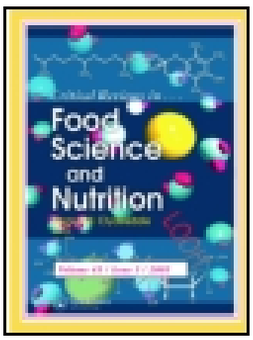

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REVIEW

Carotenoids supplementation and inflammation: a systematic review and meta-analysis of randomized clinical trials

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ABSTRACT

The aim of this study was to perform a systematic review and meta-analysis on randomized controlled trials investigating the effects of carotenoids on selected inflammatory parameters. PubMed, SCOPUS, and Web of science were searched from inception until April 2021. The random-effect model was used to analyze data and the overall effect size was computed as weighted mean difference (WMD) and corresponding 95% of confidence interval (CI). A total of 26 trials with 35 effect sizes were included in this meta-analysis. The results indicated significant effects of carotenoids on C-reactive protein (CRP) (WMD: −0.54 mg/L, 95% CI: −0.71, −0.37, $P < 0.001$), and interleukin-6 (IL-6) (WMD: −0.54 pg/mL, 95% CI: −1.01, −0.06, $P = 0.025$), however the effect on tumor necrosis factor-alpha (TNF-α) was not significant (WMD: −0.97 pg/ml, 95% CI: −1.98, 0.03, $P = 0.0.059$). For the individual carotenoids, astaxanthin, (WMD: −0.30 mg/L, 95% CI: −0.51, −0.09, $P = 0.005$), lutein/zeaxanthin (WMD: −0.30 mg/L, 95% CI: −0.45, −0.15, $P < 0.001$), and β-cryptoxanthin (WMD: −0.35 mg/L, 95% CI: −0.54, −0.15, $P < 0.001$) significantly decreased CRP level. Also, only lycopene (WMD: −1.08 pg/ml, 95%CI: −2.03, −0.12, $P = 0.027$) led to a significant decrease in IL-6. The overall results supported possible protective effects of carotenoids on inflammatory biomarkers.

KEYWORDS

Carotenoids; inflammatory biomarkers; CRP; IL-6; TNF-α

1. Introduction

The consumption of a diet rich in fruits and vegetables strongly contributes to the prevention and management of various chronic disturbances such as cardiovascular diseases (CVD) and diabetes mellitus (Coyne et al. 2005; Hozawa et al. 2006; Riccioni et al. 2010). Many research has suggested that carotenoids are one of the most contributing components of health benefits of plant-based diets including fruits and vegetables for (Kesse-Guyot et al. 2013; Liu 2013). The compounds belong to the tetraterpenes family which can be structurally categorized into two classes including carotenes (such as α- and β-carotenes and lycopene) which are non-polar carotenoids containing only a hydrocarbon chain without any functional group, and xanthophylls (such as lutein, zeaxanthin, β-cryptoxanthin, astaxanthin, and crocin) which are polar molecules with at least one oxygen atom (Britton 1995). There is also a function-based classification of carotenoids including provitamin A carotenoids (β-cryptoxanthin, α- and β-carotenes), and the non-provitamin A carotenoids (Waris and Ahsan 2006). Further, lutein and its isomer zeaxanthin are the important carotenoids that highly concentrated in the macula.

Decades of research have focused on the potential protective role of carotenoids on visual health; however, there is an increasing evidence that these lipophilic pigments may provide additional protection against obesity, CVD, dyslipidemia, diabetes mellitus, insulin resistance, neurocognitive disorders, and malignancies (Coyne et al. 2005; Hajizadeh-Sharafabad et al. 2020; Hoffmann et al. 2015; Hozawa et al. 2006; Mounien, Tourniaire, and Landrier 2019; Niranjana et al. 2015; Riccioni et al. 2010).

Based on evidence, many potential benefits of carotenoids in human health are mainly derived from their antioxidant activities (Fiedor and Burda 2014). The absence of any electrophilic group in carotenes leads to quenching of free radicals only in the lipid phase (El-Agamey and McGarvey 2008); whereas, polar xanthophylls not only exhibited more antioxidant activities than the carotenes, but also may scavenge radicals in both lipophilic and aqueous environments (Bouayed and Bohn 2012). In addition to antioxidant

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behavior, several biological activities have been also identified for carotenoids including control of cell cycle and differentiation; apoptosis; regulation of intracellular signaling pathways; immunity enhancement; and photo-protection (Stahl, Ale-Agha, and Polidori 2002). The encouraging results coming from in vitro and animal models (Harari et al. 2008; J.-H. Kim et al. 2008; Kuhad, Sethi, and Chopra 2008; Linnewiel-Hermoni et al. 2014; Ni et al. 2015), incited the association between carotenoids and inflammation. Moreover, some observational studies provided solid evidence for inverse associations between plasma levels or dietary intake of carotenoids and inflammatory mediators (Mohsen Mazidi et al. 2018; Walston et al. 2006; L. Wang et al. 2008). Despite several randomized controlled trials (RCT) examining true effect of carotenoids on the inflammation, there is no comprehensive conclusion on the exact effect of carotenoids on inflammation. Thus, the aim of this paper was to systematically review and analyze the clinical evidence derived from all published RCTs investigating the effects of carotenoids on circulating levels of CRP, IL-6, and TNF-α, the most targeted biomarkers of inflammatory response.

2. Methods

2.1. Eligibility criteria

Human studies investigating the effects of carotenoids intervention on at least one marker of inflammation included in this systematic review. The selection criteria were: RCTs on participants aged $\geq$ 17 y, with no limit for the gender, ethnicity, and health status, in which intervention group received either α-carotene, β-carotene, lycopene, lutein with or without zeaxanthin, β-cryptoxanthin, astaxanthin, or crocin; RCTs with control group which the difference between the groups was only the intake of carotenoids; trials used purified or extracted carotenoids that the quantitative carotenoid dosage was reported; and the trials provided sufficient data for baseline and endpoint values of at least one of the inflammatory biomarkers including CRP, IL-6, and TNF-α. Trials were excluded, if (1) the effect of carotenoids was not independently examined; (2) there was no appropriate control group or no randomization process; (3) the information for the dosage, duration of the intervention, and type of carotenoids was not provided; (4) studies used nonpurified carotenoid-rich foods or extracted carotenoids without mentioning quantitative carotenoid dose; (5) RCTs involving pregnant or lactating women, and (6) studies were not published in English language. Nonclinical studies, reviews, editorials, non-research letters, or conference abstracts were also excluded.

2.2. Search strategy and data extraction

This systematic review was performed in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA). We employed a PICOS (population, intervention, comparator, outcomes and setting) approach to design the present systematic review as follows:

Population: human populations;

Intervention: supplementation of common carotenoids including α-carotene, β-carotene, lycopene, lutein with or without zeaxanthin, β-cryptoxanthin, astaxanthin, or crocin;

Comparator: using a placebo or active control, on the condition that the difference between the groups was only intake of carotenoids;

Outcomes: circulating levels of CRP, IL-6, and TNF-α;

Setting: RCT.

A systematic search was carried out on electronic databases PubMed, ISI Web of Science, Scopus, using relevant key words/terms from the earliest to April 2021.

Key search terms were "carotenoids" OR "carotene" OR "beta-carotene" OR "alpha-carotene" OR "cryptoxanthin" OR "beta-cryptoxanthin" OR "lutein" OR "lycopene" OR "zeaxanthin" OR "astaxanthin" OR "crocin" OR "xanthophyll" AND "inflammation" OR "Tumor Necrosis Factor" OR "TNF-α" OR "Tumor Necrosis Factor-alpha" OR "TNF alpha" OR "TNF" OR "IL-6" OR "Interleukin 6" OR "C-reactive protein" OR "CRP" OR "hs-CRP" OR "high sensitivity-CRP".

Two investigators independently screened titles and/or abstracts of obtained studies to evaluate eligibility criteria. The full-texts articles were separately appraised by the same reviewers based on inclusion criteria and then the eligible studies were left for data extraction. The data extraction template included author/date/origin, type, dosage, and duration of the intervention, characteristics of participants, study design, and inflammatory measures. At each stage, any disagreements were resolved by discussion within the research team.

2.3. Study quality and risk of bias within the studies

Quality of articles was independently determined by two investigators using criteria of Cochrane Handbook for Systematic Reviews of Interventions which is a 7-item instrument allowing the assessment of random sequence generation, allocation concealment, blinding of participants and personnel, blinding of outcome assessment, selective outcome reporting, incomplete outcome data, and other sources of bias. Interpretation of the scale was conducted at the study level, whereby each study was classified as either high risk of bias (high risk of bias for one or more key domains), or unclear risk of bias (low or unclear risk of bias for all key domains), and low risk of bias (low risk of bias for all key domains).

2.4. Statistical analysis

The means and standard deviations (SDs) at the baseline and endpoint were directly extracted from included studies. If not provided by the author, SDs were computed from either standard errors, interquartile ranges, or 95% CIs. In cases where the outcomes were not expressed in the main

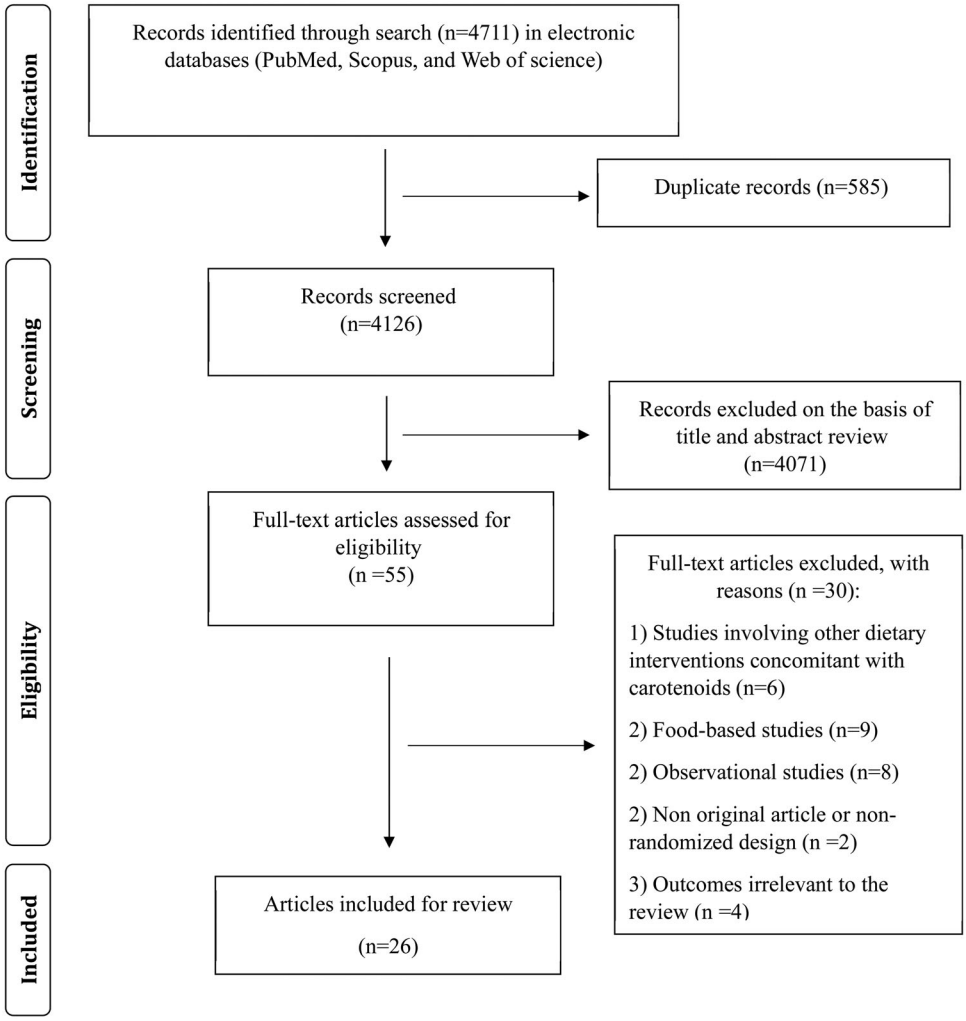

Figure 1. Flow diagram of the literature search and study selection process.

text or tables, the data were extracted from the graphs. Where the changes in outcomes were assessed at multiple time points, only data at the longest duration of follow-up was extracted for meta-analysis. The effect sizes for CRP, IL-6, and TNFα in each trial were determined as weighted mean difference (WMD) and its 95% confidence interval (CI). Pooled WMDs and their 95%CIs are presented in forest plots to evaluate the overall effects of carotenoids in comparison with control group. The pooled effects were estimated using random-effect model that considers the heterogeneity of studies. The inter-study heterogeneity was evaluated by the Cochran Q test (considered significant for P values <0.10) and I-squared (I^2) index, with values of 25, 50, and 75 to present as low, moderate, and high heterogeneity, respectively. To explore possible sources of inter-study heterogeneity, subgroup analyses were conducted based on dosage, intervention duration, participants' age, and health condition. Publication bias was determined by Begg's regression test, where more than ten articles were available for those outcomes. Regarding outcomes for which there are less than ten articles, publication bias was examined through visual inspection of funnel plots as well as Egger's regression test. In addition, the trim-and-fill analysis was used to compensate any significant publication bias observed. To evaluate whether a special study might affect overall results, sensitivity analysis was employed using the one-study remove approach. All of statistical analyses were conducted using Comprehensive Meta Analysis V2 software.

