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1,1′-Dimethylvanadocene *α*-amino acid complexes: synthesis, characterization and antimicrobial behavior toward *Escherichia coli B*

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Eight water-soluble 1,1′-dimethylvanadocene amino acid complexes have been prepared via the reaction of $(MeCp)_2VCl_2$ (2) with one equivalent of amino acid (aa) in water affording $[(MeCp)_2V(aa)]Cl$, where aa is glycine (3), L-alanine (4), L-valine (5), L-leucine (6), L-isoleucine (7), L-phenylalanine (8), L-histidine (9) and L-tryptophane (10). All prepared complexes have been characterized by EPR, IR and Raman spectroscopy, elemental analysis and mass spectrometry. Molecular structures of $[(MeCp)_2V(ala)]BPh_4 \cdot CH_3OH$ (11), $[(MeCp)_2V(leu)]PF_6$ (12) and $[(MeCp)_2V(ile)]PF_6$ (13) were determined by X-ray diffraction analysis. Cytotoxic properties of complexes 2–10 were investigated toward *Escherichia coli B* and compared with analogical unsubstituted vanadocene compounds (1, 14–21). The results showed that 1,1′-dimethylvanadocene amino acid complexes have identical or slightly higher antiproliferative activity then their unsubstituted analogs. Copyright © 2006 John Wiley & Sons, Ltd.

KEYWORDS: bent metallocene; antitumor agents; 1,1′-dimethylvanadocene dichloride; X-ray structural analysis; EPR spectroscopy; *Escherichia coli*

INTRODUCTION

The metallocene dichlorides (Cp_2MCl_2, Cp = cyclopentadienyl, M = Ti, V, Mo, Nb, Zr, Hf) are known as ''bent-sandwich'' complexes, in which two cyclopentadienyl rings contain η^5 bonded and positioned in a bent conformation with respect to the central metal atom.[1] Some of these metallocenes containing transition metal in oxidation state IV+ exhibit antitumor activity both *in vitro* and *in vivo*.[2] In contrast to the cisplatin, their adverse side effects are markedly lower[3–5] and neutral dihalido complexes, e.g. Cp_2TiCl_2 and Cp_2VCl_2 (1), have emerged as promising alternatives to cisplatin.[6] Biological experiments have shown that both the high solubility and the stability of the bent $[Cp_2M]^{2+}$ fragment in aqueous solution are limiting factors in the application of such medicines.

Derivatives of Cp_2MX_2 type can be prepared either by substitution of the chloride ligands with X[7,8] or through modification of the cyclopentadienyl ring.[9–11] It seems that replacement of chloride ligands with biogenic substituents (e.g. amino acid or carboxylic acid) is more suitable due to the physiological acceptability of the compound.[12–15]

Functionalization of the cyclopentadienyl ring with electron-donating alkyl groups generally diminishes the cytotoxicity of the drug, probably by lowering its solubility in aqueous media. However, recently published works have shown that some ring-substituted metallocenes have comparable or higher cytotoxic activity than corresponding unsubstituted analogs.[16–19]

In this paper we report the preparation, structural characterization and antimicrobial study of 1,1′-dimethylvanadocene complexes with biogenic *α*-amino acids.

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RESULTS AND DISCUSSION

Preparation of compounds

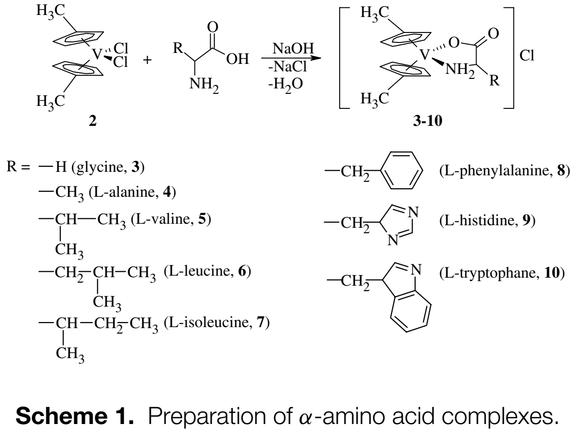
The 1,1′-dimethylvanadocene amino acid complexes [MeVD(**aa**)]Cl (**aa** = glycine **3**, L-alanine **4**, L-valine **5**, L-leucine **6**, L-isoleucine **7**, L-phenylalanine **8**, L-histidine **9**, L-tryptophane **10**) were prepared by the reaction of 1,1′-dimethylvanadocene dichloride (MeVDC, **2**) with appropriate amino acid in aqueous solution at neutral pH; see Scheme 1. The eight new complexes (**3–10)** were isolated in reasonable yields after evaporating the solvent from the reaction mixture. These compounds are extremely soluble in polar solvents such is H_2O or CH_3OH and almost insoluble in non-polar organic solvents. Attempts to grow single crystals of very soluble **3–10** were unsuccessful, but suitable crystals of corresponding large anion (PF_6^- or BPh_4^-) salts **11–13** were prepared and analyzed by X-ray diffraction.

