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1 **Carotenoid Metabolites, their Tissue and Blood Concentrations in Humans and further Bioactivity**
2 **via Retinoid Receptor-Mediated Signalling**

3
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5
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20
21
22 **Short title:** Carotenoid metabolites human tissue levels

26 **Abstract:**

27 Many epidemiological studies have emphasised the relation between carotenoid dietary intake and their
28 circulating concentrations and beneficial health effect, such as lower risk of cardiometabolic diseases and
29 cancer. However, there is dispute as to whether the attributed health benefits are due to native carotenoids
30 or they are rather induced by their metabolites. Several categories of metabolites have been reported,
31 most notably due to a) modifications at the cyclohexenyl-ring or the polyene chain, such as epoxides and
32 geometric isomers, b) excentric cleavage metabolites with also alcohol-, aldehyde- or carboxylic acid-
33 functional groups or c) centric cleaved metabolites with additional hydroxyl-, aldehyde- or carboxyl-
34 functionalities, not counting their potential phase-II glucuronidated/sulphated derivatives. Of special
35 interest are the apo-carotenoids, which originate in the intestine and other tissues from carotenoids
36 cleavage by beta-carotene oxygenases 1/2 in a symmetrical / non-symmetrical fashion. These are more
37 water soluble and more electrophilic, and therefore putative candidates for interactions with transcription
38 factors such as NF-kB and Nrf2, as well as ligands for RAR-RXR nuclear receptor interactions. In this
39 review, we discuss *in vivo* detected apo-carotenoids, their reported tissue concentrations, and potential
40 associated health effects, focussing **exclusively on the human situation and based on quantified / semi-**
41 **quantified carotenoid-metabolites proven to be present in humans.**

42
43 **Key-words:** Apo-carotenoids, apo-lycopenoids, cleavage products, tissue concentrations, liver.

1. Introduction

Carotenoids are typically colourful, mostly C-40 based pigments, which are generally obtained via plant food items. Over 1100 different carotenoids have been recognized [1], and additional new carotenoids are being discovered, including shorter (C-30), and longer (C-50) analogues of bacterial origin [2]. Likewise, the apo-carotenoids, their breakdown products formed in plants [3] or after human ingestion [4], can be considered to belong to this group.

The interest in these secondary plant compounds has been thoroughly increased in the last 2-3 decades, due to the relation of their intake and circulating plasma concentrations with chronic disease risk. A high carotenoid intake within a plant-food rich diet and concentrations in plasma have been related, among others, to a reduced risk of type-2 diabetes [5], age-related macular degeneration [6], some types of cancer such as those of the prostate [7], and even total mortality [8]. The underlying mechanisms for such associated health-benefits are not quite clear and the topic of controversial discussions, but have included direct antioxidant effects such as quenching of singlet oxygen and lipid peroxides [9], interactions with transcription factors related to inflammatory pathways (e.g., NF-kB) and oxidative stress (e.g., Nrf-2) [10], and also their interaction with the nuclear factors retinoid-X receptors (RXRs) and retinoic acid receptors (RARs) together with peroxisome proliferator-activated receptors (PPARs) [11-13].

It has also been postulated that the potential health benefits are conveyed not necessarily by the native carotenoids, following their absorption in the small intestine, but by their metabolites / cleavage products. Carotenoids as lipophilic constituents are absorbed following their micellization into the enterocytes, where they may partly undergo cleavage by carotenoid-oxygenases, namely BCO1 and BCO2, resulting in the formation of symmetrical or non-symmetrical cleavage products [14].

While some of the symmetrical cleavage products have vitamin A activity (following e.g. cleavage of β -carotene or β -cryptoxanthin) by BCO1, the biological role of the other cleavage products remains uncertain. These cleavage products or apo-carotenoids have been proposed to be bioactive. For instance,

71 in *in vitro* studies, lycopene derivatives were shown to have higher affinity to Nrf-2 and NF-kB, due to
72 their higher electrophilicity, and perhaps better aqueous solubility [15-19]. Lycopene has been shown to
73 act in part similarly to vitamin A metabolites in normalizing a vitamin A-deficient diet in rats/mice [20].
74 It cannot also be excluded that bacteria in the colon produce more hydrophilic metabolites of carotenoids
75 that are bioavailable and bioactive [21].

76
77 Therefore, there has been increased interest in carotenoid-metabolites and their potential connection to
78 health benefits. A limitation of their detection in human specimens is the lack of commercial standards, in
79 addition to their lower concentration and the lower sensitivity of UV-detection, the most common
80 technique employed in their quantification, due to the shortened delocalized electron system in the
81 molecule.

82
83 In this review, we strive to present the current state of knowledge of metabolites and breakdown product
84 of carotenoids in humans, their known concentration ranges, and potential health benefits involved, as
85 well as pointing out gaps and potential ways forward in this research domain. **In this review, we**
86 **exclusively focussed on the human situation, based on the proven presence of the described carotenoids**
87 **and carotenoid-metabolites in humans.**

90 **2. Carotenoid metabolites in plasma and tissues**

91 2.1. Rationale for interest on metabolites and overview on metabolites

92 Carotenoids, with major human food relevance (Figure 1 and Table 1), were mainly investigated for their
93 metabolism in the human body and it is uncertain whether the native compounds alone or rather their
94 metabolites are responsible for the attributed health effects. Mainly nuclear hormone receptor-mediated
95 effects were in the focus of these studies [22, 23]. These ligand-activated receptors include especially
96 RARs and RXRs, which may become activated, resulting in altered gene expression of a large set of
97 genes involved in inflammation, differentiation, proliferation and lipid metabolism / **homeostasis [24-27].**

99 **The activation** of RARs and / or RXRs was shown to be related to physiologically- and nutritionally-
100 relevant levels of endogenous carotenoid metabolites [28-31]. Thus, native carotenoids may not interact
101 on their own with gene-regulatory pathways, but rather via their metabolites, the apo-carotenoids, here
102 conclusively the apo-15-carotenoids / retinoids **and potentially others, such as apo-13/14-carotenoids that**
103 **might interact with the binding grooves of RARs and RXRs [32-34]. Here, a focus for activating**
104 **compounds is put on apo-carotenoids with an acid functionality, while apo-carotenoids with aldehyde or**
105 **alcohol functionalities might result in low affinity activators / antagonistic compounds [34].**
106 Consequently, knowing more on their identity, concentration, metabolic pathways and homeostatic
107 control and further RAR-RXR-mediated signalling appears critical for estimating potential health
108 benefits of carotenoids [35-39] (Tables 1 and 2).

109

110 Individual carotenoids, listed in Table 1 including their endogenous levels in serum / plasma as well as
111 selected organs, may be either cleaved by BCO1 (centric cleavage) or BCO2 (excentric cleavage) to
112 produce a variety of apo-carotenoids / retinoids (Figure 1, Table 1,2) [40, 41].

113

114 In general, there are three different kinds of carotenoid metabolites, which occur in human plasma /
115 serum and tissues and have been detected especially after carotenoid supplementation: a) non-cleaved
116 carotenoids with modifications at the cyclohexenyl ring or the polyene chain, such as epoxy-carotenoids,
117 geometric isomers and metabolites resulting from further rearrangement pathways, b) excentrically-
118 cleaved metabolites with also alcohol-, aldehyde- or carboxylic acid-functionalities and, c) centrically-
119 cleaved metabolites with additional alcohol-, aldehyde- or carboxylic acid-functionalities (Figure 1,
120 Table 1,2). Of note, glucuronidated products are also formed, following phase II conjugation, prior to
121 their excretion via the kidney, as reported e.g. for retinoic acids [42-44].

122

123 **The origin of selected apo-15-carotenoids / retinoid-derivatives, such as retinyl esters, retinol, retinal and**
124 **retinoic acids, might occur from various metabolic pathways including a) central cleavage of individual**

125 carotenoids such as β -carotenes or β -cryptoxanthins (Figure 1) by BCO1-cleavage [45-47] b) by
126 interaction of these previously-mentioned carotenoids with environmental or endogenous oxidants and
127 following cleavage [10, 48, 49] or c) by BCO1-cleavage of individual apo-carotenoids, which might
128 originate from food directly or by mitochondrial-based BCO2-cleavage in the human organism [14, 45,
129 47].

130
131 Alternatively, these apo-15-carotenoids / retinoids might originate from food derived apo-15-carotenoids
132 present at high concentration in animal derived food matrices, such as retinol and retinyl-esters, or from
133 bio-active retinoids, for instance retinoic acids and retinal, which are present in low amounts in the food
134 matrix. Unfortunately, it is not possible to quantitatively describe which derivative originated from which
135 individual pathway, or even at which percentile amount, due to the large variety of individually-
136 consumed food sources and individual enzymatic pathways present in humans [49-51].

137
138 Interestingly, some studies reported that blood and tissue concentrations of active vitamin A retinoids
139 differ significantly between disease and health state [reviewed in 52]. These results raise the question as
140 to whether the differences in such levels are caused by the disease or the low intake of carotenoids has
141 led to the development of these conditions. It appears that at least in inflammation-related diseases,
142 vitamin A active compounds are often less abundant in plasma, likely as a consequence and not as a
143 cause of the disease [52], as a potential feedback to counteract inflammation mediated by bioactive
144 vitamin A derivatives induced by pro-inflammatory RAR- and RXR-mediated signalling [53, 54].

145
146 In many countries, shortage of food and especially vitamin A deficiencies are still common [55], and
147 supplementation with provitamin A carotenoids / vitamin A appears to be a prudent strategy. However, in
148 our Western society, vitamin A intakes are very often quite high, while carotenoid intake is generally
149 lower [56, 57]. This has partly been associated with pathophysiological situations [58-63]. Whether
150 increased all-*trans*-retinoic acid (ATRA) concentrations in plasma or tissue following carotenoid
151 supplementation are purely beneficial has thus been discussed controversially [64-67]. The lipid hormone

152 ATRA has been described to be associated to cell differentiation, proliferation and apoptosis with
153 beneficial relevance mainly for cancer prevention [68, 69], and various diseases related to reduced
154 inflammatory competence [70, 71]. Unfortunately, ATRA has also been associated with toxic effects,
155 especially embryonic toxicity [72, 73].

156
157 Recently, ATRA has been discussed more controversially in the context of diabetes, obesity, allergies
158 and osteoporosis [74-76]. Especially the adverse effects of retinoids regarding inflammatory processes,
159 related to many diseases in Western societies, and altering local and systemic lipid metabolism and
160 homeostasis are regarded as critical [73, 77, 78]. If supplementation in such countries with retinoids /
161 carotenoids is in general beneficial must therefore be carefully evaluated.

162 163 2.2. General properties of metabolites originating from β -carotene and β -cryptoxanthin

164 When focusing on β -carotene, we may obtain a large variety of known and yet unknown, although partly
165 postulated, metabolites (Figure 1). In this chapter β -carotene isomers such as α - or γ -isoforms of
166 carotene, geometric isomers of these carotenes were included in addition we add also the provitamin A
167 carotenoid β -cryptoxanthin as a relevant precursor for the later mentioned carotenoid-metabolites under
168 this sub-chapter (Figure 1, Table 1).

169
170 Firstly, several chain-modified carotenoid metabolites have been identified, also in mammals and human
171 serum, with epoxy-, oxo- and hydroxyl-containing functional groups located at the cyclohexenyl ring or
172 at the polyene chain, as well as additional isomers [79, 80]. Whether these metabolites originate from
173 plant-based metabolism or from mammalian endogenous metabolism is not always obvious.
174 Concentrations of these potential metabolites, which are usually lower than those of its parent direct /
175 indirect nutritional precursor all-*trans*- β -carotene, are rarely reported. Problematic is their precise
176 quantification, i.e. lack of commercially available standards and also lower UV-Vis sensitivity.

178 Several similar compounds may also be generated during digestion or during food processing [80]. For
179 example, upon gastrointestinal exposure to oxidizing agents, such as iron, a large variety of degradation
180 products in the intestine have been reported, including several β -apo-carotenals [81], epoxides, and
181 diketones [82]. On the other hand, many reports have stated that carotenoids from plant matrices remain
182 relatively stable upon *in vitro* digestion, as demonstrated for β -carotene [83], lutein [84] and lycopene
183 [85]. Whether such degradation products can be absorbed, and whether they are then further metabolized
184 *in vivo*, remains unknown [81].

