

## SYSTEMATIC REVIEW

### The role of vitamin D supplementation on levator ani muscle remodeling post-delivery

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Article Info	ABSTRACT
Received Feb 20, 2024 Revised Apr 23, 2024 Accepted May 17, 2024 Published Aug 1, 2024  <b>*Corresponding author:</b> Rahajeng rahajeng.fk@ub.ac.id  <b>Keywords:</b> Levator ani Vitamin D supplementation Post-delivery Maternal health	<b>Objective:</b> Vitamin D is considered a crucial vitamin for the restoration of levator ani muscle strength. This study aimed to evaluate the association between vitamin D and levator ani muscle remodeling in the post-delivery period. <b>Materials and Methods:</b> The literature search was conducted across three electronic databases, PubMed, Google Scholar, and Springerlink. Our investigation yielded a total of 2613 studies, out of which 8 studies were found to meet the inclusion criteria and were subsequently included in our study. Among these, 4 studies specifically examined the impact of vitamin D micronutrient status on the levator ani/pelvic muscles during the post-delivery period. <b>Results:</b> The mean maximum contraction strength of the levator ani muscles following the administration of vitamin D supplement was $26.77 \pm 7.15$ cmH <sub>2</sub> O. The analysis conducted utilizing a paired t-test yielded a p-value of less than 0.05, indicating statistical significance. Additionally, a coefficient correlation of 0.831 was observed, with a p-value also less than 0.05. The findings of this study indicate a significant correlation between the levels of vitamin D and the magnitude of levator ani muscle contractions, as evidenced by a statistically significant p-value of less than 0.05. <b>Conclusion:</b> The administration of vitamin D supplements has been found to play a significant role in the remodeling of the levator ani muscle during the post-delivery period. This is evidenced by the observed increase in the strength of the levator ani muscles following vitamin D supplementation.

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#### Highlights:

1. In pregnant women, vitamin D insufficiency may play a role in the development of post-delivery illness.
2. Vitamin D supplementation is significant in remodeling of post-delivery levator ani muscle.

## INTRODUCTION

The levator ani muscle comprises both striated and smooth muscle fibers and exhibits a complex, cone-like structure. Positioned bilaterally within the lower pelvis, it plays a crucial role in supporting and elevating the pelvic floor, thus facilitating the mobility of adjacent pelvic structures. Functioning in conjunction with the coccygeus muscle, the levator ani is an integral component of the pelvic floor musculature. This muscle is further divided into three distinct parts: the puborectalis, pubococcygeus, and iliococcygeus. Innervation primarily arises from the inferior hypogastric plexus, the levator ani muscles, and the pudendal nerves, providing essential neural connections to most of these muscle groups (Figure 1).

According to Timoh et al.,<sup>2</sup> birth-related levator ani injuries seen on magnetic resonance images in affected women (Figure 2) have been linked to a much higher risk of vaginal prolapse and a notable 40% drop in pelvic floor muscle strength. An instrumented speculum is a specialized device commonly utilized to assess the isometric strength of the pelvic floor muscles. This tool measures peak voluntary contraction force applied to close the vaginal opening in the mid-sagittal plane. Interestingly, neither the resting force exerted by the vaginal muscles nor the increase in maximal contraction force demonstrated significant declines with age. This finding is notable, as striated muscle typically loses 30–40% of its volume over the aging process.

Extensive research has shown that isometric muscle strength declines, and axial or appendicular striated muscle strength increases at a slower rate in healthy older adults.<sup>3</sup> Injury to the levator ani muscle can result in pelvic organ prolapse, a condition that many women experience, often leading to a decrease in overall quality of life. Pelvic organ prolapse may cause functional disturbances in the digestive, sexual, and urinary systems and can also induce psychological, social, and emotional challenges, potentially leading to symptoms such as depression, social withdrawal, and anxiety.

The accurate identification of a medical condition is crucial in order to administer appropriate and efficient therapeutic interventions. Levator ani muscle injuries are commonly misdiagnosed, and the therapeutic interventions typically involve limited vaginal repairs. Insufficient focus may be directed towards the dome, also known as the upper part of the vagina, or the descent of the uterus.<sup>4,5</sup>

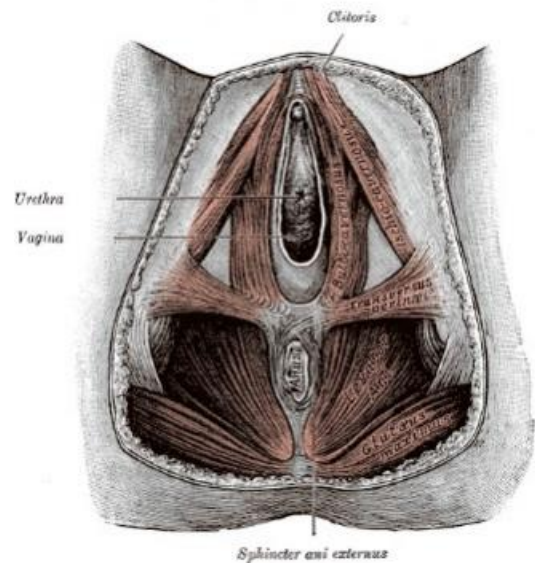


Figure 1. Female perineum, clitoris, urethra, vagina, sphincter ani externus, anus, gluteus maximus, levator ani, transversus perineae.<sup>3</sup>

