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Low-Temperature NMR *J*-Based Configurational Analysis of Flexible Acyclic Systems

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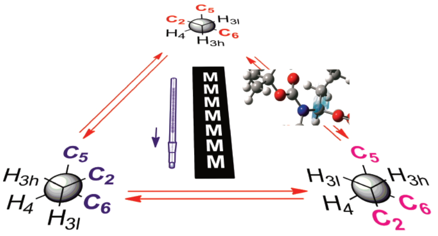
Received July 23, 2010



A new strategy for the *J*-based configurational analysis in acyclic systems is applied where the existence of a multiple conformer equilibrium precludes the application of the Murata methodology.

Introduction

NMR chemical shifts are, along with coupling constants, the most widely used key parameters to deduce skeleton frameworks and three-dimensional arrangements in a molecule with an unknown stereochemistry.

Several distinct research areas—such as asymmetric synthesis, medicinal chemistry, chemical biology, and in particular, natural products chemistry—rely on NMR spectroscopy to assign unequivocally the full configuration of an organic compound. The correct assessment of stereochemical features is crucial because all chemical or biological properties are fully dependent on all stereocenters in a chiral substrate. The predictable conformational behavior in cyclic compounds allows the deduction, in most cases, of the relative configuration by analysis of basic NMR parameters such as proton−proton coupling constants or by NOE/ROE correlations. However, as soon as the compound contains a flexible moiety, such as a polysubstituted open chain, the elucidation of the stereochemistry becomes more challenging and sometimes this is the bottleneck in its full characterization.[1]

Several approaches have become very popular in the last 10 years to study the relative stereochemistry in these acyclic compounds. In 1999, Murata and co-workers developed a robust and logical method, named *J*-based configurational analysis that has been widely adopted by the scientific community.[2]

This methodology is based on the extensive use of the proton−proton and carbon−proton coupling constants, and it has been applied with great success in dimethine systems that contain 1,2 and 1,3 hydroxyl,[3] hydroxymethyl, amino,[4] or chlorine[5] substituents attached to chiral carbons. An improvement in this methodology was developed by Bifulco, Gomez-Paloma, and co-workers, and this involved ab initio DFT methods[6] to calculate the appropriate coupling constants and comparison of these with the experimental values obtained in NMR experiments such as *J*-HMBC or HETLOC to elucidate more accurately the relative stereochemistry of such systems.[7]

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(1) Bifulco, G.; Dambruoso, P.; Gomez-Paloma, L.; Riccio, R. *Chem. Rev.* **2007**, *107*, 3744–3779.

(2) Matsumori, N.; Kaneno, D.; Murata, M.; Nakamura, H.; Tachibana, K. *J. Org. Chem.* **1999**, *64*, 866–876.

(3) Some examples: (a) Bassarello, C.; Bifulco, G.; Zampella, A.; D'Auria, M. V.; Riccio, R.; Gomez-Paloma, L. *Eur. J. Org. Chem.* **2001**, 39–44. (b) Campagnuolo, C.; Fattorusso, C.; Fattorusso, E.; Ianaro, A.; Pisano, B.; Taglialatela-Scafati, O. *Org. Lett.* **2003**, *5*, 673–676. (c) Cimino, P.; Bifulco, G.; Evidente, A.; Abouzeid, M.; Riccio, R.; Gomez-Paloma, L. *Org. Lett.* **2002**, *4*, 2779–2782. (d) Espelt, L.; Parella, T.; Bujons, J.; Solans, C.; Joglar, J.; Delgado, A.; Clapes, P. *Chem.—Eur. J.* **2003**, *9*, 4887–4899. (e) Plaza, A.; Bifulco, G.; Keffer, J. L.; Lloyd, J. R.; Baker, H.; Bewley, C. A. *J. Org. Chem.* **2009**, *74*, 504–512.

(4) (a) Randazzo, A.; Bifulco, G.; Giannini, C.; Bucci, M.; Debitus, C.; Cirino, G.; Gomez-Paloma, L. *J. Am. Chem. Soc.* **2001**, *123*, 10870–10876. (b) Ardá, A.; Rodríguez, J.; Nieto, R. M.; Bassarello, C.; Gomez-Paloma, L.; Bifulco, G.; Jiménez, C. *Tetrahedron* **2005**, *61*, 10093–10098. (d) Ardá, A.; Jiménez, C.; Rodríguez, J. *Eur. J. Org. Chem.* **2006**, *16*, 3645–3651.