185
186 Various apo-carotenoids originating from excentric cleavage of carotenoids were identified in the
187 mammalian and partly in the human organism after carotenoid-supplementation [86, 87]. Both BCO1 and
188 BCO2 appear able to cleave β -carotene. While BCO1 appears to favour full-length provitamin A
189 carotenoids resulting in centric cleavage, BCO2 appears to cleave both provitamin A carotenoids and
190 xanthophylls excentrically [40, 41, 88] and is induced in *Bcmo1*^{-/-} mice adipose tissue, leading to a β -apo-
191 10'-carotenol accumulation [89]. It is possible that some of these metabolites are themselves substrates
192 for BCO1/2, as indicated for β -apo-8'-carotenal, β -apo-10'-carotenal, β -apo-12'-carotenal and β -apo-14'-
193 carotenal in chicken and rats [86, 90, 91]. Unfortunately, as already outlined, a clear ordination which
194 individual carotenoid-metabolite is created by which specific individual metabolic pathway with specific
195 substrate / product derivatives is not possible due to the large diversity of food sources and individual
196 human enzymatic pathways [49, 92]. This large variety of food sources with individual carotenoid-
197 metabolite precursors and endogenous enzymatic pathways is an important feature of the mammalian
198 organism [49, 92]. It entails the use of various regional and timely-restricted available food sources to
199 create and degrade ligands for nuclear hormone receptors to enable normal healthy biological functions.
200 This also includes auto-regulative metabolic and uptake pathways to regulate ligand creation and
201 degradation, as exemplified and described in particular detail in a review relevant for β -carotene [49],
202 outlined in Figure 1.

204 Different apo-carotenals and apo-carotenoic acids of various chain lengths were found after β -carotene
205 supplementation [86, 87]. These were then synthesized *ex vivo* and further studied in molecular biological
206 experiments, and partly identified after direct supplementation of β -carotene and food items rich in β -
207 carotene. These described apo-carotenoids are of different chain lengths, ranging from apo-8'-, apo-10'-,
208 apo-12'- and apo-14'-carotenals, and can further be oxidized to apo-carotenoic acids (Figure 1).
209 Contrarily, endogenously produced levels have rarely been described, only for selected derivatives [93].
210 Following the ingestion of a high β -carotene tomato juice (360 mL, 30 mg β -carotene, 35 μ g apo-
211 carotenoids / day), only apo-10'- and 12'-carotenal were **claimed to be detected in plasma of some**
212 **individuals under the set quantification limit, though unfortunately without any added visualized**
213 **analytical confirmation** [94]. It could not be distinguished whether these were absorbed or formed *de*
214 *novo in vivo* [81, 94]. In a study by Kopec *et al.* [81], (^{13}C -10)- β -carotene was administered to healthy
215 subjects. Though non-symmetrical β -apo-carotenals were found in the gut, none were observed in the
216 plasma TRL fraction, suggesting a low bioavailability.

217
218 Following excentric cleavage, the shorter products such as β -ionone, β -cyclocitral and related derivatives,
219 have also been described *in vitro* as well as in animals [41, 95]. Carotenoid metabolites, originating from
220 two-side carotenoid cleavage, were also described as carotenedials *in vitro*, including rosafluene and
221 crocetindial [45], but these have not yet been identified *in vivo* and thereby were also not further
222 investigated regarding physiologically-relevant nuclear hormone mediated-signalling.

223
224 Finally, and possibly most important for the biological activity of carotenoids, centric-cleavage
225 metabolites have been described. These metabolites of β -carotene, α -carotene, and β -cryptoxanthin are
226 the apo-15-carotenoic acids, termed retinoic acids [96]. Retinoic acids are well-known endogenous
227 derivatives, functioning as lipid hormone receptor ligands, responsible for activating two major families
228 of nuclear hormone receptors, i.e. the RARs and RXRs. These receptors can, following ligand-activation,
229 modify transcription of receptor specific genes [22, 23]. The major products are retinoic acids, mainly in
230 the form of ATRA, the endogenous ligand of the RARs (RAR α , β , γ), as reviewed earlier [31].

231 Endogenous levels of ATRA in serum / plasma were in the range of 0.8 – 2.8 ng/ml (2.7 – 9.3 nM) and
232 up to 6 ng/g (20 nM) in the pancreas and 16 ng/g (53 nM) in the liver (Table 2). Thus, these
233 concentrations are at least 1-2 magnitudes lower than those of β -carotene in the bloodstream, with
234 concentrations around 0.1 - 2 μ M (Table 1, [97]). While these centric cleavage products are the main
235 activators of RARs and RXRs [38, 39], the excentric apo-carotenoid apo-13-carotenone is present at
236 lower endogenous levels of 0.8 – 1.3 ng/ml (3–5 nM) and has been demonstrated to act as “antagonist” or
237 low affinity partial agonist or competitive antagonist, but the physiological and nutritional relevance is
238 not yet known [32, 87]. The physiological- and nutritional-relevance of the “antagonism” / partial agonist
239 activity was never convincingly determined for humans, though in *in vitro* experiments, with weak and
240 questionable prediction potential for humans, but is deemed plausible when considering endogenous
241 concentrations in human serum (3-5 nM, Table 2 and Figure 1).

242
243 In addition to ATRA, other geometric isomers were identified endogenously, such as 13-*cis*-, 9,13-*dicis*-,
244 and 9-*cis*-retinoic acid [98-100], with low concentrations (Table 2). A large focus was placed on 9-*cis*-
245 retinoic acid (9CRA), which was postulated as “an” or even “the” endogenous ligand of RXRs (RXR α , β ,
246 γ) [29, 30]. However, this is seen as controversial by the authors / additional experts in the field of
247 retinoid-lipidomics [36, 101, 102] focusing on ultrasensitive retinoid-lipidomics analysis, as its
248 endogenous presence and function as a physiologically-relevant lipid hormone could not be confirmed.
249 Alternative endogenous geometric isomers of retinoic acid, including 13-*cis*-, 9,13-*dicis*- and 11-*cis*-
250 retinoic acid were not described to be of relevant major biological activity mediated via the activation of
251 RARs-RXRs [29]. For retinal, the endogenous cycle between all-*trans*-retinal and 11-*cis*-retinal in the
252 visual cycle in the eye is well established [103, 104], but it is of no systemic relevance for the whole
253 human organism.

254
255 For ATRA, increased serum levels of 1.2 towards 2.0 ng/ml (4.0 \rightarrow 6.7 nM) were found following
256 supplementation of β -carotene-rich foods [37]. Whether these increased serum levels reflect also tissue
257 levels and increased RAR-mediated signalling was and could not be identified. The physiological and

258 nutritional relevance in humans could also not be evaluated. This intervention with food items rich in β -
259 carotene resulted in low and non-significant alterations of interleukin (IL) secretion and immune response
260 as indicators of RAR-mediated signalling [105, 106]. Whether such β -carotene interventions are
261 beneficial for humans is questionable. Interestingly, the strongest effects were identified in the carotenoid
262 wash-out phase prior to intervention, resulting in reduced IL-2, natural killer (NK) cell cytotoxicity and
263 lymphocyte proliferation, a potential consequence of β -carotene (or general carotenoid) or even vitamin
264 A deficiency and possibly reduced RAR-RXR-mediated signalling [106]. These reductions were rapidly
265 recovered after β -carotene- or lycopene-supplementations, likely as a consequence of recovered RAR-
266 RXR-mediated signalling [106]. In animal studies, β -carotene-supplementation resulted in the recovery
267 of vitamin A deficiency indicated by visualized RARE-mediated signalling. In addition serum, but not
268 liver ATRA concentrations were improved, while retinol levels recovered and even increased [20]. It can
269 be assumed that β -carotene supplementation can reinstate basal retinol and ATRA concentrations and
270 RAR-mediated signalling. However, no further increase in ATRA concentrations in organs and enhanced
271 RAR-mediated signalling could be observed as a result of increased storage and transport of retinol due
272 to a highly regulated homeostasis of retinoid / vitamin A / RAR-mediated signalling pathways.

273
274 Nutritionally-relevant β -carotene intake is mainly contributing to the anti-infective properties of vitamin
275 A, which is commonly identified as its major activity besides ocular functions [12, 107]. It is suggested
276 that provitamin A carotenoids are relevant for maintaining vitamin A activity, while being of no further
277 physiologically- and nutritionally-proven relevance.

278
279 In contrast, long term high-dose supplementation of pure synthetic all-*trans*- β -carotene, studied in
280 tobacco smoke exposed ferrets, may alter RAR-RXR-mediated signalling by a negative feedback
281 regulation [108], thereby strongly reducing RAR β - and ATRA levels in the lung, as the target organ [109,
282 110].

284 In addition, it is questionable whether higher than basal RAR-mediated signalling is more beneficial or
285 whether it can be considered as detrimental, while increased RXR-mediated signalling may be considered
286 mainly beneficial [25]. Based on these limited studies we conclude that β -carotene can prevent general
287 vitamin A deficiency [37, 106], reaching a plateau, while higher and pure β -carotene supplementation
288 seems unrelated to improved health status [49]. That moderate or even high dietary consumption of
289 natural food items rich in β -carotene and additional bioactive derivatives including other carotenoids
290 transmits non-beneficial effects seems unlikely.

291
292 Recently, dihydro-metabolites of apo-15-carotenoids were described in mice, likely originating from
293 13,14-dihydroretinol [111] (Figure 1; Table 2). In a larger cohort study, 13,14-dihydroretinol and the
294 novel identified endogenous all-*trans*-13,14-dihydroretinoic acid [112, 113] and 9-*cis*-13,14-
295 dihydroretinoic acid [33, 36, 102] were analysed in human serum [114] as well as adipose tissue (*Rühl et*
296 *al. unpublished*). All-*trans*-13,14-dihydroretinoic acid was described as a medium affinity endogenous
297 RAR-ligand [38, 115] and recently, 9-*cis*-13,14-dihydroretinoic acid (9CDHRA) became a focus of
298 attention, as it appears to be an or even the physiologically- and nutritionally-relevant RXR-ligand in
299 mammals, serving as a novel endogenous lipid hormone [33, 36, 102]. Further nutritionally-relevant
300 precursors of 9CDHRA, such as 9-*cis*-13,14-dihydroretinol, 9-*cis*-dihydrocarotenoids and even the well-
301 known 9-*cis*- β -carotene were recently postulated [116] and confirmed [117] as even being a new
302 independent vitamin A signalling pathway, termed vitamin A5 (Figure 1) [118].

303 304 2.3. Metabolites of lycopene

305 In addition to β -carotene, lycopene is one of the major carotenoids present in the diet, resulting in high
306 tissue and blood concentrations (Figure 1 and Table 1, 2). However, the metabolism of lycopene has been
307 studied to a much lesser extent compared to that of β -carotene and especially when focusing on the
308 human situation.

310 Oxidative metabolism of lycopene and of additional acyclic carotenoids such as phytoene and
311 phytofluene (Table 1) were described [119], while such metabolism was neither **conclusively** observed
312 nor the focus in studies employing lutein and other carotenoids with hydroxyl- / oxo-functional groups,
313 such as zeaxanthin, canthaxanthin, β -cryptoxanthin and astaxanthin, which would have **broader relevance**
314 **for the human situation. Selected xanthophylls were described to interact and block apo-carotenoid**
315 **mediated signalling [120, 121] while no mechanism involving xanthophyll-metabolites was mentioned**
316 **and outlined.** Both excentric and centric metabolism was described for lycopene [40, 41]. Except for
317 lycopenoids, there was no further focus on the identification of potential endogenous derivatives or
318 molecular biological examination to investigate their biological activities [122-124]. Various lycopenals
319 were identified and predicted in the food matrix and in the human organism after a tomato product
320 intervention. Human serum levels were reported to be low (Figure 1 / Table 2 [125]).

321
322 While many studies display a complex pattern of lycopene metabolism via various pathways [40, 125-
323 129], and potential lycopene metabolites were found after supplementing high amounts of lycopene in
324 experimental animal models [124, 130-132], a direct association of human relevance was only recently
325 indirectly concluded [133]. An indirect evidence of lycopene activity and a further lycopene-metabolite
326 for RAR-activation was revealed, using a RARE-luciferase expressing mouse model [20, 134]. Based on
327 RARE-mediated signalling, a partial vitamin A activity following lycopene intervention was found [20].
328 An identification of the involved functional metabolites was only partly achieved, and apo-15-lycopenoic
329 acids were claimed to be present endogenously, especially after lycopene-supplementation [124, 135].

