Acta Chemica Scandinavica, 1999: 53: 114 123
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ACTA
CHEMICA SCANDINAVICA
ISSN 0904 213X

Stereoisomerization of Allenic Carotenoids – Kinetic, Thermodynamic and Mechanistic Aspects

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Refvem, T., Strand, A., Kjeldstad, B., Haugan, J. A. and Liaaen-Jensen, S., 1999. Stereoisomerization of Allenic Carotenoids Kinetic, Thermodynamic and Mechanistic Aspects. – Acta Chem. Scand. 53: 114–123. © Acta Chemica Scandinavica 1999.

Diphenyl diselenide mediated photoisomerization of the allenic carotenoids peridinin and fucoxanthin in benzene solution have been studied, with the aim of optimizing synthetic yields of (6′*S*)-allenes. The effects of catalyst concentration and of light quality and intensity have been investigated using no catalyst, diphenyl diselenide or iodine as catalysts.

Similar quasi-equilibrium mixtures of geometrical isomers were obtained with both catalysts, indicating a thermodynamic equilibrium.

Considerably higher *R* to *S* allenic conversion was obtained with diphenyl diselenide than with iodine. Increased allenic isomerization rate upon UVA radiation and the effect of radical inhibition support a radical mechanism initiated by photolysis of diphenyl diselenide. The suggested mechanism for the diphenyl diselenide mediated isomerization may account for the high yield obtained [≈88% (6′*S*)-allene of recovered carotenoid].

Recently we have demonstrated that diphenyl diselenide (Ph_2Se_2) represents a good alternative to iodine (I_2) as a mediator for photochemical stereoisomerization of carotenoids. Geometrical (*E*/*Z*) as well as allenic (*R*/*S*) isomerization may be achieved under selected and reproducible conditions.[1] In line with the radical mechanism proposed by Barton and co-workers for Ph_2Se_2 mediated isomerization of the central double bond of the triene system in ergocalciferol,[2] we have proposed a simplified radical mechanism for the isomerization of the allenic bond in carotenoids by means of Ph_2Se_2.[3] This paper addresses kinetic, thermodynamic and mechanistic aspects in a comparative study using either Ph_2Se_2 or I_2 as the mediator for allenic photoisomerization of selected carotenoids, aimed at optimizing yields of (6′*S*)-allenes.

Results and discussion

The allenic peridinin (**1**), Scheme 1, was selected for this study because a suitable HPLC system had previously been developed for the simultaneous separation of *E*/*Z*-isomers of both the allenic (6′*R*)- and (6′*S*)-series.[4–6] Moreover, the all-*trans*, 9′-*cis*-, 11-*cis*- and 13-*cis*-isomers of the (6′*R*)-series (**1**) as well as all-*trans*-(6′*S*)-peridinin (**2**) have been characterized including ^{1}H NMR assignments.[6] The 9′-*cis*-, 11-*cis*- and 13-*cis*-isomers of the (6′*S*)-series have been identified from isomerization evidence, VIS absorption data and relative abundance.[6] The *cis*/*trans* nomenclature is preferred for in-chain substituted carotenoids including peridinin (**1**).[7]

Some experiments were conducted with fucoxanthin (**3**), Scheme 1. In this case a suitable HPLC system was also available,[8] and the geometrical isomers of the (6′*R*)-allenic (**3**)[9] and (6′*S*)-allenic (**4**)[8] series have previously been characterized.

The effect of light quality and intensity. The use of glass containers restricts the application of light to $\lambda \geqslant 320$ nm. In Fig. 1 is presented the UVA–VIS absorption spectra of Ph_2Se_2 and I_2 in benzene solution. For comparison peridinin (**1**) has λ_{max} 475 nm and $\varepsilon = 83\,500$ (ethanol).[10] Relative amounts of carotenoid and catalyst are given for the individual experiments. It was of particular interest to examine (i) the efficiency of radiation at $\lambda < 410$ nm (mainly UVA) for Ph_2Se_2-mediated stereoisomerization of carotenoids in comparison with that of (ii) visible light and (iii) visible light and near UV. The three different light qualities were obtained by use of

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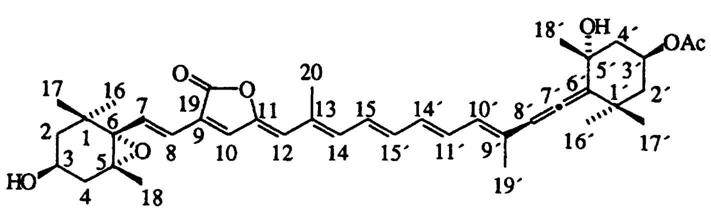
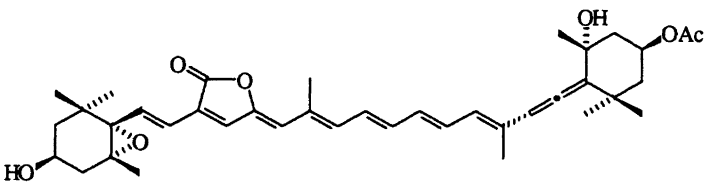


all-*trans*-(6′*R*)-Peridinin (**1**)



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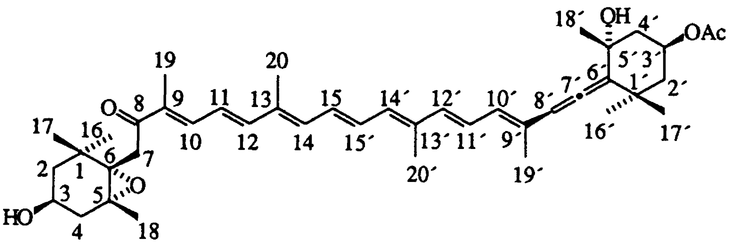
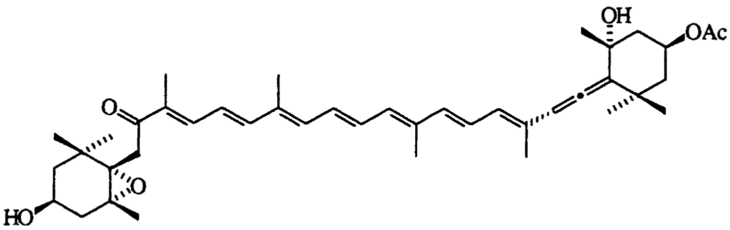


all-*E*-(6′*R*)-Fucoxanthin (**3**)



all-*E*-(6′*S*)-Fucoxanthin (**4**)

Scheme 1.