There is a pressing necessity for the establishment of nutritional guidelines to prevent levator ani muscle injury. The baseline characteristics encompass the average daily energy and nutrient intakes expressed as a proportion of the recommended dietary allowances among women in the post-delivery period. The study revealed that the post-delivery diet exhibited sufficient intake levels (above 80% of the Recommended Dietary Allowance) for energy (82.6%), protein (80.6%), carbohydrates (99.5%), vitamin C (88.7%), vitamin B2 (95.1%), and vitamin B12 (170.8%). However, inadequate intake levels (below 35%) were observed for vitamin D (12.3%), iron (28.3%), and folate (33.8%).<sup>6</sup>

Vitamin D is widely regarded as essential for the health of numerous organ systems. Vitamin D deficiency, however, has become increasingly prevalent. Multiple studies indicate that vitamin D deficiency remains common in Indonesia, as well as in Australia and the United States. Previous research has associated lower serum vitamin D levels with reduced tone and strength in skeletal muscles. Notably, the pelvic floor muscles weaken between 3 and 8 days postpartum but regain strength between 6 and 10 weeks post-delivery. It is widely believed that inadequate vitamin D contributes to decreased skeletal muscle mass and strength.<sup>7</sup> The binding of  $1,25(\text{OH})_2\text{D}_3$  to the vitamin D receptor (VDR) initiates protein transcription, promoting calcium utilization in the body.

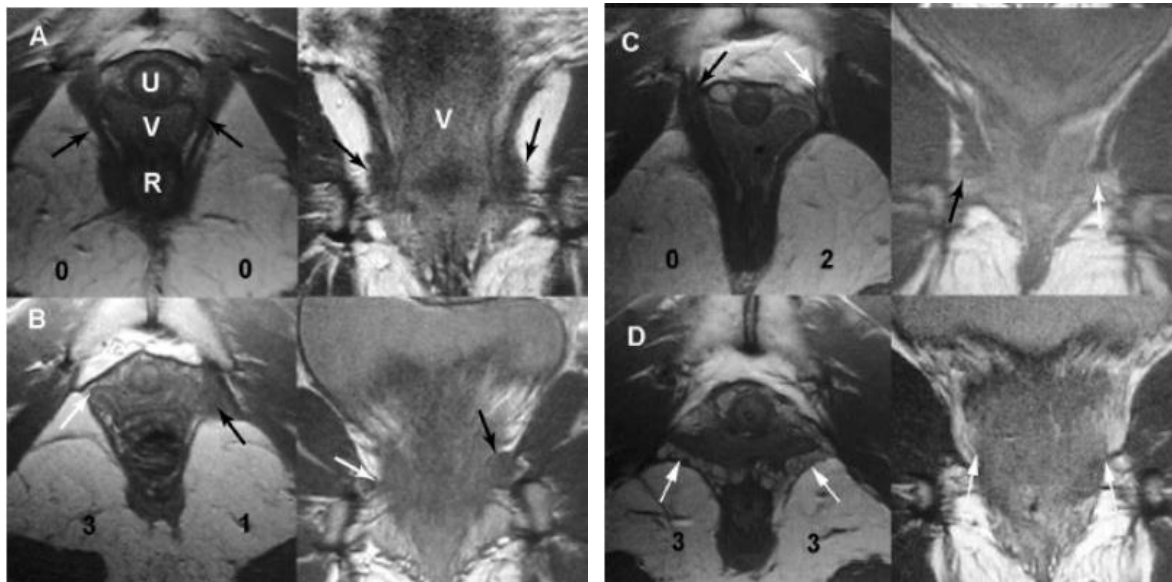


Figure 2. On axial and coronal magnetic resonance images, these examples show the different levels of damage to the levator ani pubovisceral muscle. A: A woman whose muscles are normal; B and D: Three women who each have a major disability; and C: Three women who each have a minor disability. The scores for each side are shown by the handicap scores on the left. There are black arrows that show where the muscle should be and white arrows that show where the muscle is damaged or should be. U stands for the urethra, V for the vaginal canal, and R for the rectum.<sup>2</sup>

Vitamin D has been shown to influence skeletal muscle strength and function significantly, with a strong association observed between vitamin D deficiency and muscle weakness. The levator ani and coccygeus muscles, both skeletal muscles, are essential components of the pelvic floor, and vitamin D levels may impact their function. Research suggests that adequate vitamin D intake can improve skeletal muscle performance. Determining the optimal micronutrient requirements for treating post-delivery levator ani muscle injury remains a significant challenge for healthcare providers. Therefore, this study aimed to evaluate the role of vitamin D in promoting levator ani muscle remodeling during the post-delivery period by conducting a comparative analysis of various micronutrient interventions for this purpose.

