Optical Resolution of Rotenoids

S. L. Abidi

U. S. Fish and Wildlife Service, National Fishery Research Laboratory
P. O. Box 818, La Crosse, Wisconsin 54602-0818

Received December 1, 1986

Optical resolution of selected rotenoids containing 1-3 asymmetric centers in dihydrobenzopyranofurobenzopyranone and dihydrobisbenzopyranopyranone series has been achieved on two chiral high-performance liquid chromatographic (hplc) stationary phases. In most cases, the absolute stereochemistry at the *cis*-B/C ring junction of the rotenoidal antipodes can be related to their elution order. Generally, the 6aα,12aα-enantiomers were more strongly retained by the chiral substrate than their corresponding optical antipodes. The elution-configuration relationship provides potential utility for predicting the absolute configuration of related rotenoidal compounds. Chiral phase hplc on amino-acid-bonded-silica yielded results explicable in terms of Pirkle's bonding schemes for chiral recognition. Resolution data for 12a-hydroxy-, 12a-methoxy-, and 12-hydroxyiminorotenoids further corroborate the mechanistic rationale, and demonstrate that nonpolar π-π interactions appeared to be important for enantiomeric separation on helic poly-triphenylmethylacrylate-silica (CPOT). In the latter system, steric effects and conformational factors in association with the modification of E-ring structures might play significant roles in the chiral separation process in view of the reversal to the elution order observed for all methoxylated rotenoids and elliptone derivatives including the parent deguelin. The unique separability (α = 1.44) of 12a-hydroxyelliptone on CPOT was suggestive of structural effects of the 5′-side chain on the resolution of the rotenoids having a five-membered-E-ring. The results obtained with two different types of chiral phases are complementary and useful for optical resolution of a wide variety of natural and synthetic rotenoidal compounds.

J. Heterocyclic Chem., **24**, 845 (1987).

Introduction.

Rotenoids derived from derris roots have been used as agricultural insecticides and have remained attractive heterocyclic ring systems for stereochemical and synthetic investigations. (-)-Rotenone (1A, Figure 1), which is also a widely used fish toxicant, can be regarded as a structural prototype for the various rotenoidal compounds isolated from the derris plants. As part of our ongoing programs directed toward evaluation of degradation kinetics and fates of rotenone in the environment, it was desirable to obtain optical isomers of a series of rotenoids for studying their biological activity and for identifying optically active components among a host of degradation products. Structure-biological activity relationships of some rotenone

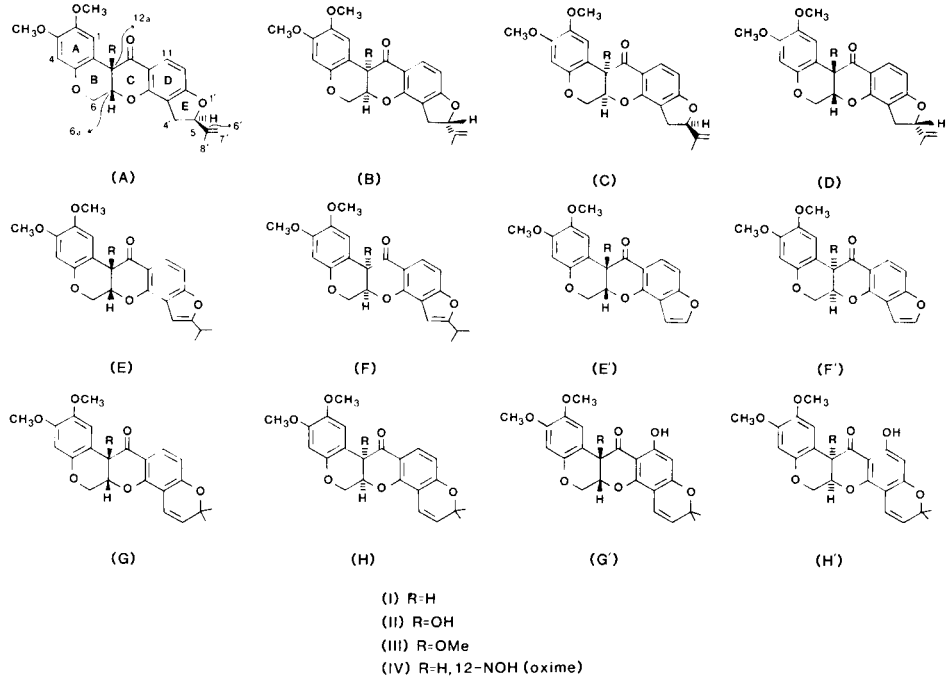

Figure 1. Structures of enantiomeric rotenoids: type (i), rotenone series (**A-D**); type (ii), isorotenone series (**E-F'**); type (iii), deguelin series (**G-H'**).

compounds of agricultural and pharmaceutical interest have been studied previously [1-3]. These studies showed that optical isomers of rotenoidal compounds exhibited different degrees of biological activities. Generally, the *cis*-6aβ, 12aβ-compounds possess higher potency of the inhibition of mitochondrial respiratory activity than the corresponding enantiomers (*cis*-6aα, 12aα-isomers), or other diastereomers with trans B/C ring junction. Most recently, we reported the high-efficiency high-performance liquid chromatographyic (hplc) separation [4] and spectral characterization [5] of epimeric rotenone and related tetrahydrobenzopyranofurobenzopyranones. The essence of these earlier studies signifies notable stereochemical effects of the B/C ring junction on the chromatographic behavior as well as on the spectral characteristics of epimeric rotenone compounds. Similar influence of the B/C ring junction stereochemistry on the optical resolution of rotenone and related rotenoids on chiral stationary phases in hplc systems would be anticipated.

