of 5E, the intensity of the shoulder at 1360 cm^{-1} increases markedly and the characteristic band of the ferrous high-spin species grows at 1474 cm^{-1}. Of particular interest in spectrum 5D are the upward shifts of the ν_{10}, ν_2, and ν_3 bands; these bands are identified at 1632, 1582, and 1497 cm^{-1}, respectively, for spectrum 5D, and at 1630, 1580, and 1494 cm^{-1} for spectrum 5E. It should be kept in mind that the corresponding bands have been observed at 1629, 1579, and 1493 cm^{-1} for Fe^{III}(OEP)I (Figure 5F), at 1627, 1579, and 1493 cm^{-1} for Fe^{III}(OEP)(2-MeIm) (Figure 1A), and at 1606, 1580, and 1474 cm^{-1} for Fe^{II}-(OEP)(2-MeIm) (Figure 5A). Therefore, the upward-shifted frequencies of the ν_{10}, ν_2, and ν_3 bands cannot be accounted for by a change of the spectral contributions from these three species. The broad feature of these bands in spectrum 5D increased steady-state concentration of an intermediate, which should have the three bands at higher frequencies than those of the three complexes cited above. The three bands are shifted to further higher frequencies in spectrum 5C, suggesting that the relative spectral contribution from the intermediate increases in spectrum 5C. In spectrum 5B, however, the intensities of the ν_{10} and ν_3 bands slightly decrease, and the ν_2 band exhibits a downward shift, probably due to the decreased contribution from the intermediate. Taking account of the presence of band-overlapping, it seems most likely to locate the ν_{10} and ν_3 bands of the intermediate at 1635–1640 and 1500–1505 cm^{-1}, respectively.

The well-known correlation of the RR frequencies of the ν_{10} and ν_3 bands with a structure of iron–porphyrin[16,17] allows us to deduce that the intermediate adopts the ferric low-spin or ferrous intermediate-spin state. Since Fe^{III}(OEP)I does not give a low-spin complex with 2-MeIm under the present condition,[22] it is very unlikely to assign the intermediate to the ferric low-spin species. Consequently, we infer that the intermediate is the intermediate-spin Fe^{II}(OEP) with no axial ligand. In fact, Fe^{II}(OEP) is known to give the ν_{10} and ν_3 bands at 1640 and 1504 cm^{-1}, respectively.[23] Thus the following mechanism appears plausible

$$Fe^{III}(OEP)X \rightarrow Fe^{III}(OEP)(2\text{-MeIm}) \overset{h\nu}{\rightleftharpoons} Fe^{II}(OEP) \rightleftharpoons Fe^{II}(OEP)(2\text{-MeIm})$$

The light excitation of Fe^{III}(OEP)(2-MeIm) causes a charge transfer from 2-MeIm to Fe^{III}(OEP), inducing the dissociation of 2-$MeIm^+$ from the axial coordination position. When there is an excess of 2-MeIm, another 2-MeIm molecule coordinates to the ligand free Fe^{II}(OEP), stabilizing the ferrous high-spin 2-MeIm complex. When the amount of 2-MeIm is too small, Fe^{III}(OEP)(2-MeIm) is not sufficiently supplied. Therefore, the photoreduction depends on the concentration of 2-MeIm. The mechanism described above would be consistent with the observed action spectrum which suggested that the irradiation in the Fe^{III} ← 2-MeIm CT band was most effective for photoreduction. This sort of mechanism was originally proposed by Bartocci et al.[12] from the ESR spin-trapping experiment for photoreduction of Fe^{III}(PP)(py) (PP = protoporphyrin, py = pyridine). Although their proposal was based on the ESR signals of the leaving ligand, we support this from the Raman signals of porphyrin side. It is unexpected that the Fe^{III}–(2-MeIm) stretching Raman band of Fe^{III}(OEP)(2-MeIm) or internal modes of 2-MeIm were not resonance enhanced upon excitation at 441.6 and 488.0 nm. This may be attributed to very small absorbance of the CT transition.

Acknowledgment. We express our gratitude to Dr. T. Ogura, Institute for Molecular Science, for his help in measuring the action spectrum. This work was supported by the Joint Studies Program (1985) of the Institute for Molecular Science and by a Grant-in-Aid for Scientific Research (61740354) to Y.O. and (60470018) to T.K.

(22) The 441.6-nm excited RR spectrum of the aerobic mixture of Fe^{III}-(OEP)I (1.85 mM) with 2-MeIm (185 mM) shown in Figure 1A demonstrates that Fe^{III}(OEP)I forms exclusively the high-spin complex with 2-MeIm.

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Pairwise Additivity in Exciton-Coupled CD Curves of Multichromophoric Systems

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Abstract: The general validity of pairwise additivity in exciton split CD curves was demonstrated by using methyl α-D-glucopyranoside as a model system with *p*-bromobenzoate (λ_{max} = 245 nm) and *p*-methoxycinnamate (λ_{max} = 311 nm) esters as chromophores. All 24 possible dichromophore diacetates (six dicinnamates, six dibenzoates, and 12 monocinnamate monobenzoates) were prepared to represent the degenerate and nondegenerate pairwise interactions contributing to the complex CD curves of multichromophoric cases. In the 20 tri- and tetrachromophoric cases prepared to check the additivity, spectral summation of the three or six corresponding constituent pairwise interactions, respectively, yielded excellent empirical calculations of the observed CD curves throughout the 200–400 nm region. The accuracy of such additive calculations provides a general affirmation of the Principle of Pairwise Additivity and justifies a retroadditive approach to spectral interpretation. Application of a "bichromophoric" derivatization to sterochemical problems including glycosidic linkage determinations promises to be a valuable extension of the exciton chirality method.

When a molecule contains two asymmetrically perturbed chromophores, interpretation of ORD and CD spectra have often benefited from an "additive" approach. Djerassi demonstrated that ORD spectra of diketosteroids[1] and diketoterpenes[2] could be calculated empirically by superimposition of the corresponding monoketone dispersion curves, provided that no "vicinal" interaction between the two carbonyl groups occurred. Snatzke later applied this approach to CD spectra of diketo compounds,[3] yet again such an approach was successful only when one keto group

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was sufficiently distant from the other; summation of independent effects has also been applied to the calculations of ORD[4] and CD[5] spectra of homodichiral or heterodichiral carotenoids with remote chiral end groups. However, in cases where two or more chirally perturbed chromophores are located nearby in space, the "additivity" of monochromophoric contributions becomes invalid.[6]

When two chromophores are located nearby in space, their electronic transitions couple. Chiral disposition of coupled chromophores was treated theoretically by Kuhn,[7] whose classical coupled oscillator model was followed by Kirkwood's quantum mechanical polarizability theory[8] and Moffit's exciton formulation.[9] Exciton theory offers a straightforward, spectroscopic method for determining the absolute configurations of molecules containing chromophores with known transition moments.