(5) Nilewski, C.; Geisser, R. W; Ebert, M.-O.; Carreira, E. M. *J. Am. Chem. Soc.* **2009**, *131*, 15866–15876.

(6) Bifulco, G.; Bassarello, C.; Riccio, R.; Gomez-Paloma, L. *Org. Lett.* **2004**, *6*, 1025–1028.

DOI: 10.1021/jo101331e

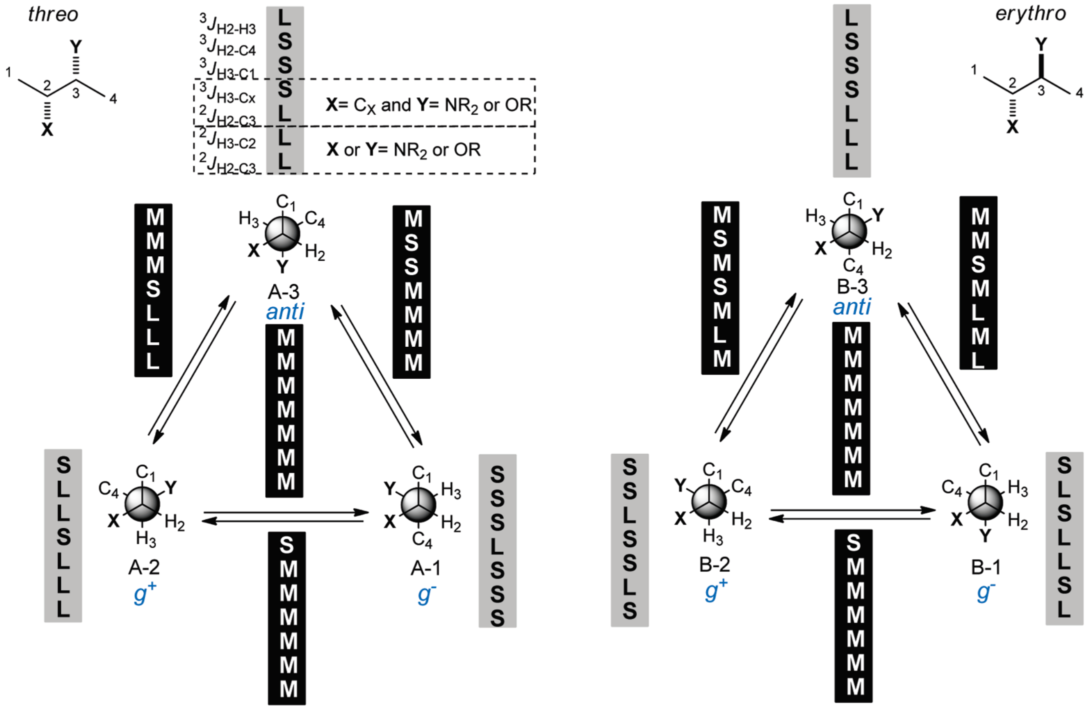

FIGURE 1. Coupling constant patterns in 1,2-dimethine systems with single or equilibrating rotamers: (L) large, M (medium), S (small).

There are a large number of examples in the literature where these systems completely satisfy Murata's methodology, and we have named these Murata models. The application of simple rules to each alternate conformer of every carbon−carbon bond in 1,2-dimethine-substituted systems allows the elucidation of the relative configuration.[1] Indeed, the measurements of proton−proton and carbon−proton coupling constants of the six staggered Newman projections in the two possible relative *threo* and *erythro* configurations allows these to be distinguished. A pattern of large (L), medium (M), or small (S) for the $^3J_{HH}$, $^2J_{CH}$, and $^3J_{CH}$ values can be pictured when a single rotamer is present (gray labels in Figure 1). Because the magnitude of the heteronuclear coupling constants depends upon the electronegativity of the substituents directly connected to the carbons of interest, the range of variability has to be known for each specific substitution pattern. These values can be computed as a function of the dihedral angles[8] and have been estimated for 1,2 dimethine non-, mono-, and dioxygenated systems.[2,9,10] For aminated frameworks, the values are qualitatively similar to those of oxygenated systems, although the $^2J_{CH}$ values fall in a tighter interval (−4/-6 large and 2/−1 small).[11]

The analysis can also be extended to methylene-spaced stereopairs (1,3-dimethine substituted systems) because the methylene diastereotopic protons (H_h and H_l)[12] can be considered as different in a combination of two 1,2-substituent systems.