Optical resolution of synthetic unnatural antipodes of rotenone isomers and natural racemates of rotenoids has not been described in the literature. In a recent publication [6], Unai *et al.* reported that successive treatment of natural rotenone (**IA**) with one mole equivalent of boron tribromide and aqueous sodium bicarbonate provided a unique synthetic route to the 5′*S*-epimer (**IC**). This is the only method available to date for obtaining other 5′*S*-rotenoids. Thus, optical isomers of rotenone have been exclusively prepared by these chemical transformation methods. Because of the lack of suitable functionalities for facile derivation, attempted resolution of racemic rotenone-like compounds has suffered from methodological limitations. Hence, rotenoids in general are difficult to form diastereomeric mixtures with chiral auxiliaries for separation by conventional fractional recrystallization. With the advent of modern separation technologies, direct resolution of rotenoids can now be realized by chromatographic means through the use of chiral stationary phases (CSP's). In consideration of the thermal instability of rotenoids, hplc techniques for separation of optical isomers should be preferred to the gc and other chromatographic methods. The relatively low reactivity of rotenoids toward auxiliary chiral reagents as observed in solution chemistry should preclude the possibility of using a chiral mobile phase in a hplc system for optical resolution. Therefore, a chiral phase hplc technique with an achiral mobile phase should be ideally suited for optical resolution of rotenoids.

In connection with another study on enantiomeric separation of cyclopropanes [7], we were particularly interested in the two fundamentally different types of CSP's: (i) an amino acid chiral phase (PCOV) (Pirkle's covalently bonded phase) developed by Pirkle *et al* [8,9], and (ii) and optically active poly(triphenylmethylmethacrylate) stationary phase (CPOT) [(Chiralpack-(+)-OT)] developed by Okamoto *et al* [10-12]. These chiral packings have been useful in the direct resolution of a variety of chiral compounds most of which contain aromatic groups. We have evaluated the resolution potential of these two CSP's for different types of rotenoids (Figure 1). The relation between absolute stereochemistry and elution order of these rotenoids has been examined in this study. The separability of optical antipodes is discussed in terms of conformational effects of the heterocyclic ring system, B/C ring junction stereochemistry and other structural variables. The approaches used in the present study should be not only invaluable in the isolation of rotenoidal enantiomers for structure-biological studies but also of methodological importance in the total synthesis of chiral rotenoidal compounds from achiral precursors.

Results and Discussion.

Because of the scarcity of available rotenoids, the compounds chosen for study are confined to the three general structural types (Figures 1 and 2): (i) rotenone type, (ii) isorotenone type, and (iii) deguelin type. In rotenone series, there are three asymmetric centers at 6a-, 12a-, and 5′-carbons in each optical isomer (Figure 1, A, B, C, and D). For compounds containing two asymmetric centers as in both the isorotenone (Figure 1, E-F′) and deguelin (Figure 1, G-H′) series, the chiral centers are situated at the B/C ring junction (6a- and 12a-carbons) while the E-ring is devoid of any asymmetric carbons. In addition to the three types of rotenoids depicted in Figure 1, we included in this study three enantiomeric pairs of related rotenoids (Figure 2) that contain one asymmetric center in each of them. As shown in Figure 2, these racemic mixtures represent the 6a,12a-, 12a,12-, and 12a,12-dehydro-analogues of rotenone (**I**), isorotenone (**J**), and deguelin (**K**), respectively. The molecular chirality of an optical isomer in this instance is obviously attributed to the only asymmetric carbon at 5′-position for **I**, and at 6a-position for both **J** and **K**.

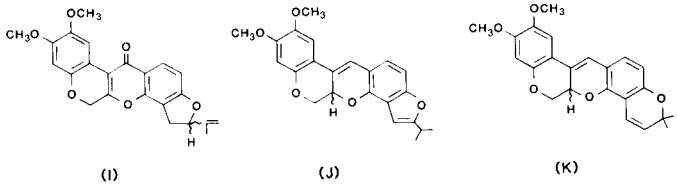

Figure 2. Structures of the dehydro-analogues of rotenone (**I**), isorotenone (**J**), and deguelin (**K**).

