

Review Article

Vitamin K and its Role in Diabetic Vascular Complications and Low-Grade Inflammation

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Abstract

This review summarizes the involvement of vitamin K and the extrahepatic Gla proteins matrix Gla protein (MGP), Osteocalcin (OC) and growth arrest-specific gene 6 protein (Gas6) in the development and progression of diabetes: in particular, complications related to angiopathy and inflammation. High vitamin K intake has been associated with a decreased risk of type 2 diabetes. Furthermore, in type 2 diabetic patients, the extent of artery calcification correlates to levels of the uncarboxylated Gla protein MGP, and supplementation with vitamin K has been shown to reduce oxidative stress markers as well as metabolic risk markers for diabetes.

Keywords: Diabetes; Gas6; Inflammation; MGP; Vitamin K; Vascular Disease

Introduction

Diabetes mellitus refers collectively to a heterogeneous set of diseases that share the common feature of hyperglycemia as a result of impaired metabolism [1]. The incidence of type 1 diabetes has steadily increased since the 1950s [2], and the current rise of type 2 diabetes [3] is likely to result in an even greater public health burden. Longtime diabetic complications include cardiovascular disease, chronic kidney disease and diabetic retinopathy, which contribute to increased mortality and an average lifespan reduction of 7 years [4]. In addition to preventive measures, it is therefore of utmost importance to further improve treatment and management of diabetes worldwide.

Fat-soluble nutrient vitamin K occurs naturally as vitamin K1 in leafy vegetables and as vitamin K2 in fermented food, such as cheese and curd. In addition, synthetic forms of vitamin K (K3-K5) exist. Structurally, all subspecies of vitamin K have an identical naphthoquinone ring structure accompanied by side chains of varying length and saturation [5]. Following absorption in the intestines, vitamin K is transported to the liver and redistributed

to extrahepatic tissues. Vitamin K undergoes a series of enzymatic modifications, referred to as the vitamin K cycle [6]. Vitamin K antagonists, such as warfarin, inhibit the enzyme vitamin K epoxide reductase, which halts vitamin K recycling.

The most established role of vitamin K is to function as a co-factor in post-translational γ -carboxylation of hepatic coagulation factors II, VII, IX and X as well as proteins C, S and Z. This modification enables the protein to bind calcium ions, upon which the protein adopts a tertiary structure and becomes biologically active [7]. The vitamin K-dependent proteins are termed Gla proteins, referring to the shift from glutamate (Glu) to γ -carboxyglutamate (Gla) in conjunction with γ -carboxylation. In addition to the aforementioned coagulation factors, a number of Gla proteins that originate from extrahepatic tissues have been identified. The most extensively researched extrahepatic Gla proteins are Matrix Gla Protein (MGP), Osteocalcin (OC) and growth arrest-specific gene 6 protein (Gas6), which are described in the following section.

Vitamin K has mainly been considered in the context of coagulation. However, since the discovery of its involvement in a vast number of other functions, such as atherosclerosis, bone metabolism, inflammation and cancer, it would be of interest to

review whether any relationship to diabetes exists.

The Extrahepatic Gla Proteins

MGP

MGP is mainly expressed by fibroblasts, chondrocytes and vascular smooth muscle cells and is a potent inhibitor of tissue calcification. The significance of MGP for vascular health was demonstrated by generating MGP-depleted mice, which died from aortic rupture before reaching two months of age [8]. Ectopic secretion of MGP from the liver did not reverse the calcification. However, with a local knock-in of MGP, the vascular abnormalities were avoided, indicating that the process is regulated locally and not systemically [9]. MGP has also been shown to bind and inhibit bone morphogenic protein 2 (BMP-2), which facilitates chondrogenesis and bone formation [10]. For the described actions, MGP requires carboxylation [11]. In addition to carboxylation, post-translational phosphorylation can modify MGP and is believed to affect transportation routes and the rate of secretion to extracellular surroundings [12]. Hence, MGP may be present in the circulation in different states of carboxylation and/or phosphorylation (p), and can be measured as p-cMGP, p-ucMGP, dp-cMGP and dp-ucMGP (dp=desphospho). Previous studies have confirmed an association between dp-ucMGP and vitamin K status [13] as well as a role for dp-ucMGP as an indicator of changes in the systemic vitamin K levels [14].

