegible]els were compatible with insufficiency (20 mL), according to the Endocrine Society p[illegible]s-al. Participants were divided into three groups [illegible] 20 patients in each group and one of the chole[illegible]ciferol or placebo treatments was given. Patie[illegible] in Group 1 were given 500 IU/d[illegible]olecalci[illegible]erol, while patients in Group 2 [illegible]1000 IU/day of cholecalciferol. Patie[illegible] grou[illegible]ere not given cholecalciferol treat[illegible] (place[illegible]. Patients in all groups underwent el[illegible]ces[illegible]. Amniotic fluid sample[illegible]re [illegible]after the fetal membranes were[illegible]and be[illegible]e fetal parts were manually re[illegible]d.

RESULTS: Th[illegible]tic fluid NF[illegible]evel of the control g[illegible]p w[illegible]d not receive VD replacement [illegible]9.33±2.[illegible]/mL. The amniotic fluid NF-[illegible]evel of the 5[illegible]day VD replacement g[illegible] was found to b[illegible]2±1.23 ng/mL. Comp[illegible]d to th[illegible]ontrol group, NF-κB levels of pre[illegible]t wom[illegible]given 500 IU/day VD replacemen[illegible]e s[illegible]nificantly[illegible]wer (9.33±2.02 ng/mL *vs.* 6.1[illegible]g/mL, [illegible]03). The amniotic fluid [illegible]B le[illegible]the[illegible] IU/day VD replacement g[illegible]was [illegible]e 3.09±0.44 ng/mL. Compar[illegible]o the co[illegible] group, amniotic fluid NF-κB leve[illegible]f pregnant women given 1000 IU/day VD re[illegible]re significantly lower (9.33±2.02 [illegible]L *vs.* [illegible]9±0.44 ng/mL, $p<0.01$). When the [illegible] replacement groups were compared among [illegible]selves, the amniotic fluid NF-κB level de[illegible]d approximately twice as much in the 1000 [illegible]/day replacement group compared to the 500 IU/day replacement group (3.09±0.44 ng/mL *vs.* 6.12±1.23 n[illegible]L, $p<0.0$[illegible]ega[illegible]correlation was fo[illegible]etween am[illegible]uid NF-κB level and V[illegible]=-0.789, $p<0$[illegible].

CONC[illegible]ON[illegible] present study showed for the first time tha[illegible]iotic fluid NF-κB levels decr[illegible]in pregnan[illegible]en who underwent VD[illegible]ment dose dep[illegible]dent manner.

Words:
[illegible]egnancy, Vita[illegible]D, Amniotic fluid, NF-κB.

Introduction

[illegible]ugh the changes caused by pregnancy in [illegible]tam[illegible]D (VD) metabolism are not known exactly, biochemically low VD levels are frequently encountered during pregnancy[1,2]. Deficiency in dietary intake of ergocalciferol or cholecalciferol or increased skin pigmentation due to pregnancy may lead to a decrease in VD levels[3]. On the other hand, the increased need for calcium and phosphate during pregnancy due to the fetus and its appendages may also lead to a decrease in VD levels. As it is known, the fetus is completely dependent on the mother in terms of VD. Therefore, maternal and umbilical cord VD levels are highly correlated[4]. The VD passes through the placenta and reaches the fetus and its appendages[5]. While maternal serum ionised calcium levels are kept stable during pregnancy, active 1,25(OH)2 D levels are increased[5]. Trying to keep calcium and phosphate metabolism stable and increasing 1,25(OH)2 D during pregnancy are important in providing immune tolerance to the fetus[6].