## MATERIALS AND METHODS

### Search strategy

The authors conducted a comprehensive literature search across three electronic databases: PubMed, Google Scholar, and SpringerLink. This search yielded 2,163 studies in total, of which 8 met the predefined inclusion criteria and were subsequently incorporated

into the analysis. These 8 studies provided detailed data on 4 studies examining the vitamin D micronutrient status and 4 studies assessing collagen micronutrient status, specifically concerning the levator ani and other pelvic muscles post-delivery. Boolean operators "AND" and "OR" were employed alongside keywords related to the clinical questions to optimize search precision. The findings were reported in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines.

### Article selection

The articles obtained through the search process underwent a second round of screening based on predefined inclusion and exclusion criteria. Inclusion criteria for this analysis required studies to be published in English or Indonesian, use quasi-experimental or observational designs, assess the percentage of micronutrient requirements for levator ani muscle repair, and involve human subjects as participants. Exclusion criteria included interventional studies, studies lacking meta-analytic data, regularly published literature sources, and data derived solely from reviews or abstracts. Only the standard deviation of score coloring was reported, with no additional data provided.

Following these criteria, a total of 19 relevant articles were ultimately included in the analysis.

### Eligibility criteria

The authors undertook a systematic approach to summarizing and assessing the available evidence using a standardized data abstraction form. Prior to initiating the abstraction and review process, the team pilot tested the screening and abstraction forms with a sample of articles. The screening and data collection forms were subsequently refined based on team feedback.

### Data synthesis

A quantitative synthesis was not performed for four reasons: there were significant variations in the operational definitions of the condition under treatment across studies, and there was a limited range of interventions with substantial diversity. This study aimed to replicate the trial using similar interventions but with distinct primary and secondary outcome measures.

### Data Extraction

The data obtained from the identified publications encompassed various aspects, such as the study design and corresponding outcomes, the total number of patients involved, the duration of follow-up during the intervention, details regarding the intervention itself, the effectiveness of the intervention, and any additional comments provided.

## RESULTS AND DISCUSSION

A total of 2163 studies were identified through our search process. After applying the inclusion criteria, a final selection of 8 studies was included in our study. Figure 1 illustrates the flowchart representing the progression of literature through the grading process for the purpose of this review update.