Mason first applied this theory to a natural product in determining the absolute configuration of the alkaloid calycanthine.[10] However, an inaccurate understanding of transition orientation may lead to misinterpretation of chiroptic data.[11,12] The most reliable and easily interpretable applications of exciton theory involve derivatizations of organic molecules with "ideal" exciton chromophores, i.e., those having large linear transitions in established directions. Thus, the absolute stereochemistry of diols is best determined according to the dibenzoate chirality rule,[13] the chirality indicated by the signs of the bisignate CD curves. Further extension of this rule led to the exciton chirality method,[14] which has proven to be a powerful and versatile approach to sterochemical problems.[15]

Application of this method to provide molecules with more than two chromophores has shown that an additivity relation exists in the amplitudes (A values) of split CD curves. Thus in hexopyranoside[16] and trichothecene[17] tri- and tetrabenzoates and, more recently, pyranoside benzylates,[18] the A values of split CD curves can be approximated by the sum of constituent pairwise interactions between the benzoate chromophores. This pairwise additivity relation was found for the nearly degenerate benzoate (230 nm)/enone (244 nm) interactions in complex ecdysteroids as well.[19]

Such pairwise additivity follows the largely neglected Principle of Pairwise Additivity proposed by Kauzmann[20] in 1961. While successful application of this principle has remained limited (to molecules possessing a D_{2d} skeleton symmetry, for example[21]), application to exciton-coupled systems seems appropriate because Kauzmann's principle is based upon the coupled oscillator theories of Kuhn and Kirkwood. A conclusive test of pairwise additivity in exciton systems would require accurate predictions of entire CD spectra of molecules containing nondegenerate as well as degenerate exciton interactions.

We have prepared all 24 permutational isomers of methyl α-D-glucopyranoside containing two acetates and two exciton chromophores: *p*-bromobenzoate(s) and/or the red-shifted *p*-methoxycinnamate(s). The CD spectra of these 24 derivatives represent a *basis set of all possible pairwise interactions in tri- and tetrachromophoric derivatives*. We herein report the general validity of pairwise additivity in exciton-coupled systems by demonstrating that the complex CD spectra of 20 methyl α-D-glucopyranoside *p*-bromobenzoate *p*-methoxycinnamates can be fully calculated by spectral summation of all degenerate and nondegenerate constituent pairwise interactions.[22]

Materials and Methods

All compounds were prepared by various sequences of partial acetylations, *p*-bromobenzoylations, and *p*-methoxycinnamylations of the parent methyl α-D-glucopyranoside. Sequences followed the typical reaction order for esterification of the four hydroxyl groups: 6 ≫ 2 > 3 ≫ 4. For example, partial *p*-methoxycinnamylation of the methyl glucoside (*p*-methoxycinnamyl chloride,[23] pyridine, DMAP as acylation catalyst,[24] 60 °C) afforded the following 11 *p*-methoxycinnamate derivatives: 2,3,4,6-tetracinnamate, 2,3,6- and 2,4,6- tricinnamates, 2,3-, 2,4-, 2,6-, 3,6-, and 4,6-dicinnamates, and the 2-, 3-, and 6-monocinnamates. These were separated first by a gradient $MeOH/CH_2Cl_2$ solvent system (0–10%) on silica followed by further purifications on silica with EtOAc/hexane. Each of these products were then further derivatized at the remaining free hydroxyls with acetate (acetic anhydride) and/or bromobenzoate (*p*-bromobenzoyl chloride) substituents. The 4,6-derivatives were obtained from the 2,3-diacetates, which were conveniently prepared by acetylation of the 4,6-benzylidene followed by deprotection (5% TsOH/MeOH, 0 °C, 40 min).

All intermediates and final products were characterized by ^1H NMR (Brucker WM250, 250 MHz, $CDCl_3$ solvent) with decoupling of ring protons whenever necessary to confirm substitution pattern; the data are given in Supplementary Material. We have observed that the relative positions of protons 6-H_R and 6-H_S were indicative of the substitution pattern for the 76 6-acylates: (1) in 20 derivatives with a cinnamate at the 6-position and any other acyl group at the 4-position, these two protons were chemical shift equivalent;[25] however, in the 4,6-dicinnamates with the 3-position underivatized (2 examples), 6-H_R was upfield of 6-H_S. (2) In all 18 derivatives with a benzoate at the 6-position and any other acyl group at the 4-position, 6-H_R was also upfield of 6-H_S. (3) In all 36 other 6-acylated cases (all 6-acetates; 6-benzoates and 6-cinnamates with the 4-position underivatized) the 6-H_R proton was always *downfield* of 6-H_S.

UV measurements were performed on a Perkin Elmer 320 UV spectrophotometer. CD spectra were recorded on a JASCO 500A spectropolarimeter driven by a JASCO DP500N data processor (four scans were taken of each compound from 200–400 nm). An IBM-PC operating with JASCO software was used to normalize all CD spectra to 1.0×10^{-5} M as well as to perform all spectral summations for empirical calculations, which simply involved addition of the normalized CD spectra of the three or six appropriate dichromophoric derivatives corresponding to the constituent pairwise interactions of the tri- or tetrachromophoric derivatives, respectively.