With the exception of (-)-6a*S*,12a*S*,5′*R*-rotenone (**AI**) which exists in nature as a pure optical isomer, racemates of rotenoids in other series under study are commonly found in the rotenoid extracts of the natural plants and

often coexist with pure enantiomers in variable proportions. Using **AI** as the starting material, enantiomeric rotenone **BI**, **CI**, and **DI** were obtained by treatment of **AI** with either anhydrous sodium acetate in absolute ethanol (epimerization at the B/C ring junction) or boron tribromide (epimerization at the 5′-carbon), or by sequential treatment of **AI** with both reagents [6,13]. The structural relationships of the two sets of optical antipodes **AI-BI** and **CI-DI** are illustrated in Figure 1. Isorotenone **EI** was prepared from natural rotenone **AI** by isomerization of the 6′,7′-double bond of **AI** [13,14]. Racemization of **EI** [or elliptone, **E′** (the desisopropyl-analogue of isorotenone)] in the presence of potassium carbonate and acetone led to a mixture consisting of **EI** (or **E′**) and **FI** (or **F′**) [15], the components of which were separated and isolated by preparative hplc on a chiral stationary phase [16]. In a similar manner, a racemate of the naturally occurring deguelin **(GI-HI)** was resolved, and pure **GI** and **FI** were isolated by the chiral phase hplc technique. Mild dehydrogenation of **AI** (or **CI**) afforded 5′*R*-dehydrorotenone (5′*R*-**I**) (Figure 2) in quantitative yield [6]. Its antipode, 5′*S*-**I**, was similarly obtained from **BI** (or **DI**). For preparation of racemates of 12a,12-dehydroisorotenone (**J**) and of 12a,12-dehydrodequelin (**K**), respective racemic mixtures of **EI-FI** and **GI-HI** were reduced with potassium borohydride followed by dehydration with sulfuric acid yielding **J** and **K** [17] (Figure 2). Optical resolution of the racemates by preparative chiral phase hplc provided pure enantiomers 6a*S*-**J**, 6a*R*-**J**, 6a*S*-**K**, and 6a*R*-**K**. The pure individual optical isomers (**A-K**, Figure 1 and Figure 2) obtained in the above fashion were then subjected to structural verification by high resolution mass spectrometry and optical rotation measurements. To gain further insight into the chiral recognition mechanisms for enantiomeric separation of rotenoids on the two types of CPS's (PCOV and CPOT) studied, the asymmetric carbon 12a of compounds **A-H′** was functionalized either with a hydroxy- or a methoxy-group; and the 12-carbonyl was derivatized to an oxime as shown in Figure 1. These reactions proceeded with retention of configurations at the chiral centers [13,17].

Results of optical resolution of type (i) rotenoid in rotenone series are summarized in Table I. The retention (k′) and separability (α) data for the compounds in this series clearly demonstrate that the success of enantiomeric separation depended heavily upon the nature of CPS's employed. Thus, with the PCOV phase, resolution of the hydroxy-containing compounds represented by rotenolones (**AII-BII** and **CII-DII**) and rotenone oximes (**AIV-BIV** and **CIV-DIV**) produced well-resolved components of an-

Table I

Optical Resolution of Type (i) Rotenoids in Rotenone Series on Chiral Stationary Phases [a]

Compound	Chiral stationary phase [b]					
	PCOV			CPOT		
	k′	α	(6a,12a,5′) [a]	k′	α	(6a,12a,5′) [a]
Type (i) Rotenone series:						
Rotenone						
AI	8.24			2.70		(*S,S,R*)
BI	8.24	1.00		5.90	2.19	
CI	8.24			3.38		
DI	8.24	1.00		2.70	1.25	(*S,S,S*)
Rotenolone						
AII	8.94		(*R,R,R*)	1.50		
BII	11.2	1.25		1.50	1.00	
CII	11.2			1.50		
DII	8.94	1.25	(*R,R,S*)	1.50	1.00	
12a-Methoxyrotenone						
AIII	3.82			3.06		
BIII	3.82	1.00		2.92	1.05	(*S,S,S*)
CIII	3.82			3.06		(*S,S,R*)
DIII	3.82	1.00		4.32	1.41	
Rotenone oxime						
AIV	11.1		(*S,S,R*)	1.76		(*S,S,R*)
BIV	18.7	1.68		2.44	1.39	
CIV	18.4			2.16		
DIV	11.1	1.64	(*S,S,S*)	1.76	1.23	(*S,S,S*)

[a] (6a,12a,5′) = The asymmetric carbon positions of the less retained enantiomer. [b] PCOV = Pirkle's covalently bonded silica phase; CPOT = Chiralpack-(+)-OT polymer phase.

tipodal pairs each of which exhibited respective separation factors (α) of 1.25 and 1.68. Under the same chromatographic conditions, there was no discernible resolution of enantiomeric mixtures of rotenones (**AI-BI** and **CI-DI**) and methoxyrotenones (**AIII-BIII** and **CIII-DIII**). The α values were equal to unity in these cases. On the other hand, separation of enantiomers in the same series on the CPOT stationary phase yielded results which were in contrast with those observed with the PCOV phase described above. In chiral phase separation on CPOT, the optical antipodes of rotenones, and of the corresponding 12a-methoxy- and oxime-derivatives were resolved with the α values varying from 1.05 to 2.19; whereas the more polar hydroxy-containing rotenolones were not at all resolved (α = 1.00) under identical experimental conditions (Table I).