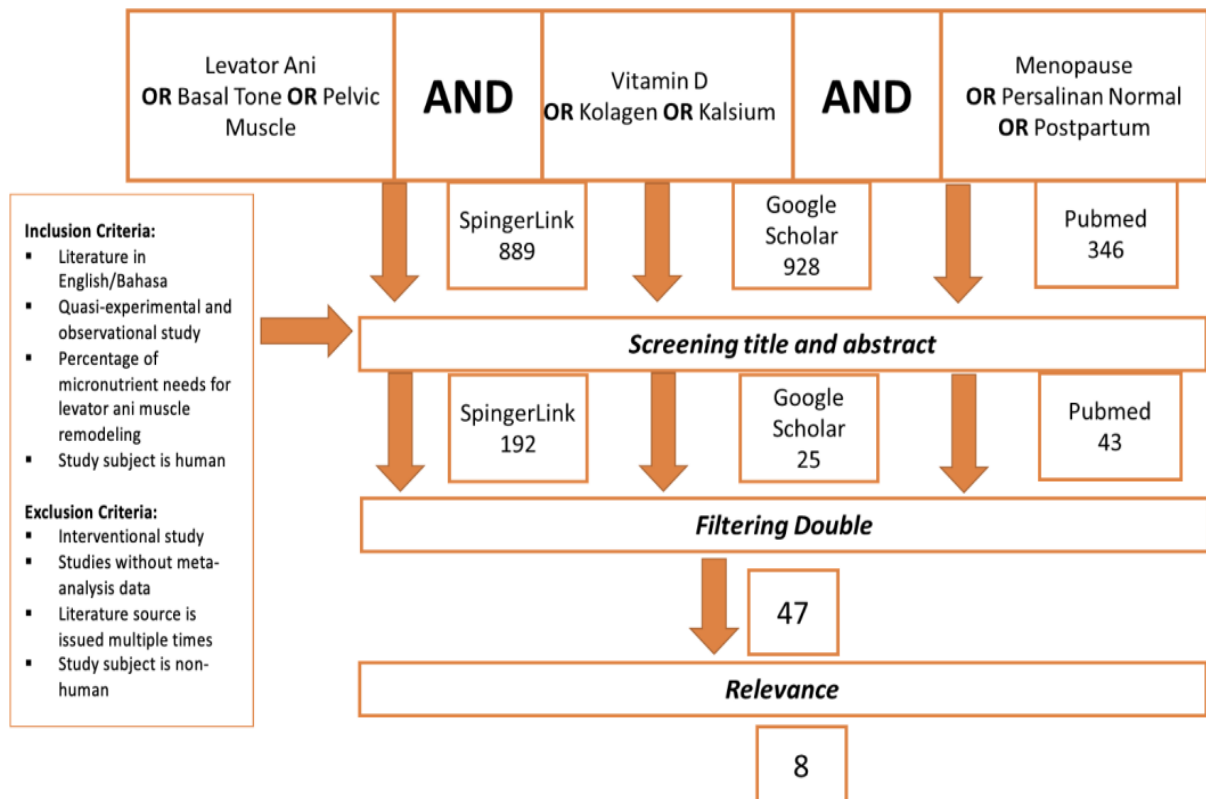


Figure 1. Flowchart of data filtering.

### Effects of vitamin D on levator ani muscle remodeling

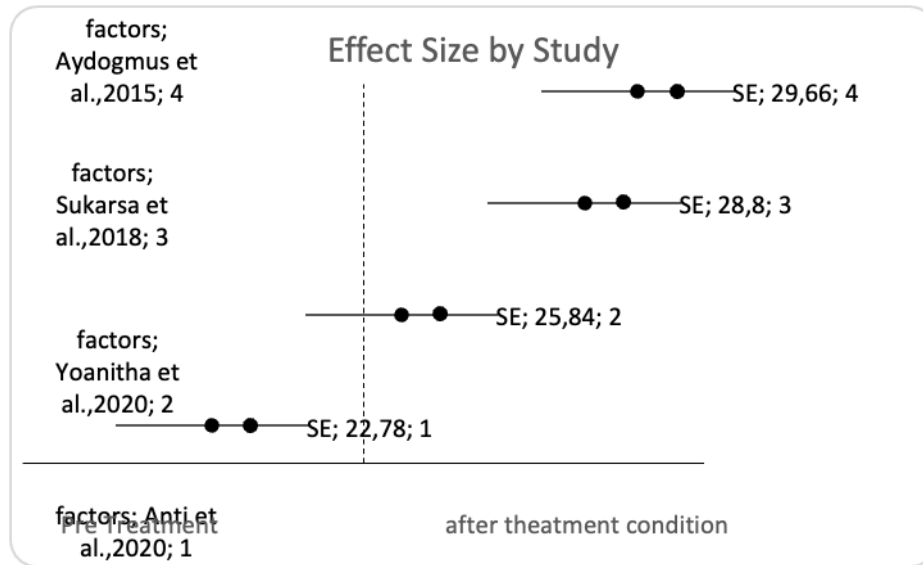


Figure 2. Forest plot effect of vitamin D on levator ani muscle strength.

Research conducted on both animals and humans has demonstrated that Vitamin D3 can enhance muscular strength and function in striated muscle. The clinical manifestation of weakened pelvic floor muscles may manifest as symptoms indicative of pelvic floor dysfunction. There is a notable correlation between the administration of Vitamin D supplementation and muscle, as evidenced by trials conducted by Aydogmus et al.<sup>8</sup> (SMD = 29.66, 95% CI: 19.36; 39.96) in comparison to Sukarsa et al.<sup>7</sup> (SMD = 22.78, 95% CI: 18.88; 26.68) and Yoanitha et al.<sup>9</sup> (SMD = 25.84, 95% CI: 18.73; 32.95), and The current study examined the average maximum pelvic floor muscle contraction strength after vitamin D3 supplementation. The measured value was  $26.77 \pm 7.15$  cmH2O. The results were statistically significant because the paired t-test p-value was less than 0.05. They also had a 0.83 correlation coefficient. This suggests a link between vitamin D levels and levator ani muscle strength. Vitamin D levels and levator ani muscle contraction strength are statistically significant at 0.05 or higher. Vitamin D has many effects on striated muscle strength and function.<sup>7,10</sup>

Research indicates that vitamin D supplementation significantly reduces TGF- $\beta$ 3 levels. Vitamin D's antioxidant properties may cause this. These properties reduce reactive oxygen species (ROS), which inhibits

MMP. However, Vitamin D increases cell structure-produced type I collagen, elastin, and fibronectin.<sup>11-13</sup> Vitamin D affects striated muscle genomically and non-genomically. Genomically, vitamin D controls gene transcription in striated muscle. By activating vitamin D receptors in muscle nuclear membranes. Muscle cells differentiate and multiply through the insulin growth factor (IGF) pathway after activation, resulting in hypertrophy. When 1,25(OH)D binds to membrane receptors, it has a nongenomic effect. Signal transmission activates the MAPK and PLC pathways. Calcium entry into cell structures is affected by these pathways.