Prior to measurement of CD spectra, all compounds were purified by HPLC (EtOAc/hexane solvent systems, silica column) to ensure accuracy of concentration determinations. Acetonitrile[26] solutions were prepared which were $0.5–1.5 \times 10^{-5}$ M, the concentrations of which were determined on the basis of the experimentally determined average *p*-methoxycinnamate UV ε's at 311 nm (where bromobenzoates are transparent): mono, ε 24 000; di, ε 45 000; tri, ε 68 000. In the case of the diacetate dibromobenzoates, the average dibromobenzoate ε 38 200 at 245 nm is used.[27]

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Table I. CD Data for "Homo" Interactions: The Six Dibenzoate Diacetates and the Six Dicinnamate Diacetates of the Basis Set

entry	compd[a]	λ[b] (Δε)	λ (Δε)	Δε = 0[c]	A[d]
1	BAAB	235 (+5)	255 (−3)	247	−8
2	BBAA	235 (−22)	253 (+47)	243	+69
3	ABAB	234 (−5)	252 (+11)	237	+16
4	BABA	237 (+4)	252 (−6)	245	−10
5	ABBA	236 (+21)	254 (−40)	243	−61
6	AABB	235 (−4)	252 (+20)	240	+24
7	CACA	283 (+4)	324 (−6)	308	−10
8	AACC	285 (−11)	321 (+21)	303	+32
9	ACAC	287 (−5)	320 (+10)	303	+15
10	CAAC	290 (+2)	324 (−3)	307	−5
11	ACCA	289 (+38)	323 (−63)	307	−101
12	CCAA	287 (−34)	323 (+61)	306	+95

[a] Derivatization of the four positions is represented by the order 2, 3, 4, 6 by A = acetate, B = *p*-bromobenzoate, and C = *p*-methoxycinnamate. [b] Extrema in nm. [c] Point where Δε changes sign, in nm. [d] *A* value or difference between extrema.

Table II. CD Data for "Hetero" Interactions: The Twelve Monobenzoate Monocinnamate Diacetates of the Basis Set

entry	compd[a]	λ[b] (Δε)	λ (Δε)	λ (Δε)	Δε = 0[c]
1	AACB		256 (−3)	306 (+1)	261
2	AABC		244 (−5)	303 (+5)	261
3	BACA		242 (+5)	300 (−2)	255
4	CABA		238 (−4)	304 (−3)	
5	ACAB	237 (+1)	253 (−2)	302 (+2)	242, 262
6	ABAC		243 (−3)	291 (+3)	258
7	CAAB	241 (+2)	253 (−2)	305 (+2)	251, 266
8	BAAC		238 (+4)	311 (+0.4)	260
9	CBAA		246 (−15)	311 (+13)	262
10	BCAA		248 (−13)	304 (+12)	263
11	ACBA		254 (+11)	302 (−12)	263
12	ABCA		248 (+10)	308 (−10)	263

[a] Derivatization of the four positions is represented in the order 2, 3, 4, 6 by A = acetate, B = *p*-bromobenzoate, and C = *p*-methoxycinnamate. [b] Extrema in nm. [c] Point where Δε changes sign, in nm.

Results

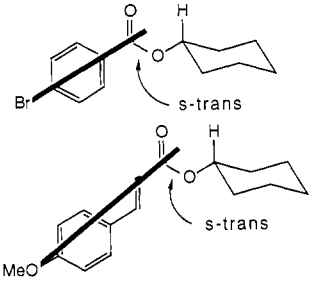
In connection with attempts to develop an exciton chirality microanalysis of glycosidic linkages,[28] we have found[22] that addition of a red-shifted chromophore to hexopyranoside *p*-bromobenzoates (λ_{max} 244 nm) results in complex CD spectra in the 220–360 nm range which are unique for each substitution pattern. These characteristics, while desirable for a discriminating methodology, indicated the need to develop a clear understanding of the effects contributing to such spectra.

The closer the λ_{max} of interacting chromophores, the more efficient the coupling, yet a split CD is observed even when the λ_{max} values differ by as much as 80 nm.[28a,29] The *p*-methoxycinnamate chromophore (λ_{max} 311 nm, ε 24 000) was chosen: benzoate/benzoate (B/B) and cinnamate/cinnamate (C/C) degenerate ("homo") interactions are well separated, yet strong benzoate/cinnamate (B/C) nondegenerate "hetero" interactions are still observed. Such "bichromophoric" systems can have three types of interactions, i.e., B/C, B/B, and/or C/C.

To determine the contribution of every possible pairwise interaction, all 24 possible methyl α-D-glucopyranosides containing these two chromophoric esters were prepared and measured, thus constituting a "basis set" of paired interactions (Tables I and II). The free hydroxyls were converted to "nonchromophoric" acetates to best simulate both ring and 6-position conformations in the tetraester test cases. Studies have shown[30] the sensitivity of exciton coupling to conformational changes; this is due to theoretically predicted and experimentally demonstrated dependency of coupling magnitude upon both dihedral angle and interchromophoric distance between transitions.[15,27]

Interactions involving chromophores at the 6-position are particularly sensitive because there are three stable rotamers about the C5–C6 bond (see Figure 6 below). It is important that the rotamer populations of the dichromophoric "basis set" compounds match those of the test case compounds for an accurate representation of the pairwise interactions involving chromophores at position 6. ^{1}H NMR coupling constants indicate that the acetate groups are in certain cases important for conformational mimicry, especially in 2,6-substituted species, and indeed acetylation of the 2,6-dichromophoric derivatives results in marked changes in the CD spectra. In most other cases, however, the CD spectra of glucopyranosides containing the B and C chromophores are practically unchanged by acetylation of the free hydroxyls. In certain cases (discussed below), acetates do not adequately mimic the steric bulk of benzoates, although they still provide some improvement over free hydroxyls.

The six dibenzoate diacetate interactions result in split CD's with extrema around 235/253 nm (Table I, entries 1–6), while the six dicinnnamate diacetates show typical splitting with extrema around 287/322 nm (entries 7–12). Due to the large number of derivatives of the same parent glucoside, simple four-letter abbreviations are used throughout the tables, figures, and text. Derivatization of the four positions is represented in the order 2, 3, 4, 6 by A = acetate, B = *p*-bromobenzoate, C = *p*-methoxycinnamate, and O = underivatized hydroxyl. The *A* values of the stronger absorbing dicinnamates are generally larger, though in two cases (entries 7 and 10) slightly smaller than the dibenzoate couplings, a difference based not only upon the larger extinction coefficient (in homointeracting systems, *A* is linearly proportional to the chromophore ε[31]) but also upon the differing geometry of the cinnamate ester. While both benzoate and cinnamate may be expected to adopt the normal s-trans configuration with the carbonyl staggered 30° to either side of the carbinyl hydrogen,[32] the cinnamate transition is centered farther from the pyranoside ring, and it adopts a different angle relative to it.