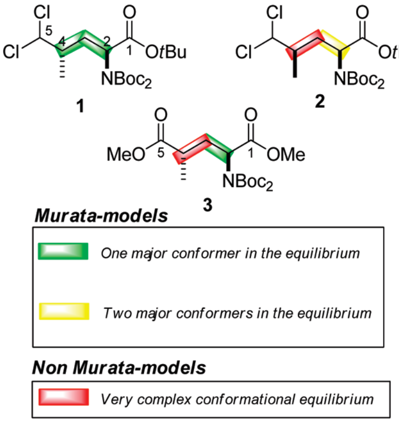

FIGURE 2. Examples of Murata-model and non-Murata-model compounds used in this study.

Results and Discussion

When two rotamers exist in equilibrium, averaged values for the coupling constants (black labels in Figure 1) still allow the relative configuration to be deduced. In these cases, it is assumed that there is an equilibrium involving the aforementioned staggered interconverting rotamers, and this allows unequivocal relative configuration assignments. In the case when gauche-(+)/gauche-(−) (g^+ and g^- in Figure 1) are the rotamers in the equilibrium, both the *threo* and *erythro* series are spectroscopically indistinguishable, and

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(7) HETLOC: (a) Kurz, M.; Schmieder, P.; Kessler, H. *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 1329–1331. (b) Wollborn, U.; Leibfritz, D. *J. Magn. Reson.* **1992**, *98*, 142–146. *J*-HMBC: (c) Meissner, A.; Sørensen, O. W. *Magn. Reson. Chem.* **2001**, *39*, 49–52.

(8) (a) Karplus, M. *J. Chem. Phys.* **1959**, *30*, 11–15. (b) Haasnoot, C. A. G.; De Leeuw, F. A. A. M.; Altona, C. *Tetrahedron* **1980**, *36*, 2783–2792. (c) Karplus, M. *J. Phys. Chem.* **1960**, *64*, 1793–1798. (d) Karplus, M. *J. Am. Chem. Soc.* **1963**, *85*, 2870–2871.

(9) (a) Oikawa, M.; Adachi, S.; Kusumoto, S. *Org. Lett.* **2005**, *7*, 661–664.

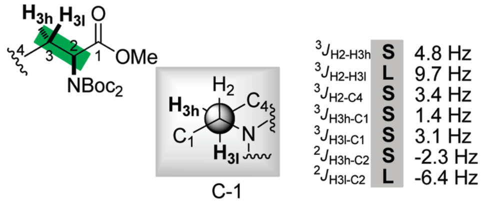
(10) $^3J_{HH}$: 8/12 Hz large; 0/4 Hz small. $^3J_{CH}$: 5/8 Hz large; 1−3 Hz small. $^2J_{CH}$: −4/−6 Hz large; 2/−2 Hz small. See ref 1.

(11) Contreras, R. H.; Peralta, J. E. *Prog. Nucl. Magn. Reson. Spectrosc.* **2000**, *37*, 321–425.

(12) In this paper, diastereotopic protons are labeled as Hh (proton at higher δ value) and Hl (proton at lower δ value).

therefore, relative configuration analysis cannot be achieved. Furthermore, a major drawback is found when the three rotamers belong to a fast multiple conformer equilibrium, and in this case, the methodology is unable to deduce the stereochemistry from the averaged NMR coupling constants. We have named the cases when a carbon−carbon framework cannot be deduced by the original Murata methodology as non-Murata models (Figure 2).

All of these cases can be visualized by considering compounds **1** and **3**, which were prepared by us during the synthesis of (−)-dysithiazolamide.[13] The presence of one major conformer around each of the C2−C3 and C3−C4 bonds (only *anti* conformers) in compound **1** allowed this to be classified as a Murata model.[4b] However, the medium/large proton−proton coupling constant values observed around the C2−C3 ($^3J_{H2-H3h}$ 8.4 and $^3J_{H2-H3l}$ 6.3 Hz) and C3−C4 ($^3J_{H4-H3h}$ 6.4 and $^3J_{H4-H3l}$ 6.5 Hz) bonds in *syn*-dichloroleucine derivative **2**, indicated the presence of a mixture of two conformers for each bond in rapid interconversion. This compound is therefore classified as a non-Murata model.[4d]