Similar results were obtained from optical resolution of type (ii) rotenoids in the isorotenone series (Table II). We failed to resolve the enantiomers of isorotenones (**EI-FI**) and methoxyisorotenones (**EIII-FIII**) by hplc on PCOV (α = 1.00). Enantiomers of isorotenolones (**EII-FII**) were found to be indistinguishable by differential chiral interactions with CPOT. However, optical resolution of the antipodal pairs **EII-FII** and **EIV-FIV** was accomplished with the PCOV phase to give respective separation factors 1.21 and 1.50; while the racemates **EI-FI**, **EIII-FIII** and **EIV-FIV** were resolved on CPOT yielding α values in the range of 1.24-1.42. For type (iii) rotenoids in the deguelin series, the results (Table III) were somewhat different from those observed in the other two series described. Resolution of both the racemic tephrosins **GII-HII** (α = 1.20) and deguelin oximes **GIV-HIV** (α = 1.58) was achieved with the PCOV chiral phase despite its inability to differentiate the enantiomers of deguelins **GI-HI** and methoxydeguelins **GIII-HIII** (α = 1.00). Nevertheless, using the COPT phase in the hplc system, we were able to resolve the racemates **GI-HI** (α = 1.90), **GII-HII** (α = 1.63) and **GIV-HIV** (α = 1.88). Surprisingly, the methoxy compounds **GIII-HIII** were not resolved. The abnormally high degree of optical resolution achieved for the tephrosin racemate **GII-HII** and the total loss of enantiomeric differentiability for the methoxydeguelin pair **GIII-HIII** contradict our general observation of a plausible correlation between molecular polarity at the B/C ring junction and chiral phase characteristics.

Table II

Optical Resolution of Type (ii) Rotenoids in Isorotenone Series on Chiral Stationary Phases

Compound	Chiral stationary phase [b]					
	PCOV			CPOT		
	k'	α	(6a,12a) [a]	k'	α	(6a,12a) [a]
Type (ii) Isorotenone series:						
Isorotenone						
EI	6.29			3.14		(*S,S*)
FI	6.29	1.00		3.88	1.24	
Isorotenolone						
EII	6.41		(*R,R*)	1.58		
FII	7.73	1.21		1.58	1.00	
12a-Methoxyisorotenone						
EIII	2.61			4.06		
FIII	2.61	1.00		2.92	1.39	(*S,S*)
Isorotenone oxime						
EIV	8.45		(*S,S*)	2.31		(*S,S*)
FIV	12.7	1.50		3.29	1.42	
Elliptone						
E'I	9.84			2.92		(*S,S*)
F'I	9.84	1.00		3.34	1.14	
12a-Hydroxyelliptone						
E'II	10.2		(*R,R*)	1.78		
F'II	11.8	1.15		1.24	1.44	(*S,S*)
12a-Methoxyelliptone						
E'III	4.42			4.40		
F'III	4.42	1.00		2.48	1.77	(*S,S*)
Elliptone oxime						
E'IV	14.2		(*S,S*)	1.98		
F'IV	19.1	1.35		1.78	1.11	(*R,R*)

[a] (6a,12a) = The asymmetric carbon positions of the less retained enantiomer. [b] For PCOV and CPOT abbreviations, see footnote to Table I.

Table III

Optical Resolution of Type (iii) Rotenoids in Deguelin Series and The Dehydro-analogues of Investigated Rotenoids on Chiral Stationary Phases

Compound	Chiral stationary phase [b]					
	PCOV			CPOT		
	k'	α	Isomer [a]	k'	α	Isomer [a]
Type (iii) Deguelin series:						
Deguelin						
GI	6.83			5.01		
HI	6.83	1.00		2.64	1.90	(*R,R*)
Tephrosin						
GII	7.07		(*R,R*)	1.44		(*R,R*)
HII	8.46	1.20		2.34	1.63	
(α)-Toxicarol						
G'II	6.80			3.84		
H'II	6.80	1.00		2.94	1.31	(*R,R*)
12a-Methoxydeguelin						
GIII	3.10			3.16		
HIII	3.10	1.00		3.16	1.00	
11-Methoxydeguelin						
G'III	5.83			4.80		
H'III	5.83	1.00		2.98	1.61	(*R,R*)
Deguelin oxime						
GIV	15.5			4.31		
HIV	9.84	1.58	(*R,R*)	2.29	1.88	(*R,R*)
Dehydro-analogues:						
6a,12a-Dehydrorotenone						
5β-**I**	18.4			2.86		(*R*)
5α-**I**	18.4	1.00		4.52	1.58	
12a,12-Dehydroisorotenone						
6aβ-**J**	4.06			3.19		
6aα-**J**	4.06	1.00		2.75	1.16	(*R*)
12a,12-Dehydrodeguelin						
6aβ-**K**	3.91			2.56		(*S*)
6aα-**K**	3.91	1.00		3.41	1.32	

[a] Isomer = the less retained enantiomer with asymmetric centers shown. [b] For PCOV and CPOT abbreviations, see footnote to Table I.

As summarized in Table III, hplc separation of the dehydro-analogues of the investigated rotenoids on PCOV led to no resolution of the optical isomers in all cases. On the contrary, comparatively high degrees of resolution were attained when racemates of the dehydro-compounds were subjected to chiral phase hplc on CPOT. The separation factors (α) of the antipodal pairs were 1.58 for (5β-**I**)-(5α-**I**), 1.16 for (6aβ-**J**)-(6aα-**J**), and 1.32 for (6aβ-**K**)-(6aα-**K**). The greater magnitude of the α value for **I** may indicate enhanced π-π interactions contributed by the extra conjugated 12-carbonyl.

