

## Photoactivated Ferric Chloride Oxidation of Carotenoids by Near-UV to Visible Light

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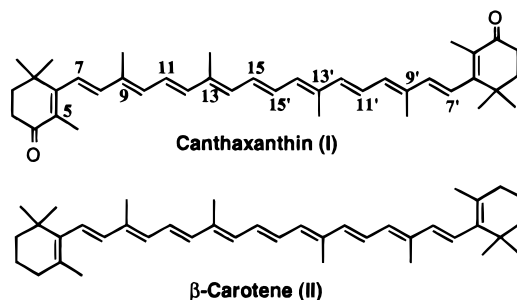
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When canthaxanthin (**I**) and  $\beta$ -carotene (**II**) dichloromethane solutions are treated with small amounts of ferric chloride ( $\leq 0.26$  mol equiv), extensive photodegradation of the remaining unreacted neutral carotenoid occurs upon subsequent irradiation with near-UV to visible light. The rate of this photodegradation is independent of the neutral carotenoid concentration at a given initial  $\text{FeCl}_3$  concentration and first-order in initial  $\text{FeCl}_3$  ( $k_1 = 1.43$  and  $3.30 \text{ min}^{-1}$  for **I** and **II**, respectively). The data are consistent with a mechanism in which  $\text{Fe(II)}$  is photochemically converted in the presence of  $\text{CH}_2\text{Cl}_2$  to  $\text{Fe(III)}$ , which then oxidizes unreacted neutral carotenoids. We also report that canthaxanthin radical cations in dichloromethane form cation dehydromers at low temperature or upon irradiation with near-UV to visible light, which have an absorption maximum near 770 nm and a reduction peak at 20 mV in cyclic voltammetry. It is proposed that radical cations associate with parent molecules and then undergo two subsequent steps to form the final cation dehydromers. In the presence of supporting electrolytes, canthaxanthin radical cations form ion pairs with the anions  $\text{PF}_6^-$ , resulting in a 10 nm blue shift of the absorption maximum of the radical cations.

## Introduction

Carotenoids are widely distributed among plants, animals, and certain bacteria. Around 600 naturally occurring carotenoids have been isolated and identified.<sup>1</sup> It is well-known that certain carotenoids have important biochemical and biological functions, such as light harvesting and photoprotection in photosynthesis<sup>2,3</sup> and providing the essential provitamin A compounds.<sup>4</sup> Carotenoids are also good antioxidants<sup>5</sup> and have been implicated in cancer prevention and treatment,<sup>6</sup> the efficacy of which may be due to their antioxidant and free radical quenching activity,<sup>7</sup> immune enhancement effect,<sup>8</sup> and their *in vivo* function as antioxidants.<sup>9</sup> The structures of two typical carotenoids, all-trans canthaxanthin (**I**) and  $\beta$ -carotene (**II**), are shown below.



To understand the mechanism of the photoprotective action in photosynthesis and utilize the photoprotective properties of carotenoids in artificial solar devices, it is essential to know the decomposition processes of carotenoids. For example, it has been shown that carotenoids within the photosystem II (P680) complex can undergo light-induced oxidation reactions<sup>10,11</sup> that lead to irreversible and rapid degradation of the pigment in the absence of oxygen,<sup>12,13</sup> and it was found that the photooxidation of the carotenoid by  $\text{P680}^+$  can protect P680 from photodamage in isolated photosystem II reaction centers. It was proposed<sup>14–16</sup> that in the presence of oxygen carotenoids

protect photoactivated chlorophyll from destruction by quenching triplet chlorophyll and/or harmful singlet oxygen. Carotenoids can also deactivate other reactive chemical species, such as peroxy and alkoxy radicals, which can be generated within cells and might otherwise initiate harmful oxidative reactions;<sup>17,18</sup> these radical-initiated oxidations of carotenoids are followed by irreversible decomposition of the pigments.

In this study optical and spectroelectrochemical techniques were employed to investigate the fate and associative properties of the carotenoid radical cations generated by electrolysis and chemical methods. Photochemical experiments were also carried out to explore the properties of ferric chloride-doped carotenoid solutions. The impetus for these experiments stemmed from the following: (1) iron and carotenoid have been found to occur in the reaction centers (RC) of photosystems, e.g., bacterial RCs consist of a non-heme iron center and a carotenoid;<sup>19</sup> (2) carotenoid radical cations have been observed optically to form at the photosystem II reaction center;<sup>16</sup> (3) approximately molar equivalents of carotenoid radical cations are formed by oxidation with ferric chloride in solution;<sup>20</sup> (4) our recent observation that exposure to near-UV to visible light of carotenoid solutions pretreated with only small amounts of  $\text{FeCl}_3$  ( $\leq 0.26$  mol equiv) results in significant degradation of the remaining neutral carotenoid. To better understand the carotenoid/ferric chloride/ $h\nu$  system, further investigation of this *in vitro* iron-catalyzed photodegradation were deemed to be particularly important.

## Experimental Section

All-trans canthaxanthin and  $\beta$ -carotene and tetrabutylammonium hexafluorophosphate (TBAHFP) were purchased from Fluka. Anhydrous dichloromethane, ferric chloride, and ferrous chloride were obtained from Aldrich.