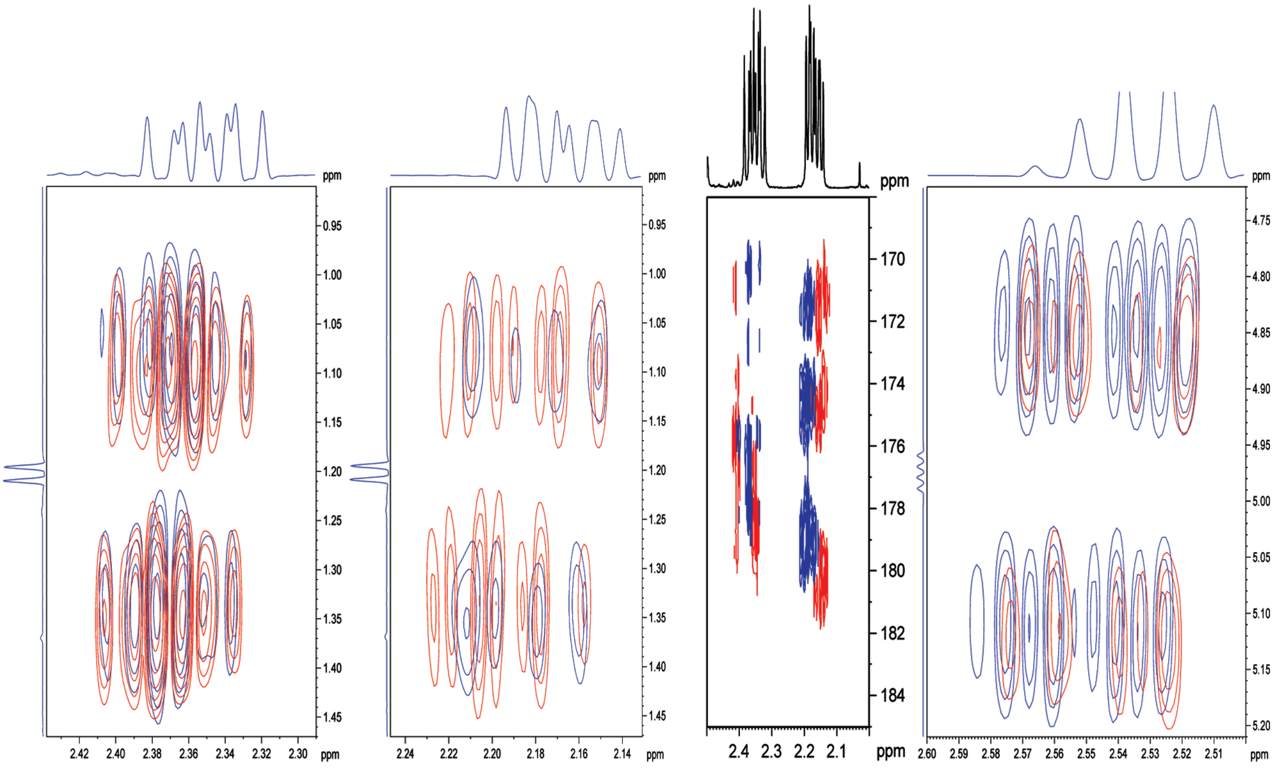
$^3J_{H2-H3h}$	S	4.8 Hz
$^3J_{H2-H3l}$	L	9.7 Hz
$^3J_{H2-C4}$	S	3.4 Hz
$^3J_{H3h-C1}$	S	1.4 Hz
$^3J_{H3l-C1}$	S	3.1 Hz
$^2J_{H3h-C2}$	S	-2.3 Hz
$^2J_{H3l-C2}$	L	-6.4 Hz

FIGURE 3. Major rotamer found for the C2−C3 bond in **3**.

The diester **3**,[14] used in the synthesis of *anti*-dichloroleucine derivative **1**, has two different situations around the first bond and a very complex conformational equilibrium around the C3−C4 bond. This compound is thus considered as a non-Murata model.

In order to solve this problem and similar ones, we describe here a new approach that involves the measurement of NMR proton−proton and carbon−proton coupling constants at lower temperatures. In complex conformer equilibrium systems, we have observed a variation in the coupling constant values from room to lower temperatures because of a shift in the relative population to the most stable staggered conformers. In this way, a pattern was found for the coupling constant values of a particular rotamer at lower temperature, with changes from the medium values observed in complex equilibrium cases.