The 12 monobenzoate monocinnamate diacetates show hetero split CD interactions with extrema around 246/305 nm (Table II) and broad shoulders around 285 and 310 nm. The magnitude of these "hetero" B/C interactions is generally on the order of one quarter to one third of the magnitude of the "homo" interactions. This is in accord with theoretical calculations[28a,29] for a λ_{max} separation of 65 nm in this region. The shoulders can be accounted for by coupling of the ^1L_b transition of the bromobenzoate (283 nm, ε 500) with the major transition of the cinnamate (311 nm). This ^1L_b transition, being perpendicular to the major ^1L_a transition at 245 nm, would thus be expected to couple with the cinnamate to afford a small contributing split CD with extrema near 283/310 nm with a sign *always* opposite to that of the major split CD. Addition of this small coupling to the coupling of the major transitions would result in a CD curve (Σ) with a shoulder near 283 nm and a relative depression near 310 nm as shown in the following schematic diagram. The maxima of hetero spectra, occurring around 303 nm rather than the expected 310

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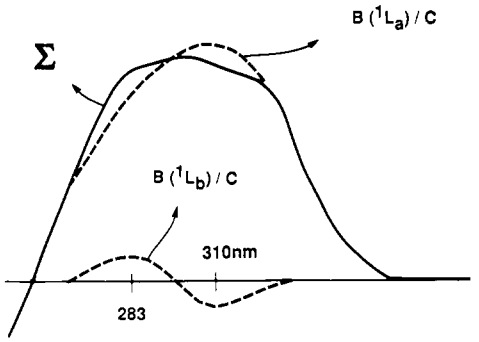
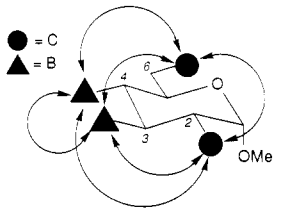


nm, seems to confirm this rationale. Thus a hetero B/C interaction is probably the sum of a major B(^1L_a)/C and a minor B(^1L_b)/C interaction.

Additivity studies involved summation of the appropriate three or six dichromophore spectra which correspond, respectively, to the contributing pairwise interactions in a tri- or tetrachromophoric case. For example, the six pairwise interactions (one B/B, one C/C, and four B/C) expected to contribute to the spectrum of the 3,4-dibenzoate 2,6-dicinnamate (CBBC) are shown. The



spectra of the six basis set compounds representing all six pairwise interactions in CBBC are shown in Figure 1. Note that while the four hetero interactions (Figure 1a) are generally smaller than the homo interactions (Figure 1b), they are more numerous. The spectra representing the four hetero interactions shown in Figure 1a are added together (Σ hetero), and this sum is then added to the two appropriate homo interactions ABBA and CAAC (Figure 1b) to give the sum total (Σ total) of all contributing pairwise interactions, the empirically calculated CD spectrum of methyl α-D-glucopyranoside 3,4 di-*p*-bromobenzoate 2,6-di-*p*-methoxycinnamate.

The excellent agreement between calculated and observed spectra for this (Figure 2b, CBBC) and the other five dibenzoate dicinnamates is shown in Figure 2. While the calculation of BCBC (Figure 2d) overestimates the magnitude of the contributing C/C interaction, this difference is due to a conformational mismatch as demonstrated by NMR and model studies (discussed later). Calculations are also excellent for the four monobenzoate tricinnamates (Figure 3) and the four monocinnamate tribenzoates (Figure 4) as well. In these two groups, the calculated spectra are also the sum of contributions from six interactions; however, here the sum of three homo and three hetero interactions is required. In trichromophoric species, such as the dicinnamate monobenzoate monoacetates and the dibenzoate monocinnamate monoacetate (Figure 5), calculations require only summation of three (one homo and two hetero) pairwise contributions. Agreement in these less complex cases is also excellent. In addition, we have recently demonstrated pairwise additivity in galactoside derivatives as well, where presence of the axial 4-position generally leads to more pronounced CD spectra[33] than the glucose derivatives having all equatorial substituents.

Discussion

The precision with which these empirical calculations simulate observed CD spectra for tri- and tetrachromophoric species clearly demonstrates that the optical activity of exciton-coupled systems is due to a summation of all pairwise interactions involved. Such

(33) Vázquez, J. T.; Wiesler, W. T.; Nakanishi, K., submitted for publication.

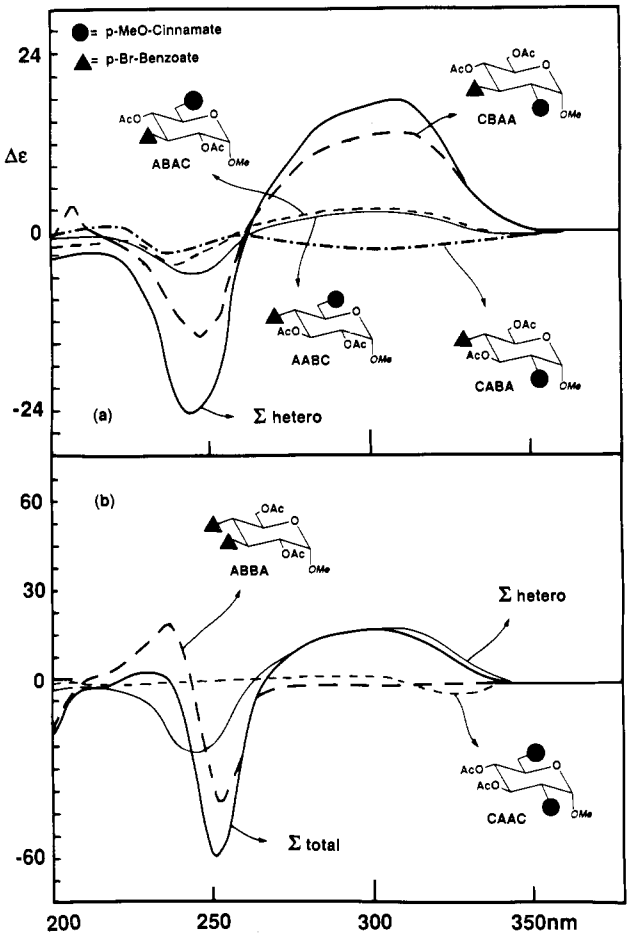


Figure 1. Calculation of the CD curve of methyl α-D-glucopyranoside 3,4-dibromobenzoate 2,6-dimethoxycinnamate CBBC. (a) The CD curves of the four contributing hetero interactions. The sum of these four "hetero" interactions is shown as Σ hetero. (b) Summation of the two "homo" interactions ABBA and CAAC and the summed "hetero" interactions (Σ hetero, Figure 1a) give the empirically calculated CD spectrum (Σ total) of the 3,4-dibenzoate 2,6-dicinnamate. For an addition example of this type of calculation, see ref 22.

a result represents a general confirmation of the Principle of Pairwise Interactions of Kauzmann and, in the case of exciton coupled systems, demonstrates that consideration of three-way or higher interaction terms need not be considered.