VD replacement in pregnant women increases umbilical cord VD levels as well as neonatal serum VD levels[7]. In addition to providing bone mineral density and muscle development of the fetus, VD also plays an important role in the continuation of the strong immunomodulatory effect mediated by T and B cells and the prevention of

Corresponding Author: Naziye Gurkan, MD; e-mail: nazeyg@yahoo.com

inflammatory reactions[8]. Chorioamnionitis with antenatal inflammation, which is thought to develop in VD deficiency, may lead to the emergence of different pathophysiological processes[9,10]. In VD deficiency, the fetus becomes more prone to many acute and chronic inflammatory diseases especially respiratory system and fetal membranes[11,12]. It has been reported that low VD induces premature rupture of membranes and uterine contractions through chorioamnionitis and may lead to preterm delivery[13,14]. However, the researchers reached this conclusion with observational studies and did not investigate the inflammatory changes that are claimed to occur in the fetal membranes.

Nuclear Factor-κB (NF-κB) is an important member of the basic cellular inflammatory pathway that regulates inflammation and apoptosis[15]. NF-κB is inactive in the cytosol by complexing with inhibitor kB[16,17]. VD regulates its anti-inflammatory and immunomodulatory effects via T and B cells as well as NF-κB pathway. Thinning and tearing of fetal membranes is a multifactorial regulated process. In addition to hormonal factors and uterine contractions, inflamm[illegible]n of the chorioamniotic membranes also p[illegible] important role in the initiation of labor[17]. [illegible]to its anti-inflammatory and immunomodulator[illegible] fects, VD may play a role in the initiation of la[illegible] Decreased tensile strength and [illegible]re of fet[illegible] membranes in VD deficiency[illegible]egulated via the NF-κB pathway. Th[illegible]ason [illegible]preterm rupture of membranes in[illegible] deficie[illegible]y be the activation of the NF[illegible]kB[illegible] the triggering of infl[illegible]mator[illegible]ion[13,18]. This study was designe[illegible] determin[illegible]iotic fluid NF-κB levels in[illegible]t women un[illegible]ing VD replacement. [illegible]e a[illegible]tic fluid NF-κB levels are found t[illegible] b[illegible] higher [illegible]gnant women who did not u[illegible]go VD replace[illegible] we can say that the inc[illegible]ed frequency of pr[illegible]m labor in VD defici[illegible]y may [illegible]F-κB-dependent.

[illegible]ient[illegible]d Methods

Pa[illegible]t Selec[illegible] and Grouping

S[illegible]atients who received antenatal vitamin D s[illegible]n from the 14th week of pregnancy [illegible]l delivery were included in the study. Partici[illegible] were selected among those whose serum vita[illegible] levels were compatible with insufficiency (20-[illegible] ng/mL), according to the Endocrine Society propsal[19]. Sixty pregnant women were divided into three groups with 20 patients in each group and one of the cholecalciferol or placebo treatments was given. Patients in Group 1 were give[illegible]/day of cholecalciferol, while patients [illegible] were given 1000 IU/day of cholecal[illegible]ol. Cholecalciferol doses were determined [illegible]rding to the Maternal Vitamin D Osteoporosis [illegible] (MAVIDOS)[20]. The patients in gr[illegible]3 we[illegible] given cholecalciferol treatment ([illegible]bo). All the[illegible] in group 1 and 2 remai[illegible]in the [illegible]udy unt[illegible] livery. Missed dose [illegible]e-adm[illegible]tered up to 7 days. Preparations co[illegible]ing[illegible]cium, f[illegible] acid and iron were al[illegible]ovi[illegible]each gr[illegible] during vitamin D trea[illegible]t[21].

Inclusion [illegible] were deter[illegible]as follows. Women a[illegible]20[illegible] and older wi[illegible] spontaneous pregnancy[illegible]nd 14[illegible]leted weeks of gestation accordi[illegible]to USG or la[illegible]strual period (LMP). Fo[illegible]ith more than [illegible]USG record, the first [illegible]d was considered. Accordingly, we consid[illegible] 14 weeks +[illegible]ys to be included in the study. [illegible]mated date o[illegible]th was determined according t[illegible]P. If there[illegible] a difference of more than ten day[illegible]G and LMP, the estimated date of birth [illegible]etermined according to second trimester USG. Those with a history of hypersensi[illegible]VD, hypercalcemia, or kidney stones, as [illegible]ose with active tuberculosis, parathyroid pathology, liver or kidney disease, hypertension or proteinuria were excluded from the study. Multiple gestations, IVF/ICSI pregnancies, oligohydramnios and epilepsy drug users were also excluded from the study. Patients who planned for normal vaginal delivery were not included in the study because it was not suitable for the study design. Those with a history of PPROM, patients with placenta previa or ablatio placenta, patients with a history of diabetes mellitus or gestational diabetes mellitus, and those with a C/S decision due to preeclampsia and eclampsia were not included in the study. Since the cervical softening mechanism is different in breech presentations, these patients were excluded from the evaluation.

