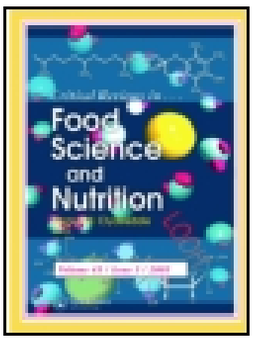

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REVIEW

Insights into the role of bacteria in vitamin A biosynthesis: Future research opportunities

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ABSTRACT

Significant efforts have been made to address the hidden hunger challenges due to iron, zinc, iodine, and vitamin A since the beginning of the 21st century. Prioritizing the vitamin A deficiency (VAD) disorders, many countries are looking for viable alternative strategies such as biofortification. One of the leading causes of VAD is the poor bioconversion of β-carotene into retinoids. This review is focused on the opportunities of bacterial biosynthesis of retinoids, in particular, through the gut microbiota. The proposed hypothesis starts with the premise that an animal can able to store and timely convert carotenoids into retinoids in the liver and intestinal tissues. This theory is experimental with many scientific insights. The syntrophic metabolism, potential crosstalk of bile acids, lipocalins and lipopolysaccharides of gut microbiota are reported to contribute significantly to the retinoid biosynthesis. The gut bacteria respond to these kinds of factors by genetic restructuring driven mainly by events like horizontal gene transfer. A phylogenetic analysis of β-carotene 15, 15′-mono (di) oxygenase enzymes among a selected group of prokaryotes and eukaryotes was carried out to validate the hypotheses. Shedding light on the probiotic strategies through non-genetically modified organism such as gut bacteria capable of synthesizing vitamin A would address the VAD disorders.

KEYWORDS

Retinoid; β-carotene; microbiota; vitamin A deficiency; probiotics; retinal

1. Introduction

A nutrient of global importance, an essential nutrient, and its deficiency can cause a pervasive hidden hunger is the vitamin A. A current worldwide issue and still an inexcusable scenario about the vitamin A deficiency (VAD) is about one-third of children aged 6–59 months is still affected by this illness. VAD can develop serious health issues such as visual impairment, frailty and increased risk of mortality to the infants below five years of age and pregnant ladies. The highest rates of the vulnerable children are in Sub-Saharan Africa (48%) and South Asia (44%) (UNICEF 2018). Although various fortification programs with pseudo vitamins and dietary supplements have reduced significant morbidity due to VAD, it also reported causing nutrition imbalance and many other adverse effects. Moreover, poor bioavailability, challenges in large-scale extension and the cost of manufacturing vitamin formulations result in an ineffective VAD eradication so far.

The lipophilic fat-soluble vitamins (A, D, E, and K) have been the focus of investigation for almost a century. In particular, many chemical and biological approaches have been made to synthesis carotenoids and retinoids. Biofortification approaches such as genetic engineering of the carotenoid pathway in crops like corn, rice, and potato have not been successful due to the risk of unintended effects and poor support offered by the public and government policies (Tang et al. 2012). Though various investigators have made efforts to decipher the viable vitamin A alternative, they have managed to find only synthetic analogs. The chemically produced retinoid compounds may exhibit less biological activity and create undesirable by-products (Parker, Smith, and Baxendale 2016). The synthetic receptor-selective retinoids also may result in altered toxicity patterns (Curley and Robarge 1997). It is the time to explore natural sources to identify the biological agents which carrying the retinoid activity to overcome the synthetic retinoid products (Uray, Dmitrovsky, and Brown 2016).

In the biological system, the animals including humans create retinoids from carotenoids obtained from plants, while a plant cannot produce retinoids. The entire retinoid biosynthesis pathway including the carotenoid synthesis might have embodied only in lower level organisms such as microbes. Numerous studies have attempted to decode the impacts of the gut microbiome on host metabolism, innate immunity, and metabolic disorders. The human colon is the host of a diverse microbiota with more than 50 bacterial phyla dominated by Bacteroidetes, Firmicutes, Proteobacteria, and Actinobacteria (Eckburg et al. 2005). The impact of host genetics and green diet on the gut microbial diversity has been detailed by human microbiome project (Lozupone et al. 2012; Blaser 2016). The microbial adaptability to eukaryotic tissues and the symbiotic

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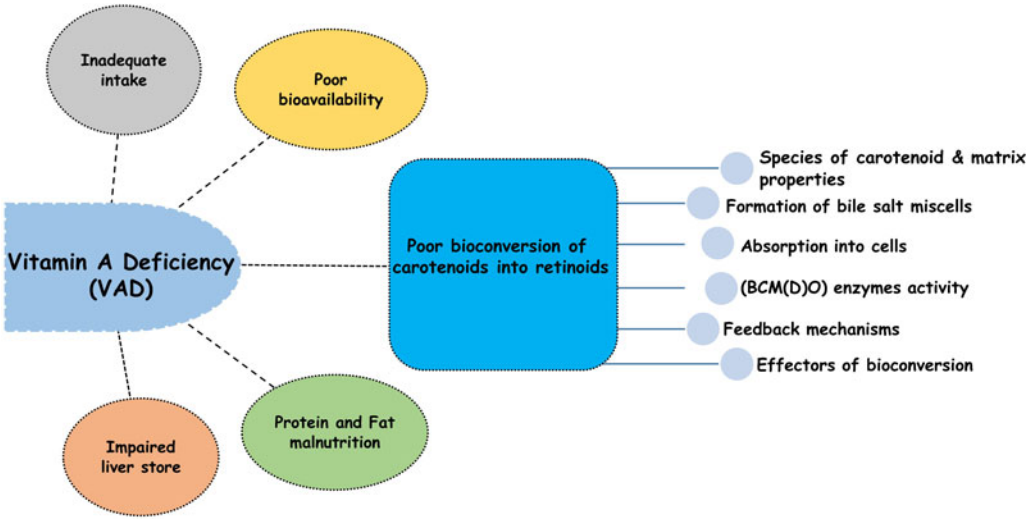

Figure 1. Major risk factors for VAD. Inadequate intake, poor bioavailability, impaired liver store (Akhtar et al. 2013), protein and fat malnutrition (He et al. 2013) and poor bioconversion due to factors such as enzyme efficiency (Scherzinger and Al-Babili 2008) are presented in the diagram.

metabolism of nutrients fuels through opportunistic genetic alterations within individual gut microbes (Harms et al. 2015). Many bacterial strains such as *Bifidobacteria* and *Lactobacillus* have been shown to exhibit vitamin producing capacity although their metabolic pathways have not been elaborated (Linares et al. 2017).

Though some gut bacteria possess the genes for carotenoids synthesis, if they acquire a rhodopsin type gene, it could be possible to synthesis retinoids from dietary carotenoids (Culligan et al. 2014). Some bacteria create retinoids as a prosthetic group for bacteriorhodopsin (Jang et al. 2011).

The carotenoids such as β-carotene, α-carotene, and β-cryptoxanthin are the primary provitamin compounds reported to present abundantly in the human tissues. In particular, people around the world consume vitamin A in the form of β-carotene through fruits and vegetables. While the recent debates about β-carotene-cleaving enzymes are more focusing on eukaryotic β-carotene 15, 15′-mono (di) oxygenase (BCM(D)O), no attention has been paid to bacterial sources. The β-carotene-cleavage enzymes were principally reported in animal liver, intestine, yolk sac, placenta and the embryo (Redmond et al. 2001). Some researchers outlined the microbial vitamin A synthesis through bacteria synthesizing retinoid-like compounds and the gut microbiome with BCM(D)O mimicking activity.

In particular, this review highlights the scientific predictions that are underpinning the hypothesis "Biosynthesis of retinoids from carotenoids dependent and independent pathways through gut microbiota." *First,* the liver and or intestine of the animals are the storage and bioconversion sites for synthesizing retinoids from carotenoids. The carotenoids are metabolized through the carotenoid cleavage enzymes synthesized by the epithelial cells that include the cells lining the liver. Since, a diverse group of microbes in the liver and intestine relay on that organ for its nutritional requirements, they could have developed the capability to take part or entire metabolism of carotenoids and retinoids. Also, the microbial role in retinol transportation to multiple sites of the body through bacterial compounds mimicking the retinol-binding protein (RBP) is an area of concern. *Second,* the host-symbiont interactions through the hologenome adaptation effect. The microbes could have remodeled its genetic make-up, generation by generation through various intragenomic recombination processes with the host cells to increase the adaptability with the gut environment. *Third,* through lateral or horizontal gene transfer phenomenon, the genes for the biosynthesis and catabolism of carotenoids and retinoids might have been transferred to bacteria from the host tissues. *Fourth,* some putative enzymes that synthesis retinoids of different biological activity could have been synthesized and utilized by microbes to sustain the niche. *Finally,* there is always a bidirectional pressure exists between the prokaryotic cells and the host eukaryotic cells in the organs. The demand could have resulted in inter-bacterial changes that alter the microbes to play a role in vitamin A synthesis.

A vital organ, nutritional powerhouse and one of the top vitamin A source is the beef liver. The liver also acts as a downstream of the gastrointestinal tract in the digestive system and harbors trillions of bacteria (Hartstra, Nieuwdorp, and Herrema 2016). Hence, this review strongly emphasizes and proposes a critical role for the liver or intestine-friendly bacteria in Vitamin A biosynthesis. These proposed conceptual points with literature evidence have been detailed, and the alternative strategies to overcome VAD are illustrated. This review aims to focus exclusively on bacterial metabolism of retinoid and carotenoid and revealed some natural and viable approaches such as probiotics and synbiotic foods for VAD.

2. Influencing factors of VAD

VAD develops mainly among the children and pregnant woman due to inadequate intake, poor bioavailability of stored vitamin due to protein and fat malnutrition, impaired liver store and especially poor bioconversion ratio of β-carotene to retinoids (Schaub et al. 2017). An insufficient or

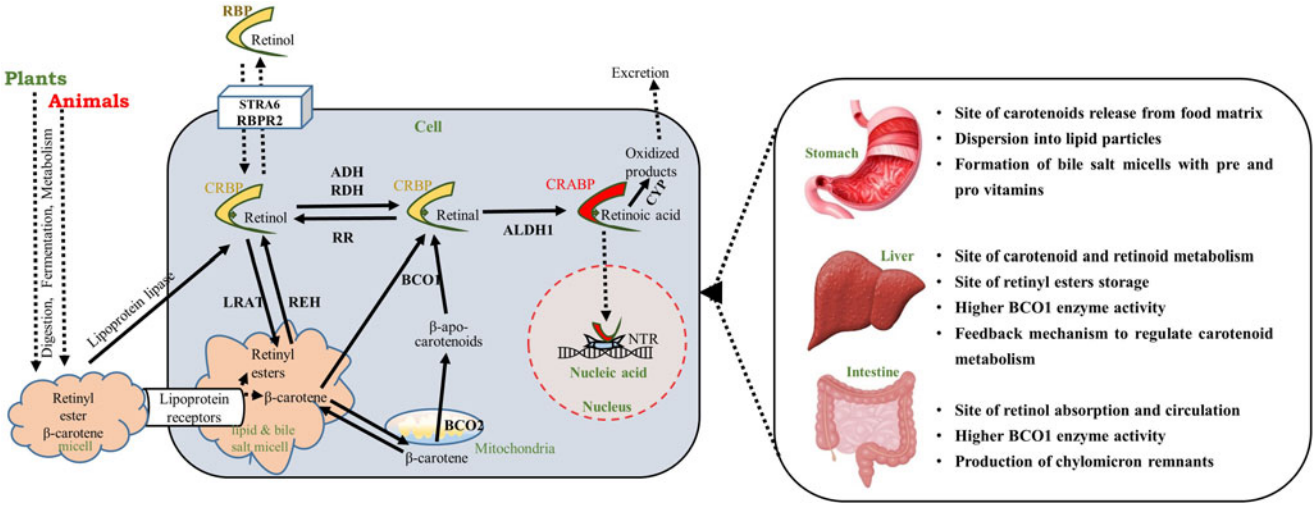

Figure 2. Overview of retinoid and β-carotene metabolism in a eukaryotic cell and physiological role of the specified organs. The important sites of bioconversion of carotenoids to retinoids are stomach, liver, and intestine with varied metabolic functions. The carotenoids digested from the plant-sourced foods and animal tissues form lipoprotein micelles during digestion, fermentation and metabolism processes. The miscelles containing retinyl esters and β-carotene with bile salts are circulated in the blood. Through endocytosis, the micelles are internalized by lipoprotein receptors. The retinyl esters are hydrolyzed to retinol by retinyl ester hydrolase (REH), which can also be oxidized back to retinyl esters by lecithin: retinol acyltransferase (LRAT). The active retinol binds with cellular binding protein (CRBP) and transported to the target tissues through specific surface receptors (STRA6, RBPR2) and attaches with retinol binding protein (RBP). Besides, the circulating RBP bind retinol can be internalized into the cells through the same receptors and can be stored as retinyl esters through REH enzymes. The retinol can be reversibly transformed to retinal through retinaldehyde reductase (RR) and back to retinol through alcohol dehydrogenase (ADH) or retinol dehydrogenase (RDH). The retinal is irreversibly oxidized to retinoic acid (RA) through the action of aldehyde dehydrogenase (ALDH1). However, the main pathways are symmetric cleavage of β-carotene into retinal by the enzyme β-carotene-15, 15′-oxygenase (BCOI) and asymmetrical cleavage by β-carotene-9′, 10′-oxygenase (BCOII). The mitochondrial BCOII products can be transformed to retinal through BCOI. The retinoic acid is transferred to the nucleus through cellular retinoic acid binding protein (CRABP). The retinoic acid exerts genomic transcriptional modulating activity by binding with nuclear transcription receptor (NTR). The retinoic acid is oxidized by the cytochrome enzymes P450 (CYP) and eliminated from the cell (Bonet et al. 2015).

marginal intake will lead to limited stores of carotenoids, and a sudden demand will deplete the reserves. The carotenoids and retinoids of plant and animal sources upon ingestion exhibit a poor absorption due to errors in the hydrolysis process of esterified compounds and unhealthy gastrointestinal status (Reboul 2013). The diseased or malfunctioned liver cannot store as much retinal or make as much retinol binding proteins. Children with adequate vitamin A but severely malnourished and protein deficient are also vulnerable to VAD. This disorder is due to the poor transportation of retinoids to the target sites of the body (Akerstrom, Flower, and Salier 2000). The primary causative factors affecting the vitamin A status and leading to VAD associated disorders are depicted in Fig. 1.