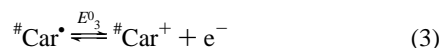
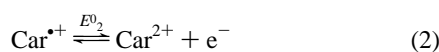
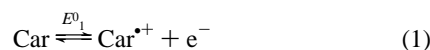
Optical absorption spectra in the range 190–1100 nm were measured using a Shimadzu UV-1601 UV–vis spectrophotometer. The quartz cuvettes (1 cm optical path length) used for irradiation and spectral measurements containing 3.0 mL solutions were closed with stoppers wrapped with Parafilm. The quartz cell used for simultaneous spectroelectrochemical measurements was described in refs 21 and 22. The solutions were

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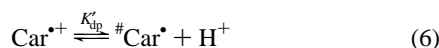
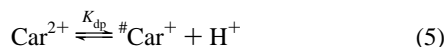
<sup>®</sup> Abstract published in *Advance ACS Abstracts*, September 1, 1997.

## SCHEME 1

electrode reactions



homogeneous reactions



#Car represents the carotenoid with one less proton

stirred during bulk electrolysis but not during the spectral measurements. For low-temperature spectroelectrochemical measurements, methanol at  $-10^\circ\text{C}$  was circulated through the cuvette holder and dry nitrogen was continuously passed through the cuvette chamber to prevent condensation of water.

Cyclic voltammetry (CV) and bulk electrolysis were carried out using the Bio Analytical Systems BAS-100W electrochemical analyzer. For CV measurements, a glassy carbon disk electrode (diameter = 3.0 mm) was used as the working electrode, the auxiliary electrode was a platinum wire, and the reference electrode was a Ag/AgCl electrode with 3 M NaCl. For bulk electrolysis a platinum gauze was used as the working electrode, and a silver wire was used as pseudoreference electrode. Except as noted, measurements were carried out at  $23^\circ\text{C}$ .

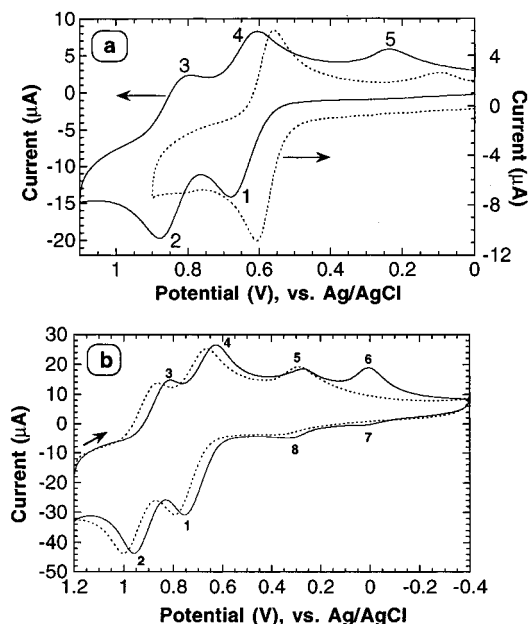
A 250 W xenon lamp (ILC Technology) at a distance of 20 cm was used to irradiate the solutions. Deionized water contained in a cylindrical glass cell (path length, 10 cm) was used as an IR cutoff filter. A filter (350 nm) was used to cut off most of the UV light. The photoenergy at the external surface of the cuvette, measured with a power meter (Newport, Model 818-SL), was  $0.4 \text{ mW cm}^{-2}$ .

Saturated stock solutions of ferric chloride in dichloromethane were prepared at room temperature. For CV measurements, dichloromethane solutions of canthaxanthin or  $\beta$ -carotene (both 1 mM) and of TBAHFP (0.1 M) were used. All solutions were prepared in a drybox under a nitrogen atmosphere.

## Results and Discussion

**Canthaxanthin. Cyclic Voltammetry.** Previous investigations<sup>21–25</sup> have established that the electrode and homogeneous reactions shown in Scheme 1 take place during the electrochemical oxidation of most carotenoids at ambient temperatures. The redox reactions (eqs 1–3) can be detected by cyclic voltammetry (CV), as illustrated in Figure 1a for canthaxanthin and  $\beta$ -carotene in dichloromethane ( $\text{CH}_2\text{Cl}_2$ ). For canthaxanthin, peak 1 corresponds to the one-electron oxidation of the neutral species (eq 1) and peak 2 results from the subsequent one-electron oxidation of the radical cation (eq 2). Peaks 3 and 4 are due to sequential one-electron reductions of the dication and the radical cation, respectively. Peak 5 results from the reduction (reverse of eq 3) of the transient intermediate,  $\# \text{Car}^+$ , which is formed by loss of a proton from the dication (eq 5). The anodic peaks 1 and 2 and the cathodic peaks 3 and 4 of  $\beta$ -carotene coincide, since the redox potentials of reactions 1 and 2 are nearly the same.

A CV of canthaxanthin obtained at lower temperature ( $-10^\circ\text{C}$ ) showed an additional pair of peaks (Figure 1b, peaks 6 and



**Figure 1.** (a) Cyclic voltammograms of 1 mM all-trans canthaxanthin (—) and  $\beta$ -carotene (···) in  $\text{CH}_2\text{Cl}_2$  containing 0.1 M TBAHFP at scan rate of  $0.1 \text{ V s}^{-1}$ , starting at 0 V. (b) Cyclic voltammograms of 1 mM all-trans canthaxanthin in  $\text{CH}_2\text{Cl}_2$  containing 0.1 M TBAHFP at scan rate of  $0.5 \text{ V s}^{-1}$ , starting at 1.2 V: (—)  $-10^\circ\text{C}$ ; (···)  $23^\circ\text{C}$ .