330
331 **Other lycopenoic acids** might also be bioactive, as it was shown earlier in a mouse study that the potential
332 lycopene metabolite apo-10'-lycopenoic acid [131] reduced hepatic fat accumulation [136]. The
333 physiological and nutritional relevance of apo-10'-lycopenoic acid was only shown in ferrets [131], but
334 could not be confirmed *in vivo* in mice and *ex vivo* in humans [130]. Alternatively, due to extensive
335 metabolism, a dihydro-apo-10'-lycopenoic acid analogue was identified and based on UV and MS-
336 characteristics predicted to be 7,8-dihydro-apo-10'-lycopenoic acid. How lycopene is metabolized to

337 dihydro-apo-10'-lycopenoic acid and whether apo-10'-lycopenoic acid is a potential intermediate are yet
338 unanswered questions. Likely these dihydro-apo-10-lycopenoids are direct precursors of dihydro-apo-15-
339 lycopenoids, which might be highly potent RAR- and / or RXR-ligands, as postulated in [124].

341 2.4. Summary for carotenoid metabolites

342 Thus, for many metabolites it remains inconclusive whether they derive from human metabolism or are
343 ingested via animal origin as pre-formed carotenoid metabolites in the forms of retinol and mainly retinyl
344 esters [12, 137]. In addition, the biological function and the concentration-dependent activity of various
345 carotenoid metabolites besides ATRA has generally not been studied, mostly due to the lack of available
346 standard compounds and established sensitive and selective analytical methods. Furthermore, the direct
347 link between carotenoid intake and RAR-RXR-mediated transcriptional signalling as a multi-step
348 procedure has not yet been proven. However, each step of this cascade has been clearly demonstrated
349 with experimental data: a) higher carotenoid supplementation resulting in higher carotenoid levels in
350 supplemented individuals [105, 138]. b) higher β -carotene levels correlating and resulting in increased
351 ATRA concentrations [37, 49], c) higher ATRA levels causing increased RAR-mediated signalling [134];
352 and d) higher RAR-mediated signalling resulting in increased individual specific immune responses [52,
353 77, 139, 140] and altered lipid metabolism [141, 142], with partially beneficial or detrimental effects.

354
355 Recently, a novel class of bioactive carotenoid-metabolites, namely strigolactones, was described to be
356 enzymatically synthesized in certain plants, such as carlactones [143-145] and identified as plant-relevant
357 hormones during germination [143] and branching-inhibition [146]. If these derivatives are also of direct
358 or indirect relevance for the human organisms remains speculative.

359
360 In summary, human supplementation studies with food items rich in β -carotene / lycopene or
361 supplemented β -carotene / lycopene, focusing on multi-targeted analyses, and identifying β -carotene /
362 lycopene and retinoid concentrations and further RARE-mediated signalling, have not yet been
363 performed and should be addressed. Due to the access of multi-omic techniques, serum markers or novel

transcriptional markers of diseases [147, 148], possibly also co-associated with vitamin A / carotenoid deficiency or reduced RAR-RXR-mediated dysfunction [25, 149], should be compared to carotenoid intake and serum / plasma carotenoid / retinoid concentrations to obtain valuable correlations.

3. Discussion and Perspectives

Several carotenoids are implicated in health-related outcomes, from AMD (lutein and zeaxanthin) to possible effects regarding cardio-metabolic diseases (predominantly, β -carotene) and diabetes / cancer (predominantly, lycopene). The dietary intake of carotenoids has also changed over time. While lycopene intake was uncommon in the pre-industrialized human diet, especially considering the primarily European-focused world sight, it strongly increased in the Western society, due to a high consumption of tomatoes and tomato products [150].

Additionally, it became obvious that light irradiation [151] and more practically relevant thermal food processing [152], as also reviewed by Khoo et al. [153], including cooking $>100^{\circ}\text{C}$ appears to constitute an important mechanisms for carotenoid isomerisation, yielding different precursor carotenoids for different functional apo-carotenoids, as well as a non-endogenous human generated apo-carotenoids, serving as easy accessible substrates for functional apo-carotenoids [154]. This highlights cooking and food-processing as important cultural achievement for generating bioactive derivatives for enabling a healthy and well-functioning human organism [155].

However, carotenoids are generally considered as lipid precursors (mainly for bioactive vitamin A / retinoids) in the diet, while their complex and multi-step metabolic pathways and the relationship to health beneficial effects are still poorly understood. In this review we summarised all available relevant information focussing on the human organism with implication of mechanistic results from further *in vitro* to *in vivo* experiments. Unfortunately, these experimental results are difficult to compare with the human because carotenoids obtain a non-similar nutri-kinetics pattern [156] and different eating

behaviour in contrast to the pure vegetarian dietary pattern of rodents, which are frequently used as experimental animal models .

4. Conclusions

As a cornerstone, we suggest that besides benchmark concentrations for carotenoids, also retinoids should be suggested, including both “normal” and deficiency threshold ranges. These ranges should correlate with well-defined and established nuclear hormone receptor signalling cascade markers, disease markers, prognostic early markers of diseases as well as markers of impairments of physiologically-important functions based on novel “omics” markers such as transcriptomics, lipidomics and proteomics, which are now frequently published for various target diseases [157]. In the case of diseases and dysfunctions related to carotenoid and vitamin A deficiency, underlying molecular mechanisms such as RAR-RXR- / RXR-plus additional nuclear hormone receptor (NHR)-dysfunctional signalling [22, 25, 158] (i.e. signalling not associated with a healthy condition as present in various diseases of the Western society), should also be considered.

Based on these two ranges, targeted supplementation strategies may be recommended to overcome deficiencies and towards reaching and maintaining “normal” concentration ranges. A correlation between dietary intake, serum levels and bioactive carotenoid metabolites and further examination of RXR-RAR / RXR-NHR in an easy accessible compartment such as peripheral blood mononuclear cells (PBMCs), plus target genes of relevant diseases are desperately missing in carotenoid / retinoid nutritional research.

The basal benchmark concentration indicating a higher risk for chronic diseases appears to constitute a total carotenoid plasma / serum concentration <1.000 nM and should further focus on endogenous retinoids. The second benchmark concentration reflecting “normal” carotenoid intake are average plasma / serum concentrations of individual and total carotenoids indicating, and here defined as, a healthy varied diet. Such levels can then be translated into the intake of relevant food items rich in carotenoids,

418 based on correlations between reported average intakes for β -carotene and lycopene with serum
419 concentrations and considering intervention with carotenoid-rich foods [97].
420

421 In this review article, we summarised the current mechanisms of carotenoid metabolism including
422 reference levels of bioactive carotenoid metabolites with pure relevance on the human organism. To
423 summarise, carotenoid to bioactive metabolite metabolism is an important knowledge to justify which
424 biological-response pathway of carotenoids is enabled to elicit valuable beneficial effects. This is
425 important in order to evaluate if there might be a problem in individual dietary intake of food enriched in
426 specific carotenoids is present or if a genetic hereditary problem in metabolism of carotenoids to
427 bioactive carotenoids based on genetic polymorphisms is the cause of disturbed occurrence of bioactive
428 carotenoid metabolites.

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443 **Table 1.** Concentrations of carotenoids in various tissues, all data in nM (nmol/kg or L), adapted
 444 from [97].

tissue	BCAR	ACAR	BCRY	LYC	PHYE	PHYF	ref.
serum / plasma	360 ± 10	120 ± 10	230 ± 10	740 ± 10	40 ± 20	170 ± 70	[159]
serum / plasma	ATBC: 823 ± 277 9CBC 22 ± 13 13CBC 29 ± 22 sum: 874			ATLYC 190 ± 25 5CLYC 130 ± 20 9CLYC 9 ± 5 13/15CLYC 55 ± 25 sum: 384			[160, 161]
abdominal adipose tissue	1472 ± 286	280 ± 74	417 ± 462	3329 ± 448			[162]
liver	5900 ± 6300			8400 ± 11,500			[50]
skin [§]	430 ± 45	95 ± 20	225 ± 35	695 ± 45	320 ± 90	46 ± 20	[163-165]
lung	350 ± 440	230 ± 270	420 ± 750	570 ± 1110			[166]
kidney	550 ± 730	300 ± 400	450 ± 1040	620 ± 620			[166]
brain [§]	10-30		<10				[167]
adrenals	5600 *** (680-31,830)	1220 *** (110-7520)	660 *** (10-2900)	1900 *** (190-5600)			[168]
testes	2680 *** (750-4770)	370 *** (140-610)	160 *** (10-290)	4340 *** (410-9380)			[168]
bone	745 ± 95	95 ± 35	125 ± 35	280 ± 35	825 ± 185	275 ± 45	[165]
colon tissue	60 ± 30						[169]
breast milk	60-200	20-40	2-10	5-25			[170]
uterus	503 [‡]	870					[171]
prostate	600		300	100	700		[172]

445
 446 All values represent mean ± SD; “blank” represents non determined carotenoids or no data available; [§]:
 447 infants, prefrontal cortex, frontal cortex, hippocampus, auditory cortex and occipital cortex; [‡]: values given in
 448 literature as “carotenes”; [&]: dermis and epidermis of back, forehead, inner forearm and hand; *** : including
 449 upper and lower level of this range; ACAR: α-carotene; BCAR: β-carotene; BCRY: β-cryptoxanthin; LYC:
 450 lycopene; PHYE: phytoene; PHYF: phytofluene.
 451

452 **Table 2.** Levels of major carotenoid metabolites / retinoids in blood plasma / serum and
 453 tissues including molecular weight in Dalton (Da) and molecular formula of each retinoid.