OC

OC is normally found in osteoblasts and odontoblasts. In its carboxylated form (cOC), it binds hydroxyapatite crystals with high affinity and inhibits mineral formation inside type I collagen fibrils, which are the main constituent of bone extracellular matrix [15]. Previous studies have used Fourier-transform infrared micro spectroscopy to demonstrate differences in mineral content; these differences are suggestive of impaired bone maturation in OC-deficient animals [16]. Studies suggest diabetic patients have altered bone metabolism and decreased bone turnover, demonstrated by lower OC and C-terminal cross-linked telopeptide concentrations [17].

Gas6

Gas6 shares similar structural features with protein S and has, since its discovery, been implicated in cell proliferation [18]. Gas6-mediated actions are achieved by binding receptor tyrosine kinases of the Tyro3, Axl and MerTK (TAM) family with subsequent activation of downstream signaling pathways [19]. Several studies have suggested involvement of Gas6 in diverse functions such as carcinogenesis [20], endothelial dysfunction [21], diabetes [22] and inflammation [23]. Results conflict in regard to Gas6 involvement in inflammatory signalling as both positive and negative correlations have been demonstrated. It has

been suggested that TAM receptors negatively regulate dendritic cells and macrophages but promote maturation of natural killer cells [24]. Similarly, studies are inconsistent regarding Gas6's relationship with diabetes [22,25-27] factors such as age, sex, Gas6 genotype and degree of γ -carboxylation as well as the complex nature of TAM signaling, could partially explain the discrepancy between the results. Large-scale studies elucidating the role of Gas6 are unfortunately lacking to date.

Vitamin K and Glucose Homeostasis - Preclinical Studies

It has been suggested that the extrahepatic Gla protein OC is involved in glucose homeostasis, following reports that OC-deficient mice demonstrated hyperglycemia, hypoinsulinemia and increased visceral fat as compared to wild type animals [28]. It was later proposed that the uncarboxylated form of OC (ucOC) is responsible for the endocrine properties by interacting with pancreatic β -cells through the G protein-coupled receptor family C group 6-member A (Gprc6a) receptor [29]. However, results are inconsistent and it is unclear whether these results can be extrapolated to humans. In studies investigating the relationship of OC to fat mass and body weight, no correlation was established with ucOC whereas cOc and adiponectin were independently associated with these variables [30].

Further supporting the theory that vitamin K has a positive impact on body composition, vitamin K2 (MK-4) has been shown to inhibit adipogenesis, stimulate alkaline phosphatase activity and reduce the expression of nuclear factor kappaB ligand/osteoclast differentiation factor in bone marrow cells in vitro [31]. These effects were not obtained with vitamin K1. This is in line with previous research indicating that vitamin K2 (MK-4) interacts with nuclear steroid and xenobiotic receptor, which is involved in multiple signaling pathways [32].

Vitamin K and Glucose Homeostasis

To date, large-scale studies investigating the relationship between vitamin K and glucose homeostasis are scarce. In the Framingham Offspring Study, which comprised 2719 participants, a high vitamin K1 intake was correlated to greater insulin sensitivity in both sexes [33]. Furthermore, in a cohort of 38,094 Dutch men and women, dietary intake of both vitamin K1 and vitamin K2 were associated with decreased risk of type 2 diabetes, with vitamin K2 also being correlated to more favorable lipid profiles and lower CRP levels [34]. The PREDIMED study demonstrated that dietary vitamin K₁, estimated from a food frequency questionnaire, was inversely associated with the risk of type 2 diabetes [35]. A cross-sectional study investigating vitamin K1 intake and its relation to the metabolic syndrome showed an inverse association between these two variables [36]. Similarly, vitamin K2 intake and high vitamin K status measured by dp-ucMGP have been associated

with lower occurrence of the metabolic syndrome [37]. When interpreting results related to vitamin K1 intake, it should be kept in mind that its main source is leafy greens and, therefore, a high vitamin K1 intake might only be a surrogate for a healthier life style.