#### Effect of collagen type I on pelvic muscles

A change (rs1800012) in the gene that codes for type I, alpha 1 collagen at the collagen binding site of Sp1 has been shown to change gene expression and the way transcription factors bind. A small link has been found between minor alleles and lower bone mineral density and a higher risk of breaking a bone in people with osteoporosis. The vaginal epithelium and endopelvic fascia are primarily composed of collagen type I and alpha 1. The gene and protein expression data in pelvic tissue from women with prolapse or stress incontinence who took part in a previous study are very different, showing that the data needs to be improved and refined even more.



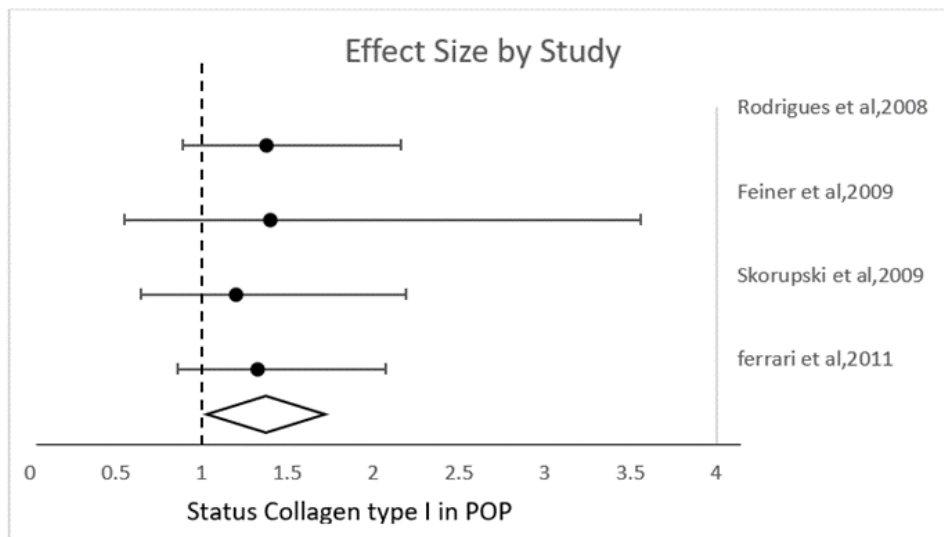


Figure 3. Forest plot status collagen type I in POP

Anatomical pelvis organ prolapse (POP) has been linked to the genetic variant rs1800012 in five different studies. These studies were done in Brazil, Israel, Poland, Italy, and Korea. Because all 30 people who took part in a study had the GG allele, they were not included in the statistical analysis. There was some disagreement between the last four studies, but not a lot. Their effect sizes were still statistically important (OR, 1.33; 95% CI, 1.02-1.73). We regarded that the bias could not be ruled out completely because the QC genotypes were not known and the two samples may have been affected by population stratification. One sample is very different from Hardy-Weinberg equilibrium, which suggests a large amount of bias, but it had no effect on the results.<sup>14,15</sup>

The study discovered a strong correlation between vitamin D levels and both the levator ani's basal tone and its maximum contractions. Some factors that can change a person's vitamin D levels and pelvic floor muscles are their age, body mass index, race, vaginal delivery, diet, and time spent in the sun. Sukarsa et al.<sup>7</sup> found that women of childbearing age and pregnant women in their first trimester are more likely to have nutritional problems because of the way their bodies work during their periods and during pregnancy. Vitamin D deficiency can be fixed by making more vitamin D, which can be done through fortification, supplements, and other means. A study looked at 25 primiparas and 20 multiparas who had a spontaneous vaginal delivery between 36 and 42 weeks of pregnancy. The results were different for the same women 3–8 days after giving birth, 6–10 weeks after giving birth, and 9–15 months after giving birth. Pain during contractions and/or pressure inside the abdomen

during the exam have both been linked to the chance of perimetric assessments being biased.<sup>16–19</sup> Our study showed that taking extra vitamin D is important for remodeling the levator ani muscle after giving birth.

If the vitamin D is insufficient, the calcium balance in the pelvic floor muscles gets off, which makes them less effective and can cause problems with the pelvic floor. A small amount of vitamin D has been shown to improve the function of skeletal muscles. PFMT is meant to make the levator ani muscles stronger, which are very important for a woman's ability to control her bladder. This should be the first line of defense when dealing with SUI, OAB, UUI, or FI. Studies using random assignments have shown that strengthening the muscles in the pelvic floor can help reduce urinary incontinence by 54 to 75%. Low 25(OH)D levels may impair skeletal muscle function, which the urethra needs. The levator ani, extrinsic urethral, and external anal sphincters may work better with normal vitamin D levels. Thus, vitamin D levels may affect how well behavioral therapy helps PFMT women control urinary and fecal incontinence. More research is needed to determine how vitamin D affects the levator ani muscle and how vitamin D supplements and PFMT may treat pelvic floor symptoms.