3. Results

The flow diagram of study selection process is depicted in Figure 1. A total of 4711 records from the database search were identified, of which 585 duplicate articles were omitted, 3921 of the remaining 4126 records were excluded based on title and abstract screening and 205 were excluded because they did not meet the inclusion criteria. Ultimately, 55 full-text articles were reviewed for eligibility, and 26 studies were identified as being RCTs of carotenoids on selected inflammatory biomarkers and included in the systematic review and meta-analysis. Where the published articles did not provide sufficient data on inflammatory outcomes, we tried to contact the authors of the trials by email to access full data.

3.1. Characteristics of included studies

Table 1 outlined the main characteristics of the 26 included studies. All studies were RCTs, of which only one had a

Table 1. Overview of the characteristics and main findings of the clinical trials included in the systematic review.

Author/date/country	Study design	Study participants	Nutritional intervention			Main outcomes
			Groups	Dose of supplement	Duration (week)	
Poursamimi et al./2020/Iran (Poursamimi et al. 2020)	Parallel RCT	35 Osteoarthritis Patients; Mean age: 58.1yrs, Mean BMI: 25.8 kg/m^2	Crocin vs. placebo	15 mg/d	16	CRP
Chan et al./2019/Taiwan (Chan, Chen, and Chen 2019)	Parallel RCT	Mean age: 59.6 yrs 54 Type 2 diabetic; patients; Mean BMI: 25.8 kg/m^2	Astaxanthin vs. placebo	Study1: 6 mg/d Study2: 12 mg/d	8	hs-CRP, IL-6, TNF-α
Ghaderi et al./2019/Iran (Ghaderi et al. 2019)	Parallel RCT	53 Patients undergoing MMT; Mean age: 45.0 yrs; Mean BMI: 24.8 kg/m^2	Crocin vs. placebo	30 mg/d	8	hs-CRP
Stringham et al./2019/USA (Stringham, Holmes, and Stringham 2019)	Parallel RCT	59 Healthy subjects; Mean age: 21.5 yrs Mean BMI: 18.5–27 kg/m^2.	Macular xanthophylls vs. placebo	13 or 27 mg/d	24	IL-6, TNF-α
Estévez-Santiago et al./2019/Spain (Estévez-Santiago et al. 2019)	Parallel RCT	46 Postmenopausal women; Mean age: 59 yrs Mean BMI: 24.7 kg/m^2	Lutein + anthocyanins vs. anthocyanins alone	6 mg/d lutein + 2 mg/d zeaxanthin	32	CRP, IL-6
Ghiasian et al./2019/Iran (Ghiasian et al. 2019)	Parallel RCT	40 Patients with multiple sclerosis; Mean age: 30.2 yrs; Mean BMI: Not reported.	Crocin vs. placebo	30 mg/d	8	TNF-α
Haidari et al./2019/Iran (Haidari et al. 2020)	Parallel RCT	46 Patients with NAFLD; Mean age: 37.1 yrs; Mean BMI: 33.25 kg/m^2	Study1: β-cryptoxanthin + energy-restricted high-protein diet vs. placebo + energy-restricted high-protein diet Study2: β-cryptoxanthin + energy-restricted normal protein diet vs. placebo + energy-restricted normal protein diet	6 mg/d	12	hs-CRP, IL-6
Chen et al./2017/Japan (Chen and Kotani 2017)	Parallel RCT	29 healthy climacteric women; Mean age: 52 yrs; Mean BMI: 21.6 kg/m^2	Astaxanthin vs. placebo	12 mg/d	12	hs-CRP
Nosrati et al./2017/Iran (M. Nosrati et al., 2018)	Parallel RCT	58 individuals with metabolic syndrome; Mean age: 41.1 yrs, Mean BMI: 34.1 kg/m^2.	Crocin vs. placebo	30 mg/d	8	hs-CRP
Nieman et al./2017/USA (Nieman et al. 2018)	Cross over RCT	20 Runners; Mean age: 37.35 yrs Mean BMI: Not reported.	Lycopene prior to running vs. placebo	11 mg/d	4	CRP, IL-6
Mousavi et al./2015/Iran (Mousavi et al. 2015)	Parallel RCT	61 Healthy men; Mean age: 48.5 yrs Mean BMI: Not reported.	Crocin vs. placebo	30 mg/d	12	CRP
Coombes et al./2015/Australia (Coombes, Sharman, and Fassett 2016)	Parallel RCT	58 Renal transplant recipients; Mean age: 49.9 yrs, Mean BMI: 26.8 kg/m^2	Astaxanthin vs. placebo	12 mg/d	48	CRP
Baralic et al./2015/Serbia (Baralic et al. 2015)	Parallel RCT	40 Trained male soccer players; Mean age: 17.75 yrs; Mean BMI: 22.3 kg/m^2	Astaxanthin vs. placebo	4 mg/d	12	hs-CRP
Ramezani et al./2014/Iran (Ramezani et al. 2014)	Parallel RCT	44 Type 2 diabetic patients; Mean age: 55.3 yrs Mean BMI: 28.4 kg/m^2	Carrot juice fortified with beta-carotene vs. normal carrot juice	10 mg/d	8	CRP, IL-6
Gajendragadkar et al./2014/UK (Gajendragadkar et al. 2014)	Parallel RCT	72 Patients with Cardiovascular Disease/Healthy Volunteers; Mean age: 64.8 yrs; Mean BMI: 27.2 kg/m^2	Lycopene vs. placebo	7 mg/d	8	hs-CRP, TNF-α, IL-6

Mohamadpour et al./2013 (Mohamadpour et al. 2013)		44 Healthy Volunteers; Mean age: 31.1 yrs; Mean BMI: 24.9 kg/m^2	Crocin vs. placebo	20 mg/d	4	hs-CRP
Wang et al./2013/China (Wang et al. 2013)	Parallel RCT	116 Healthy nonsmokers; Mean age: 55.1 yrs Mean BMI:23.4 kg/m^2.	Lutein vs. placebo	Study1: 10 mg/d Study2: 20 mg/d	12	CRP
Graydon et al./2012/UK (Graydon et al. 2012)	Parallel RCT	75 Healthy subjects; Mean age: 37.6 yrs Mean BMI: 25.1 kg/m^2	Lutein + zeaxanthin vs. placebo β-Carotene vs. placebo	Study1: 10 mg/d lutein + 5 mg/d zeaxanthin Study2: 15 mg/d β-Carotene	8	CRP
Kırkıl et al./2012/Turkey (Kirkil et al. 2012)	Parallel RCT	30 COPD patients; Mean age: 64.9 yrs Mean BMI: Not reported.	Lycopene vs. placebo	20 mg/d	16	IL-6, TNF-α
Thies et al./2012/Scotland (Thies et al. 2012)	Parallel RCT	144 Moderately overweight, disease-free, middle-aged adults; Mean age: 51.1 yrs; Mean BMI: 26.7 kg/m^2	Control diet supplemented with lycopene capsule vs. control diet	10 mg/d	16	hs-CRP, IL-6
Xu et al./2012/China (Xu et al. 2013)	Parallel RCT	65 Early atherosclerosis patients; Mean age: 57.0 yrs, Mean BMI: 25.7 kg/m^2	Lutein vs. placebo	20 mg/d	12	IL-6
Petyaev et al./2012/Russia (Petyaev et al. 2012)	Parallel RCT	40 Patients with prehypertension; Mean age: 51.6 yrs, Mean BMI: 27.5 kg/m^2	Study1: lycopene vs. placebo Study2: whey protein isolates embedded into lycopene micelles vs. whey protein	7 mg/d	4	CRP
Kim et al./2011/Korea (J. Y. Kim et al. 2011)	Parallel RCT	116 Healthy men; Mean age: 34.3 yrs; Mean BMI: 24.7 kg/m^2	Lycopene vs. placebo	Study1: 6 mg/d Study2: 15 mg/d	8	hs-CRP
Park et al./2010/USA (Park et al. 2010)	Parallel RCT	42 healthy adult female human subjects; Mean age: 21.5 yrs; BMI: 24.7 kg/m^2: 21.6	Astaxanthin vs. placebo	Study1: 2 mg/d Study2: 8 mg/d	8	CRP, IL-6, TNF-α
Karppi et al./2007/Finland (Karppi et al. 2013)	Parallel RCT	39 healthy nonsmoking men; Mean age: 24.4 yrs; BMI: 24.7 kg/m^2: 23.8	Astaxanthin vs. placebo	8 mg/d	12	hs-CRP, IL-6
Briviba et al./2004/Germany (Briviba et al. 2004)	Parallel RCT	55 Healthy smokers and nonsmokers ; Mean age: 34.2yrs, Mean BMI: Not reported	Tomato extract capsules vs. placebo	14.64 mg/d	2	TNF-α

RCT, randomized controlled trial; BMI, body mass index; CRP, C-reactive protein; hs-CRP, high sensitive c-reactive protein; IL-6, interleukin-6; TNF-α, tumor necrosis factor; MMT, methadone maintenance treatment; NAFLD, nonalcoholic fatty liver disease.

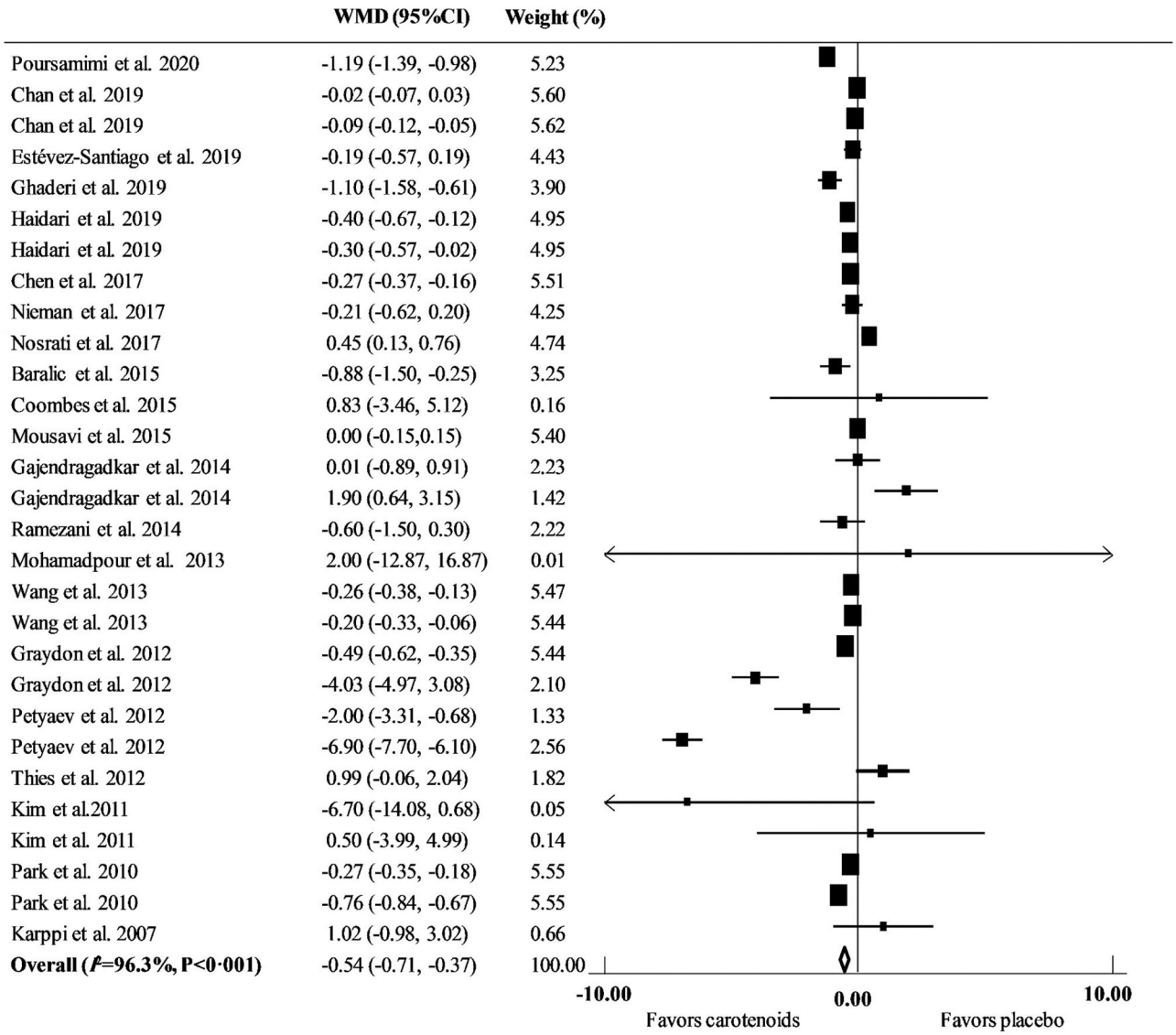

Figure 2. Forest plot of pooled estimate effects of carotenoids on CRP. Data are presented as weighted mean difference (WMD) between treatment and control groups with 95% CIs.