Likewise, one of the critical conditions leading to VAD is bioconversion efficiency of the gut cells mediating the transformation of β-carotene into retinoids. Various factors influence the catalytic property of the carotenoid oxygenase enzymes. The enzyme BCM(D)O depends on ferrous iron as a cofactor and exhibits variable enzyme activity based on host genetic factors such as single nucleotide polymorphisms (Esaki, Malkaram, and Zempleni 2012). Iron or zinc deficiency considerably alters the vitamin A metabolism (Himoto and Masaki 2018). The preferential absorption of carotenoids into lipid micelles depends on the all-trans isomer configuration of the compound and formation of secondary and tertiary bile acids. Moreover, the availability of lipopolysaccharides and lipoproteins specific to the absorption and transportation of carotenoids and retinoids have significant implications on vitamin A status (He et al. 2013). Also, the excess intake of carotenoids leads to poor uptake owing to the limited capacity of micelle-carotenoid incorporation (Perera and Yen 2007).

The bioconversion efficiency of β-carotene to retinal in various animal species differs based on a number of factors, in particular, the poor converter phenotype of enzyme BCMO (Green and Fascetti 2016). It is evident that vitamin A status is greatly influenced by the bioconversion ability of the epithelial gut cells lining the intestine, liver, and other target organs.

3. Carotenoids to retinoids: bioavailability to bioconversion to bioefficiency

Concerning the bioconversion of carotenoids into retinoids, the site, rate, and efficiency of the process is specific and are carried out at particular localities of the body. An overview of the retinoid cycle and β-carotene metabolism from feed to bioconversion is depicted detailly in Fig. 2 adopted from Bonet et al. (2015). The carotenoids intake through green plants and animal products are absorbed and metabolized majorly in the stomach, liver and intestinal organs. In diets, the carotenoids and retinoids found majorly in two forms: in oil droplets and fruits and vegetable matrices (Olson 1999). Upon ingesting the carotenoid-rich plants or retinoids rich animals, the compounds are released from the food matrix through the physical and chemical metabolic processes such as chewing and fermentation. The epithelial tissues covering the organs carry the retinoid metabolizing enzymes and the necessary receptors. In addition to the passive diffusion of preformed retinoid, the carotenoids such as β-carotene are absorbed intact by the intestine, and also

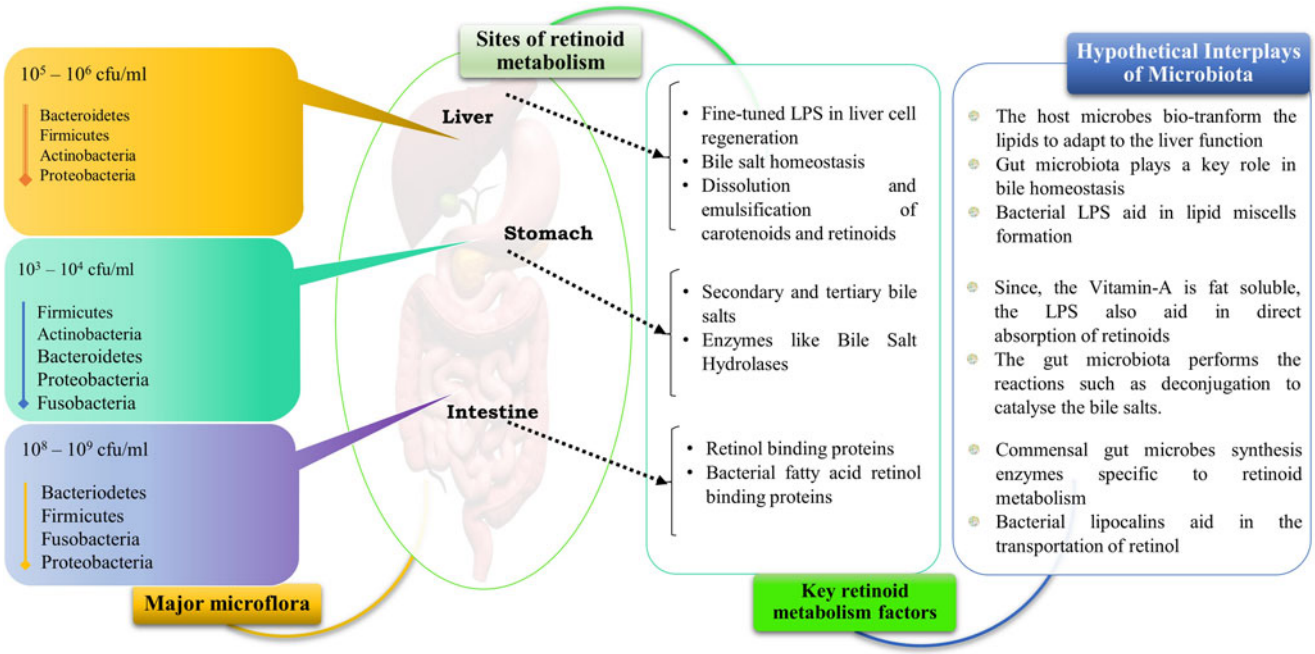

Figure 3. The hypothetical interplay among gut microbiota, bile salt, dietary lipids and cellular retinol binding proteins in retinoid metabolism. The figure illustrates the richness of gut microbiota in order of abundance in the specified organs and some crucial carotenoid and retinoid metabolism factors such as lipopolysaccharides (LPS), retinol binding proteins and bile salts.

circulated through chylomicrons and their remnants (Redlich et al. 1996, 1999). The digestive fluid of the stomach, enzymes such as gastric lipase and gut microbiota aid in dispersion of carotenoids and retinoids into lipid micelles along with bile salt. Some microbial proteins such as *E. coli* Blc and dietary proteins such as cow milk lipocalins with the potential to bind retinol and β-carotene also transport them into enterocytes (Borel and Desmarchelier 2017).

The bioconversion of β-carotene into retinal is also depending on the type of cleavage enzyme, health condition and even the rate of fat intake and its digestion. The β-carotene split into retinal through oxidative enzyme BCM(D)O. The enzyme β-carotene-9′,10′-oxygenase (BCO2) mediates the asymmetric cleavage of β-carotene into apocarotenoids. The excess retinoic acid is cleared through oxidization by the cytochrome enzymes P450 (CYP) (Bonet et al. 2015). The enzyme activity has been reported in mammalian organelles such as jejunum enterocytes, intestinal mucosa, liver, kidney, lung, and brain (Wolf 2009; During et al. 1996; Scherzinger and Al-Babili 2008). However, the major sites of carotenoid metabolism are revealed as liver and intestine. The bioconversion of carotenoids to retinoid and its bioavailability in the rat intestine in relation to the abundant of gut microflora was studied by Grolier et al. (1998). The findings suggest that the intestinal microflora have some impact on absorption of carotenoids and also on the biological activities of carotenoids and retinoids.

On account of β-carotene bioconversion efficiency, the factor is calculated from the proportion of absorbed provitamin A carotenoid into retinoid. Though, various populations having different nutritional habits, we gathered the data by searching the PubMed listed publications to derive the recent data. The conversion ratio is also varied by the type of food matrix, body weight and condition of the human beings. The mean conversion ratio of vegetable β-carotene matrix to vitamin A by weight as 26:1 and for fruits like orange as 12:1 and for the golden rice, the proportion is 3.6:1 (Tang 2010). With a breastfeeding woman, the ratio is 12:1 for fruits and 28:1 for vegetables by weight (Khan et al. 2007) and with an anemic child with VAD, the conversion factor of vitamin A rich food is 26:1 by weight (de Pee et al. 1998). Despite the various conclusions, the recent research data of mean conversion ratio of a healthy well-fed man is 19:1 of β-carotene to retinol activity equivalent (RAE) by weight (Schaub et al. 2017).

Therefore, it is evident that the gut cells, in particular, the liver and intestine mediate the vitamin A homeostasis and hence the symbiotic bacteria could contribute to the functions significantly.

4. Role of gut bacteria in bile acids, dietary lipids, and retinol binding protein regulation

Recently, much scientific evidence has been documented on the crucial role of gut microbiota in biosynthesis or co-metabolism of the essential vitamins (Roberfroid et al. 1995). There is a multidirectional benefit exists between gut microflora, bile acid homeostasis and retinoid metabolism (Sommer and Backhed 2013). This section reviews the symbiotic relationship between the gut microbiota and the metabolism of bile acids, dietary lipids and cellular proteins in support of vitamin A synthesis. The ability of the body to convert β-carotene to retinoids is stimulated or impeded by numerous factors which include: gene variant, microbiota, dysbiosis, insulin resistance, bile acid status, carotenoid oxygenases enzymes, hydrochloric acid content, lipid level, and celiac diseases (Saeed et al. 2017). A synergy between the microbiota variation and vitamin A absorption supports the

notion that there are cross-talks among bile acid, lipids and cellular proteins with gut microorganisms (Leung et al. 2009). The microbes may provide the host with specific and functional enzymes and altering the biochemical pathways.

The scientific knowledge about the number of human gut bacterial species from a few hundred in the 1970s to many thousands in the recent times is due to the advancements in metagenomics analysis. With the improvements in molecular techniques, the gut microbiota is estimated at between 100 to 200 trillion microbes in 1000 different species. The human stomach, liver, and intestine harbors complex microbial communities in the range of 10^3–10^4, 10^4–10^8 and 10^8–10^{14} colonies per gram respectively (Hooper, Midtvedt, and Gordon 2002). It is clear that the density of microflora is relatively lower in the stomach, high in the liver and intestinal organs. The common dominant microbiota belongs to the Bacteroidetes, Firmicutes, Actinobacteria, Proteobacteria and *Fusobacteria phylum*. In a healthy adult intestine, Bacteroidetes and Firmicutes dominate (around 95%) the total microflora (Ohland and Jobin 2015) and constitute more than 85% in the liver metabolic studies (Liu, Hu, and Wan 2016). The hypothetical interplays between the gut microflora and vitamin A biosynthesis are illustrated in Fig. 3 which is completely or partially supported through the scientific relevance.

Around 2844–5653 intestinal genes were regulated by the habitat microbiota including the retinoid metabolic pathways (Johansson et al. 2008). The gut microbiota aid in absorption, metabolism, and transport of carotenoids and retinoid by synthesizing secondary and tertiary bile salts through various biochemical reactions. *Lactobacillus*, *Bifidobacterium*, *Bacteroides*, *Listeria*, and *Clostridium* performs deconjugation; *Clostridium*, *Bacteroides, Eggerthella, Escherichia, Eubacterium, Ruminococcus*, and *Peptostreptococcus* conducts oxidation and epimerization; *Eubacterium* and *Clostridium* performs dihydroxylation; *Eubacterium*, *Bacteroides*, and *Lactobacillus* conducts esterification and *Peptococcus, Clostridium, Pseudomonas* and *Fusobacterium* carry out desulfation (Gerard 2014). The hydrolysis and conjugation reactions are catalyzed by the intestinal bacterial enzymes such as bile salt hydrolases (Kellogg 1974). The dihydroxylation reactions such as conversion of primary bile salts into successive salts are mediated by bacterial species such as Clostridia (Zwicker and Agellon 2013). The retinoid metabolism genes such as retinoid X receptor (RXR) are modified in relation to the gut microbial alterations. The significant pathways in retinoid biosynthesis like a canonical pathway, farnesoid X receptor (FXR) and RXR are regulated by the secondary and tertiary bile acids transformed by the gut microbiota (Swann et al. 2011).