The chromatographic behavior of the investigated rotenoids on the amino acid derived PCOV phase paralells that of a diversity of alcohols reported by Pirkle *et al* [9]. Consistent with Pirkle's hypothetical model of three point interactions for chiral recognition are our observations of successful separations of the hydroxy-containing enantiomers as in groups **II** and **IV** rotenoids in which the required structural moieties serving as π-donors, and hydrogen-acceptors and donors are present. Of the required elements for specific interactions, the polar hydrogen-bonding interactions appeared to be most important. In consideration of the polar nature of the PCOV chiral phase, it is logical to envisage the chiral recognition mechanism proceeding *via* the predominent polar interactions involving hydrogen acceptor-donor complexation between the rotenoidal solutes and the chiral substrates. Because of the apparent absence of such polar interactions essential for chiral recognition in this system, no separation of optical isomers was observed in all the cases where the rotenoids are void of the hydroxy functionality as in groups **I** and **III** rotenoids. An exceptional circumstance was encountered in the hplc of toxicarols (**G'II**-**H'II**) where the 11-hydroxy group is at least three-carbon

remote from the chiral centers at the B/C ring fusion, all attempts to reslove this racemate on PCOV were unsuccessful (α = 1.00) (Table III). The result suggests that the structural prerequisite for hydroxy-rotenoids is the position of the OH-group to be directly attached to or in the proximity of the chiral center. Comparisons of the separability data (α values) (Table I-III) for the 12a-hydroxy compounds (group **II** rotenoids) with those for the ketoximes (group **IV** rotenoids) show that the greater magnitude of α values for the latter compounds in group **IV** rotenoids is indicative of a higher degree of differential multiple polar interactions presumably entailing the 12-hydroxyimino-group in the chiral complexation process.

The results from optical resolution of the majority of investigated rotenoids with the CPOT chiral phase are complementary to those found in the PCOV systems. The rotenoidal enantiomers that failed to separate on PCOV were resolvable on CPOT exhibiting α values between 1.00 and 2.19 (Table I-III). It is worthy of mention that the chiral phase hplc of rotenoids on CPOT is the first example of successful direct resolution of group **I** rotenoids of pesticidal interest. With a few exceptions as in the cases of 12a-hydroxylated elliptone (**E'II-F'II**) and deguelin (**GII-HII**), optical resolution on the CPOT phase seemed to be in favor of the rotenoids devoid of 12a-hydroxy groups. The experimental data for the dehydro-analogues (Table III) are also illustrative of the same trend of structural preference. In light of the high degree of aromaticity inherent in the CPOT chiral phase and the chromatographic outcome of enantiomeric separation, it was highly likely that the optical antipodes of rotenoids were distinguished by chiral recognition through differential non-polar π-π and other van der Waals interactions between the enantiomeric analytes and the helical CPOT substrates [10].

The hydrogen bonding participation in the chiral recognition process may not be important. The chromatographic behavior of type (iii) rotenoids in the deguelin series appeared to deviate somewhat from the prototypical characteristics noted in the other two series. While the α value for the deguelin pair was 1.90 as expected, the respective α values for the hydroxylated-(**GII-HII**) and methoxylated-(**GIII-HIII**) compounds were 1.63 and 1.00 (Table III). A reversal of the latter values would otherwise be expected on the basis of the simple non-polar interaction rationale for chiral recognition. Without any additional experimental evidence, we speculate that a steric factor probably of conformational origin in the heterocyclic rotenoidal ring system may play an important role in the enantiomeric differentiation mechanism. The inclusion of an OH (or OCH_3) to the deguelin structure may presumably result in destabilization of the nonpolar interactions of the helical polymer chain of CPOT with the enantiomers derived from deguelin. With regard to the resolution of the methoxylated compounds (group **III** rotenoids) on CPOT, an interesting feature was noteworthy. Without exception, a reverse elution order with respect to the parent rotenoids was seen in every antipodal pair of the methoxylated rotenoids examined.

As demonstrated throughout this study, optical resolution of rotenoids tended to be governed, to a large extent, by the stereochemistry of the asymmetric centers at the B/C ring junction and, to a much lesser extent, by the chiral center at the 5' carbon (applicable only for compounds in rotenone series) (Table I). Although diastereomeric pairs **BI-CI**, **BII-CII**, **BIII-CIII**, and **BIV-CIV** were separated on CPOT, the diastereomers having the same ring junction stereochemistry (as in A-D pairs) were generally not resolved on either CSP. None of the B-C pairs was separable on PCOV (Table I). The apparent lack of differentiability in these instances may be associated with the low molecular dissymmetry inherent in the diastereomers that bear an epimeric relationship at the 5'-asymmetric center. Cursory survey of the data in Table I-III indicates that in experiments with PCOV, variations in E-ring structures had a relatively small effect on the separability (α) of enantiomers. However, in chiral phase hplc on CPOT, it was found that the magnitude of α values was dramatically influenced by the change in the structure of the E-ring in going from five membered- (type i and type ii rotenoids) to the six membered-oxygen-heterocyclic ring (type iii rotenoids).