crossover design (Nieman et al. 2018). A total of 1481 participants aged 17–65 years were included in the meta-analysis from all 26 RCTs. Based on body mass index (BMI) values, 9 studies included overweight population (Chan, Chen, and Chen 2019; Coombes et al., 2016; Gajendragadkar et al. 2014; Graydon et al. 2012; Petyaev et al. 2012; Poursamimi et al. 2020; Ramezani et al. 2014; Thies et al. 2012; Xu et al. 2013), 2 studies were conducted on obese (Haidari et al. 2020; Mina Nosrati et al. 2018), 5 studies didn't report BMI (Briviba et al. 2004; Ghiasian et al. 2019; Kirkil et al. 2012; Mousavi et al. 2015; Nieman et al. 2018), and remaining included subjects within healthy range of BMI. Twelve of included RCTs were carried out in the healthy subjects (Briviba et al. 2004; Chen and Kotani 2017; Estévez-Santiago et al. 2019; Graydon et al. 2012; Karppi et al. 2013; J. Y. Kim et al. 2011; Mohamadpour et al. 2013; Mousavi et al. 2015; Park et al. 2010; Stringham, Holmes, and Stringham 2019; Thies et al. 2012; Wang et al. 2013) and two in athletes (Baralic et al. 2015; Nieman et al. 2018). Other RCTs conducted in type 2 diabetes mellitus (Chan, Chen, and Chen 2019; Ramezani et al. 2014), metabolic syndrome (M. Nosrati et al. 2018), osteoarthritis patients (Poursamimi et al. 2020), patients with multiple sclerosis (Ghiasian et al. 2019), nonalcoholic fatty liver disease (Haidari et al. 2020), CVD (Gajendragadkar et al. 2014), Chronic obstructive pulmonary disease (Kirkil et al. 2012), early atherosclerosis (Xu et al. 2013), prehypertension (Petyaev et al. 2012), renal transplant recipients (Coombes, Sharman, and Fassett 2016), moderately overweight (Thies et al. 2012) and patients undergoing methadone maintenance therapy MMT (Ghaderi et al. 2019). Lycopene with 7 RCTs and astaxanthin and crocin each with 6 trials, comprised the largest number of RCTs, followed by lutein and/or zeaxanthin with 5 trials, β-carotene with 2 trials, and β-cryptoxanthin with 1 trial. Daily dosage ranged from 2 to 30 mg/d (lycopene 6–20 mg, ataxanthin 2–12 mg, crocin 15–30 mg, lutein/zeaxanthin 8–20 mg, β-Carotene 10–15 mg, and β-cryptoxanthin 6 mg). The duration of supplementation ranged from 1 to 12 months. Carotenoids were used as purified form or carotenoid-rich extract, all of which were

Table 2. Subgroup analyses for the effects of carotenoids on inflammatory cytokines in the participants of included RCTs.

		Meta-analysis			Heterogeneity	
	Effect sizes, *n*	WMD (95% CI)	*P* within group	*P* between group	I^2	*P*-heterogeneity
Subgroup analysis for CRP						
Overall	29	−0.54 (−0.71, −0.37)	**<0.001**		96.3	<0.001
Intervention duration						
<12 weeks	17	−0.78 (−1.03, −0.52)	**<0.001**	**<0.001**	97.5	<0.001
≥12 weeks	12	−0.32 (−0.54, −0.10)	**<0.001**		89.5	<0.001
Dosage						
≤10 mg/d	16	−0.61 (−0.92, −0.31)	**<0.001**	**<0.001**	97.0	<0.001
>10 mg/d	13	−0.49 (−0.73, −0.25)	**<0.001**		95.0	<0.001
Health status						
Healthy individuals	17	−0.39 (−0.60, −0.18)	**<0.001**	**<0.001**	91.1	<0.001
Individuals with impaired health	12	−0.72 (−0.99, −0.46)	**<0.001**		97.5	<0.001
Participants' age						
<50	15	−0.51 (−0.76, −0.26)	**<0.001**	**<0.001**	90.7	<0.001
≥50	14	−0.57 (−0.81, −0.33)	**<0.001**		96.6	<0.001
Subgroup analysis for IL-6						
Overall	16	−0.54 (−1.01, −0.06)	**0.025**		82.2	<0.001
Intervention duration						
<12	8	−0.15 (−0.79, 0.47)	0.626	0.095	73.5	0.007
≥12	8	−1.14 (−2.11, −0.17)	**0.021**		87.3	<0.001
Dosage						
≤10 mg/d	11	−0.24 (−0.69, 0.20)	0.278	0.086	53.6	0.017
>10 mg/d	5	−1.72 (−3.18, −0.26)	**0.020**		93.6	<0.001
Health status						
Healthy individuals	9	−0.13 (−0.61, 0.35)	0.597	0.140	67.1	0.002
Individuals with impaired health	7	−1.11 (−2.09, −0.14)	**0.024**		88.0	<0.001
Participants' age						
<50	7	−0.16 (−0.95, 0.62)	0.675	0.059	72.9	0.001
≥50	9	−0.93 (−1.73, −0.12)	**0.023**		86.2	<0.001
Subgroup analysis for TNF-α						
Overall	11	−0.97 (−1.98, 0.03)	0.059		85.7	<0.001
Intervention duration						
<12	9	−0.67 (−1.68,0.34)	0.195	0.172	82.7	<0.001
≥12	2	−6.94 (−20.7, 6.81)	0.323		95.8	<0.001
Dosage						
≤10 mg/d	5	0.26 (−0.31, 0.83)	0.368	0.996	0.0	0.62
>10 mg/d	6	−2.02 (−3.62, −0.43)	**0.013**		91.5	<0.001
Health status						
Healthy individuals	5	−0.41 (−1.44, 0.61)	0.426	0.208	79.7	<0.001
Individuals with impaired health	6	−1.58 (−3.92, 0.76)	0.186		90.6	<0.001
Participants' age						
<50	6	−0.75 (−1.96, 0.45)	0.219	0.071	89.2	<0.001
≥50	5	−1.85 (−4.18, 0.48)	0.120		82.8	<0.001

CRP, C-reactive protein; IL-6, interleukin-6; RCT, randomized controlled trial; TNF-α, tumor necrosis factor.
Data are pooled weighted mean differences (95% CIs) by a random-effects model.

provided as capsules or tablets. Eight RCTs were included in the analysis twice: four compared various dosages of supplements (Chan, Chen, and Chen 2019; J. Y. Kim et al. 2011; Park et al. 2010; Wang et al. 2013); two compared the effect of one carotenoid with two different control groups (Haidari et al. 2020; Petyaev et al. 2012); one assessed the impact of carotenoid supplement separately on patients with CVD and healthy subjects (Gajendragadkar et al. 2014); and one used two distinct carotenoids as intervention (Graydon et al. 2012). Only 16 out of 26 studies measured compliance rates using pill counting (Ghaderi et al. 2019; M. Nosrati et al. 2018), serum concentration of carotenoids (Briviba et al. 2004; Graydon et al. 2012; Kirkil et al. 2012; Ramezani et al. 2014; Thies et al. 2012), or both pill counting and measuring serum carotenoid level (Chan, Chen, and Chen 2019; Coombes, Sharman, and Fassett 2016; Gajendragadkar et al. 2014; Haidari et al. 2020; J. Y. Kim et al. 2011; Nieman et al. 2018; Stringham, Holmes, and Stringham 2019; Wang et al. 2013; Xu et al. 2013). Compliance rates measured by pill counting were reported around 90% for most of the studies (Coombes, Sharman, and Fassett 2016; Gajendragadkar et al. 2014; Ghaderi et al. 2019; Haidari et al. 2020; Wang et al. 2013; Xu et al. 2013). Meanwhile, six articles measured serum lycopene level (Briviba et al. 2004; Gajendragadkar et al. 2014; J. Y. Kim et al. 2011; Kirkil et al. 2012; Nieman et al. 2018; Thies et al. 2012), two measured serum astaxanthin level (Chan, Chen, and Chen 2019; Coombes, Sharman, and Fassett 2016), four measured serum lutein/zeaxanthin level (Graydon et al. 2012; Stringham, Holmes, and Stringham 2019; Wang et al. 2013; Xu et al. 2013), two measured serum β-carotene level (Graydon et al. 2012; Ramezani et al. 2014), and one measured serum β-cryptoxanthin level (Haidari et al. 2020) to evaluate adherence rates with study protocol and all reported significant increases in serum levels of relevant carotenoid compared to controls. Only 11 studies evaluated side effects during the study period, all of which did not report serious side effects (Briviba et al. 2004; Coombes, Sharman, and Fassett 2016; Gajendragadkar et al. 2014; Ghaderi et al. 2019; Haidari et al. 2020; Karppi et al. 2013; J. Y. Kim et al. 2011;

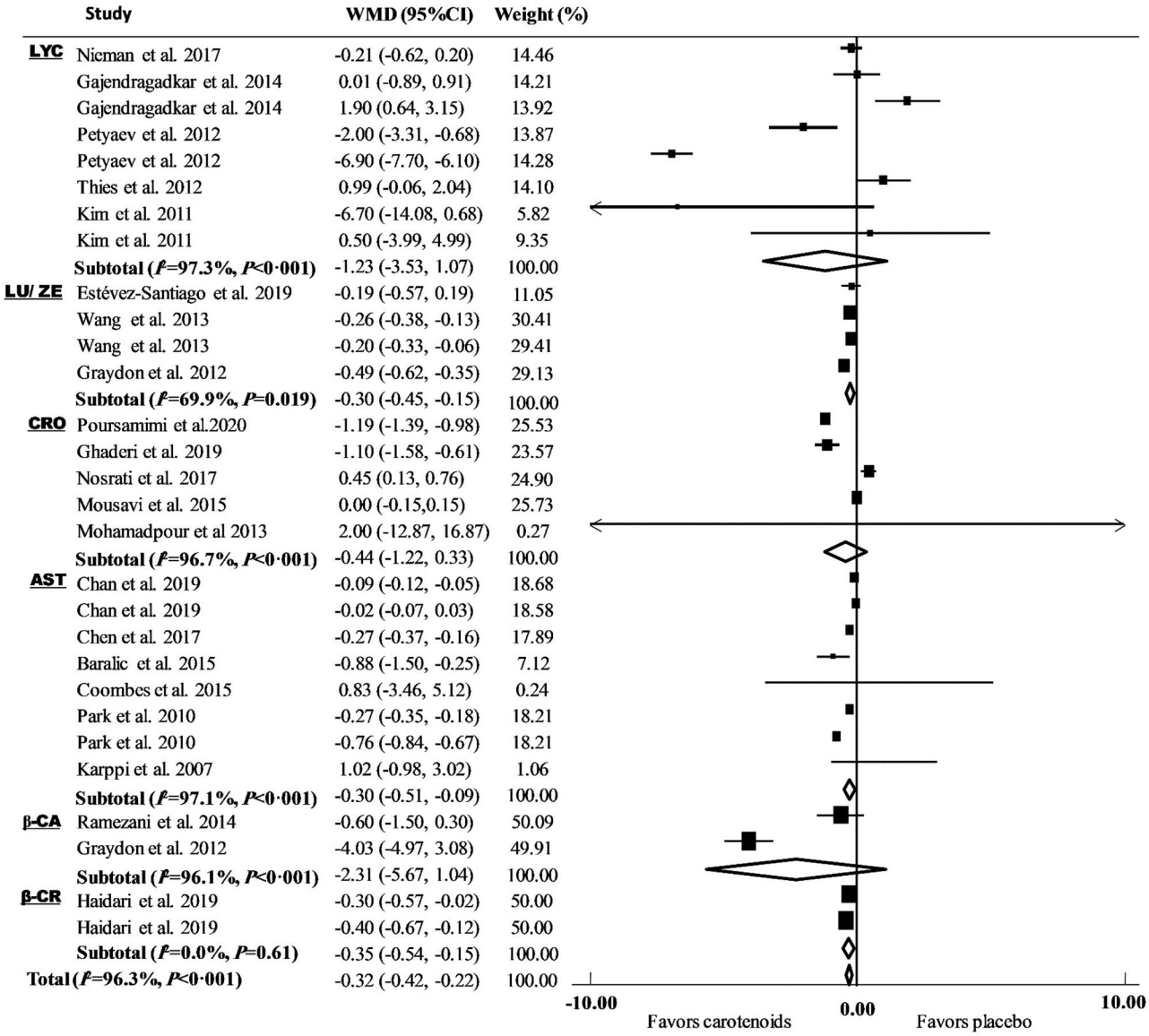

Figure 3. Forest plot of pooled estimate effects of carotenoids on CRP, as stratified by the type of carotenoids (AST, astaxanthin; β-CA, β-carotene; β-CR, β-cryptoxanthin; CRO, crocin; LU/ZE, lutein/zeaxanthin; LYC, lycopene). Data are presented as weighted mean difference (WMD) between treatment and control groups with 95% CIs.