Tests and applications of Kauzmann's principle have always used rigid ring backbones to provide a fixed geometry between chromophores. An application of the principle by Johnson[34] to simulate CD spectra of pyranoid monosaccharides in the vacuum UV range (165–190 nm) suggested the validity of the principle. Summation of "fragment" spectra of nearest neighbor interactions within sugars were used to provide good simulations of monosaccharide spectra. However, the "fragment" spectra were obtained as difference spectra (assuming additivity), and therefore the successful summation of difference spectra does not represent proof of the pairwise principle.

Successful tests of both this principle and of the group theoretical Theory of Chirality Functions,[35] another basis for semiempirical approach, have been limited, remarkably, to molecules with D_{2d} skeleton symmetry.[21] This is likely due to the inability to account for chromophore/skeleton interactions unless they vanish as a result of symmetry. Such effects are negligible in the exciton-coupled systems we have studied. Interchromophoric interactions are orders of magnitude greater than chromo-

(34) (a) Johnson, W. C. *Carbohyr. Res.* **1977**, *58*, 9. (b) Nelson, R. G.; Johnson, W. C. *J. Am. Chem. Soc.* **1976**, *98*, 4290. (c) Nelson, R. G.; Johnson, W. C. *Ibid.* **1976**, *98*, 4296.

(35) Ruch, E.; Schönhofer, A. *Theor. Chim. Acta* **1968**, *10*, 91; **1970**, *19*, 225.

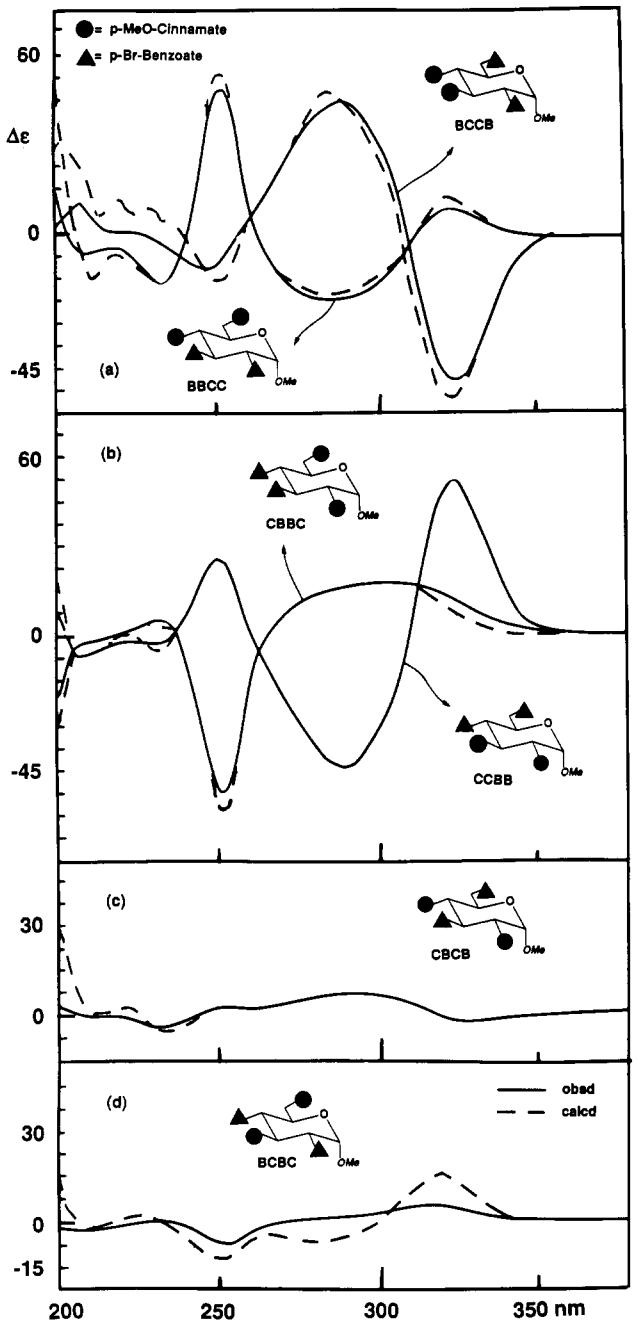


Figure 2. Comparison of calculated and observed CD spectra for the six dibenzoate dicinnamates of methyl α-D-glucopyranoside.

phore/skeleton interactions, and thus exciton systems represent "idealized" systems to study chiroptic effects.

Deviations between pairwise additive calculations and observed CD spectra are generally due to one or more conformational mismatches between a contributing basis set compound and the interaction which it represents. These mismatches generally involve rotamer population differences around the C5–C6 bond, important when a chromophore is at the 6-position. The pairwise interactions in each of the three stable rotamers differ both in sign and magnitude. The three rotamers and their 3,6- and 4,6-coupling modes are shown in Figure 6. The fact that experimental values for both 3,6- and 4,6-interactions are always positive (Table I, entries 3, 6, 8, and 9) indicates that the GL rotamer with the chromophore in the exo conformation over the pyranose ring is favored.

NMR studies by Nishida et al.[36] used coupling constants to show the rotamer distribution in various D-glucopyranose deriv-

(36) Nishida, Y.; Ohrui, H.; Meguro, H. *Tetrahedron Lett.* **1984**, *25*, 1575.