Carotenoid metabolite	parent carotenoid	serum/ tissue	metabolite concentration in ng/g (ml) in nM		remark	ref.	occurrence
all-trans-retinoic acid (C ₂₀ H ₂₈ O ₂ / 300.4 Da)	BCAR	serum	2.8 ± 0.8 ng/ml	9.3 ± 2.6 nM	*** German children Turkish children *** *** fasted adults -before, -after suppl. of food rich in BCAR	[173] [67] [67] [35] [174] [175]	end.
			1.4 ± 2.3 ng/ml	4.7 ± 7.7 nM			
			1.2 ± 1.6 ng/ml	4.0 ± 5.3 nM			
			1.4 ± 0.3 ng/ml	4.7 ± 1.0 nM			
			0.8 ± 0.2 ng/ml	2.7 ± 0.7 nM			
			0.9 ± 0.2 ng/ml	3.0 ± 0.6 nM			
			1.2 ± 0.3 ng/ml	4.0 ± 1.0 nM		[37]	
		skin	0.7 / 2.1 ng/g	2.3 / 7.0 nM	***	[173]	end.
		pancreas	5.9 ± 2.1 ng/g	19.7 ± 7.0 nM	***	[176]	end.
		liver	15.8 ± 8.3 ng/g	52.7 ± 27.7 nM	***	[35]	end.
9-cis-retinoic acid (*)	BCAR	serum	(*) 0.03 ng/ml	(*) 0.1 nM	fasted adults	[175]	end.
		liver	(*) 0.6 ± 0.2 ng/g	(*) 2.0 ± 0.7 nM	***	[35]	end.
13-cis-retinoic acid (#)	BCAR	serum	1.8 ± 1.0 ng/ml	6.0 ± 3.3 nM	*** *** fasted adults	[35] [174] [175]	end.
			1.1 ± 0.2 ng/ml	3.7 ± 0.7 nM			
			1.2 ± 0.3 ng/ml	3.9 ± 1.0 nM			
		liver	1.5 ± 0.4 ng/g	5.0 ± 1.3 nM	***	[35]	end.
9,13-dicis-retinoic acid (#)		serum	1.0 ± 0.3 ng/ml	0.3 ± 0.1 nM	fasted adults	[175]	end.
all-trans-13,14-dihydroretinoic acid (C ₂₀ H ₃₀ O ₂ / 302.5 Da)	BCAR	serum	ecnd		***	[117]	end.
		pancreas	88 ± 72 ng/g	290 ± 238 nM	***	[113]	end.
9-cis-13,14-dihydroretinoic acid	BCAR	serum	4.8 ± 0.7 ng/ml	15.8 ± 2.3 nM	***	[114, 117]	end.
9-cis-4-oxo-13,14-dihydroretinoic acid (C ₂₀ H ₂₈ O ₃ / 316.4 Da)	BCAR	liver	10.3 ng/g	32.6 nM	***	[35]	end.
all-trans-4-oxo-retinoic acid (C ₂₀ H ₂₆ O ₃ / 314.4 Da)	BCAR/CA ₍₁₎	serum	0.6 ± 0.3 ng/ml	1.9 ± 0.9 nM	***	[174]	end.
13-cis-4-oxo-retinoic acid	BCAR/CA ₍₁₎	serum	2.4 ± 1.8 ng/ml	7.6 ± 5.7 nM	***	[35]	end.
all-trans-apo-13'-carotenone (C ₁₈ H ₂₇ O ₁ / 255.4 Da)	BCAR	serum	0.8 – 1.3 ng/ml	3 - 5 nM	***	[32]	end.
all-trans-apo-14'-carotenonic acid (C ₂₂ H ₂₃ O ₂ / 323.5 Da)	BCAR	serum	1,3 ± 0,6 ng/ml	4,0 ± 1,9 nM	***	[177]	end.
all-trans-retinoyl-glucuronide (C ₂₆ H ₃₆ O ₈ / 476.6 Da)	BCAR	serum	3.2 ± 1.9 ng/ml	6.8 ± 4.0 nM	***	[178]	end.
all-trans-retinol (C ₂₀ H ₃₀ O ₁ / 286.5 Da)	BCAR	serum	510 ± 217 ng/ml	1777 ± 759 nM	*** *** ***	[173] [35] [174]	end.
			980 ± 110 ng/ml	3414 ± 383 nM			
			641 ± 99 ng/ml	2233 ± 345 nM			
		skin	207 / 253 ng/g	724 / 885 nM	***	[173]	end.
		pancreas	96 ± 86 ng/g	336 ± 301 nM	***	[176]	end.
all-trans-3,4-dehydroretinol (C ₂₀ H ₂₈ O ₁ / 288.5 Da)	BCAR	skin	9 - 70 ng/g	31.2 – 240 nM	** , ***	[179]	end.
all-trans-13,14-dihydroretinol (C ₂₀ H ₃₂ O ₁ / 284.4 Da)	BCAR	serum	ecnd		***	[117]	end.
9-cis-13,14-dihydroretinol	BCAR	serum	0.9 ng/ml	3.1 nM	***	[117]	end.
all-trans-retinal (C ₂₀ H ₂₈ O ₁ / 284.4 Da)	BCAR	eye	ecnd		***	[103]	end.
11-cis-retinal	BCAR	eye	ecnd		***	[103]	end.
apo-6'-lycopenal (C ₃₂ H ₄₂ O ₁ / 443.1 Da)	LYC	plasma	<0.1 ± <0.0 ng/ml	0.1 ± 0.1 nM	after tomato suppl. diet	[125]	after supp.
apo-8'-lycopenal (C ₃₀ H ₄₀ O ₁ / 416.6 Da)	LYC	plasma	0.2 ± 0.1 ng/ml	0.6 ± 0.3 nM	"	[125]	after supp.
apo-10'-lycopenal (C ₂₇ H ₃₆ O ₁ / 376.6 Da)	LYC	plasma	0.1 ± <0.0 ng/ml	0.3 ± 0.1 nM	"	[125]	after supp.
apo-12'-lycopenal (C ₂₅ H ₃₄ O ₁ / 350.1 Da)	LYC	plasma	0.2 ± 0.1 ng/ml	0.7 ± 0.4 nM	"	[125]	after supp.
apo-14'-lycopenal (C ₂₂ H ₃₀ O ₁ / 310.5 Da)	LYC	plasma	0.03 ± <0.0 ng/ml	0.1 ± <0.0 nM	"	[125]	after supp.

References:

just in mouse

all-trans-retinal (C ₂₀ H ₂₈ O ₁ / 284.4 Da)	BCAR	serum (m)	0.6 ng/ml 9.1 ± 1.8 ng/ml	~2 nM 32.2 ± 6.2 nM		[180] [181]	just mouse
		WAT (m)	~8.5 – 11.4 ng/g 17.9 ± 1.4 ng/g	~30 - 40 nM 63 ± 5 nM		[180] [181]	just mouse
all-trans-retinol	BCAR	serum (m)	257 ± 31 ng/ml 170 ± 10 ng/ml	900 ± 110 nM 595 ± 35 nM	****	[181] [35]	
9-cis-retinol	BCAR	serum (m)	8.6 ± 2.9 ng/ml	30 ± 10 nM		[181]	just mouse
13-cis-retinol	BCAR	serum (m)	11.4 ± 2.9 ng/ml	40 ± 10 nM		[181]	just mouse
dihydro-apo-10'- lycopenoic acid (C ₂₇ H ₃₈ O ₂ / 394.6 Da)	LYC	WAT (m)	?	?	*****	[130]	just mouse

454
 455 *: likely just an isomerisation product of ATRA during sample preparation; **: present in different concentrations
 456 in different zones of the human skin; ***: healthy adults; ****: all-trans-retinol levels in mouse are just used as
 457 reference for comparison to 9-cis- and 13-cis-retinol levels, which were just determined in mouse serum and not in
 458 humans; *****: derivatives which were predicted by analytical studies ; #: 9,13-dicis- and 13-cis-retinoic acid
 459 usually co-elute during HPLC-separation and are not identified separately in many described studies; ##: this
 460 concentration is based on a total amount of 0.1 ng in a retina, which is calculated on a predicted retina weight of 10-
 461 80 mg; (1): 4-oxo-retinoic acid was described as an *in vitro* metabolite of canthaxanthin [182]; BCAR: β-carotene;
 462 LYC: lycopene; CA: canthaxanthin; WAT: white adipose tissue; end.: endogenous; supp.: supplementation, ecnd –
 463 exact concentration was not determined.
 464

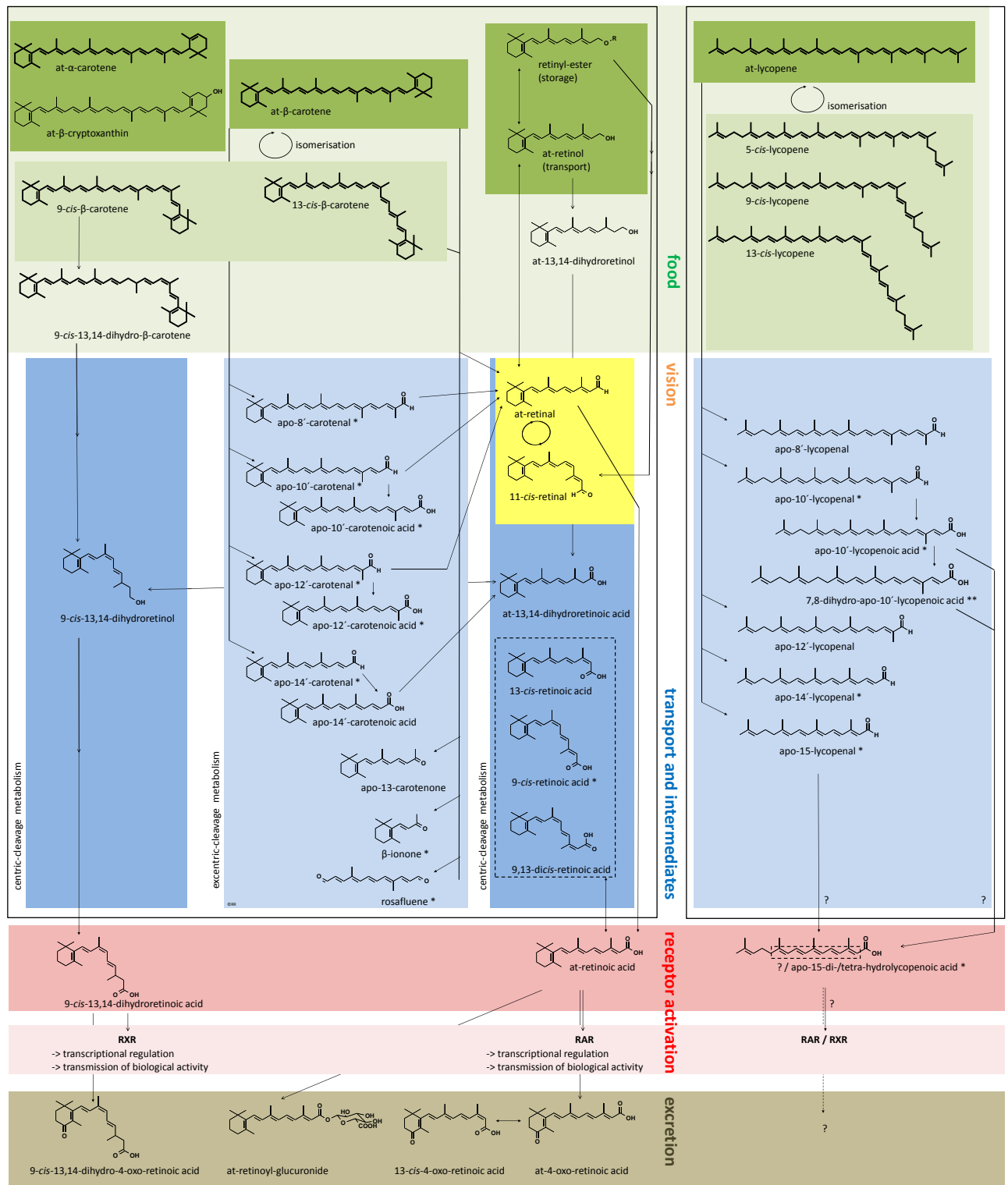
465 **Figure headings**

466

467 **Figure 1:** Metabolic pathway starting from all-*trans*- β -carotene and all-*trans*-lycopene via a)
468 geometric isomerisation, b) excentric cleavage metabolism and c) centric-cleavage mechanisms.

469 Starting from food, towards transport and intermediate derivatives, nuclear hormone receptor
470 activating ligands including further regulation of transcription and thereby major mediation of
471 biological signalling of carotenoids and further deactivation / excretion metabolites. Arrows in
472 the figure indicate potential and simplified metabolic pathways. Derivatives which were not
473 conclusively identified to be present endogenously in humans were marked with a star (*) and
474 represent derivatives which were suggested as metabolites and identified in *in vitro* or *in vivo*
475 experimental approaches. Additional derivatives which were predicted based on analytical
476 studies were indicated by two stars (**). Abbreviations: AT: all-*trans*-, RAR: retinoic acid
477 receptor, RXR: retinoid-X receptor.