Patients in all three groups underwent elective cesarean section. Amniotic fluid samples were collected after the fetal membranes were cut and before the fetal parts were manually removed. Sterile 10 cc injectors were used for amniotic fluid collection. Care was taken not to contaminate the samples with blood. Amniotic fluid samples containing dense blood and vernix caseosa were not included in the study. Samples containing mild to moderate blood or vernix were included in the study after centrifugation. The primary

outcome was to determine the amniotic fluid NF-κB levels of pregnant women who received and did not receive VD. The secondary outcome was to correlate the amniotic fluid NF-κB with the dose of vitamin D administered and other laboratory, demographic parameters. All procedures performed in this study were in accordance with the Ethical Standards of International Research Committee and local approval was obtained from the OMU. All patients recruited to the study were fully counseled and written informed consent was obtained.

NF-κB Analysis in Amniotic Fluid

NF-κB levels were measured using ELISA after thawing the amniotic fluid samples of all groups that were taken during cesarean section and frozen in RNA later. The immunological kit used can measure NF-κB levels in biological fluids with great sensitivity (CusabioBiotechCo., Ltd., WUHAN, CHINA). The kit can make *in vitro* and quantitative measurements in humans. Thawed amniotic fluids were treated with phosphate buffer and centrifuged at 2500 rpm for 5 minutes. Thus, blood and vernix were rem NF-κB was analyzed according to the specified by the manufacturer in the kit. R are given in ng/mL. The detection range o kit was 0.3 to 20 ng/mL and the minimum m surable level was 0.078 ng/mL. Th tra- and i ter-assay coefficients of varia kit were <8% and <10%, respectivel est res are expressed as ng/mL.

Statistical Analysi

The data obtain llowing placement was analyzed wi se of the Sta l Package for Social nce ware 21.0 for windows package software (SPSS Inc., Armonk, NY, USA). Normality of data was examined by nonparametric Kolmogorov–Smirnov test. Continu ables were analyzed using parametri ANOVA test. Pearson's correlatio alysis was used to determine the correlati tween amniotic fluid NF-κB levels and oth ameters. Data are presented as mean D. A p <0.05 was considered statistical gnificant.

Results

Demograph aborator and perinatal charac s of patients nd without VD repla ch resented in able I. There was no sig ficant nce between the groups in ter f serum VD gravida, parity, and ge age. The VD l s of all experimental a placebo groups were determined as 20-30 ng/ consistent insufficiency. Post-replace- VD levels not evaluated. There was no d nce betw the groups in terms of fetal birt tive cesarean delivery was performed w t complications in all groups.

The amniotic fluid NF-κB level of the control gro o did not receive VD replacement was 55 2 ng/mL. The amniotic fluid NF-κB level of the 500 IU/day VD replacement group was found to be 6.12±1.23 ng/mL. Compared to the control group, NF-κB levels of pregnant women given 500 IU/day VD replacement were significantly lower (9.33±2.02 ng/mL *vs.* 6.12±1.23 ng/mL, p<0.03). The amniotic fluid NF-κB level of the 1000 IU/day VD replacement group was found to be 3.09±0.44 ng/mL. Compared to the control group, amniotic fluid NF-κB levels of pregnant women given 1000 IU/day VD replace-

Table I ographic and laborator haracteristics of vitamin D replacement and placebo groups.

