

## HUMAN NUTRITION

**Title:** Metabolism of Vitamin K Forms in Fresh Pork - #17-003 IPPA

**Investigator:** Xueyan Fu, PhD

**Institution:** Jean Mayer USDA Human Nutrition Research Center on Aging  
at Tufts University

**Date Submitted:** March 28, 2019

**Scientific Abstract:**

**Objective:** Vitamin K (VK) exists in multiple forms. Plant-based phylloquinone (PK) is considered the predominant dietary VK form. However, recent studies have shown bacterial and animal-based forms of VK, called menaquinones (MK<sub>n</sub>; n=number of prenyl units in side chain), are prevalent in foods such as dairy, fermented products, and meat, including pork. Bacterially-produced MK<sub>n</sub> are also synthesized by the gut microbiota. It is unknown if MK<sub>n</sub> are absorbed and metabolized. The objective of this study was to compare MK<sub>n</sub> concentrations in blood, tissue, and feces of mice given purified and food-based MK<sub>n</sub> to mice fed a VK-deficient (VKD, “control”) diet.

**Methods:** Sixty male and 60 female 10-week old C57BL6 mice were acclimated on a VKD diet. After 4 weeks, mice were randomized to six groups and given a diet containing purified dietary VK forms (“PK”, “MK4”, “MK9”, or an equimolar combination, “Combo”), pork (24% of diet, “Pork”), or maintained on “VKD” diet for 4 weeks. VK forms in diets, blood, tissues, and feces were measured using LC-MS, and compared by diet group and sex using 2-way ANOVA. Bacterial DNA was extracted from fecal samples and sequenced using Illumina HiSeq to analyze bacterial community composition. Differences in composition by sex and diet group were determined using non-parametric PERMANOVA, and bacterial richness was compared by sex and by diet group using Welch’s t-test.

**Results:** The VK content of the study diets were as follows (in µg/kg diet): **control:** 20.6 ± 2.0 PK; **PK:** 3200 ± 932 PK; **MK4:** 10.2 ± 0.4 PK and 2150 ± 184 MK4; **MK9:** 9.2 ± 1.6 PK and 3820 ± 598 MK9; **Combo:** 925 ± 173 PK, 821 ± 130 MK4, and 1310 ± 214 MK9; and **Pork:** 11.1 ± 0.6 PK, 38.5 ± 7.8 MK4, 34.7 ± 9.8 MK9, and 33.2 ± 10.5 MK10. In all analyzed tissues, tissue accumulation of VK was greater in females as compared to males. VK content of the small intestine and liver reflected vitamin K present in the diet, such that PK, MK4, and MK9 were significantly higher in groups supplemented that form as compared to the control group (all p<0.03). Female mice had significantly higher bacterial richness compared to male mice (Welch’s t-test, t = 2.2699, df = 92.739, p-value = 0.02553) while no significant differences in bacterial richness were observed between dietary regiments.

---

These research results were submitted in fulfillment of checkoff-funded research projects. This report is published directly as submitted by the project’s principal investigator. This report has not been peer-reviewed.

---

For more information contact:

National Pork Board • PO Box 9114 • Des Moines, IA 50306 USA • 800-456-7675 • Fax: 515-223-2646 • pork.org

---

**Conclusions:** MKn forms in small intestine and liver reflect dietary MKn intake. There is sex difference for VK forms absorption. Fecal MKn partially reflected intake, but endogenous production of MKn was largely unaffected by dietary VK. Despite abundant endogenous MKn production, tissue VK was low in the control group suggesting tissue accumulation of VK is independent of MKn produced in the gut.