A limited number of trials investigating the effect of vitamin K supplementation on metabolic disease have been published. When prediabetic women received supplementation with vitamin K1 for four weeks, the subjects showed improved glycemic status and increased insulin sensitivity [38]. In a study investigating the effects of vitamin K1 supplementation during 36 months on older, non-diabetic men and women (n = 355), results indicated that vitamin K reduced insulin resistance progression rate in men. This effect was not seen in women [38]. Another study, investigating 12 months of daily supplementation with vitamin K1 to postmenopausal women (n = 21), failed to demonstrate any association between vitamin K supplementation and glucose metabolism. Furthermore, changes in ucOC did not affect glucose metabolism [40].

Study findings on vitamin K2 supplementation are contradictory. Whereas some studies report improved insulin sensitivity [41], other studies that investigated supplementation with vitamin K2 found no association between MK-4 supplementation and circulating levels of leptin and adiponectin [30,42]. A systematic review of eight trials comprising 1077 participants concluded that vitamin K supplementation has no effect on insulin sensitivity. [43] However, this review admits to several limitations, such as a small number of studies and substantial heterogeneity in the analysis of fasting glucose levels and adiponectin, and emphasized the need for larger, well-designed Randomized Controlled Trials (RCTs). A recent randomized controlled intervention study not included in the aforementioned meta-analysis demonstrated that three years of supplementation with MK-7 may support a reduction of body weight, especially in subjects with a strong increase of cOC [44].

In conclusion, it is difficult to establish the role of vitamin K in glucose homeostasis and whether it has any clinical significance, based on the available data. Of the subspecies, it appears that vitamin K2 has produced the most promising results, which is in line with previous reviews on this topic [45,46]. Furthermore, based on the studies included in this review, the effect does not appear to be mediated by ucOC, but is rather related to increased cOC.

Vitamin K and Vascular Disease

Since studies have shown an increased prevalence of cardiovascular disease [47] as well as peripheral artery calcification [48] among diabetes patients, substantial effort has been made to survey different mechanisms that contribute to these complications. The vascular complications of diabetes not only increase the risk of premature death, but may also lead to crippling lower extremity

ulcers and amputations.

The role of vitamin K in vascular calcification has been extensively studied. High intake of vitamin K2 has been linked to decreased coronary artery calcification [49], an effect attributed mainly to increased carboxylation of extrahepatic Gla protein MGP. Correlations between levels of circulating dp-ucMGP and coronary artery calcification have been demonstrated in healthy women [50]. Furthermore, dp-ucMGP has been independently associated with the extent of peripheral arterial calcification among patients with type 2 diabetes [51]. High levels of circulating dp-ucMGP have also been correlated to increased risk of cardiovascular disease in patients with type 2 diabetes [52]. In another recent study, dp-ucMGP was associated with carotid-femoral pulse wave velocity (CF-PWV), a measure of large artery stiffness, in patients with type 2 diabetes [53]. Positive associations between PWV and circulating dp-ucMGP levels have also been observed in population-based studies [54].

Low levels of vitamin K result in reduced γ -carboxylation with a subsequent decrease in Gla protein activity. Poor vitamin K status could be due to inadequate intake, as studies suggest daily recommendations are not sufficient to include extrahepatic carboxylation. However, vitamin K deficiency could also result from decreased recycling, following impaired activity among enzymes involved in the vitamin K cycle. Type 2 diabetes patients demonstrate a higher frequency of the vitamin K epoxide reductase complex subunit 1 (VKORC1) AA genotype, which is further associated with decreased VKORC1 activity and functions as a predictor of intima-media calcification [55].

The accumulating evidence supporting the role of MGP in preventing vascular calcification has motivated several clinical trials. In healthy postmenopausal women, supplementation with MK-7 for three years improved arterial stiffness [56]. In addition, several ongoing trials are expected to reach completion in the near future, which hopefully will aid in establishing the role of vitamin K in vascular pathology [57].

Diabetes, Vitamin K and Inflammation

Diabetic angiopathy affects both small and large vessels, and results from inflammatory processes related to oxidative stress, protein glycation and endothelial dysfunction. Advanced glycation end products increase the production of reactive oxygen species and induce transcription of nuclear factor kappa-light-chain-enhancer of activated B cells (NF κ B), which in turn promotes inflammation and hypercoagulability [58].