The lipopolysaccharide (LPS) of the stomach or rumen fluid aids in the absorption of carotenoids and retinoids from the feed digesta and also mediating the circulation (German and Dillard 2006). LPS ingested through feed and metabolized through the digestive fluid are further tuned into fat micelles suitable for carotenoids transportation to the active sites by the gut microbiota. Bacterial LPS also plays a crucial role in liver nutrient absorption (Heuman 1989). The liver regeneration also needs the role of commensal microbes in harnessing the signaling cascade molecules such as LPS (Karin and Clevers 2016). The secondary and tertiary bile acids tuned for carotenoids and retinoids metabolism is essential for vitamin homeostasis (He et al. 2013). In the intestine, the gut microbiota synthesis enzymes or complement proteins such as bacterial lipocalins which could contribute to the intestinal transport of retinoid (Bishop 2005).

Berry et al. (2012) experimented with the protein made in bacteria to evaluate the human STRA6 mediated retinol uptake process in retinol or RBP complex. He elucidated that the microbial proteins contribute to the RBP concentration in human blood. BCM(D)O activity is regulated by the feedback mechanism and varied protein levels (Gronowska-Senger and Wolf 1970). The bacterial lipocalins are lipophilic molecules with diverse functional activities including retinol transportation (Akerstrom, Flower, and Salier 2000). The lipocalins are majorly found in gram-negative bacteria with a 3D structure (Newcomer et al. 1984). A significant similarity is observed between the structures of bacterial lipocalins and serum retinol binding protein (Bishop 2005).

A deeper understanding of the gut microbiota in VAD affected individuals could lead to preventive strategies and natural therapies. Further, targeting the interplay between vitamin A metabolism, bile acids, and lipopolysaccharides and proteins of bacterial source seems to be one of the promising avenues for the treatment of VAD disorders, but more research is needed.

5. Comparison of enzymes and their genes reported in the bacterial, plant and animal metabolism of ß-carotene and retinoids

The scientific fact that the gut organs of the animals can synthesis retinoids through the carotenoids of green forage propels the concept that there is a role for gut friendly microflora in the production of carotenoids and or vitamin A. The characterization and illustration of the retinoid metabolic pathway describing the bioconversion process in eukaryotes were investigated (Goodman et al. 1966, 1967). The enzyme BCMO was first identified and isolated as a cytosolic enzyme by the product retinal (Olson and Hayaishi 1965). The molecular characterization of BCM(D)O have been reported in various species, including chickens (Wyss et al. 2000), zebra fishes (Lampert et al. 2003), humans (Yan et al. 2001), fruit flies (von Lintig and Vogt 2000) and mice (Paik et al. 2001). In prokaryotic organisms, the bacteria usually use the mevalonate (MVA) pathway over 2-C-methyl-D-erythritol-4-phosphate (MEP) pathway for carotenoid production (Vranova, Coman, and Gruissem 2013). The bacterial biosynthesis of carotenoids has been detailed and discussed by various researchers in different organisms (Armstrong 1994). However, studies explaining the biosynthesis of retinoid cycle intermediates through microbial cells are limited and small attention has been paid.

Carotenogenesis occurs in bacteria through various pathways and can synthesis 700 different types of carotenoids

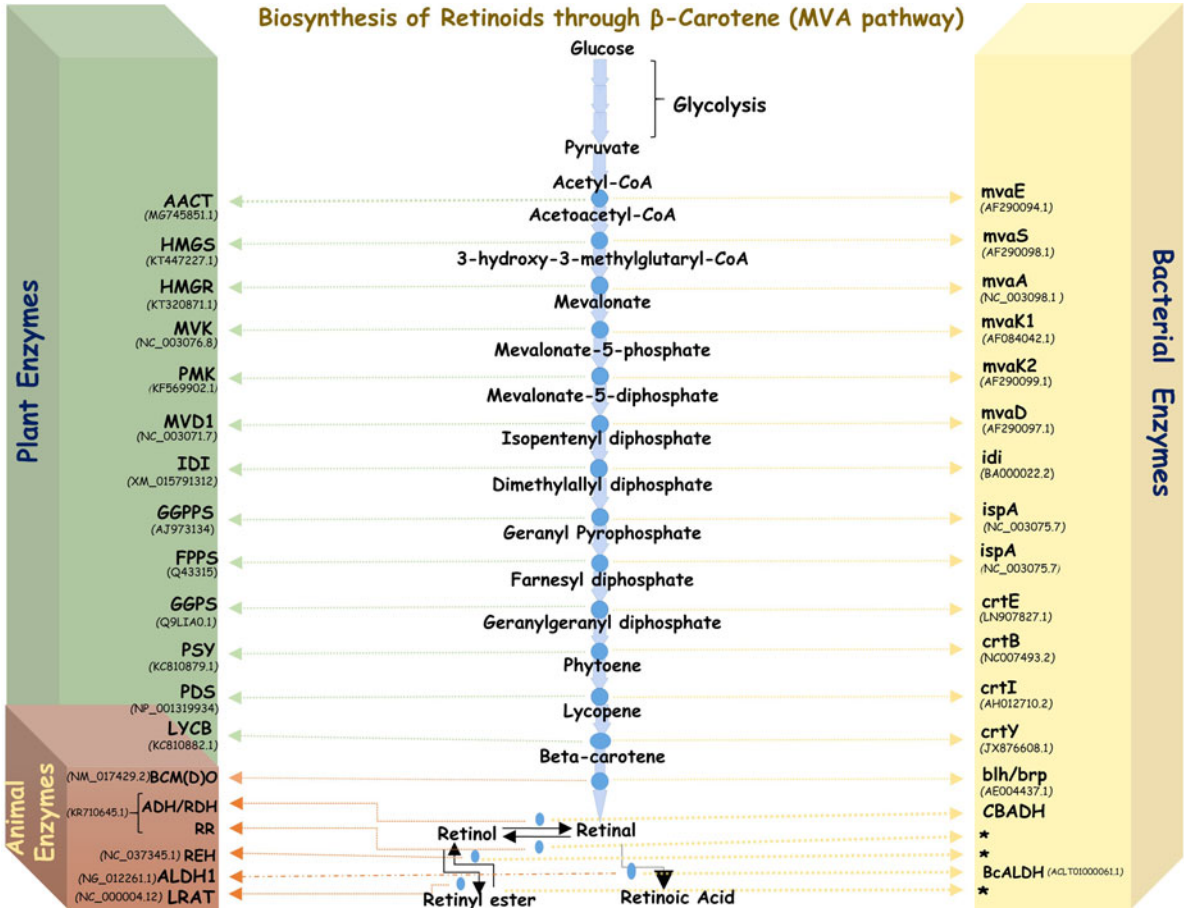

Figure 4. The biosynthesis pathway of β-carotene (MVA pathway) in plants and retinoid synthesis in animals and its equivalent steps in bacteria (modified from Bai et al. 2011). For simplicity, only major intermediates are shown. The gene accession numbers of the enzymes are given below the gene symbol in parentheses with the published authors. Enzymes: AACT, acetoacetyl-CoA thiolase; HMGS, hydroxymethylglutaryl-CoA synthase; HMGR, hydroxymethylglutaryl-CoA reductase; MVK, mevalonate kinase; PMK, phosphomevalonate kinase; MVD1, phosphomevalonate decarboxylase; IDI, isopentenyl diphosphate delta isomerase; GGPPS, geranylgeranyl diphosphate synthase; FPPS, farnesyl pyrophosphate synthase; GGPS, geranylgeranyl pyrophosphate synthase; PSY, phytoene synthase; PDS, phytoene desaturase; LYCB, lycopene β-cyclase; BCM(D)O, β-carotene 15,15′-mono(di)oxygenase; RR, retinal reductase; ADH, alcohol dehydrogenases; RDH, retinol dehydrogenase; ALDH1, aldehyde dehydrogenases; LRAT, lecithin retinol acyltransferase; REH, retinyl ester hydrolase; mvaE, Acetyl-CoA acetyltransferase; mvaS, HMG-CoA synthase; mvaA, HMG-CoA reductase; mvaK1, mevalonate kinase, mvaK2, phosphomevalonate kinase; mvaD, mevalonate diphosphate decarboxylase; idi, Isopentenyl-diphosphate Delta-isomerase; ispA, geranyl diphosphate synthase; crtE, bacterial GGPP synthase; crtB, bacterial phytoene synthase; crtI, carotenoid isomerase; crtY, bacterial lycopene cyclase; BCO, β-carotene oxygenases; blh, bacteriorhodopsin-related protein-like homolog protein; brp, bacterioopsin-related protein; BcALDH, *Bacillus cereus* Aldehyde dehydrogenase, CBADH, *Clostridium beijerinckii* alcohol dehydrogenases; ALDH1a/2, aldehyde dehydrogenase and *indicates no enzymes were reported.

(Hedl et al. 2002). All the isoprenoid compounds are derived from five-carbon (C5) units and in particular through the MVA pathway (Putter et al. 2017). A list of bacterial, plant and animal enzymes with corresponding genes and their accession number reported to be responsible for synthesis and metabolism of carotenoids and retinoids are outlined in Fig. 4. Since most of the prokaryotic carotenoid metabolism takes place through the MVA pathway starting with acetyl co-A, the important intermediates were given with reference. The genes and the enzymes identified for prokaryotic carotenoid metabolism were derived from a web server called ProCarDB (http://bioinfo.imtech.res.in/servers/procardb/) (Nupur et al. 2016). Most cyanobacteria and some pigment-producing bacteria having the phytoene synthase (CrtB) enzyme which mediates the phytoene synthesis step (Sabehi et al. 2005). The following steps such as phytoene to lycopene and β-carotene are catalyzed by phytoene desaturase (CrtI) and lycopene β-cyclase (CrtY) respectively. Some bacteria such as *Streptococcus* and *Staphylococcus* have carotenoid desaturase enzymes which can catalyze the entire pathway steps of the C40 carotenoids desaturation (Marshall and Wilmoth 1981). These data illustrate the ability of the bacteria to synthesize the β-carotene and to metabolize it into the retinoid products.

However, the carotenoid cleavage dioxygenases (CCD) enzymes of gut friendly bacteria have not been studied so far. Recently, some of the Proteobacteria such as *Sphingopyxis alaskensis, Novosphingobium aromaticivorans,* and mycobacteria such as *Mycobacterium tuberculosis* reported undergoing the cleavage reaction (Liang, Zhu, and Jiang 2017). The two bacterioopsin proteins thought to convert β-carotene into retinoids are bacteriorhodopsin-related protein-like homolog protein (Blh) and bacterioopsin-related protein (Brp).

The role of bacteria in the synthesis of retinoid intermediates such as retinoic acid, retinal and retinol demonstrated by different researchers is summarized here. Sabehi et al. 2005 revealed that the proteorhodopsin genes were

linked with *blh* genes that can encode the carotenoid cleavage enzymes. These genes were reported in multiple microorganisms like *Halobacterium halobium* (Shand and Betlach 1994), *Haloarcula marismortui* (Baliga et al. 2004), *Haloquadratum walsbyi*, and *Salinibacter ruber* (Mongodin et al. 2005) and *Halobacterium* sp. NRC-1 (Ng et al. 2000). Although these enzymes exhibit the capability to metabolize the carotenoids into retinoids, they differ in various properties from the mammalian BCM(D)O enzymes (Kim et al. 2009). While mining the genome of *Donghaeana dokdonensis* DSW-6, the *blh* gene, whose product is responsible for the cleavage of β-carotene into retinal molecules were unraveled (Kwon et al. 2013). Hong et al. 2016 identified aldehyde dehydrogenase enzyme from *Bacillus cereus* that having the potential to biotransform retinal to retinoic acid. Further, he elucidated that the amino acid sequence from the enzyme aldehyde dehydrogenase of *B. cereus* and mammalian were closely related. This bacterium is also reported to have the catalyzing power for converting the all-trans retinal to all-trans retinoic acid with NAD(P)+ and could reduce all-trans retinal to all-trans-retinol with the catalytic residues of the enzyme Glu266 and Cys300 along with NADPH. The retinaldehyde dehydrogenase enzyme was expressed in *E. coli* and observed that it functions through complexing with cellular retinol binding protein (Penzes, Wang, and Napoli 1997).

Sobreira et al. (2011) performed the phylogenetic analysis of retinol dehydrogenase enzymes in eukaryotes and prokaryotes through NCBI BLASTp 16. A large number of bacteria have shown closest hits majorly the Cyanobacteria, Bacteroidetes, and Planctomycetes. Further, he added that the enzymes of bacteria and animal orthologs are considered as the same clade since they are closely linked with each other. The metagenomics analysis uncovered certain gram-positive bacteria such as *Enterococcus* and *Streptococcus* spp. with *brp/blh* genes that are mimicking the (BCM(D)O) genes (Culligan et al. 2012).

In a human gut metagenomic study, certain gram-positive and gram-negative bacteria possessing the putative genes *brp/blh* were identified mimicking the BCM(D)O activity. The relevant genes might have undergone lateral gene transfer from multicellular eukaryotic cells to microorganisms in the aphotic nature of the gut environment (Sleator 2011, 2013). Culligan et al. (2012) revealed the genes encoding the enzyme homology to BCMO in human gut metagenomics library. The metabolic pathways of vitamin synthesis were highly represented in the enterotypes of 22 fecal metagenomes and implied a synergetic interrelationship with the host (Arumugam et al. 2011). The role of microbiota in vitamin A synthesis and transport should be explored in depth with relevant scientific insights to fill the gaps in VAD disease complications.