Mohamadpour et al. 2013; Mousavi et al. 2015; Wang et al. 2013; Xu et al. 2013). These 26 RCTs originated from the Iran (n = 8), USA (n = 3), UK (n = 2), China (n = 2), and single studies from Russia, Japan, Turkey, Australia, Taiwan, Germany, Spain, Scotland, Finland, Korea and Serbia.

3.2. Meta-analysis

3.2.1. Effect on plasma level of CRP

A total of 29 WMD from 21 RCTs, including 1365 subjects (708 treated and 657 controls) provided data on the effect of carotenoids on plasma level of CRP. The quantitative meta-analysis indicated a significant decrease in CRP level in subjects receiving carotenoids supplements compared with controls (Figure 2; WMD: −0.54 mg/L, 95% CI: −0.71, −0.37, $P < 0.001$). Considering I^2 index (96.3%) and Cochrane Q test ($P < 0.001$), a high inter-trial heterogeneity was detected. A sensitivity analysis revealed that excluding each of the studies had no significant effect on the pooled effect size. The results from subgroup analyses showed that the high heterogeneity among the studies was due to the type and dosage of carotenoids, intervention duration, participants' age, and health status. Pooling change from the trials with intervention duration of <12 weeks, studies that prescribed ≤10 mg/day carotenoids, individuals with impaired health subjects, and subjects aged ≥50 years resulted in significant reductions of CRP when compared with their counterparts (Table 2). When the effect of individual carotenoids on CRP level was assessed, the association was only significant for astaxanthin (Figure 3; WMD: −0.30 mg/L, 95% CI: −0.51, −0.09, $P = 0.005$), lutein and/or zeaxanthin (WMD: −0.30 mg/L, 95% CI: −0.45, −0.15, $P < 0.001$), and β-cryptoxanthin (WMD: −0.35 mg/L, 95% CI: −0.54, −0.15, $P < 0.001$). High heterogeneity was found for astaxanthin (based on I^2=97.1% and $P < 0.001$) and moderate for lutein and/or zeaxanthin (based on I^2=69.9% and $P = 0.019$). Considering

Table 3. Subgroup analyses for the effects of carotenoids on serum levels of CRP in the participants of included RCTs.

	Meta-analysis				Heterogeneity	
	Effect sizes, *n*	WMD (95% CI)	*P* within group	*P* between group	I^2	*P*-heterogeneity
Lycopene						
Overall	8	−1.23 (−3.53, 1.07)	0.295		97.3	<0.001
Intervention duration						
<12 weeks	7	−1.60 (−4.22, 1.01)	0.229	0.210	97.6	<0.001
≥12 weeks	1	0.99 (−0.06, 2.04)	0.066		0.0	–
Dosage						
≤10 mg/d	6	−0.98 (−4.22, 2.25)	0.551	0.372	97.8	<0.001
>10 mg/d	2	−2.36 (−8.35, 3.62)	0.439		66.1	0.086
Health status						
Healthy individuals	5	0.57 (−0.70, 1.84)	0.379	0.611	75.0	0.003
Individuals with impaired health	3	−2.97 (−7.56, 1.62)	0.205		98.4	<0.001
Participants' age						
<50	3	−0.65 (−3.04, 1.72)	0.589	0.408	34.5	0.217
≥50	5	−1.21 (−4.71, 2.29)	0.498		98.2	<0.001
Astaxanthin						
Overall	8	−0.30 (−0.51, −0.09)	**0.005**		97.1	<0.001
Intervention duration						
<12	4	−0.28 (−0.54, −0.02)	**0.032**	**0.011**	98.7	<0.001
≥12	4	−0.37 (−0.89, 0.14)	0.158		44.7	0.143
Dosage						
≤10 mg/d	5	−0.38 (−0.78,−0.001)	**0.011**	**0.010**	98.1	<0.001
>10 mg/d	3	−0.17 (−0.33,−0.002)	**0.047**		79.3	0.008
Health status						
Healthy individuals	6	−0.45 (−0.76, −0.14)	**0.004**	**0.025**	93.8	<0.001
Individuals with impaired health	2	−0.05 (−0.12, 0.01)	0.101		79.8	0.026
Participants' age						
<50	4	−0.53 (−0.94, −0.11)	**0.013**	**0.005**	95.4	<0.001
≥50	4	−0.11 (−0.20, −0.01)	**0.021**		83.0	0.001
Lutein and/or zeaxanthin						
Overall	4	−0.30 (−0.45, −0.15)	**<0.001**		69.9	0.019
Intervention duration						
<12	1	−0.49 (−0.62, −0.35)	**<0.001**	**<0.001**	0.0	–
≥12	3	−0.23 (−0.32, −0.14)	**<0.001**		0.0	0.800
Dosage						
≤10 mg/d	2	−0.20 (−0.32, −0.07)	**0.002**	**<0.001**	0.0	0.960
>10 mg/d	2	−0.37 (−0.60, −0.14)	**0.001**		82.7	0.016
Participants' age						
<50	1	−0.49 (−0.62, −0.35)	**<0.001**	**<0.001**	0.0	–
≥50	3	−0.23 (−0.32, −0.14)	**<0.001**		0.0	0.800
Crocin						
Overall	5	−0.44 (−1.22, 0.33)	0.260		96.7	<0.001
Intervention duration						
<12	3	−0.29 (−1.77, 1.19)	0.701	0.307	92.7	<0.001
≥12	2	−0.59 (−1.75, 0.57)	0.319		98.8	<0.001
Health status						
Healthy individuals	1	2.0 (−12.8, 16.88)	0.792	0.262	0.0	–
Individuals with impaired health	4	−0.45 (−1.23, 0.32)	0.255		97.5	<0.001
Participants' age						
<50	4	−0.17 (−0.80, 0.45)	0.581	**<0.001**	89.0	<0.001
≥50	1	−1.19 (−1.39, −0.98)	**<0.001**		0.0	–

CRP, C-reactive protein; RCT, randomized controlled trial.
Data are pooled weighted mean differences (95% CIs) by a random-effects model.

lycopene, when we restricted the analysis to the RCTs that assessed levels of compliance based on plasma concentration of lycopene (Gajendragadkar et al. 2014; J. Y. Kim et al. 2011; Nieman et al. 2018; Thies et al. 2012), pooled WMD in subjects being in the highest quartile of lycopene (Number of effect size: 6, WMD: 0.34, 95%CI: −0.83, 1.51, $P = 0.568$) didn't show significant decrease in CRP level compared with those in the lowest quartile (WMD: 0.20, 95%CI: −1.74, 2.14, $P = 0.839$). As Table 3 shows, astaxanthin had significant effect on CRP in subgroups of dosages ≤10 mg/day, intervention duration <12 weeks, healthy individuals and ages <50 years. Subgroup analysis also revealed that lutein and/or zeaxanthin supplementation substantially decreased CRP level in subgroups of dosages ≤10 mg/day, intervention duration <12 weeks, and ages <50 years. Although, crocin did not show a significant overall effect on CRP, in subgroups of studies conducted in subjects aged ≥50 years, it is demonstrated a significant reduction in CRP levels. Although, visual inspection of the funnel plot showed evidence of asymmetry, no publication bias was found among studies based on Begg's ($P = 0.721$), Egger's ($P = 0.055$) tests.

3.2.2. Effect on plasma level of IL-6

The pooled analysis of 16 effect sizes from 12 RCTs, with 714 subjects (374 treated and 340 controls) revealed a significant decrease in IL-6 level following carotenoids

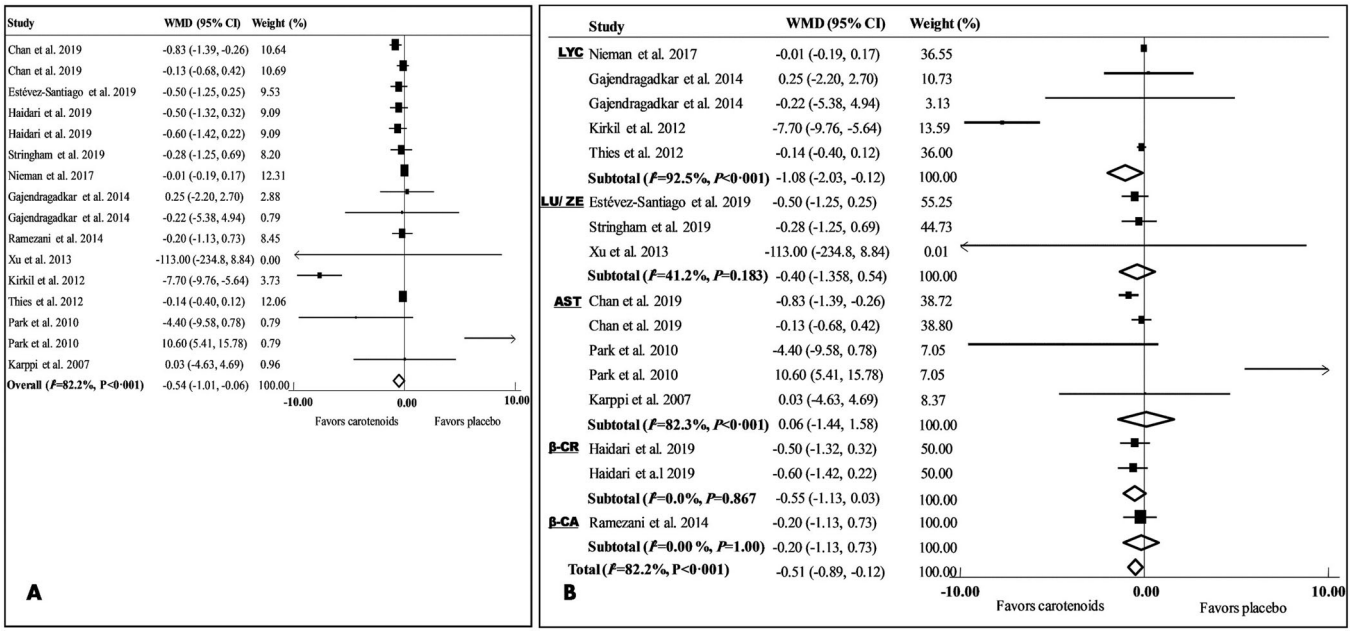

Figure 4. Forest plot of pooled estimate effects of carotenoids on IL-6. (A) Overall effect of carotenoids; and (B) stratified by the type of carotenoids (AST, astaxanthin; β-CA, β-carotene; β-CR, β-cryptoxanthin; LU/ZE, lutein/zeaxanthin; LYC, lycopene). Data are presented as weighted mean difference (WMD) between treatment and control groups with 95% CIs.

supplementation (Figure 4; WMD: −0.54 pg/mL, 95% CI: −1.01, −0.06, $P = 0.025$). The I^2 index (82.2%) and Cochrane Q test ($P < 0.001$) revealed a high inter-trial heterogeneity. Further a sensitivity analysis showed that the effect of the carotenoids on IL-6 level largely depended on the study conducted by Kirkil et al.; after exclusion of this study, the pooled WMD turned insignificant (WMD: −0.28 pg/mL, 95% CI: −0.58 to 0.02, $P = 0.076$). Subgroup analyses based on dosage of carotenoids, intervention duration, participants' age and health status could not change the overall effect of carotenoid on IL-6 level (Tables 2 and 4). However, subgrouping based on carotenoid type resulted in a significant decrease in IL-6 level following lycopene supplementation (Figure 4; WMD: −1.08 pg/ml, 95%CI: −2.03, −0.12, $P = 0.027$), irrespective of dosage, intervention duration, health status, and age. Moreover, astaxanthin supplementation significantly reduced IL-6 level at dosage >10 mg/d (Table 4). Regarding lycopene, when we restricted the analysis to the RCTs that evaluated participants' compliance to study protocol by measuring plasma level of lycopene (Gajendragadkar et al. 2014; Kirkil et al. 2012; Nieman et al. 2018; Thies et al. 2012), pooled WMD in subjects being in the highest quartile of lycopene (Number of effect size: 5, WMD: 0.0 pg/ml, 95%CI: −0.31, 0.31, $P = 0.979$) didn't show any significant decrease in IL-6 level compared with those in the lowest quartile (WMD: −2.63 pg/ml, 95%CI: −8.33, 3.06, $P = 0.365$). Visual inspection of the funnel plot did not show evidence of asymmetry. Furthermore, the results from Begg's ($P = 0.558$) and Egger's ($P = 0.198$) tests didn't confirm the likely of publication bias across studies.