The study's correlation analysis supports the proposed explanation, aligning with numerous studies that have examined the relationship between vitamin D and muscle strength. Aydogmus et al. discovered a link between the strength of the levator ani muscle and the amount of vitamin D a woman had before giving birth. When women had enough vitamin D, their pelvic floor muscles were stronger after giving birth.<sup>8</sup> Low vitamin

D levels in the third trimester have been linked to weaker pelvic floor muscles after giving birth.<sup>20-23</sup> A study by Barat et al. observed 394 women who said they had problems with their pelvic floor and found that insufficient vitamin D was linked to more urinary incontinence and other colorectal symptoms that made their quality of life worse. In a study of 99 girls ages 12 to 14, it was found that vitamin D levels were linked to better measures of strength, speed, and vertical jump. Other studies have looked at what happens to muscle tissue when people take extra vitamin D. Teenage girls between the ages of 12 and 14 who took 150,000 IU of vitamin D supplements every three months for a year showed improvements in their strength, height, and efficiency when jumping.<sup>24-26</sup>

Wang et al. did an in vitro study and found that the sacrouterine ligament in the neck may be able to hold up to 17 kg of weight before the hip gives way. The ECM is made up of elastin, collagen, and fibronectin. It is the main structure-supporting protein.<sup>27</sup> There is a special structure to the ligaments of a woman's genitalia because elastin and collagen can change shape during the reproductive stages of her life. The component goes back to its pre-pregnancy levels after giving birth, even though it rose a lot during pregnancy. POP is more likely in older women, multi-pregnant women, and vaginal birthers. Oxford Family Planning found that POP hospital admissions increased four times for women with one child, eight times for two children, and ten times for more than two children in a prospective study of 17,000 women.

New studies show that vitamin D deficiency increases POP risk. Vitamin D supports skin and heart connective tissues better than bone building and mineral density. Maymon et al.<sup>28</sup> explain the biomolecular processes that regulate vitamin D intake or exposure in POP. TGF-3 levels dropped significantly after vitamin D exposure. Due to its antioxidant properties, vitamin D reduces ROS and matrix metalloproteinase. Vitamin D boosts type I collagen, elastin, and fibronectin production. It was also found that vitamin D grows fibroblast growth factor II, which makes elastin and collagen. Vitamin D makes it easier for skin and heart tissues to make collagen types I and II. Along with the transcription process, this way of copying is thought to be passed down from parent to child.<sup>29</sup>

### Strength and limitation of the study

Strength of this study are there is specific micronutrient and specific vitamin D observed on this study. The limitation of this study is that there is wide gap of age of post-partum woman observed and there isn't exact measurement of levator ani muscle strength.

## CONCLUSION

During pregnancy and childbirth, the levator ani muscles experience significant stress due to both mechanical and neurological factors. However, additional factors may also contribute to these stresses. Pregnant women with inadequate vitamin D levels may face an increased risk of postpartum complications. Strength improvements in the levator ani muscle following postpartum vitamin D3 supplementation suggest that vitamin D plays a critical role in post-delivery muscle remodeling.

## DISCLOSURES

### Acknowledgment

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### Conflict of interest

There is no conflict of interest to disclose.

### Funding

There is no financial conflict of interest to disclose.

### Author contribution

All of the authors worked on this study in some way, including planning, collecting and analysing data, writing, and getting permission to publish.

## REFERENCES

1. Němec M, Horčíčka L, Dibonová M, et al. Analýza stavu muskulo-fasciální složky pánevního dna pomocí MRI u pacientek před plánovaným vaginálním rekonstrukčním výkonem pro symptomatický sestup pánevního dna [MRI analysis of the musculo-fascial component of pelvic floor in woman before planned vaginal reconstruction procedur for symptomatic pelvic organ prolapse]. *Ceska Gynekol.* 2018 Summer;83(2):84-93. Czech. [PMID: 29869505](https://pubmed.ncbi.nlm.nih.gov/29869505/).
2. Nyangoh Timoh K, Bessede T, Zaitouna M, et al. Anatomie du muscle élévateur de l'anus et applications en gynécologie obstétrique [Anatomy of the levator ani muscle and implications for obstetrics and gynaecology]. *Gynécologie Obstétrique & Fertilité.* 2015;43(1):84-90. [doi: 10.1016/j.gyobfe.2014.11.015](https://doi.org/10.1016/j.gyobfe.2014.11.015).