The human microbiome being considered as a second genome is one of the important sources of genetic diversity and metabolic adaptability. The genetic make-up of the bacteria depending on the liver, intestine, and gut may transform into retinoid producers through any one of the proposed phenomena explained in the following sections.

6. Testable hypotheses: microbial metabolism of carotenoids and retinoids

There are a considerable data supports and reveal the biochemistry behind the bacterial synthesis of folates, riboflavin, cobalamin, and other K and B vitamins. Qin et al. (2010) published the fact that the human gut microorganism can synthesize B-vitamins through pubSEED, a metagenomics analysis platform. In that study, subsystems of eight B-vitamin synthesizing genes were identified and used to predict the capable gut bacteria. Few studies have demonstrated that the bacterial enzymes mimicking the BCM(D)O can produce retinoids from β-carotene. However, the natural enzymes of the bacteria to biotransform the retinoids into various functional forms and the biochemical pathways is not well exploited (Peck et al. 2001; Sabehi et al. 2005). The importance of studying the role of gut microbiota in the biosynthesis of retinoids and bioconversion capability from carotenoids is emphasized through the following contextual scientific backgrounds.

6.1 Mechanisms proposed for microbial retinoid synthesis: symbiotic relationship pressures in acquiring the metabolic capabilities

One of the hidden metabolic organs that play a significant role in nutrition metabolisms such as bioaccessibility, bioavailability and bioconversion processes is the gut microbiota. The microbiome adapts rapidly with the human tissues through multiple factors that necessitate a symbiotic relationship. The gut commensal microbes are concerned about their wellbeing and response favorable to host in all the conditions. It is hypothesized that the microbiome may directly participate in host metabolic activities and support the host with the essential nutrients on a mutual benefit basis (Fukuda and Ohno 2014). Lack of retinoid affects the normal cellular functions and interferes with the nutrient digestion process (Reboul 2013). Higher β-carotene stores and demand in retinoid availability might impact the association between gut microbiota and the host. As a consequence of the demand due to poor bioconversion of β-carotene to retinoids, the gut bacteria could have developed the potentials which could also prolong beyond the demand period of retinoids. The phenomena of carotenoid metabolic gene transfer between the host and microbe and in turn the secondary changes to stabilize the same are part of the interactional symbiosis (Moran and Jarvik 2010). Hence, the assumption is that the gut microbes would play key roles as enzymes, mediators, and transporters in retinoid biosynthesis as part of the symbiotic relationship.

6.2 Mechanisms proposed for microbial retinoid synthesis: evolutionary and hologenomic adaptational effect

The microbiome adaptational effect to support the gut bacterial biosynthesis of vitamin A can be projected based on the following significant assumptions. The inter variations in

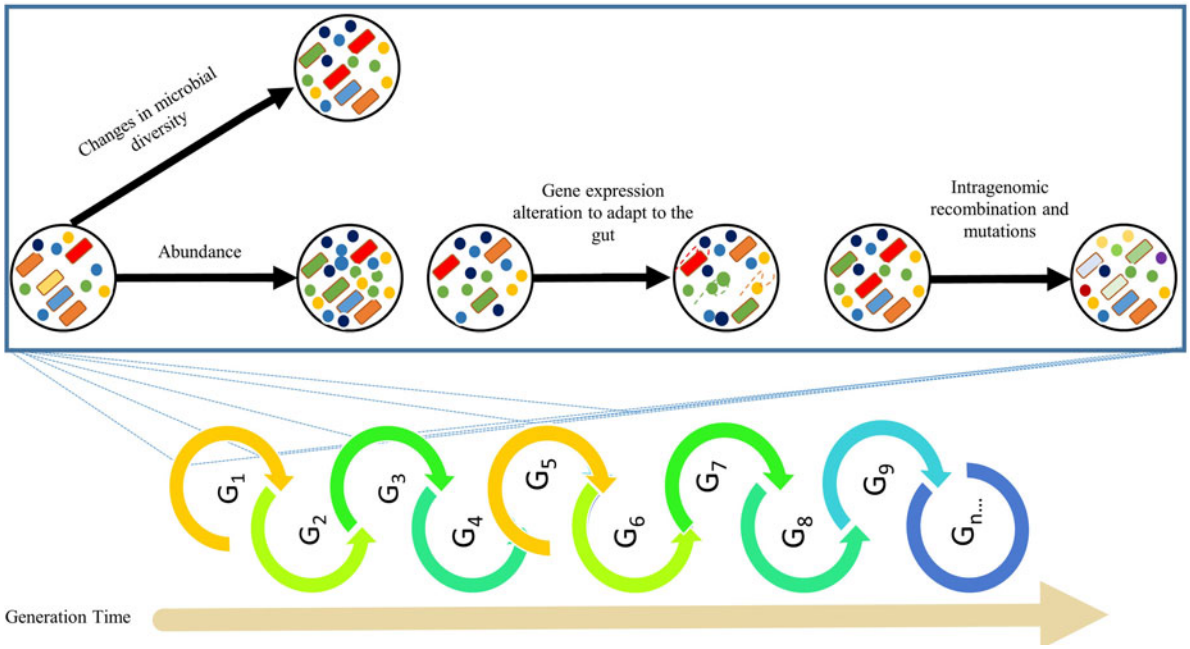

Figure 5. Proposed process in support of the adaptation of microbiome to host environment and through ecological and genetic changes due to the short generation times and large population size. G_1 to G_n denotes the generation period (adapted from, Li et al. 2016).

microbial diversity in response with host metabolic activities (high vitamin metabolism activity in liver and intestine), gene expression compatibility with the host condition and mobile genetic elements that could transfer the genes from the host to bacteria (Fig. 5). The principal phenomenon of remodeling its bacterial cells to associate with the eukaryotic cells is intragenomic recombination processes. Various clusters of microbial genes are activated or engineered in response to the adapted habitat (Hill et al. 2002). The epigenetic switches (Li et al. 2016), site-specific recombination (Van der Woude 2011), spontaneous mutations (Harms et al. 2015), and phenotypic switches work in a bi-directional manner. The genetic novelty in the gut microbes through mutators might also facilitate the biosynthesis of required compounds within the bacterial populations due to the fluctuating environments (Travis and Travis 2002). Although, the beneficial mutation ratio is comparatively lesser than the neutral or detrimental mutations (e.g., *E. coli* beneficial mutation per genome per replication is 2×10^{-9}), the associated microbes of the organs tend to adapt faster (Imhof and Schlotterer 2001). It could be proposed that the retinoid metabolizing genes acquired by the microbiota also should have been conserved in successive generations due to the shorter multiplication times and larger population sizes.

6.3 Mechanisms proposed for microbial retinoid synthesis: horizontal transfer of retinoid metabolizing gene

One of the natural phenomena and principle behind the adaptation of living organisms in their host environment is horizontal gene transfer. Even a complex biosynthetic pathway can be gained by this process and transform into a favorable host organism (Malys, Campbell, and Malys 2015). The constraints in host carotenoid metabolic pathways were addressed by the microbial enzyme and coevolved with the local ecosystem. The selective evolution of biosynthetic pathway and the ability to integrate the microbial genome with the host to function in the receipt metabolic network is one of the crucial possible mechanisms. The protein encoding transcripts including retinal biosynthetic enzymes were characterized in Flavobacterium species (Kimura et al. 2011). In Bacteroidetes genomes, the β-carotene gene is flanked only by *blh* and also suggests it is by the lateral gene transfer of rhodopsin genes and is a frequent process (Pinhassi et al. 2016). A proportion of carotenoids found in the membrane of haloarchae and the symbiotic *Salinibacter* sp. produced red colonies of similar appearance. The genes responsible for the conversion of carotenoids to retinoids may be evolved through horizontal gene transfer from ancestral organisms (Anton et al. 2002; Lutnaes et al. 2004).

6.3.1 Phylogenetic analysis to elucidate the gene transfer phenomena from eukaryotes to prokaryotes

The most relevant mechanism of gut bacterial evolution can be explained by the lateral gene transfer (LGT). Numerous forces and positive selection pressure influence the LGT (Gillings 2017). Only a few researchers have elucidated the LGT event from a eukaryote to prokaryote. Arias et al. (2012) speculated the transfer of glycoside hydrolase gene from Archaeplastida (eukaryote) to Bacteroides through Bayesian and bootstrapped maximum likelihood methods. The evolutionary proximity of retinoid metabolism pathways among eukaryotic organisms such as animals and the bacteria was investigated and narrated through phylogenetic analysis of the responsible enzymes (Kiser, Golczak, and Palczewski 2014). The aldehyde dehydrogenase catalyzing the transformation of retinaldehyde to retinoic acid is reasoned and proved with standard models of evolution

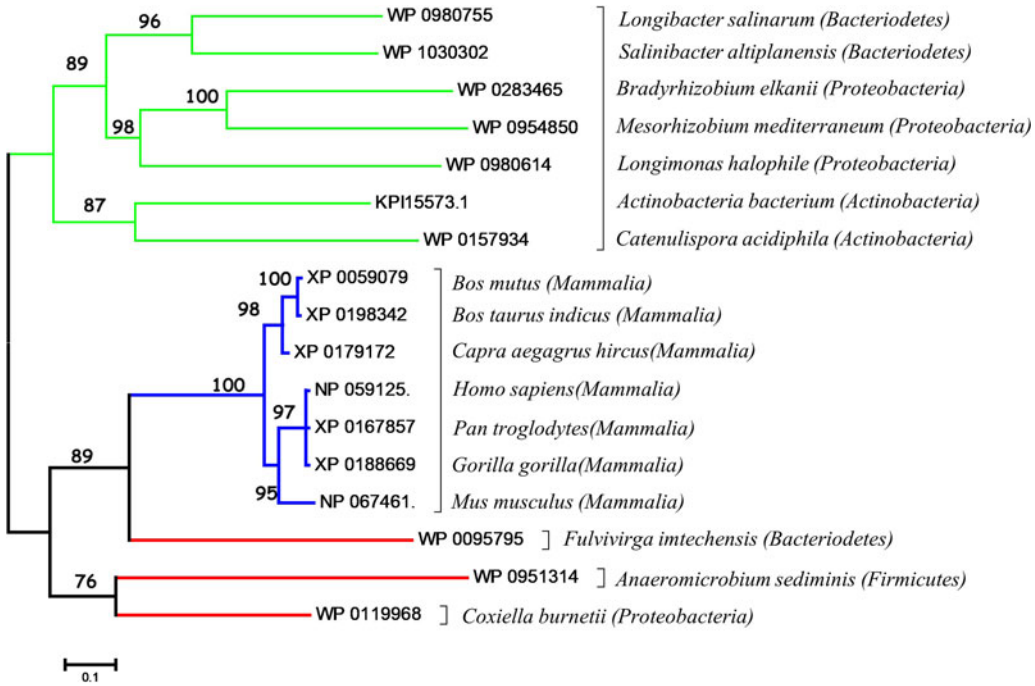

Figure 6. Maximum-likelihood phylogeny unrooted tree of β-carotene mono(di) oxygenases enzymes in selected prokaryotes and eukaryotes.

(Millard et al. 2014). Doolittle (1998) demonstrated the transformation of genes from protist to food bacteria through LGT phenomenon.

In this review, we identified and retrieved the protein homologs of BCM(D)O from selected eukaryotic and prokaryotic organisms by BLASTp search at the NCBI non-redundant database. Redundant sequences with >90% non-pairing amino acid sequences were removed, and entirely aligned sequences were kept for phylogenetic reconstruction. The sequences were aligned through MAFFT version 7 (Katoh and Standley 2013). The best protein substitution model JTT + I + G + F fit for the sequences was generated using Protest (Perez Sirkin et al. 2017) and was used to perform maximum likelihood (ML) estimation with RAxML ver.8.2.4 (Bohm et al. 2015). For the best-scoring tree with well-supported nodes, 1000 bootstrap replicates were done to generate the ML tree topology. The unrooted tree generated through the ML method is given in Fig. 6.

The RAxML bootstrap values more than 70 are indicated at the branches. Genebank accession numbers are provided at the end of the branch. The eukaryotic organisms with BC(M)DO enzymes were indicated as blue color and the green color highlights the bacteria. The red color highlights the bacteria closely related to the eukaryotic organisms. In particular, *Fulvivirga imtechensis* belonging to the phylum Bacteroidetes subclades from the mammalian animals indicates that a maximum homology found among the protein sequences which indicates the possibility of horizontal gene transfer like phenomena.

Fulvivirga imtechensis, a gram-negative aerobic bacterium is reported to have some eukaryotic characters through evolutionary phylogenetic analysis. In the comparative genomics study using *in silico* bacterial carotenoid biosynthesis pathway, Bacteroidetes which is a sub-lineage from Actinobacteria is proposed to possesses carotenoid oxygenases and desaturases enzyme families. The evolutionary mechanism suggested for this phenomena may be explained through horizontal gene transfer, co-evolution with other biochemical reactions, gene acquisition followed by gene loss and positive selection (Klassen 2010). In multiple sequence alignment of specific bacterial annexin proteins with the eukaryotic domains, the sequence motifs are strongly conserved in various sites of *Fulvivirga imtechensis.* This phenomenon might be explained by horizontal gene transfer between eukaryotic organisms and bacteria through protozoan eukaryotes (Kodavali et al. 2014).

