



Review

Advances in the biosynthesis of β -carotene and its derivatives in yeast

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HIGHLIGHTS

- The bioproduction of both xanthophyll and apocarotenoid derivatives in yeast is emphasized.
- Yeasts as producers of β -carotene derivatives by naturally or heterologously are highlighted.
- Key metabolic engineering and fermentation strategies that improved the production are summarized.

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ABSTRACT

β -Carotene and its derivatives have been gaining huge interest due to its applications as food supplements, nutraceuticals, pharmaceuticals, pigments, etc. Owing to their high values, sustainable microbial production has been a heated research topic. Traditional production methods, such as plant extraction and chemical synthesis, face challenges in scalability, cost, and environmental impact. With advances in synthetic biology, yeast-based biosynthesis has emerged as a promising alternative. This review provides a comprehensive summary of recent progress in the metabolic engineering strategies and fermentation optimization approaches of yeast, particularly *Saccharomyces cerevisiae* and *Yarrowia lipolytica*, for the production of β -carotene and its derivatives. In contrast to previous reviews, this work emphasizes the shared biosynthetic logic underlying structurally related derivatives, classifying them into two major groups: xanthophylls (canthaxanthin, zeaxanthin, astaxanthin, and violaxanthin) and apocarotenoids (crocin, retinol, β -ionone, β -cyclocitral, and strigolactones). Representative cases and transferable engineering/fermentation strategies are highlighted. Advantages and limitations of yeast species as production hosts are thoroughly compared, and potential strategies to improve the production are discussed. Future work may focus on broadening product diversity in different yeast hosts and enhancing biosynthetic efficiency for a more sustainable production.

1. Introduction

β -Carotene and its derivatives are a group of chemicals that can be synthesized in various plants and microorganisms (Rapoport et al., 2021). Based on their molecular structures, β -carotene and its derivatives exhibit remarkable diversity. Carotenoids containing oxygenated hydrocarbon (e.g., zeaxanthin, astaxanthin, canthaxanthin, and violaxanthin) are classified as xanthophylls (Fig. 1) (Gong and Bassi, 2016), while those produced through oxidative cleavage, like crocin,

β -ionone, β -cyclocitral, retinol, and strigolactones, are categorized into apocarotenoids (Fig. 1) (Saini and Keum, 2018).

In plants, they play an essential role in photosynthesis with an ability to absorb light and convert light into usable chemical energy (Hashimoto et al., 2016). In microorganisms, including bacteria, yeasts, and molds, they can protect cells from reactive oxygen species and radiation (Di Mascio et al., 1991). Except for their inherent functions in plants and microorganisms, β -carotene and its derivatives also contribute a lot to humans, considering their wide applications in food

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additives (Catarina et al., 2021; Gordon and Bauernfeind, 1982), nutraceuticals (Garcia et al., 2022; Mori et al., 2019), cosmetics ingredients (Lima et al., 2021; Tominaga et al., 2012), and medicines (Avraham et al., 2021; Lambrianidou et al., 2020; Moshiri et al., 2015).

Traditionally, carotenoids are manufactured by extraction from plants or chemical synthesis, which cannot meet the vast demand in the market. These processes also have shown the additional problems of the unstable raw plant materials supply (Bogacz-Radomska and Harasym, 2018) or the need to use toxic extraction agents that are not favorable for customers (Adadi et al., 2018). Moreover, large-scale agricultural cultivation or petrochemical-based synthesis can lead to high land and energy consumption, contributing to environmental degradation and increased production costs. In contrast, microbial synthesis offers a promising and sustainable alternative. Enabled by synthetic biology, this approach allows the controlled and scalable production of β -carotene and its derivatives in engineered microbial hosts. Compared to traditional methods, microbial biosynthesis significantly reduces reliance on natural plant resources, enables continuous and stable production in bioreactors, lowers operational and raw material costs, and minimizes environmental destruction, aligning with the principles of green chemistry and the global push toward a more sustainable bioeconomy (Loiseau et al., 2016). Among various microorganisms, yeasts like *Saccharomyces cerevisiae* and *Yarrowia lipolytica* are widely accepted as promising heterologous hosts for bioproduction due to their safety status (GRAS), excellent sustainability in manufacturing (Love et al., 2018), relatively clear genetic background (Sharma, 2016), abundant synthetic biology tools (Korpys-Woźniak et al., 2022), and so on. Several metabolic engineering strategies and fermentation technologies have been applied in yeast for the biosynthesis of β -carotene and its derivatives, but there is still no standardized and systematic regulation for their best production, which is a non-negligible bottleneck for their industrial manufacturing and commercialization.

Many recent reviews have explored the sources, synthesis, and functions of carotenoids (Jing et al., 2022; Wang et al., 2021; Zhang et al., 2022b). However, only a few have examined their microbial

bioproduction through the lens of metabolic engineering, particularly within specific chassis organisms. Among carotenoids, β -carotene has long been a heated topic (Guo et al., 2024). Yet, its structurally diverse derivatives, especially apocarotenoids, with great commercial and therapeutic potential, have received comparatively limited attention (Mai et al., 2025). To address these gaps, this review focuses specifically on the yeast-based biosynthesis of β -carotene and its derivatives. While most existing studies concentrate on engineering pathways for a single target compound (Wang et al., 2024; Zhang et al., 2020a; Zhang et al., 2018b), our review instead considers the shared structural features and common biosynthetic pathways among β -carotene derivatives. By doing so, we provide a systematic summary of metabolic and fermentation engineering strategies that have the potential to be broadly applicable and transferable across multiple similar products. This integrative review aims to guide future research toward more versatile, standardized, and scalable bioproduction platforms using yeast chassis.

2. Yeast as chassis for β -carotene and its derivatives

2.1. Limitations of traditional production methods and the rise of yeast biosynthesis

The traditional method of obtaining β -carotene and its derivatives relies on extraction from plant materials using physicochemical techniques. However, this approach has several major drawbacks, including high production costs, dependence on geographical factors (requiring extensive land and water resources) (Cravens et al., 2019), and the seasonality of raw material availability (Bogacz-Radomska and Harasym, 2018), thus causing insecurity and instability of supply chains. Since the 1950s, chemical synthesis has been developed and worked as the major source of carotenoid pigments (Bogacz-Radomska and Harasym, 2018). However, the synthesis of certain derivatives remains challenging due to their complex stereochemistry (Cravens et al., 2019; Serra, 2015). Furthermore, some studies have raised concerns about the potential carcinogenic activity of synthetic β -carotene (Black, 2004).

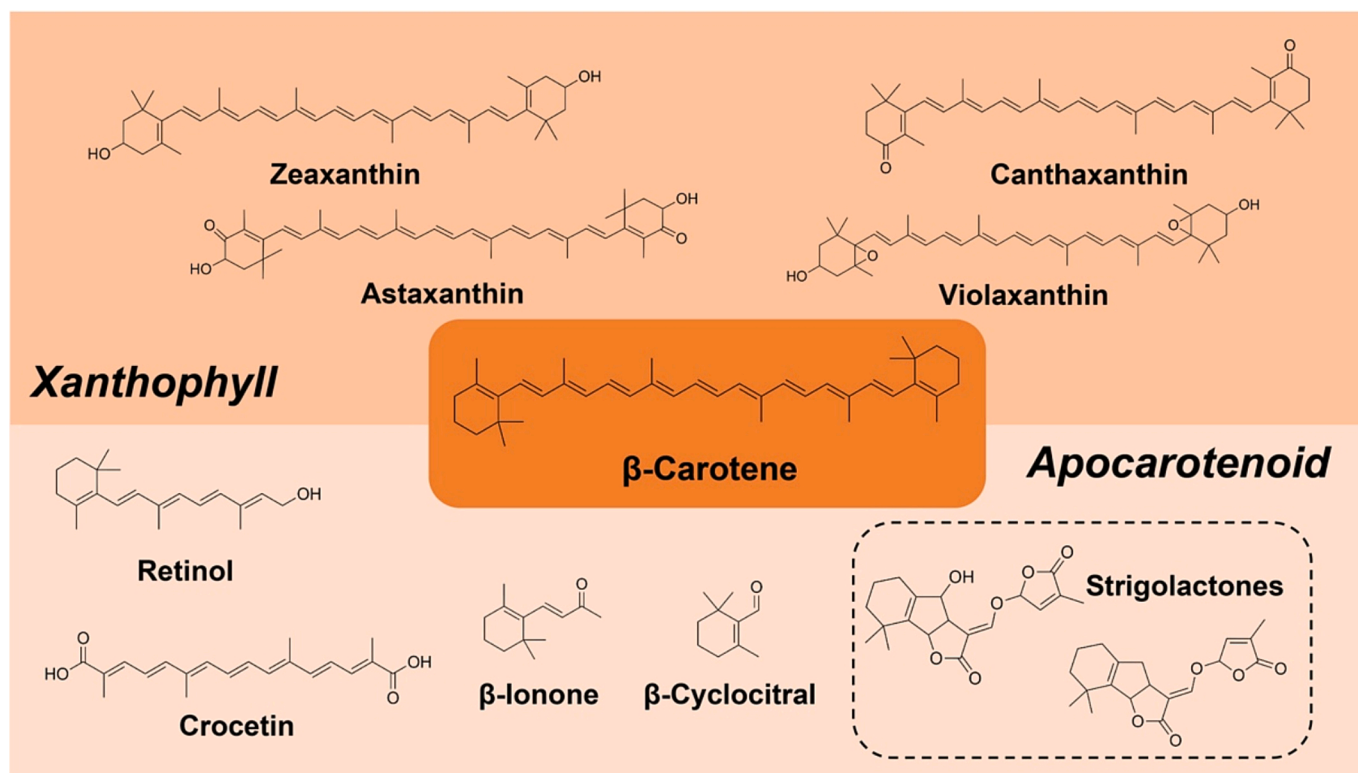


Fig. 1. Chemical structures of β -carotene and its typical derivatives.

These limitations associated with plant extraction and chemical synthesis have driven researchers to explore alternative methods for producing β -carotene and its derivatives. Among various approaches, microbial biosynthesis has garnered significant attention due to its ability to provide a scalable and controlled synthesis process via fermentation (Avraham et al., 2021; Cravens et al., 2019), eliminating concerns related to pests, extreme weather conditions, or hazardous chemical catalysts.

Various microorganisms have demonstrated significant potential as homologous or heterologous hosts for the production of β -carotene derivatives. Many microalgae have been reported as naturally accumulating strains (Gong and Bassi, 2016). For instance, *Dunaliella salina* is rich in zeaxanthin and β -carotene (Xu et al., 2018), and astaxanthin can be extracted from the biomass of *Haematococcus pluvialis* (Villaró et al., 2021). However, the natural accumulation of pigments in microalgae faces challenges such as low growth rates and difficulties in dewatering algae cultures for biomass production (Khan et al., 2018). Additionally, microalgae rely on photosynthesis, requiring controlled light intensity and CO₂ supply, making the large-scale cultivation technically demanding and energy-intensive. Certain bacteria species, including *Rhodococcus maris* and *Rhodobacter sphaeroides*, have also been reported to possess β -carotene-producing capabilities (Bogacz-Radomska and Harasym, 2018). Because of a lack of organelles or lipid bodies, their ability to synthesize and store large amounts of hydrophobic molecules like carotenoids is often restricted in bacteria. Apart from microalgae and bacteria, yeasts are also another natural source of these value-added pigments. A known example is the red yeast *Phaffia rhodozyma*, considered a valuable source of astaxanthin (Elwan et al., 2019).