478



479
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Figure 1

485 **5 References**

486

- 487 1. Yabuzaki J: **Carotenoids Database: structures, chemical fingerprints and distribution among**
488 **organisms**. *Database (Oxford)* 2017, **2017**(1).
- 489 2. Rodriguez-Concepcion M, Avalos J, Bonet ML, Boronat A, Gomez-Gomez L, Hornero-Mendez D,
490 Limon MC, Meléndez-Martínez AJ, Olmedilla-Alonso B, Palou A *et al*: **A global perspective on**
491 **carotenoids: Metabolism, biotechnology, and benefits for nutrition and health**. *Prog Lipid Res*
492 2018, **70**:62-93.
- 493 3. Felemban A, Braguy J, Zurbriggen MD, Al-Babili S: **Apocarotenoids Involved in Plant**
494 **Development and Stress Response**. *Front Plant Sci* 2019, **10**:1168.
- 495 4. Harrison EH, Quadro L: **Apocarotenoids: Emerging Roles in Mammals**. *Annu Rev Nutr* 2018,
496 **38**:153-172.
- 497 5. Sluijs I, Cadier E, Beulens JW, van der AD, Spijkerman AM, van der Schouw YT: **Dietary intake of**
498 **carotenoids and risk of type 2 diabetes**. *Nutr Metab Cardiovasc Dis* 2015, **25**(4):376-381.
- 499 6. Zhou H, Zhao X, Johnson EJ, Lim A, Sun E, Yu J, Zhang Y, Liu X, Snelling T, Shang F *et al*: **Serum**
500 **Carotenoids and Risk of Age-Related Macular Degeneration in a Chinese Population Sample**.
501 *Invest Ophthalmol Visual Sci* 2011, **52**(7):4338-4344.
- 502 7. Dulińska-Litewka J, Hałubiec P, Łazarczyk A, Szafranski O, Sharoni Y, McCubrey JA, Gąsioriewicz
503 B, Bohn T: **Recent Progress in Discovering the Role of Carotenoids and Metabolites in**
504 **Prostatic Physiology and Pathology-A Review-Part II: Carotenoids in the Human Studies**.
505 *Antioxidants (Basel, Switzerland)* 2021, **10**(2):319.
- 506 8. Zhao LG, Zhang QL, Zheng JL, Li HL, Zhang W, Tang WG, Xiang YB: **Dietary, circulating beta-**
507 **carotene and risk of all-cause mortality: a meta-analysis from prospective studies**. *Sci Rep*
508 2016, **6**:26983.
- 509 9. Krinsky NI, Johnson EJ: **Carotenoid actions and their relation to health and disease**. *Mol*
510 *Aspects Med* 2005, **26**(6):459-516.
- 511 10. Bohn T: **Carotenoids and markers of oxidative stress in human observational studies and**
512 **intervention trials – implications for chronic diseases**. *Antioxidants* 2019, **8**:pii: E179.
- 513 11. Bonet ML, Canas JA, Ribot J, Palou A: **Carotenoids in adipose tissue biology and obesity**.
514 *Subcell Biochem* 2016, **79**:377-414.
- 515 12. Rühl R: **Effects of dietary retinoids and carotenoids on immune development**. *Proc Nutr Soc*
516 2007, **66**(3):458-469.
- 517 13. Mounien L, Tourniaire F, Landrier J-F: **Anti-Obesity Effect of Carotenoids: Direct Impact on**
518 **Adipose Tissue and Adipose Tissue-Driven Indirect Effects**. *Nutrients* 2019, **11**(7):1562.
- 519 14. Amengual J, Widjaja-Adhi MA, Rodriguez-Santiago S, Hessel S, Golczak M, Palczewski K, von
520 Lintig J: **Two carotenoid oxygenases contribute to mammalian provitamin A metabolism**. *J*
521 *Biol Chem* 2013, **288**(47):34081-34096.
- 522 15. Linnewiel K, Ernst H, Caris-Veyrat C, Ben-Dor A, Kampf A, Salman H, Danilenko M, Levy J,
523 Sharoni Y: **Structure activity relationship of carotenoid derivatives in activation of the**
524 **electrophile/antioxidant response element transcription system**. *Free Radic Biol Med* 2009,
525 **47**(5):659-667.
- 526 16. Linnewiel-Hermoni K, Motro Y, Miller Y, Levy J, Sharoni Y: **Carotenoid derivatives inhibit**
527 **nuclear factor kappa B activity in bone and cancer cells by targeting key thiol groups**. *Free*
528 *Radic Biol Med* 2014, **75**:105-120.

- 529 17. Nidhi B, Sharavana G, Ramaprasad TR, Vallikannan B: **Lutein derived fragments exhibit higher antioxidant and anti-inflammatory properties than lutein in lipopolysaccharide induced inflammation in rats.** *Food Funct* 2015, **6**(2):450-460.
- 530
- 531
- 532 18. Farkhondeh T, Samarghandian S: **The effect of saffron (*Crocus sativus* L.) and its ingredients on the management of diabetes mellitus and dislipidemia.** *Afr J Pharm Pharmacol* 2014, **8**:541-549.
- 533
- 534
- 535 19. Bruzzone S, Ameri P, Briatore L, Mannino E, Basile G, Andraghetti G, Grozio A, Magnone M, Guida L, Scarfi S *et al*: **The plant hormone abscisic acid increases in human plasma after hyperglycemia and stimulates glucose consumption by adipocytes and myoblasts.** *The FASEB Journal* 2012, **26**(3):1251-1260.
- 536
- 537
- 538
- 539 20. Aydemir G, Kasiri Y, Bartok EM, Birta E, Frohlich K, Bohm V, Mihaly J, Rühl R: **Lycopene supplementation restores vitamin A deficiency in mice and possesses thereby partial provitamin A activity transmitted via RAR signaling.** *Mol Nutr Food Res* 2016, **60**(11):2413-2420.
- 540
- 541
- 542 21. Dingeo G, Brito A, Samouda H, La Frano MR, Bohn T: **Phytochemicals as Modifiers of Gut Microbial Communities.** *Food and Function* 2020, **11**:8444-8471.
- 543
- 544 22. Evans RM, Mangelsdorf DJ: **Nuclear receptors, RXR, and the big bang.** *Cell* 2014, **157**(1):255-266.
- 545
- 546 23. Mangelsdorf DJ, Thummel C, Beato M, Herrlich P, Schutz G, Umesono K, Blumberg B, Kastner P, Mark M, Chambon P *et al*: **The nuclear receptor superfamily: the second decade.** *Cell* 1995, **83**(6):835-839.
- 547
- 548
- 549 24. Balmer JE, Blomhoff R: **Gene expression regulation by retinoic acid.** *J Lipid Res* 2002, **43**(11):1773-1808.
- 550
- 551 25. Desvergne B: **RXR: from partnership to leadership in metabolic regulations.** *Vitam Horm* 2007, **75**:1-32.
- 552
- 553 26. Tourniaire F, Musinovic H, Gouranton E, Astier J, Marcotorchino J, Arreguin A, Bernot D, Palou A, Bonet ML, Ribot J *et al*: **All-trans retinoic acid induces oxidative phosphorylation and mitochondria biogenesis in adipocytes.** *J Lipid Res* 2015, **56**(6):1100-1109.
- 554
- 555
- 556 27. Karkeni E, Bonnet L, Astier J, Couturier C, Dalifard J, Tourniaire F, Landrier JF: **All-trans-retinoic acid represses chemokine expression in adipocytes and adipose tissue by inhibiting NF- κ B signaling.** *J Nutr Biochem* 2017, **42**:101-107.
- 557
- 558
- 559 28. Mangelsdorf DJ, Ong ES, Dyck JA, Evans RM: **Nuclear receptor that identifies a novel retinoic acid response pathway.** *Nature* 1990, **345**(6272):224-229.
- 560
- 561 29. Levin AA, Sturzenbecker LJ, Kazmer S, Bosakowski T, Huselton C, Allenby G, Speck J, Kratzeisen C, Rosenberger M, Lovey A *et al*: **9-cis retinoic acid stereoisomer binds and activates the nuclear receptor RXR alpha.** *Nature* 1992, **355**(6358):359-361.
- 562
- 563
- 564 30. Heyman RA, Mangelsdorf DJ, Dyck JA, Stein RB, Eichele G, Evans RM, Thaller C: **9-cis retinoic acid is a high affinity ligand for the retinoid X receptor.** *Cell* 1992, **68**(2):397-406.
- 565
- 566 31. Petkovich M, Brand NJ, Krust A, Chambon P: **A human retinoic acid receptor which belongs to the family of nuclear receptors.** *Nature* 1987, **330**(6147):444-450.
- 567
- 568 32. Eroglu A, Hruszkewycz DP, dela Sena C, Narayanasamy S, Riedl KM, Kopec RE, Schwartz SJ, Curley RW, Jr., Harrison EH: **Naturally occurring eccentric cleavage products of provitamin A beta-carotene function as antagonists of retinoic acid receptors.** *J Biol Chem* 2012, **287**(19):15886-15895.
- 569
- 570
- 571
- 572 33. Krezel W, Rühl R, de Lera AR: **Alternative retinoid X receptor (RXR) ligands.** *Mol Cell Endocrinol* 2019, **491**:110436.
- 573

- 574 34. Schierle S, Merk D: **Therapeutic modulation of retinoid X receptors - SAR and therapeutic**
575 **potential of RXR ligands and recent patents.** *Expert Opin Ther Pat* 2019, **29**(8):605-621.
- 576 35. Schmidt CK, Brouwer A, Nau H: **Chromatographic analysis of endogenous retinoids in tissues**
577 **and serum.** *Anal Biochem* 2003, **315**(1):36-48.
- 578 36. de Lera AR, Krezel W, Rühl R: **An endogenous mammalian retinoid X receptor ligand, at last!**
579 *ChemMedChem* 2016, **11**(10):1027-1037.
- 580 37. Rühl R, Bub A, Watzl B: **Modulation of plasma all-trans retinoic acid concentrations by the**
581 **consumption of carotenoid-rich vegetables.** *Nutrition* 2008, **24**(11-12):1224-1226.
- 582 38. Allenby G, Bocquel MT, Saunders M, Kazmer S, Speck J, Rosenberger M, Lovey A, Kastner P,
583 Grippo JF, Chambon P *et al*: **Retinoic acid receptors and retinoid X receptors: interactions with**
584 **endogenous retinoic acids.** *Proc Natl Acad Sci U S A* 1993, **90**(1):30-34.
- 585 39. Allenby G, Janocha R, Kazmer S, Speck J, Grippo JF, Levin AA: **Binding of 9-cis-retinoic acid and**
586 **all-trans-retinoic acid to retinoic acid receptors alpha, beta, and gamma. Retinoic acid**
587 **receptor gamma binds all-trans-retinoic acid preferentially over 9-cis-retinoic acid.** *J Biol*
588 *Chem* 1994, **269**(24):16689-16695.
- 589 40. dela Sena C, Narayanasamy S, Riedl KM, Curley RW, Jr., Schwartz SJ, Harrison EH: **Substrate**
590 **specificity of purified recombinant human beta-carotene 15,15'-oxygenase (BCO1).** *J Biol*
591 *Chem* 2013, **288**(52):37094-37103.
- 592 41. dela Sena C, Sun J, Narayanasamy S, Riedl KM, Yuan Y, Curley RW, Jr., Schwartz SJ, Harrison EH:
593 **Substrate specificity of purified recombinant chicken beta-carotene 9',10'-oxygenase (BCO2).**
594 *J Biol Chem* 2016, **291**(28):14609-14619.
- 595 42. Barua AB, Olson JA: **Retinoyl beta-glucuronide: an endogenous compound of human blood.**
596 *Am J Clin Nutr* 1986, **43**(4):481-485.
- 597 43. Sass JO, Forster A, Bock KW, Nau H: **Glucuronidation and isomerization of all-trans- and 13-cis-**
598 **retinoic acid by liver microsomes of phenobarbital- or 3-methylcholanthrene-treated rats.**
599 *Biochem Pharmacol* 1994, **47**(3):485-492.
- 600 44. Radomska A, Little JM, Lehman PA, Samokyszyn V, Rios GR, King CD, Green MD, Tephly TR:
601 **Glucuronidation of retinoids by rat recombinant UDP: glucuronosyltransferase 1.1 (bilirubin**
602 **UGT).** *Drug Metab Dispos* 1997, **25**(7):889-892.
- 603 45. Amengual J, Lobo GP, Golczak M, Li HN, Klimova T, Hoppel CL, Wyss A, Palczewski K, von Lintig
604 J: **A mitochondrial enzyme degrades carotenoids and protects against oxidative stress.** *Faseb J*
605 2011, **25**(3):948-959.
- 606 46. von Lintig J, Vogt K: **Filling the gap in vitamin A research. Molecular identification of an**
607 **enzyme cleaving beta-carotene to retinal.** *J Biol Chem* 2000, **275**(16):11915-11920.
- 608 47. Kiefer C, Hessel S, Lampert JM, Vogt K, Lederer MO, Breithaupt DE, von Lintig J: **Identification**
609 **and characterization of a mammalian enzyme catalyzing the asymmetric oxidative cleavage**
610 **of provitamin A.** *J Biol Chem* 2001, **276**(17):14110-14116.
- 611 48. Caris-Veyrat C, Schmid A, Carail M, Böhm V: **Cleavage products of lycopene produced by in**
612 **vitro oxidations: characterization and mechanisms of formation.** *J Agric Food Chem* 2003,
613 **51**(25):7318-7325.
- 614 49. Bohn T, Desmarchelier C, El SN, Keijer J, van Schothorst E, Rühl R, Borel P: **beta-Carotene in the**
615 **human body: metabolic bioactivation pathways - from digestion to tissue distribution and**
616 **excretion.** *Proc Nutr Soc* 2019, **78**(1):68-87.

