

Magnetic Anisotropy in a Dysprosium/DOTA Single-Molecule Magnet: Beyond Simple Magneto-Structural Correlations**

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Lanthanides are strategic materials in current technology^[1] and play a remarkable role in magnetism thanks to their large magnetic moments and large anisotropy.^[2] Molecular magnetism is not an exception to this trend, and lanthanides are becoming almost ubiquitous among single-molecule magnets (SMMs).^[3] In SMMs an anisotropy barrier opposes the reversal of the molecular giant spin at low temperature.^[4] In these conditions, individual molecules exhibit a memory effect, thus offering a new means for information storage and processing through single-spin manipulation.^[5]

Lanthanide-based SMMs have shown higher blocking temperatures and coercivity than transition-metal-based ones,^[6] as well as unusual behavior, such as the vortex arrangement of the magnetic moments in dysprosium(III) triangles.^[7] A fascinating result was the observation of SMM behavior in complexes comprising a single anisotropic lanthanide ion; this SMM behavior was first encountered in a terbium(III) bis(phthalocyaninato) complex.^[8] The phenomenon was associated with the presence of axially symmetric coordination environments to reduce the admixing of states on opposite sides of the barrier, which is responsible of fast quantum tunneling relaxation. This discovery stimulated the successful design of other single-ion SMMs based on polyoxymetalates,^[9] an organometallic double-decker structure,^[10] or exploiting a symmetric macrocycle.^[11] The crystallographic symmetry of the above-mentioned derivatives is however very low and direct experimental confirmation of the presence of uniaxial anisotropy is lacking.

The anion of 1,4,7,10-tetraazacyclododecane N,N',N'',N''' -tetraacetic acid, H_4 DOTA, is well known for the exceptional

stability of its adducts with lanthanides,^[12] and provides a coordination environment with the idealized tetragonal symmetry of a capped square antiprism (Figure 1).^[13] Four nitrogen atoms and the four oxygen atoms are on the corners of two parallel squares. The capping atom is an oxygen atom of a labile water molecule that is exploited to generate contrast in magnetic resonance imaging.^[14]

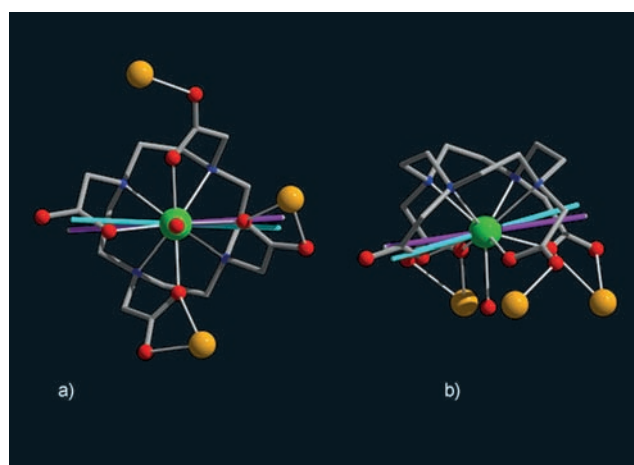


Figure 1. Two orthogonal views of the crystal structure of **1**: a) parallel to the pseudotetragonal axis, that is, the Dy–O_w bond; (b) orthogonal to the pseudotetragonal axis. Color scheme: Dy, green; Na, gold; O, red; N, blue; C, gray. The violet rod represents the orientation of the experimental easy axis of the magnetization, while the pale blue rod represents the calculated one.

We have recently investigated the magnetic properties of a polycrystalline sample of $[Na\{Dy(DOTA)(H_2O)\}]\cdot 4H_2O$ (**1**) and found that the compound behaves like a SMM, thus showing a giant field dependence on the relaxation time of the magnetization. In zero static field a temperature-independent underbarrier mechanism, strongly influenced by spin–spin interactions, is observed, while the application of a weak magnetic field induces a thermally activated regime with an effective barrier of ca. 60 K and an increase in the relaxation time, compared to zero static field, of six orders of magnitude at 1.8 K.^[15]

A closer look at the crystal structure of **1** reveals that three out of the four carboxylate arms are coordinated to the Na⁺ ions, which themselves are coordinated to two symmetry-related complexes (see Figure S1 in the Supporting Information). The axial water molecule forms hydrogen bonds to another water molecule and to a carboxylate oxygen, both of which are coordinated to Na⁺ ions.