Moreover, with advancements in synthetic biology, several model microorganisms have been genetically engineered to enhance the production of various β -carotene derivatives. For example, *Escherichia coli* has been successfully modified to biosynthesize lycopene (Liu et al., 2020), β -carotene (Wu et al., 2020), zeaxanthin (Li et al., 2015), astaxanthin (Zhang et al., 2022a), crocetin (Wang et al., 2019b) and canthaxanthin (Scaife et al., 2012). Nonetheless, considering the potential application of these chemicals in food and medicine after biosynthesis, immune recognition of endotoxin (a lipid component in the *E. coli* cell membrane) (Das et al., 2014) is an inevitable problem when using *E. coli* as the heterogenous manufacturing chassis (Wilding et al., 2019). Compared to *E. coli*, yeasts, especially those considered as GRAS ones, have emerged as robust host cells for the biosynthesis of β -carotene and its derivatives. Furthermore, yeasts exhibit superior capabilities in producing larger proteins, particularly those requiring complex post-translational modifications, a feature in which *E. coli* is limited (Demain and Vaishnav, 2009). Consequently, yeast-based biosynthesis for β -carotene derivative production has attracted growing research interest in recent years.

2.2. Limitations on native yeast producers

With the expanding global market for carotenoids and the growing trend of replacing traditional chemical synthesis with bioproduction, native carotenoid-producing microorganisms have attracted significant attention and have been increasingly utilized in industrial applications (Li et al., 2020a). Among various yeasts, *P. rhodozyma* (now reclassified as *Xanthophyllomyces dendrorhous*) is one of the best-known astaxanthin native producers. Several systematic reviews have been published focusing on the *X. dendrorhous*, highlighting its strong potential for commercial-scale production (Visser et al., 2003; Zhang et al., 2020a). In addition to *X. dendrorhous*, other species of the genera *Rhodospiridium*, *Rhodotorula*, *Sporobolomyces*, and *Sporidiobolus* commonly referred to as red yeasts are also recognized as natural carotenoid producers. The taxonomy and carotenoid biosynthesis potential of these four red yeast genera has been comprehensively analyzed and summarized by Mannazzu et al. (Mannazzu et al., 2015).

As research in this field continues to advance, new natural yeast

producers are being discovered. For instance, *Pichia fermentans*, a yeast strain capable of naturally synthesizing β -carotene and β -cryptoxanthin (an intermediate in the β -carotene-to-zeaxanthin conversion pathway), was isolated in southern Brazil (Otero et al., 2019). Additionally, newly identified red yeast genera such as *Buckleyzyma*, *Cystofilobasidium*, *Erythrobasidium* (Kot et al., 2023), and *Occultifur* (Promsai et al., 2024) have recently been isolated. Building upon these naturally occurring yeast producers, various strategies have been employed to enhance the biosynthesis of targeted carotenoids (Table 1).

Despite their natural capabilities, these native yeast strains exhibit several limitations. Most of them are restricted to the production of β -carotene or its oxygenated derivatives (xanthophylls), while the biosynthesis of structurally diverse apocarotenoids remains inaccessible. In addition, native yeasts often lack the genetic tools, industrial robustness, or metabolic flexibility required for further pathway expansion and optimization. Crucially, among yeast species, only two non-carotenoid-producing strains *S. cerevisiae* and *Y. lipolytica*, have a clear GRAS status (Kulczyk-Majsa and Bogusławska-Was, 2025), which means that the regulatory distinction may pose challenges for the commercial application of native producers in food and pharmaceutical industries. These constraints collectively highlight the necessity of developing heterologous biosynthesis platforms of β -carotene or its derivatives in GRAS-designated yeasts.

2.3. GRAS yeasts as preferred hosts for heterologous biosynthesis

Heterologous biosynthesis of β -carotene and its derivatives has already been successfully achieved in many yeast species. For example, the exogenous carotenoid biosynthetic genes originated from the epiphytic bacterium *Erwinia uredovora* and the marine bacterium *Agrobacterium aurantiacum* were introduced to *Candida utilis*, enabling it to synthesize lycopene, β -carotene, and astaxanthin (Miura et al., 1998). Similarly, the biosynthesis of β -carotene derivatives in *Pichia pastoris* has been primarily accomplished in strain X-33 through the sequential introduction of exogenous carotenoid biosynthetic genes (Bhataya et al., 2009; Veiga-Crespo et al., 2018). However, they face the same safety problem (lack of GRAS status) as native producers.

As GRAS-designated yeasts, *S. cerevisiae* and *Y. lipolytica* have emerged as the most prominent chassis for heterologous biosynthesis of β -carotene and its derivatives. As a well-characterized model organism, *S. cerevisiae* has been widely adopted due to its extensive synthetic biology toolkit (Chen et al., 2018), robust genetic background (Goffeau et al., 1996), and compatibility with plant-derived enzymes such as cytochrome P450s (Paramasivan and Mutturi, 2017)—key players in downstream carotenoid modification. Its tolerance to high sugar concentrations and ethanol production also provides competitive advantages in industrial-scale fermentation (Parapouli et al., 2020). To date, *S. cerevisiae* has been successfully engineered to produce a wide range of β -carotene derivatives (Table 2), and recent developments in universal terpenoid biosynthesis platforms have further enhanced its potential as a versatile production host.

In contrast, *Y. lipolytica*, although classified as a non-conventional yeast (Cui et al., 2017), is gaining momentum due to its oleaginous nature (Salvador López et al., 2022) and strong flux toward acetyl-CoA, an essential precursor for carotenoid biosynthesis (Pesantes-Munoz and Ledesma-Amaro, 2021). Its ability to store lipophilic compounds within lipid bodies (Larroude et al., 2018) makes it particularly suited for producing high levels of β -carotene and its derivatives. While *Y. lipolytica* does not naturally produce carotenoids, recent studies have demonstrated its remarkable capacity as an engineered host, achieving the highest reported β -carotene titer (39.5 g/L) (Ma et al., 2022) and enabling the production of other derivatives such as zeaxanthin, astaxanthin, crocetin, β -ionone, and retinol (Table 3). However, compared to *S. cerevisiae*, the full potential of *Y. lipolytica* for synthesizing more diverse β -carotene derivatives remains less explored.

Together, these two GRAS yeasts provide complementary strengths,

Table 1Production of β -carotene and its various derivatives in native yeast producers.

Strain	Compound	Biosynthetic strategies	Production	Reference
<i>Rhodospiridium</i> sp.	β -carotene	optimize extraction method	5.112 mg/L	(Kim et al., 2010)
<i>Rhodotorula glutinis</i>	β -carotene	supply vegetable oil	320 mg/L	(Yen et al., 2019)
<i>R. glutinis</i>	β -carotene	screen mutants	185 mg/L in 14-L bioreactor	(Bhosale and Gadre, 2001)
<i>Rhodotorula acheniorum</i>	β -carotene	supply additional yeast extract		
		mutate via UV, ethymethanesulfonate, and nitrosoguanidine	262.12 mg/L	(Nasrabadi and Razavi, 2011)
<i>Rhodotorula mucilaginosa</i>	β -carotene	use molasses as the substrate	621 μ g/L	(Cheng and Yang, 2016)
<i>R. mucilaginosa</i>	β -cryptoxanthin	optimize media	360 μ g/L (total carotenoids)	(Otero et al., 2019)
<i>Rhodotorula alborubescens</i>	β -carotene	isolate from sour cream	406.50 μ g/g biomass (total carotenoids)	(Kot et al., 2023)
<i>Rhodospiridium kratochvilovae</i>	β -carotene	optimize fermentation conditions	60–75% of total carotenoids (2.59 mg/L)	(Sereti et al., 2023)
<i>Rhodospiridium toruloides</i>	astaxanthin	optimize media by response surface methodology	927.11 μ g/L	(Tran et al., 2019)
<i>Sporidiobolus pararoseus</i>	β -carotene and β -cryptoxanthin	optimize media	905.3 μ g/L (total carotenoids)	(Otero et al., 2019)
<i>S. pararoseus</i>	β -carotene	provide demethanolized crude glycerol	54.43 mg/L	(Chaiyaso and Manowattana, 2018)
<i>P. fermentans</i>	β -carotene and β -cryptoxanthin	add olive oil	500 μ g/L (total carotenoids)	(Otero et al., 2019)
<i>Buckleyzyma aurantiaca</i>	β -carotene	optimize media		
		isolate from air	255.79 μ g/g biomass (total carotenoids)	(Kot et al., 2023)
<i>Cystofilobasidium macerans</i>	β -carotene	isolate from wild strawberry fruit	224.59 μ g/g biomass (total carotenoids)	(Kot et al., 2023)
<i>Erythrobasidium hasegawianum</i>	β -carotene	isolate from air, street	234.72 μ g/g biomass (total carotenoids)	(Kot et al., 2023)
<i>Oocultifur</i> sp.	β -carotene	optimize fermentation conditions	Not mentioned	(Promsai et al., 2024)
<i>X. dendrorhous</i>	astaxanthin	combine classical mutation and biosynthetic gene overexpression	9 mg/g DCW	(Gassel et al., 2014)
<i>X. dendrorhous</i>	astaxanthin	delete diploid CYP61 genes to decrease ergosterol biosynthesis	1.6 mg/L	(Yamamoto et al., 2016)
<i>X. dendrorhous</i>	astaxanthin	feed glutamate to upregulate biosynthetic genes	1.14 mg/g DCW	(Wang et al., 2019a)
<i>X. dendrorhous</i>	astaxanthin	lower pH	0.7 g/kg cultural broth	(Schewe et al., 2017)

S. cerevisiae offers genetic tractability and pathway flexibility, while *Y. lipolytica* provides a favorable metabolic background for lipid-associated product accumulation, forming a solid foundation for future metabolic engineering of β -carotene derivatives.

In addition to the two most extensively studied yeast chassis for heterologous engineering, the biosynthesis of β -carotene and its derivatives has also been successfully achieved in other yeast species. For example, the exogenous carotenoid biosynthetic genes originated from the epiphytic bacterium *Erwinia uredovora* and the marine bacterium *Agrobacterium aurantiacum* were introduced to *Candida utilis*, enabling it to synthesize lycopene, β -carotene, and astaxanthin (Miura et al., 1998). Similarly, the biosynthesis of β -carotene derivatives in *Pichia pastoris* has been primarily accomplished in strain X-33 through the sequential introduction of exogenous carotenoid biosynthetic genes (Bhataya et al., 2009; Veiga-Crespo et al., 2018).

3. Biosynthetic pathways of β -carotene and its derivatives in yeast

β -Carotene and its derivatives are widely synthesized in various plant cells through common isoprene (C5) precursor isopentenyl diphosphate (IPP) and its isomer, dimethylallyl diphosphate (DMAPP) (Fig. 2) (Yang et al., 2016; Zhang et al., 2022b). In higher plants, β -carotene and its derivatives can be synthesized via both the mevalonate (MVA) pathway and methylerythritol phosphate (MEP) pathway (Yang et al., 2016). Some marine algae are also equipped with both pathways (Chandran et al., 2011). The MEP pathway in most gram-negative bacteria, green algae, and cyanobacteria can be used in the biosynthesis of β -carotene forming units (Ren et al., 2020).