Close scrutiny of the chromatographic elution data in Table I-III shows that, in most cases, a useful relationship between the elution order and the configuration [6,18-20] at the B/C ring junction exists for rotenoids in the same groups (or series). Thus, all the enantiomers in group **II** rotenoids (**AII, DII, EII, E'II** and **GII**) that were less retained by PCOV had 6a*R* and 12a*R* chiral centers as shown in Figure 1; whereas, except for **HIV**, the less retained components in group **IV** rotenoids on the same chiral phase were the 6a*S*,12a*S*-enantiomers that had the same configurations at the 6a and 12a chiral centers as those in group **II** rotenoids just described. On the other hand, the early eluting isomers from the CPOT phase were the 6a*S*,12a*S*-enantiomers for all of the type (i) (rotenone series) and type (ii) (isorotenone series) rotenoids with **F'IV** as an exception. For type (iii) rotenoids in deguelin series, the less retained rotenoidal components were the 6a*R*,12a*R*-enantiomers. In this regard, it should be pointed out that the configurations of the early eluting enantiomers of all methoxy-rotenoids and elliptone derivatives including two deguelin compounds **HI** and **HIV** are not the same as that of **IA** and other 6aβ,12aβ-rotenoids as shown in Figure 1. Examination of the hplc data for the dehydro-analogues (Table III) indicates that both (5*R*)-**I** (dehydrorotenone) and (6a*R*)-**J** (de-

hydroisorotenone) were less retained by CPOT. The early eluting component of the dehydrodeguelin 6a-**K** was the 6a*S*-enantiomer. Based on the generality of the elution order-configuration relationship, it became apparent that the fusion of a six-membered pyranyl ring (E-ring) onto the dihydrobenzopyranobenzopyranone (A-B-C-D-ring) system seemed to alter the stereochemical requirements for specific chiral interactions with chiral phases. Inspection of the Dreiding molecular models revealed that appreciable changes in conformational preference were possible for the pentacyclic rotenoidal structures as result of the variation in the size of the E-ring. As seen from the models, there are several conformations of the flexible *cis*-B/C fusion in a rotenoidal structure: the B ring can be a quasi-boat or quasi-chair. It is not obvious how such changes determine the elution order in the system under consideration.

To demonstrate the effect of 5′-side chain structures on resolution of type (i) and (ii) rotenoids, the chromatographic data for elliptone and its derivatives (**E′I-IV** and **F′I-IV**) are incorporated in Table II. While hplc on PCOV displayed strikingly similar patterns of enantiomeric separation as those exhibited by other rotenoids, optical antipodes of all four elliptone compounds were unexpectedly resolved on CPOT with α values in the range 1.11-1.77. In the unique case of 12a-hydroxylated elliptones, the nonpolar π-π interactions appeared to be considerably more important than those in the corresponding 5′-substituted-analogues **AII**, **BII**, **CII**, **DII**, **EII**, and **FII** where such interactions might be hampered by the presence of the 5′-substituents. Also, it is highly likely that the presence of the 5′-isopropyl (or isopropenyl) on the side chain may change the conformation of the rotenoidal heterocyclic ring system to accomodate the chiral recognition requirements for interactions with the helic polymer chain of the CPOT phase. These explanations seem to be reasonable to account for the anomalous elution order observed for all three enantiomeric pairs **E′II-F′II**, **E′III-F′III**, and **E′IV-F′IV**. The exact nature of this substituent effect is not clear.

The complementary capability of the two CSP's do not limit the applicability of the methods in the event that only one of the CPS's is available. For resolution of rotenoids devoid of hydroxyl groups on PCOV, we converted the rotenoids into the corresponding oximes which were then subjected to chiral phase hplc on PCOV followed by mild hydrolysis [21] to afford the antipodal components of parent rotenoids. In another approach, we adopted a scheme of conversions analogous to published methods [22]. The rotenoids were hydroxylated to the group **II** rotenoids which were subsequently resolved on PCOV. The optical antipodes of the hydroxy-derivatives were trimethylsilylated prior to protiodesilylation for the regeneration of the optical antipodes of the parent rotenoids. The same scheme of transformations was equally applicable for the resolution of 12a-hydroxy-rotenoids on CPOT.

In conclusion, optical resolution of rotenoids containing 1-3 asymmetric centers can be accomplished by chiral phase hplc on either PCOV or CPOT. The elution-configuration relationship provides potential utility for predicting the absolute configuration of related rotenoidal compounds. These versatile methods are applicable to the resolution of synthetic racemates of rotenoids and related structures.

EXPERIMENTAL

Analytical and preparative hplc experiments were performed on a Varian Model 5020 liquid chromatograph. The full details of hplc procedures have been described [7,16]. The Pirkle's column (PCOV) (Pirkle's covalently bonded phase) derived from silica covalently bonded to (*R*)-N-3,5-dinitrobenzoylphenylglycine was obtained from Regis Chemicals (Morton Grove, IL). The Chiralpack-(+)-OT (CPOT) [(Chiralpack-(+)-OT)] column was purchased from Daicel Chemical Industries (New York, NY). The ^{13}C nmr spectra (22.5 MHz) [5] of the optically active rotenoids were superimposable with authentic racemates. Specific rotations were measured on a Rudolph Autopol III polarimeter. Mass spectral analyses of rotenoid enantiomers were performed on an AEI MS-902 high-resolution mass spectrometer (hrms). Elemental analyses were carried out on a Hewlett-Packard 185B CHN analyzer.