(i) two UVA fluorescent tubes, (ii) a tungsten lamp with a special Plexi-glass filter (cut-off λ < 390 nm) and (iii) a tungsten lamp. Varied intensities of the light qualities (ii) and (iii) were also employed by changing the distance between the sample and irradiation sources, see Table 1.

The application of Ph_2Se_2 as a mediator for photochemical stereoisomerization, previously demonstrated for fucoxanthin (**3**),[1] was examined for peridinin (**1**) in visible light and near UV (conditions iii) and with UVA irradiation (conditions i). The results presented in Table 2 demonstrate that *E*/*Z* stereomutation as well as reversible allenic *R*/*S* isomerization was achieved. A quasi-equilibrium was reached with 200 mol% Ph_2Se_2 at high light intensity within 3–4 h and in UVA within 0.25–1.25 h. The reproducible, rate-determining effect of different mediator concentrations is demonstrated in Fig. 2.

When the near UV contribution from the tungsten lamp was reduced (cf. conditions ii, Table 1) the rate of allenic isomerization of peridinin (**1**) was considerably reduced both with 40 mol% and 200 mol% Ph_2Se_2 at high light intensities, Fig. 3.

Stereoisomerization effected with UVA, Fig. 4, revealed that Ph_2Se_2 was a particularly efficient mediator for the allenic isomerization in this wavelength region. Similar effects were observed with both 40 mol% and 200 mol% Ph_2Se_2.

Comparative studies were conducted with peridinin (**1**) using I_2 as the catalyst. The same quasi-equilibrium mixture of *E*/*Z*-isomers of the allenic (6′*R*)-(**1**) and (6′*S*)-(**2**) series as previously reported[6] was obtained, independent of light conditions and catalyst (I_2) concentration.

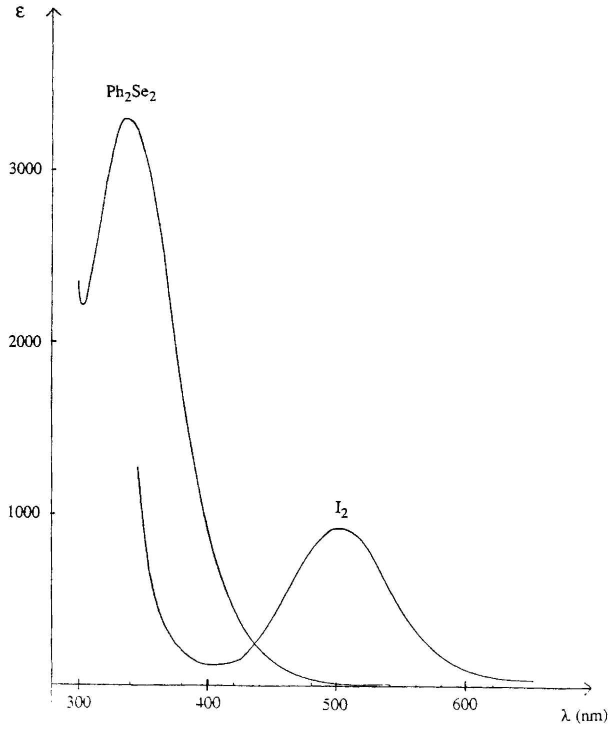


Fig. 1. Absorption spectra of diphenyl diselenide and iodine in benzene solution.

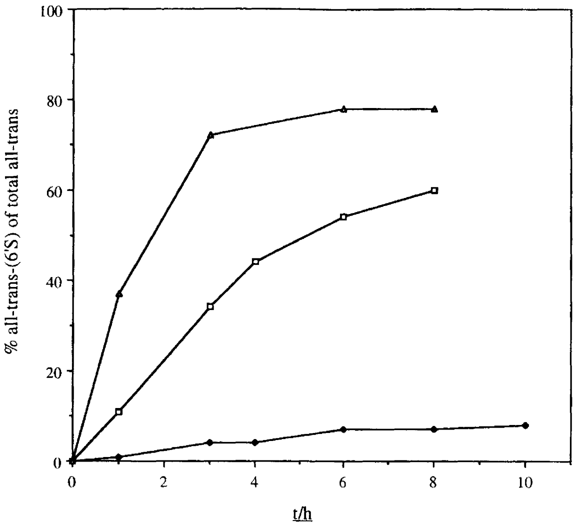


Fig. 2. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high intensity visible light + near UV, with 4 mol% (◆), 40 mol% (□) and 200 mol% (▲) Ph$_2$Se$_2$.

The effect of light intensity upon visible and near UV irradiation, on the rate of allenic *R*/*S* isomerization, is demonstrated in Fig. 5 with 5 mol% I$_2$. The results were consistent with the previously observed problems in obtaining allenic isomerization at low sunlight intensity.[9] At high light intensity the effect of increasing the I$_2$ concentration to 50 mol% was less pronounced than for Ph$_2$Se$_2$, Fig. 6. At 250 mol% I$_2$ decomposition of the carotenoid occurred. The rate of allenic isomerization with 5 mol% and 50 mol% I$_2$ was the same for near UVA

Table 1. Light quality and intensity at the test tube employed for stereoisomerizations of allenic carotenoids in benzene solution.

