

# INTRODUCTION

## Breastfeeding

Breastfeeding is the gold standard of infant feeding. The recognition of breastfeeding as the most favorable method of infant feeding has spread worldwide.<sup>(1)</sup> The numerous maternal and child health benefits of breastfeeding have been widely acknowledged, with an increase in the awareness of the advantages of breast milk over formula.<sup>(2)</sup> Yet breastfeeding rates continue to be unsatisfactory despite the known advantages of breastfeeding. Evidence from the literature overwhelmingly indicates that this optimal method of infant feeding should be encouraged in order to ensure the best possible health outcomes for women and their children.<sup>(3)</sup> Moreover, breastfeeding is now globally accepted as the gold standard for infant nutrition. The current recommendations are exclusive breastfeeding for the first six months of the newborn's life, and then continued breastfeeding at least through the first year.<sup>(4-6)</sup>

## Breastfeeding Rates

### Global Breastfeeding Rates

Despite these known benefits, critically low breastfeeding rates persist worldwide. The occurrence of suboptimal breastfeeding practices has significantly affected maternal and infant health and morbidity and mortality worldwide.<sup>(7,8)</sup> Among children less than five years old in developing countries, up to 1.5 million deaths per year and 10% of disease morbidity are attributed to lack of immediate and exclusive breastfeeding in infancy.<sup>(7,8)</sup>

According to the WHO (2003), less than 35% of infants worldwide are exclusively breastfed during the first four months of life; supplementary feeding either begins too early or too late, and liquid and solid foods given to infants and children are often nutritionally inadequate and unsafe.<sup>(9)</sup> Improving the global health problem of malnutrition among children can be addressed through focusing on infant nutrition and breastfeeding. Engaging in breastfeeding promotion efforts that target mothers of infants is an attempt to ensure that each child is provided with the highest standard of health.<sup>(10)</sup>

### National breast feeding rates

Although breastfeeding rates are slowly increasing, they remain still very low. The government has made very little efforts to support and promote breastfeeding, despite its well known impact on reducing under 5 infant and neonatal mortality rates. The major causes of infant mortality in 2008 were: neonatal causes (61%); pneumonia (7%); diarrhea (5%).<sup>(11)</sup>

A study was conducted on 2011 to determine prevalence of exclusive breastfeeding during the first 6 months of life in rural areas in Egypt found 95.8% of mothers (1,015 of 1,059) were breastfeeding their babies, whereas only 9.7% (103 of 1,059) were exclusively breastfeeding their infants for 6 months. Among the different sociodemographic, maternal, and infant factors studied, only antenatal care (four or more visits), early breastfeeding

initiation after delivery, male infant, and absence of breastfeeding difficulties were the significant predictors associated with higher chance for exclusive breastfeeding.<sup>(12)</sup>

### **Benefits of Breast feeding**

Breastfeeding has psychological, nutritional, and immunological benefits for the infant in addition to maternal and economic benefits. A new baby benefits from breast milk in a myriad of ways. Both short-term and lifelong advantages to a breastfed child's health have been documented by research. In the first months of an infant's life and beyond, breast milk provides nutrition and immune protection that breast milk substitutes cannot emulate. A live substance, breast milk is constantly changing to protect an infant against illness.<sup>(13)</sup>

**For the infants**, according to the American Academy of Pediatrics, breastfeeding decreases "the incidence and/or severity of a wide range of infectious diseases including bacterial meningitis, bacteremia, diarrhea, respiratory tract infection, necrotizing enterocolitis, otitis media, urinary tract infection, and late-onset sepsis in preterm infants".<sup>(6)</sup> Additionally, studies show a decreased risk of sudden infant death syndrome (SIDS) as well as decreases in rates of "type 1 diabetes, type 2 diabetes, lymphoma, leukemia, Hodgkin disease, overweight and obesity, hypercholesterolemia, and asthma in older children and adults who were breastfed, compared with individuals who were not breastfed".<sup>(14)</sup>

**For the mothers** in the short term, breastfeeding has been known to decrease the risk of postpartum hemorrhage, increase pregnancy spacing as a result of lactational amenorrhea, and aid in post-pregnancy weight loss.<sup>(15)</sup> There is also a decreased risk of premenopausal breast cancer and ovarian cancer in women who breastfeed.<sup>(14)</sup> Breastfeeding also decreases the risk of type two diabetes and uterine cancer and osteoporosis.<sup>(16)</sup>

### **Breast milk composition**

Breast milk contains all the nutrients that an infant needs in the first 6 months of life, including fat, carbohydrates, proteins, vitamins, minerals and water.<sup>(17)</sup> It is easily digested and efficiently used. Breast milk also contains bioactive factors that augment the infant's immature immune system, providing protection against infection, and other factors that help digestion and absorption of nutrients.<sup>(18)</sup>