The extrahepatic Gla protein Gas6 interacts with TAM receptors in order of decreasing affinity, Axl, Tyro3 and Mer which are involved in cell survival and migration as well as inflammatory signaling [59]. Various signaling pathways activated downstream of Gas6 have been proposed [60]; for example,

Gas6-mediated phosphorylation of Axl, which activates PI3/AKT. Peripheral glucose uptake is mediated by the glucose transporter type 4 protein, which translocate to the cell membrane upon insulin stimulation through PI3/AKT signaling [61]. Human microvascular endothelial cells exposed to high glucose levels showed decreased Gas6/Axl mRNA and down-regulation of AKT phosphorylation [62]. Gas6/TAM has also been shown to decrease inflammation by down-regulating toll-like receptors [63] and to stimulate proliferation in rat pancreatic beta cells [64].

In a study investigating Gas6 levels among adult healthy subjects and patients with insulin resistance or type 2 diabetes, decreased levels of Gas6 were demonstrated among diabetic patients. Gas6 levels were further negatively correlated to fasting glucose levels and inflammatory markers, such as tumor necrosis factor- α (TNF- α) and interleukin 6 (IL-6) [21]. However, in another study by the same research team with adolescent subjects, opposite results were demonstrated, indicating Gas6 was positively correlated with inflammation markers, fat mass and insulin resistance [65]. None of these studies considered carboxylation status of Gas6 or any marker for vitamin K status. Dietary intake of vitamin K1 has been shown to reduce the inflammatory cytokine burden and to improve metabolic risk markers for type 2 diabetes in high-risk populations [66].

In addition to anti-inflammatory effects mediated by Gla proteins, it is possible that vitamin K itself functions as an antioxidant. In healthy, postmenopausal women, supplementation with vitamins D3, K1 and B6 over one-year improved vitamin K status, as measured by decreased ucOC content, and reduced oxidative stress markers [67]. Vitamin K has also been shown to protect developing oligodendrocytes against oxidative damage by inhibiting activation of 12-lipoxygenase [68].

It was recently shown that initiating glucose-lowering treatment with metformin in VKA-treated patients decreased the activity of the anticoagulant therapy [69]. Since warfarin is primarily metabolized by cytochrome P450 (CYP)2C9 [70], the authors hypothesized that lowered insulin levels resulted in subsequent reduction of pro-inflammatory cytokines and higher expression of cytochrome p450 enzymes.

Conclusion

The research focus on vitamin K has shifted from hemostasis to include a broader range of functions, such as carcinogenesis, atherosclerosis and inflammation. High vitamin K intake has been associated with a decreased risk of type 2 diabetes but, due to conflicting results and study heterogeneity, larger well-designed RCTs are warranted. Furthermore, in type 2 diabetic patients, the extent of artery calcification correlates to levels of the uncarboxylated Gla protein MGP, and supplementation with vitamin K can reduce oxidative stress markers as well as metabolic

risk markers for diabetes. Trials investigating whether vitamin K2 supplementation may reduce or halt progression of vascular calcification are ongoing. In conclusion, the relationship between vitamin K2 subspecies and diabetes as well as their dose response effects on complications should be highlighted in future studies.

Conflicts of interest: There are no conflicts of interest for the authors of this review.

Ethics: There is no ethical background conflict or concern in this review as being a review not involving any original patient or experimental data no institutional or national ethical committee approval has been included in the manuscript.