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[**] This work was supported by the European Research Council through ERC-AdG “MolNanoMaS”. The stimulating discussions with L. Sorace, D. Gatteschi, and A. Cornia are gratefully acknowledged.

Supporting information for this article is available on the WWW under <http://dx.doi.org/10.1002/ange.201107453>.

Tetragonal symmetry is therefore limited to the first coordination sphere and an experimental investigation was necessary to avoid deceptive oversimplification. The complex crystallizes in a triclinic space group with all the molecules equally oriented in the crystal, therefore the magnetic anisotropy can be directly extracted from the angular dependence of the single-crystal magnetic susceptibility.^[16] Herein we report a magnetic single-crystal and luminescence characterization, supported by ab initio calculations, and thus reveal that simple analysis based on the idealized symmetry of the lanthanide complex can be misleading.

To investigate the angular dependence of the magnetization we employed a procedure we recently established that consists of gluing a crystal on the face of a millimetric Teflon cube.^[17] The three orthogonal faces of the cube are then indexed in the reference frame of the crystal with noninteger Miller indices, and the normals to these faces define the laboratory *xyz* reference frame (see the Supporting Information for details). The cube attached to the crystal is then mounted on a horizontal rotator and placed in a SQUID magnetometer with a vertical solenoid to generate the magnetic field.

The outcome of three isothermal (*T* = 1.8 K) rotations along *x*, *y*, and *z* respectively is reported in Figure 2. No significant shifts of the maxima and minima are seen on increasing the temperature (see Figure S2).

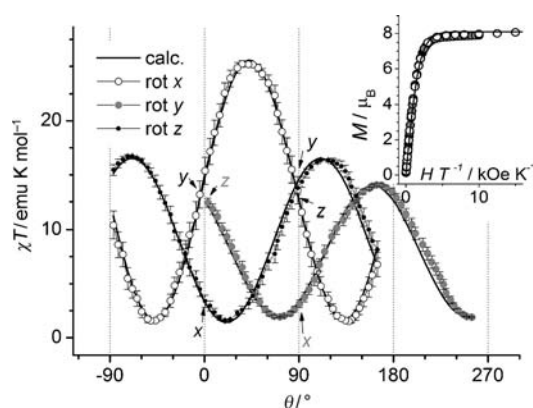


Figure 2. Angular dependence of the magnetic susceptibility of **1** multiplied by temperature measured at *T* = 1.8 K and *H* = 1.0 kOe for the rotation along the *x*, *y*, and *z* axes of the laboratory reference frame. The solid line represents the calculated values (see text). Inset: Field dependence of the magnetization of a single crystal along the easy axis at 2.0, 5.0, and 10.0 K plotted against the reduced variable *H/T*. The solid line corresponds to the Brillouin function for *S*_{eff} = 1/2 and *g*_{eff} = 16.2.

The data in Figure 2 suggests a strong magnetic anisotropy, which is not at all surprising as Dy³⁺ exhibits a strong splitting of the ⁶H_{15/2} ground configuration and often an Ising-type magnetic anisotropy. At low temperature the ground-state doublet is in fact frequently treated as an effective spin *S*_{eff} = 1/2 with gyromagnetic tensors *g*_{||} → 20 and *g*_⊥ → 0.^[18]

By exploiting the tensorial character of the susceptibility, and taking into account that we measure the component of

the magnetization along the applied field, the data in Figure 2 has been simulated according to [Eq. (1)],

$$\chi^{\text{rot}}(\theta) = \chi_{\alpha\alpha} \cos^2\theta + \chi_{\beta\beta} \sin^2\theta + 2\chi_{\alpha\beta} \cos\theta \sin\theta \quad (1)$$

with cyclic permutation of the *α*, *β*, and *γ* indices over the *xyz* reference frame and *θ* the angle the magnetic field forms with the *α* axis.