Consequently, the key factor in this strategy was to study the *tendency of medium* (*M*) *coupling constant values observed*



a)
$^3J_{H3l-C6}$ 4.5 Hz
$^3J_{H3l-C6}$ 4.1 Hz

b)
$^3J_{H3h-C6}$ 4.4 Hz
$^3J_{H3h-C6}$ 4.0 Hz

c)
$^3J_{H3l-C5}$ 3.6Hz
$^3J_{H3l-C5}$ 2.0 Hz
$^3J_{H3h-C5}$ 5.5 Hz
$^3J_{H3h-C5}$ 6.3 Hz

d)
$^3J_{H2-C4}$ 3.5 Hz
$^3J_{H2-C4}$ 2.9 Hz

FIGURE 4. Heteronuclear coupling constant measurements at different temperatures for **3**: (a) HETLOC detail of $^3J_{H3l-C6}$ at 293 (blue) and 213 K (red); (b) HETLOC detail of $^3J_{H3h-C6}$ at 293 (blue) and 213 K (red); (c) *J*-HMBC detail of $^3J_{H3h-C5}$ and $^3J_{H3l-C5}$ at 293 K (blue) and 213 K (red); (d) HETLOC detail of $^3J_{H2-C4}$ at 293 K (blue) and 213 K (red). The signals in a), b), and d) are shifted for comparison.

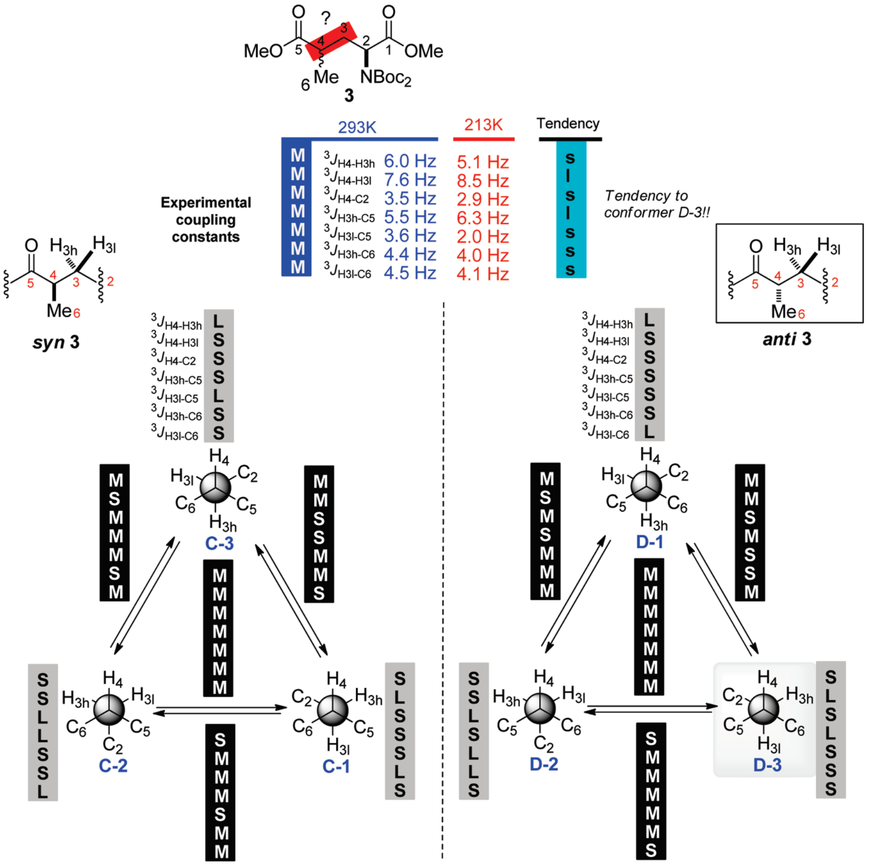


FIGURE 5. Comparison of coupling constant patterns and experimentally determined values for the C3–C4 bond in a non-Murata system (**3**): L (large), M (medium), S (small). Tendencies: l (larger), s (smaller).

at room temperature to become larger or smaller (*absolute value*) *at lower temperature*. In particular, the g^{+}/g^{-} equilibrated conformers shown in Figure 1 for both the *erythro* and *threo* series can be spectroscopically distinguished using this methodology by simply inspecting such a tendency.