References

1. Kerner W, Bruckel J (2014) Definition, classification and diagnosis of diabetes mellitus. *Experimental and clinical endocrinology & diabetes: official journal, German Society of Endocrinology [and] German Diabetes Association* 122: 384-386.
2. DIAMOND Project Group (2006). Incidence and trends of childhood Type 1 diabetes worldwide 1990-1999. *Diabet Med* 23:857-866.
3. Sharma M, Nazareth I, Petersen I (2016) Trends in incidence, prevalence and prescribing in type 2 diabetes mellitus between 2000 and 2013 in primary care: a retrospective cohort study. *BMJ open* 6: e010210.
4. Morgan CL, Currie CJ, Peters JR (2000) Relationship between diabetes and mortality: a population study using record linkage. *Diabetes care* 23: 1103-1107.
5. Shearer MJ (1992) Vitamin K metabolism and nutrition. *Blood reviews* 6: 92-104.
6. Tie JK, Stafford DW (2016) Structural and functional insights into enzymes of the vitamin K cycle. *Journal of thrombosis and haemostasis JTH* 14: 236-247.
7. Vermeer C (1990) Gamma-carboxyglutamate-containing proteins and the vitamin K-dependent carboxylase. *The Biochemical journal* 266: 625-636.
8. Luo G, Ducey P, McKee MD, Pinero GJ, Loyer E, et al. (1997) Spontaneous calcification of arteries and cartilage in mice lacking matrix GLA protein. *Nature* 386: 78-81.
9. Murshed M, Schinke T, McKee MD, Karsenty G (2004) Extracellular matrix mineralization is regulated locally; different roles of two gla-containing proteins. *The Journal of cell biology* 165: 625-630.
10. Sweatt A, Sane DC, Hutson SM, Wallin R (2003) Matrix Gla protein (MGP) and bone morphogenetic protein-2 in aortic calcified lesions of aging rats. *Journal of thrombosis and haemostasis JTH* 1: 178-185.
11. Wallin R, Schurgers L, Wajih N (2008) Effects of the blood coagulation vitamin K as an inhibitor of arterial calcification. *Thromb Res* 122: 411-417.
12. Wajih N, Borrás T, Xue W, Hutson SM, Wallin R (2004) Processing and transport of matrix gamma-carboxyglutamic acid protein and bone morphogenetic protein-2 in cultured human vascular smooth muscle cells: evidence for an uptake mechanism for serum fetuin. *The Journal of biological chemistry* 279: 43052-43060.