3.2.3. Effect on plasma level of TNF-α

A total of 12 effect sizes from 8 RCTs, including 375 subjects (206 treated and 169 controls) evaluated the effect of carotenoids on plasma level of TNF-α. The pooled analysis of these studies showed that administration of carotenoids caused a non-significant decrease in TNF-α level (Figure 5; WMD: −0.97 pg/ml, 95% CI: −1.98, 0.03, $P = 0.0.059$). Again, a high inter-trial heterogeneity was observed based on I^2 index = 85.7% and $P < 0.001$. A sensitivity analysis revealed that two effect sizes from one RCT conducted by Park et al. had the largest influence on TNF-α level. After exclusion of the effect size related to minimum (WMD: −1.17 pg/mL, 95% CI: −2.30 to −0.05, $P = 0.041$) and maximum (WMD pg/mL: −1.21, 95% CI: −2.30 to −0.12, $P = 0.029$) dosage of astaxanthin used in Park et al. study, the reduction in TNF-α level remained significant as compared to that from the main analysis. In stratified analyses, there was no evidence of variation between subgroups of studies based on type or dosage of carotenoids, intervention duration, participants' age, and health status (Tables 2 and 4). Regarding lycopene, when we restricted the analysis to the RCTs that assessed levels of compliance based on plasma concentration of lycopene (Briviba et al. 2004; Gajendragadkar et al. 2014; Kirkil et al. 2012), pooled WMD in subjects being in the highest quartile of lycopene (Number of effect size: 5, WMD: −0.43, 95%CI: −1.06, 0.20, $P = 0.887$) didn't show significant decrease in TNF-α concentration compared with those in the lowest quartile (WMD: −4.79, 95%CI: −14.04, 4.46, $P = 0.311$). In contrary to visual inspection of the funnel plot that showed evidence of asymmetry, there was no publication bias among studies based on Begg's ($P = 0.073$) and Egger's ($P = 0.134$) tests.

3.3. Assessment of the quality and risk of bias

The risk of bias in individual studies was shown in Table 1, supplementary material. Twenty-five of the 26 RCTs provided sufficient data on random sequence generation; 13

Table 4. Subgroup analyses for the effects of carotenoids on serum levels of IL-6 and TNF-α in the participants of included RCTs.

	Meta-analysis				Heterogeneity	
	Effect sizes, *n*	WMD (95% CI)	*P* within group	*P* between group	I^2	*P*-heterogeneity
Subgroup analysis for IL-6						
Lycopene						
Overall	5	−1.08 (−2.03, −0.12)	**0.027**		92.5	<0.001
Intervention duration						
<12 weeks	3	−0.009 (−0.18, 0.17)	0.923	0.903	0.0	0.976
≥12 weeks	2	−3.84 (−11.25, 3.55)	0.309		98.0	<0.001
Dosage						
≤10 mg/d	3	−0.13 (−0.40, 0.13)	0.315	0.299	0.0	0.953
>10 mg/d	2	−3.78 (−11.31, 3.75)	0.325		98.1	<0.001
Health status						
Healthy individuals	3	−0.05 (−0.20, 0.09)	0.504	0.492	0.0	0.729
Individuals with impaired health	2	−3.75 (−11.54, 4.03)	0.345		95.7	<0.001
Participants' age						
<50	1	−0.01 (−0.19, 0.17)	0.913	0.877	0.0	–
≥50	4	−2.05 (−6.05, 1.93)	0.313		94.1	<0.001
Astaxanthin						
Overall	5	0.06 (−1.44, 1.58)	0.930		82.3	<0.001
Intervention duration						
<12	4	0.09 (−1.53, 1.72)	0.907	0.909	86.7	<0.001
≥12	1	0.03 (−4.63, 4.69)	0.990		0.0	–
Dosage						
≤10 mg/d	4	1.36 (−3.26, 5.98)	0.564	**0.005**	84.2	<0.001
>10 mg/d	1	−0.83 (−1.39, −0.26)	**0.004**		0.0	–
Health status or participants' age						
Healthy individuals or age <50	4	2.06 (−6.39, 10.51)	0.633	0.186	88.3	<0.001
Individuals with impaired health or age ≥50	1	−0.47 (−1.16, 0.20)	0.172		0.0	–
Subgroup analysis for TNF-α						
Lycopene						
Overall	5	−1.83 (−4.10, 0.42)	0.112		82.8	<0.001
Intervention duration						
<12	4	−0.41 (−1.04, 0.21)	0.198	0.066	0.0	0.992
≥12	1	−14.25(−19.8,−8.66)	**<0.001**		0.0	–
Dosage						
≤10 mg/d	2	0.006 (−3.07, 3.09)	0.997	0.180	0.0	0.927
>10 mg/d	3	−2.78 (−5.72, 0.15)	0.066		91.3	<0.001
Participants' age						
<50	2	−0.43 (−1.07, 0.21)	0.188	0.166	0.0	0.887
≥50	3	−4.70 (−13.69, 4.28)	0.305		88.5	<0.001
Astaxanthin						
Overall	4	0.10 (−0.47, 0.68)	0.722		23.1	0.272
Dosage						
≤10 mg/d	3	0.27 (−0.39, 0.93)	0.425	0.808	23.0	0.273
>10 mg/d	1	−0.42 (−1.45, 0.61)	0.427		0.0	–
Health status or participants' age						
Healthy individuals or age <50	2	0.58 (−0.13, 1.29)	0.110	0.684	0.0	0.524
Individuals with impaired health or age ≥50	2	−0.39 (−1.11, 0.33)	0.293		0.0	0.935

IL-6, interleukin-6; RCT, randomized controlled trial; TNF-α, tumor necrosis factor.
Data are pooled weighted mean differences (95% CIs) by a random-effects model.

RCTs clarified allocation concealment; 22 RCTs conducted blinding of the participants and researchers; in 7 RCTs, outcome assessors were blinded to the study protocol as well; 23 RCTs elucidated the completeness of outcome data such as attrition and exclusions from the analysis; 16 RCTs reported all outcomes measured; and 14 RCTs addressed variations in BMI, dietary intake during the intervention or differences in baseline values of outcomes as confounding factors in statistical analyses. Based on judgments of risk of bias for each item, 20 studies were classified as "high" and 3 studies (Park et al. 2010; Ramezani et al. 2014; Stringham, Holmes, and Stringham 2019) as "unclear" risk of bias. Only 3 studies (Gajendragadkar et al. 2014; Haidari et al. 2020; Nieman et al. 2018) could be classified as high quality because of being all key domains at low risk of bias.

4. Discussion

This systematic review and meta-analysis assessed the effectiveness of carotenoids supplementation on serum levels of inflammatory mediators, including CRP, IL-6, and TNF-α for the first time. In the overall assessment, carotenoids reduced CRP level compared with control groups, which was highly stable to sensitivity analysis. Carotenoids also showed significant beneficial effects on IL-6 level, but non-significant decrease in serum level of TNF-α, which both were less stable to sensitivity analysis. For the individual carotenoids, astaxanthin, lutein with or without zeaxanthin, and β-cryptoxanthin resulted in a significant decrease in CRP level, however, the effects from lycopene, crocin, and β-carotene were not statistically significant. Besides, lycopene was the only carotenoid that led to a significant reduction in IL-6 level.

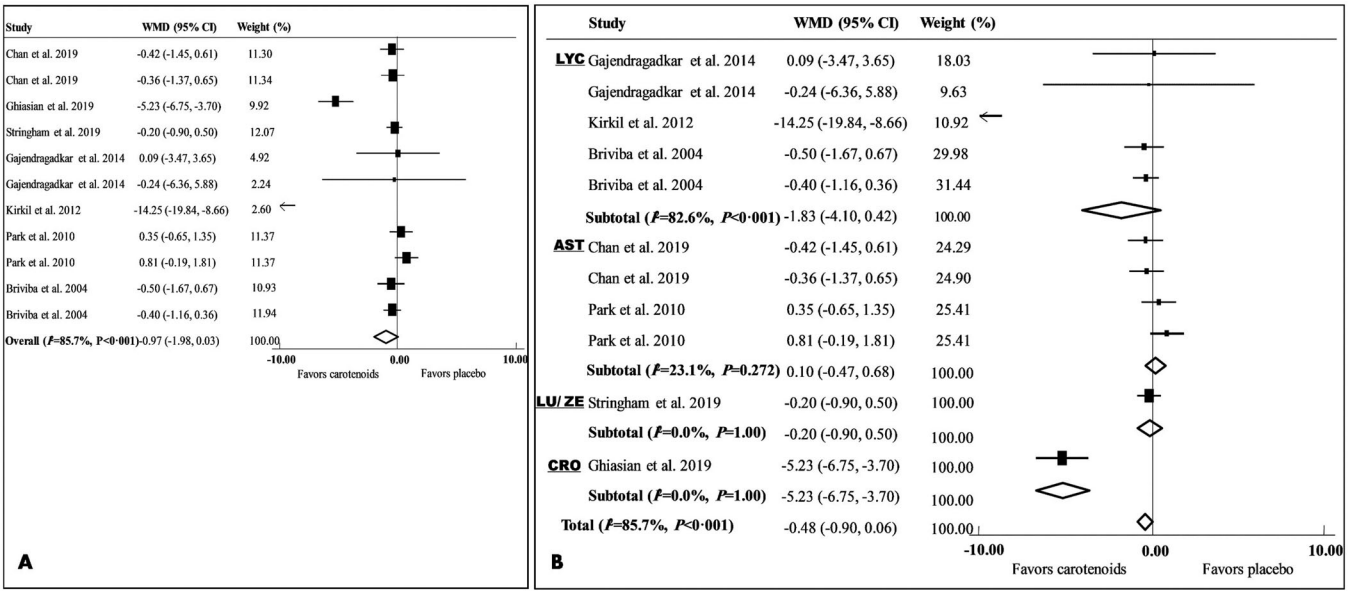

Figure 5. Forest plot of pooled estimate effects of carotenoids on TNF-α. (A) Overall effect of carotenoids; and (B) stratified by the type of carotenoids (AST, astaxanthin; CRO, crocin; LU/ZE, lutein/zeaxanthin; LYC, lycopene). Data are presented as weighted mean difference (WMD) between treatment and control groups with 95% CIs.

IL-6, a pro-inflammatory cytokine mainly released from macrophages and T cells, has been well known to trigger hepatic production of CRP. Thus, concentrations of CRP and IL-6 are physiologically interrelated. Serum concentrations of CRP and IL-6 may independently predict cardiovascular events and all-cause mortality (Baune et al. 2011; Danesh et al. 2008; Morrow and Ridker 2000; Shah et al. 2009). For instance, evidence from Women's Health Study suggested that elevated levels of CRP or IL-6 independently associated with the incidence of CVD (Volpato et al. 2001). In a meta-analysis conducted by the US Preventive Services Task Force was also found 60% higher risk of incident coronary heart disease in the individuals with CRP levels more than 3 mg/L compared to levels less than 1 mg/L (Buckley et al. 2009).

In the analyses on TNF-α, the studies with the lowest baseline levels of TNF-α gave the lowest responses to carotenoid supplementation and also affected the overall WMD. Thus, a sensitivity analysis was performed and observed that the overall WMD was statistically significant after removal of the study by Park et al. (2010) in which subjects had lower level of TNF-α at baseline than other subjects. The reduction in IL-6 level was non-significant, after excluding the study conducted by Kirkil et al. (2012) in which baseline values of IL-6 were much more than other studies included in the meta-analysis. Therefore, baseline values of IL-6 and TNF-α may determine the effectiveness of carotenoids on post-intervention levels of these biomarkers; meaning that the higher the baseline values of pro-inflammatory cytokines, the higher response to the carotenoid supplementation may be occurred.

Based on subgroup analyses, carotenoids totally decreased CRP levels much more effectively at the dosage of ≤10 mg/d, intervention duration of <12 weeks, and studies that recruited subjects aged ≥50 years or with impaired health status than comparators. When the analyses were conducted based on dosage, intervention duration, health status and participants' age, there was no change on the response of IL-6 and TNF-α to the carotenoids. However, it seems that effectiveness of carotenoids in diverse subgroups varies depending on the type of carotenoids which is discussed in the next sections.

