

Highly Recommended Dose of MK4 for Osteoporosis

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The recommended dose of Menatretene is 45 mg three times a day; however, the compliant in daily practice is not convenient. This study shows the twice dose per day is inferior to the recommended dose. This study used the level of Gla protein in osteocalcin as a parameter for the comparison. The mean of three-time dose a day is 11.27 nanogram per milliliter while the mean of the other group is 6.07 nanogram per milliliter after the three-month treatment.

Keywords: Dose, Menatretene

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Vitamin K is commonly used in practice. There are two forms, *i.e.* vitamin K1 has a prominent action on blood coagulation while vitamin K2 enhances the formation of osteocalcin. Practically, the selection of proper vitamin K will be a great benefit to the treatment. The first step of osteocalcin formation is formed the undercarboxylated osteocalcin. This process needs vitamin D as a cofactor for forming the immature osteocalcin

The osteocalcin has three residues of glutamic acids at the seventeenth, twenty first and twenty fourth positions of the osteocalcin molecule. These positions in the further step will be changed into the gamma carboxylic acids by vitamin K2 being a cofactor; the osteocalcin will later be matured. The protein is characterized by the presence of three residues of the gamma glutamic acid (Gla). This reactions are inhibited by warfarin⁽¹⁾.

Material and Method

The randomized control trial study (RCT) comprised of the patients participating in two groups; each group took 800 mg of elemental calcium and active form of vitamin D3 0.5 microgram per day. The

different doses of Vitamin K2, or Menatretene-4 given to both groups were: 45 mg/day and 30 mg/day. We used the level of GLA protein and Procollagen type 1 nitrogenous peptide (PINP) as the parameters for evaluation of efficacy of both groups.

The serum analysis for Gla-type osteocalcin EIA kit (Takara) is an *in vitro* enzyme immunoassay (EIA) kit for the quantitative determination of human Gla-OC in serum. The principle is based on the sandwich method that utilizes two-mouse monoclonal anti-Gla-OC antibodies to detect Gla-OC by the two-step procedure. Firstly, one of the anti-Gla-OC is fixed on the micro titer plate and blocked against the non-specific binding. The samples have to be prepared by being incubated in the microtiterplate. The second step is to wash the plate and add the second anti-OC labeled with peroxidase. During the incubation, the Gla-OC is bound to the anti Gla-OC on one side and tagged on the other by peroxidase. This reaction consequently produces different levels of color shades of which the intensities are proportional to the amount of Gla-OC presented in the samples, and then the standards are used in comparison. The accurate sample concentrations of Gla-OC can be determined by comparing their specific absorbance with those obtained for the standard plot on a standard curve.

The serum also investigated for bone formation marker, PINP (Roche).

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Patient characteristic

Type of group	Number	Year of menopause	Genders	Age
Three-time group (45 mg/d)	40	> 6 years	Female	50-70
Twice-a-day group (30mg/d)	40	> 6 years	Female	50-70

Results

The results were analyzed by paired-sample t-test, SPSS version 10. The group of 45 mg per day showed Gla protein before the vitamin K2 intake was 5.681 ng/ml after three months, the level of Gla-protein was increased, 11.27 ng/ml significantly ($p = 0.001$). The other groups taking 30 mg per day of vitamin K2: both the Gla-protein intake before and after were not changed (Table 1). The tid-group gave the high level of PINP significantly after adjusted the values of CTx ($p = 0.001$) comparing with the other group (Fig. 2).

Discussion

The osteocalcin formation needs vitamin D which induces the immature osteocalcin (Under-carboxylated osteocalcin, UcOC), but its function cannot perfectly completed. Thus, the formation is progressively developed to another step by using vitamin K2 as a cofactor for encouraging the immature osteocalcin to be the mature protein by changing the glutamic acid (Glu) at 17th, 21st, 24th positions into the gamma glutamic acid (Gla). At present, we can detect

the Gla protein by the enzyme immunoassay (EIA) (Takara)⁽²⁾. This examination is the evaluation of vitamin K2. The recommended dose of vitamin K2 is