#### **Fats**

Breast milk contains about 3.5 g of fat per 100 ml of milk, which provides about one half of the energy content of the milk. The fat is secreted in small droplets, and the amount increases as the feed progresses. As a result, the hindmilk secreted towards the end of a feed is rich in fat and looks creamy white, while the foremilk at the beginning of a feed contains less fat and looks somewhat bluish-grey in colour. Breast-milk fat contains long chain polyunsaturated fatty acids {docosahexaenoic acid(DHA) and arachidonic acid (ARA)} that are not available in other milks. These fatty acids are important for the neurological development of a child. DHA and ARA are added to some varieties of infant formula, but this does not confer any advantage over breast milk, and may not be as effective as those in breast milk.<sup>(18)</sup>

### **Carbohydrates**

The main carbohydrate is the special milk sugar lactose, a disaccharide. Breast milk contains about 7 g lactose per 100 ml, which is more than in most other milks, and is another important source of energy. Another kind of carbohydrate present in breast milk is oligosaccharides, or sugar chains, which provide important protection against infection.<sup>(17)</sup>

### **Protein**

Breast milk protein differs in both quantity and quality from animal milks, and it contains a balance of amino acids which makes it much more suitable for a baby. The concentration of protein in breast milk (0.9 g per 100 ml) is lower than in animal milks. The much higher protein in animal milks can overload the infant's immature kidneys with waste nitrogen products. Breast milk contains less of the protein casein, and this casein in breast milk has a different molecular structure. It forms much softer, more easily-digested curds than that in other milks. Among the whey, or soluble proteins, human milk contains more alpha-lactalbumin; cow milk contains betalactoglobulin, which is absent from human milk and to which infants can become intolerant.<sup>(17)</sup>

### **Anti-infective factors**

Breast milk contains many factors that help to protect an infant against infection including: sIgA and IgM transferred to the infant via breast milk play an important role in the innate mucosal immune protection of the infant. These antibodies can block the adherence and entry of microorganisms and cause inactivation, neutralization, or agglutination of viruses. Secretory IgA and IgM in human milk are active against a litany of viruses including enteroviruses, herpes viruses, respiratory syncytial virus, rubella, and rotavirus. Many bacteria are targeted by sIgA in human milk, including *E. coli*, *Shigella*, *Salmonella*, *Campylobacter*, *Vibrio cholerae*, *H. influenzae*, *S. pneumoniae*, *Clostridium difficile*, and *C. botulinum*, *Klebsiella pneumoniae*, as well as the parasite: *Giardia* and the fungus: *Candida albicans*.<sup>(19)</sup>

Other factors contained in breast milk that act primarily at the mucosal level include: lactoferrin, lysozyme, casein, oligosaccharides (prebiotics), glycoconjugates, and lipids.<sup>(19)</sup>

Lactoferrin has a high affinity for iron, which may limit the available iron required by microorganisms for growth. Also it has separate bactericidal and antiviral properties as well. Partially hydrolyzed lactoferrin seems to block adsorption or penetration of specific viruses, such as herpes simplex virus, cytomegalovirus, and even HIV. Lactoferrin can interfere with the adhesion of enteral pathogens Enterotoxigenic *E. coli* and *Shigella flexneri*. Lactoferrin may also increase the growth of probiotic intestinal bacterial.<sup>(19)</sup>

Lysozyme, which seems to act by lysing bacteria, maintains high concentrations throughout lactation. Casein inhibits the adherence of microorganisms to mucosal and epithelial cells (eg, *Helicobacter pylori*, *S. pneumoniae*, *H. influenzae*). A fragment of proteolysis of k-casein promotes the growth of *Bifidobacterium bifidum*, an important organism in the infant's microflora and a recognized probiotic bacterium.<sup>(19)</sup>

Glycoconjugates and oligosaccharides function as ligands, binding to bacteria, toxins, and viruses, blocking the ability of these harmful organisms to bind to the infant's epithelial cells.<sup>(19)</sup>

Mucin-1, lacadherin, and glycosaminoglycan are specifically identified antimicrobial components in the milk-fat globule membrane. There are also immune modulating agents within breast milk especially cytokines and growth factors which can act at the level of the mucosa. IL-10 and IFN- act to modulate epithelial barrier integrity.<sup>(19)</sup>

### **Other bioactive factors**

**Bile-salt stimulated lipase** facilitates the complete digestion of fat once the milk has reached the small intestine. Fat in artificial milks is less completely digested.<sup>(17)</sup>

**Epidermal growth factor** stimulates maturation of the lining of the infant's intestine, so that it is better able to digest and absorb nutrients, and is less easily infected or sensitised to foreign proteins. It has been suggested that other growth factors present in human milk target the development and maturation of nerves and retina.<sup>(20)</sup>

### **Vitamins and minerals**

Breast milk normally contains sufficient vitamins for an infant, unless the mother herself is deficient.<sup>(21)</sup> The exception is vitamin D. The infant needs exposure to sunlight to generate endogenous vitamin D or, if this is not possible, a supplement. The minerals iron and zinc are present in relatively low concentration, but their bioavailability and absorption is high. Provided that maternal iron status is adequate, term infants are born with a store of iron to supply their needs; only infants born with low birth weight may need supplements before 6 months. Delaying clamping of the cord until pulsations have stopped (approximately 3 minutes) has been shown to improve infants' iron status during the first 6 months of life.<sup>(22,23)</sup>