The susceptibility tensor (*χ*) has been diagonalized (see the Supporting Information), thus providing the principal values of the susceptibility and their orientation. The largest principal value multiplied by the temperature provide *χ*_{||}*T* = 27.2 emu K mol^{−1} corresponding to *g*_{||} = 17.0 for an effective spin *S*_{eff} = 1/2, thus confirming the Ising anisotropy of the Dy³⁺ ion in this coordination environment. A small rhombic anisotropy is observed for the other two components of *g* (see Table 1). These values should be considered as an upper limit because they are very sensitive to spurious angular independent contributions.

Table 1: Principal components of the *g* tensor assuming for the ground-state doublet *S*_{eff} = 1/2 and energy gap between the ground doublet and the first excited one.

Model	<i>g</i> ₁	<i>g</i> ₂	<i>g</i> ₃	Δ ₁ /cm ^{−1}
exp.	17.0(1)	4.8(1)	3.4(1)	53(8) ^[a]
A	18.6	0.9	0.2	64
B	19.7	< 0.1	< 0.1	120
C	18.7	1.33	0.44	20
D	18.1	2.18	0.55	34
A'	18.3	1.50	0.44	13

[a] From luminescence spectra. exp. = experimental.

In the inset of Figure 2 are shown the magnetization versus field curves for a crystal aligned with the magnetic field parallel to the easy axis. Interestingly, the magnetization data recorded at 2.0, 5.0, and 10 K are superimposable if plotted against the rescaled variable *H/T* and can be well simulated with the Brillouin function for a *S*_{eff} = 1/2 spin state with *g*_{eff} = 16.2 ± 0.1. This observation, at a first glance, is in contradiction with the presence of magnetic anisotropy, but indeed confirms that only the ground-state doublet of this highly anisotropic system is populated at these temperatures. A preliminary investigation of the magnetization dynamics substantially confirms the peculiar behavior of the polycrystalline powder sample (further investigations will be discussed in a separate publication).

We focus here on the orientation of the magnetic anisotropy tensor in respect to the coordination environment of the Dy³⁺ ion. We show in Figure 1 the easy axis, as a pale blue rod, superimposed on the molecular structure (see the Supporting Information for the complete tensor). It is evident that the easy axis is not linked to the idealized symmetry axis of the complex, but is almost perpendicular to it.

To investigate the origin of the unexpected orientation of the anisotropy tensor we have performed post Hartree–Fock ab initio calculations. The chosen method was the CASSCF/CASPT2-RASSI-SO^[19] as implemented in the quantum chemistry package MOLCAS 7.4^[20] (see the Supporting

Information for details). This approach has recently been demonstrated to be able to predict the magnetic anisotropy of Dy^{3+} ions in a low symmetry environment.^[7a,21] In our calculations we have investigated several finite clusters of atoms to model the material. The most accurate model (model A) includes all the atoms of the complex, the three coordinated sodium ions as well as other fragments to mimic the crystalline environment of the complex as shown in Figure 3. The effective g tensor of the ground-state doublet of the ${}^6\text{H}_{15/2}$ has also been evaluated (for more details see the Supporting Information).

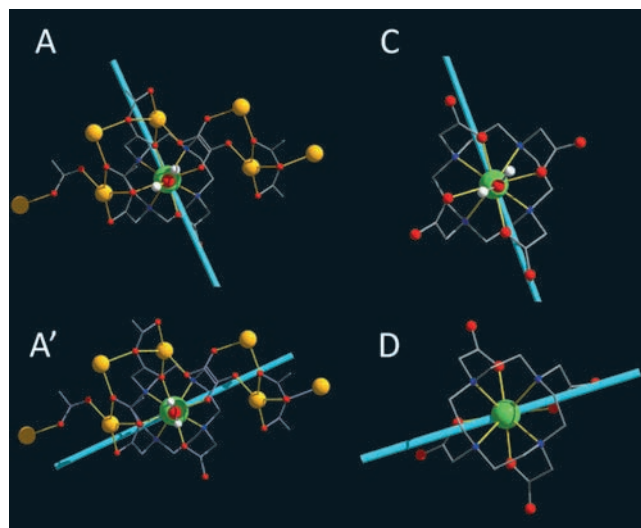


Figure 3. View of four molecular models (A, A', C, and D) employed for the ab initio calculations together with the evaluated easy axis of the magnetization (pale blue rod). Color code is the same as in Figure 1 with hydrogen atoms of coordinated water depicted as small white spheres.