- 617 50. Bohn T, Desmarchelier C, Dragsted LO, Nielsen CS, Stahl W, Ruhl R, Keijer J, Borel P: **Host-**
618 **related factors explaining interindividual variability of carotenoid bioavailability and tissue**
619 **concentrations in humans.** *Mol Nutr Food Res* 2017, **61**:1600685.
- 620 51. Bohn T, Bonet ML, Borel P, Keijer J, Landrier JF, Milisav I, Ribot J, Riso P, Winklhofer-Roob B,
621 Sharoni Y *et al*: **Mechanistic aspects of carotenoid health benefits - where are we now?** *Nutr*
622 *Res Rev* 2021:1-27.
- 623 52. Rubin LP, Ross AC, Stephensen CB, Bohn T, Tanumihardjo SA: **Metabolic effects of**
624 **inflammation on vitamin A and carotenoids in humans and animal models.** *Adv Nutr* 2017,
625 **8(2):197-212.**
- 626 53. Al Senaidy AM: **Serum vitamin A and beta-carotene levels in children with asthma.** *J Asthma*
627 2009, **46(7):699-702.**
- 628 54. Nogueira C, Borges F, Lameu E, Franca C, Rosa CL, Ramalho A: **Retinol, beta-carotene and**
629 **oxidative stress in systemic inflammatory response syndrome.** *Revista da Associacao Medica*
630 *Brasileira (1992)* 2015, **61(2):116-120.**
- 631 55. Akhtar S, Ahmed A, Randhawa MA, Atukorala S, Arlappa N, Ismail T, Ali Z: **Prevalence of vitamin**
632 **A deficiency in South Asia: causes, outcomes, and possible remedies.** *J Health Popul Nutr*
633 2013, **31(4):413-423.**
- 634 56. EFSA Panel on Dietetic Products N, and Allergies (NDA), European Food Safety Authority
635 (EFSA): **Scientific opinion on dietary reference values for vitamin A.** *EFSA Journal* 2015,
636 **13:4028.**
- 637 57. Biehler E, Alkerwi A, Hoffmann L, Krause E, Guillaume M, Lair ML, Bohn T: **Contribution of**
638 **violaxanthin, neoxanthin, phytoene and phytofluene to total carotenoid intake: Assessment**
639 **in Luxembourg.** *J Food Comp Anal* 2012, **25:56-65.**
- 640 58. Aage S, Kiraly N, Da Costa K, Byberg S, Bjerregaard-Andersen M, Fisker AB, Aaby P, Benn CS:
641 **Neonatal vitamin A supplementation associated with increased atopy in girls.** *Allergy* 2015,
642 **70(8):985-994.**
- 643 59. Rühl R: **Non-pro-vitamin A and pro-vitamin A carotenoids in atopy development.** *Int Arch*
644 *Allergy Immunol* 2013, **161(2):99-115.**
- 645 60. Milner JD, Stein DM, McCarter R, Moon RY: **Early infant multivitamin supplementation is**
646 **associated with increased risk for food allergy and asthma.** *Pediatrics* 2004, **114(1):27-32.**
- 647 61. Michaelsson K, Lithell H, Vessby B, Melhus H: **Serum retinol levels and the risk of fracture.** *N*
648 *Engl J Med* 2003, **348(4):287-294.**
- 649 62. Ruiz-Castell M, Le Coroller G, Landrier JF, Kerkour D, Weber B, Fagherazzi G, Appenzeller BMR,
650 Vaillant M, Bohn T: **Micronutrients and Markers of Oxidative Stress and Inflammation Related**
651 **to Cardiometabolic Health: Results from the EHES-LUX Study.** *Nutrients* 2020, **13(1):5.**
- 652 63. Crandall C: **Vitamin A intake and osteoporosis: a clinical review.** *J Womens Health (Larchmt)*
653 2004, **13(8):939-953.**
- 654 64. Mihaly J, Gericke J, Lucas R, de Lera AR, Alvarez S, Torocsik D, Ruhl R: **TSLP expression in the**
655 **skin is mediated via RARgamma-RXR pathways.** *Immunobiology* 2016, **221(2):161-165.**
- 656 65. Rühl R, Taner C, Schweigert FJ, Wahn U, Gruber C: **Serum carotenoids and atopy among**
657 **children of different ethnic origin living in Germany.** *Pediatr Allergy Immunol* 2010,
658 **21(7):1072-1075.**
- 659 66. Gericke J, Ittensohn J, Mihaly J, Dubrac S, Ruhl R: **Allergen-induced dermatitis causes**
660 **alterations in cutaneous retinoid-mediated signaling in mice.** *PLoS One* 2013, **8(8):e71244.**

- 661 67. Gruber C, Taner C, Mihaly J, Matricardi PM, Wahn U, Ruhl R: **Serum retinoic acid and atopy**
662 **among children of different ethnic origin living in Germany.** *J Pediatr Gastroenterol Nutr* 2012,
663 **54(4):558-560.**
- 664 68. Wei S, Kozono S, Kats L, Nechama M, Li W, Guarnerio J, Luo M, You MH, Yao Y, Kondo A *et al*:
665 **Active Pin1 is a key target of all-trans retinoic acid in acute promyelocytic leukemia and**
666 **breast cancer.** *Nat Med* 2015, **21(5):457-466.**
- 667 69. Connolly RM, Nguyen NK, Sukumar S: **Molecular pathways: current role and future directions**
668 **of the retinoic acid pathway in cancer prevention and treatment.** *Clin Cancer Res* 2013,
669 **19(7):1651-1659.**
- 670 70. Tan X, Sande JL, Pufnock JS, Blattman JN, Greenberg PD: **Retinoic acid as a vaccine adjuvant**
671 **enhances CD8+ T cell response and mucosal protection from viral challenge.** *J Virol* 2011,
672 **85(16):8316-8327.**
- 673 71. Saiag P, Pavlovic M, Clerici T, Feauveau V, Nicolas JC, Emile D, Chastang C: **Treatment of early**
674 **AIDS-related Kaposi's sarcoma with oral all-trans-retinoic acid: results of a sequential non-**
675 **randomized phase II trial. Kaposi's Sarcoma ANRS Study Group. Agence Nationale de**
676 **Recherches sur le SIDA.** *Aids* 1998, **12(16):2169-2176.**
- 677 72. Tzimas G, Nau H: **The role of metabolism and toxicokinetics in retinoid teratogenesis.** *Curr*
678 *Pharm Des* 2001, **7(9):803-831.**
- 679 73. David M, Hodak E, Lowe NJ: **Adverse effects of retinoids.** *Med Toxicol Adverse Drug Exp* 1988,
680 **3(4):273-288.**
- 681 74. Govind Babu K, Lokesh KN, Suresh Babu MC, Bhat GR: **Acute Coronary Syndrome Manifesting**
682 **as an Adverse Effect of All-trans-Retinoic Acid in Acute Promyelocytic Leukemia: A Case**
683 **Report with Review of the Literature and a Spotlight on Management.** *Case Reports in*
684 *Oncological Medicine* 2016, **2016:9.**
- 685 75. Henning P, Conaway HH, Lerner UH: **Retinoid receptors in bone and their role in bone**
686 **remodeling.** *Front Endocrinol (Lausanne)* 2015, **6:31.**
- 687 76. Trasino SE, Gudas LJ: **Vitamin A: a missing link in diabetes?** *Diabetes Manag (Lond)* 2015,
688 **5(5):359-367.**
- 689 77. Rühl R, Garcia A, Schweigert FJ, Worm M: **Modulation of cytokine production by low and high**
690 **retinoid diets in ovalbumin-sensitized mice.** *Int J Vitam Nutr Res* 2004, **74(4):279-284.**
- 691 78. Wansley DL, Yin Y, Prussin C: **The retinoic acid receptor-alpha modulators ATRA and Ro415253**
692 **reciprocally regulate human IL-5+ Th2 cell proliferation and cytokine expression.** *Clin Mol*
693 *Allergy* 2013, **11(1):4.**
- 694 79. Khachik F, Spangler CJ, Smith JC, Jr., Canfield LM, Steck A, Pfander H: **Identification,**
695 **quantification, and relative concentrations of carotenoids and their metabolites in human**
696 **milk and serum.** *Anal Chem* 1997, **69(10):1873-1881.**
- 697 80. Britton G, Liaaen-Jensen S, Pfander H: **Carotenoids Handbook.** Basel: Birkhäuser; 2008.
- 698 81. Kopec RE, Caris-Veyrat C, Nowicki M, Gleize B, Desmarchelier C, Borel P: **Production of**
699 **asymmetric oxidative metabolites of ¹³C β-carotene during digestion in the gastrointestinal**
700 **lumen of healthy men.** *Am J Clin Nutr* 2018, **108:803-813.**
- 701 82. Sy C, Dangles O, Borel P, Caris-Veyrat C: **Iron-induced oxidation of (all-E)-beta-carotene under**
702 **model gastric conditions: kinetics, products, and mechanism.** *Free Radical Biol Med* 2013,
703 **63:195-206.**

- 704 83. Ferruzzi MG, Lumpkin JL, Schwartz SJ, Failla M: **Digestive stability, micellarization, and uptake**
705 **of beta-carotene isomers by Caco-2 human intestinal cells.** *J Agric Food Chem* 2006,
706 **54(7):2780-2785.**
- 707 84. Granado-Lorencio F, Olmedilla-Alonso B, Herrero-Barbudo C, Perez-Sacristan B, Blanco-Navarro
708 I, Blazquez-Garcia S: **Comparative in vitro bioaccessibility of carotenoids from relevant**
709 **contributors to carotenoid intake.** *J Agric Food Chem* 2007, **55(15):6387-6394.**
- 710 85. Richelle M, Sanchez B, Tavazzi I, Lambelet P, Bortlik K, Williamson G: **Lycopene isomerisation**
711 **takes place within enterocytes during absorption in human subjects.** *Br J Nutr* 2010,
712 **103(12):1800-1807.**
- 713 86. Eroglu A, Harrison EH: **Carotenoid metabolism in mammals, including man: formation,**
714 **occurrence, and function of apocarotenoids.** *J Lipid Res* 2013, **54(7):1719-1730.**
- 715 87. Harrison EH, dela Sena C, Eroglu A, Fleshman MK: **The formation, occurrence, and function of**
716 **beta-apocarotenoids: beta-carotene metabolites that may modulate nuclear receptor**
717 **signaling.** *Am J Clin Nutr* 2012, **96(5):1189s-1192s.**
- 718 88. Bandara S, Thomas LD, Ramkumar S, Khadka N, Kiser PD, Golczak M, von Lintig J: **The Structural**
719 **and Biochemical Basis of Apocarotenoid Processing by β -Carotene Oxygenase-2.** *ACS Chem*
720 *Biol* 2021, **16(3):480-490.**
- 721 89. Amengual J, Gouranton E, van Helden YG, Hessel S, Ribot J, Kramer E, Kiec-Wilk B, Razny U, Lietz
722 G, Wyss A *et al*: **Beta-carotene reduces body adiposity of mice via BCMO1.** *PLoS One* 2011,
723 **6(6):e20644.**
- 724 90. Kim YS, Oh DK: **Biotransformation of carotenoids to retinal by carotenoid 15,15'-oxygenase.**
725 *Appl Microbiol Biotechnol* 2010, **88(4):807-816.**
- 726 91. Harrison EH: **Carotenoids, β -Apocarotenoids, and Retinoids: The Long and the Short of It.**
727 *Nutrients* 2022, **14(7):1411.**
- 728 92. Böhm V, Lietz G, Olmedilla-Alonso B, Phelan D, Reboul E, Bánati D, Borel P, Corte-Real J, de Lera
729 AR, Desmarchelier C *et al*: **From carotenoid intake to carotenoid blood and tissue**
730 **concentrations – implications for dietary intake recommendations.** *Nutr Rev* 2021, **79(5):544-**
731 **573.**
- 732 93. Ho CC, de Moura FF, Kim S-H, Clifford AJ: **Excentral cleavage of β -carotene in vivo in a healthy**
733 **man.** *Am J Clin Nutr* 2007, **85(3):770-777.**
- 734 94. Cooperstone JL, Riedl KM, Cichon MJ, Francis DM, Curley RW, Schwartz SJ, Novotny JA, Harrison
735 EH: **Carotenoids and apo-carotenoids in human plasma after continued consumption of high**
736 **β -carotene or high lycopene tomato juice.** *The FASEB Journal* 2017, **31(1 Supplement):635.613-**
737 **635.613.**
- 738 95. Sommerburg O, Langhans CD, Arnhold J, Leichsenring M, Salerno C, Crifo C, Hoffmann GF,
739 Debatin KM, Siems WG: **Beta-carotene cleavage products after oxidation mediated by**
740 **hypochlorous acid--a model for neutrophil-derived degradation.** *Free Radic Biol Med* 2003,
741 **35(11):1480-1490.**
- 742 96. von Lintig J, Wyss A: **Molecular analysis of vitamin A formation: cloning and characterization**
743 **of beta-carotene 15,15'-dioxygenases.** *Arch Biochem Biophys* 2001, **385(1):47-52.**
- 744 97. Böhm V, Borel P, Corte-Real J, de Lera A, Desmarchelier C, Dulinska-Litewka J, Landrier J-F, Lietz
745 G, Milisav I, Nolan J *et al*: **From carotenoid intake to carotenoid blood and tissue**
746 **concentrations – implications for dietary intake recommendations.** *Nutr Rev* 2021, **79:544-**
747 **573.**