	Group 1 (n=20) 500IU/day Vitamin D	Group 2 (n=20) 1000 IU/day Vitamin D	Group 3 (n=20) Placebo
Age (yea	27.9±6.10	29.1±5.30	28.3±7.20
el (ng	24.4±4.51	25.9±6.03	26.5±8.22
Gra y	3.20±1.02	2.90±1.44	2.87±0.50
P	2.11±0.33	1.87±0.40	1.90±0.21
ational a (weeks)*	37.2±8.33	36.6±5.31	37.3±7.10
al birthweight (gr)	2895.1±149.2	2945.4±202.1	2830.4±104.5
ic fluid NF-kB (ng/mL)**	6.12±1.23	3.09±0.44	9.33±2.02

*Estimated date of birth was determined according to LMP or USG. **Amnotic fluid NF-kB levels of both groups who received 500 or 1000 IU/day VD replacement were found to be significantly lower than the controls.

ment were significantly lower (9.33±2.02 ng/mL *vs.* 3.09±0.44 ng/mL, $p<0.01$). When the VD replacement groups were compared among themselves, the amniotic fluid NF-κB level decreased approximately twice as much in the 1000 IU/day replacement group compared to the 500 IU/day replacement group (3.09±0.44 ng/mL *vs.* 6.12±1.23 ng/mL, $p<0.01$). A negative correlation was found between amniotic fluid NF-κB level and VD dose (r=-0.789, $p<0.04$). No significant correlation was found between other maternal and perinatal parameters and NF-κB levels.

Discussion

There is no consensus on both the definition of VD deficiency in pregnancy and the dose of VD to be used for supplementation. The threshold values used for the definition of deficiency vary from country to country. In this study, we selected the patient groups among pregnant women with VD insufficiency (25-35 ng/mL). In general, the recommended dose for VD support during pregnancy varies between 400-600 IU/day. However, the World H th Organization has not determined a specific VD support during pregnancy[22]. On the other d, while the Endocrine Society determines the t limit of the maximum safe VD dose as 10,000 day in pregnant women with VD ficiency, t dose is 600 IU/day in pregnant o do not have VD deficiency[19]. Due t these ates, we administered 500 IU/day t group ticipants and 1000 IU/day of the order not to cause an terin al complications. Since these d were far t he safe upper limits recom l by the Endo Society, we did not enc er a ternal and fe al complications relate t VD thre t the study.

The m isms that init hor are not fully known al and systemic hor nes, inflammator cytol whose hesis and release in fetal membra e inc ed as a sult of the synchronized activa fetal a l cortex, maternal brain placen mpa nt. While cascading local stemic cause inflammation of the mem nes, ma metalloproteinases cause the infl membranes to rupture and stimulate the c NF-κB may play a role in this reaction cascade as the main regulator of inflammation. release of adhesion molecules such as E-selectin, xcellular adhesion molecule 1, and vascular cell a hesion molecule 1 is regulated by the NF-κB pathway[23]. VD may contrbute to the regulation of inflamatuar pathways in fetal membranes with its immunomodulatory and anti-inflammatory effects. In good agreement with this in the LPS d placental inflammation model, Vit D tion has been shown to inhibit both p tal NF-kB signaling and VD receptor expres In the same study, it was emphasized that VD e anti-inflammatory effect in placenta sue via B and VD receptors[24]. On the oth and, since target of VD in the infl atory thway is κB[25,26], the onset of la may be ed in case of VD deficiency[13,18]. Our is t rst clini tudy to present data o effe D repl ent on amniotic fluid N B levels. ni fluid NF-κB levels of l ups in which erwent VD replaceme ere ficantly lowe han the group that did n receive eplacement therapy (placebo). W found a nega rrelation between the gi dose and the re ion in amniotic fluid N B. Compared to the group given 500 IU VD y, the amnoti id NF-κB levels of the patients n 1000 IU/da D showed a two-fold decrease. do not cl y know the rationale for the dec B levels in parallel with the increase D dose. Since this is not a dose-response study, we cannot clearly state what the VD dose is that reduces amniotic fluid F-k evels. By choosing the doses, we chose the VD that have close to the minimum feto-maternal side effects according to the Endocrine Society criteria. We showed that VD at a dose of 500 IU/day significantly reduced amniotic fluid NF-κB levels. The inhibitory effect of VD on the fetal membrane NF-κB pathway may also be dose-dependent, since the dose of VD required for the improvement of clinical and laboratory findings in paratroid dysfunction is higher than the pharmacological VD dose[27]. However, more detailed dose-response studies are needed to determine the minimum VD dose required to meet the needs of both mother and fetus during pregnancy.