Spectroscopic measurements

Both infrared and Raman spectra of compounds **3–10** have similar patterns (see Table 1), with intensive bands characteristic for the presence of the η^5-bonded MeCp ring. The strong Raman band at 296 ± 4 cm^{-1} of the ring tilting vibration, κ(MeCp), indicates that bent $(\eta^5\text{-MeCp})_2V^{2+}$ unit has remained unchanged in all prepared complexes.[20,21]

The presence of amino acid in the molecule is evident from ν_{as}(COO) stretching mode at 1634 ± 1 cm^{-1}; observed values are slightly shifted to higher wavelengths (20–30 cm^{-1}) when compared with those in corresponding free α-amino acids.[22] Similar values were previously reported for complexes with α-amino acids chelate-bonded to Cp_2V^{2+} moiety.[12,23] A symmetric stretching mode ν_s(COO) band can be expected[22] at wavelengths 1350–1390 cm^{-1}, but we observed more than one IR band in this region and therefore the assigning of band to ν_s(COO) mode was not accomplished.

EPR spectroscopy appeared to be a very efficient method for investigation of such paramagnetic complexes. The paramagnetism of the central metal atom d^1 V(IV) and its nuclear spin (^{51}V: I = 7/2, abundance 99.8%) make vanadocene(IV) complexes suitable as a EPR structural probe.



Scheme 1. Preparation of α-amino acid complexes.

The EPR spectra contain information about the coordination environment of the central atom. In particular, the isotropic hyperfine coupling (HFC) constant bears information about the delocalization of unpaired electron between the central atom and ligands.[24]

The EPR spectra of all amino acids complexes **3–13** (prepared as described above) dissolved in methanol give a simple eight-line pattern indicating the presence of only one paramagnetic species and observed HFC tensors are in a very narrow range of $62.0 \pm 0.3 \times 10^{-4}$ cm^{-1}. Therefore, we can presume that in **3–13** the same type of bonding occurs. Furthermore, the structure of complexes **11–13** determined by X-ray analysis unambiguously demonstrated the presence of a chelate-type bond. Based on similarity of spectroscopic data for all studied compounds, we can assume the same type of amino acid bonding to the $[(MeCp)_2V]^{2+}$ fragment.

Attempts to prepare bis(amino acid) complexes were unsuccessful even in the presence of the large excess (more than 10 : 1) of amino acid. The EPR spectra measured immediately after neutralization as well as the EPR spectra measured after dissolving crude reaction products in methanol correspond to complexes **3–10**.

Mass spectrometry

The base peak of the first-order positive-ion ESI mass spectra of all studied compounds is the [M-Cl]$^+$ ion. Tandem mass spectra of [M-Cl]$^+$ also provide common features, because the $[(MeCp)_2V]^+$ ion at m/z 208 is the base peak or the second most abundant peak in all cases. The fragmentation behaviour is in accordance with the expected structures.

X-ray analysis

Molecular structures of compounds **11–13** are shown in Figs 1–3. Table 2 summarizes important structural parameters. The cationic parts of **11–13** have a typical bent metallocenes structure in which nitrogen and oxygen atoms of amino acid and two η^5-bonded methylcyclopentadienyl rings occupy the pseudotetrahedral coordination sites around the vanadium(IV) centre. The V-Cg distances (1.9663–1.9809 Å) and Cg-V-Cg angles (132.92–134.01°) are comparable to those reported for other 1,1′-dimethylvanadocene complexes.[14,25] The methylcyclopentadienyl rings have the staggered conformation with the methyl groups positioned outside the space segment defined by N–V–O angle. The unit cell of complex **12** includes two crystallographically independent, but essentially identical molecules **12A** and **12B** (see Table 2).