The evidence supporting a protective association with inflammatory biomarkers is more potent for total carotenoids than for each individual carotenoid, as numerous observational studies showed inverse association between plasma level of total carotenoids and CRP (Beydoun et al. 2012; Karppi et al. 2013; Mohsen Mazidi et al. 2018; Lu Wang et al. 2008). For individual carotenoids, however, the findings were mixed. In the present meta-analysis, astaxanthin consumption could significantly decrease CRP levels. Contrary to our results, a previous meta-analysis involving fewer RCTs than the present study reported no significant changes in CRP level following astaxanthin supplementation (Xia et al. 2020). Like overall carotenoids, astaxanthin decreased CRP level in the dosage of ≤10 mg/d. The dosage of astaxanthin used in the trials included in this meta-analysis ranged between 2–12 mg/d which was much lower than the average dosage of other carotenoids used in the trials. For CRP, the most beneficial effects of astaxanthin were observed at the 2–6 mg/d and not at higher intakes. This suggests that, to possess a significant decrease in serum levels of CRP after astaxanthin supplementation, there is likely a minimum dose that must be considered. Likewise, a clinical trial comparing the dosages of 5 and 20 mg/d of astaxanthin, recommended daily dose of 5 mg of astaxanthin as an optimal dose to decrease the oxidative stress (Choi et al. 2011). Unlike CRP, for IL-6 level, beneficial effects were observed at only the higher dosages of astaxanthin. This may be due to chance, because there were less studies in subgroup analyses of astaxanthin for IL-6 than CRP. Astaxanthin also exhibited more benefit on CRP levels in

the healthy individuals, subgroups of ages <50 years and intervention duration <12 weeks.

In line with our recent systematic review in which most studies highlighted protective effects of lutein consumption against inflammatory mediators (Hajizadeh-Sharafabad et al. 2019), this meta-analysis indicated that lutein/zeaxanthin supplementation significantly reduced CRP levels. The clinical trials used purified lutein at doses 5 to 10 times higher than regular dietary intake doses (<2mg/d) at which serum concentrations of lutein can reach the levels required to play an anti-inflammatory role. In support of this claim, the present meta-analysis showed that doses of >10 mg/d lutein decreased plasma level of CRP, whereas doses of ≤10 mg/d did not. Additionally, in subgroup analysis, lutein showed potential for reducing CRP in subgroups of ages <50 years and intervention duration <12 weeks.

The variation in the dose-dependent effect between astaxanthin and lutein may come from the fact that lutein content of regular diets is much more than astaxanthin; therefore, the higher dosage of lutein may be required to exert a potent impact on inflammation. Moreover, possible explanation for the observed beneficial effects following astaxanthin and lutein/zeaxanthin supplementation in subgroups of ages <50 years is that a majority of elderly individuals had different chronic disorders likely to be undergoing pharmacotherapy which would make it hard to reach additional reductions in inflammatory biomarkers through dietary interventions. Moreover, considering higher baseline level of CRP in subjects aged <50 years, the most pronounced effects on the CRP in this subgroup, implies that the baseline serum level of CRP is likely involved in the response to astaxanthin and lutein/zeaxanthin supplementation. Although, such analyses should be treated with caution due to limited number of trials available for subgrouping.

The only available RCT investigating anti-inflammatory effects of β-cryptoxanthin showed a significant reduction in serum levels of CRP, but not IL-6 (Haidari et al. 2020). For lycopene supplementation, significant reduction was observed in IL-6 level. Unexpectedly, the subgroup analysis did not influence the response of subjects to lycopene supplementation. When the analysis was restricted to the studies reporting results for blood concentration of lycopene, no significant association was observed between blood levels of lycopene in treatment group and inflammatory mediators. Previous meta-analysis of RCTs assessing the effects of tomato and lycopene supplementation on inflammatory markers showed that neither tomato nor lycopene consumption did not affect CRP level; however, tomato but not lycopene intervention significantly reduced IL-6 level (Cheng et al. 2017). A large cohort study found that participants being in the highest quartile of lycopene and tomato consumption had lower CRP levels compared with those in the lowest quartile (M. Mazidi et al. 2020); although, residual confounding as a limitation of all observational studies may bias the results. In the present study, β-carotene supplementation did not affect inflammatory mediators; however, the number of RCTs was too small to draw a definite conclusion about anti-inflammatory role of β-carotene. Contrary to the clinical evidence, the observational studies tended to favor a protective effect of β-carotene against inflammation (Erlinger et al. 2001; Hu et al. 2004; Jing et al. 2018; Suzuki et al. 2010). Since the serum levels of carotenoids are often considered biomarkers of fruit and vegetable consumption or a healthier lifestyle, causality of these inverse associations cannot be confirmed (Gallicchio et al. 2008).

In overall estimate, crocin did not change inflammatory biomarkers significantly, while in subgroup analysis, this carotenoid showed potential for lowering CRP level in studies that recruited participants with ≥50 years old. In a recent meta-analysis, Asbaghi et al. analyzed data of clinical trials on the effect of saffron, as one of the richest source of crocin on inflammatory markers and found no significant changes in serum levels of CRP, IL-6, and TNF-α (Asbaghi et al. 2020).

It is well known that the presence of intracellular redox state strongly potentiates the inflammatory processes (Salzano et al. 2014). The ability of carotenoids to quench reactive oxygen species (ROS) may be considered the most underlying mechanism through which these compounds mitigate inflammatory response (Kaulmann and Bohn 2014). The antioxidant activity of carotenoids mainly depends on conjugated double bonds as well as the position and number of functional groups of carotenoids. The unique chemical features of astaxanthin including placing keto groups in the fourth and fourth prime positions in cyclohexene structure, and hydroxyl groups at the third and third prime position, make it superior to other carotenoids (Martínez-Delgado, Khandual, and Villanueva–Rodríguez 2017). More importantly, contrary to nonpolar carotenoids (i.e. β-carotene and lycopene) which are located in the interior of membrane, polar carotenoids such as astaxanthin and macular xanthophylls span the cell membrane bilayer. The intramembranous alignment of the polar carotenoids allows the molecules to scavenge ROS in the internal and external surface of cells as well as in the interior of the membrane (Goto et al. 2001). On the other hand, more extended π-electron-conjugated chain gives lycopene more power to quench free radicals. Carotenoids stimulate nuclear factor erythroid 2–related factor 2 (Nrf2) signaling, which resulted in the inhibition of redox-induced activation of the nuclear factor-κB (NF-κB) pathway (Ben-Dor et al. 2005; Farruggia et al. 2018; J. E. Kim et al. 2012; Wu et al. 2015). Apart from antioxidant mechanism, carotenoids can directly inhibit NF-κB signaling pathway (Bhuvaneswari et al. 2014; Gouranton et al. 2011; J.-H. Kim et al. 2008; Ni et al. 2015). Few trials investigated the effects of carotenoids on NF-κB pathway. In a clinical trial, carotenoid enrichment of enteral nutrition significantly reduced NF-κB lymphocyte expression as well as oxidative stress (Vaisman et al. 2006). Lycopene have been shown that inhibits NF-κB DNA binding and expression of NF-κB p65 and consequently reduces gene expression of pro-inflammatory cytokines (Sahin et al. 2010; Simone et al. 2011). The interaction of electrophilic groups of xanthophylls with cysteine residues of IκB kinase (IKK) and NF-κB p65 subunits may potentially decrease the phosphorylation levels of IKK and p65 leading to suppressing

NF-κB signaling pathway (Linnewiel-Hermoni et al. 2014). Moreover, data from mechanistic studies showed that astaxanthin and lutein could act as agonists for peroxisome proliferator-activated receptor (PPAR) γ in the cell context of macrophages or in the proinflammatory state and thereby regulate transcription of various genes involved in the inflammatory response such as NF-κB and activator protein 1 (Inoue et al. 2012; Selvaraj and Klasing 2006; Selvaraj, Shanmugasundaram, and Klasing 2010).

Overall, this meta-analysis supported the potential anti-inflammatory effects of carotenoids, in particular for astaxanthin, lutein/zeaxanthin, and lycopene. Given the growing interest in carotenoids as well as their potential anti-inflammatory effects observed in the present meta-analysis, more RCTs of high quality should be performed to determine the therapeutic doses required for various population subgroups.

5. Knowledge gaps and future directions

The present meta-analysis had the strengths as follows: inclusion of comprehensive overview of the available trials, appropriate meta-analytic methodology, and inclusion of a large number of studies with a randomized controlled design that used a pure or extracted carotenoids, instead of food. These items reduced possible bias in concluding about the anti-inflammatory effects of carotenoids, however, gap knowledge should be considered before subsuming carotenoids into clinical practice. There was high heterogeneity between studies in terms of study quality, characteristics of participants, the type and dose of carotenoids, and the duration of the intervention. However, we used random-effect model as well as subgroup and sensitivity analyses, still the heterogeneity remained high. Although our meta-analysis included carotenoids from five subclasses, the limited number of RCTs on β-carotene and β-cryptoxanthin didn't allow us to draw definitive conclusions about these subclasses. Compared to CRP, fewer studies evaluated the effect of individual carotenoids such as lutein/zeaxanthin and crocin on IL-6 and TNF-α, making it difficult to conclude certainly about their effects on these cytokines. Only 10 studies controlled the dietary intake using validated dietary records. Compliance rates were also assessed only in 16 studies. Many of studies did not report information on some lifestyle habits including physical activity level and smoking habits. To reach firm conclusions on health claims regarding carotenoids, it is necessary to consider the following issues: controlling variations in weight, nutritional habits, and physical activity; evaluation of compliance rate, particularly through biomarkers for carotenoids consumption; and restricting dietary intake of carotenoids.

6. Conclusion

The results of current meta-analysis showed that in the overall assessment, carotenoids reduced CRP and IL-6 levels compared with controls, while the effect on TNF-α level was not statistically significant. For the individual carotenoids, astaxanthin, lutein with or without zeaxanthin, and β-cryptoxanthin significantly decreased CRP level. Also, lycopene was the only carotenoid that caused a significant reduction in IL-6 level. Further high quality and adequately powered RCTs that consider current knowledge gaps, should be conducted, to further enhance our understanding of the efficacy of carotenoids on inflammatory conditions.

Author contributions

FH-S: Contributed to the study conception, design and data collection, and interpretation and drafting the manuscript; MA and RZ: Participated in revising the paper critically and approving the version of the manuscript being submitted; ESZ, MM: screened articles and extracted data; and all authors: contributed to the manuscript, and read and approved the final manuscript.

Disclosure statement

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Abbreviations

BMI	body mass index
CRP	C-reactive protein
CVD	cardiovascular diseases
IKK	IκB kinase
IL-6	interleukin-6
NF-κB	nuclear factor-κB
Nrf2	nuclear factor erythroid 2–related factor 2
PPAR	peroxisome proliferator-activated receptor
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
RCT	randomized controlled trials
ROS	reactive oxygen species
SD	standard deviations
TNF-α	tumor necrosis factor-alpha
WMD	weighted mean difference

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References

Asbaghi, O., M. Sadeghian, O. Sadeghi, S. Rigi, S. C. Tan, A. Shokri, and S. M. Mousavi. 2021. Effects of saffron (Crocus sativus L.) supplementation on inflammatory biomarkers: A systematic review and meta-analysis. *Phytotherapy Research* 35 (1):20–32. doi:10.1002/ptr.6748.

Baralic, I., M. Andjelkovic, B. Djordjevic, N. Dikic, N. Radivojevic, V. Suzin-Zivkovic, S. Radojevic-Skodric, and S. Pejic. 2015. Effect of astaxanthin supplementation on salivary IgA, oxidative stress, and inflammation in young soccer players. *Evidence-Based Complementary and Alternative Medicine* 2015:1–9. doi:10.1155/2015/783761

Baune, B. T., M. Rothermundt, K. H. Ladwig, C. Meisinger, and K. Berger. 2011. Systemic inflammation (Interleukin 6) predicts all-

cause mortality in men: Results from a 9-year follow-up of the MEMO Study. *Age* 33 (2):209–17. doi: 10.1007/s11357-010-9165-5.

Ben-Dor, A., M. Steiner, L. Gheber, M. Danilenko, N. Dubi, K. Linnewiel, A. Zick, Y. Sharoni, and J. Levy. 2005. Carotenoids activate the antioxidant response element transcription system. *Molecular Cancer Therapeutics* 4 (1):177–86.

Beydoun, M. A., J. A. Canas, H. A. Beydoun, X. Chen, M. R. Shroff, and A. B. Zonderman. 2012. Serum antioxidant concentrations and metabolic syndrome are associated among US adolescents in recent national surveys. *The Journal of Nutrition* 142 (9):1693–704. doi: 10.3945/jn.112.160416.

Bhuvaneswari, S., B. Yogalakshmi, S. Sreeja, and C. V. Anuradha. 2014. Astaxanthin reduces hepatic endoplasmic reticulum stress and nuclear factor-κB-mediated inflammation in high fructose and high fat diet-fed mice. *Cell Stress and Chaperones* 19 (2):183–91. doi: 10.1007/s12192-013-0443-x.

Bouayed, J., and T. Bohn. 2012. Dietary derived antioxidants: Implications on health. In *Nutrition, well-being and health*, 1–22. Rijeka, Croatia: InTech.

Britton, G. 1995. Structure and properties of carotenoids in relation to function. *Faseb Journal* 9 (15):1551–8.

Briviba, K., S. E. Kulling, J. Möseneder, B. Watzl, G. Rechkemmer, and A. Bub. 2004. Effects of supplementing a low-carotenoid diet with a tomato extract for 2 weeks on endogenous levels of DNA single strand breaks and immune functions in healthy non-smokers and smokers. *Carcinogenesis* 25 (12):2373–8. doi: 10.1093/carcin/bgh249.