VD exerts its inhibitory effect on fetal membrane NF-κB pathway via the VD receptor (VDR). VDR is intensely expressed in the placenta and decidua as well as in all reproductive organs[24,28]. In addition to those in the systemic circulation, 1,25(OH)2D3 production also takes place locally at the maternal-fetal interface. However, we do not have data on whether VD affects the VDR in fetal membranes in a genomic or nongenomic way. Since VD exerts its anti-inflammatory effect on fetal membranes through uterine natural killer cells, macrophages, and T lymphocytes, it may also reduce NF-κB in fetal membranes by

a similar mechanism[24,28,29]. The inhibition of the decidual cells differentiation by 1,25(OH)2D3[30,31] may explain the reduction of NF-κB synthesis in fetal membranes by VD. The increased synthesis of pro-inflammatory cytokines in VDR knock-out mice[32] is an important proof that the NF-κB-blocking effect of VD is mediated by VD receptors. Moreover, in line with our results, another strong evidence is that 1,25(OH)2D3 blocks cytokine release in uNK cells[33].

VD inhibits the NF-κB pathway through different mechanisms in different cells. While it blocks the binding of NF-κB to DNA in some cells, it decreases RelB and VD receptor expression in others and sometimes prevents the nuclear translocation of p65[34-36]. On the other hand, it is not known exactly how VD blocks NF-κB in fetal membranes. VD replacement may increase the synthesis of many local and systemic factors that block NF-κB translocation to the nucleus[37-39]. Since the patients were sent to elective cesarean section we could not establish a correlation between the delivery times of the VD repecement groups and NF-κB levels. Since active labor can affect NF-κB levels by stimulating inflammation in fetal membranes, w d to design the study in this way.

Conclusions

We showed for the first tim tic fluid NF-κB levels decreased in nant v en who underwent VD replaceme ur resu clini- cally important in terms o ns g of the NF-κB pathw the on of labor. In addition, this may ope horizons in the treatmen stetric emer s such as preterm la and ature rupture of fetal membranes du to the i ry effect of VD on the place NF-κB signali thway[24].

Co of est

The a re no c cts of interest.

Eth Comm

Eth tandards of International Research Committ- t roval was obtained from the OMU.

ed Consent

All ents recruited to the study were fully counseled and written informed consent was obtained.