It is also inferred that the animals like *Bos mutus* (domestic yak), *Bos Taurus* (cow) and *Capra hircus* (goat) with a close relationship with human BCM(D)O enzymes and their liver as an abundant source of the vitamin A is a promising area of concern. *In vitro* examination of the microbial flora of these animals using suitable culture system may unravel some significant outbreaks in the biosynthesis of vitamin A. Owing to the consequence of the long-term coevolution among the gut bacteria and the eukaryote, it can be speculated that the bacteria could acquire the BCM(D)O genes because of an LGT like event.

6.4 Mechanisms proposed for microbial retinoid synthesis: Putative enzymes reported synthesizing retinoids from carotenoids

Zhang et al. (2018) discovered the inherent enzymes of *E. coli* capable of converting retinal into retinol or retinyl esters. A group of genes in the microbes encodes the enzymes with retinol reductase activity. Kim et al. (2009) find out the salt tolerance genes that encode a putative carotenoid metabolizing enzyme with homology to a BCMO protein, which capable of cleaving a β-carotene to two molecules of retinal. This enzyme also shares more similarity with a *brp/blh*-family 15, 15′-β-carotene monooxygenase

from *Prevotella marshii* DSM 16973 and with *Bifidobacterium bifidum* BGN 4 (Kim et al. 2009; Peck et al. 2001). In the bacteria *Staphylococcus aureus,* the orange carotenoid staphyloxanthin is synthesized by acyltransferase enzymes (Pelz et al. 2005). *Synechocystis* sp. PCC 6803, a cyanobacterium encodes the retinal synthesizing enzyme which can metabolize the β-apo-carotenals (Ruch et al. 2005). In the carotenoid biosynthesis studies, specific putative ADP-binding sites in the N-terminal portion of the carotenoid dehydrogenase enzymes were identified. The metabolic pathway could have preserved evolutionarily in bacteria. The bacterial enzymes mimicking the carotenoid metabolism were purified and characterized (Armstrong, Alberti, and Hearst 1990). These findings show the probability of putative enzymes in the bioconversion of carotenoids into retinoids.

6.5 Mechanisms proposed for microbial retinoid synthesis: literature insights behind the bacterial enzymes in the synthesis of retinoids

The *Halobacterium* strains are proven with the capability to synthesis retinal from carotenoids. However, these bacteria possess carotenoid synthesizing genes and not the carotenoids oxygenase homologs. Hence it can be postulated that retinal might have biosynthesized through some independent enzymatic pathways (Ruch et al. 2005). There are some speculations behind the retinoid production that there is a direct biosynthesis pathway without the carotenoids and its cleavage mechanism (Peck et al. 2001). Besides, the proven carotenoid oxygenases enzymes BCOI (β-carotene-15,15′-oxygenase) and BCOII (β-carotene-9,9′-oxygenase); a third enzyme retinyl ester binding protein could exist rather than the oxygenases (Mata et al. 2004).

About six enzymes are required to biosynthesis β-carotene from isopentenyl diphosphate (IPP) (Gómez-Consarnau et al. 2007; Kimura et al. 2011), and the *blh* gene can produce enzymes mimicking the BCMO which transform β-carotene into retinoids. The genes responsible for all these enzymes were identified in the genome of the strains of *Dokdonia* sp. PRO95, 4H-3-735, MED134, and R-8282. These organisms lack the genes for carotenoid metabolism (*crtO* gene). This phenomenon indicates that they possess a new group of rhodopsins which aid in catabolizing the carotenoids to retinoids (Riedel et al. 2013). The biosynthetic capacity for retinal was found in *Nostoc,* whereas, in the genome, no sequence similarity with *blh* was found (Bouvier et al. 2005; Moise, von Lintig, and Palczewski 2005). Based on *Synechocystis* enzyme apocarotenoid 15,15′-oxygenase assumption, *Nostoc* might utilize the apocarotenals to synthesis retinoids through an alternative pathway without carotenoids cleavage enzymes (Ruch et al. 2005).

Overall, with the above postulated scientific assumptions, research insights on the role of gut microflora in vitamin A metabolism in carotenoid and retinoid production pathway would be a solution to overcome VAD disorders.

7. Genetically engineered microbes in vitamin a synthesis

Owing to the disadvantages of chemical synthesis and complicated extraction process, researchers have developed competent bacteria through genetic engineering. The carotenoid biosynthesis is successful through natural and genetically modified biosynthetic pathways in several bacteria (Miura et al. 1998). The sensitivity towards oxygen and aqueous culture system is a challenging issue in synthesis and extraction of retinoid compounds. Plasmid constructed genes such as pAC-CAR16DcrtX and its expression are successfully demonstrated in *E. coli* for producing β-carotene. The *E. coli* genetically engineered with *brp/blh* genes coding for BCM(D)O enzymes produced retinal, and the microbial cells acquired the orange or red color (Martinez et al. 2007; Beja et al. 2000; Wang et al. 2012). The engineered *E. coli* reported to produce a high level of β-carotene can be modified with oxygenase enzymes specific to β-carotene oxidation. Further, the *E. coli* produce protein-bound retinal which is not appropriate for therapeutic applications (Zhang et al. 2018). A two-phase culture system comprising a dodecane layer and the culture broth was effective to minimize the degradation of retinoids. Recombinant *E. coli* produces retinal through MVA pathway from acetyl co-A. (Jang et al. 2011). The *blh* gene was isolated from uncultured marine bacterium 66A03 and expressed in *E. coli.* The expressed enzyme is capable of cleaving β-carotene at 15, 15′ double bond to yield two molecules of all-trans-retinal (Kim et al. 2009)

The role of surfactants on retinoid production such as agitation, pH, and temperature in bioreactors using genetically engineered *E. coli,* was optimized and proved useful for mass production (Lee et al. 2012). A recombinant *E. coli* constructed with chicken BCMO gene demonstrated good survivability in cattle rumen conditions with stable expression of the target genes. Further, the bacteria are tested for generally regarded as safe (GRAS) organism (Garcia-Lopez et al. 2012). Benefits aside, the genetically modified bacteria have been a bone of contention and considered as a threat to human health and the environment.

8. Strategies and predictable routes for the identification of retinoid synthesizing bacteria

The modern genetic tools such as 16s rRNA sequencing provide genus and species level identification of both culturable and unculturable archaea and bacteria. Also, the bioinformatics tools such as cluster linkage analysis enable screening of target genes among the prokaryotes and eukaryotes. Taking these advantages, by integrating the modern molecular and microbiological techniques, metabolomics approach would be a feasible route for identifying a bacterium with retinoid synthesizing genes.

In the recent decades, some of the researchers experimented the interactions between vitamin A biosynthesis and rumen microflora. The major sources of vitamin A are pre-vitamins like retinyl acetate from foods of animal origin such as beef liver (4–20 mg retinol/100 g) and pro-vitamins

like β-carotene from plant-based foods (roughly 60–120 mg β-carotene per 100 g) (USDA 2004). The highest concentrations of preformed vitamin A are avian and mammalian livers. The cattle fed with good quality forages and free-range cattle stores β-carotene in adipose tissue and also nutritionally superior due to its higher contents of vitamins and conjugated linoleic acids (Rohrle et al. 2011). However, the animals cannot synthesize retinoid "de novo," and hence they rely on BCM(D)O to transform carotenoids into retinal. The sheep excreted more β-carotene in the feces than they consumed which could be due to the gut microbial organisms (McGillivray 1951). King, Ohman, and Smith (1962) reported that β-carotene is bio-converted into pre-vitamin compounds in steer and sheep rumen. Certain strains of bacteria such as *Staphylococcus, Streptococcus*, and *Bacillus* are known to synthesize carotenoids (Aono and Horikoshi 1991). The bioavailability of carotenoids is affected by cattle rumen microflora, and an indirect mechanism may be responsible for this effect. A modulation in the microbial population is altering the liver storage of vitamin A and a decrease in β-carotene (Almquist and Maurer 1955).

Given this scientific evidence that the commensal gut microflora is one of the vital sources of a range of vitamins to the host, showing light on the potential of gut bacteria in organisms like cattle would be an ideal strategy. The carotenoid intake by the ruminants is not affected by ruminal fermentation and is absorbed in the liver and small intestine where the enzyme BCM(D)O cleaves it into two retinal molecules (Garcia-Lopez et al. 2012). The bioconversion of β-carotene into retinoids occurs rapidly in the liver and intestinal tissues of the grass-fed cattle which could be influenced by several factors (Green and Fascetti 2016). Further, investigations have to be made to spot out the probiotic bacteria that can colonize and metabolize or co-metabolize beta-carotene and retinoids.

9. Future research projections to combat VAD: probiotics and synbiotics

One of the general therapeutic strategies that can beneficially influence the host metabolism to improve the health is pre or probiotic functional foods. The retinoid producing probiotics may provide a complementary source of vitamin and would prevent VAD. The prebiotics can stimulate the growth and or activity of the colonizing probiotic beneficial bacteria. While a synbiotic, the combination of probiotic microbes and prebiotic substances could effectively mediate the microbe-directed therapeutics (Pandey, Naik, and Vakil 2015). The synbiotic products such as oligosaccharides with beneficial microbes such as lactobacilli and bifidobacteria for the production of short chain fatty acids are successful therapy in practice (Rastall and Gibson 2015). Apart from induction of specific antimicrobial peptides, chemokines, competitive inhibition of pathogens and immune homeostasis, the probiotic bacteria can intermediate the organ-specific metabolic pathways. Interfering with the gut metabolism with targeted beneficial microbes would be an attractive future therapy design.

The probiotic gut-friendly bacteria have been documented for various therapeutic purpose in clinical trials. Formulating the vitamin producing microorganisms in nutritive drinks or foods which can mutually associate with the gut, synthesis carotenoids and or retinoids from β-carotene would be a promising solution. Long-term association and tendency to release the biosynthesized molecules over a prolonged period are some of the advantages in developing a retinoid producing synbiotic food. Pompei et al. 2007 used lyophilized bifidobacteria in the synbiotic formulation to increase the serum folate concentration. The probiotic strain *E. coli* Nissle 1914 isolated from World War I soldier been used to treat bowel diseases without side effects in Europe (Schultz 2008). Such a probiotic bacterium with the capacity to produce and deliver vitamin A from β-carotene would be a natural alternative to combat VAD.

10. Conclusion

In conclusion, this review elucidates the hypothetical facts behind the role of microorganisms in particular gut microbiota in vitamin A biosynthesis. An inexpensive, readily available source of vitamin A apart from dietary intake, fortification, and direct dose would be finding probiotic bacteria capable of producing carotenoids and retinoids. Consumption of probiotic bacteria in live supplement or synbiotic form would also become a strategy for improving the bioavailability of the retinoid compounds in liver and other target sites of the body. Further, a finely focused approach to search for gut-friendly vitamin A-producing bacterium would provide new avenues of research about the alternative biosynthetic strategies. A complex task and a necessary goal at the moment would be mapping the gut microbiota, especially from the animal such as free-range cattle liver in carotenoid and retinoid metabolism. If this task could be accomplished, fighting and eliminating the VAD is not-too-distant in future.

Acknowledgments

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Abbreviations

AACT	Acetoacetyl-CoA thiolase
ADH	Alcohol dehydrogenase
ALDH1	Aldehyde dehydrogenase
BCM(D)O	β-carotene 15,15′-mono (di) oxygenase
BCMO	β-carotene 15, 15′-monooxygenase
BCOI	β-carotene-15, 15′-oxygenase
BCOII	β-carotene-9′, 10′-oxygenase
Blh	Bacteriorhodopsin-related protein-like homolog protein
Brp	Bacterioopsin-related protein
CCD	Carotenoid cleavage dioxygenases
CRBP	Cellular retinol-binding protein
CYP	Cytochrome enzyme P450
FPPS	Farnesyl pyrophosphate synthase
FXR	Farnesoid X receptor

GGPPS	Geranylgeranyl diphosphate synthase
GGPS	Geranylgeranyl pyrophosphate synthase
GRAS	Generally regarded as safe
HMGR	Hydroxymethylglutaryl-CoA reductase
HMGS	Hydroxymethylglutaryl-CoA synthase
IDI	Isopentenyl diphosphate delta isomerase
LGT	Lateral gene transfer
LPS	Lipopolysaccharide
LRAT	Lecithin: retinol acyltransferase
LYCB	Lycopene β-cyclase
MEP	2-C-methyl-D-erythritol-4-phosphate
ML	Maximum likelihood
MVA	Mevalonate
MVD1	Phosphomevalonate decarboxylase
MVK	Mevalonate kinase
NTR	Nuclear transcription receptor
PDS	Phytoene desaturase
PMK	Phosphomevalonate kinase
PSY	Phytoene synthase
RBP	Retinol-binding protein
RDH	Retinol dehydrogenase
REH	Retinyl ester hydrolase
RR	Retinaldehyde reductase
RXR	Retinoid X receptor
UNICEF	United Nations children's fund
VAD	Vitamin A deficiency

References

Akerstrom, B., D. R. Flower, and J. P. Salier. 2000. Lipocalins: Unity in diversity. *Biochimica et Biophysica Acta* **1482**:1–8. doi:10.1016/S0167-4838(00)00137-0..