The V–O and V–N bond distances of 1.9917–2.0092 and 2.1406–2.1783 Å, respectively, are close to corresponding ones in unsubstituted complexes $[Cp_2V(val)][PF_6]$ (V–O 2.004 Å, V–N 2.148 Å) and $[Cp_2V(ile)][PF_6]$ (V–O 2.010 Å, V–N 2.142 Å).[12] Both hydrogen atoms of the -NH_2 group are involved inintermolecular contacts in all crystals, one forming a hydrogen bond with oxygen of carbonyl moiety [**11**, N(1)···O(2) 3.015(2) Å; **12A**, N(1)···O(2) 2.917(5) Å; **12B**,

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Table 1. Selected IR and Raman data for complexes **3–10**

	ν(C–H) Cp		ν(C–H) CH_3		ν(C=O) aa	ν(C–C) Cp		δ(C–H) CH_3	δ(C–C) Cp	δ(C–H) Cp	γ(C–H) Cp	κ(MeCp) Cp
	IR	RA	IR	RA	IR	IR	RA	IR	IR	IR	IR	RA
3	3087	3120 3097	2964	2930	1634	1495	1505	1454	1261	1029	860	298
4	3087	3112 3102	2965	2929	1634	1496	n.o.	1456	1262	1029	859	289
5	3099	3112 3096	2963	2924	1635	1500	n.o.	1463	1261	1034	856	296
6	3096	n.o.	2958	n.o.	1635	1494	n.o.	1454	1260	1031	858	n.o.
7	3086	3102	2966	2927	1635	1499	1501	1456	1260	1031	862	296
8	3092	n.o.	2965	n.o.	1635	1496	n.o.	1455	1262	1029	860	n.o.
9	n.o.	n.o.	2967	n.o.	1634	1495	n.o.	1456	1265	1025	851	n.o.
10	3099	3112 3094	2964	2923	1634	1495	1503	1455	1262	1031	854	289

n.o., Not observed.

Table 2. Selected bond lengths (Å) and angles (deg) of **11–13**

	11	**12A**	**12B**	**13**
V–Cg1	1.9663(9)	1.9764(11)	1.9759(11)	1.9810(11)
V–Cg2	1.9806(8)	1.9736(11)	1.9633(9)	1.9670(11)
Cg1–V–Cg2	133.35(4)	132.93(5)	132.92(5)	134.03(4)
V–N1	2.1458(15)	2.1783(18)	2.1602(19)	2.1403(14)
V–O1	2.0092(12)	1.9917(15)	2.0081(16)	2.0007(13)
N1–V–O1	77.38(5)	78.01(6)	77.2(7)	76.98(5)

Cg1 = centroid of ring C1–C5; Cg2 = centroid of ring C6–C10.

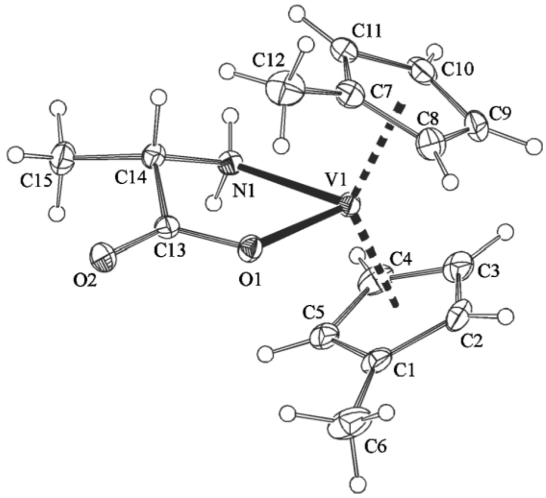

Figure 1. ORTEP drawing of cationic part of **11** with atom numbering scheme (ellipsoids: 30% probability).

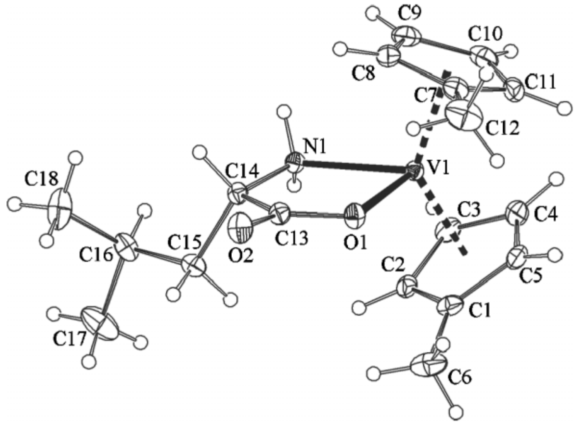

Figure 2. ORTEP drawing of cationic part of **12** with atom numbering scheme (ellipsoids: 30% probability).