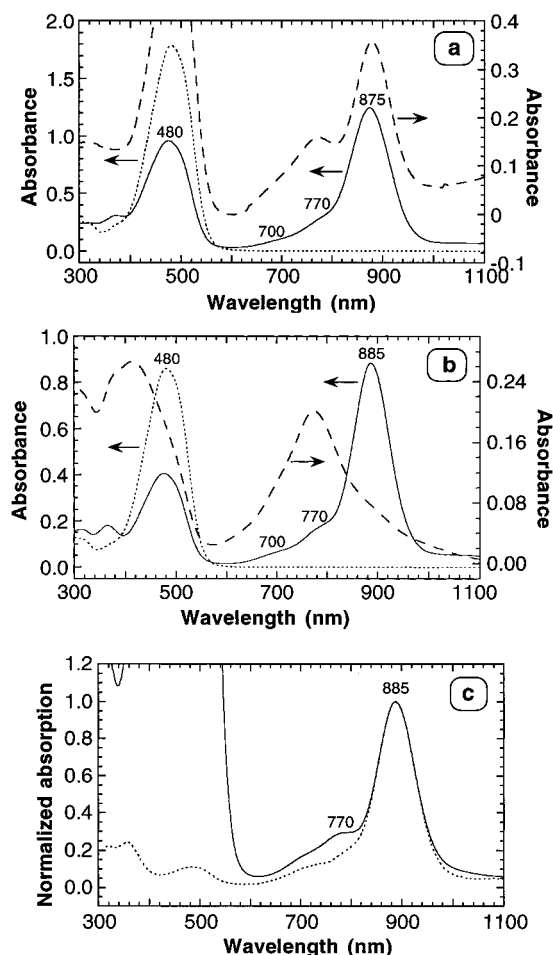
7). The probable origin of this pair of peaks will be discussed in the next section.

**Optical Absorption Spectroscopy.** Carotenoid radical cations formed by bulk electrolysis in  $\text{CH}_2\text{Cl}_2$  can be observed by simultaneous optical absorption spectroscopy.<sup>26</sup> The optical absorption spectrum of neutral all-trans canthaxanthin is shown in Figure 2 (dotted lines). The solid line in Figure 2a is the spectrum obtained after anodic bulk electrolysis for 10 min at  $23^\circ\text{C}$ . The additional absorption maximum at 875 nm is due to radical cations that are generated during the bulk electrolysis (BE).<sup>20</sup> As determined from the rate of decrease of this absorption, the half-life of the radical cation of canthaxanthin in  $\text{CH}_2\text{Cl}_2$  is 570 s; i.e., this species is relatively stable in this solvent.

Chemical oxidation of carotenoids of, for example, canthaxanthin by ferric chloride, can also generate radical cations (Figure 2b, solid line, 885 nm). There is a 10 nm difference between the absorption maxima of the radical cations generated by BE and  $\text{FeCl}_3$  oxidation. Formation of ion pairs between the radical cations and the supporting electrolyte anions  $\text{PF}_6^-$  can account for the blue shift of the absorption of radical cations (solid line in Figure 2a). Such a hypsochromic shift has been previously observed and attributed to contact ion pairs of radical cations of a similarly conjugated diphenylpolyene and  $\text{PF}_6^-$  in 1,2-dichloroethane.<sup>27</sup>

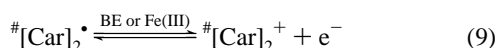
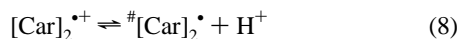
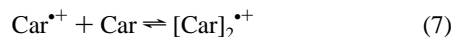
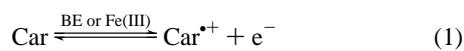
In addition to the 885 absorption due to the radical cation, the spectra (Figure 2, solid lines) display shoulders near 770 and 700 nm. The absorption near 700 nm is attributable to the canthaxanthin dication that is formed, nearly exclusively, in the presence of excess ( $>2$  molar equiv)  $\text{FeCl}_3$  according to earlier studies.<sup>20</sup> A possible source of the 770 nm absorption is described below.

Bulk electrolysis of canthaxanthin at lower temperature ( $-10^\circ\text{C}$ ) resulted in enhanced absorption at 770 nm (Figure 2a, dashed line). Further, when a  $\text{CH}_2\text{Cl}_2$  solution of canthaxanthin and 0.5 mol equiv of  $\text{FeCl}_3$  (Figure 2b, solid line) was briefly (1.5 min) irradiated with UV-vis light, only the 770 nm maximum was observed; little, if any, absorption of the radical cations (885 nm) was present (Figure 2b, dashed line). These observations indicate that the new species is derived from the



**Figure 2.** (a) Optical absorption spectra of 15  $\mu\text{M}$  canthaxanthin in  $\text{CH}_2\text{Cl}_2$  containing 0.05 M supporting electrolyte TBAHFP: (···) neutral canthaxanthin; (—) after bulk electrolysis at 1.0 V, 23  $^\circ\text{C}$ , for 10 min; (---) after bulk electrolysis of 7  $\mu\text{M}$  canthaxanthin at  $-10^\circ\text{C}$  for 5 min. (b) Optical absorption spectra of 7  $\mu\text{M}$  canthaxanthin in  $\text{CH}_2\text{Cl}_2$ : in the presence of (···) 0  $\mu\text{M}$  and (—) 3.5  $\mu\text{M}$   $\text{FeCl}_3$ ; (---) after irradiating the 3.5  $\mu\text{M}$   $\text{FeCl}_3$ -doped solution for 1.5 min. (c) Optical absorption spectra of canthaxanthin in  $\text{CH}_2\text{Cl}_2$  in the presence of 2.5  $\mu\text{M}$   $\text{FeCl}_3$ : (···) 3  $\mu\text{M}$  and (—) 50  $\mu\text{M}$  canthaxanthin. The spectra are normalized at 885 nm.