### **Vitamin K in human milk**

**Human milk** is a poor source of vitamin K, containing 1000– 4000 ng/l, with high intra- and interindividual variability. Vitamin K levels in cord blood are also very low, about 1/30 of the levels in maternal plasma.<sup>(24)</sup> Vitamin K, in breast milk at 2, 6, 12, and 26 weeks was averaging 1170±700, 950±500, 1150±620, and 870±500 ng/l, respectively. This may be secondary to low maternal vitamin K intakes.<sup>(25)</sup>

The low vitamin K plasma concentration in neonates is determined by a small transplacental passage from the mother to the infant and compared to adults they have virtually no hepatic stores of menaquinones (vitamin K<sub>2</sub>).<sup>(26)</sup> This vitamin is synthesized by bacteria of the gut. Most of the bacteria comprising the normal intestinal flora of human milk fed infants do not produce menaquinones, including *Bifidobacterium*, *Lactobacillus* and *Clostridium* species. Bacteria that produce menaquinones include *Bacteroides fragilis* and *Escherichia coli*, which are more common in formula fed infants.<sup>(27)</sup>

## **Vitamin K**

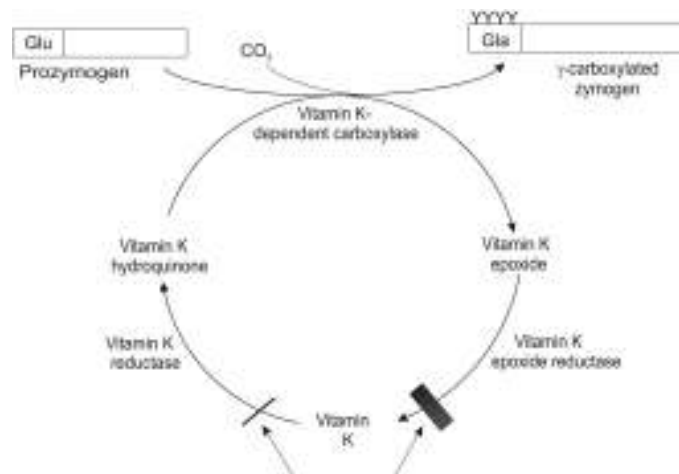
Vitamin K is an essential fat-soluble micronutrient which is needed for a unique post-translational chemical modification in a small group of proteins with calcium-binding properties, collectively known as vitamin K - dependent proteins or Gla-proteins. Thus far, the only obvious role of vitamin K in health is in the maintenance of normal coagulation. The vitamin K- dependent coagulation proteins are synthesized in the liver and comprise factors II, VII, IX, and X, which have a haemostatic role (i.e., they are procoagulants that arrest and prevent bleeding), and proteins C and S, which have an anticoagulant role (i.e., they inhibit the clotting process). Vitamin K dependent proteins synthesized by other tissues include the bone protein osteocalcin and matrix Gla protein; their functions remain to be clarified.<sup>(28)</sup>

### **Biological role of vitamin K**

Vitamin K is the family name for a series of fat-soluble compounds, which have a common 2-methyl-1, 4-naphthoquinone nucleus but differ in the structures of a side chain at the 3-position. They are synthesized by plants and bacteria. In plants the only important molecular form is phyloquinone (vitamin K<sub>1</sub>), which has a phytyl side chain. Bacteria synthesize a family of compounds called menaquinones (vitamin K<sub>2</sub>), which have side chains based on repeating unsaturated 5-carbon (prenyl) units. These are designated menaquinone-n (MK-n) according to the number (n) of prenyl units. Vitamin K<sub>3</sub> (menadione or menaphthone) is a synthetic compound with structural similarities of naturally occurring vitamin K. It is a naphthoquinone with a methyl group at position 2 differing from natural vitamin K side chains at position 3.<sup>(29, 30)</sup>

Vitamin K<sub>1</sub> is essential for hemostasis in humans. Vitamin K<sub>2</sub> has been more recently found to be essential in healthy calcium disposition in bones, prevention of arterial calcification, and gene regulation of cell growth. Vitamin K<sub>3</sub> exhibits some of the actions of natural vitamin K. It is used as a nutritional supplement in livestock and pets because it is a precursor for vitamin K<sub>2</sub>.<sup>(31)</sup>