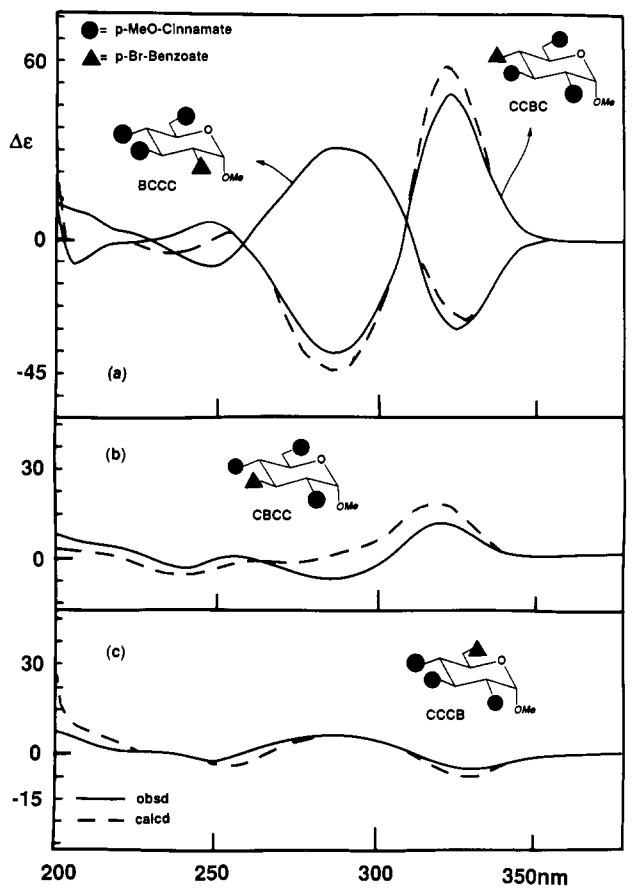


Figure 3. Comparison of calculated and observed CD spectra for the four monobenzoate tricinnamates.

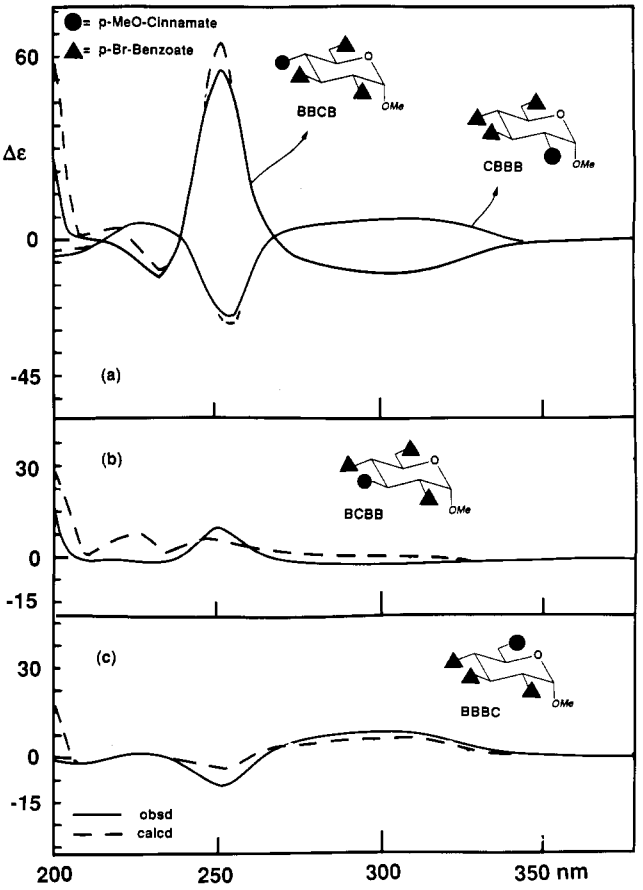


Figure 4. Comparison of calculated and observed CD spectra for the four monocinnamate tribenzoates.

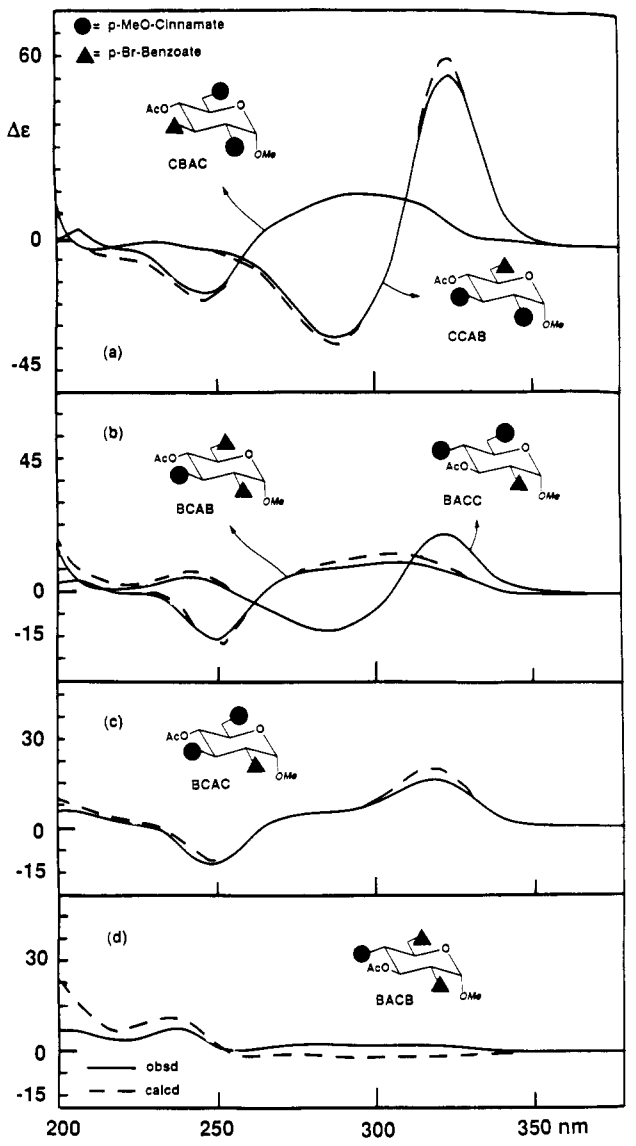


Figure 5. Comparison of calculated and observed CD spectra for six trichromophoric derivatives.