13. Shea MK, O'Donnell CJ, Vermeer C, Magdeleyns EJ, Crosier MD, et al. (2011) Circulating uncarboxylated matrix gla protein is associated with vitamin K nutritional status, but not coronary artery calcium, in older adults. *The Journal of nutrition* 141: 1529-1534.
14. Cranenburg EC, Koos R, Schurgers LJ, Magdeleyns EJ, Schoonbrood TH, et al. (2010) Characterisation and potential diagnostic value of circulating matrix Gla protein (MGP) species. *Thrombosis and haemostasis* 104: 811-822.
15. Hunter GK, Hauschka PV, Poole AR, Rosenberg LC, Goldberg HA (1996) Nucleation and inhibition of hydroxyapatite formation by mineralized tissue proteins. *The Biochemical journal* 317: 59-64.
16. Boskey AL, Gadaleta S, Gundberg C, Doty SB, Ducey P, et al. (1998) Fourier transform infrared microspectroscopic analysis of bones of osteocalcin-deficient mice provides insight into the function of osteocalcin. *Bone* 23: 187-196.
17. Starup-Linde J, Eriksen SA, Lykkeboe S, Handberg A, Vestergaard P (2014) Biochemical markers of bone turnover in diabetes patients--a meta-analysis, and a methodological study on the effects of glucose on bone markers. *Osteoporosis international: a journal established as result of cooperation between the European Foundation for Osteoporosis and the National Osteoporosis Foundation of the USA* 25: 1697-1708.
18. Manfioletti G, Brancolini C, Avanzi G, Schneider C (1993) The protein encoded by a growth arrest-specific gene (gas6) is a new member of the vitamin K-dependent proteins related to protein S, a negative coregulator in the blood coagulation cascade. *Mol Cell Biol* 13: 4976-4985.
19. Fernandez-Fernandez L, Bellido-Martin L, Garcia de Frutos P (2008) Growth arrest-specific gene 6 (GAS6). An outline of its role in haemostasis and inflammation. *Thrombosis and haemostasis* 100: 604-610.
20. Wu G, Ma Z, Hu W, Wang D, Gong B, et al. (2017) Molecular insights of Gas6/TAM in cancer development and therapy. *Cell death & disease* 8: e2700.
21. Hung YJ, Lee CH, Chu NF, Shieh YS (2010) Plasma protein growth arrest-specific 6 levels are associated with altered glucose tolerance, inflammation, and endothelial dysfunction. *Diabetes care* 33: 1840-1844.
22. Fouad NA, Eltahir SM, Abdullah OA, Metwally RA (2015) Serum Level of Growth Arrest-Specific 6 (Gas6) Protein and Genetic Variations in the Gas6 Gene in Patients with Type 2 Diabetes Mellitus. *The Egyptian journal of immunology* 22: 41-47.
23. Sen P, Wallet MA, Yi Z, Huang Y, Henderson M, et al. (2007) Apoptotic cells induce Mer tyrosine kinase-dependent blockade of NF-kappaB activation in dendritic cells. *Blood* 109: 653-660.
24. Lemke G, Rothlin CV (2008) Immunobiology of the TAM receptors. *Nature reviews Immunology* 8: 327-336.
25. Lee CH, Chu NF, Shieh YS, Hung YJ (2012) The growth arrest-specific 6 (Gas6) gene polymorphism c.834+7G>A is associated with type 2 diabetes. *Diabetes research and clinical practice* 95: 201-206.
26. Wu KS, Hung YJ (2015) The Involvement of GAS6 Signaling in the Development of Obesity and Associated Inflammation: 202513.
27. Kuo FC, Hung YJ, Shieh YS, Hsieh CH, Hsiao FC, et al. (2014) The levels of plasma growth arrest-specific protein 6 is associated with insulin sensitivity and inflammation in women. *Diabetes research and clinical practice* 103: 304-309.