Akhtar, S., A. Ahmed, M. A. Randhawa, S. Atukorala, N. Arlappa, T. Ismail, and Z. Ali. 2013. Prevalence of vitamin a deficiency in South Asia: Causes, outcomes, and possible remedies. *Journal of Health, Population, and Nutrition* **31** (4):413–423. doi:10.3329/jhpn.v31i4.19975.

Almquist, H. J., and S. Maurer. 1955. The effect of antibiotic, antioxidant, and fat on conversion of carotene to vitamin a in the chicken. *Archives of Biochemistry and Biophysics* **55** (1):297–298. doi:10.1016/0003-9861(55)90572-5.

Anton, J., A. Oren, S. Benlloch, F. Rodriguez-Valera, R. Amann, and R. Rossello-Mora. 2002. *Salinibacter ruber* gen. nov., sp. nov., a novel, extremely halophilic member of the bacteria from saltern crystallizer ponds. *International Journal of Systematic and Evolutionary Microbiology* **52**:485–491. doi:10.1099/00207713-52-2-485.

Aono, R., and K. Horikoshi. 1991. Carotenes produced by alkaliphilic yellow pigmented strains of *Bacillus*. *Agricultural and Biological Chemistry* **55** (10):2643–2645. doi:10.1271/bbb1961.55.2643.

Arias, M. C., E. G. J. Danchin, P. Coutinho, B. Henrissat, and S. Ball. 2012. Eukaryote to gut bacteria transfer of a glycoside hydrolase gene essential for starch breakdown in plants. *Mobile Genetic Elements* **2** (2):81–87. doi:10.4161/mge.20375.

Armstrong, G. A. 1994. Eubacteria show their true colors: Genetics of carotenoid pigment biosynthesis from microbes to plants. *Journal of Bacteriology* **176** (16):4795–4802. doi:10.1128/jb.176.16.4795-4802.1994.

Armstrong, G. A., M. Alberti, and J. E. Hearst. 1990. Conserved enzymes mediate the early reactions of carotenoid biosynthesis in nonphotosynthetic and photosynthetic prokaryotes. *Proceedings of the National Academy of Sciences of the United States of America* **87** (24):9975–9979. doi:10.1073/pnas.87.24.9975.

Arumugam, M., J. Raes, E. Pelletier, D. Le Paslier, T. Yamada, D. R. Mende, G. R. Fernandes, J. Tap, T. Bruls, J.-M. Batto, et al. 2011. Enterotypes of the human gut microbiome. *Nature* **473** (7346):174–180. doi:10.1038/nature09944.

Bai, C., R. M. Twyman, G. Farre, G. Sanahuja, P. Christou, T. Capell, and C. Zhu. 2011. A golden era-pro-vitamin a enhancement in diverse crops. *In Vitro Cellular & Developmental Biology – Plant* **47** (2):205–221. doi:10.1007/s11627-011-9363-6

Baliga, N. S., R. Bonneau, M. T. Facciotti, M. Pan, G. Glusman, E. W. Deutsch, P. Shannon, Y. Chiu, R. S. Weng, and R. R. Gan. 2004. Genome sequence of *Haloarcula marismortui*: A halophilic Archaeon from the Dead sea. *Genome Research* **14** (11):2221–2234. doi:10.1101/gr.2700304.

Beja, O., L. Aravind, E. V. Koonin, M. T. Suzuki, A. Hadd, L. P. Nguyen, S. B. Jovanovich, C. M. Gates, R. A. Feldman, and J. L. Spudich. 2000. Bacterial rhodopsin: Evidence for a new type of phototrophy in the sea. *Science* **289** (5486):1902–1906. doi:10.1126/science.289.5486.1902.

Berry, D. C., C. M. Croniger, N. B. Ghyselinck, and N. Noy. 2012. Transthyretin blocks retinol uptake and cell signalling by the holo-retinol-binding protein receptor stra6. *Molecular and Cellular Biology* **32** (19):3851–3859. doi:10.1128/MCB.00775-12.

Bishop, R. E. 2005. The lipid a palmitoyltransferase PagP: Molecular mechanisms and role in bacterial pathogenesis. *Molecular Microbiology* **57** (4):900–912. doi:10.1111/j.1365-2958.2005.04711.x.

Blaser, M. J. 2016. Antibiotic use and its consequences for the normal microbiome. *Science (New York, N.Y.)* **352** (6285):544–545. doi:10.1126/science.aad9358.

Bohm, M.-E., C. Huptas, V. M. Krey, and S. Scherer. 2015. Massive horizontal gene transfer, strictly vertical inheritance and ancient duplications differentially shape the evolution of *Bacillus cereus* enterotoxin operons *hbl, cytK* and *nhe*. *BMC Evolutionary Biology* **15**:246. doi:10.1186/s12862-015-0529-4.

Bonet, M. L., J. A. Canas, J. Ribot, and A. Palou. 2015. Carotenoids and their conversion products in the control of adipocyte function, adiposity and obesity. *Archives of Biochemistry and Biophysics* **572**:112–125. doi:10.1016/j.abb.2015.02.022.

Borel, P., and C. Desmarchelier. 2017. Genetic variations associated with vitamin a status and vitamin a bioavailability. *Nutrients* **9** (3):246. doi:10.3390/nu9030246.

Bouvier, F., J. C. Isner, O. Dogbo, and B. Camara. 2005. Oxidative tailoring of carotenoids: A prospect towards novel functions in plants. *Trends in Plant Science* **10** (4):187–194. doi:10.1016/j.tplants.2005.02.007.

Culligan, E. P., R. D. Sleator, J. R. Marchesi, and C. Hill. 2012. Functional metagenomics reveals novel salt tolerance loci from the human gut microbiome. *The ISME Journal* **6** (10):1916–1925. doi:10.1038/ismej.2012.38.

Culligan, E. P., R. D. Sleator, J. R. Marchesi, and C. Hill. 2014. Metagenomic identification of a novel salt tolerance gene from the human gut microbiome which encodes a membrane protein with homology to a brp/blh-family β-Carotene 15,15′-monooxygenase. *PLoS One* **9** (7):e103318. doi:10.1371/journal.pone.0103318.

Curley, R. W., and M. J. Robarge. 1997. Retinoid structure, chemistry, and biological active derivatives. In *Retinoids: Their physiological function and therapeutic potential, Advances in organ biology*, eds. Bittar, E. E., and G. V. Sherbet, vol. **3**, 1st ed, 1–34. Bingley, England: JAI Press Inc. doi:10.1016/s1569-2590(08)60051-8.

de Pee, S., C. E. West, D. Permaesih, S. Martuti, S. Muhilal, and J. G. Hautvast. 1998. Orange fruit is more effective than are dark-green, leafy vegetables in increasing serum concentrations of retinol and beta-carotene in school-children in Indonesia. *The American Journal of Clinical Nutrition* **68**:1058–1067. doi:10.1093/ajcn/68.5.1058.

Doolittle, W. F. 1998. You are what you eat: A gene transfer ratchet could account for bacterial genes in eukaryotic nuclear genomes. *Trends in Genetics* **14**:307–311. doi:10.1016/S0168-9525(98)01494-2.

During, A., A. Nagao, C. Hoshino, and J. Terao. 1996. Assay of beta-carotene 15,15′-dioxygenase activity by reverse-phase high-pressure liquid chromatography. *Analytical Biochemistry* **241** (2):199–205. doi:10.1006/abio.1996.0400.

Eckburg, P. B., E. M. Bik, C. N. Bernstein, E. Purdom, L. Dethlefsen, M. Sargent, S. R. Gill, K. E. Nelson, and D. A. Relman. 2005. Diversity of the human intestinal microbial flora. *Science* **308** (5728):1635–1638. doi:10.1126/science.1110591.

Esaki, S., S. A. Malkaram, and J. Zempleni. 2012. Effects of single-nucleotide polymorphisms in the human *holocarboxylase synthetase*

gene on enzyme catalysis. *European Journal of Human Genetics* **20** (4):428–433. doi:10.1038/ejhg.2011.198.

Fukuda, S., and H. Ohno. 2014. Gut microbiome and metabolic diseases. *Seminars in Immunopathology* **36** (1):103–114. doi:10.1007/s00281-013-0399-z.

Garcia-Lopez, E., A. Gonzalez-Gallardo, A. Antaramian, M. L. González-Dávalos, A. Shimada, A. Varela-Echavarria, and O. Mora. 2012. In vitro conversion of β-carotene to retinal in bovine rumen fluid by a recombinant β-carotene-15, 15 mooxygenase. *International Journal for Vitamin and Nutrition Research* **82**:94–103. doi:10.1024/0300-9831/a000098.

Gerard, P. 2014. Metabolism of cholesterol and bile acids by the gut microbiota. *Pathogens* **3** (1):14–24. doi:10.3390/pathogens3010014.

German, J. B., and C. J. Dillard. 2006. Composition, structure and absorption of milk lipids: A source of energy, fat-soluble nutrients and bioactive molecules. *Critical Reviews in Food Science and Nutrition* **46** (1):57–92.

Gillings, M. R. 2017. Lateral gene transfer, bacterial genome evolution, and the anthropocene: Lateral gene transfer in the anthropocene. *Annals of the New York Academy of Sciences* **1389** (1):20–36. doi:10.1111/nyas.13213.

Gómez-Consarnau, L., J. M. González, M. Coll-Llado, P. Gourdon, T. Pascher, R. Neutze, C. Pedros-Alio, and J. Pinhassi. 2007. Light stimulates growth of proteorhodopsin-containing marine flavobacteria. *Nature* **445**:210–213. doi:10.1038/nature05381.

Goodman, D. S., H. S. Huang, M. Kanai, and T. Shiratori. 1967. The enzymatic conversion of all-trans beta-carotene into retinal. *Journal of Biological Chemistry* **242**:3543–3554.

Goodman, D. S., R. Blomstrand, B. Werner, H. S. Huang, and T. Shiratori. 1966. The intestinal absorption and metabolism of vitamin a and beta-carotene in man. *Journal of Clinical Investigation* **45** (10):1615–1623. doi:10.1172/JCI105468.

Green, A. S., and A. J. Fascetti. 2016. Meeting the vitamin a requirement: the efficacy and importance of β-carotene in animal. *The Scientific World Journal* **7393620**:22. doi:10.1155/2016/7393620.

Grolier, P., P. Borel, C. Duszka, S. Lory, M. C. Alexandre-Gouabau, V. Azais-Braesco, and L. Nugon-Baudon. 1998. The bioavailability of alpha- and beta-carotene is affected by gut microflora in the rat. *The British Journal of Nutrition* **80** (2):199–204. doi:10.1017/S0007114598001111.

Gronowska-Senger, A., and G. Wolf. 1970. Effect of dietary protein on the enzyme from rat and human intestine which converts β-Carotene to retinal. *The Journal of Nutrition* **100** (3):300–308. doi:10.1093/jn/100.3.300.

Harms, V., C. Nowak, S. Carl, and V. Muñoz-Fuentes. 2015. Experimental evaluation of genetic predator identification from saliva traces on wildlife kills. *Journal of Mammalogy* **96** (1):138–143. doi:10.1093/jmammal/gyu014.

Hartstra, A. V., M. Nieuwdorp, and H. Herrema. 2016. Interplay between gut microbiota, its metabolites and human metabolism: dissecting cause from consequence. *Trends in Food Science and Technology* **57**:233–243. doi:10.1016/j.tifs.2016.08.009.

He, Y., L. Gong, Y. Fang, Q. Zhan, H. Liu, Y. Lu, G. L. Guo, L. Lehman-McKeeman, J. Fang, and Y. J. Wan. 2013. The role of retinoic acid in hepatic lipid homeostasis defined by genomic binding and transcriptome profiling. *BMC Genomics* **14** (1):575. doi:10.1186/1471-2164-14-575.

Hedl, M., A. Sutherlin, E. I. Wilding, M. Mazzulla, D. McDevitt, P. Lane, J. W. Burgner, K. R. Lehnbeuter, C. V. Stauffacher, M. N. Gwynn, and V. W. Rodwell. 2002. *Enterococcus faecalis* acetoacetyl-coenzyme a thiolase/3-hydroxy-3-methylglutaryl-coenzyme a reductase, a dual-function protein of isopentenyl diphosphate biosynthesis. *Journal of Bacteriology* **184** (8):2116–2122. doi:10.1128/JB.184.8.2116-2122.2002.

Heuman, D. M. 1989. Quantitative estimation of the hydrophilic-hydrophobic balance of mixed bile salt solutions. *Journal of Lipid Research* **30** (5):719–730.

Hill, C., P. D. Cotter, R. D. Sleator, and C. G. M. Gahan. 2002. Bacterial stress response in *Listeria monocytogenes*: Jumping the hurdles imposed by minimal processing. *International Dairy Journal* **12** (2–3):273–283. doi:10.1016/S0958-6946(01)00125-X.