Table III. Rotamer Populations about the C5–C6 Bond in 6-Acrylates

entry no.	compd class[a]	no. of Compounds	$J_{5,6}$ av (Hz)		rotamers[b] (%)		
			$J_{5,6R}$	$J_{5,6S}$	GL	GR	T
1	XXOB	9	4.5	2.2	66	29	5
2	XXAB	6	4.6	2.5	63	29	8
3	XXCB	8	4.9	2.8	59	31	10
4	XXBB	4	4.8	2.9	59	29	11
5	XXOA	5	4.4	2.2	67	28	5
6	XXAA	4	4.6	2.3	64	30	6
7	XXCA	5	4.8	2.5	62	31	7
8	XXBA	2	5.3	2.6	56	36	7
9	XXOC	10	3.9	2.0	73	23	4
10	XXAC	8	3.6				
11	XACC						
	XCCC	7	4.1				
	XBCC						
12	XOCC	2	4.9	3.3	56	29	15
13	XXBC	5	4.2				

[a] Derivatization of the four positions is represented in the order 2, 3, 4, 6 by A = acetate, B = bromobenzoate, C = methoxycinnamate, O = underivatized hydroxyl, X = A, B, C, or O. [b] Calculations use the equations from Nishida et al., ref 36.

atives as GL > GR ≫ T. We have applied the equations from this study to calculate similar rotamer distributions shown in Table III; however, they could not be applied to 6-cinnamates which were derivatized at the 4-position (entries 10, 11, and 13). In these cases the chemical shifts of the 6_R and 6_S protons are always equivalent, thus providing only a single doublet rather than the usual, clearly differentiated doublet of doublets for each proton. Rotamer distributions could, however, be calculated for 6-cinnamates underivatized at the 4-position (entry 9), and these indicate that the GL rotamer is even more dominant in 6-cinnamates than in 6-benzoates or acetates.[37]

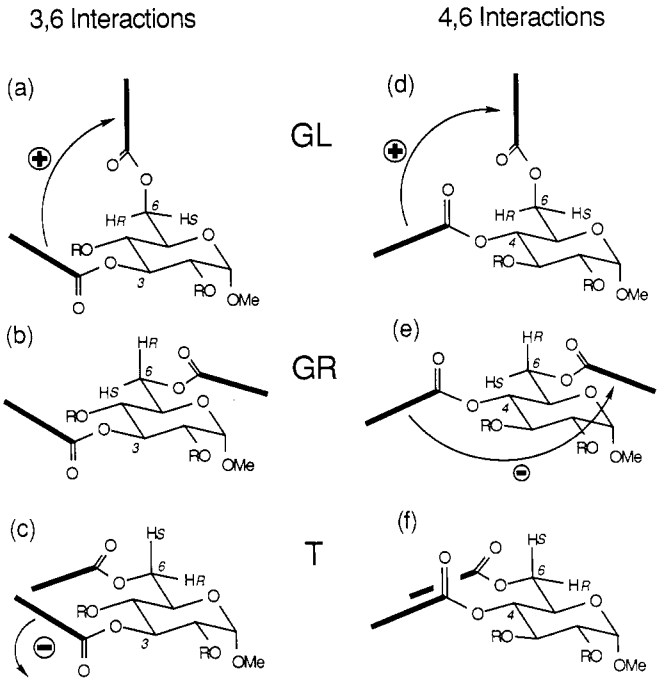


Figure 6. The three stable rotamers (GL, GR, and T) about the C5–C6 bond and the coupling of the chromophore at C-6 to a chromophore at the 3-position (a–c) and the 4-position (d–f) for each of the three rotamers.

An increase in the bulk of the 4-position substituent results in a decreased proportion of GL rotamer as calculated from coupling constants. For example, the proportion of GL rotamer in 6-benzoates (entries 1–4) is calculated to decrease as the substituent in the 4-position goes from the underivatized free hydroxyl (66%) to acetate (63%) and finally to cinnamate and benzoate (59%). A similar pattern is observed in the 6-acetate series (entries 5–8). While the rotamer distributions can be calculated for only a few 6-cinnamates (entry 9, XXOC, 73% GL and entry 12, XOCC, 56% GL), a similar pattern is indicated by the coupling constant $J_{5,6}$ increasing from 3.6 Hz in the 4-acetates to 4.1 Hz in 4-cinnamates and to 4.2 Hz in the 4-benzoates, corresponding to a decrease of the GL rotamer.

Comparison of coupling constants in additivity test derivatives to corresponding coupling constants in the contributing basis set compounds provides an indication of the accuracy with which each basis set compound mimics the conformation of the system with the pairwise interaction which it is intended to simulate.[38] Discrepancies indicate a few clear cases of conformationally mismatched basis set and tetrachromophoric compounds, thus accounting for the differences between calculated and observed CD spectra for the few cases where agreement is less than excellent.[39] For example, in the dibenzoate dicinnamate series, the

(37) The larger, more positive split CD curves obtained from 6-cinnamates vs. 6-benzoates, i.e., AABC > AACB and ABAC > ACAB (Table II), also indicate the increased preference for the GL rotamer when the 6-position is derivatized as cinnamate. Furthermore, equivalence of the CD spectra of ABAC and OBOC, for example, makes it reasonable to extend this conclusion to derivatives for which distributions cannot be directly calculated.

(38) Most of the 20 additivity test cases show good coupling constant matching with the basis set compounds contributing to the empirically calculated CD spectra. As an example, for CCBB, $J_{2,3} = J_{3,4} = 9.8$; $J_{5,6} = 3.0$, 4.8. These are then compared to the following: CCAA, $J_{2,3} = 9.8$; ACBA, $J_{3,4} = 9.8$; ACAB, $J_{5,6} = 2.5$, 4.7; AABB, $J_{5,6} = 3.1$, 4.8. The basis set compounds show good agreement with the additivity test case.