The biologic role of vitamin K is to act as a cofactor for a specific carboxylation reaction that transforms selective glutamate (Glu) residues to gg-carboxyglutamate (Gla) residues. The reaction is catalysed by a microsomal gg-glutamyl or vitamin K -dependent carboxylase enzyme, which in turn is linked to a cyclic salvage pathway known as the vitamin K epoxide cycle (figure 1). The vitamin K-epoxide cycle is pivotal to both the function of vitamin K and to the conservation of the microsomal cellular stores of vitamin K. The cycle includes both the glutamyl g-carboxylation step of vitamin K-dependent proteins and the recovery of the cofactor vitamin K quinol that is oxidized to vitamin K epoxide metabolite as a consequence of g-carboxylation. It involves 2 major integral membrane proteins: g-glutamyl carboxylase (GGCX) and vitamin K epoxide reductase (VKOR).<sup>(32)</sup>



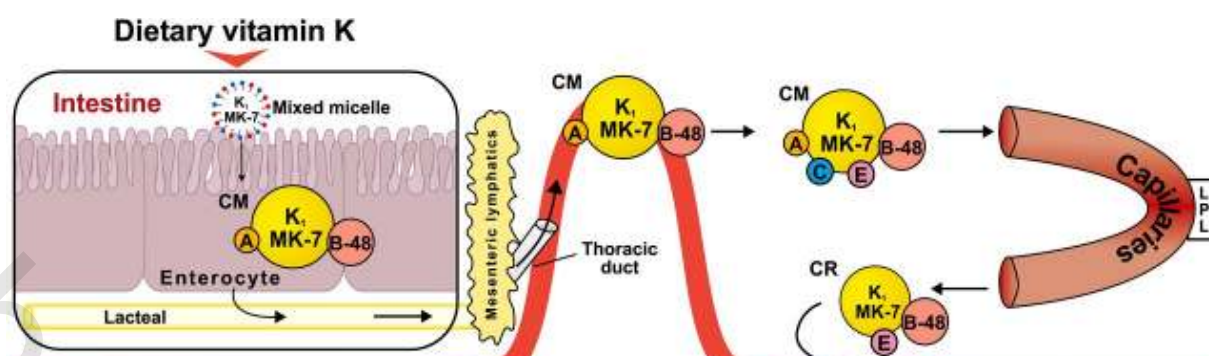
**Fig. (1): Vitamin K epoxide cycle.**<sup>(32)</sup>

Two other vitamin K-dependent proteins called protein C and protein S play a regulatory role in the inhibition of coagulation. The function of protein C is to degrade phospholipid-bound activated factors V and VIII in the presence of calcium. Protein S acts as a synergistic cofactor to protein C by enhancing the binding of activated protein C to negatively charged phospholipids. Yet another vitamin K-dependent plasma protein (protein Z) is suspected to have a haemostatic role but its function is currently unknown.<sup>(34)</sup>

Apart from the coagulation proteins, several other vitamin K-dependent proteins have been isolated from bone, cartilage, kidney, lungs, and other tissues.<sup>(35)</sup> Only two, osteocalcin and matrix Gla protein (MGP), have been well characterized. Both are found in bone but MGP also occurs in cartilage, blood vessel walls, and other soft tissues. There is evidence that protein S is synthesized by several tissues including the vessel wall and bone and may have other functions besides its well-established role as a coagulation inhibitor. It also seems likely that one function of MGP is to inhibit mineralization.<sup>(36)</sup> Although vitamin K is also required for the biosynthesis of some other proteins found in the plasma, bone and kidney, defective coagulation of the blood is the only major sign of vitamin K deficiency.<sup>(29)</sup>

## **Metabolism of vitamin K**

The intestinal absorption of vitamin K follows a well-established pathway that applies to most dietary lipids, which includes bile salt- and pancreatic-dependent solubilization, uptake of mixed micelles into the enterocytes, the packaging of dietary lipids into chylomicrons, and their exocytosis into the lymphatic system.<sup>(37)</sup> A simplified diagram of how this absorption is shown in Figure 2.



**Fig. (2): Intestinal absorption of dietary phylloquinone (K<sub>1</sub>) and menaquinone-7.<sup>(37)</sup>**

The absorption of a physiological dose (20  $\mu$ g) of free C-phyloquinone varied according to the type of accompanying meal. Significantly more phylloquinone tracer was absorbed when consumed with animal-based foods than with the meal pattern characterized by fast foods.<sup>(38)</sup>

Early studies using radiolabeled phylloquinone showed that after intestinal absorption, vitamin K first appears in lymph and then enters the blood stream associated with chylomicrons.<sup>(39)</sup> Later investigations of lipoprotein transport showed that the majority of phylloquinone was associated with triglyceride rich lipoprotein during the postprandial phase of absorption.<sup>(40, 41)</sup>

**Uptake of vitamin K by the liver:** The majority of vitamin K is delivered to the liver within the chylomicron remnants generated during the postprandial phase of intestinal absorption (Figure 2). The uptake process is complex and involves different apoproteins on the surface of lipoproteins, cell surface low affinity binding sites of heparin sulfate proteoglycan, and high-affinity lipoprotein receptors that mediate internalization of the lipoprotein particles.<sup>(42)</sup> A simplified scheme of the postulated mechanism of vitamin K tissue uptake is shown in Figure 3.<sup>(29)</sup>