For this purpose, we selected a non-Murata-model diester **3** to validate the method.[15] The C2–C3 bond in **3** behaves as a Murata system because the relative configuration of the nitrogenated substituent at C2 in relation to that of protons H_{3l} and H_{3h} was unquestionably defined from the set of coupling constant values found at room temperature (293 K), as shown in figure 3. The homonuclear coupling constants were measured in the ^{1}H NMR spectrum of **3** and $^{3}J_{\mathrm{H2-H3h}}$ 4.8 Hz (small) and $^{3}J_{\mathrm{H2-H3l}}$ 9.7 Hz (large) values were found. On the other hand, the heteronuclear coupling constants were measured by HETLOC and *J*-HMBC experiments and $^{3}J_{\mathrm{H2-C4}}$ 3.4 Hz (small), $^{3}J_{\mathrm{H3h-C1}}$ 1.4 Hz (small), $^{3}J_{\mathrm{H3l-C1}}$ 3.1 Hz (small), $^{2}J_{\mathrm{H3h-C2}}$ −2.3 Hz (small) and $^{2}J_{\mathrm{H3l-C2}}$ −6.4 Hz (large) values were obtained. These data indicate the presence of a major conformer (C-1 following Murata's work) where the C1 and C4 carbons display an *anti* disposition, as do the protons H_{3l} and H2 (see Figure 3).

However, the situation around C3–C4 in **3** was found to be more challenging because all of the $^{3}J_{\mathrm{H4-H3h}}$, $^{3}J_{\mathrm{H4-H3l}}$, $^{3}J_{\mathrm{H2-C4}}$, $^{3}J_{\mathrm{H3h-C5}}$, $^{3}J_{\mathrm{H3l-C5}}$, $^{3}J_{\mathrm{H3h-C6}}$, and $^{3}J_{\mathrm{H3l-C6}}$ coupling constants measured at room temperature (293 K) displayed medium values, which is indicative of the existence of a very complex equilibrium. When we ran the same *J*-HMBC and HETLOC experiments at lower temperature (213 K), a slight tendency toward a gauche-(+) rotamer (D-3) was observed. Now, the medium values of the aforementioned coupling constants became smaller, larger, smaller, larger, smaller, smaller, and smaller, respectively, as it can be seen from the red spots in Figure 4 and the data shown in Figure 5, where the values in red and the small letters display this tendency.

Once the conformer around the C3–C4 bond had been deduced from this tendency, the presence of a single rotamer around the C2–C3 bond and the stereorelationship to the diastereotopic methylene proton pair allowed us to calculate the relative configuration of this compound **3**.[14]

A similar situation around the C3–C4 bond is found in the *syn*-dichloroleucine **2** in relation to that of compound **3**. However, this case is simpler because only two conformers in equilibrium around C2–C3 were observed. For this reason, we decided to apply our new temperature-dependent *J*-based strategy to determine the major conformer around the C2–C3 bond in **2**. A pattern of MMMSLM values was found for $^{3}J_{\mathrm{H2-H3h}}$, $^{3}J_{\mathrm{H2-H3l}}$, $^{2}J_{\mathrm{H3h-C1}}$, $^{3}J_{\mathrm{H3l-C1}}$, $^{3}J_{\mathrm{H3h-C2}}$, and $^{3}J_{\mathrm{H3l-C2}}$ coupling constants at room temperature (298 K), indicating the existence of an *anti*/G^{+} conformer equilibrium around the bond in question (denoted as D-1+D-3 conformers in part a of Figure 6). Now, the *J*-HMBC and HETLOC NMR experiments at lower temperature