		Light intensity	
Light quality	Light source	UVA (mW cm^{-2})	VIS (µE m^{-2} s^{-1})
(i) UVA	2 Fluorescent tubes	≈20	7
(ii) Visible, HL[a]	Tungsten lamp	≈0.2	3150
Visible, LL[b]	with Plexi-glass	≈0.01	500
(iii) Visible + near UV, HL[a]	Tungsten lamp	≈2	3150
Visible + near UV, LL[b]		≈0.2	500
Visible + near UV, VLL[c]		—	100

[a] High light intensity. [b] Low light intensity. [c] Very low light intensity. Intensity refers to the visible light level (not UV irradiance).

Table 2. Stereoisomeric composition at quasi-equilibrium upon isomerization of all-*trans*-(6′*R*)-peridinin (**1**) and reversible isomerization of all-*trans*-(6′*S*)-peridinin (**2**) under different light conditions [UVA or high light intensity visible light + near UV (HL)] and exposure times using 200 mol% Ph$_2$Se$_2$.

		% Total peridinin (**1, 2**)			
		all-*trans*-(6′*R*) (**1**)[b]		all-*trans*-(6′*S*) (**2**)[b]	
Optical isomer	Geometrical isomer	4 h, HL	1.25 h, UVA	3 h, HL	0.25 h, UVA
6′*R*	all-*trans* (**1**)[a]	15	12	6	5
6′*S*	all-*trans* (**2**)	54	59	61	64
	9′-*cis* (**2a**)	17	17	19	18
	11-*cis* (**2b**)	5	3	5	5
	13-*cis* (**2c**)	9	8	9	9

[a] Other geometrical isomers in the (6′*R*)-series presumably present. [b] Initial isomer.

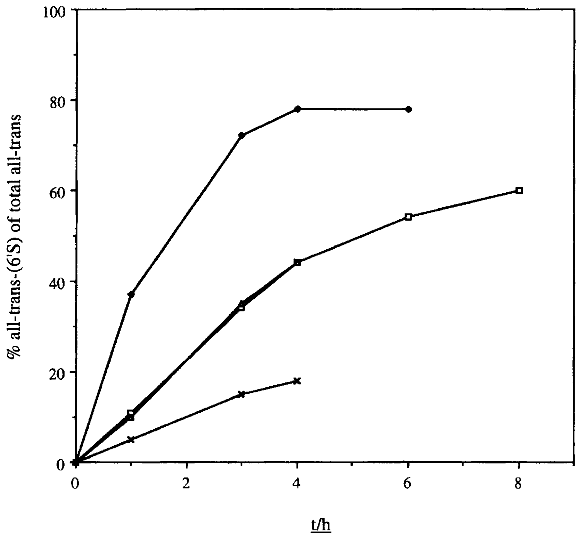


Fig. 3. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high intensity visible light + near UV, with 200 mol% (◆) and 40 mol% (□) Ph_2Se_2, and at high intensity visible light with reduced UV with 200 mol% (▲) and 40 mol% (×) Ph_2Se_2.

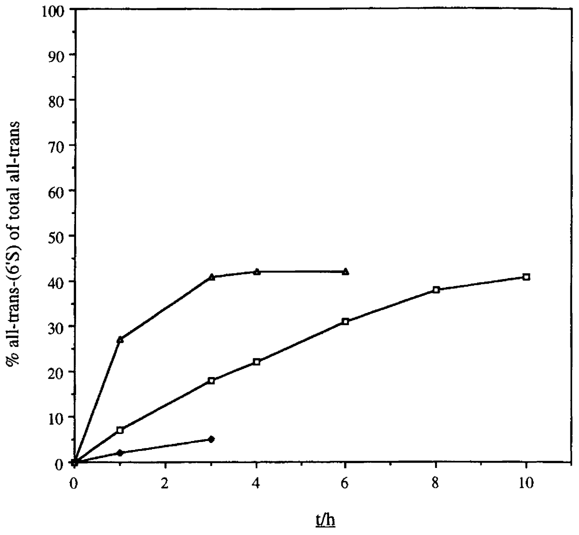


Fig. 5. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at visible light + near UV irradiation at very low light (◆), low light (□) and high light (▲) intensity, with 5 mol% I_2.

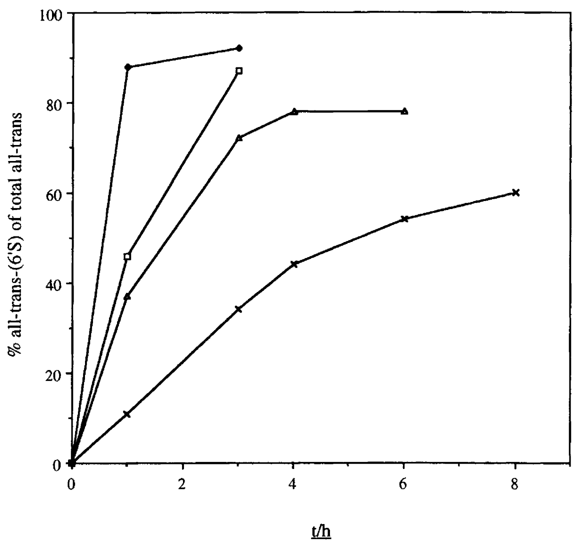


Fig. 4. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at UVA with 200 mol% (◆) and 40 mol% (□) Ph_2Se_2, and at high intensity visible light + near UV with 200 mol% (▲) and 40 mol% (×) Ph_2Se_2.