- 748 98. Horst RL, Reinhardt TA, Goff JP, Nonnecke BJ, Gambhir VK, Fiorella PD, Napoli JL: **Identification**
749 **of 9-cis,13-cis-retinoic acid as a major circulating retinoid in plasma.** *Biochemistry* 1995,
750 **34(4):1203-1209.**
- 751 99. Kane MA, Chen N, Sparks S, Napoli JL: **Quantification of endogenous retinoic acid in limited**
752 **biological samples by LC/MS/MS.** *Biochem J* 2005, **388**(Pt 1):363-369.
- 753 100. Sass JO, Nau H: **Single-run analysis of isomers of retinoyl-beta-D-glucuronide and retinoic acid**
754 **by reversed-phase high-performance liquid chromatography.** *J Chromatogr A* 1994,
755 **685(1):182-188.**
- 756 101. Wolf G: **Is 9-cis-retinoic acid the endogenous ligand for the retinoic acid-X receptor?** *Nutr Rev*
757 2006, **64(12):532-538.**
- 758 102. Rühl R, Krzyzosiak A, Niewiadomska-Cimicka A, Rochel N, Szeles L, Vaz B, Wietrzyk-Schindler
759 M, Alvarez S, Szklenar M, Nagy L *et al*: **9-cis-13,14-dihydroretinoic acid is an endogenous**
760 **retinoid acting as RXR ligand in mice.** *PLoS Genet* 2015, **11(6):e1005213.**
- 761 103. Palczewski K: **Chemistry and biology of vision.** *J Biol Chem* 2012, **287(3):1612-1619.**
- 762 104. von Lintig J, Moon J, Babino D: **Molecular components affecting ocular carotenoid and retinoid**
763 **homeostasis.** *Prog Retin Eye Res* 2021, **80:100864.**
- 764 105. Watzl B, Bub A, Brandstetter BR, Rechkemmer G: **Modulation of human T-lymphocyte**
765 **functions by the consumption of carotenoid-rich vegetables.** *Br J Nutr* 1999, **82(5):383-389.**
- 766 106. Watzl B, Bub A, Briviba K, Rechkemmer G: **Supplementation of a low-carotenoid diet with**
767 **tomato or carrot juice modulates immune functions in healthy men.** *Ann Nutr Metab* 2003,
768 **47(6):255-261.**
- 769 107. Underwood BA: **Was the "anti-infective" vitamin misnamed?** *Nutr Rev* 1994, **52(4):140-143.**
- 770 108. Lee LM, Leung CY, Tang WW, Choi HL, Leung YC, McCaffery PJ, Wang CC, Woolf AS, Shum AS: **A**
771 **paradoxical teratogenic mechanism for retinoic acid.** *Proc Natl Acad Sci U S A* 2012,
772 **109(34):13668-13673.**
- 773 109. Wang XD, Liu C, Bronson RT, Smith DE, Krinsky NI, Russell M: **Retinoid signaling and activator**
774 **protein-1 expression in ferrets given beta-carotene supplements and exposed to tobacco**
775 **smoke.** *J Natl Cancer Inst* 1999, **91(1):60-66.**
- 776 110. Lotan R: **Lung cancer promotion by beta-carotene and tobacco smoke: relationship to**
777 **suppression of retinoic acid receptor-beta and increased activator protein-1?** *J Natl Cancer*
778 *Inst* 1999, **91(1):7-9.**
- 779 111. Moise AR, Kuksa V, Imanishi Y, Palczewski K: **Identification of all-trans-retinol:all-trans-13,14-**
780 **dihydroretinol saturase.** *J Biol Chem* 2004, **279(48):50230-50242.**
- 781 112. Moise AR, Alvarez S, Dominguez M, Alvarez R, Golczak M, Lobo GP, von Lintig J, de Lera AR,
782 Palczewski K: **Activation of retinoic acid receptors by dihydroretinoids.** *Mol Pharmacol* 2009,
783 **76(6):1228-1237.**
- 784 113. Bazhin AV, Bleul T, de Lera AR, Werner J, Ruhl R: **Relationship between all-trans-13,14-dihydro**
785 **retinoic acid and pancreatic adenocarcinoma.** *Pancreas* 2016, **45(6):e29-31.**
- 786 114. Lucas R, Szklenar M, Mihály J, Szegedi A, Töröcsik D, Rühl R: **Plasma Levels of Bioactive Vitamin**
787 **D and A5 Ligands Positively Correlate with Clinical Atopic Dermatitis Markers.** *Dermatology*
788 2022:1-8.
- 789 115. Sani BP, Venepally PR, Levin AA: **Didehydroretinoic acid: retinoid receptor-mediated**
790 **transcriptional activation and binding properties.** *Biochem Pharmacol* 1997, **53(7):1049-1053.**

- 791 116. Rühl R, Krezel W, de Lera AR: **9-Cis-13,14-dihydroretinoic acid, a new endogenous mammalian**
792 **ligand of retinoid X receptor and the active ligand of a potential new vitamin A category:**
793 **Vitamin A5.** *Nutr Rev* 2018, **76**(12):929-941.
- 794 117. Kręzel W, Rivas A, Szklenar M, Ciancia M, Alvarez R, de Lera AR, Rühl R: **Vitamin A5/X, a New**
795 **Food to Lipid Hormone Concept for a Nutritional Ligand to Control RXR-Mediated Signaling.**
796 *Nutrients* 2021, **13**(3):925.
- 797 118. Bohn T, Hellmann-Regen J, de Lera Á R, Böhm V, Rühl R: **Human nutritional relevance and**
798 **suggested nutritional guidelines for Vitamin A5/X and Provitamin A5/X.** *submitted to*
799 *Nutrition Reviews* 2022.
- 800 119. Khachik F, Carvalho L, Bernstein PS, Muir GJ, Zhao DY, Katz NB: **Chemistry, distribution, and**
801 **metabolism of tomato carotenoids and their impact on human health.** *Exp Biol Med*
802 *(Maywood)* 2002, **227**(10):845-851.
- 803 120. van den Berg H: **Effect of lutein on beta-carotene absorption and cleavage.** *Int J Vitam Nutr*
804 *Res* 1998, **68**(6):360-365.
- 805 121. van den Berg H, van Vliet T: **Effect of simultaneous, single oral doses of beta-carotene with**
806 **lutein or lycopene on the beta-carotene and retinyl ester responses in the triacylglycerol-rich**
807 **lipoprotein fraction of men.** *Am J Clin Nutr* 1998, **68**(1):82-89.
- 808 122. Caris-Veyrat C, Garcia AL, Reynaud E, Lucas R, Aydemir G, Rühl R: **Lycopene-induced nuclear**
809 **hormone receptor signalling in inflammation and lipid metabolism via still unknown**
810 **endogenous apo-10'-lycopenoids.** *Int J Vitam Nutr Res* 2016, **86**(1-2):62-70.
- 811 123. Ford NA, Erdman JW, Jr.: **Are lycopene metabolites metabolically active?** *Acta Biochim Pol*
812 2012, **59**(1):1-4.
- 813 124. Aydemir G, Kasiri Y, Birta E, Beke G, Garcia AL, Bartok EM, Rühl R: **Lycopene-derived bioactive**
814 **retinoic acid receptors/retinoid-X receptors-activating metabolites may be relevant for**
815 **lycopene's anti-cancer potential.** *Mol Nutr Food Res* 2013, **57**(5):739-747.
- 816 125. Kopec RE, Riedl KM, Harrison EH, Curley RW, Jr., Hruszkewycz DP, Clinton SK, Schwartz SJ:
817 **Identification and quantification of apo-lycopenals in fruits, vegetables, and human plasma.** *J*
818 *Agric Food Chem* 2010, **58**(6):3290-3296.
- 819 126. Ferreira AL, Yeum KJ, Russell RM, Krinsky NI, Tang G: **Enzymatic and oxidative metabolites of**
820 **lycopene.** *J Nutr Biochem* 2003, **14**(9):531-540.
- 821 127. Reynaud E, Aydemir G, Rühl R, Dangles O, Caris-Veyrat C: **Organic synthesis of new putative**
822 **lycopene metabolites and preliminary investigation of their cell-signaling effects.** *J Agric Food*
823 *Chem* 2011, **59**(4):1457-1463.
- 824 128. Lindshield BL, Canene-Adams K, Erdman JW, Jr.: **Lycopenoids: are lycopene metabolites**
825 **bioactive?** *Arch Biochem Biophys* 2007, **458**(2):136-140.
- 826 129. Wang XD: **Lycopene metabolism and its biological significance.** *Am J Clin Nutr* 2012,
827 **96**(5):1214s-1222s.
- 828 130. Gouranton E, Aydemir G, Reynaud E, Marcotorchino J, Malezet C, Caris-Veyrat C, Blomhoff R,
829 Landrier JF, Rühl R: **Apo-10'-lycopenoic acid impacts adipose tissue biology via the retinoic**
830 **acid receptors.** *Biochim Biophys Acta* 2011, **1811**(12):1105-1114.
- 831 131. Hu KQ, Liu C, Ernst H, Krinsky NI, Russell RM, Wang XD: **The biochemical characterization of**
832 **ferret carotene-9',10'-monooxygenase catalyzing cleavage of carotenoids in vitro and in vivo.**
833 *J Biol Chem* 2006, **281**(28):19327-19338.
- 834 132. Gajic M, Zaripheh S, Sun F, Erdman JW, Jr.: **Apo-8'-lycopenal and apo-12'-lycopenal are**
835 **metabolic products of lycopene in rat liver.** *J Nutr* 2006, **136**(6):1552-1557.