Himoto, T., and T. Masaki. 2018. Associations between zinc deficiency and metabolic abnormalities in patients with chronic liver disease. *Nutrients* **10**(1):88. doi:10.3390/nu10010088.

Hong, S. H., H. P. Ngo, H. K. Nam, K. R. Kim, L. W. Kang, and D. K. Oh. 2016. Alternative biotransformation of retinal to retinoic acid or retinol by an aldehyde dehydrogenase from *Bacillus cereus*. *Applied and Environmental Microbiology* **82** (13):3940–3946. doi:10.1128/AEM.00848-16.

Hooper, L. V., T. Midtvedt, and J. I. Gordon. 2002. How host-microbial interactions shape the nutrient environment of the mammalian intestine. *Annual Review of Nutrition* **22** (1):283–307. doi:10.1146/annurev.nutr.22.011602.092259.

Imhof, M., and C. Schlotterer. 2001. Fitness effects of advantageous mutations in evolving *Escherichia coli* populations. *Proceedings of the National Academy of Sciences of the United States of America* **98** (3):1113–1117. doi:10.1073/pnas.98.3.1113.

Jang, H. J., S. H. Yoon, H. K. Ryu, J. H. Kim, C. L. Wang, J. Y. Kim, D. K. Oh, and S. W. Kim. 2011. Retinoid production using metabolically engineered *Escherichia coli* with a two-phase culture system. *Microbial Cell Factories* **10** (1):59. doi:10.1186/1475-2859-10-59.

Johansson, M. E., M. Phillipson, J. Petersson, A. Velcich, L. Holm, and G. C. Hansson. 2008. The inner of the two Muc2 mucin-dependent mucus layers in Colon is devoid of bacteria. *Proceedings of the National Academy of Sciences of the United States of America* **105** (39):15064–15069. doi:10.1073/pnas.0803124105.

Karin, M., and H. Clevers. 2016. Reparative inflammation takes charge of tissue regeneration. *Nature* **529** (7586):307–315. doi:10.1038/nature17039.

Katoh, K., and D. M. Standley. 2013. MAFFT multiple sequence alignment software version 7: Improvements in performance and usability. *Molecular Biology and Evolution* **30** (4):772–780. doi:10.1093/molbev/mst010.

Kellogg, T. F. 1974. Steroid balance and tissue cholesterol accumulation in germfree and conventional rats fed diets containing saturated and polyunsaturated fats. *The Journal of Lipid Research* **15** (6):574–579.

Khan, N. C., C. E. West, S. de Pee, D. Bosch, H. D. Phuong, P. J. Hulshof, H. H. Khoi, H. Verhoef, and J. G. Hautvast. 2007. The contribution of plant foods to the vitamin a supply of lactating women in Vietnam: A randomized controlled trial. *The American Journal of Clinical Nutrition* **85** (4):1112–1120. doi:10.1093/ajcn/85.4.1112.

Kim, Y. S., N. H. Kim, S. J. Yeom, S. W. Kim, and D. K. Oh. 2009. *In vitro* characterization of a recombinant blh protein from an uncultured marine bacterium as a β-Carotene 15,15′-dioxygenase. *Journal of Biological Chemistry* **284** (23):15781–15793. doi:10.1074/jbc.M109.002618.

Kimura, H., C. R. Young, A. Martinez, and E. F. Delong. 2011. Light-induced transcriptional responses associated with proteorhodopsin-enhanced growth in a marine flavobacterium. *The Isme Journal* **5** (10):1641–1651. doi:10.1038/ismej.2011.36.

Klassen, J. L. 2010. Phylogenetic and evolutionary patterns in microbial carotenoid biosynthesis are revealed by comparative genomics. *PloS One* **5** (6):e11257. doi:10.1371/journal.pone.0011257.

King, T. B., T. G. Ohman, and G. S. Smith. 1962. Evidence of rumeno reticular losses of vitamin a and carotene. *Journal of Animal Science* **21**:1002.

Kiser, P. D., M. Golczak, and K. Palczewski. 2014. Chemistry of the retinoid (Visual) cycle. *Chemical Reviews* **114** (1):194–232. doi:10.1021/cr400107q.

Kodavali, P. K., M. Dudkiewicz, S. Pikuła, and K. Pawłowski. 2014. Bioinformatics analysis of bacterial annexins-putative ancestral relatives of eukaryotic annexins. *PloS One* **9** (1):e85428. doi:10.1371/journal.pone.0085428.

Kwon, S. K., B. K. Kim, J. Y. Song, M. J. Kwak, C. H. Lee, J. H. Yoon, T. K. Oh, and J. F. Kim. 2013. Genomic makeup of the marine flavobacterium *nonlabens (Donghaeana) dokdonensis* and identification of a novel class of rhodopsins. *Genome Biology and Evolution* **5** (1):187–199. doi:10.1093/gbe/evs134.

Lampert, J. M., J. Holzschuh, S. Hessel, W. Driever, K. Vogt, and J. von Lintig. 2003. Provitamin a conversion to retinal via the β, β-carotene-15,15′-oxygenase (*bcox*) is essential for pattern formation and differentiation during zebrafish embryogenesis. *Development* **130** (10):2173–2186. doi:10.1242/dev.00437.

Lee, J. H., J. G. Choi, Y. S. Kim, K. R. Kim, S. W. Kim, and D. K. Oh. 2012. Enhancement of retinal production by supplementing the surfactant span 80 using metabolically engineered *Escherichia coli*. *Journal of Bioscience and Bioengineering* **113** (4):461–466. doi:10.1016/j.jbiosc.2011.11.020.

Leung, W. C., S. Hessel, C. Meplan, J. Flint, V. Oberhauser, F. Tourniaire, J. E. Hesketh, J. von Lintig, and G. Lietz. 2009. Two common single nucleotide polymorphisms in the gene encoding beta-carotene 15,15′-monoxygenase alter beta-carotene metabolism in female volunteers. *The FASEB Journal* **23** (4):1041–1053. doi:10.1096/fj.08-121962.

Li, J., J.-W. Li, Z. Feng, J. Wang, H. An, Y. Liu, Y. Wang, K. Wang, X. Zhang, Z. Miao, et al. 2016. Epigenetic switch driven by DNA inversions dictates phase variation in *streptococcus pneumonia*. *PLoS Pathogens* **12** (7):e1005762. doi:10.1371/journal.ppat.1005762.

Liang, M. H., J. Zhu, and J. G. Jiang. 2017. Carotenoids biosynthesis and cleavage related genes from bacteria to plants. *Critical Reviews in Food Science and Nutrition* **13**:1–20. https://doi.org/10.1080/10408398.2017.1322552. doi:10.1080/10408398.2017.1322552.

Linares, D. M., C. Gomez, E. Renes, J. M. Fresno, M. E. Tornadijo, R. P. Ross, and C. Stanton. 2017. Lactic acid bacteria and bifidobacteria with potential to design natural biofunctional health-promoting dairy foods. *Frontiers in Microbiology* **8**:846. doi:10.3389/fmicb.2017.00846.

Liu, H. X., Y. Hu, and Y. J. Wan. 2016. Microbiota and bile acid profiles in retinoic acid-primed mice that exhibit accelerated liver regeneration. *Oncotarget* **7** (2):1096–1106. doi:10.18632/oncotarget.6665.

Lozupone, C. A., J. I. Stombaugh, J. I. Gordon, J. K. Jansson, and R. Knight. 2012. Diversity, stability and resilience of the human gut microbiota. *Nature* **489** (7415):220–230. doi:10.1038/nature11550.

Lutnaes, B. F., A. Strand, S. K. Petursdottir, and S. Liaaen-Jensen. 2004. Carotenoids of thermophilic bacteria – *Rhodothermus marinus* from submarine icelandic hot springs. *Biochemical Systematics and Ecology* **32** (5):455–468. doi:10.1016/j.bse.2003.09.005.

Malys, M. K., L. Campbell, and N. Malys. 2015. Symbiotic and antibiotic interactions between gut commensal microbiota and host immune system. *Medicina (Kaunas)* **51** (2):69–75. doi:10.1016/j.medici.2015.03.001.

Marshall, J. H., and G. J. Wilmoth. 1981. Proposed pathway of triterpenoid carotenoid biosynthesis in *Staphylococcus aureus:* Evidence from a study of mutants. *Journal of Bacteriology* **147**:914–919.

Martinez, A., A. S. Bradley, J. R. Waldbauer, R. E. Summons, and E. F. DeLong. 2007. Proteorhodopsin photosystem gene expression enables photophosphorylation in a heterologous host. *Proceedings of the National Academy of Sciences of the United States of America* **104** (13):5590–5595. doi:10.1073/pnas.0611470104.

Mata, N. L., W. N. Moghrabi, J. S. Lee, T. V. Bui, R. A. Radu, J. Horwitz, and G. H. Travis. 2004. Rpe65 is a retinyl ester binding protein that presents insoluble substrate to the isomerase in retinal pigment epithelial cells. *Journal of Biological Chemistry* **279** (1):635–643. doi:10.1074/jbc.M310042200.

McGillivray, W. A. 1951. The apparent intestinal synthesis of carotene by sheep. *British Journal of Nutrition* **5** (02):223–228. doi:10.1079/BJN19510028.

Millard, A., D. J. Scanlan, C. Gallagher, A. Marsh, and P. C. Taylor. 2014. Unexpected evolutionary proximity of eukaryotic and cyanobacterial enzymes responsible for biosynthesis of retinoic acid and its oxidation. *Molecular Biosystems* **10** (3):380–383. doi:10.1039/c3mb70447e.

Miura, K. K., T. Saito, H. Shimada, P. D. Faraser, and N. Misawa. 1998. Production of the carotenoids lycopene, beta-carotene and astaxanthin in the food yeast *Candida utilis*. *Applied and Environmental Microbiology* **64**:1226–1229.

Moise, A. R., J. von Lintig, and K. Palczewski. 2005. Related enzymes solve evolutionarily recurrent problems in the metabolism of carotenoids. *Trends in Plant Science* **10** (4):178–186. doi:10.1016/j.tplants.2005.02.006.

Mongodin, E. F., K. E. Nelson, S. Daugherty, R. T. DeBoy, J. Wister, H. Khouri, J. Weidman, D. A. Walsh, R. T. Papke, G. Sanchez Perez, et al. 2005. The genome of *Salinibacter ruber*: Convergence and gene exchange among hyperhalophilic bacteria and archaea. *Proceedings of the National Academy of Sciences of the United States of America* **102** (50):18147–18152. doi:10.1073/pnas.0509073102.

Moran, N. A., and T. Jarvik. 2010. Lateral transfer of genes from fungi underlies carotenoid production in aphids. *Science* **328** (5978):624–627. doi:10.1126/science.1187113.

Newcomer, M. E., T. A. Jones, J. Aqvist, J. Sundelin, U. Eriksson, L. Rask, and P. A. Peterson. 1984. The three-dimensional structure of retinol-binding protein. *The EMBO Journal* **3** (7):1451–1454. doi:10.1002/j.1460-2075.1984.tb01995.x.

Ng, W. V., S. P. Kennedy, G. G. Mahairas, B. Berquist, M. Pan, H. D. Shukla, S. R. Lasky, N. S. Baliga, V. Thorsson, J. Sbrogna, et al. 2000. Genome sequence of *Halobacterium* species NRC-1. *Proceedings of the National Academy of Sciences of the United States of America* **97** (22):12176–12181. doi:10.1073/pnas.190337797.

Nupur, L., A. Vats, S. K. Dhanda, G. P. Raghava, A. K. Pinnaka, and A. Kumar. 2016. Procardb: A database of bacterial carotenoids. *BMC Microbiology* **16** (1):96. doi:10.1186/s12866-016-0715-6.

Ohland, C. L., and C. Jobin. 2015. Microbial activities and intestinal homeostasis: a delicate balance between health and disease. *Cellular and Molecular Gastroenterology and Hepatology* **1** (1):28–40. doi:10.1016/j.jcmgh.2014.11.004.

Olson, J. A., and O. Hayaishi. 1965. The enzymatic cleavage of beta-carotene into vitamin a by soluble enzymes of rat liver and intestine. *Proceedings of the National Academy of Sciences of the United States of America* **54**(5):1364–1370.

Olson, J. A. 1999. Carotenoids. In *Modern nutrition in health and disease*, ed. M. E. Shils, J. A. Olson, M. Shike, and A. C. Ross, 9th ed., 525–541. Baltimore: Williams and Wilkins.

Paik, J., A. During, E. H. Harrison, C. L. Mendelsohn, K. Lai, and W. S. Blaner. 2001. Expression and characterization of a murine enzyme able to cleave β-carotene. The formation of retinoids. *Journal of Biological Chemistry* **276** (34):32160–32168. doi:10.1074/jbc.M010086200.

Pandey, K. R., S. R. Naik, and B. V. Vakil. 2015. Probiotics, prebiotics and synbiotics- a review. *Journal of Food Science and Technology* **52** (12):7577–7587. doi:10.1007/s13197-015-1921-1.

Parker, G. L., L. K. Smith, and L. R. Baxendale. 2016. Ferrocene derivatives of liquid chiral molecules allow assignment of absolute configuration by X-ray crystallography. *Tetrahedron* **72** (13):1645–1652. doi:10.1016/j.tetasy.2017.09.002.