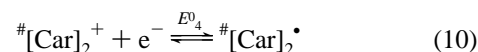
## SCHEME 2



radical cations. The neutral carotenoid must also be involved in the formation of this species, as shown by the following. Even at room temperature, in the absence of irradiation, the relative intensity of the 770 nm compared to the 885 nm absorption is enhanced when the ratio  $[\text{Car}]/[\text{FeCl}_3]$  is large. As shown in Figure 2c, in the presence of only a small excess of neutral canthaxanthin ( $[\text{Car}]/[\text{FeCl}_3] = 1.2$ ) the neutral carotenoid is nearly exclusively converted to the radical cation (eq 1). When a 20-fold excess of carotenoid is present (Figure 2c, solid line), in addition to the absorption due to  $\text{Car}^{\bullet+}$  (885 nm), the 770 nm band is clearly evident.

The series of reactions shown in Scheme 2 can account for the above and other results. The radical cation initially formed in the one-electron oxidation (eq 1) reacts with excess neutral carotenoid to give a radical cation dimer (eq 7). This species

then deprotonates (eq 8) and the resulting “dimeric” radical ( ${}^{\#}[\text{Car}]_2^{\bullet}$ ) is oxidized to the deprotonated dimeric cation ( ${}^{\#}[\text{Car}]_2^+$ , eq 9). Deprotonation (eq 8) of an excited species formed upon irradiation is expected to be facile. Although all transient species are expected to be more stable at lower temperatures and any one of those formed in reactions 7–9 could, a priori, give rise to the 770 nm absorption, results of other experiments are best interpreted in terms of the  ${}^{\#}[\text{Car}]_2^+$  species. Thus, at  $-10^\circ\text{C}$ , when the intensity of the 770 nm band is enhanced, an additional cathodic peak is observed at low potential (Figure 1b, solid line, peak 6). This low value seems to be incompatible with the reduction of the  $[\text{Car}]_2^{\bullet+}$  species, since other radical cations, e.g.,  $\text{Car}^{\bullet+}$ , are reduced at very much higher positive potential ( $E^0_2 = 0.65$  V). Similarly, the deprotonated dimeric radical ( ${}^{\#}[\text{Car}]_2^{\bullet}$ ) is unlikely, since it is expected to be reduced at much lower (negative) potential. We ascribe the low potential peak ( $E^0_4 = 20 \pm 5$  mV) to the reduction of  ${}^{\#}[\text{Car}]_2^+$  (eq 10):



The reduction potential ( $E^0_3$ ) of monomeric canthaxanthin cations ( $\text{Car}^{\bullet+}$ ) is  $290 \pm 5$  mV (Figure 1).

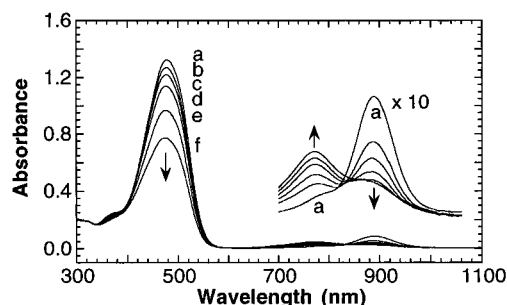
Only minor amounts of canthaxanthin radical cations generated by BE in the presence of TBAHFP (0.05 M) are converted to the “dimeric” species  ${}^{\#}[\text{Car}]_2^+$  upon irradiation. This is not surprising, since the neutral carotenoid molecules are surrounded by the ionic electrolyte that is present in large amounts and since ion pairs are formed between the radical cations and the electrolyte anion,  $\text{PF}_6^-$  (see above).

The tendency of radical cations to form dimers has long been known for aromatics and olefins.<sup>28–32</sup> Formation of radical cation dimers by association of the shorter chain carotenoids (four chain double-bonds) retinal, retinoic acid, and methyl retinoate radical cations with their own parent polyenes by pulse radiolysis in acetone has been discussed.<sup>33</sup> In addition, formation of neutral retinal and retinoic acid dimers in dry alkane solvents has also been well studied.<sup>34,35</sup> The dimers of retinoic acid have a tail-to-tail structure formed by hydrogen bonding but a partial sandwich configuration for retinal. The radical cation monomer/dimer equilibrium is especially favorable for polyenes containing terminal oxygen atoms.