28. Lee NK, Sowa H, Hinoi E, Ferron M, Ahn JD, et al. (2007) Endocrine regulation of energy metabolism by the skeleton. *Cell* 130: 456-469.
29. Wei J, Hanna T, Suda N, Karsenty G, Ducey P (2014) Osteocalcin promotes beta-cell proliferation during development and adulthood through Gprc6a. *Diabetes* 63: 1021-1031.
30. Knapen MH, Schurgers LJ, Shearer MJ, Newman P, Theuvsissen E, et al. (2012) Association of vitamin K status with adiponectin and body composition in healthy subjects: uncarboxylated osteocalcin is not associated with fat mass and body weight. *The British journal of nutrition* 108: 1017-1024.
31. Takeuchi Y, Suzawa M, Fukumoto S, Fujita T (2000) Vitamin K (2) inhibits adipogenesis, osteoclastogenesis, and ODF/RANK ligand expression in murine bone marrow cell cultures. *Bone* 27: 769-776.
32. Shearer MJ, Newman P (2008) Metabolism and cell biology of vitamin K. *Thrombosis and haemostasis* 100: 530-547.
33. Yoshida M, Booth SL, Meigs JB, Saltzman E, Jacques PF (2008) Phylloquinone intake, insulin sensitivity, and glycemic status in men and women. *The American journal of clinical nutrition* 88: 210-215.
34. Beulens JW, van der AD, Grobbee DE, Sluijs I, Spijkerman AM, et al. (2010) Dietary phyloquinone and menaquinones intakes and risk of type 2 diabetes. *Diabetes care* 33: 1699-1705.
35. Ibarrola-Jurado N, Salas-Salvado J, Martinez-Gonzalez MA, Bullo M (2012) Dietary phyloquinone intake and risk of type 2 diabetes in elderly subjects at high risk of cardiovascular disease. *The American journal of clinical nutrition* 96: 1113-1118.
36. Pan Y, Jackson RT (2009) Dietary phyloquinone intakes and metabolic syndrome in US young adults. *Journal of the American College of Nutrition* 28: 369-379.
37. Dam V, Dalmeijer GW, Vermeer C, Drummen NE, Knapen MH, et al. (2015) Association Between Vitamin K and the Metabolic Syndrome: A 10-Year Follow-Up Study in Adults. *The Journal of clinical endocrinology and metabolism* 100: 2472-2479.
38. Rasekhi H, Karandish M, Jalali MT, Mohammad-Shahi M, Zarei M, et al. (2015) The effect of vitamin K1 supplementation on sensitivity and insulin resistance via osteocalcin in prediabetic women: a double-blind randomized controlled clinical trial. *European journal of clinical nutrition* 69: 891-895.
39. Yoshida M, Jacques PF, Meigs JB, et al. (2008) Effect of vitamin K supplementation on insulin resistance in older men and women. *Diabetes care* 31: 2092-2096.
40. Kumar R, Binkley N, Vella A (2010) Effect of phyloquinone supplementation on glucose homeostasis in humans. *The American journal of clinical nutrition* 92: 1528-1532.
41. Choi HJ, Yu J, Choi H, et al. (2011) Vitamin K2 supplementation improves insulin sensitivity via osteocalcin metabolism: a placebo-controlled trial. *Diabetes care* 34: e147.
42. Koitaya N, Sekiguchi M, Tosen Y, Nishide Y, Morita A, et al. (2014) Low-dose vitamin K2 (MK-4) supplementation for 12 months improves bone metabolism and prevents forearm bone loss in postmenopausal Japanese women. *Journal of bone and mineral metabolism* 32: 142-150.
43. Suksomboon N, Poolsup N, Darli Ko Ko H (2017) Effect of vitamin K supplementation on insulin sensitivity: a meta-analysis. *Diabetes, metabolic syndrome and obesity: targets and therapy* 10: 169-177.