**Uptake of phylloquinone by bone:** There are now studies that have specifically addressed the question of how phylloquinone is delivered to osteoblasts (Figure 3). The importance of this area for vitamin K centers on knowledge that bone matrix contains several Gla proteins [e.g., osteocalcin, matrix Gla protein (MGP), Gla-rich protein] that require vitamin K for their function and by findings that undercarboxylated species of osteocalcin and MGP normally circulate in healthy people.<sup>(43, 44)</sup>

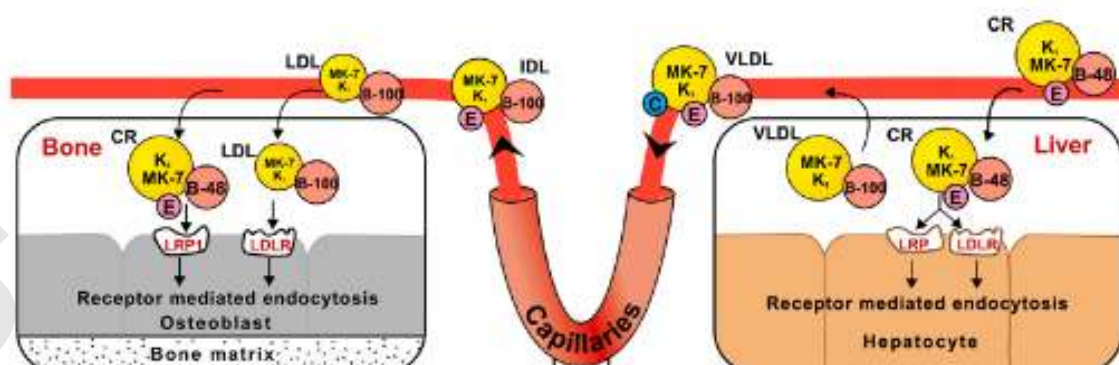


Fig. (3): Uptake of phyloquinone (K<sub>1</sub>) and Menaquinones-7 by liver and by bone.<sup>(33)</sup>

**Catabolism of vitamin K:** Humans excrete phyloquinone and MK by a common degradative pathway whereby the polyisoprenoid side chain is first shortened to 2 major carboxylic acid metabolites with 7- and 5-carbon side chains, respectively; the metabolites are then conjugated, mainly with glucuronic acid, and excreted in the bile and urine.<sup>(29)</sup> One notable feature of vitamin K metabolism compared to other fat-soluble vitamins is that the most abundant dietary form, phyloquinone is poorly retained in the body.<sup>(45)</sup>

## Food sources of vitamin K

### Phylloquinone (vitamin K<sub>1</sub>):

Phylloquinone is the primary dietary source of vitamin K. In general, green leafy vegetables contain the highest known phylloquinone concentrations and contribute approximately 60% of total phylloquinone intake.<sup>(46)</sup> As indicated in Table 1, spinach and collards, which have concomitant high concentrations of chlorophyll associated with the photosynthetic process, hence, dark leaf color have substantially higher concentrations of phylloquinone compared to the more commonly consumed iceberg lettuce, which is substantially paler, hence, lower chlorophyll concentrations. The other plant sources of phylloquinone are certain plant oils including soybean, canola (also known as rapeseed), cottonseed, and olive (Table 1).<sup>(47)</sup>

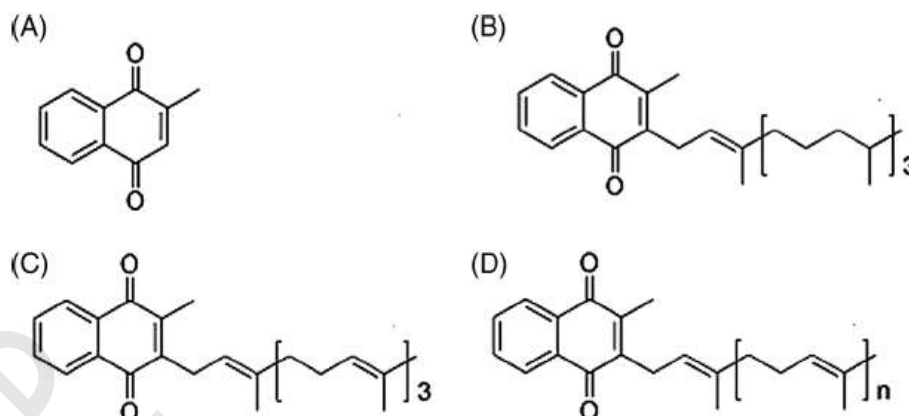
### 2', 3'-Dihydrophylloquinone:

The commercial hydrogenation of phylloquinone rich oils results in a transformation of phylloquinone into a hydrogenated form 2',3' Dihydrophylloquinone.<sup>(47)</sup> In the US food supply commercial hydrogenation of plant oils was a common practice that prolonged shelf life of the oil-based products. Many foods sold in fast-food restaurants and frozen prepared products such as fast-food French fries, doughnuts, and breaded fish sticks contained high concentrations of 2',3'-Dihydrophylloquinone (Table 1).<sup>(48)</sup>