- 836 133. Moran NE, Thomas-Ahner JM, Fleming JL, McElroy JP, Mehl R, Grainger EM, Riedl KM, Toland
837 AE, Schwartz SJ, Clinton SK: **Single nucleotide polymorphisms in beta-carotene oxygenase 1**
838 **are associated with plasma lycopene responses to a tomato-soy juice intervention in men**
839 **with prostate cancer.** *J Nutr* 2019, **149**(3):381-397.
- 840 134. Aydemir G, Carlsen H, Blomhoff R, Rühl R: **Lycopene induces retinoic acid receptor**
841 **transcriptional activation in mice.** *Mol Nutr Food Res* 2012, **56**(5):702-712.
- 842 135. Ben-Dor A, Nahum A, Danilenko M, Giat Y, Stahl W, Martin HD, Emmerich T, Noy N, Levy J,
843 Sharoni Y: **Effects of acyclo-retinoic acid and lycopene on activation of the retinoic acid**
844 **receptor and proliferation of mammary cancer cells.** *Arch Biochem Biophys* 2001, **391**(2):295-
845 302.
- 846 136. Ip BC, Liu C, Lichtenstein AH, von Lintig J, Wang XD: **Lycopene and apo-10'-lycopenoic acid**
847 **have differential mechanisms of protection against hepatic steatosis in beta-carotene-9',10'-**
848 **oxygenase knockout male mice.** *J Nutr* 2015, **145**(2):268-276.
- 849 137. Rühl R: **[Retinoids, vitamin A and pro-vitamin A carotenoids. Regulation of the immune**
850 **system and allergies].** *Pharm Unserer Zeit* 2009, **38**(2):126-131.
- 851 138. Muller H, Bub A, Watzl B, Rechkemmer G: **Plasma concentrations of carotenoids in healthy**
852 **volunteers after intervention with carotenoid-rich foods.** *Eur J Nutr* 1999, **38**(1):35-44.
- 853 139. Rühl R, Hanel A, Garcia AL, Dahten A, Herz U, Schweigert FJ, Worm M: **Role of vitamin A**
854 **elimination or supplementation diets during postnatal development on the allergic**
855 **sensitisation in mice.** *Mol Nutr Food Res* 2007, **51**(9):1173-1181.
- 856 140. Zunino SJ, Storms DH, Stephensen CB: **Diets rich in polyphenols and vitamin A inhibit the**
857 **development of type I autoimmune diabetes in nonobese diabetic mice.** *J Nutr* 2007,
858 **137**(5):1216-1221.
- 859 141. Landrier JF, Kasiri E, Karkeni E, Mihaly J, Beke G, Weiss K, Lucas R, Aydemir G, Salles J, Walrand S
860 *et al*: **Reduced adiponectin expression after high-fat diet is associated with selective up-**
861 **regulation of ALDH1A1 and further retinoic acid receptor signaling in adipose tissue.** *Faseb J*
862 2017, **31**:203-211.
- 863 142. Rühl R, Landrier JF: **Dietary regulation of adiponectin by direct and indirect lipid activators of**
864 **nuclear hormone receptors.** *Mol Nutr Food Res* 2016, **60**(1):175-184.
- 865 143. Al-Babili S, Bouwmeester HJ: **Strigolactones, a novel carotenoid-derived plant hormone.** *Annu*
866 *Rev Plant Biol* 2015, **66**:161-186.
- 867 144. Jia K-P, Baz L, Al-Babili S: **From carotenoids to strigolactones.** *JExB* 2018, **69**(9):2189-2204.
- 868 145. Alder A, Jamil M, Marzorati M, Bruno M, Vermathen M, Bigler P, Ghisla S, Bouwmeester H,
869 Beyer P, Al-Babili S: **The path from β -carotene to carlactone, a strigolactone-like plant**
870 **hormone.** *Science* 2012, **335**(6074):1348-1351.
- 871 146. Gomez-Roldan V, Fermas S, Brewer PB, Puech-Pagès V, Dun EA, Pillot J-P, Letisse F, Matusova R,
872 Danoun S, Portais J-C *et al*: **Strigolactone inhibition of shoot branching.** *Nature* 2008,
873 **455**(7210):189-194.
- 874 147. Casamassimi A, Federico A, Rienzo M, Esposito S, Ciccodicola A: **Transcriptome profiling in**
875 **human diseases: new advances and perspectives.** *Int J Mol Sci* 2017, **18**(8).
- 876 148. Pedrotty DM, Morley MP, Cappola TP: **Transcriptomic biomarkers of cardiovascular disease.**
877 *Prog Cardiovasc Dis* 2012, **55**(1):64-69.
- 878 149. Altucci L, Leibowitz MD, Ogilvie KM, de Lera AR, Gronemeyer H: **RAR and RXR modulation in**
879 **cancer and metabolic disease.** *Nat Rev Drug Discov* 2007, **6**(10):793-810.

- 880 150. Jenab M, Ferrari P, Mazuir M, Tjonneland A, Clavel-Chapelon F, Linseisen J, Trichopoulou A,
881 Tumino R, Bueno-de-Mesquita HB, Lund E *et al*: **Variations in lycopene blood levels and**
882 **tomato consumption across European countries based on the European Prospective**
883 **Investigation into Cancer and Nutrition (EPIC) study.** *J Nutr* 2005, **135**(8):2032s-2036s.
- 884 151. Xu Y, Harvey PJ: **Red Light Control of β -Carotene Isomerisation to 9-cis β -Carotene and**
885 **Carotenoid Accumulation in *Dunaliella salina*.** *Antioxidants (Basel, Switzerland)* 2019, **8**(5):148.
- 886 152. Schieber A, Carle R: **Occurrence of carotenoid cis-isomers in food: Technological, analytical,**
887 **and nutritional implications.** *Trends Food Sci Technol* 2005, **16**(9):416-422.
- 888 153. Khoo HE, Prasad KN, Kong KW, Jiang Y, Ismail A: **Carotenoids and their isomers: color pigments**
889 **in fruits and vegetables.** *Molecules* 2011, **16**(2):1710-1738.
- 890 154. Cooperstone JL, Novotny JA, Riedl KM, Cichon MJ, Francis DM, Curley RW, Jr., Schwartz SJ,
891 Harrison EH: **Limited appearance of apocarotenoids is observed in plasma after consumption**
892 **of tomato juices: a randomized human clinical trial.** *Am J Clin Nutr* 2018, **108**(4):784-792.
- 893 155. Rosati A: **The dietary practice coincided with increases in brain size, evidence suggests.** *SciAm*
894 2018, [https://www.scientificamerican.com/article/food-for-thought-was-cooking-a-pivotal-](https://www.scientificamerican.com/article/food-for-thought-was-cooking-a-pivotal-step-in-human-evolution/assessed)
895 [step-in-human-evolution/assessed](https://www.scientificamerican.com/article/food-for-thought-was-cooking-a-pivotal-step-in-human-evolution/assessed) July 15, 2022).
- 896 156. Lee CM, Boileau AC, Boileau TW, Williams AW, Swanson KS, Heintz KA, Erdman JW, Jr.: **Review**
897 **of animal models in carotenoid research.** *J Nutr* 1999, **129**(12):2271-2277.
- 898 157. Olivier M, Asmis R, Hawkins GA, Howard TD, Cox LA: **The Need for Multi-Omics Biomarker**
899 **Signatures in Precision Medicine.** *Int J Mol Sci* 2019, **20**(19).
- 900 158. Szanto A, Narkar V, Shen Q, Uray IP, Davies PJ, Nagy L: **Retinoid X receptors: X-ploring their**
901 **(patho)physiological functions.** *Cell Death Differ* 2004, **11 Suppl 2**:S126-143.
- 902 159. Al-Delaimy WK, Slimani N, Ferrari P, Key T, Spencer E, Johansson I, Johansson G, Mattisson I,
903 Wirfalt E, Sieri S *et al*: **Plasma carotenoids as biomarkers of intake of fruits and vegetables:**
904 **ecological-level correlations in the European Prospective Investigation into Cancer and**
905 **Nutrition (EPIC).** *Eur J Clin Nutr* 2005, **59**(12):1397-1408.
- 906 160. Schierle J, Bretzel W, Bühler I, Faccin N, Hess D, Steiner K, Schüep W: **Content and isomeric**
907 **ratio of lycopene in food and human blood plasma.** *Food Chemistry* 1997, **59**(3):459-465.
- 908 161. Fröhlich K: **Lycopin-Isomere in Lebensmitteln und Humanplasma - Strukturaufklärung,**
909 **antioxidative Aktivität, Gehalte und relative (E)-(Z)-Verhältnisse.** Friedrich-Schiller-Universität
910 Jena, Germany; 2007.
- 911 162. Chung HY, Ferreira AL, Epstein S, Paiva SA, Castaneda-Sceppa C, Johnson EJ: **Site-specific**
912 **concentrations of carotenoids in adipose tissue: relations with dietary and serum carotenoid**
913 **concentrations in healthy adults.** *Am J Clin Nutr* 2009, **90**(3):533-539.
- 914 163. Alaluf S, Heinrich U, Stahl W, Tronnier H, Wiseman S: **Dietary carotenoids contribute to normal**
915 **human skin color and UV photosensitivity.** *J Nutr* 2002, **132**(3):399-403.
- 916 164. Ermakov IV, Sharifzadeh M, Ermakova M, Gellermann W: **Resonance Raman detection of**
917 **carotenoid antioxidants in living human tissue.** *J Biomed Opt* 2005, **10**(6):064028.
- 918 165. Ermakov IV, Ermakova MR, Bernstein PS, Chan GM, Gellermann W: **Resonance Raman based**
919 **skin carotenoid measurements in newborns and infants.** *Journal of biophotonics* 2013,
920 **6**(10):793-802.
- 921 166. Schmitz HH, Poor CL, Wellman RB, Erdman JW, Jr.: **Concentrations of selected carotenoids and**
922 **vitamin A in human liver, kidney and lung tissue.** *J Nutr* 1991, **121**(10):1613-1621.
- 923 167. Vishwanathan R, Kuchan MJ, Sen S, Johnson EJ: **Lutein and preterm infants with decreased**
924 **concentrations of brain carotenoids.** *J Pediatr Gastroenterol Nutr* 2014, **59**(5):659-665.

- 925 168. Stahl W, Schwarz W, Sundquist AR, Sies H: **cis-trans isomers of lycopene and beta-carotene in**
926 **human serum and tissues.** *Arch Biochem Biophys* 1992, **294**(1):173-177.
- 927 169. Pappalardo G, Maiani G, Mobarhan S, Guadalaxara A, Azzini E, Raguzzini A, Salucci M, Serafini
928 M, Trifero M, Illomei G *et al*: **Plasma (carotenoids, retinol, alpha-tocopherol) and tissue**
929 **(carotenoids) levels after supplementation with beta-carotene in subjects with precancerous**
930 **and cancerous lesions of sigmoid colon.** *Eur J Clin Nutr* 1997, **51**(10):661-666.
- 931 170. Gossage CP, Deyhim M, Yamini S, Douglass LW, Moser-Veillon PB: **Carotenoid composition of**
932 **human milk during the first month postpartum and the response to β -carotene**
933 **supplementation.** *Am J Clin Nutr* 2002, **76**(1):193-197.
- 934 171. Czczuga-Semeniuk E, Wolczynski S: **Dietary carotenoids in normal and pathological tissues of**
935 **corpus uteri.** *Folia Histochem Cytobiol* 2008, **46**(3):283-290.
- 936 172. Clinton SK, Emehiser C, Schwartz SJ, Bostwick DG, Williams AW, Moore BJ, Erdman JW, Jr.: **cis-**
937 **trans lycopene isomers, carotenoids, and retinol in the human prostate.** *Cancer Epidemiol*
938 *Biomarkers Prev* 1996, **5**(10):823-833.
- 939 173. Mihaly J, Gamlieli A, Worm M, Ruhl R: **Decreased retinoid concentration and retinoid**
940 **signalling pathways in human atopic dermatitis.** *Exp Dermatol* 2011, **20**(4):326-330.
- 941 174. Arnhold T, Tzimas G, Wittfoht W, Plonait S, Nau H: **Identification of 9-cis-retinoic acid, 9,13-di-**
942 **cis-retinoic acid, and 14-hydroxy-4,14-retro-retinol in human plasma after liver consumption.**
943 *Life Sci* 1996, **59**(12):PI169-177.
- 944 175. Arnold SL, Amory JK, Walsh TJ, Isoherranen N: **A sensitive and specific method for**
945 **measurement of multiple retinoids in human serum with UHPLC-MS/MS.** *J Lipid Res* 2012,
946 **53**(3):587-598.
- 947 176. Bleul T, Ruhl R, Bulashevskaya S, Karakhanova S, Werner J, Bazhin AV: **Reduced retinoids and**
948 **retinoid receptors' expression in pancreatic cancer: A link to patient survival.** *Mol Carcinog*
949 2015, **54**(9):870-879.
- 950 177. Aydemir G, Domínguez M, de Lera AR, Mihaly J, Törőcsik D, Rühl R: **Apo-14'-Carotenoic Acid Is**
951 **a Novel Endogenous and Bioactive Apo-Carotenoid.** *Nutrients* 2019, **11**(9):2084.
- 952 178. Barua AB, Sidell N: **Retinoyl beta-glucuronide: a biologically active interesting retinoid.** *J Nutr*
953 2004, **134**(1):286s-289s.
- 954 179. Vahlquist A, Lee JB, Michaelsson G, Rollman O: **Vitamin A in human skin: II Concentrations of**
955 **carotene, retinol and dehydroretinol in various components of normal skin.** *J Invest Dermatol*
956 1982, **79**(2):94-97.
- 957 180. Wang J, Yoo HS, Obrochta KM, Huang P, Napoli JL: **Quantitation of retinaldehyde in small**
958 **biological samples using ultrahigh-performance liquid chromatography tandem mass**
959 **spectrometry.** *Anal Biochem* 2015, **484**:162-168.
- 960 181. Kane MA, Folias AE, Napoli JL: **HPLC/UV quantitation of retinal, retinol, and retinyl esters in**
961 **serum and tissues.** *Anal Biochem* 2008, **378**(1):71-79.
- 962 182. Stahl W, Hanusch M, Sies H: **4-oxo-retinoic acid is generated from its precursor canthaxanthin**
963 **and enhances gap junctional communication in 10T1/2 cells.** *Adv Exp Med Biol* 1996, **387**:121-
964 128.
- 965