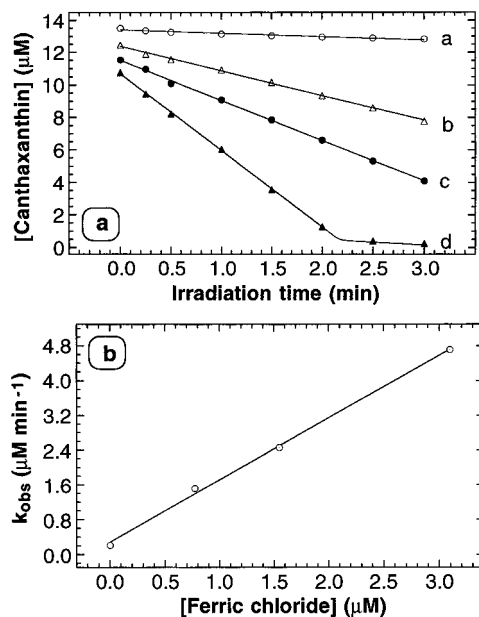
For the longer chain carotenoids (nine chain double bonds), although no discussion of dimer formation has been reported to our knowledge, the synthetic  $\alpha,\omega$ -diphenylcarotene formed polymers upon electrochemical oxidation in dichloromethane.<sup>36,37</sup> Formation of carbon–carbon bonds at the C7 and C7' positions has been suggested.

The structure of the dehydrodimer radical cation is unlikely to be of the aggregate or associative type, since parallel alignment and close approach of the monomeric units would be prevented by the bulky terminal trimethylcyclohexene substituents. It is possible that formation of a sigma bond is involved.

**Photocatalytic Degradation of Canthaxanthin by Ferric Chloride.** In the absence of  $\text{FeCl}_3$ , after irradiating a  $\text{CH}_2\text{Cl}_2$  solution of all-trans canthaxanthin with near-UV to visible light for three min, the only detectable change in the absorption spectrum is a small decrease (<3%) in the absorbance of the neutral species (480 nm). In contrast, when the solution also contains small amounts of  $\text{FeCl}_3$  (0.06 mol equiv), which results in immediate formation of approximately stoichiometric amounts of radical cations (Figure 3, trace a) and  $\text{Fe(II)}$  in the absence of light, subsequent irradiation for increasing periods of time causes not only a rapid decrease in both the neutral (480 nm) and the radical cation (885 nm) species but also an increase in



**Figure 3.** Optical absorption spectra after irradiating 12  $\mu\text{M}$  all-trans canthaxanthin in  $\text{CH}_2\text{Cl}_2$  in the presence of 0.7  $\mu\text{M}$   $\text{FeCl}_3$  with near-UV to visible light for (a) 0, (b) 0.25, (c) 0.5, (d) 1.0, (e) 2.0, and (f) 3.0 min.



**Figure 4.** (a) Photodegradation of 13.5  $\mu\text{M}$  canthaxanthin in  $\text{CH}_2\text{Cl}_2$  by irradiation with near-UV to visible light for increasing periods of time in the presence of (a) 0, (b) 0.77, (c) 1.55, and (d) 3.1  $\mu\text{M}$   $\text{FeCl}_3$ . (b) Apparent photodegradation reaction rate constants ( $k_{\text{obs}}$ ) of canthaxanthin as a function of  $\text{FeCl}_3$  concentration. Data are from part a.

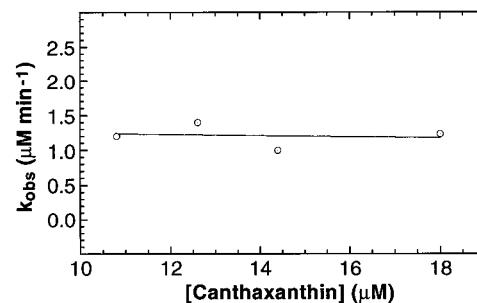
the absorption at 770 nm (Figure 3, traces b–f) as was discussed above. For a given initial concentration of  $\text{FeCl}_3$ , the concentration of the remaining neutral canthaxanthin shows a linear dependence on the irradiation time (Figure 4a), i.e., the photodegradation reaction is zero-order in canthaxanthin. With larger amounts of  $\text{FeCl}_3$  the rates of these changes increase. The photoreaction is first-order in  $\text{FeCl}_3$ , since the observed zero-order constants for the disappearance of canthaxanthin increase linearly with the initial  $\text{FeCl}_3$  concentration (Figure 4b). That the reaction is zero-order in canthaxanthin is also evident from Figure 5 in which the slopes ( $k_{\text{obs}}$ ) of  $[\text{Car}]$  vs irradiation time at constant  $[\text{FeCl}_3]$  are plotted vs various initial canthaxanthin concentrations.

The rate of this photodecay of canthaxanthin can be described by eq 11:

$$-\frac{d[\text{Car}]}{dt} = k' + k_1[\text{FeCl}_3]_0 = k_{\text{obs}} \quad (11)$$

where  $k'$  ( $\text{M min}^{-1}$ ) is the zero-order rate constant of the photodecay in the absence of  $\text{FeCl}_3$ ,  $k_1$  ( $\text{min}^{-1}$ ) is the first-order rate constant,  $[\text{FeCl}_3]_0$  ( $\mu\text{M}$ ) is the initial  $\text{FeCl}_3$  concentration, and  $k_{\text{obs}}$  ( $\mu\text{M min}^{-1}$ ) is the observed zero-order rate constant. Numerical data are given in Table 1.