**Table (1): Vitamin K content of common foods.**

| <b>Food</b>                          | <b>Major form of vitamin K</b> | <b>Concentration (mg/100 g)</b> |
|--------------------------------------|--------------------------------|---------------------------------|
| <b>Vegetables <sup>(49)</sup></b>    |                                |                                 |
| Collards                             | Phylloquinone                  | 440                             |
| Spinach                              | Phylloquinone                  | 380                             |
| Broccoli                             | Phylloquinone                  | 180                             |
| Cabbage                              | Phylloquinone                  | 145                             |
| Iceberg lettuce                      | Phylloquinone                  | 35                              |
| <b>Fats and oils <sup>(48)</sup></b> |                                |                                 |
| Soybean oil                          | Phylloquinone                  | 193                             |
| Canola oil                           | Phylloquinone                  | 127                             |
| Cottonseed oil                       | Phylloquinone                  | 60                              |
| Olive oil                            | Phylloquinone                  | 55                              |
| <b>Mixed dishes <sup>(50)</sup></b>  |                                |                                 |
| Fast food french fries               | Dihydrophylloquinone           | 59                              |
| Fast food nachos                     | Dihydrophylloquinone           | 60                              |
| Frozen, breaded fish sticks          | Dihydrophylloquinone           | 16                              |
| Margarine with hydrogenated oil      | Dihydrophylloquinone           | 102                             |
| <b>Other foods <sup>(51)</sup></b>   |                                |                                 |
| Natto                                | Menaquinone-7                  | 998                             |
| Hard cheeses                         | Menaquinone-9                  | 51.1                            |
| Soft cheeses                         | Menaquinone-9                  | 39.5                            |

## Menaquinones (vitamin K<sub>2</sub>):



**Fig. (4): Forms of vitamin K.** (A) Menadione, which is present in animal feed; (B) phylloquinone, which is the primary dietary source; (C) menaquinone-4, which is a conversion product from menadione or phylloquinone; and (D) menaquinones, which can vary in length from MK-4 to MK-13.<sup>(52)</sup>

Menaquinones are the other category of vitamin K present in the food supply (Figure 4). Menaquinones are often referred to as Vitamin K<sub>2</sub>, which is somewhat misleading given that all menaquinones are not alike in their origin or their function. Menaquinones are primarily of bacterial origin (*Bacteroides fragilis* and *Escherichia coli*), and differ in structure from phylloquinone in their 3-substituted lipophilic side chain.<sup>(52)</sup>

There is ongoing debate on whether bacterial synthesis of vitamin K in the intestine provides a significant supply of this vitamin in humans. The colon contains a large reservoir of bacterial vitamin K<sub>2</sub> but it is now undeniable that this pool represents only about 10% of normal human requirements and is insufficient to satisfy these requirements. Furthermore, there is some evidence of poor bioavailability of this intestinal source of vitamin K.<sup>(53)</sup>

Bile salts are necessary for effective absorption of vitamin K, but are not present in the colon, and the intestinal synthesis of vitamin K is not sufficient to compensate for deficiency due to biliary obstruction. Moreover, intestinal menaquinones are enveloped within the bacterial membranes and are, therefore, poorly available for intestinal adsorption. These data argue against the concept of the colon as a significant source of vitamin K for human use, so that patients at risk of deficiency remain those who cannot absorb vitamin K from the small intestine.<sup>(53)</sup>

## Dietary intakes of vitamin K:

The current US dietary guidelines for intakes of vitamin K are 90 and 120 µg/day for women and men, respectively. These guidelines are termed adequate intakes (AI).<sup>(54)</sup> The average intake of phylloquinone in infants fed human milk during the first 6 months of life has been reported to be less than 1 µg/day; this is approximately 100-fold lower than the intake in infants fed a typical supplemented formula. This big disparity between intakes is reflected in plasma levels (Table 2).<sup>(55)</sup>

The recommended dietary intake of vitamin K<sub>1</sub> is 2 µg/day for infants in the first 6 months of life and 2.5 µg/day for infants aged 7-12 months. After this age, the adequate intake progressively increases from 30 µg/day in children aged 1-3 years, up to 75 µg/day in adolescents.<sup>(53)</sup>

**Table (2): Dietary intakes and plasma levels of phyloquinone in human-milk-fed *versus* formula-fed infants aged 0-6 months.**<sup>(55)</sup>

| Age (weeks) | Phylloquinone intake (µg/day) |                          | Plasma phylloquinone (µg/l) |             |
|-------------|-------------------------------|--------------------------|-----------------------------|-------------|
|             | Human milk fed <sup>a</sup>   | Formula fed <sup>b</sup> | Human milk fed              | Formula fed |
| 6           | 0.55                          | 45.4                     | 0.13                        | 6.0         |
| 12          | 0.74                          | 55.5                     | 0.20                        | 5.6         |
| 26          | 0.56                          | 52.2                     | 0.24                        | 4.4         |

<sup>a</sup>Breast-milk concentrations averaged 0.86, 1.14, and 0.87 µg/l of phyloquinone at 6, 12, and 26 weeks, respectively.