In yeasts, the MVA pathway has long been recognized as the sole route for synthesizing terpenoids, including β -carotene and its derivatives. However, a novel perspective was introduced by Dissook et al. in 2021 suggesting the potential presence of an MEP pathway in the oleaginous yeast *Y. lipolytica*. (Dissook et al., 2021). Their study

proposed a possible co-existence of both pathways under nitrogen-limiting conditions, challenging the conventional understanding of isoprenoid biosynthesis in yeast. While this finding awaits further validation, it presents an intriguing avenue for future research into alternative metabolic routes under specific environmental conditions.

In this section, we will focus on the widely accepted MVA pathway in yeast. The complete biosynthetic pathway of β -carotene and its derivatives via the MVA pathway can be systematically divided into upstream and downstream modules based on the distinct subcellular localization of enzymatic reactions, as illustrated in Fig. 2 (Paniagua-Michel et al., 2012).

3.1. Upstream MVA pathway

The upstream module of the MVA pathway consists of six enzymatic steps (Miziorko, 2011), in which acetyl-CoA is converted to universal precursors IPP and DMAPP in the cytosol. Firstly, three molecules of acetyl-CoA are needed in the beginning two steps of the MVA pathway. The enzymes ERG10 (acetoacetyl-CoA thiolase) and ERG13 (also known as HMGS, hydroxymethylglutaryl-CoA synthase) catalyze the condensation reactions to form hydroxymethylglutaryl-CoA (HMG-CoA) (Ferguson and Rudney, 1959). Secondly, HMG-CoA is reduced by HMGR (hydroxy-3-methyl glutaryl coenzyme A reductase) (Durr and Rudney, 1960). Then, ERG12 (mevalonate kinase) and ERG8 (phosphomevalonate kinase) catalyze the phosphorylation reaction to form mevalonate-5-phosphate (MVA-5-P) and mevalonate diphosphate (MVA-PP) via MVA, sequentially (Hellig and Popják, 1961; Tchen, 1958). An ATP-dependent decarboxylation reaction is the last step before IPP (Bloch et al., 1959), and MVD1 (also known as ERG19) acts as the diphosphomevalonate decarboxylase in yeast. IPP can then be isomerized into DMAPP by isopentenyl diphosphate isomerase (IDI), providing the key precursors for downstream biosynthesis in yeast.

Several genes within the MVA pathway, including *ERG12*, *ERG8*, and *IDI*, have been identified as potential rate-limiting factors (Li et al.,

Table 2
Production of β -carotene and its various derivatives in *S. cerevisiae*.

Type	Compound	Biosynthetic strategies	Production	Reference
β -carotene	β -carotene	integrate multicopy of biosynthetic genes overexpress <i>tHMG</i>	5.9 mg/g DCW	(Verwaal et al., 2007)
	β -carotene	overexpress sterol ester synthesis genes <i>ARE1</i> and <i>ARE2</i> delete phosphatidate phosphatase genes	8.98 mg/g DCW	(Zhao et al., 2021)
	β -carotene	integrate a xylose utilizing pathway	772.8 mg/L in 3-L bioreactor	(Sun et al., 2020)
	β -carotene	site-specific mutation of <i>crtI</i> and <i>crtYB</i> evaluate HMG-CoA reductase from different origins	390 μ g/g DCW	(Li et al., 2013)
	β -carotene	express lipases from <i>Y. lipolytica</i> enabling hydrophobic substrates usage	477.9 mg/L (46.5 mg/g DCW)	(Fathi et al., 2021)
	β -carotene	regulate lipid droplet-triacylglycerol metabolism down-regulate <i>ERG9</i> expression	142 mg/L (11.4 mg/g DCW)	(Bu et al., 2022)
xanthophyll	zeaxanthin	screen promoters integrate a xylose utilizing pathway	0.74 mg/L	(Sun et al., 2012)
	zeaxanthin	screen <i>CrtZ</i> from different origins adjust copy number	96.2 mg/L	(Mei et al., 2016)
	zeaxanthin	screen and identify <i>CrtW</i> and <i>CrtZ</i> enzymatic variants	70.5 μ g/L	(Promdonkoy et al., 2024)
	astaxanthin	overexpress GGPP synthase	29 μ g/g DCW	(Ukibe et al., 2009)
	astaxanthin	adjust copy number supply iron cofactor	4.7 mg/g DCW	(Zhou et al., 2015)
	astaxanthin	atmospheric and room temperature plasma (ARTP)-driven gene mining and overexpression	217.9 mg/L (13.8 mg/g DCW) in 5-L bioreactor	(Jin et al., 2018)
	astaxanthin	engineer <i>CrtZ</i> and <i>CrtW</i>	81.0 mg/L in 5-L bioreactor	(Wang et al., 2017a)
	astaxanthin	fuse <i>crtZ</i> and <i>crtW</i> error-prone PCR based mutant library	1.87 mg/g DCW	(Ding et al., 2023)
	astaxanthin	<i>N</i> -terminal truncation of β -carotene hydroxylase modeling-based mutations of β -carotene hydroxylase	26.0 mg/L 3.6 mg/g DCW	(Du et al., 2025)
	canthaxanthin	subcellular re-localization of β -carotene ketolase variant adjust the copy number of β -carotene ketolase variant	1.44 g/L in 5-L bioreactor	(Chen et al., 2022a)
	canthaxanthin	overexpress <i>Pdr1</i> and <i>Pdr3</i> to improve stress tolerance screen and identify <i>CrtW</i> and <i>CrtZ</i> enzymatic variants	60.36 mg/L in flask, 5-L bioreactor	(Promdonkoy et al., 2024)
	canthaxanthin	delete the <i>GAL80</i> gene sucrose fermentation		
apocarotenoid	violaxanthin	screen several <i>CrtZ</i> and <i>ZEP</i> genes from different origins express fused <i>CrtZ-ZEP</i> <i>N</i> -terminal truncation of zeaxanthin epoxidase co-express redox partner <i>RFNRI/FD3</i> increase copy number of carotenogenic genes	7.3 mg/g DCW	(Cataldo et al., 2020)
	crocetin	screen <i>CrtZ</i> and <i>CCD</i> from different origins adjust copy number	1219 μ g/L in flask, 6278 μ g/L in 5-L bioreactor	(Chai et al., 2017)
	crocetin	block the genes involved in glyoxylate cycle introduce fusion enzymes	1.95 mg/L in flask, 12.43 mg/L in 5-L bioreactor	(Song et al., 2020)
	crocetin	CsCCD2 access tunnel design (S323A mutation)	20 mg/L in flask, 107 mg/L in 5-L bioreactor	(Liang et al., 2021)
	β -ionone	overexpress <i>tHMG</i> and <i>ERG20</i> downregulate <i>ERG9</i> delete <i>LPP1</i> and <i>DPP1</i> multiple copies of <i>crtYB</i> extra copy of <i>BTS1</i>	0.63 mg/g DCW in flask, 1 mg/g DCW in 2.5-L bioreactor	(López et al., 2015)
	β -ionone	multiple copies of <i>CCD1</i> subcellular localization of <i>CCD1</i> by a membrane destination peptide	33 mg/L	(López et al., 2020)
	β -ionone	<i>PhCCD1</i> K164L mutations subcellular localization by membrane-anchoring peptides	184 mg/L	(Werner et al., 2019)
	strigolactones	<i>S. cerevisiae</i> - <i>E. coli</i> coculture <i>E. coli</i> producer to provide carlactone precursor	47.3 μ g/L 5-deoxystrigol, 3.46 μ g/L 4-deoxyorobanchol, 19.36 μ g/L orobanchol	(Wu et al., 2021)
	retinol	introduce xylose utilization pathway BCMO to convert β -carotene to retinol	1256 mg/L in 3-L bioreactor	(Sun et al., 2019)
	retinol	select heterologous <i>RDHs</i> from different origins introduce xylose utilization pathway express <i>noxE</i> to avoid excess intracellular NADH	123.1 mg/L	(Lee et al., 2022)
	retinol	mine β -carotene 15,15'-mono(di)oxygenase from different sources overexpress <i>POS5Δ17</i>	588 mg/L in flask, 5.21 g/L in 1-L bioreactor	(Shi et al., 2025)
	β -cyclocitral	integrate multi-copy of the key rate-limiting step gene <i>Mbblh</i> identify novel CCDs from <i>Vitis vinifera</i> L cv. Pinotage	About 0.03 μ g/g DCW	(Meng et al., 2019)

2020b). These bottlenecks present promising targets for genetic engineering strategies aimed at optimizing the metabolic flux and enhancing the supply of IPP and DMAPP precursors for improved terpenoid biosynthesis.

3.2. Downstream biosynthetic pathway of β -carotene and its derivatives

In the downstream module, IPP and DMAPP undergo sequential condensation to form β -carotene through the introduction of key carotenoid biosynthetic genes (Veiga-Crespo et al., 2018). This process begins with *CrtE*, which encodes geranylgeranyl pyrophosphate (GGPP)

Table 3
Production of β -carotene and its various derivatives in *Y. lipolytica*.

Type	Compound	Biosynthetic strategies	Production	Reference
β -carotene	β -carotene	screen the optimum promoter-gene pairs overexpress <i>HMG1</i>	1.5 g/L in flask, 6.5 g/L (90 mg/g DCW) in 5-L bioreactor	(Larroude et al., 2018)
	β -carotene	copy number adjustment overexpress MVA genes (<i>HMG1</i> and <i>ERG13</i>)	4.5 g/L in 5-L bioreactor	(Zhang et al., 2020b)
	β -carotene	overexpress biosynthetic genes overexpress the native diacylglycerol O-acyltransferase (<i>DGA1</i>) overexpress <i>HMG</i> , <i>ERG12</i> , and <i>IDI</i>	164 mg/L	(Arnesen et al., 2020)
	β -carotene	overexpress <i>HXX</i> to accelerate glucose consumption overexpress <i>ERG13</i>	2.4 g/L in 50-L bioreactor	(Qiang et al., 2020)
	β -carotene	overexpress <i>tHMG</i> and <i>CarRP</i> downregulate <i>ERG9</i>	4 g/L in 2-L bioreactor	(Gao et al., 2017)
	β -carotene	multicopy of <i>ERG10</i> , <i>ERG13</i> , <i>ERG19</i> , <i>ERG12</i> , <i>ERG8</i> , <i>IDI</i> and <i>ERG20</i> source the relevant biosynthetic genes point mutation of lycopene cyclase to increase selectivity overexpress <i>tHMG</i> , <i>ERG12</i> , <i>IDI</i> and <i>ERG20</i> introduce isopentenol utilization pathway high copy numbers of <i>GGPPsa</i> , <i>CarB</i> , and <i>CarRP</i>	7.5 g/L (360.8 mg/g DCW) in flask, 39.5 g/L (494 mg/g DCW) in 3-L bioreactor	(Ma et al., 2022)
	β -carotene			
Xanthophyll	zeaxanthin	compare three <i>CrtZ</i> from different organismshigh-copy-number integration	21.98 mg/L in flask, 3.20 mg/g DCW	(Xie et al., 2021)
	zeaxanthin	modular assembly of <i>IDI</i> & <i>GGS1</i> and <i>ERG20Y</i> and <i>GGS1</i> to improve β -carotene production enhance MVA synthesis transfer RFNR1 to generate more reduced-Fd cofactors	775.3 mg/L	(Zhang et al., 2023)
	zeaxanthin	screen suitable <i>Y. lipolytica</i> strains hybrid promoter copy number adjustment	1575.09 mg/L	(Zhou et al., 2025)
	astaxanthin	overexpress <i>Hmg1</i> , <i>Erg12</i> and <i>Erg20</i> screen biosynthetic genes from different origins increase copy number fusion of key biosynthetic enzymes	587.3 mg/L in 3-L bioreactor, 812.3 mg/L in 100-L bioreactor	(Yuzbasheva et al., 2023)
	astaxanthin	fuse <i>CrtZ</i> and <i>CrtW</i> target fusion enzyme to subcellular organelles	139.4 mg/L in flask, 858 mg/L (16.7 mg/g DCW) in 3-L bioreactor	(Ma et al., 2021)
	astaxanthin	compare genes from different origins copy number adjustment	44 mg/L in flask, 285 mg/L in 1-L bioreactor	(Tramontin et al., 2019)
	astaxanthin	overexpress <i>HMG</i> -CoA reductase and <i>GGPP</i> synthase downregulate squalene synthase fuse <i>CrtZ</i> and <i>CrtW</i> copy number optimization of fused enzyme <i>CrtZ</i> - <i>CrtW</i>	54.6 mg/L (3.5 mg/g DCW) in microtiter plate	(Kildegaard et al., 2017)
Apocarotenoid	crocin	screen suitable <i>Y. lipolytica</i> strains hybrid promoter copy number adjustment two-step temperature-shift fermentation	30.17 mg/L	(Zhou et al., 2025)
	β -ionone	introduce <i>NphT7</i> from <i>Streptomyces</i> sp. and <i>IPI</i> from <i>Haemotoccus pluvialis</i> overexpress native MVA genes	380 mg/L in 2-L bioreactor	(Czajka et al., 2018)
	β -ionone	strong constitutive promoters overexpress <i>tHMG</i> , <i>GGS1</i> , <i>ERG10</i> , <i>ERG13</i> , <i>ERG8</i> , <i>ERG12</i> , <i>IDI</i> and <i>ERG20</i> introduce PK-PTA pathway to enhance cytosolic acetyl-CoA supply	358 mg/L in flask, 1 g/L in 3-L bioreactor	(Lu et al., 2020)
	β -ionone	PhCCD1 K164L mutation	4 g/L	(Chen et al., 2022b)
	retinol	overexpress <i>HMG1</i> , <i>crtYB</i> , <i>crtI</i> , <i>GGPPS1</i> , <i>ERG10</i> , <i>ERG13</i> , <i>GGS1</i> and <i>FAD1</i> <i>GGS1</i> F43I mutation delete <i>DGA1</i> , <i>SLC1</i> , <i>OPI3</i> , <i>POX4-6</i> , <i>DOS2</i> , <i>ERG5</i> , <i>EXG1</i> and <i>CRF1</i> select <i>BCO</i> from different sources multicopy of <i>BCO</i> genes	437 mg/L (0.0175 g/g DCW) in flask, 4.86 g/L in 5-L bioreactor	(Park et al., 2022)