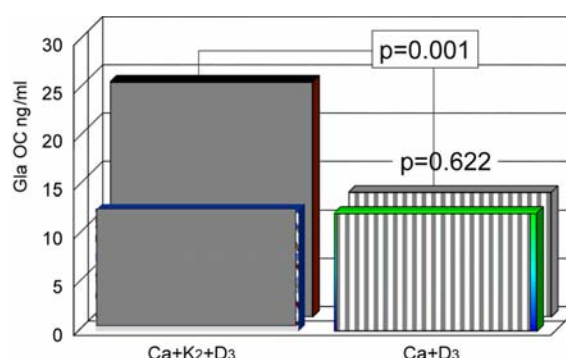


Fig. 1 The amount of Gla osteocalcin significantly increased in the group of vitamin K2 (Dark bar) compared to the base line control .The other group (Striped bar) of non vitamin K2 showed the Gla protein osteocalcin which was not changed after 3 months ($p = 0.622$). The final result could be concluded that the group of vitamin K2 dramatically expressed more Gla protein when compared with the other group ($p = 0.001$)

Table 1. The number of Gla-proteins of both major groups categorized into pre/post-tid and pre/post-bid. Pre/post-tid means a group of 45 mg/day intake and pre/post-bid means a group of 30 mg/day intake

	Gla protein	SD	p-value
Pre-tid	5.681	2.392	
Post-tid	11.27	4.848	0.001
Pre-bid	6.23	2.330	
Post-bid	6.075	2.154	0.622
	PINP	SD	p-value
Pre-tid	68.58	2.542	
Post-tid	84.22	3.121	0.001
Pre-bid	48.54	3.4	
Post-tid	52.42	4.12	0.26

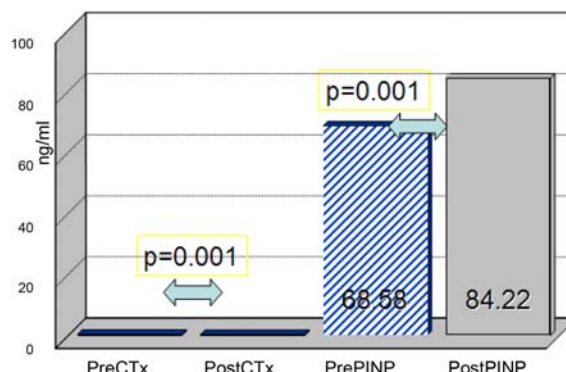


Fig. 2 The group of Menatretene + Calcium+ Active vitamin D3 showed mark increase of Procollagen type 1 nitrogenous peptide (PINP) after three months ($p = 0.001$), the other group had no response of increase PINP

normally three times a day or 45 mg per day, but the dose of twice a day is a practically good compliance. This study showed that the dose of two-time a day was inferior to the recommended dose (Fig. 1). The vitamin K2, Menatretene-4 has a short half life which cannot maintain the blood level of this vitamin so 45mg dose can keep the therapeutic level at all day. This study also supports that the two-time a day dose is not suitable for the osteocalcin formation.

Reference

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การศึกษาขนาดของยา วิตามิน เค สอง 45 มิลลิกรัม ให้ระดับของ Gla protein และเพิ่มการสร้างกระดูก (PINP) ดีกว่าขนาด 30 มิลลิกรัม

ณรงค์ บุญยะรัตเวช, นิพัทธ์ กิตติมานนท์, ธนัยชนก จิตติวิไล, บุญสม ตั้งทองทิพย์

การศึกษาเปรียบเทียบขนาดของยา วิตามิน เค สอง (Menatrenone-4) ในสตรีวัยหมดประจำเดือนสองกลุ่ม รวม 80 คน โดยได้รับแคลเซียม 800 มิลลิกรัม และวิตามิน ดี $1,25(\text{OH})_2 \text{D}_3$ ขนาด 0.5 ไมโครกรัมต่อวัน กลุ่มแรกได้รับยาวิตามิน เค สองขนาด 45 มิลลิกรัมต่อวัน กลุ่มที่สองได้ วิตามินเคสองขนาด 30 มิลลิกรัมต่อวัน เมื่อครบ 3 เดือน นำเลือดมาตรวจเปรียบเทียบกับเลือดก่อนได้รับยาโดยการตรวจหา Gla protein ด้วยวิธี immunoassay (EIA), Takara พบว่ากลุ่มที่ได้ วิตามินเค สองให้ Gla protein มากกว่าอีกกลุ่ม ($p = 0.001$).นอกจากนี้ ในกลุ่มนี้ยังให้ การสร้างคอลลาเจนมากกว่าอีกด้วย ($p = 0.001$) คือค่า PINPสูงขึ้นในเดือนที่สาม ดังนั้นวิตามินเค สอง ขนาด 45 มิลลิกรัมต่อวันจะให้ประสิทธิภาพของยาได้ดีกว่าตามที่สลากแนะนำไว้