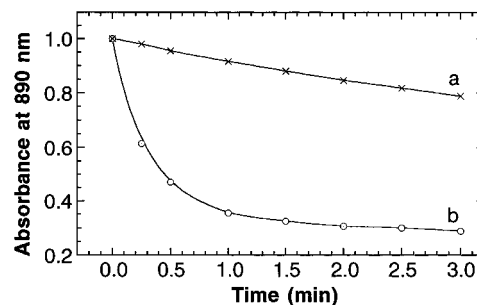
As stated above, canthaxanthin radical cations generated by chemical oxidation with ferric chloride also undergo rapid



**Figure 5.** Apparent photodegradation reaction rate constants ( $k_{\text{obs}}$ ) as a function of canthaxanthin in the presence of 0.43  $\mu\text{M}$   $\text{FeCl}_3$ . Other conditions are the same as in Figure 4a.

**TABLE 1: Effect of Initial Ferric Chloride Concentration on the Photodegradation Rates**

canthaxanthin		$\beta$ -carotene	
[ferric chloride] ( $\mu\text{M}$ )	$k_{\text{obs}}$ ( $\mu\text{M min}^{-1}$ )	[ferric chloride] ( $\mu\text{M}$ )	$k_{\text{obs}}$ ( $\mu\text{M min}^{-1}$ )
0	0.21 ( $k'$ )	0	0.79 ( $k'$ )
0.77	1.52	0.19	1.42
1.55	2.46	0.39	2.11
3.10	4.71	0.78	3.31
$k_1 = 1.43 \text{ min}^{-1}$		$k_1 = 3.30 \text{ min}^{-1}$	

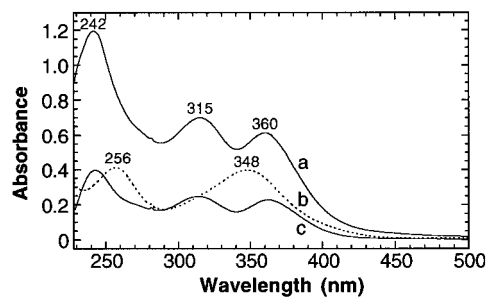


**Figure 6.** Time dependence of absorbance of canthaxanthin radical cations formed by  $\text{FeCl}_3$  in  $\text{CH}_2\text{Cl}_2$ : (a) without irradiation; (b) upon irradiation for increasing periods of time.

degradation upon irradiation (Figure 3, 885 nm). For comparison, curve a in Figure 6 shows the relatively slow natural decay of the radical cations generated by ferric chloride. The half-life of the radical cations decreases from several minutes to only about 30 s upon irradiation with near-UV to visible light in the presence of ferric chloride. This is due to increased interaction between the radical cations and their parent molecules to form “dimers” as discussed above. However, since  $\text{Fe(III)}$  in the excited state is a much stronger oxidizing agent than it is in the ground state,<sup>38</sup> competing oxidation of the radical cations cannot be ruled out; the products would be dications.

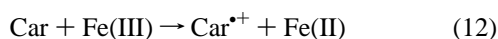
**Photocatalytic Degradation Mechanism.** Similar irradiation experiments with solutions that were degassed with dried and  $\text{CH}_2\text{Cl}_2$ -saturated nitrogen for 15 min to remove any possible oxygen showed the same results, indicating that the enhanced photodecay is not due to a reaction involving oxygen.

Other studies have shown that ferric chloride is photocatalytically active in the oxidation and degradation of organic compounds. In the presence of ferric chloride, photodegradation of polyolefins,<sup>39</sup> polypropylene,<sup>40</sup> and oxalic acid,<sup>41</sup> photochemical oxidation of alcohols,<sup>38</sup> photoinduced hydrogen transfer reactions,<sup>42</sup> and photocatalyzed reactions between alcohols and alkyl isocyanates<sup>43</sup> have been reported. In addition to direct energy transfer from excited  $\text{FeCl}_3$ , promotion of oxidation and/or degradation has also been ascribed to free chlorine or hydroxy radicals that are formed upon irradiation of  $\text{Fe(III)}$  in aqueous systems with UV light.<sup>39,43,44</sup>



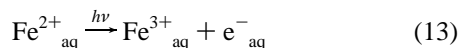
**Figure 7.** Optical absorption spectra of  $\text{CH}_2\text{Cl}_2$  solutions of (a)  $\text{FeCl}_3$  and (b)  $\text{FeCl}_2$  and (c) after irradiating solution b with near-UV to visible light for 1 min. Solution b was purged with dry  $\text{N}_2$  for 10 min prior to irradiation.

The photocatalyzed oxidation of canthaxanthin by  $\text{Fe(III)}$  in  $\text{CH}_2\text{Cl}_2$  does not seem to involve the above mechanisms because the initially added small amounts of  $\text{Fe(III)}$  species ( $<0.26$  mol equiv) are rapidly converted to  $\text{Fe(II)}$  according to eq 12:



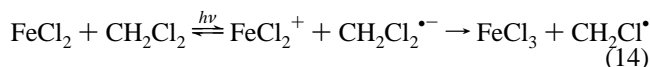
When  $\text{Fe(III)}$  is not in excess, each mole of  $\text{Fe(III)}$  generates about 1 equiv of canthaxanthin radical cations at room temperature in the absence of light (Figure 2b, solid line). The photodecay reaction therefore must involve the  $\text{Fe(II)}$  species. Since the rate of photodecay is first-order in initial concentration of ferric chloride, it seemed likely that  $\text{Fe(II)}$  in  $\text{CH}_2\text{Cl}_2$  is photooxidized to  $\text{Fe(III)}$ , which then reacts with excess neutral carotenoid.