As summarized in Table 1, the largest g component, $g_z = 18.6$, approaches the Ising limit, $g = 20$, and its orientation, shown in Figure 1 as a pale blue rod, is calculated to be very close (10°) to the experimental one, as deviations are comparable to experimental uncertainty. Our results suggest that complex **1** has no axial symmetry, in contrast with that deduced from DFT calculations.^[22] The gap between the ground-state and the first excited doublet is estimated as 64 cm^{-1} . To investigate which structural features play a key role in determining the unexpected orientation of the easy axis, ab initio calculations were performed on models where the number of atoms around the complex were progressively reduced. In model B the description of the complex surroundings is limited to the three coordinated Na^+ ions, and for this model the first energy gap of the ${}^6\text{H}_{15/2}$ manifold is significantly increased (see Table 1) but there is no change in the orientation of the easy axis (see the Supporting Information, Table S3 and Figure S4).

A similar orientation of the easy axis of the magnetization is obtained if the naked complex (model C) is considered to be as shown in Figure 3. When the apical water molecule is removed (model D) an abrupt change in the anisotropy

tensor is observed. The easy axis is now rotated by 90° but it is still orthogonal to the pseudotetragonal axis (see Figure 3). The coordinated water molecule seems to play a key role in the unusual orientation of the easy axis, therefore our model A was then modified by rotating by 90° the position of the water hydrogen atoms along the $\text{Dy}-\text{O}_w$ axis (model A'), although the hydrogen bonds hamper any rotation of the water molecule in the crystal. Interestingly in model A' the calculated easy axis of the magnetization rotates as well, as is clearly shown in Figure 3. A significant reduction of the first energy gap is also observed.

Owing to the unexpected large influence of the orientation of the apical water molecule on the magnetic anisotropy, calculations were repeated using different basis sets to exclude a spurious origin of the effect (see Table S4 of the Supporting Information). The same simplified model C has been used to limit computation time in the analysis of the orientation of the easy axis as a function of the angle of rotation of the water molecule. The reorientation is not gradual but occurs rather abruptly in correspondence to the level crossing between the ground-state and the first excited doublets (see the Supporting Information for more details).

Although the role of the second coordination sphere on the magnetic anisotropy was already theoretically evidenced for cobalt(II) ions,^[23] it is rather unexpected for three positive lanthanide ions. A possible explanation can be found in the partial charge transfer from the ligand atoms to the Dy 5d orbitals, evidenced by the ab initio calculations. The rotation of the water molecule could then affect the relative population of the Dy 5d orbitals through a π interaction with the oxygen atom.

A comparison of single-crystal magnetization data with ab initio calculations suggests that these calculations are an outstanding method to predict and rationalize the nature and orientation of the magnetic anisotropy, provided that structural details are correctly taken into account, while, less easy to assess is the ability to correctly quantify the crystal field level splitting using ab initio calculations. Up to now the comparison has been in fact limited to the estimation provided by the dynamics of the magnetization.^[7a,17] It would be therefore interesting to get an independent evaluation of the splitting of the ${}^6\text{H}_{15/2}$ manifold through electronic spectroscopy.

Lanthanide complexes with DOTA ligands are well known for their strong luminescence,^[24] but the solid-state electronic spectra have been reported only for the Europium isostructural complex.^[25] We therefore recorded the luminescence spectrum of **1**, which had an average quantum yield of 1.54%. Figure 4 shows the emission spectrum recorded at room temperature on a polycrystalline sample excited at 365 nm. The excitation spectrum is shown in the Supporting Information, Figure S5.