synthase, catalyzing the formation of GGPP from IPP and DMAPP. Subsequently, *CrtB*, encoding phytoene synthase, catalyzes the head-to-head condensation of two GGPP molecules to form phytoene, marking the first committed step in carotenoid biosynthesis. *CrtI*, encoding phytoene desaturase, then introduces multiple conjugated double bonds into phytoene, converting it into lycopene, a key intermediate. Finally, *CrtY*, encoding lycopene β -cyclase, cyclizes both ends of the linear lycopene molecule to produce β -carotene, a central compound in the carotenoid pathway.

In addition to these discrete mono-functional enzymes, several bifunctional or multifunctional enzymes have been identified in different organisms, enhancing the efficiency and compactness of the pathway. For instance, *CarRP*, found in filamentous fungi such as *Mucor*

circinelloides (Velayos et al., 2000), and *CrtYB*, identified in *X. dendrorhous* (Verdoes et al., 1999), both possess dual enzymatic activities: phytoene synthase (*CrtB* activity) and lycopene cyclase (*CrtY* activity). These proteins can thus catalyze the conversion from GGPP to phytoene and from lycopene to β -carotene, potentially increasing catalytic efficiency and reducing metabolic burden in heterologous hosts. Moreover, a putative trifunctional β -carotene synthase, named *CrtIBY*, was identified in the genome of *Anomurantiocytrium* sp. KH105 (Iwasaka et al., 2018), and is proposed to catalyze three sequential steps: phytoene synthesis, desaturation, and lycopene cyclization. Since β -carotene serves as the universal precursor for a wide range of downstream carotenoids, the construction of a high-yield β -carotene-producing chassis strain is of central importance. Such a strain can serve as a

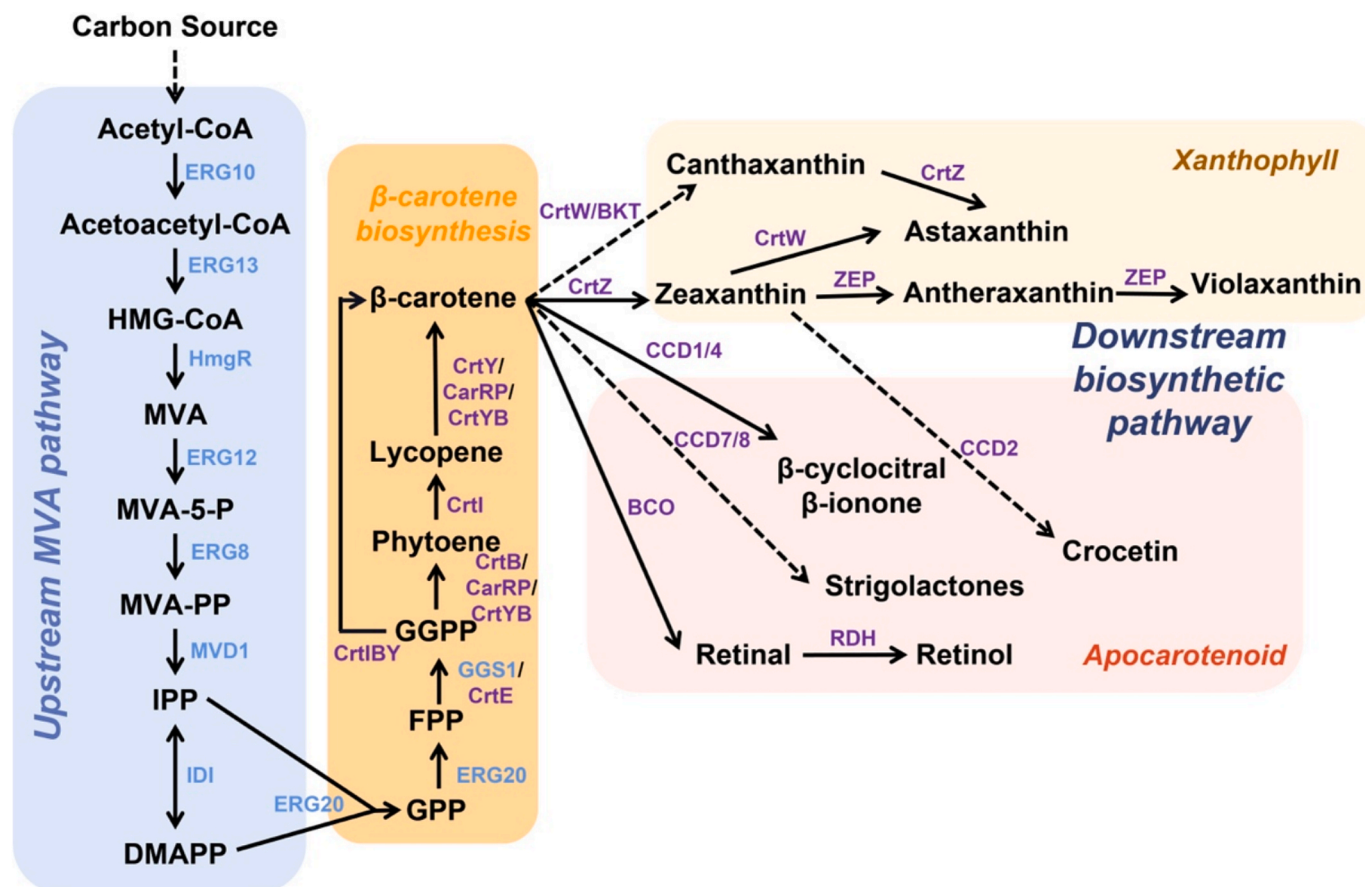


Fig. 2. Biosynthetic pathway of β -carotene and its derivatives in yeast. Yeast endogenous genes are labeled in blue, whereas heterologous ones are in purple; dotted arrows indicate multi-step reactions.

robust and flexible platform for the efficient production of various value-added carotenoid derivatives through pathway extension and metabolic engineering (Lin et al., 2025).

To generate specific β -carotene derivatives, additional pathway-specific enzymes are introduced to extend the core carotenoid biosynthetic pathway toward the production of high-value compounds with distinct structures and functions. Each derivative requires specific catalytic steps mediated by dedicated enzymes: CrtZ (encoding β -carotene hydroxylase) catalyzes the stepwise hydroxylation of β -carotene at the β -ionone rings, producing β -cryptoxanthin and subsequently zeaxanthin; CrtW or BKT (encoding β -carotene ketolase) introduces keto groups at the β -ionone rings of β -carotene or zeaxanthin, leading to the formation of canthaxanthin and astaxanthin; ZEP (encoding zeaxanthin epoxidase) catalyzes the epoxidation of zeaxanthin to violaxanthin via antheraxanthin as an intermediate; CCD (carotenoid cleavage dioxygenase) represents a diverse family of enzymes capable of cleaving carotenoids at specific double bonds to yield apocarotenoids. For instance, CCD1 cleaves β -carotene to form volatile compounds, CCD2 cleaves zeaxanthin to saffron compounds, while CCD7 and CCD8 are involved in plant hormone strigolactones' biosynthesis; BCO (β -carotene monooxygenase, particularly BCO1 in mammals) cleaves β -carotene symmetrically at the central 15, 15'-double bond to produce two molecules of retinal, which is further reduced to retinol by RDH (retinol dehydrogenase).

4. Metabolic engineering strategies to produce β -carotene and its derivatives

Biosynthesis of β -carotene and its derivatives in yeast has attracted significant interest, especially in two well-characterized and promising

platforms, *S. cerevisiae* and *Y. lipolytica*. However, the efficient production of xanthophylls and apocarotenoids in the engineered yeasts remains challenging due to multiple intrinsic and extrinsic metabolic bottlenecks as follows:

Low enzyme activity or substrate specificity: Enzymes such as CrtZ and CrtW often exhibit suboptimal catalytic efficiency and broad substrate specificity, leading to incomplete conversion of intermediates and low final product yields (Ma et al., 2021). Most CCDs are also reported to have a poor conversion rate from carotenoids to apocarotenoids in heterologous hosts (Chai et al., 2017; Chen et al., 2022b; Meng et al., 2019; Werner et al., 2019; Zhou et al., 2025), further hindering yield.

Limited substrate accessibility: Due to their hydrophobic nature, most carotenoids preferentially localize to cellular membranes or lipid bodies (Larroude et al., 2018). This membrane-bound sequestration can reduce the availability of intermediates to downstream enzymes (Ma et al., 2021), thereby limiting pathway throughput and product conversion efficiency.

Metabolic competition and flux imbalance: Multiple pathway branches compete for common intermediates such as FPP or β -carotene, leading to unbalanced metabolic flux distribution. Without precise regulation of lipid metabolism (Bu et al., 2022), this competition can divert resources away from the target product pathway, decreasing overall biosynthetic efficiency.