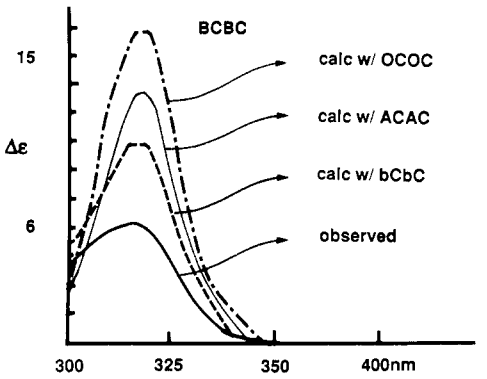


Figure 7. Comparison of three empirical calculations vs. the observed CD spectrum in the C/C coupling (300–350 nm) region for BCBC (Figure 2d). Increasing the bulk of the C-4 substituent from hydroxyl to acetate to benzoate (b) decreases the proportion of the positively coupling GL rotamer (Figure 6a) and thus reduces the magnitude of C/C coupling.

only basis compound with a coupling constant which differs by more than 0.5 Hz from the corresponding test compound was ACAC ($J_{5,6}$ = 3.4 Hz) representing the C/C interaction in BCBC ($J_{5,6}$ = 4.1 Hz). Because the smaller $J_{5,6}$ of the basis set compound indicates it to have a greater proportion of the positively coupled GL rotamer (Figure 6a), the calculated spectrum would be expected to overestimate the magnitude of the C/C interaction, and, as shown in Figure 2d, this is found to occur. To test this rationale, bCbC (b = unsubstituted benzoate, a weaker and blue-shifted chromophore relative to *p*-bromobenzoate) was prepared. The weakly interacting benzoate at C-4 should affect the C/C coupling only insofar as it dictates a distribution of rotamers which matches the distribution in BCBC. As expected, the CD spectrum showed smaller C/C interaction than ACAC. The b/b and b/C interactions in this compound are negligible, and its use as a C/C basis set compound contributing to the CD spectra of BCBC leads to improved calculations. Calculations of BCBC using three different C/C basis spectra are shown in Figure 7. The calculation improves with increasing bulk of the "nonchromophoric" substituents on the C/C basis set derivative. Its use also improves the calculation of CCBC (Figure 3a). Similarly, the overcalculated hetero B/C contribution to the calculation of CBCC (Figure 3b) may be understood as a mismatch between the 3,6-interactions in ABAC ($J_{5,6}$ = 3.6 Hz) and in CBCC ($J_{5,6}$ = 3.9 Hz).

In conclusion, discrepancies between calculation and observed spectra arise in cases of conformational mismatch, usually because an acetate at the 4-position in basis set compounds inadequately mimics larger groups as regards its influence on the distribution of 6-cinnamate rotamers. Aside from this particular conformational change, we have generally found that the CD spectra of di- and trichromophoric derivatives are nearly unchanged upon acetylation of the free hydroxyls.

Conclusion

While we have clearly demonstrated pairwise additivity in "bichromophoric" (i.e., two types of chromophores) exciton-coupled systems, we can expect pairwise additivity to be valid for exciton-coupled systems containing any number of different chromophore types.

The general validity of pairwise additivity in exciton systems expands the utility of the exciton chirality method, especially when dealing with any multichromophoric systems. The CD curves of exciton systems suggest the types, magnitudes, and signs of the *pairwise* interactions involved. For example, the spectra of the 4-cinnamate 2,3,6-tribenzoate, BBCB (Figure 4a), is clearly dominated by B/B interaction, while in the case of CBAC (Figure 5a), hetero coupling dominates the spectrum.

The CD spectrum of any derivative is largely dependent upon the vicinal couplings 2/3 (positive, i.e., B/B interaction in BBCC, Figure 2a) and 3/4 (negative, i.e., B/B interaction in CBBC, Figure 2b). However, when the 2/3 and 3/4 interactions are of the same type, i.e., both B/C, then the contributions of these two oppositely signed pairwise interactions cancel, resulting in smaller CD spectra with less definition (see, for example, CBCB, Figure 2c, or BCBC, Figure 2d). Thus, the glucosides studied here generally have less pronounced spectra than the corresponding galactosides (2/3 and 3/4 interactions both positive due to the axial 4-position) or mannosides (2/3 and 3/4 interactions both negative due to the axial 2-position), and yet the spectrum of each glucoside derivative is still unique and *indicative of substitution pattern.*

An important consequence of pairwise additivity in "bichromophoric" derivatives is the *ability to assign substitution pattern and configuration directly from the CD spectrum.* This is in marked contrast to the use of the octant rule to interpret ORD or CD spectra, which has been proven[40] to be a "one-way rule": prediction of the sign of a Cotton effect of a known configuration is possible, but assignment of configurations by the Cotton effect without further structural information is not. We have found it possible to assign bichromophoric exciton spectra of hexopyranoside derivatives to the appropriate sugar, substitution pattern, and D or L enantiomer on the basis of UV (to determine chromophoric ratios) and CD alone. For example, the spectrum of BBCC (Figure 2a) shows a large positive B/B interaction and a smaller positive C/C interaction. The large positive B/B interaction can only be due to 2,3-substitution of a glucoside or either 2,3- or 3,4-substitution of a galactoside. Of these three choices, only the 2,3-dibenzoate of glucopyranoside could have a positive C/C interaction between the remaining (4,6) positions.[16]

We are currently developing methods to apply selective "bichromophoric" derivatization to various structural problems. While use of only one type of chromophore provides for a single split CD curve of a particular sign and magnitude, use of two types of chromophores in a "bichromophoric" approach provides for information-rich yet easily interpretable CD spectra. Such an approach is currently being applied to oligosaccharide microanalysis, wherein the free hydroxyls are derivatized with one type of chromophore, and the linkage positions are tagged with a second chromophore. The resulting subunits, of which the tetrachromophoric pyranosides are representative, are then fully characterizable by UV and CD measurements. In addition to indicating sugar type and linkage pattern, this approach provides for clear distinction between D and L enantiomers. The highly characteristic nature of the CD spectra is notable, especially considering that all spectra were recorded by using only 20 nmol of material. High sensitivity, combined with straightforward spectral interpretation due to a clearly demonstrated pairwise additivity, should lead to further exploitations of the exciton chirality method.

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Supplementary Material Available: ^{1}H NMR data and CD data for all acylates (10 pages). Ordering information is given on any current masthead page.

(39) Note that ^{1}H NMR spectra were recorded in $CDCl_3$, while CD spectra were taken in CH_3CN. While it is conceivable that the two solvents may show preference for different conformations, and, specifically, different rotamer populations of a particular glucoside derivative, NMR spectra of several derivatives recorded in CD_3CN indicate no appreciable conformational differences.

(40) Dugundi, J.; Marquarding, D.; Ugi, I. *Chem. Scr.* **1976**, *9*, 74.