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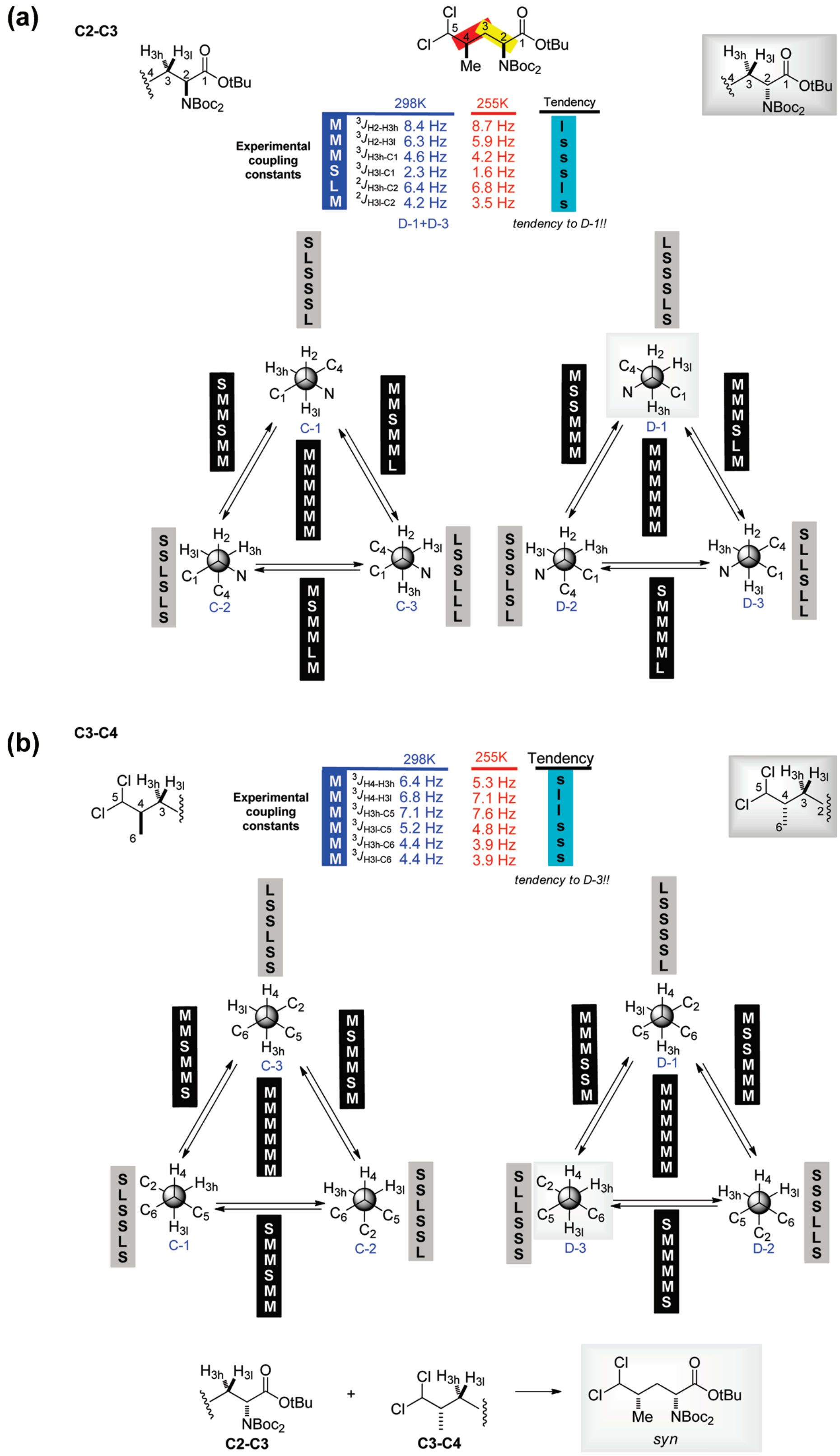
(13) Ardá, A.; Soengas, R. G.; Nieto, M. I.; Jimenez, C.; Rodríguez, J. *Org. Lett.* **2008**, *10*, 2175–2178.

(14) Hanessian, S.; Margarita, R. *Tetrahedron Lett.* **1998**, *39*, 5887–5890.

(15) Ardá, A.; Jiménez, C.; Rodríguez, J. *Tetrahedron Lett.* **2004**, *45*, 3241–3243.



(a) C2-C3

		298K	255K	Tendency
M	$^3J_{H2\text{-}H3h}$	8.4 Hz	8.7 Hz	l
M	$^3J_{H2\text{-}H3l}$	6.3 Hz	5.9 Hz	s
M	$^3J_{H3h\text{-}C1}$	4.6 Hz	4.2 Hz	s
S	$^3J_{H3l\text{-}C1}$	2.3 Hz	1.6 Hz	s
L	$^2J_{H3h\text{-}C2}$	6.4 Hz	6.8 Hz	l
M	$^2J_{H3l\text{-}C2}$	4.2 Hz	3.5 Hz	s

(b) C3-C4

		298K	255K	Tendency
M	$^3J_{H4\text{-}H3h}$	6.4 Hz	5.3 Hz	s
M	$^3J_{H4\text{-}H3l}$	6.8 Hz	7.1 Hz	l
M	$^3J_{H3h\text{-}C5}$	7.1 Hz	7.6 Hz	l
M	$^3J_{H3l\text{-}C5}$	5.2 Hz	4.8 Hz	s
M	$^3J_{H3h\text{-}C6}$	4.4 Hz	3.9 Hz	s
M	$^3J_{H3l\text{-}C6}$	4.4 Hz	3.9 Hz	s