Cofactor limitations: Several enzymes, like CrtY (Hornero-Méndez and Britton, 2002), depend on redox cofactors such as NAD(P)H for bioactivity. However, microbial hosts often have limited intracellular pools of these cofactors. The insufficient cofactor supply in microorganisms can limit the overall reaction rate.

Product toxicity and accumulation: Most carotenoids accumulate

intracellularly because of their membrane-impermeable properties (Son et al., 2022). While intracellular accumulation is essential for storage and product recovery, excessive buildup can pose challenges, including membrane disruption and overall cytotoxicity, ultimately impairing cell growth and productivity.

Substrate feedback inhibition: Overaccumulation of intermediates can trigger feedback inhibition on upstream enzymes, thereby reducing pathway flux. A notable example is the bifunctional enzyme CarRP, which exhibits reduced activity in the presence of elevated levels of lycopene, one of its downstream products (Ma et al., 2022). The product-mediated inhibition can create a regulatory bottleneck, leading to precursor accumulation and compromised pathway efficiency.

Given these multifaceted challenges, it is crucial to implement systematic and rational metabolic engineering strategies to overcome key bottlenecks. Moreover, due to structural similarities and shared biosynthetic precursors among these β -carotene derivatives, strategies developed for enhancing the production of one derivative may be transferable to others. In this section, we aim to summarize a range of effective engineering approaches and provide insights into selecting appropriate strategies tailored to the biosynthesis of specific carotenoids.

4.1. Selection of target genes from different origins

The origin of biosynthetic genes plays a crucial role in the production efficiency of β -carotene and its derivatives in yeast. Selecting suitable heterologous genes is the first and most critical step before further metabolic engineering, as it significantly influences pathway efficiency (Fig. 3A).

From IPP and DMAPP precursors to β -carotene, all different derivatives share the same synthetic pathway, including *GGSI*, *CrtB*, and *CrtI*. GGPP synthase (*GGSI*), also referred to as *BTS1*, is endogenous to yeast (Dai et al., 2012; Larroude et al., 2018; Verwaal et al., 2007; Zhang et al., 2019a). Its overexpression has been shown to enhance carotenoid production, making it a common target for upregulation alongside other heterologous genes (Gao et al., 2017; Xie et al., 2021). Frequently, *GGSI* overexpression is combined with the heterologous expression of *CrtE* (encodes an enzyme with the same function as *GGSI*) for further enhanced production (Jing et al., 2022; Verwaal et al., 2007; Zhang et al., 2019a). Among various species, *X. dendrorhous* is a popular origin of *CrtB* and *CrtI* (Ukibe et al., 2009; Verwaal et al., 2007), and they have been described as being sufficient to enable carotenoid production in yeast (Verwaal et al., 2007). Other species, such as *Pantoea agglomerans* (Zhang et al., 2020b) and *M. circinelloides* (Zhang et al., 2019b), have also been explored as alternative sources for these genes.

For subsequent heterogeneous genes, *CrtZ* and *CrtW*, bacteria and algae-derived genes were evaluated (Tramontin et al., 2019). After comparing 9 different *CrtZ*s from *Agrobacterium aurantiacum*, *Alcaligenes* sp. PC-1, *Erwinia uredovora*, *Pantoea agglomerans*, *Pantoea stewartii*, *Sulfolobus solfataricus*, *Brevundimonas* sp. SD212, *Brevundimonas vesicularis* DC263, and *Haematococcus pluvialis*, respectively, Chai et al. found the highest zeaxanthin production from the expression of *CrtZ* from *E. uredovora* in *S. cerevisiae* (Chai et al., 2017). This conclusion was further supported by Ukibe's (Ukibe et al., 2009) and Xie's (Xie et al., 2021) research, indicating that *E. uredovora* could be the most suitable source of *CrtZ*, up to now. Through combinational expression of *CrtZ* and *CrtW* from distinguished sources, a 3.1 mg/g DCW of astaxanthin yield is reached with *CrtW* from *B. vesicularis* DC263 and *CrtZ* from *Alcaligenes* sp. strain PC-1 in *S. cerevisiae* (Wang et al., 2017a). Meanwhile, *CrtZ* and *CrtW* from algae were introduced to *S. cerevisiae* and reached a 4.7 mg/g DCW astaxanthin production (Zhou et al., 2015), also showing a great potential for algae-derived genes. Furthermore, plant-derived genes have been evaluated for β -carotene derivative biosynthesis in yeast (Wang et al., 2020; Zhou et al., 2019). Notably, *CsCCD2* from *Crocus* in crocetin biosynthesis was successfully expressed in *S. cerevisiae* (Chai et al., 2017) and *Y. lipolytica* (Zhou et al., 2025), leading to the

heterologous production of crocetin.

4.2. Modulation of the gene expression level

4.2.1. Overexpression of the desired pathway

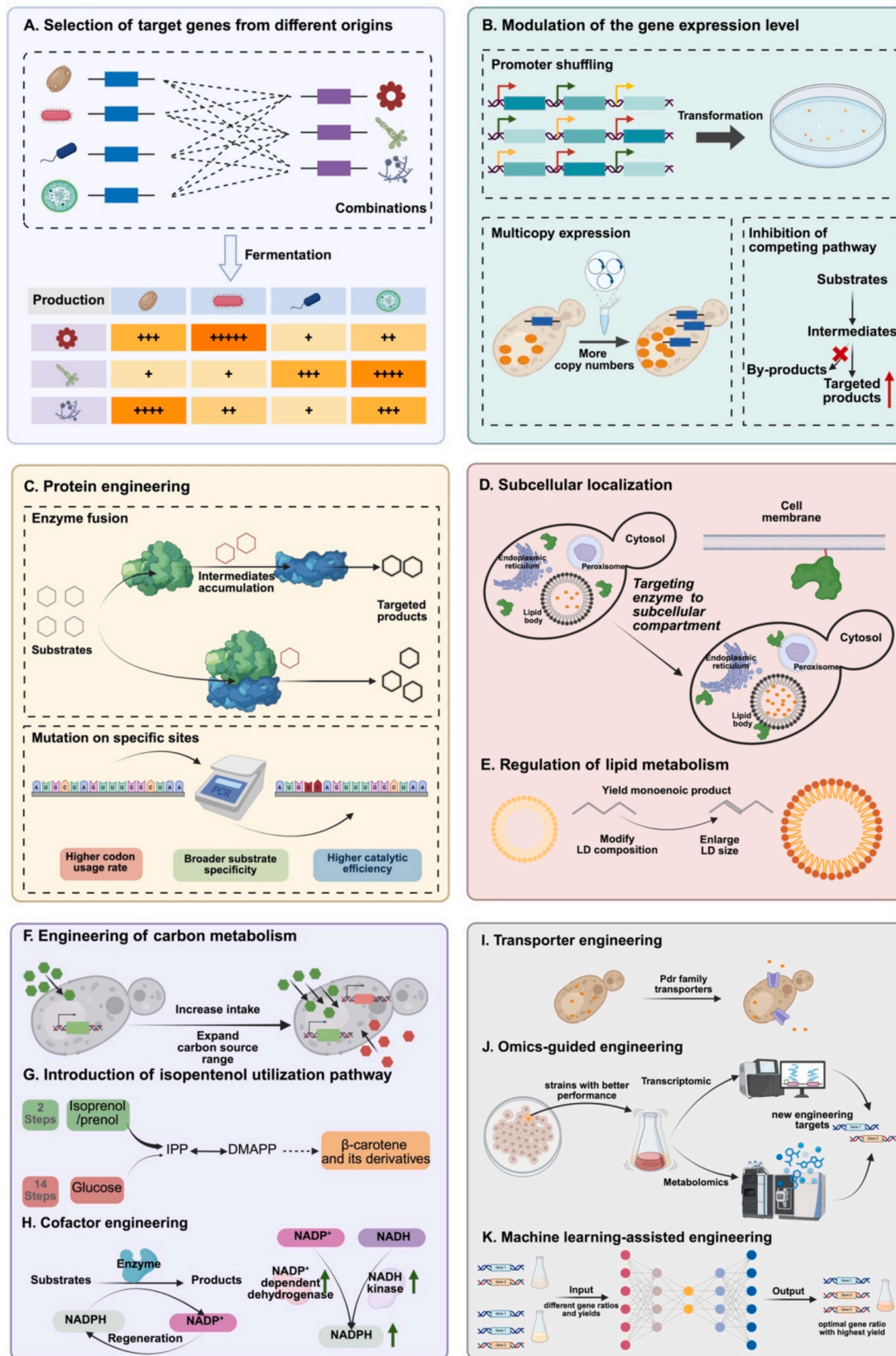
Promoter shuffling and multicopy expression are two widely used metabolic engineering strategies to modulate gene expression levels for enhanced β -carotene and derivative production in yeast (Fig. 3B). Both approaches can be applied to overexpress genes in the upstream MVA pathway and downstream biosynthetic pathway to optimize yield.

Different promoters can quantitatively regulate the gene expression levels and product formation rates (Jing et al., 2022; Ma et al., 2015). A replacement of the promoter of *CtrZ* from pFBA1 to pTEF1 in *S. cerevisiae* led to an increase in astaxanthin yield by 30.4% (Wang et al., 2017a). An automatic recommendation tool for promoter combinations was developed in recent years (Radivojević et al., 2020), hinting at the significance of promoter shuffling for direct regulation of product biosynthesis. For example, three different promoters (pTEF1, pPGM1, and pGAP) were randomly assembled with three target genes for β -carotene by a Golden Gate system in *Y. lipolytica* (Larroude et al., 2018). After screening based on the orange color, *Y. lipolytica* strain bearing *CrtE*, *CrtB*, and *CrtI* with strong promoter pTEF1 was selected as the best producer (Larroude et al., 2018). Similarly, this strategy has also been applied to assemble gene-combination libraries for lycopene production in *S. cerevisiae* (Liu et al., 2019).

Multicopy integration of biosynthetic genes has been applied in the biosynthesis of β -carotene and its derivatives in both *S. cerevisiae* and *Y. lipolytica*. Among MVA genes, *IDI* is the most frequently selected one, demonstrating its indispensable role in the whole pathway (Liu et al., 2019). *HMGR* and *ERG20*, responsible for catalyzing rate-limiting steps (Zhang et al., 2018a), are also frequently overexpressed. In the downstream biosynthetic pathway, multicopy integration is of great value to further increase desired pigment production. *CrtE* (Zhang et al., 2019b; Zhang et al., 2020b), *CrtB* (Zhang et al., 2019b; Zhang et al., 2020b), *CrtI* (Gao et al., 2017; Verwaal et al., 2007), and *CrtZ* (Tramontin et al., 2019; Xie et al., 2021) have all been selected as multicopy overexpression targets. However, copy number optimization is crucial to avoid metabolic burden (Ma et al., 2015). A good example is that a further increase of *HMGR* and *CrtI* copy numbers in the engineered strain already containing 3 copies of *HMGR* and 4 copies of *CrtI* had no significant effect on β -carotene production in *Y. lipolytica* (Gao et al., 2017). Similarly, when comparing strains with two, three, and four copies of *CrtW*, the strain with three copies achieved the highest canthaxanthin titer in *S. cerevisiae* (Chen et al., 2022a). In addition to chromosomal integration, plasmid-based expression can also regulate gene expression levels. When using single-copy plasmid pRS416 in *S. cerevisiae*, crocetin production was limited to 633 μ g/L in *S. cerevisiae* but increased nearly by two fold to 1219 μ g/L when using the multicopy plasmid pRS426 (Chai et al., 2017).