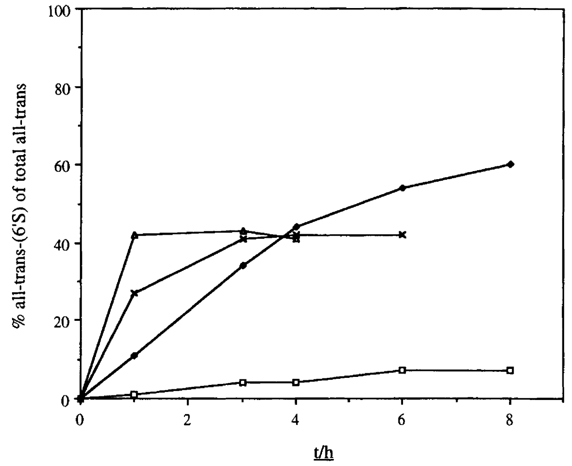


Fig. 6. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high intensity visible light + near UV, with 40 mol% (◆) Ph_2Se_2, 4 mol% (□) Ph_2Se_2, 50 mol% (▲) I_2 and 5 mol% (×) I_2.

and visible light exposure for irradiation times longer than 1 h (Table 3). However, the intensity seems to play a role at low (5 mol%) catalytic concentrations of I_2 (Fig. 5). Reciprocity (dose response) appears to be valid for 5 mol% I_2, since the threshold isomerization ratio of 40% *S*-allene was achieved after less than 3 h irradiation with high light intensity, while approximately 10 h or more were required with a low intensity level (Fig. 5, Table 3). Reducing the UVA contribution of the irradiation or applying UVA radiation did not effect the rate of allenic isomerization with 50 mol% I_2 as the catalyst, Fig. 7.

In conclusion, UVA irradiation had a pronounced effect on the rate of stereoisomerization of peridinin (**1**) using Ph_2Se_2 as the mediator. At high light intensity (conditions iii, Table 1) I_2 caused faster isomerization than Ph_2Se_2 employing 4–50 mol% catalyst, Fig. 6.

Pigment recoveries. Experiments were performed under an atmosphere of nitrogen. The carotenoid concentration (40 μg ml^{-1} benzene; 6.34×10^{-5} M) was kept constant.

Table 3. Stereoisomeric composition at quasi-equilibrium upon isomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high intensity visible light + near UV (HL) with 50 mol% or 5 mol% iodine as the catalyst.

		% Total peridinin (**1**, **2**)	
Optical isomer	Geometrical isomer	HL, 50 mol% I_2, 1 h	HL, 5 mol% I_2, 3 h
6′*R*	all-*trans* (**1**)	37	37
	9′-*cis* (**1a**)	9	9
	11-*cis* (**1b**)	5	6
	13-*cis* (**1c**)	1	2
6′*S*	all-*trans* (**2**)	27	26
	9′-*cis* (**2a**)	8	8
	11-*cis* (**2b**)	5	5
	13-*cis* (**2c**)	5	4
Not identified		3	3

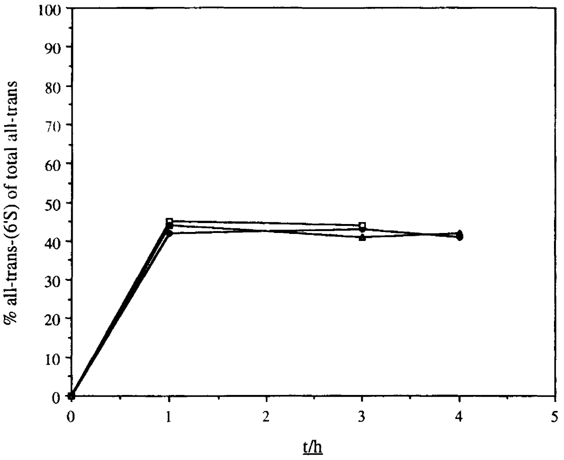


Fig. 7. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1**, **2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high light intensity visible light + near UV (◆), visible light (□) and UVA (▲), with 50 mol% I_2.

Pigment recoveries were comparable when using the same light conditions and mol% (in the range ca. 4–50 mol%) Ph_2Se_2 or I_2 relative to the carotenoid. However, pigment recoveries decreased with increasing catalyst concentration and 250 mol% I_2 caused extensive pigment decomposition.

The pigment recoveries decreased when the intensity of visible light was changed from low to high, and further to visible and near UV at high light intensities. Prolonged irradiation by UVA alone resulted in the lowest recoveries.

In general recoveries of 20–85% were achieved at quasi-equilibrium for *R*/*S* allenic isomerization under different isomerization conditions with Ph_2Se_2 or I_2 catalysis.

Radical inhibition. The results discussed above supported homolysis of Ph_2Se_2 upon UVA radiation. The effect of adding hydroquinone as a general radical quencher[11] is illustrated in Fig. 8. These experiments were performed with 200 mol% Ph_2Se_2 under conditions favoring *R*/*S* allenic isomerization, with UVA or high intensity visible light and near UV irradiation, but with an inhibitor concentration of 10 or 400 molar equivalents, relative to Ph_2Se_2.

A pronounced decrease in the isomerization rate was observed at high light intensity with 10 mol equivalents of hydroquinone. The effect of further increasing the inhibitor concentration was small. The results lend support to a radical mechanism.

Under UVA conditions the radical inhibitor effected only a small reduction of the isomerization rate, compatible with the high efficiency of UVA irradiation for allenic isomerization, cf. also Fig. 10.

Similar experiments, Fig. 9, were performed with I_2 as the catalyst under optimum conditions for *R*/*S* allenic isomerization (5 mol% I_2 in high intensity visible light, cf. Fig. 5) with a hydroquinone concentration of 10 mol equivalents relative to I_2, and with no effect on the isomerization. However, with 400 mol equivalents of the inhibitor, a decreased isomerization rate and a new allenic *R*/*S* quasi-equilibrium, with less (6′*S*)-peridinin (**2**) (~30% of total **1** + **2** versus ~42% in the absence of hydroquinone), was observed. One possible rationalization for the effect at very high inhibitor concentration is the gradual destruction of the catalyst.