Well-resolved multiline emissions are observed around 20800, 17500, and 15200 cm^{-1} , and attributed to ${}^4\text{F}_{9/2} \rightarrow {}^6\text{H}_{15/2}$, ${}^4\text{F}_{9/2} \rightarrow {}^6\text{H}_{13/2}$, and ${}^4\text{F}_{9/2} \rightarrow {}^6\text{H}_{11/2}$ transitions, respectively (Figure 4).^[26] Energy gaps between the ground-state multiplet of ca. 3550 cm^{-1} and 6400 cm^{-1} were calculated using model A (see the Supporting Information, Table S3), and these energy gaps are in reasonable agreement with the observed spec-

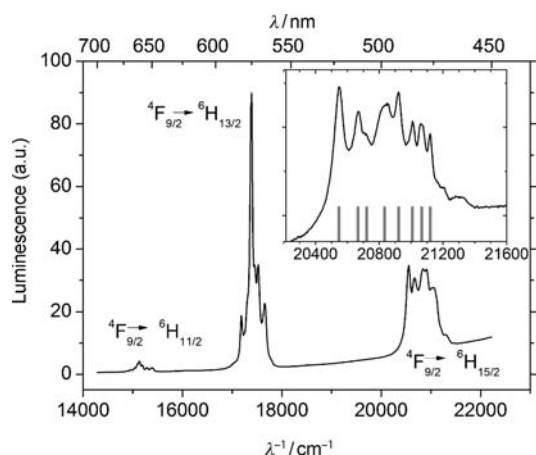


Figure 4. Luminescence spectrum of a polycrystalline powder sample of **1** excited at 365 nm at room temperature. In the inset, an enlarged view of the ${}^4F_{9/2} \rightarrow {}^6H_{15/2}$ emission. The gray bars correspond to the emission transitions extracted from the simulation of the spectrum (see the Supporting Information, Figure S8).

trum. The emission line involving the ground-state ${}^6H_{15/2}$ manifold shown in the inset of Figure 4 shows a well-resolved structure. The luminescence spectra are recorded at room temperature, thus “hot” bands, involving excited doublets of the ${}^4F_{9/2}$ manifold as the starting level, can also be present. The shape of the emission suggests however that the shoulders observed around 21 200 cm^{-1} and above are “hot” in nature, while the narrower peaks evidenced by the grey bars in Figure 4 have been assumed to be representative of the splitting of the ground ${}^6H_{15/2}$ state. From the spectrum simulation, using a convolution of Gaussian lines (see the Supporting Information, Figure S8), the separation between the two lines at highest energy is evaluated to be $53 \pm 8 \text{ cm}^{-1}$, which is slightly larger than the value of the activation barrier estimated from the dynamics of the magnetization, ca. 42 cm^{-1} . This observation suggests that the population of the first excited doublet is sufficient to reverse the magnetization. To the best of our knowledge the correlation between the luminescence spectra and the mechanism of relaxation of a SMM is unprecedented.

If we focus on the entire ${}^6H_{15/2}$ splitting (see Figure S4 in the Supporting Information), the observed experimental overall splitting of ca. 500 cm^{-1} is approximately reproduced by all models that include the apical water molecule, while the splitting is almost doubled for the octacoordinated complex (see the Supporting Information, Figure S4).

In conclusion, single-crystal magnetic investigations, which are still seldom used in molecular magnetism, have been shown to be required to study molecular systems that are based on anisotropic lanthanide ions. Our detailed experimental and theoretical investigation on one of the most studied lanthanide coordination compounds has provided several key results.

The most important one is that simple magneto-structural correlations based on the coordination environment can fail to predict, even approximately, the correct magnetic anisotropy of anisotropic lanthanide ions in a low-symmetry environment. Subtle structural details, like the position of

hydrogen atoms and the consequent orientation of the nonbonding orbitals of the axial ligand can overcome the symmetry imposed by the coordination polyhedron. We can also expect significant changes in the magnetic anisotropy once this type of complex is anchored to substrates, a procedure commonly employed on lanthanide/DOTA complexes to enhance their properties as contrast agents in MRI.^[27]

The well-resolved luminescence spectra, which allow the energy splitting of the ${}^6H_{15/2}$ multiplet to be determined, have provided precious indications, which confirm that the first excited doublet plays a key role in the magnetization dynamics, as there is a reasonable agreement between the estimated barrier and the first energy gap.