ORCID

N. Gurkan 0000-0003-1088-018X.

References

1) Blomberg Jensen M. Vit D and epro- duction. Nat Rev End ology 2014; 186.
2) Zhang JY, Lucey A organ nny LC, K y M. Impact of preg n vi D stat a lon- gitudinal study r J 1-7.
3) McAree T, s B, M vasa , Sivalo- kanathan nnan L, Bas ow S, Blair M. Vita iciency in preg still a public healt e. Child Nutr 20 ; 9: 23-30.
4) Magh oli Z, H Nezhad A, Shafaei AR, K i F, Madani F ani B. Vitamin D status ers and their ne ns in Iran. BMC Preg- ancy Childbirth 2007; 7: 1.
5) Curtis EM, M RJ, Harvey NC, Cooper C. Ma- ternal vitamin upplementation during pregnan- y. Br Med B 018; 126: 57-77.
6) blyn JA wison M, Wagner CL, Bulmer JN, unological role of vitamin D at the mate tal interface. J Endocrinol 2015; 224: R107-21.
7) ke OG, Brown IR, Bone CD, Carter ND, e HJ, Maxwell JD, Robinson VP, Winder SM. Vitamin D supplements in pregnant Asian women: effects on calcium status and fetal growth. Br Med J 1980; 280: 751-754.
8) Guillot X, Semerano L, Saidenberg-Kermanac'h N, Falgarone G, Boissier MC. Vitamin D and inflammation. Joint Bone Spine 2010; 77: 552-557.
9) Adams JS, Hewison M. Update in vitamin D. J Clin Endocrinol Metab 2010; 95: 471-478.
10) Bikle D. Nonclassic actions of vitamin D. J Clin Endocrinol Metab 2009; 94: 26-34.
11) Brehm JM, Schuemann B, Fuhlbrigge AL, Hollis BW, Strunk RC, Zeiger RS. Childhood Asthma Management Program Research G Serum vitamin D levels and severe asthma exacerbations in the Childhood Asthma Management Program study. J Allergy Clin Immunol 2010; 126: 52-58; e55.
12) Clifford RL, Knox AJ. Vitamin D – a new treatment for airway remodelling in asthma? Br J Pharmacol 2009; 158: 1426-1428.
13) Urrutia RP, Thorp JM. Vitamin D in Pregnancy: Current Concepts. Curr Opin Obstet Gynecol 2012; 24: 57-64.
14) Palacios C, Kostiuk LK, Peña-Rosas JP. Vitamin D supplementation for women during pregnancy. Cochrane Database Syst Rev 2019; 7: CD008873.
15) Hu W, Zhou PH, Rao T. Adrenomedullin attenuates interleukin-1beta-induced inflammation and apoptosis in rat Leydig cells via inhibition of NF-kappaB signaling pathway. Exp Cell Res 2015; 339: 220-230.

16) Celik O, Celik E, Turkcuoglu I, Yilmaz E, Ulas M, Simsek Y, Karaer A, Celik N, Aydin NE, Ozerol I, Unlu C. Surgical removal of endometrioma decreases the NF-kB1 (p50/105) and NF-kB p65 (Rel A) expression in the eutopic endometrium during the implantation window. Reprod Sci 2013; 20: 762-770.

17) Fischer C, Page S, Weber M, Eisele T, Neumeier D, Brand K. Differential effects of lipopolysaccharide and tumor necrosis factor on monocytic IkappaB kinase signalsome activation and IkappaB proteolysis. J Biol Chem 1999; 274: 24625-24632.

18) Bodnar LM, Klebanoff MA, Gernand AD, Platt RW, Parks WT, Catov JM, Simhan HN. Maternal vitamin D status and spontaneous preterm birth by placental histology in the US Collaborative Perinatal Project. Am J Epidemiol 2014; 179: 168-176.

19) Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, Murad MH, Weaver CM; Endocrine Society. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. J Clin Endocrinol Metab 2011; 96: 1911-1930.

20) Harvey NC, Javaid K, Bishop N, Kennedy S, Papageorghiou AT, Fraser R, Gandhi SV, Schoenmakers I, Prentice A, Cooper C. MAVIDOS Maternal Vitamin D Osteoporosis Study: study protocol for a randomized controlled trial. The MAVIDOS Study Group. Trials 2012; 13: 13.

21) Roth DE, Gernand AD, Morris SK, Pez… Islam MM, Dimitris MC, Shanta SS, Zlotk… H, Willan AR, Ahmed T, Shah PS, Murphy KE, V… berg R, Choufani S, Shah R, Al Mahmud A. Ma… nal vitamin D supplementation during pregnan… and lactation to promote infa… in Dhaka… Bangladesh (MDIG trial): st… or a randomized controlled trial. T… 2015; …00.