All experiments with the radical quencher were performed in diethyl ether–benzene (1 : 1.7) owing to the poor solubility of hydroquinone in neat benzene. The presence of diethyl ether had no effect on the isomerization rate, as demonstrated in an experiment carried out

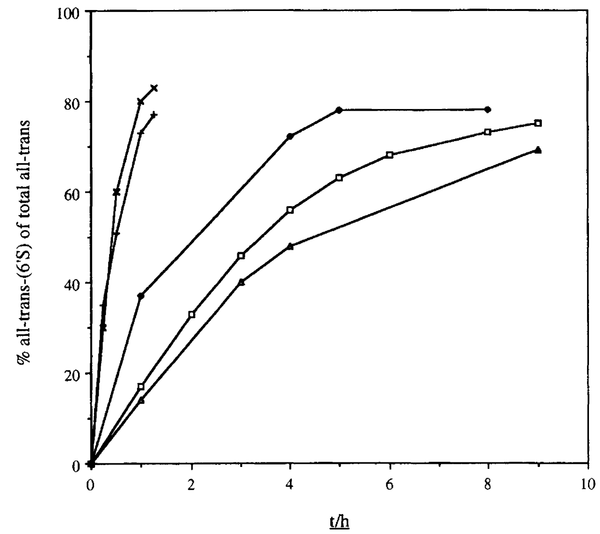


Fig. 8. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1**, **2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high light intensity visible light + near UV with 200 mol% Ph_2Se_2 without (◆) and with 10 (□) or 400 (▲) mol equivalents of hydroquinone as a radical quencher. Analogous experiments carried out in UVA with no hydroquinone (×) and with 10 mol equivalents (+) of hydroquinone.

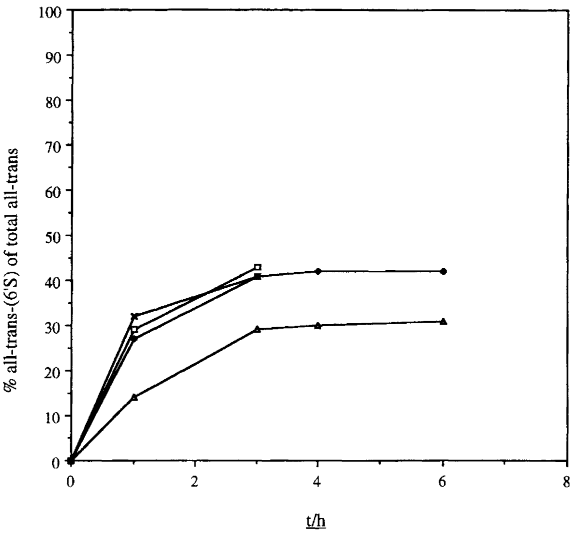


Fig. 9. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at high light intensity visible light + near UV with 5 mol% I_2 without (◆) and with 10 (□) or 400 (▲) mol equivalents of hydroquinone in diethyl ether–benzene 1 : 1.7. Parallel experiment with no hydroquinone in neat benzene solution (×).

with 5 mol% I_2 and no hydroquinone in diethyl ether–benzene (1 : 1.7), cf. Fig. 9.

Quasi-equilibrium mixtures. A similar ratio of *E*/*Z*-isomers within the allenic (6′*R*)–(**1**) and (6′*S*)-peridinin (**2**) series was obtained under quasi-equilibrium conditions using I_2 (Table 3) or Ph_2Se_2 (Table 2) catalysis. Ratios of *E*/*Z*-isomeric (6′*R*)-peridinins (**1**) under conditions not providing allenic isomerization are given in Table 4.

Since reproducible and very similar mixtures of *E*/*Z*-isomers were obtained for peridinin (**1** and **2**) with two different catalysts (≈5 mol%), it is inferred that this mixture may represent a thermodynamic equilibrium. The relative stability of different geometrical isomers (here observed in the order all-*trans*>9′-*cis*>11-*cis*≈13-*cis*) has not been calculated for peridinin (**1**). Recently reported data for fucoxanthin (**3**) revealed a closely similar ratio of different *E*/*Z*-isomers upon Ph_2Se_2- or I_2-catalyzed photoisomerization,[1] reflecting relative stability in the order all-*E*>9′*Z*>13′*Z*≈13*Z*>di-*Z*. The closely similar *E*/*Z* isomeric mixtures obtained with the two catalysts, also for fucoxanthin (**3**), support the conclusion of a thermodynamic equilibrium of *cis*/*trans* (*E*/*Z*) geometrical isomers.

However, a striking difference in the allenic (6′*S*) : (6′*R*) ratio was observed at quasi-equilibrium for peridinin (**1, 2**) when Ph_2Se_2 was employed as the mediator, ≈80:20 versus 45:55 with I_2. The same trend has recently been reported for fucoxanthin (**3, 4**),[1,8] although not yet discussed, namely a 75:25 versus 46:54 (6′*S*) to (6′*R*) ratio with Ph_2Se_2 and I_2 respectively. For Ph_2Se_2-mediated isomerization with UVA radiation, a ratio 88:12 was observed for fucoxanthin (**4**:**3**) in the present work.

The results presented in Fig. 10 demonstrate that similar *R*/*S* quasi-equilibrium mixtures were reached for peridinin (**1** or **2**) with Ph_2Se_2 irrespective of the nature, (6′*R*)- or (6′*S*)-configuration, of the starting isomer.