Finally, post Hartree–Fock ab initio calculations were confirmed to be an invaluable method for the prediction and rationalization of the magnetic anisotropy, provided that subtle structural details are also correctly taken into account. The evaluation of the energy splitting seems, however, even more critically dependent on which structural model and basis sets are used.

It is worth stressing that the relevance of our results is not limited to the field of molecular magnetism. Anisotropic lanthanide ions are currently investigated as contrast agents for high-field MRI.^[28] In modeling the relaxivity effects it is generally assumed that the orientation of the M–O_{water} of DOTA-like complexes corresponds to the anisotropy axis and that the apical water molecules lie in the cone of highest susceptibility. We have shown here that this is probably not the case and we also suspect that the mechanism of contrast in the nuclear relaxation is more complex because the labile water molecule modifies the magnetic anisotropy of the lanthanide ion.

Experimental Section

Single crystals of **1** were prepared as previously described.^[12,13] An Oxford Diffraction Xcalibur3 was employed to index the faces of the Teflon cube with a single crystal of **1** glued on it. The angular dependence of the magnetization was measured with a Quantum Design horizontal sample rotator adapted to a Cryogenic S600 SQUID magnetometer, while magnetic studies on the oriented single crystal were performed on a Quantum Design MPMS magnetometer. Luminescence measurements were collected using Horiba-Jobin Yvon Fluorolog III spectrofluorometer with a F-3018 integrating sphere at 294 K, between 450 and 700 nm under UV irradiation at 365 nm. Ab initio calculations have been performed using MOLCAS.7.4 on IBM SP6 supercomputer facility at the Italian computing center CINECA.

Received: October 22, 2011

Published online: January 12, 2012

Keywords: Ab initio calculations · dysprosium · lanthanide · luminescence · magnetic properties

[1] R. Stone, *Science* **2009**, 325, 1336–1337.

[2] J. C. G. Bünzli, C. Piguet, *Chem. Rev.* **2002**, 102, 1897–1928.

[3] R. Sessoli, A. K. Powell, *Coord. Chem. Rev.* **2009**, 253, 2328–2341.