<sup>b</sup>All infants were fed a formula containing phyloquinone at 55 µg/l.

Using the detection of PIVKA-II as a marker of sub-clinical deficiency, It was concluded that a minimum daily intake of about 100 ml of colostrum milk (that supplies about 0.2-0.3 µg of phyloquinone) is sufficient for normal haemostasis in a baby of about 3 kg during the first week of life.<sup>(55)</sup>

### Vitamin K deficiency

Vitamin K (VK) deficiency can occur in any age group but is encountered most often in infancy. In infants, the low transmission of VK across the placenta, liver prematurity with prothrombin synthesis, lack of VK in breast milk, and the sterile gut in neonates account for VK deficiency.<sup>(24, 56, 57)</sup> The increased incidence of breast-feeding in the past two decades has led to renewed interest in vitamin K deficiency in infants. Despite the significantly lower amounts of vitamin K in breast milk compared with formula and the susceptibility to vitamin K deficiency bleeding (VKDB) in infants not supplemented with vitamin K, the efficacy of supplementation continues to be questioned.<sup>(58)</sup> Neither the reported vitamin K concentrations in milk nor amounts of vitamin K consumed by infants have been correlated with risk for VKDB. This can be explained partially by individual differences in vitamin K absorption and in volumes of milk consumed. However, another factor has been the difficulty in obtaining quantitative data on vitamin K in milk because of the trace quantities present. With the advent of high performance liquid chromatography (HPLC) methodology came several reports quantitating vitamin K in human milk.<sup>(45)</sup>

### Vitamin K deficiency bleeding

Vitamin K deficiency bleeding (VKDB) is a coagulopathy that develops in infants who do not have sufficient vitamin K stores to support production of clotting factors.<sup>(59)</sup> VKDB is usually classified by aetiology (idiopathic and secondary) and by the age of onset (early, classical and late).<sup>(53)</sup> In idiopathic VKDB no cause other than breast-feeding can be demonstrated. In secondary VKDB there is usually an underlying cause, such as the effect

of drugs that have been given to the mother or infant or a hereditary hepatobiliary malabsorption disease (e.g., biliary atresia, alpha-1-antitrypsin deficiency, cystic fibrosis).<sup>(56)</sup> In addition, autosomal recessive vitamin K-dependent coagulation factor deficiencies (VKCFD), due to mutations in the gene encoding for g-glutamyl carboxylase (VKCFD type I) and in the gene encoding for vitamin K epoxide reductase (VKCFD type II), have been reported.<sup>(60)</sup>

According to the age of onset, early VKDB presents within 24 hours of birth and is almost exclusively seen in infants of mothers taking drugs which inhibit vitamin K. These drugs include anticonvulsants (carbamazepine, phenytoin and barbiturates), antituberculosis drugs (isoniazid, rifampicin), some antibiotics (cephalosporins) and vitamin K antagonists (coumarin, warfarin). The clinical presentation is often severe with cephalic haematoma and intracranial and intra-abdominal haemorrhages.<sup>(61, 62)</sup>

Classical VKDB occurs between 24 hours and 7 days of life and is associated with delayed or insufficient feeding. The clinical presentation is often mild, with bruises, gastrointestinal blood loss or bleeding from the umbilicus and puncture sites. Blood loss can, however, be significant, and intracranial haemorrhage, although rare, has been described.<sup>(56)</sup>

Late VKDB is associated with exclusive breast-feeding. It occurs between the ages of 2 and 12 weeks. The clinical presentation is severe, with a mortality rate of 20% and intracranial haemorrhage occurring in 50%. Persistent neurological damage is frequent in survivors. In fully breast-fed infants who did not receive vitamin K at birth, the incidence is between 4.4/100,000 and 7.2/100,000 births. Babies with cholestasis or malabsorption syndromes are at particular risk.<sup>(24, 62)</sup>

While the generally low, weight-adjusted intakes of phylloquinone in breast-fed infants account for the much higher prevalence of VKDB in breast-fed infants as a group, knowledge of the precipitating factors that trigger VKDB in an individual infant are less well understood. In some infants, underlying pathologies that cause cholestasis with resultant malabsorption of vitamin K may be identified, but in a substantial proportion no predisposing factor is found.<sup>(45)</sup>

### **Risk factors of vitamin K deficiency in early infancy**

The concentrations of phylloquinone in cord blood are <50 ng/L and are generally too low to be accurately measured. However, the fact that the average maternal/neonatal cord concentration gradient of phylloquinone is within the range of 20:1 to 40:1 has led to the concept of a placental barrier to phylloquinone, which is fairly unresponsive to maternal vitamin K supplementation.<sup>(63)</sup>