Peck, R. F., C. Echavarri-Erasun, E. A. Johnson, W. V. Ng, S. P. Kennedy, L. Hood, S. DasSarma, and M. P. Krebs. 2001. *Brp* and *blh* are required for synthesis of the retinal cofactor of bacteriorhodopsin in *Halobacterium salinarum*. *Journal of Biological Chemistry* **276** (8):5739–5744. doi:10.1074/jbc.M009492200.

Pelz, A., K. P. Wieland, K. Putzbach, P. Hentschel, K. Albert, and F. Gotz. 2005. Structure and biosynthesis of staphyloxanthin from *Staphylococcus aureus*. *Journal of Biological Chemistry* **280** (37):32493–32498. doi:10.1074/jbc.M505070200.

Penzes, P.,. X. Wang, and, and J. L. Napoli. 1997. Enzymatic characteristics of retinal dehydrogenase type I expressed in *Escherichia coli*. *Biochimica et Biophysica Acta* **1342** (2):175–181. doi:10.1016/S0167-4838(97)00102-7.

Perera, C. O., and G. M. Yen. 2007. Functional properties of carotenoids in human health. *International Journal of Food Properties* **10** (2):201–230. doi:10.1080/10942910601045271.

Perez Sirkin, D. I., A.-G. Lafont, N. Kamech, G. M. Somoza, P. G. Vissio, and S. Dufour. 2017. Conservation of Three-Dimensional Helix-Loop-Helix structure through the vertebrate lineage reopens the cold case of Gonadotropin-Releasing Hormone-Associated peptide. *Frontiers in Endocrinology* **8**:207. doi:10.3389/fendo.2017.00207.

Pinhassi, J., E. F. DeLong, O. Beja, J. M. González, and C. Pedrós-Alio. 2016. Marine bacterial and archaeal ion-pumping rhodopsins: Genetic diversity, physiology, and ecology. *Microbiology and Molecular Biology Reviews* **80** (4):929–954. doi:10.1128/MMBR.00003-16.

Pompei, A., L. Cordisco, A. Amaretti, S. Zanoni, S. Raimondi, D. Matteuzzi, and M. Rossi. 2007. Administration of folate-producing bifidobacteria enhances folate status in wistar rats. *The Journal of Nutrition* **137** (12):2742–2746. doi:10.1093/jn/137.12.2742.

Putter, K. M., N. van Deenen, K. Unland, D. Prüfer, and C. Schulze Gronover. 2017. Isoprenoid biosynthesis in dandelion latex is enhanced by the overexpression of three key enzymes involved in the mevalonate pathway. *BMC Plant Biology* **17**:88. doi:10.1186/s12870-017-1036-0.

Qin, J., R. Li, J. Raes, M. Arumugam, K. S. Burgdorf, C. Manichanh, T. Nielsen, N. Pons, F. Levenez, T. Yamada, et al. 2010. Human gut microbial gene catalogue established by metagenomic sequencing. *Nature* **464** (7285):59–65. doi:10.1038/nature08821.

Rastall, R. A., and G. R. Gibson. 2015. Recent developments in prebiotics to selectively impact beneficial microbes and promote intestinal health. *Current Opinion in Biotechnology* **32**:42–46. doi:10.1016/j.copbio.2014.11.002.

Reboul, E. 2013. Absorption of vitamin a and carotenoids by the enterocyte: Focus on transport proteins. *Nutrients* **5** (9):3563–3581. doi:10.3390/nu5093563.

Redlich, C. A., J. S. Chung, M. R. Cullen, W. S. Blaner, A. M. Van Bennekum, and L. Berglund. 1999. Effect of long-term beta-carotene and vitamin a on serum cholesterol and triglyceride levels among participants in the carotene and retinol efficacy trial (CARET). *Atherosclerosis* **143** (2):427–434. doi:10.1016/S0021-9150(98)00303-7.

Redlich, C. A., J. N. Grauer, A. M. Van Bennekum, S. L. Clever, R. B. Ponn, and W. S. Blaner. 1996. Characterization of carotenoid, vitamin A, and alpha-tocopheral levels in human lung tissue and pulmonary macrophages. *American Journal of Respiratory and Critical Care Medicine* **154** (5):1436–1443. doi:10.1164/ajrccm.154.5.8912761.

Redmond, T. M., S. Gentleman, T. Duncan, S. Yu, B. Wiggert, E. Gantt, and F. Cunningham. 2001. Identification, expression, and substrate specificity of a mammalian beta-carotene 15,15'-dioxygenase. *Journal of Biological Chemistry* **276** (9):6560–6565. doi:10.1074/jbc.M009030200.

Riedel, T., L. Gómez-Consarnau, J. Tomasch, M. Martin, M. Jarek, J. M. González, S. Spring, M. Rohlfs, T. Brinkhoff, H. Cypionka, et al. 2013. Genomics and physiology of a marine flavobacterium encoding a proteorhodopsin and a xanthorhodopsin-like protein. *PLoS One* **8**(3):e57487. doi:10.1371/journal.pone.0057487.

Roberfroid, M. B., F. Bornet, C. Bouley, and J. H. Cummings. 1995. Colonic microflora: nutrition and health. Summary and conclusions of an international life sciences institute (ILSI) [Europe] workshop held in Barcelona, Spain. *Nutrition Reviews* **53** (5):127–130. doi:10.1111/j.1753-4887.1995.tb01535.x.

Rohrle, F. T., A. P. Moloney, M. T. Osorio, G. Luciano, A. Priolo, P. Caplan, and F. J. Monahan. 2011. Carotenoid, colour and reflectance measurements in bovine adipose tissue to discriminate between beef from different feeding systems. *Meat Science* **88** (3):347–353. doi:10.1016/j.meatsci.2011.01.005.

Ruch, S., P. Beyer, H. Ernst, and S. Al-Babili. 2005. Retinal biosynthesis in eubacteria: *in vitro* characterization of a novel carotenoid oxygenase from *Synechocystis* sp. PCC6803. *Molecular Microbiology* **55** (4):1015–1024. doi:10.1111/j.1365-295804460.x.

Sabehi, G., A. Loy, K. H. Jung, R. Partha, J. L. Spudich, T. Isaacson, J. Hirschberg, M. Wagner, and O. Beja. 2005. New insights into metabolic properties of marine bacteria encoding proteorhodopsins. *PLoS Biology* **3** (8):e273. doi:10.1371/journal.pbio.0030273.

Saeed, A., M. Hoekstra, M. O. Hoeke, J. Heegsma, and K. N. Faber. 2017. The interrelationship between bile acid and vitamin a homeostasis. *Biochimica et Biophysica Acta* **1862** (5):496–512. doi:10.1016/j.bbalip.2017.01.007.

Schaub, P., F. Wüst, J. Koschmieder, Q. Yu, P. Virk, J. Tohme, and P. Beyer. 2017. Non-enzymatic β-carotene degradation in provitamin A-biofortified crop plants. *Journal of Agricultural and Food Chemistry* **65** (31):6588–6598. doi:10.1021/acs.jafc.7b01693.

Scherzinger, D., and S. Al-Babili. 2008. *In vitro* characterization of a carotenoid cleavage dioxygenase from *Nostoc* sp. PCC 7120 reveals a novel cleavage pattern, cytosolic localization and induction by high-light. *Molecular Microbiology* **69** (1):231–244. doi:10.1111/j.1365-2958.2008.06282.x.

Schultz, M. 2008. Clinical use of *E. coli* nissle 1917 in inflammatory bowel disease. *Inflamm. Bowel Dis* **14**(7):1012–1018. doi:10.1002/ibd.20377.

Shand, R. F., and M. C. Betlach. 1994. Bop gene cluster expression in bacteriorhodopsin-overproducing mutants of *Halobacterium halobium*. *Journal of Bacteriology* **176** (6):1655–1660. doi:10.1128/jb.176.6.1655-1660.1994.

Sleator, R. D. 2011. Phylogenetics. *Archives of Microbiology* **193** (4):235–239. doi:10.1007/s00203-011-0677-x.

Sleator, R. D. 2013. A beginner's guide to phylogenetics. *Microbial Ecology* **66** (1):1–4. doi:10.1007/s00248-013-0236-x.

Sobreira, T. J. P., F. Marletaz, M. Simoes-Costa, D. Schechtman, A. C. Pereira, F. Brunet, S. Sweeney, A. Pani, J. Aronowicz, C. J. Lowe., et al. 2011. Structural shifts of aldehyde dehydrogenase enzymes were instrumental for the early evolution of retinoid-dependent axial patterning in metazoans. *Proceedings of the National Academy of Sciences of the United States of America* **108** (1):226–231. doi:10.1073/pnas.1011223108.

Sommer, F., and F. Backhed. 2013. The gut microbiota – Masters of host development and physiology. *Nature Reviews Microbiology* **11** (4):227–238. doi:10.1038/nrmicro2974.

Swann, J. R., E. J. Want, F. M. Geier, K. Spagou, I. D. Wilson, J. E. Sidaway, J. K. Nicholson, and E. Holmes. 2011. Systemic gut microbial modulation of bile acid metabolism in host tissue compartments. *PNAS* **108** (Suppl_1):4523–4530. doi:10.1073/pnas.1006734107.

Tang, G. 2010. Bioconversion of dietary provitamin a carotenoids to vitamin a in humans. *The American Journal of Clinical Nutrition* **91** (5):1468S–1473S. doi:10.3945/ajcn.2010.28674G.

Tang, G., Y. Hu, S. Yin, Y. Wang, G. E. Dallal, M. A. Grusak, and R. M. Russell. 2012. Beta-carotene in golden rice is as good as beta-carotene in oil at providing vitamin a to children. *The American Journal of Clinical Nutrition* **96** (3):658–664. doi:10.3945/ajcn.111.030775.

Travis, J. M. J., and E. R. Travis. 2002. Mutator dynamics in fluctuating environments. *Proceedings of the Royal Society B: Biological Sciences* **269** (1491):591–597. doi:10.1098/rspb.2001.1902.

UNICEF. 2018. Coverage at a crossroads: New directions for vitamin A supplementation programmes. UNICEF New York headquarters, 2018. Accessed September 17, 2018. https://data.unicef.org/resources/vitamin-a-coverage/.

Uray, I. P., E. Dmitrovsky, and P. H. Brown. 2016. Retinoids and rexinoids in cancer prevention: from laboratory to clinic. *Seminars in Oncology* **43** (1):49–64. doi:10.1053/j.seminoncol.2015.09.002.

US Department of Agriculture (USDA). 2014. Agricultural Research Service, USDA Nutrient Database for Standard Reference. Accessed August 15, 2018. http://www.nal.usda.gov/fnic/foofcomp.

Van der Woude, M. W. 2011. Phase variation: How to create and coordinate population diversity. *Current Opinion in Microbiology* **14** (2):205–211. doi:10.1016/j.mib.2011.01.002.

Von Lintig, J., and K. Vogt. 2000. Molecular analysis of vitamin a formation: cloning and characterization of beta-carotene 15,15'-dioxygenases. *Journal of Biological Chemistry* **275** (16):11915–11920. doi:10.1006/abbi.2000.2096.

Vranova, E., D. Coman, and W. Gruissem. 2013. Network analysis of the MVA and MEP pathways for isoprenoid synthesis. *Annual Review of Plant Biology* **64**:665–700.

Wang, Z., T. J. O'Shaughnessy, C. M. Soto, A. M. Rahbar, K. L. Robertson, N. Lebedev, and G. J. Vora. 2012. Function and regulation of *Vibrio campbellii* proteorhodopsin: acquired phototrophy in a classical organoheterotroph. *PLoS One* **7** (6):e38749. doi:10.1371/journal.pone.0038749.

Wolf, G. 2009. The enzymatic cleavage of beta-carotene: still controversial. *Nutrition Reviews* **53** (5):134–137. doi:10.1111/j.1753-4887.1995.tb01537.x.

Wyss, A., G. Wirtz, W. Woggon, R. Brugger, M. Wyss, A. Friedlein, H. Bachmann, and W. Hunziker. 2000. Expression pattern and localization of beta, beta-carotene 15,15'-dioxygenase in different tissues. *Biochemical and Biophysical Research Communications* **271** (2):334–336.

Yan, W., G. F. Jang, F. Haeseleer, N. Esumi, J. Chang, M. Kerrigan, M. Campochiaro, P. Campochiaro, K. Palczewski, and D. J. Zack. 2001. Cloning and characterization of a human beta, beta-carotene-15,15'-dioxygenase that is highly expressed in the retinal pigment epithelium. *Genomics* **72** (2):193–202. doi:10.1006/geno.2000.6476.

Zhang, C., X. Chen, N. D. Lindley, and H. P. Too. 2018. A "plug-n-play" modular metabolic system for the production of apocarotenoids. *Biotechnology and Bioengineering* **115** (1):174–183. doi:10.1002/bit.26462.

Zwicker, B. L., and L. B. Agellon. 2013. Transport and biological activities of bile acids. *The International Journal of Biochemistry & Cell Biology* **45** (7):1389–1398. doi:10.1016/j.biocel.2013.04.012.