3. Gowda SN, Bordoni B. Anatomy, Abdomen and Pelvis: Levator Ani Muscle. 2022 Oct 26. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024 Jan-. PMID: [32310538](https://pubmed.ncbi.nlm.nih.gov/32310538/).
4. Rahajeng R. The increased of MMP-9 and MMP-2 with the decreased of TIMP-1 on the uterosacral ligament after childbirth. *Pan Afr Med J*. 2018;30:283. doi: [10.11604/pamj.2018.30.283.9905](https://doi.org/10.11604/pamj.2018.30.283.9905). PMID: 30637068; PMCID: PMC6317396.
5. Abbas N, Reid F. Pelvic organ prolapse: anatomical and functional assessment. *Obstet Gynaecol Reprod Med*. 2022;32(7):127–34. doi: [10.1016/j.ogrm.2022.04.006](https://doi.org/10.1016/j.ogrm.2022.04.006).
6. Aparicio E, Jardí C, Bedmar C, et al. Nutrient intake during pregnancy and post-partum: ECLIPSES Study. *Nutrients*. 2020;12(5):1325. doi: [10.3390/nu12051325](https://doi.org/10.3390/nu12051325). PMID: 32392706; PMCID: PMC7285175.
7. Sukarsa RA, Anti DN, Purwara BH, et al. Effect of vitamin D3 supplementation on levator ani muscle strength in primipara pregnancy with postpartum vitamin D3 deficiency. *Indonesian Journal of Obstetrics and Gynecology*. 2020;8(4):231–6. doi: [10.32771/inajog.v8i4.1063](https://doi.org/10.32771/inajog.v8i4.1063).
8. Aydogmus S, Kelekci S, Aydogmus H, et al. Association of antepartum vitamin D levels with postpartum pelvic floor muscle strength and symptoms. *Int Urogynecol J*. 2015;26(8):1179-84. doi: [10.1007/s00192-015-2671-3](https://doi.org/10.1007/s00192-015-2671-3). Epub 2015 Mar 20. PMID: 25792352.
9. Yoanitha N, Purwara BH, Ruslina I, et al. The effect of vitamin D3 supplementation on increases of levator ani contraction strength in women with uterine prolapse. *Indonesian Journal of Obstetrics and Gynecology*. 2020;8(3):174-8. doi: [10.32771/inajog.v8i3.1184](https://doi.org/10.32771/inajog.v8i3.1184).
10. Mangir N, Hillary CJ, Chapple CR, et al. Oestradiol-releasing biodegradable mesh stimulates collagen production and angiogenesis: An approach to improving biomaterial integration in pelvic floor repair. *Eur Urol Focus*. 2019;5(2):280-9. doi: [10.1016/j.euf.2017.05.004](https://doi.org/10.1016/j.euf.2017.05.004). Epub 2017 Jun 3. PMID: 28753895.
11. Roman S, Mangir N, Bissoli J, et al. Biodegradable scaffolds designed to mimic fascia-like properties for the treatment of pelvic organ prolapse and stress urinary incontinence. *J Biomater Appl*. 2016;30(10):1578-88. doi: [10.1177/0885328216633373](https://doi.org/10.1177/0885328216633373). Epub 2016 Feb 18. PMID: 26896234.
12. Mangir N, Bullock AJ, Roman S, et al. Production of ascorbic acid releasing biomaterials for pelvic floor repair. *Acta Biomater*. 2016;29:188-97. doi: [10.1016/j.actbio.2015.10.019](https://doi.org/10.1016/j.actbio.2015.10.019). Epub 2015 Oct 19. PMID: 26478470; PMCID: PMC4678952.
13. Cartwright R, Kirby AC, Tikkinen KA, et al. Systematic review and metaanalysis of genetic association studies of urinary symptoms and prolapse in women. *Am J Obstet Gynecol*. 2015;212(2):199.e1-24. doi: [10.1016/j.ajog.2014.08.005](https://doi.org/10.1016/j.ajog.2014.08.005). Epub 2014 Aug 8. PMID: 25111588; PMCID: PMC4342521.
14. Da Silva AS, Asfour V, Digesu GA, et al. Levator Ani avulsion: The histological composition of this site. A cadaveric study. *Neurourol Urodyn*. 2019;38(1):123-9. doi: [10.1002/nau.23847](https://doi.org/10.1002/nau.23847). Epub 2018 Oct 30. PMID: 30375038.
15. Ali ML, Kumar SP, Bjornstad K, et al. The sheep as an animal model for heart valve research. *Cardiovasc Surg*. 1996;4(4):543-9. doi: [10.1016/0967-2109\(95\)00142-5](https://doi.org/10.1016/0967-2109(95)00142-5). PMID: 8866098..
16. Scaramuzzi RJ, Campbell BK, Downing JA, et al. A review of the effects of supplementary nutrition in the ewe on the concentrations of reproductive and metabolic hormones and the mechanisms that regulate folliculogenesis and ovulation rate. *Reprod Nutr Dev*. 2006;46(4):339-54. doi: [10.1051/rnd:2006016](https://doi.org/10.1051/rnd:2006016). Epub 2006 Jul 7. PMID: 16824444.
17. Kurniadi A, Dewi AK, Sasotya RMS, et al. Effect of Vitamin D analog supplementation on levator ani strength and plasma Vitamin D receptor expression in uterine prolapse patients. *Sci Rep*. 2023;13(1):3616. doi: [10.1038/s41598-023-30842-2](https://doi.org/10.1038/s41598-023-30842-2). PMID: 36869168; PMCID: PMC9984360.
18. Rezaei H, Asefnejad A, Daliri-Joupari M, et al. In-vitro cellular and in-vivo investigation of ascorbic acid and  $\beta$ -glycerophosphate loaded gelatin/sodium alginate injectable hydrogels for urinary incontinence treatment. *Prog Biomater*. 2021;10(2):161-71. doi: [10.1007/s40204-021-00160-9](https://doi.org/10.1007/s40204-021-00160-9). Epub 2021 Jun 24. PMID: 34169484; PMCID: PMC8271082.
19. Schimpf M, Tulikangas P. Evolution of the female pelvis and relationships to pelvic organ prolapse. *Int Urogynecol J Pelvic Floor Dysfunct*. 2005;16(4):315-20. doi: [10.1007/s00192-004-1258-1](https://doi.org/10.1007/s00192-004-1258-1). Epub 2005 Jan 15. PMID: 15654501.
20. Hogervorst T, Bouma HW, de Vos J. Evolution of the hip and pelvis. *Acta Orthop Suppl*. 2009;80(336):1-39. doi: [10.1080/17453690610046620](https://doi.org/10.1080/17453690610046620). PMID: 19919389.
21. Jelovsek JE, Maher C, Barber MD. Pelvic organ prolapse. *Lancet*. 2007;369(9566):1027-38. doi: [10.1016/S0140-6736\(07\)60462-0](https://doi.org/10.1016/S0140-6736(07)60462-0). PMID: 17382829.
22. Guler Z, Roovers JP. Role of fibroblasts and myofibroblasts on the pathogenesis and treatment of pelvic organ prolapse. *Biomolecules*. 2022;12(1):94. doi: [10.3390/biom12010094](https://doi.org/10.3390/biom12010094). PMID: 35053242; PMCID: PMC8773530.
23. Ben Menachem-Zidon O, Gropp M, Reubinoff B, et al. Mesenchymal stem cell transplantation