There is limited information of liver stores of vitamin K in fetuses and young infants, the median hepatic concentration of phylloquinone in infants at term was 2.2 pmol/g compared to a median of 12 pmol/g in adults. Concentrations of ~2–4 pmol/g were detected as early as 10 wk of gestation, suggesting that the concept of a placental barrier for phylloquinone may be overplayed.<sup>(64)</sup>

The major difference between neonatal hepatic reserves and those of adults is that whereas the long-chain menaquinones (mainly MK 7–13) make up the majority of adult reserves (~90%), they are absent or very low at birth and build up slowly over several weeks. This slow buildup of hepatic MK would be consistent with the colonization of the neonatal gut by MK-producing bacteria.<sup>(65)</sup>

### **Vitamin K prophylaxis for newborns**

In 2003 the American Academy of Pediatrics recommended that vitamin K1 should be given to all neonates as a single, intramuscular dose of 0.5 to 1 mg, and this recommendation was recently reaffirmed in 2009.<sup>(66, 67)</sup> A similar recommendation was issued and reaffirmed in 2009 by the Canadian Pediatric Society and the Committee on Child and Adolescent Health, College of Family Physicians of Canada.<sup>(53)</sup>

Accordingly, it is recommended that vitamin K1 should be given as a single intramuscular dose of 0.5 mg (for babies weighing 1,500 g or less at birth) or 1.0 mg (for babies weighing more than 1,500 g at birth) to all neonates within the first 6 hours after birth following initial stabilisation of the baby and an appropriate opportunity for maternal (family)-baby interaction. Several European countries are increasingly moving towards a uniform policy.<sup>(53)</sup>

Prophylaxis with 1 mg vitamin K was endorsed by the UK Department of Health in 1998, while no preference was stated for either administration route (i.e., intramuscular or oral), concluding that this is a matter for professionals and services to agree locally.<sup>(53)</sup> In the 2008 guidelines of the UK National Health System recommended that babies weighing less than 2.5 kg should be administered 400 µg/kg, whereas the dose for babies weighing more than 2.5 kg is 1 mg. It is especially important that babies at extra risk receive vitamin K via the intramuscular route. When the intramuscular route is declined by the parent, two oral doses of 2 mg should be offered instead (the first dose within 6 hours of birth, and the second between 4 - 7 days of age).<sup>(68)</sup>

A consensus conference of the Italian Society of Neonatology held in 2004 established that 0.5 mg of vitamin K should be administered intramuscularly at birth, followed by 25 µg/day orally from the second to the fourteenth week of life.<sup>(53)</sup>

An alternative strategy accepted by the consensus conference is the administration of 2 mg of vitamin K at birth, followed by 25 µg/day from the seventh day to the fourteenth week of life.<sup>(69)</sup>

It should, however, be noted that Kumar et al., found extremely high plasma K levels on day 14 of life in premature infants (<28th gestational week) who received 1 mg of vitamin K intramuscularly shortly after birth.<sup>(70)</sup>

In another study, by Costakos et al., preterm neonates who were given 0.5 to 1 mg vitamin K prophylaxis also showed vitamin K levels that were 1,900 to 2,600 times higher (2 days afterwards) and 550 to 600 times higher (10 days afterwards) than normal adult plasma values (0.5 ng/mL).<sup>(71)</sup>

### **Risks of vitamin k prophylaxis include the following:**

- 1) Vitamin k ampoules contain phenol, propylene glycol and polyethoxylated castor oil as a non-ionic surfactant. Studies in animals given polyethoxylated castor oil have shown a severe anaphylactic reaction associated with histamine release, alterations in blood viscosity and erythrocyte aggregation (red blood cell clumping).<sup>(72,73)</sup>
- 2) The risks of injecting vitamin K into a newborn baby are nerve or muscle damage as the preparation must be injected deeply into the muscle, not subcutaneously under the skin.<sup>(74)</sup>
- 3) Infants can suffer from jaundice or kernicterus (brain damage from a build-up of bile pigments in the brain) from vitamin k. <sup>(53)</sup>
- 4) Infants who have the G6PD (glucose 6 phosphate dehydrogenase) deficiency are at particular risk from vitamin K.<sup>(75)</sup>
- 5) Possible increased chance of childhood cancer. However this suspicion was disproved by two large retrospective studies in the USA and Sweden which failed to find any evidence of a relationship between childhood cancers and vitamin K injections at birth.<sup>(53)</sup>
- 6) Marked supraphysiological tissue concentrations that for serum can be several 100-folds higher than normal. <sup>(76)</sup>

A recent study provided evidence that the combination of a raised serum vitamin K epoxide concentration with an increased proportional excretion of the 7-carbon side chain urinary metabolite may together indicate a metabolic overload of both vitamin K recycling and catabolic pathways. <sup>(77)</sup>

Exclusive breast feeding as a predisposing factor for late onset vitamin k deficiency bleeding (VKDB) is still a matter of controversy.