Rotenone (**AI**) was obtained from Aldrich Chemical Co. All other rotenoids were either obtained from various sources (see Acknowledgement) or prepared at our laboratory by known procedures (see Results and Discussion). Optically pure rotenoidal samples suitable for spectrometric confirmation were obtained by preparative hplc [7,16].

Since we used the same samples of **AI**, **CI**, **AII** and **CII** as those used in our most recent study [5], the melting points of these compounds are omitted here. The melting point data for other known compounds are recorded below (values in parentheses are the melting points reported in the literature): 6aα,12aα,5′α-rotenone (**BI**), mp 162-163° (162°) [6]; 6aβ,12aβ,5′α-rotenone (**DI**), mp 89-91° (88-95°) [6]; 6aβ,12aβ-isorotenone (**EI**), mp 175-177° (174-176°) [14]; 6aβ,12aβ-elliptone (**E′I**), mp 176-178 (178.5-179.5°) [23]; 6aβ,12aβ-isorotenolone (**EII**), mp 97-98° (96-97°) [17]; 6aα,12aα-isorotenolone (**FII**), mp 95-96° (96°) [17]; 6aβ,12aβ-deguelin (**GI**), mp 168-170° (172°) [15]; 6aβ,12aβ-tephrosin (**GII**), mp 197-199° (197-198°) [24]; 6aβ,12aβ-α-toxicarol (**G′II**), mp 126-128° (125-127°) [25]; 5′β-dehydrorotenone (5′β-**I**), mp 216-218° (217°) [6]; 5′α-dehydrorotenone (5′α-**I**), mp 216-219° (217°) [6]; 6aβ-dehydroisorotenone (6aβ-**J**), mp 178-179° (179°) [18]; 6aβ-dehydrodeguelin (6aβ-**K**), mp 173-176° (176°) [18].

The follwing rotenoids were isolated by preparative chiral phase hplc [7,16] from corresponding racemic mixtures of known structures.

6aα,12aα,5′α-Rotenolone (**BII**).

This compound exhibited ir and nmr spectra superimposable on those of known rotenolones **AII** and **CII**, mp 79-81°; hrms: m/e 410.4221 (Calcd. for $C_{23}H_{22}O_7$ m/e 410.4226).

Anal. Calcd. for $C_{23}H_{22}O_7$: C, 67.32; H, 7.97. Found: C, 67.28; H, 8.00.

6aβ,12aβ,5′α-Rotenolone (**DII**).

This compound exhibited ir and nmr spectra superimposable on those of known rotenolones **AII** and **CII**, mp 88-90°; hrms: m/e 410.4325 (Calcd. for $C_{23}H_{22}O_7$ m/e 410.4226.

Anal. Calcd. for $C_{23}H_{22}O_7$: C, 67.32; H, 7.97. Found: C, 67.25; H, 8.01.

6aα,12aα-Isorotenone (**FI**).

This compound exhibited ir and nmr spectra superimposable on those of its known optical antipode, mp 176°; hrms: m/e 394.4244 (Calcd. for

$C_{23}H_{22}O_6$ m/e 394.4232.

Anal. Calcd. for $C_{23}H_{22}O_6$: C, 70.05; H, 5.58. Found: C, 70.03; H, 5.55.

6aα,12aα-Elliptone (**F'I**).

This compound exhibited ir and nmr spectra superimposable on those of natural elliptone, mp 173-176; hrms: m/e 352.3433 (Calcd. for $C_{20}H_{16}O_6$ m/e 352.3428).

Anal. Calcd. for $C_{20}H_{16}O_6$: C, 68.18; H, 4.55. Found: C, 68.24; H, 4.60.

6aβ,12aβ-12a-Hydroxyelliptone (**E'II**).

This compound exhibited ir and nmr spectra superimposable on those of its known racemate; hrms: m/e 368.3465 (Calcd. for $C_{20}H_{16}O_7$ m/e 368.3422.

Anal. Calcd. for $C_{20}H_{16}O_7$: C, 65.22; H, 4.35. Found: C, 65.26; H, 4.31.

6aα,12aα-12a-Hydroxyelliptone (**F'II**).

This compound exhibited ir and nmr spectra superimposable on those of its known racemate: hrms: m/e 368.3448 (Calcd. for $C_{20}H_{16}O_7$ m/e 368.3422).

Anal. Calcd. for $C_{20}H_{16}O_7$: C, 65.22; H, 4.35. Found: C, 65.20; H, 4.36.

6aα,12aα-Deguelin (**HI**).

This compound exhibited ir and nmr spectra superimposable on those of natural deguelin, mp 165-168; hrms: m/e 394.4253 (Calcd. for $C_{23}H_{22}O_6$ m/e 394.4232).

Anal. Calcd. for$C_{23}H_{22}O_6$: C, 70.05; H, 5.58. Found: C, 69.98; H, 5.60.