FIGURE 6. (a) Comparison of coupling constant patterns and experimentally determined values in a 1,3-dimethine non-Murata system (C2−C3 bond): L (large), M (medium), S (small). Tendencies: l (larger), s (smaller). $^2J_{H3h-C2}$ and $^2J_{H3l-C2}$ in absolute values. (b) Comparison of coupling constant patterns and experimentally determined values in a 1,3-dimethine non-Murata system (C3−C4 bond): L (large), M (medium), S (small). Tendencies: l (larger), s (smaller). $^2J_{H3h-C2}$ and $^2J_{H3l-C2}$ in absolute values.

(255 K) for **2** showed a slight tendency toward a *population increase* in the D-1 conformer.

As a consequence of this, the pattern of MMMSLM values due to the D-1 and D-3 rotamer-equilibrium underwent a clear tendency toward the pattern of lsssls (larger, smaller, smaller, smaller, larger and smaller, see Supporting Information).

Once we had found the appropriate pattern of coupling constant values around the C2−C3 bond, the complex equilibrium around the C3−C4 bond was considered. The absence of a heteroatom linked to the chiral center at C4 meant that we only had to consider six three-bond coupling constants: $^3J_{\mathrm{H4-H3h}}$, $^3J_{\mathrm{H4-H3l}}$, $^3J_{\mathrm{H3h-C5}}$, $^3J_{\mathrm{H3l-C5}}$, $^3J_{\mathrm{H3h-C6}}$, and $^3J_{\mathrm{H3l-C6}}$. In this case, we could not identify any tendency in the $^3J_{\mathrm{H2-C4}}$ value when it was measured at 298 and 255 K, as the values were close to 0 Hz in both cases. All of the "*medium*" coupling constants as absolute values measured at room temperature (298 K) became smaller, larger, larger, smaller, smaller, and smaller, respectively, at lower temperature, indicating a clear tendency toward the labeled conformer D-3 (see part b of Figure 6). This *syn* stereorelationship between the methyl group at C4 and the di-*tert*-butoxycarbonylamino moiety at C2 are consistent with the known relative stereochemistry of this dichloroleucine derivative **2**.

Conclusions

We have found that measurement of the homonuclear and heteronuclear coupling constants at different temperatures can be applied to the resolution of complex conformer equilibria in flexible acyclic systems. This is achieved by considering the observed tendencies in these values in order to determine the relative configuration in 1,3- or adjacent 1,2-dimethine chiral systems. This new strategy, which we have called temperature-dependent *J*-based configurational analysis, can be applied where the existence of multiple conformer equilibrium precludes the application of the Murata methodology. This approach also allows spectroscopically indistinguishable gauche-(+)/gauche-(−) equilibria from a *threo* or *erythro* configuration to be solved by observing the tendency toward a unique rotamer.

Experimental Section

All NMR experiments were recorded on a 500 MHz spectrometer equipped with either a ^{1}H/^{13}C (5 mm) dual cryoprobe or a BBI inverse probehead incorporating a z-gradient coil. To measure heteronuclear coupling constants, the following general experimental conditions were applied. HETLOC: 4K data points in F_2, 512 or 1024 increments with 8 scans per F_1 increment. Delays were set to 1/2*J*(C,H) and 1/4*J*(C,H), respectively, with 40−60 ms isotropic mixing time. The data were zero filled to 4K in F_1, and a $\cos^2$ window function was applied to both dimensions before Fourier transformation. *J*-HMBC: 2K data points in F_2, 128 increments with 64 scans per F_1 increment. Scaleff around 190−230 were used to split $^{2,3}J$(C, H). Once acquired, the data were processed and zero-filled to 4K data points and a cosine window function was applied in both dimensions. For comparison of coupling constants at different temperatures, the same experimental conditions were used to determine *J* values.

Compounds **1** and **3** were synthesized using ref 13 and compound **2** following ref 4d.

Acknowledgment. This research was financially supported by a grant from the Ministerio de Educación y Ciencia (CTQ2008-04024/BQU and AGL2009-12266-C02-02). M.B. and M.I.N. thank the FPU (MEC) and Parga Pondal (Xunta de Galicia) programs, respectively, for support.

Supporting Information Available: NMR spectra at different temperatures (^{1}H, ^{13}C, HETLOC, *J*-HMBC) of compounds **2** and **3**. This material is available free of charge via the Internet at http://pubs.acs.org.