4.2.2. Inhibition of the competing pathway

To accumulate more precursors for desired pathways, undesired branched pathways can be cut off or downregulated, thus ensuring a better metabolic flux towards final products (Fig. 3B). According to Song's report, *CIT2* (encoding citric acid synthase) and *MLS1* (encoding malic acid synthase) in yeast glyoxylate cycle were modulated to enhance crocetin production (Song et al., 2020). Crocetin titer was significantly increased in the Δ *cit2* strain by 50% compared to the control strain in *S. cerevisiae* (Song et al., 2020). Similarly, to increase β -carotene accumulation for astaxanthin biosynthesis, the expression level of *SQS1* (squalene synthase) in the competing pathway was downregulated by truncation of native promoters or replacement of two alternative ones, leading to the increase in β -carotene titer from 453.9 ± 20.2 mg/L to 797.1 ± 57.2 mg/L in *Y. lipolytica* (Kildegaard et al., 2017).

Fig. 3. Metabolic engineering strategies for the biosynthesis of β -carotene and its derivatives.

4.3. Protein engineering

Protein engineering (Fig. 3C) contributes to biosynthesis by either designing new proteins or modifying existing ones to achieve desired functions (Dhanjal et al., 2019).

To enhance the conversion of β -carotene to astaxanthin, enzyme fusion strategies have been employed to improve substrate channeling and reduce by-products. Astaxanthin synthesis involves two sequential reactions, ketolation by CrtW and hydroxylation by CrtZ. Depending on the reaction order of CrtW and CrtZ, multiple pathways from β -carotene to astaxanthin are possible, resulting in the formation of various undesired intermediates (Ma et al., 2021). To solve this problem, the strategy using the fusion enzyme of CrtZ and CrtW has been applied to decrease by-products in both *S. cerevisiae* and *Y. lipolytica* (Ding et al., 2023; Kildegaard et al., 2017; Ma et al., 2021). In *Y. lipolytica*, both fusion enzymes CrtZ-W and CrtW-Z had better astaxanthin production than the control strain, harboring CrtZ and CrtW individually by 2.2-fold and 2.8-fold, respectively (Ma et al., 2021). This might be due to an improved substrate-enzyme proximity and their interactions (Ma et al., 2021). This strategy has also been applied to crocetin biosynthesis, where a CrtZ-CCD fusion protein increased crocetin production to 1.95 ± 0.23 mg/L in *S. cerevisiae* (Song et al., 2020).

Targeted mutations can enhance enzymatic performance by modifying substrate specificity, improving stability, increasing catalytic efficiency, or even introducing novel functions (Chen, 1999). Considering the low codon usage rate when expressing heterogeneous enzymes in *S. cerevisiae*, 5 codons of *CrtI* and 8 codons of *CrtB* were mutated to increase the expression, and a two-fold improvement in β -carotene production was reached through this rational mutation (Li et al., 2013). Apart from optimizing enzyme expression, the catalytic activity can also be improved by site-directed mutagenesis. For example, the CsCCD2 access tunnel (channel within a protein that allows substrates or products to enter or exit the active site) was engineered, obtaining mutants with broader substrate specificity and higher catalytic efficiency for crocetin biosynthesis in *S. cerevisiae* (Liang et al., 2021). The engineered CsCCD2 mutant S323A exhibited a 12.83-fold k_{cat}/K_m compared with the wild-type based on *in vitro* enzymatic assay (Liang et al., 2021). Unlike the rational enzyme re-design, error-prone PCR was performed by increasing $MnCl_2$ concentration (Ding et al., 2023). Random mutants with I87V, L276A, L95S, and I206L of CrtZ-CrtW fusion enzyme could significantly improve astaxanthin yield by 2.5–3.1 times in *S. cerevisiae* (Ding et al., 2023).

In conclusion, both rational and random mutation can contribute to the biosynthesis of β -carotene and its derivatives, and more enzymes, especially those catalyzing rate-limiting steps, can be mutated to realize a better performance.

4.4. Subcellular localization

β -carotene and its derivatives are generally stored in the biological membranes, including plasma and cell membranes (Wu et al., 2018). However, excessive pigment accumulation can reduce membrane fluidity, negatively affecting cell growth (Wu et al., 2018). To avoid this influence, biosynthetic pathways can be re-localized in specific subcellular organelles (Fig. 3D). In Ma's research, part of the astaxanthin synthetic pathway was targeted to the endoplasmic reticulum (ER), lipid body (LB), and peroxisome by adding targeting sequences, KDEL, oleosin, and SKL sequences, respectively, in *Y. lipolytica* (Ma et al., 2021). After single (ER, LB, or peroxisome), double (LB and ER), and triple (LB, ER, and peroxisome) re-localization of pathways, the highest production was observed in the triple organelle-engineering strain (Ma et al., 2021). Similarly, direct localization of the rate-limiting enzyme CrtW to the cell membrane—the primary site for hydrophobic compound storage in *S. cerevisiae*—has been shown to enhance the bio-production of canthaxanthin by 1.33-fold by accelerating the conversion of β -carotene (Chen et al., 2022a).

Apart from the above xanthophylls, subcellular localization is also crucial for apocarotenoids. To improve the efficiency of β -ionone biosynthesis, engineered CCD1 with better membrane affinity by adding membrane-anchoring peptides to the N-terminal domain has shown a three-fold increase of β -ionone production in *S. cerevisiae* (Werner et al., 2019).

Together, these approaches underscore the importance of subcellular localization as an effective strategy in improving β -carotene derivative biosynthesis.

4.5. Regulation of lipid metabolism

Lipid metabolism is challenging to regulate because it demands a dynamic balance for optimal production of β -carotene and its derivatives. On one hand, lipid metabolism competes with the MVA pathway for key precursors such as FPP. To redirect metabolic flux to β -carotene biosynthesis, the expression of *ERG9*, which encodes squalene synthase in sterol biosynthesis, has been downregulated in *S. cerevisiae* (Bu et al., 2022; Lin et al., 2025).

On the other hand, the production of lipophilic natural products, including carotenoids, is often limited by their cytotoxicity and restricted storage capacity (Ma et al., 2019). Aiming to solve this problem, key genes associated with fatty acid synthesis and triacylglycerol (TAG) production were overexpressed to increase lipid droplets in *S. cerevisiae* (Fig. 3E) (Ma et al., 2019). Then, the improved lipid metabolism was further regulated by overexpressing *OLE1* (encoding a fatty acid desaturase) and the deletion of *Seipin* (*FLD1*, regulating the size of lipid droplets). Through engineering lipid metabolism, the composition and size of the core lipid droplet were modified, and the lycopene production could achieve 2.37 g/L, a 25% increase compared with their original high-yield strain (Ma et al., 2019). Similarly, Zhao et al. overexpressed sterol ester synthesis genes (*ARE1* and *ARE2*) and deleted phosphatidate phosphatase genes (*PAH1*, *DPPI1*, and *LPPI1*), resulting in a 2.4-fold improvement in β -carotene production in *S. cerevisiae* (Zhao et al., 2021). These findings reveal the great potential of extended and systematic lipid engineering to boost the biosynthesis of lipophilic β -carotene and its derivatives. As an oleaginous yeast with a robust lipid metabolism (Salvador López et al., 2022), *Y. lipolytica* has provided an advantage when synthesizing lipophilic products like β -carotene and its derivatives. Using a lipid-overproducing *Y. lipolytica* as the parental strain, 8.9 mg/g DCW β -carotene production was obtained, which was 2.61 times more than the control strain (Larroude et al., 2018). Interestingly, the boosted β -carotene pathway also favored lipid synthesis in *Y. lipolytica*, offering dual benefits for industrial application (Larroude et al., 2018). The improved hydrophobic environment favors isoprenoid deposition (Wu et al., 2020) and may help remove the final product from the proximity of the biosynthetic pathway to avoid product inhibition, thus 'pulling' more flux towards isoprenoids (Tai and Stephanopoulos, 2013). These cases indicate that further optimization of lipid metabolism in oleaginous yeasts can facilitate the bioproduction of β -carotene and its derivatives.

Ideally, a dynamic regulation of lipid metabolism is preferred to maintain the balance between precursor availability and storage capacity. For instance, the supplementation of exogenous oleic acid (OA) can enhance TAG metabolism and increase storage potential, while employing OA-repressive promoter to replace native *ERG9* promoter could allow for tunable downregulation of sterol biosynthesis, ensuring sufficient flux towards desired pathways (Bu et al., 2022).

4.6. Engineering of carbon metabolism

While most metabolic engineering efforts focus on optimizing pathways downstream of the indispensable precursor acetyl-CoA, it is crucial to recognize that fundamental carbon metabolism also significantly influences the entire synthetic pathway (Fig. 3F), including the upstream MVA pathway and downstream biosynthesis of β -carotene and its

derivatives.

To enhance glucose uptake and improve β -carotene accumulation in *Y. lipolytica*, Qiang et al. selected the rate-limiting hexokinase gene *HXK* as an overexpression target to increase glucose intake, resulting in a better β -carotene accumulation in *Y. lipolytica* (Qiang et al., 2020). Beyond glycolysis, xylose and arabinose utilization pathways were introduced to yeast for a broader range of carbon sources (Sun et al., 2012; Sun et al., 2020). For example, when culturing with xylose, the xylose-fermenting *S. cerevisiae* had a higher titer than the glucose-utilizing strain by 3-fold (Sun et al., 2020). In addition, lipase genes (*LIP2*, *LIP7*, and *LIP8*) derived from *Y. lipolytica* could also be integrated into *S. cerevisiae*, enabling a direct utilization of olive oil and triolein when they serve as the sole carbon source (Fathi et al., 2021).

4.7. Introduction of isopentenol utilization pathway

β -Carotene and its derivatives are all generated from two C5 isoprenoid precursors, IPP and its isomer DMAPP, through the MVA pathway in yeast (Fig. 3G). However, the natural pathway has an unsolved limitation: intermediates or downstream products have been demonstrated to inhibit “gatekeeper” MVA-related enzymes (Chatzivasilieiou et al., 2019). For instance, CoA, acetoacetyl-CoA, and HMG-CoA can inhibit HMG-CoA synthase (encoded by *ERG13*) (Nahegowda et al., 2004); HMG, free CoA, and NAD(P)⁺/NADPH inhibit HMG-CoA reductase (encoded by *HMGR*) (Bach et al., 1986; Brooker and Russell, 1975); and IPP, DMAPP, GPP, and FPP inhibit mevalonate kinase (encoded by *ERG12*) (Primak et al., 2011).

To overcome these bottlenecks and ensure an abundant supply of IPP and DMAPP for downstream biosynthesis of β -carotene and its derivatives while avoiding product inhibition, a novel pathway termed isopentenol utilization pathway (IUP) was designed (Chatzivasilieiou et al., 2019). In the IUP, choline kinase (CK) catalyzes the phosphorylation of isoprenol or prenol into isopentenyl monophosphate (IP) or dimethylallyl monophosphate (DMP), which are then further phosphorylated to form IPP or DMAPP, respectively, by isopentenyl phosphate kinase (IPK). This two-step process effectively decouples isoprenoid biosynthesis from central carbon metabolism (Chatzivasilieiou et al., 2019). More importantly, IUP dramatically simplifies the biosynthesis of IPP and DMAPP and enables exceptionally high flux toward isoprenoid synthesis, serving as a universal strategy applicable to the biosynthesis of nearly all β -carotene derivatives. For example, introducing IUP into *Y. lipolytica* elevated the IPP/DMAPP level by 15.7-fold compared to the native MVA pathway alone, resulting in a remarkable lycopene titer of 4.2 g/L (Luo et al., 2020).