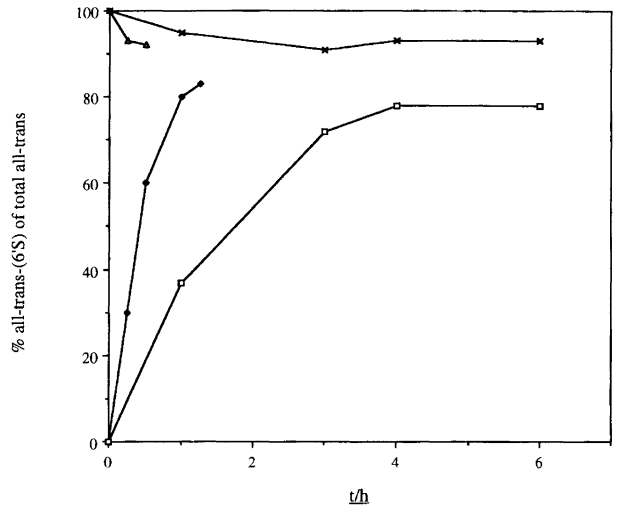
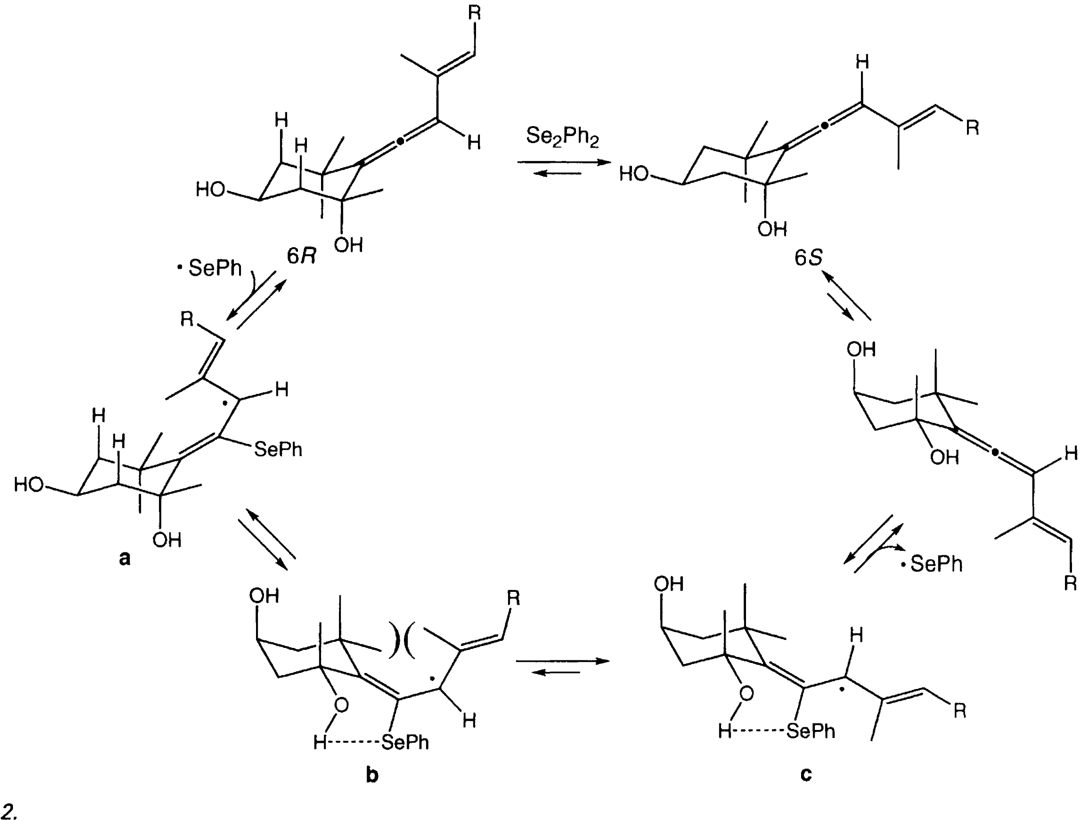


Fig. 10. Percentage all-*trans*-(6′*S*)-peridinin (**2**) of total all-*trans*-peridinin (**1, 2**) as a function of exposure time upon stereoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) at UVA (◆) and at high light intensity visible light + near UV (□), and reversible isomerization of all-*trans*-(6′*S*)-peridinin (**2**) to UVA (▲) and at high intensity visible light + near UV (×), with 200 mol% Ph_2Se_2.

Table 4. Conditions resulting in *trans*/*cis* stereomutation and no allenic isomerization of all-*trans*-(6′*R*)-peridinin (**1**).

			% Total (6′*R*)-peridinin (**1**)			
Light conditions[a]	Mol% Ph_2Se_2	Time	all-*trans*	9′-*cis*	11-*cis*	13-*cis*
VLL	0	4 h	74	14	12	0
LL	0	8 h	63	23	11	3
Darkness	40	2 months	74	17	9	0
VLL	4	1 h	63	23	10	3
LL	4	1 h	61	25	10	2
LL	4	3 h	61	25	10	3
LL	40	1 h	62	27	8	2
HL	4	1 h	59	25	12	3

[a] Darkness of visible light + near UV, for intensities cf. explanations in Table 1.



Scheme 2.

The high (6′*R*) to (6′*S*) conversion with Ph_2Se_2 for both peridinin (**1**) and fucoxanthin (**3**)[1] was unexpected since calculations have suggested similar thermodynamic stabilities for the all-*E*-(6′*R*)- and all-*E*-(6′*S*)-fucoxanthin diastereomers.[12] However, the mechanism suggested in Scheme 2 for diphenyl diselenide mediated isomerization may serve to rationalize the high yield of the (6′*S*)-allene obtained with this mediator.