- [4] D. Gatteschi, R. Sessoli, J. Villain, *Molecular nanomagnets*, Oxford University Press, Oxford, **2006**.
- [5] L. Bogani, W. Wernsdorfer, *Nat. Mater.* **2008**, *7*, 179–186.
- [6] a) J. D. Rinehart, M. Fang, W. J. Evans, J. R. Long, *Nat. Chem.* **2011**, *3*, 538–542; b) J. D. Rinehart, M. Fang, W. J. Evans, J. R. Long, *J. Am. Chem. Soc.* **2011**, *133*, 14236–14239; c) R. J. Blagg, F. Tuna, E. J. L. McInnes, R. E. P. Winpenny, *Chem. Commun.* **2011**, *47*, 10587–10589; d) R. J. Blagg, C. A. Muryn, E. J. L. McInnes, F. Tuna, R. E. P. Winpenny, *Angew. Chem. Int. Ed.* **2011**, *50*, 6530–6533.
- [7] a) L. F. Chibotaru, L. Ungur, A. Soncini, *Angew. Chem.* **2008**, *120*, 4194–4197; *Angew. Chem. Int. Ed.* **2008**, *47*, 4126–4129; b) J. Luzon, K. Bernot, I. J. Hewitt, C. E. Anson, A. K. Powell, R. Sessoli, *Phys. Rev. Lett.* **2008**, *100*, 247205.
- [8] N. Ishikawa, M. Sugita, T. Ishikawa, S. Koshihara, Y. Kaizu, *J. Am. Chem. Soc.* **2003**, *125*, 8694–8695.
- [9] M. A. Aldamen, J. M. Clemente-Juan, E. Coronado, C. Marti-Gastaldo, A. Gaita-Arino, *J. Am. Chem. Soc.* **2008**, *130*, 8874–8875.
- [10] B. W. Wang, S. J. Jiang, H. L. Sun, Z. M. Wang, S. Gao, *J. Am. Chem. Soc.* **2011**, *133*, 4730–4733.
- [11] H. L. C. Feltham, Y. Lan, F. Kloewer, L. Ungur, L. F. Chibotaru, A. K. Powell, S. Brooker, *Chem. Eur. J.* **2011**, *17*, 4362–4365.
- [12] J. F. Desreux, *Inorg. Chem.* **1980**, *19*, 1319–1324.
- [13] a) F. Benetollo, G. Bombieri, S. Aime, M. Botta, *Acta Crystallogr. Sect. C* **1999**, *55*, 353–356; b) N. Viola-Villegas, R. P. Doyle, *Coord. Chem. Rev.* **2009**, *253*, 1906–1925.
- [14] M. Bottrill, L. Kwok, N. J. Long, *Chem. Soc. Rev.* **2006**, *35*, 557–571.
- [15] R. Sessoli, P. E. Car, M. Perfetti, M. Mannini, A. Favre, A. Caneschi, *Chem. Commun.* **2011**, *47*, 3751–3753.
- [16] S. Mitra, *Prog. Inorg. Chem.* **1977**, *22*, 309–408.
- [17] I. J. Hewitt, J. Tang, N. T. Madhu, C. E. Anson, Y. Lan, J. Luzon, M. Etienne, R. Sessoli, A. K. Powell, *Angew. Chem.* **2010**, *122*, 6496–6500; *Angew. Chem. Int. Ed.* **2010**, *49*, 6352–6356.
- [18] A. Abragam, B. Bleaney, *Electron Paramagnetic Resonance of Transition Ions*, Dover, New York, **1986**.
- [19] P. A. Malmqvist, B. O. Roos, B. Schimmelpfennig, *Chem. Phys. Lett.* **2002**, *357*, 230–240.
- [20] a) J. A. Duncan, *J. Am. Chem. Soc.* **2009**, *131*, 2416–2416; b) G. Karlstrom, R. Lindh, P. A. Malmqvist, B. O. Roos, U. Ryde, V. Veryazov, P. O. Widmark, M. Cossi, B. Schimmelpfennig, P. Neogady, L. Seijo, *Comput. Mater. Sci.* **2003**, *28*, 222–239.
- [21] K. Bernot, J. Luzon, L. Bogani, M. Etienne, C. Sangregorio, M. Shanmugam, A. Caneschi, R. Sessoli, D. Gatteschi, *J. Am. Chem. Soc.* **2009**, *131*, 5573–5579.
- [22] L. Smentek, B. A. Hess, J. P. Cross, H. C. Manning, D. J. Bornhop, *J. Chem. Phys.* **2005**, *123*, 244302.
- [23] S. Petit, G. Pilet, D. Luneau, L. Chibotaru, L. Ungur, *Dalton Trans.* **2007**, 4582–4588.
- [24] L. Armelao, S. Quici, F. Barigelletti, G. Accorsi, G. Bottaro, M. Cavazzini, E. Tondello, *Coord. Chem. Rev.* **2010**, *254*, 487–505.
- [25] J. G. Kang, M. K. Na, S. K. Yoon, Y. Sohn, Y. D. Kim, I. H. Suh, *Inorg. Chim. Acta* **2000**, *310*, 56–64.
- [26] W. T. Carnall, P. R. Fields, K. Rajnak, *J. Chem. Phys.* **1968**, *49*, 4424–4442.
- [27] F. Carniato, L. Tei, M. Cossi, L. Marchese, M. Botta, *Chem. Eur. J.* **2010**, *16*, 10727–10734.
- [28] S. Viswanathan, Z. Kovacs, K. N. Green, S. J. Ratnakar, A. D. Sherry, *Chem. Rev.* **2010**, *110*, 2960–3018.