44. Knapen MHJ, Jardon KM, Vermeer C (2017) Vitamin K-induced effects on body fat and weight: results from a 3-year vitamin K2 intervention study. *European journal of clinical nutrition* 72:136-141.
45. Rees K, Guraewal S, Wong YL, Majanbu DL, Mavrodaris A, et al. (2010) Is vitamin K consumption associated with cardio-metabolic disorders? A systematic review. *Maturitas* 67: 121-128.
46. Valdes-Ramos R, Guadarrama-Lopez AL, Martinez-Carrillo BE, Benitez-Arciniega AD (2015) Vitamins and type 2 diabetes mellitus. *Endocrine, metabolic & immune disorders drug targets* 15: 54-63.
47. Barr EL, Zimmet PZ, Welborn TA, Jolley D, Magliano DJ, et al. (2007) Risk of cardiovascular and all-cause mortality in individuals with diabetes mellitus, impaired fasting glucose, and impaired glucose tolerance: the Australian Diabetes, Obesity, and Lifestyle Study (AusDiab). *Circulation* 116: 151-157.
48. Yang SL, Zhu LY, Han R, Sun LL, Li JX, et al. (2017) Pathophysiology of peripheral arterial disease in diabetes mellitus. *Journal of diabetes* 9: 133-140.
49. Beulens JW, Bots ML, Atsma F, Bartelink ML, Prokop M, et al. (2009) High dietary menaquinone intake is associated with reduced coronary calcification. *Atherosclerosis* 203: 489-493.
50. Dalmeijer GW, van der Schouw YT, Vermeer C, Magdeleyns EJ, Schurgers LJ, et al. (2013) Circulating matrix Gla protein is associated with coronary artery calcification and vitamin K status in healthy women. *The Journal of nutritional biochemistry* 24: 624-628.
51. Liabeuf S, Bourron O, Vermeer C, Theuvsissen E, Magdeleyns E, et al. (2014) Vascular calcification in patients with type 2 diabetes: the involvement of matrix Gla protein. *Cardiovascular diabetology* 13: 85.
52. Dalmeijer GW, van der Schouw YT, Magdeleyns EJ, Vermeer C, Verschuren WM, et al. (2013) Matrix Gla protein species and risk of cardiovascular events in type 2 diabetic patients. *Diabetes care* 36: 3766-3771.
53. Sardana M, Vasim I, Varakantam S, Kewan U, Tariq A, et al. (2017) Inactive Matrix Gla-Protein and Arterial Stiffness in Type 2 Diabetes Mellitus. *American journal of hypertension* 30: 196-201.
54. Pivin E, Ponte B, Pruijm M, Ackermann D, Guessous I, et al. (2015) Inactive Matrix Gla-Protein Is Associated with Arterial Stiffness in an Adult Population-Based Study. *Hypertension (Dallas, Tex : 1979)* 66: 85-92.
55. Tavridou A, Petridis I, Vasileiadis M, Ragia G, Heliopoulos I, et al. (2011) Association of VKORC1 -1639 G>A polymorphism with carotid intima-media thickness in type 2 diabetes mellitus. *Diabetes research and clinical practice* 94: 236-241.
56. Knapen MH, Braam LA, Drummen NE, Bekers O, Hoeks A, et al. (2015) Menaquinone-7 supplementation improves arterial stiffness in healthy postmenopausal women. A double-blind randomised clinical trial. *Thrombosis and haemostasis* 113: 1135-1144.
57. Caluwe R, Pyfferoen L, De Boeck K, De Vriese AS (2016) The effects of vitamin K supplementation and vitamin K antagonists on progression of vascular calcification: ongoing randomized controlled trials. *Clinical kidney journal* 9: 273-279.
58. Domingueti CP, Dusse LM, Carvalho M, de Sousa LP, Gomes KB, et al. (2016) Diabetes mellitus: The linkage between oxidative stress, inflammation, hypercoagulability and vascular complications. *Journal of diabetes and its complications* 30: 738-745.
59. Korshunov VA (2012) Axl-dependent signalling: a clinical update. *Clinical science (London, England: 1979)* 122: 361-368.
60. Dihingia A, Kalita J, Manna P (2017) Implication of a novel Gla-containing protein, Gas6 in the pathogenesis of insulin resistance, impaired glucose homeostasis, and inflammation: A review. *Diabetes research and clinical practice* 128: 74-82.
61. Thong FS, Dugani CB, Klip A (2005) Turning signals on and off: GLUT4 traffic in the insulin-signaling highway. *Physiology (Bethesda, Md)* 20: 271-284.
62. Lee CH, Shieh YS, Hsiao FC, Kuo FC, Lin CY, et al. (2014) High glucose induces human endothelial dysfunction through an Axl-dependent mechanism. *Cardiovascular diabetology* 13: 53.
63. Rothlin CV, Ghosh S, Zuniga EI, Oldstone MB, Lemke G (2007) TAM receptors are pleiotropic inhibitors of the innate immune response. *Cell* 131: 1124-1136.
64. Haase TN, Rasmussen M, Jaksch CA, Gaarn LW, Petersen CK, et al. (2013) Growth arrest specific protein (GAS) 6: a role in the regulation of proliferation and functional capacity of the perinatal rat beta cell. *Diabetologia* 56: 763-773.
65. Hsiao FC, Lin YF, Hsieh PS, Chu NF, Shieh YS, et al. (2013) Circulating growth arrest-specific 6 protein is associated with adiposity, systemic inflammation, and insulin resistance among overweight and obese adolescents. *The Journal of clinical endocrinology and metabolism* 98: E267-E274.
66. Juanola-Falgarona M, Salas-Salvado J, Estruch R, Portillo MP, Casas R, et al. (2013) Association between dietary phyloquinone intake and peripheral metabolic risk markers related to insulin resistance and diabetes in elderly subjects at high cardiovascular risk. *Cardiovascular diabetology* 12: 7.
67. Mazzanti L, Battino M, Nanetti L, Raffaelli F, Alidori A, et al. (2015) Effect of 1-year dietary supplementation with vitaminized olive oil on markers of bone turnover and oxidative stress in healthy post-menopausal women. *Endocrine* 50: 326-334.
68. Li J, Wang H, Rosenberg PA (2009) Vitamin K prevents oxidative cell death by inhibiting activation of 12-lipoxygenase in developing oligodendrocytes. *Journal of neuroscience research* 87: 1997-2005.
69. Stage TB, Pottegard A, Henriksen DP, Christensen MM1, Højlund K, et al. (2016) Initiation of glucose-lowering treatment decreases international normalized ratio levels among users of vitamin K antagonists: a self-controlled register study. *J Thromb Haemost* 14: 129-133.
70. Johnson JA, Cavallari LH (2015) Warfarin pharmacogenetics. *Trends in cardiovascular medicine* 25: 33-41.