Mechanistic aspects

Photoisomerization in the absence of catalyst. Photoisomerization in the absence of a catalyst is most effective when the emission spectrum of the irradiation source overlaps with the main absorption band of the compound.[13,14] Absorption of a photon induces a transition from the singlet ground-state (S_0) to the lowest singlet excited state (S_1). However, results have been published for β,β-carotene which suggested that no geometrical isomerization takes place in the S_1 state.[15] On the other hand, it has also been inferred that photoisomerization of polyenes by direct irradiation does not involve excited triplet states.[16] The electronic configuration involved in photoisomerizations promoted by a triplet sensitizer is better understood than that involved for isomerizations in the absence of a sensitizer. Either the *Z* or the *E* compound may be formed.[17] Triplet-sensitized isomerizations carried out with various mono-*Z*-isomers of β,β-carotene revealed that the major product was all-*E*-β,β-carotene, independent of the starting isomer.[18,19]

In Tables 5 and 6 are presented results demonstrating that geometrical isomerization of peridinin (**1**, **2**) and fucoxanthin (**3**, **4**) occurred in sunlight in the absence of a catalyst, whereas allenic isomerization occurred only to a very low extent. Apparently the activation energy for *R*/*S* allenic isomerization in carotenoids is higher than the activation energy associated with geometrical isomerization.

Diphenyl diselenide mediated photoisomerization. The present results, with increased reaction rate at UVA radiation and response towards a radical quencher, are taken

Table 5. Isomeric mixture of peridinin (**1, 2**) obtained upon photoisomerization of all-*trans*-(6′*R*)-peridinin (**1**) or all-*trans*-(6′*S*)-peridinin (**2**) in May sunlight in the absence of catalyst.

		% Total peridinin (**1, 2**)	
Optical isomer	Geometrical isomer	all-*trans*-(6′*R*)[a] (**1**), 3 h	all-*trans*-(6′*S*)[a] (**2**), 2.5 h
6′*R*	all-*trans* (**1**)	64	9
	9′-*cis* (**1a**)	20	
	11-*cis* (**1b**)	13	
	13-*cis* (**1c**)	2	
6′*S*	all-*trans* (**2**)	2	56
	9′-*cis* (**2a**)		17
	11-*cis* (**2b**)		10
	13-*cis* (**2c**)		8

[a] Initial isomer.

Table 6. Isomeric mixture of fucoxanthin (**3**, **4**) obtained upon photoisomerization of all-*E*-(6′*R*)-fucoxanthin (**3**) or all-*E*-(6′*S*)-fucoxanthin (**4**) in October sunlight in the absence of catalyst.

		% Total fucoxanthin (**3**, **4**)	
Optical isomer	Geometrical isomer	all-*E*-(6′*R*)[a] (**3**), 2.5 h	all-*E*-(6′*S*)[a] (**4**), 3 h
6′*R*	di-*Z* isomers	2	
	all-*trans* (**3**)	74	1
	13-*Z*	3	
	13′-*Z*	5	
	9′-*Z*	16	
6′*S*	all-*trans* (**4**)		71
	13-*Z*		3
	13′-*Z*		5
	9′-*Z*		20

[a] Initial isomer.

as support for a radical mechanism for allenic *R*/*S* isomerization. The selenide radical may in principle attack the allenic sp carbon from the top (β) or bottom (α) face, cf. Scheme 2. Attack from the β-face is to some extent hindered by the axial hydrogens at C-2 and C-4. Attack from the α-face promotes the formation of the (6*Z*)-bisallylic radical **a**.

The observation that the (6*S*)-allene is the major isomer upon selenide promoted isomerization suggests a pronounced thermodynamic preference for one conformer of the radical intermediate. Radical **a** may undergo a ring-flip to give radical **b**, which may be stabilized by an intramolecular hydrogen bond between selenium and the equatorial hydoxy group at C-5. Rotation around the C-7,8 carbon–carbon single bond may relieve the steric interaction between the geminal methyl groups at C-1 and the bulky polyene chain, giving rise to radical **c**. Elimination of the selenide radical then provides the (6*S*)-allene, ultimately with the preferred conformation of the cyclohexane ring.

Alternative attack of the selenide radical from the β-face of the (6*R*)-allene promotes the formation of a (6*E*)-bisallylic radical, in which no stabilizing intramolecular hydrogen bond may be formed to selenide. In this case steric preference for the radical rotamer giving rise to the (6*R*)-allene was observed in a model study. Consequently, it appears likely that the diphenyl diselenide promoted isomerization of allenic carotenoids proceeds via α-attack of the selenide radical, as discussed above and illustrated in Scheme 2.

The *E*/*Z* stereomutation of sterically unhindered double bonds would proceed by a radical mechanism, via attack at an sp^2 carbon, possibly with formation of a tertiary radical. Mild conditions promoting *E*/*Z* stereomutation, but no allenic isomerization, have recently been reported for fucoxanthin (**3**)[1] and are compiled in Table 4 for peridinin (**1**).

Iodine-catalyzed photoisomerization. No generally accepted mechanism has so far been published for the photo-induced iodine-catalyzed stereomutation of polyenes. It has been suggested that addition of iodine to a carbon–carbon double bond to generate a free polyene radical is a key step in the isomerization.[20,21] The main factors influencing iodine-catalyzed stereomutation of carotenoids include the concentration of pigment and of iodine, the absolute quantity of iodine, the pigment : iodine ratio, the light intensity employed, the exposure time[14] and the solvent.[22,23] It has been claimed that the most effective irradiation wavelengths correspond to light absorbed by iodine, 450–600 nm in hexane, and not by the carotenoid (350–500 nm).[22] The formation of a charge transfer complex between β,β-carotene and iodine has been reported.[24] The charge transfer complex exhibited a large bathochromic shift of the main absorption maximum, compared with pure β,β-carotene, from ca. 450 nm to ca. 1000 nm, compatible with the generation of an electron-delocalized carotenoid cation.[25–27] The complex was formulated as $C_{40}H_{56}\cdots I^+$, which was suggested to be the predominant form in the electronic ground state, while in the excited state the positive charge is located at the carotenoid $C_{40}H_{56}^+\cdots I$. It has been postulated that the latter form may be the species involved in iodine-catalyzed stereomutation.[24,28]

In the present work iodine-catalyzed allenic isomerization of peridinin (**1**) occurred to a small extent over longer periods in darkness at high iodine concentrations, cf. Table 7. However, considerable *E*/*Z* isomerization was noted.