4.8. Cofactor engineering

In the carotenoid synthetic pathway, NADPH is a decisive reducing cofactor directly influencing carotenoid production (Jing et al., 2021). Both enhancement and regeneration of NADPH can be used in yeast to increase the production of β -carotene and its derivatives (Fig. 3H). It has been proven that overexpressing *ZWF1* (encoding 6-phosphate dehydrogenase) and *POS5* (encoding NADH kinase) had the ability to increase the NADPH concentration in *S. cerevisiae* and then increase the yield of lycopene and β -carotene (Jing et al., 2021). Enzymatic NADPH regeneration can be realized through “coupled enzyme” or “coupled substrate” (Wang et al., 2017b), providing abundant cofactors continuously. To be more specific, glucose dehydrogenase (GDH) and formate dehydrogenase (FDH) are the two additional enzymes most widely applied for cofactor regeneration, associated with their sacrificial hydride donors (glucose and formate) when using the “coupled enzyme” strategy (Wang et al., 2017b). Meanwhile, there is no second enzyme in the “coupled substrate” strategy, meaning that one enzyme should be functional in both substrate reduction and cofactor regeneration (Wang et al., 2017b).

4.9. Transporter engineering

In *Y. lipolytica*, lipid bodies provide an ideal intracellular compartment for the accumulation of β -carotene and its derivatives. In contrast, *S. cerevisiae* lacks such dedicated storage organelles, relying mainly on the cell membrane, which often leads to excessive intracellular accumulation and consequently limits the further improvement in product yields (Liu et al., 2023). To solve this bottleneck, Liu et al. employed *in silico* molecular docking to screen potential transporters. Through a “mining-docking-construction-validation” strategy, Pdr5 (ABC transporter) was found to enhance the secretion of β -carotene and retinal (Fig. 3I) (Liu et al., 2023), offering a broadly applicable strategy for transporter discovery and secretion-based production enhancement. Similarly, Pdr family transporters were also overexpressed in *S. cerevisiae* for alleviating cell stress and then further improving canthaxanthin production (Chen et al., 2022a).

For apocarotenoid biosynthesis in yeast, crocetin has traditionally been regarded as an intracellular compound in *S. cerevisiae*. Interestingly, a potentially secretable biosynthesis has recently been reported in *Y. lipolytica*, which may partly explain the higher crocetin titer achieved in this host (Zhou et al., 2025). Although the underlying mechanism for crocetin secretion in *Y. lipolytica* remains unclear, this phenomenon suggests a promising new direction for titer improvement through transporter engineering.

4.10. Omics-guided engineering

Unlike traditional metabolic engineering based on known gene targets, new targets also can be identified from transcriptomics analysis (Fig. 3J). This has been used to identify the target genes for improving astaxanthin biosynthesis (Jin et al., 2018). The highest astaxanthin-producing *S. cerevisiae* strain screened by the darkest red color after atmospheric and room temperature plasma (ATRP) mutagenesis was further studied through genomic and transcriptomic analysis (Jin et al., 2018). Three genes *CSS1*, *YBR012W-B*, and *DAN4*, were first uncovered, being involved in regulating astaxanthin production. Notably, the deletion of *CSS1* has been demonstrated to upregulate membrane synthesis, thus improving astaxanthin yield significantly (Jin et al., 2018).

Meanwhile, metabolomics also serves as a valuable tool for gaining deeper insights into the metabolic state of engineered yeast strains (Fig. 3J). To be more specific, a comparative metabolomic analysis between β -carotene-producing *S. cerevisiae* and its parent strain revealed a lower level of central intermediates (amino acids, carbohydrates, glycolysis, and TCA cycle intermediates) in recombinant yeast (Bu et al., 2017). These findings suggest that redirecting metabolic flux toward carotenoid biosynthesis may deplete key central metabolites, providing potential targets for further metabolic engineering.

4.11. Machine learning-assisted engineering

In supervised machine learning, predictive models are trained on a limited set of input–output data pairs, such as genetic modifications and corresponding metabolite production levels. Once trained, these models can be used to predict the metabolic outcomes of new, untested genetic designs in yeast, thereby guiding targeted metabolic engineering efforts (Fig. 3K). Shimazaki et al. built a machine learning model based on expression levels of 8 genes in the MVA pathway and carotenoid productivity, successfully predicting the optimal gene expression levels for the highest carotenoid production (4.3 times higher than the parental strain) in *S. cerevisiae* (Shimazaki et al., 2024).

5. Fermentation optimization for enhanced production of β -carotene and its derivatives

Metabolic optimization is a synergistic combination of both genetic and fermentation optimization strategies (Jones and Koffas, 2016).

While many metabolic engineering strategies focused on modifying the genetic pathways of production strains, integrating these approaches with culture condition optimization and bioreactor development can further enhance the yield of industrially valuable products (Marinelli and Marcone, 2011). From the perspective of green biomanufacturing, such integration not only improves productivity but also contributes to environmental sustainability by reducing resource inputs, minimizing waste generation, and enabling energy-efficient biosynthetic routes. This holistic strategy ensures that both cellular metabolism and external environmental factors are optimized to maximize biosynthetic efficiency.

5.1. Carbon and nitrogen sources

Improvements to fermentation media composition, including carbon sources, nitrogen sources, and other essential nutrients, play a crucial role in optimizing β -carotene and its derivatives' production (Fig. 4A). As discussed earlier, alternative carbon sources such as xylose (Sun et al., 2020), arabinose (Sun et al., 2012), olive oil, and triolein (Fathi et al., 2021) have been explored. Compared with those genetic methods to optimize carbon sources, a simple change of glucose concentration is also influential to the production of β -carotene and its derivatives. A gradient glucose concentration is always used to find out the optimal concentration. For example, the astaxanthin-producing *Y. lipolytica* strain was cultured in YPD media with 20 g/L, 30 g/L, 40 g/L, and 50 g/

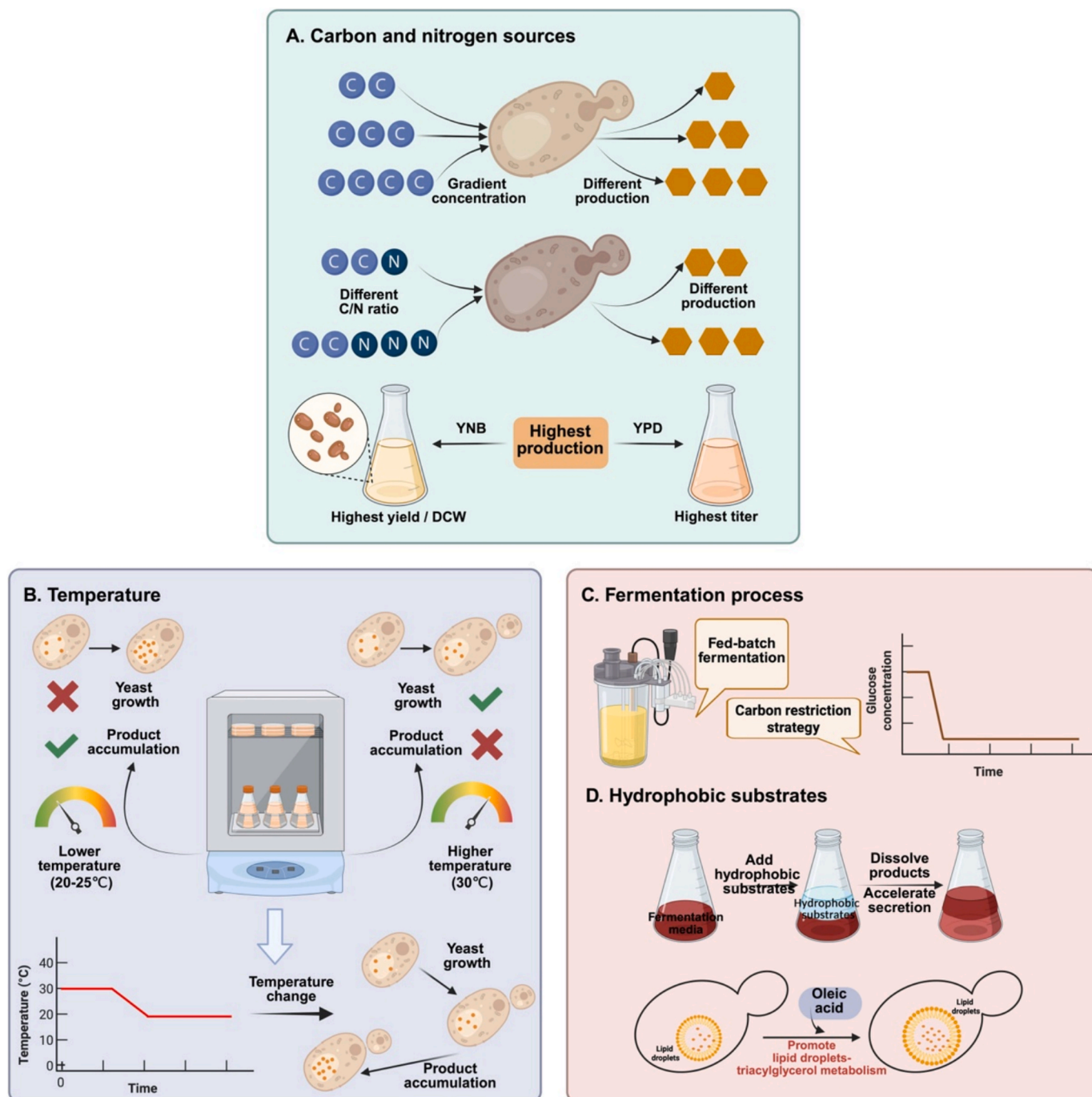


Fig. 4. Strategies on fermentation optimization for producing β -carotene and its derivatives.

L of glucose, and the best production was obtained from 40 g/L of glucose (Ma et al., 2021). Beyond individual nutrient concentrations, the carbon-to-nitrogen (C/N) ratio is another critical factor affecting productivity. Adjusting the C/N ratio from 3:1.5 to 2:3 resulted in an increase in β -carotene yield from 29.12 ± 1.93 mg/g DCW to 33 ± 1.2 mg/g DCW in *Y. lipolytica* (Gao et al., 2017). This highlights the importance of optimizing both absolute concentrations and nutrient balance to ensure stable and enhanced production.

Interestingly, different fermentation media can maximize either overall titer or yield per dry cell weight (DCW). In the case of zeaxanthin production in *Y. lipolytica*, the highest titer (21.98 ± 1.80 mg/L) was achieved in rich YPD medium, whereas the highest yield per DCW (3.20 ± 0.11 mg/g) was obtained in synthetic YNB medium (Xie et al., 2021). This suggests that researchers can tailor media selection based on specific production goals.

In line with the principles of green biomanufacturing, the use of renewable, low-cost, and waste-derived feedstocks presents a promising strategy to enhance both the sustainability and economic viability of microbial production systems. Agricultural byproducts like wheat waste have been successfully utilized as cost-effective fermentation substrates for the biosynthesis of high-value compounds like astaxanthin (Dursun and Dalgıç, 2016), significantly lowering production costs while reducing environmental impact. Similarly, in the production of β -ionone in *Y. lipolytica*, Chen et al. demonstrated the efficient conversion of food and agricultural waste into nutrient-rich hydrolysates, yielding fermentable sugar concentrations as high as 78.4 g/L glucose and 8.3 g/L fructose, or 34.7 g/L glucose and 20.1 g/L xylose (Chen et al., 2022b). This approach not only enabled robust cell growth and metabolite production but also achieved the highest β -ionone yield reported to date in microbial fermentation (Chen et al., 2022b). These examples underscore the potential of coupling metabolic engineering with circular bioeconomy strategies to create efficient, low-carbon, and scalable platforms for a more sustainable biosynthesis of β -carotene and its derivatives.