Buckley, D. I., R. Fu, M. Freeman, K. Rogers, and M. Helfand. 2009. C-reactive protein as a risk factor for coronary heart disease: A systematic review and meta-analyses for the US Preventive Services Task Force. *Annals of Internal Medicine* 151 (7):483–95. doi: 10.7326/0003-4819-151-7-200910060-00009.

Chan, K. C., S. C. Chen, and P. C. Chen. 2019. Astaxanthin attenuated thrombotic risk factors in type 2 diabetic patients. *Journal of Functional Foods* 53:22–7. doi: 10.1016/j.jff.2018.12.012.

Chen, J.-T., and K. Kotani. 2017. Effects of astaxanthin on liver and leukocyte parameters in healthy climacteric women: Preliminary data. *Journal of Medicinal Food* 20 (7):724–5. doi: 10.1089/jmf.2016.3819.

Cheng, H. M., G. Koutsidis, J. K. Lodge, A. Ashor, M. Siervo, and J. Lara. 2017. Tomato and lycopene supplementation and cardiovascular risk factors: A systematic review and meta-analysis. *Atherosclerosis* 257:100–8. doi: 10.1016/j.atherosclerosis.2017.01.009.

Choi, H. D., J. H. Kim, M. J. Chang, Y. Kyu-Youn, and W. G. Shin. 2011. Effects of astaxanthin on oxidative stress in overweight and obese adults. *Phytotherapy Research* 25 (12):1813–8. doi: 10.1002/ptr.3494.

Coombes, J. S., J. E. Sharman, and R. G. Fassett. 2016. Astaxanthin has no effect on arterial stiffness, oxidative stress, or inflammation in renal transplant recipients: A randomized controlled trial (the XANTHIN trial). *American Journal of Clinical Nutrition* 103 (1):283–9. doi: 10.3945/ajcn.115.115477.

Coyne, T., T. I. Ibiebele, P. D. Baade, A. Dobson, C. McClintock, S. Dunn, D. Leonard, and J. Shaw. 2005. Diabetes mellitus and serum carotenoids: Findings of a population-based study in Queensland, Australia. *The American Journal of Clinical Nutrition* 82 (3):685–93. doi: 10.1093/ajcn/82.3.685.

Danesh, J., S. Kaptoge, A. G. Mann, N. Sarwar, A. Wood, S. B. Angleman, F. Wensley, J. P. Higgins, L. Lennon, and G. Eiriksdottir. 2008. Long-term interleukin-6 levels and subsequent risk of coronary heart disease: Two new prospective studies and a systematic review. *PLoS Medicine* 5 (4):e78. doi: 10.1371/journal.pmed.0050078.

El-Agamey, A., and D. J. McGarvey. 2008. *Carotenoid radicals and radical ions*. In *Carotenoids*, vol 4, 119–154. Switzerland: Springer. doi: 10.1007/978-3-7643-7499-0_7.

Erlinger, T. P., E. Guallar, E. R. Miller, III, R. Stolzenberg-Solomon, and L. J. Appel. 2001. Relationship between systemic markers of inflammation and serum β-carotene levels. *Archives of Internal Medicine* 161 (15):1903–8. doi: 10.1001/archinte.161.15.1903.

Estévez-Santiago, R., J. M. Silván, C. A. Can-Cauich, A. M. Veses, I. Alvarez-Acero, M. A. Martinez-Bartolome, R. San-Román, M. Cámara, B. Olmedilla-Alonso, and S. De Pascual-Teresa. 2019. Lack of a synergistic effect on cardiometabolic and redox markers in a dietary supplementation with anthocyanins and xanthophylls in postmenopausal women. *Nutrients* 11 (7):1533. doi: 10.3390/nu11071533.

Farruggia, C., M.-B. Kim, M. Bae, Y. Lee, T. X. Pham, Y. Yang, M. J. Han, Y.-K. Park, and J.-Y. Lee. 2018. Astaxanthin exerts anti-inflammatory and antioxidant effects in macrophages in NRF2-dependent and independent manners. *The Journal of Nutritional Biochemistry* 62:202–9. doi: 10.1016/j.jnutbio.2018.09.005.

Fiedor, J., and K. Burda. 2014. Potential role of carotenoids as antioxidants in human health and disease. *Nutrients* 6 (2):466–88. doi: 10.3390/nu6020466.

Gajendragadkar, P. R., A. Hubsch, K. M. Mäki-Petäjä, M. Serg, I. B. Wilkinson, and J. Cheriyan. 2014. Effects of oral lycopene supplementation on vascular function in patients with cardiovascular disease and healthy volunteers: A randomised controlled trial. *PLOS One.* 9 (6):e99070. doi: 10.1371/journal.pone.0099070.

Gallicchio, L., K. Boyd, G. Matanoski, X. (G.). Tao, L. Chen, T. K. Lam, M. Shiels, E. Hammond, K. A. Robinson, L. E. Caulfield, et al. 2008. Carotenoids and the risk of developing lung cancer: A systematic review. *The American Journal of Clinical Nutrition* 88 (2):372–83. doi: 10.1093/ajcn/88.2.372.

Ghaderi, A., M. Rasouli-Azad, N. Vahed, H. R. Banafshe, A. Soleimani, A. Omidi, F. S. Ghoreishi, and Z. Asemi. 2019. Clinical and metabolic responses to crocin in patients under methadone maintenance treatment: A randomized clinical trial. *Phytotherapy Research* 33 (10):2714–25. doi: 10.1002/ptr.6445.

Ghiasian, M., F. Khamisabadi, N. Kheiripour, M. Karami, R. Haddadi, A. Ghaleiha, B. Taghvaei, S. S. Oliaie, M. Salehi, P. Samadi, et al. 2019. Effects of crocin in reducing DNA damage, inflammation, and oxidative stress in multiple sclerosis patients: A double-blind, randomized, and placebo-controlled trial [Article. *Journal of Biochemical and Molecular Toxicology* 33 (12):e22410. doi: 10.1002/jbt.22410.

Goto, S., K. Kogure, K. Abe, Y. Kimata, K. Kitahama, E. Yamashita, and H. Terada. 2001. Efficient radical trapping at the surface and inside the phospholipid membrane is responsible for highly potent antiperoxidative activity of the carotenoid astaxanthin. *Biochimica et Biophysica Acta (BBA) - Biomembranes* 1512 (2):251–8. doi: 10.1016/S0005-2736(01)00326-1.

Gouranton, E., C. Thabuis, C. Riollet, C. Malezet-Desmoulins, C. El Yazidi, M. Amiot, P. Borel, and J. Landrier. 2011. Lycopene inhibits proinflammatory cytokine and chemokine expression in adipose tissue. *The Journal of Nutritional Biochemistry* 22 (7):642–8. doi: 10.1016/j.jnutbio.2010.04.016.

Graydon, R., R. E. Hogg, U. Chakravarthy, I. S. Young, and J. V. Woodside. 2012. The effect of lutein- and zeaxanthin-rich foods v. supplements on macular pigment level and serological markers of endothelial activation, inflammation and oxidation: Pilot studies in healthy volunteers. *British Journal of Nutrition* 108 (2):334–42. doi: 10.1017/S0007114511005599.

Haidari, F., A. Hojhabrimanesh, B. Helli, S. S. Seyedian, and K. Ahmadi-Angali. 2020. An energy-restricted high-protein diet supplemented with β-cryptoxanthin alleviated oxidative stress and inflammation in nonalcoholic fatty liver disease: A randomized controlled trial. *Nutrition Research* 73:15–26. doi: 10.1016/j.nutres.2019.08.009.

Hajizadeh-Sharafabad, F., Z. Ghoreishi, V. Maleki, and A. Tarighat-Esfanjani. 2019. Mechanistic insights into the effect of lutein on atherosclerosis, vascular dysfunction, and related risk factors: A systematic review of in vivo, ex vivo and in vitro studies. *Pharmacological Research* 149:104477. doi: 10.1016/j.phrs.2019.104477.

Hajizadeh-Sharafabad, F., A. Tarighat-Esfanjani, Z. Ghoreishi, and M. Sarreshtedari. 2020. Lutein supplementation combined with a low-calorie diet in middle-aged obese individuals: effects on anthropometric indices, body composition and metabolic parameters. *The British Journal of Nutrition* :1–12. doi:10.1017/S0007114520004997. PMID:33298201

Harari, A., D. Harats, D. Marko, H. Cohen, I. Barshack, Y. Kamari, A. Gonen, Y. Gerber, A. Ben-Amotz, and A. Shaish. 2008. A 9-cis β-carotene–enriched diet inhibits atherogenesis and fatty liver formation in LDL receptor knockout mice. *Journal of Nutrition* 138 (10):1923–30. doi: 10.1093/jn/138.10.1923.

Zeng, L., K. C. Collins, Y. Hu, M. N. Luckyanova, A. A. Maznev, S. Huberman, V. Chiloyan, J. Zhou, X. Huang, K. A. Nelson, et al. 2015. Measuring Phonon Mean Free Path Distributions by Probing Quasiballistic Phonon Transport in Grating Nanostructures. *Scientific Reports* 5:17131 doi:10.1038/srep17131. PMC: 26612032

Hozawa, A., D. R. Jacobs, Jr., M. W. Steffes, M. D. Gross, L. M. Steffen, and D.-H. Lee. 2006. Associations of serum carotenoid concentrations with the development of diabetes and with insulin concentration: Interaction with smoking: The Coronary Artery Risk Development in Young Adults (CARDIA) Study. *American Journal of Epidemiology* 163 (10):929–37. doi: 10.1093/aje/kwj136.

Hu, P., D. B. Reuben, E. M. Crimmins, T. B. Harris, M.-H. Huang, and T. E. Seeman. 2004. The effects of serum beta-carotene concentration and burden of inflammation on all-cause mortality risk in high-functioning older persons: MacArthur studies of successful aging. *The Journals of Gerontology Series A: Biological Sciences and Medical Sciences* 59 (8):M849–M854. doi: 10.1093/gerona/59.8.M849.

Inoue, M., H. Tanabe, A. Matsumoto, M. Takagi, K. Umegaki, S. Amagaya, and J. Takahashi. 2012. Astaxanthin functions differently as a selective peroxisome proliferator-activated receptor γ modulator in adipocytes and macrophages. *Biochemical Pharmacology* 84 (5): 692–700. doi: 10.1016/j.bcp.2012.05.021.

Jing, L., M. Xiao, H. Dong, J. Lin, G. Chen, W. Ling, and Y. Chen. 2018. Serum carotenoids are inversely associated with RBP4 and other inflammatory markers in middle-aged and elderly adults. *Nutrients* 10 (3):260. doi: 10.3390/nu10030260.

Karppi, J., S. Kurl, K. Ronkainen, J. Kauhanen, and J. A. Laukkanen. 2013. Serum carotenoids reduce progression of early atherosclerosis in the carotid artery wall among Eastern Finnish men. *PLOS One* 8 (5):e64107. doi: 10.1371/journal.pone.0064107.

Kaulmann, A., and T. Bohn. 2014. Carotenoids, inflammation, and oxidative stress—implications of cellular signaling pathways and relation to chronic disease prevention. *Nutrition Research* 34 (11): 907–29. doi: 10.1016/j.nutres.2014.07.010.

Kesse-Guyot, E., N. Ahluwalia, C. Lassale, S. Hercberg, L. Fezeu, and D. Lairon. 2013. Adherence to Mediterranean diet reduces the risk of metabolic syndrome: A 6-year prospective study. *Nutrition, Metabolism and Cardiovascular Diseases* 23 (7):677–83. doi: 10.1016/j.numecd.2012.02.005.

Kim, J.-H., H.-J. Na, C.-K. Kim, J.-Y. Kim, K.-S. Ha, H. Lee, H.-T. Chung, H. J. Kwon, Y.-G. Kwon, and Y.-M. Kim. 2008. The non-provitamin A carotenoid, lutein, inhibits NF-κB-dependent gene expression through redox-based regulation of the phosphatidylinositol 3-kinase/PTEN/Akt and NF-κB-inducing kinase pathways: Role of H2O2 in NF-κB activation. *Free Radical Biology and Medicine* 45 (6):885–96. doi: 10.1016/j.freeradbiomed.2008.06.019.

Kim, J. E., R. M. Clark, Y. Park, J. Lee, and M. L. Fernandez. 2012. Lutein decreases oxidative stress and inflammation in liver and eyes of guinea pigs fed a hypercholesterolemic diet. *Nutrition Research and Practice* 6 (2):113–9. doi: 10.4162/nrp.2012.6.2.113.

Kim, J. Y., J. K. Paik, O. Y. Kim, H. W. Park, J. H. Lee, Y. Jang, and J. H. Lee. 2011. Effects of lycopene supplementation on oxidative stress and markers of endothelial function in healthy men. *Atherosclerosis* 215 (1):189–95. doi: 10.1016/j.atherosclerosis.2010.11.036.