6aα,12aα-Tephrosin (**HII**).

This compound exhibited ir and nmr spectra superimposable on those of natural tephrosin, mp 194-198°; hrms: m/e 410.4260 (Calcd. for $C_{23}H_{22}O_7$ m/e 410.4225).

Anal. Calcd. for $C_{23}H_{22}O_7$: C, 67.32; H, 7.97. Found: C, 67.31; H, 7.95.

6aα,12aα-α-Toxicarol (**H'II**).

This compound exhibited ir and nmr spectra superimposable on those of natural toxicarol, mp 123-127°; hrms: m/e 410.4254 (Calcd. for $C_{23}H_{22}O_7$ m/e 410.4225).

Anal. Calcd. for $C_{23}H_{22}O_7$: C, 67.32; H, 7.97. Found: C, 67.29; H, 7.99.

6aα-Dehydroisorotenone (6aα-**J**).

This compound exhibited ir and nmr spectral superimposable on those of its known optical antipode, mp 177-179°; hrms: m/e 378.4261 (Calcd. for $C_{23}H_{22}O_5$ m/e 378.4238).

Anal. Calcd. for $C_{23}H_{22}O_5$: C, 73.02; H, 5.82. Found: C, 73.10; H, 5.79.

6aα-Dehydrodeguelin (6aα-**K**).

This compound exhibited ir and nmr spectra superimposable on those of its optical antipode, mp 177-179°; hrms: m/e 378.4241 (Calcd. for $C_{23}H_{22}O_5$ m/e 378.4238).

Anal. Calcd. for $C_{23}H_{22}O_5$: C, 73.02; H, 5.82. Found: 72.96; H, 5.85.

Acknowledgement.

The author thanks Drs. N. Peet (Dow chemical Co), R. Bushway (University of Maine) and M. Jacobson (U. S. Department of Agriculture) for providing samples of rotenoids.

REFERENCES AND NOTES

[1] T. J. Haley, *J. Environ. Pathol. Toxicol.*, **1**, 315 (1978) and references cited therein.

[2] J. Fukami, T. Nakatsugawa and T. Narahashi, *J. Appl. Entomol. Zool.*, **3**, 259 (1959).

[3] T. Unai, *J. Pesticide Sci.*, **5**, 453 (1980).

[4] S. L. Abidi, *J. Chromatogr.*, **317**, 383 (1984).

[5] S. L. Abidi, *J. Heterocyclic Chem.*, **20**, 1687 (1983).

[6] T. Unai and I. Yamamoto., *Agr. Biol. Chem.*, **37**, 897 (1973).

[7] S. L. Abidi, *J. Chromatogr.*, **368**, 59 (1986).

[8] W. H. Pirkle, J. M. Finn, J. L. Schreiner and B. C. Hamper, *J. Am. Chem. Soc.*, **103**, 3964 (1981).

[9] W. H. Pirkle, T. C. Pochapsky, G. S. Mahler and R. E. Field, *J. Chromatogr.*, **348**, 89 (1985) and references cited therein.

[10] Y. Okamoto, K. Suzuki, K. Ohta, K. Hatada and H. Yuki, *J. Am. Chem. Soc.*, **101**, 4763 (1979).

[11] M. Nakazaki, K. Yamamoto and M. Maeda., *J. Org. Chem.*, **46**, 1985 (1981).

[12] Y. Okamoto, S. Honda, K. Hatada and H. Yuki, *J. Chromatogr.*, **350**, 127 (1985) and references cited therein.

[13] L. Crombie, P. J. Godin, D. A. Whiting and K. S. Siddalingaiah, *J. Chem. Soc.*, 2876 (1961).

[14] N. P. Peet and S. Sunder, *J. Heterocyclic Chem.*, **18**, 1123 (1981).

[15] R. S. Cahn, R. F. Phipers and J. J. Boam, *J. Chem. Soc.*, 513 (1938).

[16] S. L. Abidi, *J. Chromatogr.*, in press.

[17] L. Crombie and P. J. Godin, *J. Chem. Soc.*, 2861 (1961).

[18] G. Buchi, L. Crombie, P. J. Godin, J. S. Kaltenbronn, K. S. Siddalingaiah and D. A. Whiting, *J. Chem. Soc.*, 2843 (1961).

[19] D. Carson, L. Crombie and D. A. Whiting, *J. Chem. Soc., Chem. Commun.*, 851 (1975).

[20] C. Djerassi, W. D. Ollis and R. C. Russell, *J. Chem. Soc.*, 1448 (1961).

[21] C. H. Depuy and B. W. Ponder, *J. Am. Chem. Soc.*, **81**, 4629 (1959).

[22] D. Carson, M. W. Cass, L. Crombie, I. Holden and D. A. Whiting, *J. Chem. Soc., Perkin Trans. 1*, 7734 (1982).

[23] T. M. Meijer and D. R. Koolhaas, *Rec. Trav. Chim.*, **58**, 207 (1939).

[24] E. P. Clark, *J. Am. Chem. Soc.*, **53**, 729 (1931).

[25] F. Tattersfield and J. T. Martin, *J. Chem. Soc. Ind.*, **56**, 771 (1937).