5.2. Temperature

While yeasts like *S. cerevisiae* and *Y. lipolytica* are typically cultured at 30 °C, optimal biosynthesis of β -carotene and its derivatives often occurs at lower temperatures (Fig. 4B). Shi et al. reported that temperature could influence the β -carotene accumulation in recombinant *S. cerevisiae*, and 20 °C was the optimal temperature for its biosynthesis (Shi et al., 2014). They also conducted a transcriptional analysis by real-time PCR to figure out the transcription level of genes in yeast growing at different temperatures (Shi et al., 2014). Despite a lower growth rate of cells indicating the lower transcription level of *KGD1* (encoding α -ketoglutarate dehydrogenase of the tricarboxylic acid cycle) at 20 °C, a higher mRNA level of β -carotene synthesis genes (*Crt*) was proved (Shi et al., 2014). The lower temperature preference for producing β -carotene and its derivatives was also supported by crocetin biosynthesis in both *S. cerevisiae* (Chai et al., 2017) and *Y. lipolytica* (Zhou et al., 2025).

This temperature-dependent trade-off between yeast growth (optimal at 30 °C) and carotenoid production (optimal at 20 °C) presents a challenge for industrial-scale fermentation. To address this, a temperature-responsive GAL system was developed recently in *S. cerevisiae* (Chen et al., 2022a). In this system, the culturing temperature was kept at 30 °C during the cell growth stage until the OD₆₀₀ reached 160 and then reduced to 24 °C to initiate the canthaxanthin biosynthesis (Chen et al., 2022a). At last, 1.44 g/L canthaxanthin was harvested based on this temperature-responsive high-density fermentation (Chen et al., 2022a), offering a promising system for improved production of other β -carotene derivatives. A simpler two-step temperature-shift strategy was also successfully applied to crocetin biosynthesis in *Y. lipolytica*. In this method, yeast was cultured at 30 °C for the first two days to promote cell growth, followed by a temperature reduction to 20 °C for the remaining five days, leading to improved crocetin

production (Zhou et al., 2025).

These findings suggest that dynamic temperature regulation is an effective strategy for balancing yeast growth and product yield in carotenoid biosynthesis. One possible explanation for the enhanced biosynthetic efficiency at lower temperatures is that many plant-derived enzymes tend to exhibit higher stability and activity under mild, low-temperature conditions (Trenchard and Smolke, 2015). This aligns with the observation that the heterologous expression of recombinant proteins in yeast often benefits from reduced cultivation temperature (Çelik and Çalik, 2012). Therefore, implementing temperature-change strategies not only mitigates the trade-off between biomass accumulation and product formation but may also improve the functional expression of heterologous enzymes, ultimately leading to enhanced carotenoid yields in engineered yeasts.

5.3. Fermentation process

Bioreactor fermentation is essential for large-scale bioproduct manufacturing, as it ensures optimal conditions—such as temperature, pH, and oxygen levels—for both cell growth and metabolite production (Wang et al., 2005). To maximize β -carotene and derivative yields, engineered yeast strains are typically cultured in bioreactors using fed-batch fermentation (Fig. 4C), the most common strategy for industrial applications.

Based on the carbon restriction strategy (Chai et al., 2017), glucose concentration in bioreactors is normally restricted to maintain a semi-starvation status (Chen et al., 2022a). While a high initial glucose concentration supports cell growth, lower concentrations are maintained throughout the fermentation process. For example, glucose concentrations were limited to 0.5% for β -carotene (Qiang et al., 2020), 0.5% for crocetin fermentation (Chai et al., 2017), and 2% for astaxanthin fermentation (Wang et al., 2017a). This carbon restriction method during bioreactor fermentation processes nicely prevents the overflow metabolism, where excess glucose shifts metabolism toward ethanol production rather than biomass and product formation, diverting carbon away from desired compounds.

In addition to glucose regulation, nitrogen supplementation also plays a crucial role in fermentation. Unlike carbon sources, nitrogen-rich compounds such as yeast extract and peptone are often continuously supplied (Larroude et al., 2018; Wang et al., 2017a) probably to support sustained growth and enzyme synthesis. For instance, doubling the yeast extract and peptone concentrations in *Y. lipolytica* fermentation significantly increased β -carotene production from 2.9 g/L to 6.5 g/L (Larroude et al., 2018).

Other fermentation parameters, like dissolved oxygen, can also be optimized during the fermentation process. Lv et al. applied a constant dissolved oxygen value feeding (DO-stat) to enhance the β -carotene production during *Y. lipolytica* fermentation by reducing the Crabtree effect and improving the utilization rate of glucose (Lv et al., 2020). The data also illustrated that more dissolved oxygen increased the biomass (Lv et al., 2020).

5.4. Hydrophobic substrates

During fermentation, some hydrophobic substrates are added to the fermentation system to improve the production of β -carotene and its derivatives in yeast (Fig. 4D). Due to their hydrophobic nature, β -carotene and its derivatives can readily dissolve in exogenous oil layers added to the fermentation medium, facilitating their secretion and ultimately increasing overall production. This strategy has been particularly effective in astaxanthin production in *Y. lipolytica*, where both single and sequential oil additions have demonstrated positive effects. However, the optimal supplementation method should be carefully adapted to different carotenoids, considering their distinct accumulation patterns (Yuzbasheva et al., 2023).

Beyond serving as a solvent, hydrophobic substrates can also

enhance lipid metabolism, further promoting carotenoid biosynthesis. For example, the addition of 2 mM oleic acid improved lipid droplet-triacylglycerol metabolism in *S. cerevisiae*, resulting in a 36.4 % increase in β -carotene content (Bu et al., 2022).

These findings highlight the potential of hydrophobic substrate supplementation as a valuable strategy for optimizing yeast-based carotenoid production.

6. Comparative analysis and future research

Based on current studies, *S. cerevisiae* and *Y. lipolytica* demonstrate distinct strengths in the biosynthesis of β -carotene and its derivatives. In particular, *Y. lipolytica* outperforms the traditional yeast *S. cerevisiae* in the production of β -carotene and its xanthophyll derivatives, demonstrating superior metabolic compatibility and higher yields. One key contributing factor is its sufficient supply of acetyl-CoA, a critical precursor in the carotenoid biosynthetic pathway. More importantly, the oleaginous nature of *Y. lipolytica* dramatically facilitates the storage of the highly hydrophobic compounds like β -carotene and its xanthophyll derivatives in lipid bodies. This not only mitigates the cytotoxic effects typically associated with such compounds but also allows for greater accumulation. Additionally, *Y. lipolytica* is capable of achieving higher biomass and sustaining growth under high cell density fermentation conditions, further supporting the efficient production of these intracellular metabolites. However, despite generally low production levels, a wider variety of xanthophylls has been successfully biosynthesized in *S. cerevisiae*. In the case of apocarotenoid derivatives, the significant reduction in their hydrophobicity (Fig. 5) likely diminishes the storage advantage offered by *Y. lipolytica*. As a result, the benefits of its oleaginous nature become less relevant. Moreover, the enzymatic cleavage of β -carotene or other xanthophyll precursors to apocarotenoids remains a major bottleneck, which restricts the metabolic flux toward the target products. Therefore, there appears to be no substantial difference in production capacity between the two yeast chassis for most apocarotenoids. Interestingly, *S. cerevisiae* has demonstrated the ability to produce a broader range of apocarotenoid compounds compared to *Y. lipolytica*. This is likely due to its longer history as a model organism in synthetic biology and the availability of a more comprehensive and mature genetic engineering tools, which facilitates the modular construction and fine-tuning of complex biosynthetic pathways.

Although the biosynthesis of β -carotene and its derivatives in yeast has achieved promising progress at the laboratory scale with distinct advantages using different strains, translating these advances into cost-effective industrial-scale production remains a significant challenge. In comparison to traditional chemical synthesis, which benefits from decades of industrial refinement, mature infrastructure, and well-established processes, yeast biomanufacturing still faces hurdles such as relatively low yields, high production costs (Venil et al., 2020), and complex extraction procedures (Saini and Keum, 2018). To address the

issue of limited productivity, more systematic metabolic engineering strategies are needed to enhance pathway efficiency and boost product titers. From a fermentation standpoint, reducing production costs will also depend on optimizing key parameters, including nutrient composition (Igreja et al., 2021). The use of low-cost, waste-derived feedstocks presents a dual advantage: improving environmental sustainability while significantly lowering raw material expenses. Furthermore, the development of efficient, scalable, and economically viable downstream extraction methods (Saini and Keum, 2018) will also be essential to support the broader industrial deployment of yeast-based biomanufacturing.

7. Conclusions

β -Carotene and its derivatives are in high demand due to their multifunctional properties and wide-ranging applications across various industries. As consumer demand for these value-added natural pigments continues to grow, microbial biosynthesis presents a sustainable, eco-friendly, and economically viable alternative to traditional extraction and chemical synthesis methods.

In this review, we highlighted yeast as a chassis for β -carotene and its derivatives production, summarizing recent advancements in metabolic engineering strategies. Additionally, we explored fermentation optimization approaches, including nutrient regulation, temperature control, bioreactor strategies, and hydrophobic substrate supplementation, all of which contribute to improved yields. Furthermore, we presented a comparative analysis of yeast chassis selection, emphasizing how the physicochemical properties of different carotenoid derivatives (e.g., xanthophylls vs. apocarotenoids) influence host suitability.

By integrating precise genetic modifications with optimized fermentation conditions, the large-scale and cost-effective production of β -carotene and its derivatives in yeast can be further advanced. We hope this review provides a comprehensive and practical reference for researchers and industry professionals, facilitating the rational design and optimization of microbial cell factories, ultimately accelerating industrial applications and commercialization of β -carotene and its derivatives.

CRediT authorship contribution statement

Tingan Zhou: Writing – original draft, Investigation, Conceptualization. **Young-Kyoung Park:** Writing – review & editing, Supervision, Investigation, Conceptualization. **Rodrigo Ledesma-Amaro:** Writing – review & editing, Supervision, Project administration, Funding acquisition, Conceptualization.

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Declaration of competing interest

The authors declare that they have no known competing financial

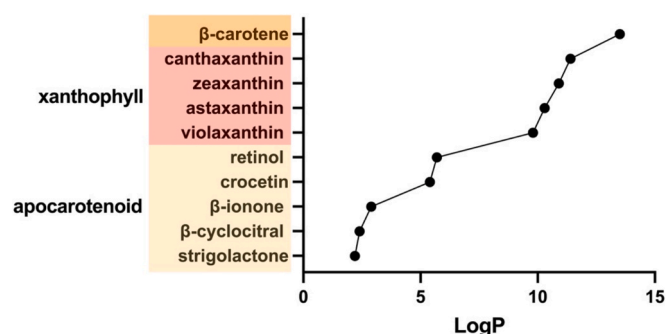


Fig. 5. Comparison of hydrophobicity between xanthophylls and apocarotenoids. LogP values are computed by XLogP3 3.0 and are available on PubChem (<https://pubchem.ncbi.nlm.nih.gov>).

interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

No data was used for the research described in the article.

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