At high light intensity the allenic *R*/*S* isomerization rate increased upon increasing I_2 concentration from 5 mol% to 50 mol% (Fig. 6). Moreover, UVA radiation had no beneficial effect (Fig. 7). At 10 equivalents of

Table 7. Iodine-catalyzed isomerization of all-*trans*-(6′*R*)-peridinin (**1**) in benzene solution in darkness at room temperature.

		% Total peridinin (**1**, **2**)					
Mol% I_2	Time	all-*trans*-**2**	Unknown	all-*trans*-**1**	9′-*cis*-**1**	11-*cis*-**1**	13-*cis*-**1**
5	20 h	0	0	88	6	5	1
5	48 h	0	0	69	24	5	1
5	13 days	0	0	65	23	9	2
5	2 months	0	1	63	24	9	3
50	2 months	2	1	61	26	8	3
525	48 h	8	0	55	28	7	3

hydroquinone relative to iodine (5 mol%) no effect of this radical quencher on the allenic isomerization was observed (Fig. 9). However, the result when employing 400 molar equivalents of hydroquinone does not exclude a radical mechanism.

The mechanism of iodine-catalyzed photoisomerization of carotenoids remains to be established.

Experimental

Carotenoid substrates. All-*trans*-(3*S*,5*R*,6*S*,3′*S*,5′*R*,6′*R*)-peridinin (**1**) was isolated from stored extracts of dinoflagellates by employing previous procedures.[29] VIS, MS and ^{1}H NMR spectra were consistent with reported data.[6,10] HPLC-pure all-*trans* **1** was used for experiments. The all-*trans*-(6′*S*)-isomer (**2**) was prepared from **1** and isolated by preparative HPLC.

All-*E*-(3*S*,5*R*,6*S*,3′*S*,5′*R*,6′*R*)-fucoxanthin (**3**) and the (6′*S*)-isomer (**4**) were isolated and characterized as previously described.[8,29]

Isomerization experiments. The equipment used has recently been specified.[1] The 1000 W tungsten lamp (Osram 64741 CY4) was used as a source of visible light. For producing visible light with reduced UV contribution a double layer of Plexi-glass (Röhm GMBH, type GS, Farblos 231, 3 mm thickness) was employed as filter between the tungsten lamp and the test tube, shielded for indirect irradiation.

Two vertical UVA fluorescent tubes (Osram L 18 W/73) were employed for UVA (maximum emission at 360 nm) irradiation. Two test tubes were placed one above the other, between and at $\approx$1 cm distance from the UVA tubes.

The intensity of visible light was measured by use of a quantum meter with a spherical sensor[1] and that of UVA with a Solar Light Co. Inc. Erythema UV and UVA intensity meter, model 3D with a flat sensor, cf. Table 1. The spectral sensitivity of the UVA sensor overlaps with the emission spectrum of the UVA tubes. Irradiances were measured at the sample position for all experiments.

The experiments without catalyst were carried out in direct sunlight (May or October) at 63 °N during the period 10 am to 3 pm.

All experiments were performed in *p.a.* benzene solution with a carotenoid concentration of 40 μg ml^{-1}. The amount of catalyst relative to the carotenoid, is given in each case. Two weight% Ph_2Se_2 corresponds to $\approx$4 mol%, and 2 weight% I_2 to $\approx$5 mol%. Procedures for maintaining an inert atmosphere are described.[1] The isomerization was monitored by HPLC and pigment recoveries calculated at the end of each experiment using the same $E^{1\%}_{1\,\mathrm{cm}}$ at $\lambda_{\max}$ as for the all-*trans* isomer.

The experiments with a radical quencher were performed with 0.11 mg **1** and 0.11 mg (200 mol%) Ph_2Se_2 in 1.7 ml benzene plus 0.4 mg or 16 mg hydroquinone (corresponding to 10 and 400 mol equivalents) in 1 ml diethyl ether. Two other experiments were carried out using 0.11 mg **1** in 1.7 ml benzene containing 2.2 μg (5 mol%) I_2 plus 0.01 mg or 0.4 mg hydroquinone (10 and 400 mol equivalents relative to I_2) in 1 ml diethyl ether. Diethyl ether was distilled from sodium–benzophenone.

Analysis. Procedures and equipment have been outlined.[1] Preparative HPLC (Hewlett Packard 1050 pump, Hewlett Packard 1040A detector, Hewlett Packard 79994A HPLC Chemstation dataprogram) was used for the isolation of all-*trans*-(6′*R*)-peridinin (**1**) and all-*trans*-(6′*S*)-peridinin (**2**). HPLC system A[4–6] Techsphere 5CN, 4.6 × 250 mm, eluent hexane–acetone–methanol (91 : 8 : 1), flow 4 ml min^{-1}, detection wavelength 445 nm; HPLC system B as above except column 10 × 250 mm, eluent hexane–acetone–methanol (93 : 6 : 1), flow 1.5 ml min^{-1}.

Retention times and VIS data ($\lambda_{\max}$, %D_B/D_{II},[30] %III/II[30]) of individual stereoisomers in comparison with published data for previously identified geometrical isomers of peridinin (**1**, **2**)[6] and fucoxanthin (**3**, **4**)[8,9] were used for identification.

Acknowledgements. A. Strand was supported by a research grant from Hoffmann–La Roche, Basel to S. L.-J. and J. A. Haugan was supported by a postdoctoral fellowship from The Norwegian Research Council.

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Received June 18, 1998.