Photoejection of electrons in aqueous solution from  $\text{Fe(II)}$  yielding  $\text{Fe(III)}$  and hydrated electrons has been extensively studied.<sup>45–48</sup> The reaction can be expressed as



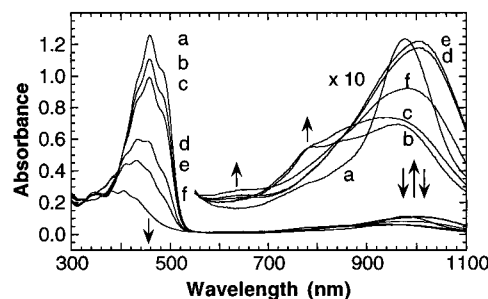
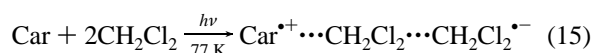
The solvated electrons then react with scavengers such as nitrous oxide in the solution.

In the case of this study, a similar mechanism occurs except that in the dichloromethane solution the solvent acts as electron acceptor. The following reactions could account for the photooxidation of  $\text{Fe(II)}$  to  $\text{Fe(III)}$  in  $\text{CH}_2\text{Cl}_2$ .



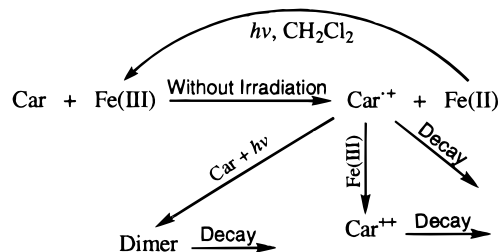
The regenerated  $\text{Fe(III)}$  species oxidizes remaining unreacted  $\text{Car}$  to  $\text{Car}^{\bullet+}$ . The amount of  $\text{Fe(III)}$  in the solution remains almost the same as the initial concentration of  $\text{FeCl}_3$  owing to its continual regeneration during photoexcitation. The following observation supports the assumption that  $\text{FeCl}_3$  is formed by irradiation of  $\text{FeCl}_2$  in dichloromethane solution. The UV spectra in  $\text{CH}_2\text{Cl}_2$  of  $\text{FeCl}_3$  (Figure 7, trace a) and  $\text{FeCl}_2$  (Figure 7, trace b) differ considerably. Brief irradiation (1 min) of a  $\text{N}_2$ -purged  $\text{CH}_2\text{Cl}_2$  solution of  $\text{FeCl}_2$  (under the same conditions used for the irradiation of the canthaxanthin/ $\text{FeCl}_3/\text{CH}_2\text{Cl}_2$  system) resulted in the characteristic spectrum of  $\text{FeCl}_3$  (Figure 7, trace c).

Although EPR evidence for the  $\text{CH}_2\text{Cl}_2$  radical anion formed in eq 14 has not been obtained, perhaps because of the low concentration of  $\text{FeCl}_2$  and/or the short lifetime of the radical, formation of solvent-separated radical ion pairs generated by photolysis of carotenoid solutions in  $\text{CH}_2\text{Cl}_2$  at 77 K has been observed.<sup>49</sup>



**Figure 8.** Optical absorption spectra after irradiating  $13 \mu\text{M}$  all-trans  $\beta$ -carotene in  $\text{CH}_2\text{Cl}_2$  in the presence of  $0.8 \mu\text{M}$   $\text{FeCl}_3$  with near-UV to visible light for (a) 0, (b) 0.25, (c) 0.5, (d) 1.5, (e) 2.0, and (f) 3.0 min.

### SCHEME 3



When the frozen solution is warmed, the  $\text{CH}_2\text{Cl}_2^{\bullet-}$  species rapidly decomposes to  $\text{CH}_2\text{Cl}^{\bullet}$  and  $\text{Cl}^-$ . The resulting  $\text{CH}_2\text{Cl}^{\bullet}$  is more stable and has been detected by EPR spectroscopy. Formation of the solvent-separated radical ion pair  $\text{Car}^{\bullet+} \cdots \text{CCl}_4 \cdots \text{CCl}_4^{\bullet-}$  has been reported<sup>50</sup> to occur at room temperature.

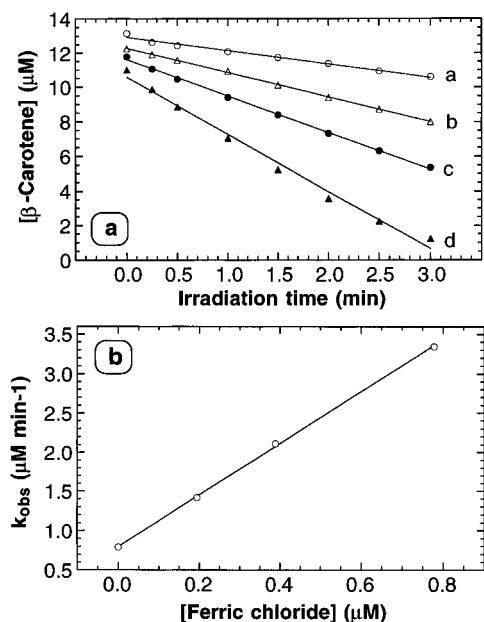
The reactions involved in the oxidation of canthaxanthin by small amounts of  $\text{FeCl}_3$  in the absence and presence of light are summarized in Scheme 3.