Kirkil, G., M. H. Muz, E. Sancaktar, D. Kaman, K. Sahin, and O. Kucuk. 2012. The effect of lycopene supplementation on chronic obstructive lung disease. *Nobel Medicus* 8 (3):98–104.

Kuhad, A., R. Sethi, and K. Chopra. 2008. Lycopene attenuates diabetes-associated cognitive decline in rats. *Life Sciences* 83 (3–4): 128–34. doi: 10.1016/j.lfs.2008.05.013.

Linnewiel-Hermoni, K., Y. Motro, Y. Miller, J. Levy, and Y. Sharoni. 2014. Carotenoid derivatives inhibit nuclear factor kappa B activity in bone and cancer cells by targeting key thiol groups. *Free Radical Biology and Medicine* 75:105–20. doi: 10.1016/j.freeradbiomed.2014.07.024.

Liu, R. H. 2013. Health-promoting components of fruits and vegetables in the diet. *Advances in Nutrition* 4 (3):384S–92S. doi: 10.3945/an.112.003517.

Martínez-Delgado, A. A., S. Khandual, and S. J. Villanueva–Rodríguez. 2017. Chemical stability of astaxanthin integrated into a food matrix: Effects of food processing and methods for preservation. *Food Chemistry* 225:23–30. doi: 10.1016/j.foodchem.2016.11.092.

Mazidi, M., N. Katsiki, E. S. George, and M. Banach. 2020. Tomato and lycopene consumption is inversely associated with total and cause-specific mortality: A population-based cohort study, on behalf of the International Lipid Expert Panel (ILEP). *British Journal of Nutrition* 124 (12):1303–10. doi: 10.1017/S0007114519002150.

Mazidi, M., A. P. Kengne, N. Katsiki, D. P. Mikhailidis, and M. Banach. 2018. Inverse association between serum antioxidant levels and inflammatory markers is moderated by adiposity: A report based on a large representative population sample of American adults. *British Journal of Nutrition* 120 (11):1272–8. doi: 10.1017/S0007114518002581.

Mohamadpour, A. H., Z. Ayati, M. R. Parizadeh, O. Rajbai, and H. Hosseinzadeh. 2013. Safety evaluation of crocin (a constituent of saffron) tablets in healthy volunteers. *Iranian Journal of Basic Medical Sciences* 16 (1):39.

Morrow, D. A., and P. M. Ridker. 2000. C-reactive protein, inflammation, and coronary risk. *Medical Clinics of North America* 84 (1): 149–61. doi: 10.1016/S0025-7125(05)70211-X.

Mounien, L., F. Tourniaire, and J.-F. Landrier. 2019. Anti-obesity effect of carotenoids: Direct impact on adipose tissue and adipose tissue-driven indirect effects. *Nutrients* 11 (7):1562. doi: 10.3390/nu11071562.

Mousavi, B., S. Z. Bathaie, F. Fadai, Z. Ashtari, N. Ali Beigi, S. Farhang, S. Hashempour, N. Shahhamzei, and H. Heidarzadeh. 2015. Safety evaluation of saffron stigma (Crocus sativus L.) aqueous extract and crocin in patients with schizophrenia. *Avicenna Journal of Phytomedicine* 5 (5):413–9.

Ni, Y., M. Nagashimada, F. Zhuge, L. Zhan, N. Nagata, A. Tsutsui, Y. Nakanuma, S. Kaneko, and T. Ota. 2015. Astaxanthin prevents and reverses diet-induced insulin resistance and steatohepatitis in mice: A comparison with vitamin E. *Scientific Reports* 5:17192. doi: 10.1038/srep17192.

Nieman, D. C., C. L. Capps, C. R. Capps, Z. L. Shue, and J. E. McBride. 2018. Effect of 4-week ingestion of tomato-based carotenoids on exercise-induced inflammation, muscle damage, and oxidative stress in endurance runners. *International Journal of Sport Nutrition and Exercise Metabolism* 28 (3):266–73. doi: 10.1123/ijsnem.2017-0272.

Niranjana, R., R. Gayathri, S. N. Mol, T. Sugawara, T. Hirata, K. Miyashita, and P. Ganesan. 2015. Carotenoids modulate the hallmarks of cancer cells. *Journal of Functional Foods* 18:968–85. doi: 10.1016/j.jff.2014.10.017.

Nosrati, M., I. Nikbakht-Jam, S. Eslami, M. M. Khademi, M. Foroutan-Tanha, F. Hadizadeh, A. H. Sahebkar, S. Tavallaie, Z. Taherzadeh, M. Ghayour-Mobarhan, et al. 2018. Effect of crocin on antibody titers to heat-shock protein 27 in subjects with metabolic syndrome. *Current Clinical Pharmacology* 12 (2):113–21. doi: 10.2174/1574884712666170509162523.

Park, J. S., J. H. Chyun, Y. K. Kim, L. L. Line, and B. P. Chew. 2010. Astaxanthin decreased oxidative stress and inflammation and enhanced immune response in humans. *Nutrition & Metabolism* 7 (1):18. doi: 10.1186/1743-7075-7-18.

Petyaev, I. M., P. Y. Dovgalevsky, V. A. Klochkov, N. E. Chalyk, and N. Kyle. 2012. Whey protein lycosome formulation improves vascular functions and plasma lipids with reduction of markers of inflammation and oxidative stress in prehypertension. *TheScientificWorldJournal* 2012:269476 doi:10.1100/2012/269476. PMC: 23326213

Poursamimi, J., Z. Shariati-Sarabi, J. Tavakkol-Afshari, S. A. Mohajeri, M. Ghoryani, and M. Mohammadi. 2020. Immunoregulatory effects of Krocina^{TM}, a herbal medicine made of crocin, on osteoarthritis

patients: A successful clinical trial in Iran. *Iranian Journal of Allergy, Asthma and Immunology* 19 (3):253–63. doi: 10.18502/ijaai.v19i3.3453.

Ramezani, A., A. Yousefinejad, M. H. Javanbakht, H. Derakhshanian, and F. Tahbaz. 2014. Effect of beta-carotene enriched carrot juice on inflammatory status and fasting blood glucose in type 2 diabetic patients. *Current Topics in Nutraceutical Research* 12 (1–2):1–7.

Riccioni, G., N. D'Orazio, L. Speranza, E. Di Ilio, M. Glade, V. Bucciarelli, L. Scotti, F. Martini, A. Pennelli, and T. Bucciarelli. 2010. Carotenoids and asymptomatic carotid atherosclerosis. *Journal of Biological Regulators & Homeostatic Agents* 24 (4):447.

Sahin, K., M. Tuzcu, N. Sahin, S. Ali, and O. Kucuk. 2010. Nrf2/HO-1 signaling pathway may be the prime target for chemoprevention of cisplatin-induced nephrotoxicity by lycopene. *Food and Chemical Toxicology* 48 (10):2670–4. doi: 10.1016/j.fct.2010.06.038.

Salzano, S., P. Checconi, E.-M. Hanschmann, C. H. Lillig, L. D. Bowler, P. Chan, D. Vaudry, M. Mengozzi, L. Coppo, S. Sacre, et al. 2014. Linkage of inflammation and oxidative stress via release of glutathionylated peroxiredoxin-2, which acts as a danger signal. *Proceedings of the National Academy of Sciences* 111 (33):12157–62. doi: 10.1073/pnas.1401712111.

Selvaraj, R. K., and K. C. Klasing. 2006. Lutein and eicosapentaenoic acid interact to modify iNOS mRNA levels through the PPARγ/RXR pathway in chickens and HD11 cell lines. *The Journal of Nutrition* 136 (6):1610–6. doi: 10.1093/jn/136.6.1610.

Selvaraj, R. K., R. Shanmugasundaram, and K. C. Klasing. 2010. Effects of dietary lutein and PUFA on PPAR and RXR isomer expression in chickens during an inflammatory response. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology* 157 (3):198–203. doi: 10.1016/j.cbpa.2010.06.172.

Shah, T., J. P. Casas, J. A. Cooper, I. Tzoulaki, R. Sofat, V. McCormack, L. Smeeth, J. E. Deanfield, G. D. Lowe, A. Rumley, et al. 2009. Critical appraisal of CRP measurement for the prediction of coronary heart disease events: New data and systematic review of 31 prospective cohorts. *International Journal of Epidemiology* 38 (1):217–31. doi: 10.1093/ije/dyn217.

Simone, R. E., M. Russo, A. Catalano, G. Monego, K. Froehlich, V. Boehm, and P. Palozza. 2011. Lycopene inhibits NF-kB-mediated IL-8 expression and changes redox and PPARγ signalling in cigarette smoke–stimulated macrophages. *PLOS One* 6 (5):e19652. doi: 10.1371/journal.pone.0019652.

Stahl, W., N. Ale-Agha, and M. C. Polidori. 2002. Non-antioxidant properties of carotenoids. *Biological Chemistry* 383 (3–4):553–8. doi: 10.1515/BC.2002.056.

Stringham, N. T., P. V. Holmes, and J. M. Stringham. 2019. Effects of macular xanthophyll supplementation on brain-derived neurotrophic factor, pro-inflammatory cytokines, and cognitive performance. *Physiology & Behavior* 211:112650. doi: 10.1016/j.physbeh.2019.112650.

Suzuki, K., T. Inoue, S. Hashimoto, J. Ochiai, Y. Kusuhara, Y. Ito, and N. Hamajima. 2010. Association of serum carotenoids with high molecular weight adiponectin and inflammation markers among Japanese subjects. *Clinica Chimica Acta* 411 (17–18):1330–4. doi: 10.1016/j.cca.2010.05.029.

Thies, F., L. F. Masson, A. Rudd, N. Vaughan, C. Tsang, J. Brittenden, W. G. Simpson, S. Duthie, G. W. Horgan, and G. Duthie. 2012. Effect of a tomato-rich diet on markers of cardiovascular disease risk in moderately overweight, disease-free, middle-aged adults: A randomized controlled trial. *American Journal of Clinical Nutrition* 95 (5):1013–22. doi: 10.3945/ajcn.111.026286.

Vaisman, N., G. R. Haenen, Y. Zaruk, C. Verduyn, J. G. Bindels, S. Verlaan, and E. P. Meijer. 2006. Enteral feeding enriched with carotenoids normalizes the carotenoid status and reduces oxidative stress in long-term enterally fed patients. *Clinical Nutrition* 25 (6):897–905. doi: 10.1016/j.clnu.2006.06.002.

Volpato, S., J. M. Guralnik, L. Ferrucci, J. Balfour, P. Chaves, L. P. Fried, and T. B. Harris. 2001. Cardiovascular disease, interleukin-6, and risk of mortality in older women: The women's health and aging study. *Circulation* 103 (7):947–53. doi: 10.1161/01.cir.103.7.947.

Walston, J., Q. Xue, R. D. Semba, L. Ferrucci, A. R. Cappola, M. Ricks, J. Guralnik, and L. P. Fried. 2006. Serum antioxidants, inflammation, and total mortality in older women. *American Journal of Epidemiology* 163 (1):18–26. doi: 10.1093/aje/kwj007.

Wang, L., J. M. Gaziano, E. P. Norkus, J. E. Buring, and H. D. Sesso. 2008. Associations of plasma carotenoids with risk factors and biomarkers related to cardiovascular disease in middle-aged and older women. *The American Journal of Clinical Nutrition* 88 (3):747–54. doi: 10.1093/ajcn/88.3.747.

Wang, M. X., J. H. Jiao, Z. Y. Li, R. R. Liu, Q. Shi, and L. Ma. 2013. Lutein supplementation reduces plasma lipid peroxidation and C-reactive protein in healthy nonsmokers. *Atherosclerosis* 227 (2):380–5. doi: 10.1016/j.atherosclerosis.2013.01.021.

Waris, G., and H. Ahsan. 2006. Reactive oxygen species: Role in the development of cancer and various chronic conditions. *Journal of Carcinogenesis* 5 (1):14. doi: 10.1186/1477-3163-5-14.

Wu, W., Y. Li, Y. Wu, Y. Zhang, Z. Wang, and X. Liu. 2015. Lutein suppresses inflammatory responses through Nrf2 activation and NF-κB inactivation in lipopolysaccharide-stimulated BV-2 microglia. *Molecular Nutrition & Food Research* 59 (9):1663–73. doi: 10.1002/mnfr.201500109.

Xia, W., N. Tang, H. K. Varkaneh, T. Y. Low, S. C. Tan, X. Wu, and Y. Zhu. 2020. The effects of astaxanthin supplementation on obesity, blood pressure, CRP, glycemic biomarkers, and lipid profile: A meta-analysis of randomized controlled trials. *Pharmacological Research* 161. doi:10.1016/j.phrs.2020.105113.

Xu, X. R., Z. Y. Zou, X. Xiao, Y. M. Huang, X. Wang, and X. M. Lin. 2013. Effects of lutein supplement on serum inflammatory cytokines. *Journal of Atherosclerosis and Thrombosis* 20 (2):170–7. doi: 10.5551/jat.14365.