22) World Health Organisati… ideline: … plementation in pregnant … G…

23) Zhong L, Simard … fluot … helial microRNAs regulating … NF-κB pat… and cell adhesion molec… during inflamm… FASEB J 2018; 32: 4…

24) Chen YH, … Z, Fu… ng H, Chen X, Zhang C, Lv Z… Xu DX. Vitam… inhibits lipopolysaccha… nduced placenta… mation through rei… ing interaction betwee… itamin D receptor … nuclear… r kappa B p65 subunit. Sci Rep … 5: 1…

25) … King N… iar R. Vitamin D and its ana… regul… of immune activation and ntigen… n… Annu Rev Nutr 2003; 23: …-145.

26) … EE, Mathi… C. Immunoregulation by 1,25-di… itamin D3: basic concepts. J Steroid Bio… …iol 2005; 97: 93-101.

… Bouillon R. Vitamin D: from photosynthesis, metabolism, and action to clinical applications, endocrinology: adult and pediatric: vitamin D, 7th ed. Philadephia, PA: Elsevier, 2015; pp. 1018-1037.

28) Arck PC, Hecher K. Fetomaternal immu… talk and its consequences for mater… 's health. Nat Med 2013; 19: 548-5…

29) Tamblyn JA, Hewison M, Wagn… , Bulmer JN, Kilby MD. Immunological role … in D at the maternal-fetal interface. J Endoc… 015; 224: R107-121.

30) Kachkache M, Rebut… neton C, D… J, Cynober E, Garab… h M. L… rine cell… er than stromal d… al cell… required for 1,25-dihydroxyvit… 3 pr… tion dur… early human pregna… 993; 33… -88.

31) Griffin MD, … W, Phan… Bachm… A, McKean DJ, … r R. Dendri… ulation by 1,α-25 d… vitamin D3 a… nalogs: a vitamin … ce… pendent pathw… that promotes a persis… state o… urity in vitro and in vivo. Proc Natl Acad Sci 2001; … 0-6805.

32) … , Kaplan AT, … ietty V, Ouyang YB, …uyang Y, Simmons CF. Vitamin D and the regu…lation of placen…al inflammation. J Immunol 2011; 186: 5968-59…

… Evans KN, N… n L, Chan J, Innes BA, Bulmer … Kilby M… ffects of 25-hydroxyvitamin D3 … roxyvitamin D3 on cytokine pro…du… human decidual cells. Biol Reprod 2006; 75: 816-822.

… Berry DM, Clark CS, Meckling-Gill KA. 1al…5-dihydroxyvitamin D3 stimulates phos…phorylation of IkappaBalpha and synergizes with TPA to induce nuclear translocation of NFkappaB during monocytic differentiation of NB4 leukemia cells. Exp Cell Res 2002; 272: 176-184.

35) Adams LS, Teegarden D. 1,25-dihydroxycholecalciferol inhibits apoptosis in C3H10T1/2 murine fibroblast cells through activation of nuclear factor kappaB. J Nutr 2004; 134: 2948-2952.

36) Ribeiro MC, Moore SM, Kishi N, Macklis JD, MacDonald JL. Vitamin D Supplementation Rescues Aberrant NF-κB Pathway Activation and Partially Ameliorates Rett Syndrome Phenotypes in Mecp2 Mutant Mice. eNeuro 2020; 7: ENEURO.0167-20.

37) Harant H, Wolff B, Lindley IJ. 1a,25-dihydroxyvitamin D3 decreases DNA binding of nuclear factor-kB in human fibroblasts. FEBS Lett 1998; 436: 329-334.

38) Sadeghi K, Wessner B, Laggner U. Vitamin D3 down-regulates monocyte TLR expression and triggers hyporesponsiveness to pathogen-associated molecular patterns. Eur J Immunol 2006; 36: 361-370.

39) Celik O, Ersahin A, Acet M, Celik N, Baykus Y, Deniz R. Disulfiram, as a candidate NF-κB and proteasome inhibitor, prevents endometriotic implant growing in a rat model of endometriosis. Eur Rev Med Pharmacol Sci 2016; 20: 4380-4389.