**$\beta$ -Carotene.** Unlike canthaxanthin, the CV of  $\beta$ -carotene displays only one pair of redox peaks (Figure 1), since the  $E^0_1$  and  $E^0_2$  values are nearly the same.<sup>21–25</sup> The lower anodic peak potential of  $\beta$ -carotene compared with that of canthaxanthin indicates that  $\beta$ -carotene is more easily oxidized.

$\beta$ -Carotene radical cations could also be formed by either bulk electrolysis or chemical oxidation with ferric chloride in  $\text{CH}_2\text{Cl}_2$ . The optical absorption spectrum obtained after addition of small amounts of ferric chloride ( $\sim 0.06$  mol equiv) to a  $\text{CH}_2\text{Cl}_2$  solution of  $\beta$ -carotene is shown in Figure 8, trace a. In addition to the absorption at 460 nm of neutral  $\beta$ -carotene, an absorption maximum at 970 nm, due to cation radicals, is observed. Furthermore, similar to the behavior of canthaxanthin, this spectrum displays a shoulder near 780 nm.

The spectral changes observed when the above solution is irradiated for increasing periods of time with near-UV to visible light indicate that the photochemical behavior of  $\beta$ -carotene is more complicated than that of canthaxanthin. Significant decay of neutral  $\beta$ -carotene is observed (Figure 8, traces b–f). Brief irradiation (0.25 min) causes a rapid decrease in the radical cation absorption (970 nm) and an increase in the 780 nm shoulder (Figure 8, trace b). However, in contrast to canthaxanthin, further irradiation causes not only an increase but also broadening and a red shift of the absorption near 970 nm (Figure 8, traces c–e). After 2 min of irradiation, the absorption near 970 nm decreases again (Figure 8, trace f). It is possible that not only dimers but also higher order (i.e.,  $n > 2$ ) aggregates might be formed during the processes. Further investigation of these phenomena need to be carried out in the future.

As for canthaxanthin, the photodegradation reaction of neutral  $\beta$ -carotene is zero-order in  $\beta$ -carotene at a given  $\text{FeCl}_3$  concentration (Figure 9a) and first-order in initial concentration of  $\text{FeCl}_3$  (Figure 9b). Numerical data are listed in Table 1. The



**Figure 9.** (a) Photodegradation of 13.2  $\mu\text{M}$   $\beta$ -carotene in  $\text{CH}_2\text{Cl}_2$  by irradiation with near-UV to visible light for increasing periods of time in the presence of (a) 0, (b) 0.19, (c) 0.39, and (d) 0.78  $\mu\text{M}$   $\text{FeCl}_3$ . (b) Apparent photodegradation reaction rate constants ( $k_{\text{obs}}$ ) as a function of  $\text{FeCl}_3$  concentration. Data are from part a.

photodegradation rate of  $\beta$ -carotene is quite significant even in the absence of  $\text{FeCl}_3$  (Figure 9a, curve a); its rate constant is much larger than that of canthaxanthin ( $k' = 0.79$  and  $0.21 \mu\text{M min}^{-1}$  for  $\beta$ -carotene and canthaxanthin, respectively), which indicates that canthaxanthin is photochemically more stable than  $\beta$ -carotene. The ferric photocatalytic degradation rate of  $\beta$ -carotene is also much larger than that of canthaxanthin ( $k_1 = 3.30$  and  $1.43 \text{ min}^{-1}$  for  $\beta$ -carotene and canthaxanthin, respectively), consistent with the fact that  $\beta$ -carotene is more easily oxidized.

## Conclusions

Canthaxanthin radical cations, generated by electrolysis, form cation dehydrodimers, which are detectable at lower temperature, or when generated by  $\text{FeCl}_3$  oxidation in dichloromethane upon irradiation at room temperature. The product has an absorption maximum near 770 nm and shows a reduction peak ( $E^0_4 = 20 \pm 5 \text{ mV}$ ) in a CV. In the presence of the supporting electrolyte TBAHFP, canthaxanthin radical cations form ion pairs with  $\text{PF}_6^-$ , resulting in a 10 nm blue shift of the absorption maximum of the radical cations.  $\beta$ -Carotene shows more complicated photochemical behavior than canthaxanthin.

Rapid photodegradation of canthaxanthin and  $\beta$ -carotene occurs upon irradiation with near-UV to visible light in the presence of small amounts of ferric chloride in  $\text{CH}_2\text{Cl}_2$ . The iron-catalyzed photodegradation rate is independent of the carotenoid concentration and first-order in the initial concentration of ferric chloride. Irradiation of  $\text{FeCl}_2$  in  $\text{CH}_2\text{Cl}_2$  results in the same UV spectrum as is observed for anhydrous  $\text{FeCl}_3$ , which, if a neutral carotenoid is present, is again reduced to ferrous species. In consequence, irradiation of carotenoid  $\text{CH}_2\text{Cl}_2$  solutions, containing much less than one mol equiv of either  $\text{Fe(III)}$  or  $\text{Fe(II)}$  species, causes the destruction of all of the carotenoid.

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