- improves biomechanical properties of vaginal tissue following full-thickness incision in aged rats. *Stem Cell Reports*. 2022;17(11):2565-78. doi: [10.1016/j.stemcr.2022.09.005](https://doi.org/10.1016/j.stemcr.2022.09.005). Epub 2022 Oct 13. PMID: 36240774; PMCID: PMC9669396.
24. Sundrani D, Narang A, Mehendale S, et al. Investigating the expression of MMPs and TIMPs in preterm placenta and role of CpG methylation in regulating MMP-9 expression. *IUBMB Life*. 2017;69(12):985-93. doi: [10.1002/iub.1687](https://doi.org/10.1002/iub.1687). Epub 2017 Nov 11. PMID: 29130646.
  25. Wieslander CK, Rahn DD, McIntire DD, et al. Quantification of pelvic organ prolapse in mice: vaginal protease activity precedes increased MOPQ scores in fibulin 5 knockout mice. *Biol Reprod*. 2009;80(3):407-14. doi: [10.1095/biolreprod.108.072900](https://doi.org/10.1095/biolreprod.108.072900). Epub 2008 Nov 5. PMID: 18987327; PMCID: PMC2805390.
  26. Barat S, Bouzari Z, Mehdinia S, et al. The serum level of Vitamin D in women with urinary incontinence due to pelvic floor disorder and prolapse: a regional case-control study on Iranian population. *International Journal of Women's Health and Reproduction Sciences*. 2015;3(3):126-31. doi: [10.15296/ijwhr.2019.11](https://doi.org/10.15296/ijwhr.2019.11).
  27. Wang HL, Zhou C, Zhang YZ. [Role of matrix metalloproteinase-2,9 and their inhibitors in premature rupture of membranes]. *Zhonghua Fu Chan Ke Za Zhi*. 2005;40(1):29-33. Chinese. PMID: [15774089](https://pubmed.ncbi.nlm.nih.gov/15774089/).
  28. Maymon E, Romero R, Pacora P, et al. A role for the 72 kDa gelatinase (MMP-2) and its inhibitor (TIMP-2) in human parturition, premature rupture of membranes and intraamniotic infection. *J Perinat Med*. 2001;29(4):308-16. doi: [10.1515/JPM.2001.044](https://doi.org/10.1515/JPM.2001.044). PMID: 11565199.
  29. Strinic T, Vulic M, Tomic S, et al. Matrix metalloproteinases-1, -2 expression in uterosacral ligaments from women with pelvic organ prolapse. *Maturitas*. 2009;64(2):132-5. doi: [10.1016/j.maturitas.2009.08.008](https://doi.org/10.1016/j.maturitas.2009.08.008). Epub 2009 Sep 17. PMID: 19765922.