N(1)···O(2) 2.826(5) Å; **13**, N(1)···O(2) 2.772(5) Å]. The second hydrogen is connected either to the solvent [**11**, N1···O(3) 2.836(2) Å or to fluorine atom of PF_6^- [**12A**, N(1)···F(2) 3.059(2); **12B**, N(2)···F(7) 3.166(3); **13**, N(1)···F(4) 3.168(2) Å].

Biological assays

The finding of filamentous growth of *Escherichia coli* detected for the first time the biological activity of cisplatin and led to the discovery its antitumor properties against various tumors.[26] Since this time, the elongation of this germ has interpreted as the possible evidence of antiproliferative activity of tested compounds.[27]

Table 3 summarizes the results of microbiological studies performed with *Escherichia coli B*. Maximum effective concentrations inducing the largest count of filaments and the maximum elongation of the filament at this concentration are listed. The most effective concentrations of all tested complexes (**3–10** and **14–21**) were in the range 0.4–3.2 mM. When the observed activity was compared with the starting

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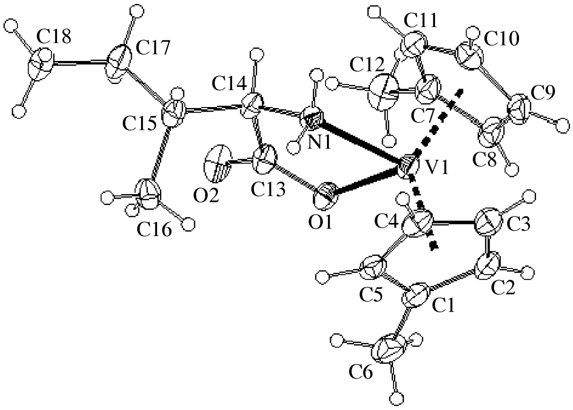

Figure 3. ORTEP drawing of cationic part of **13** with atom numbering scheme (ellipsoids: 30% probability).

Table 3. Cytotoxic activity of compounds **1–10**, **14–21** toward *E. coli B*

	$c_{ef.}$ (mM)	Maximum observed elongation		$c_{ef.}$ (mM)	Maximum observed elongation
2	0.8	30×	**1**	0.8	50×
3	0.8	15×	**14**	1.6	20×
4	0.8	50×	**15**	1.6	75×
5	1.6	30×	**16**	1.6	30×
6	0.8	75×	**17**	0.8	70×
7	0.8	50×	**18**	3.2	25×
8	0.4	60×	**19**	0.8	50×
9	3.2	30×	**20**	1.6	50×
10	1.6	25×	**21**	0.4	50×

c_{ef}, the most effective concentration evoking cell elongation.

compounds (VDC **1** or MeVDC **2**, $c_{ef.} = 0.8$ mM), it is evident that neither the replacing of chloride anion with amino acid nor the methyl substitution at Cp ring leads to significant decrease in biological activity.

Complexes **3, 4, 7** and **8** embody higher antiproliferative activity than their non-methylated analogs. The aforementioned compounds are more efficient at lower concentrations and induce similar elongation of cells. Complexes with histidine (**9**) and tryptophane (**10**) are slightly less active then their vanadocene analogs (**20, 21**).

CONCLUSIONS

Eight new 1,1′-dimethylvanadocene amino acid complexes were prepared and characterized by elemental analysis, mass spectrometry and spectroscopic measurements. The molecular structures of three studied compounds (**11–13**) were determined by X-ray analysis, confirming the chelate-type bonding of amino acid to $[(MeCp)_2V]^{2+}$ fragment realized via the nitrogen of the amino group and the oxygen atom of the carboxylic group. Based on the similarity of all spectroscopic data for complexes **3–13**, we suppose that identical bonding occurs in all prepared compounds.

The cytotoxic behaviour of 1,1′-dimethylvanadocene amino acid complexes (**3–10**) and their unsubstituted analogs (**14–21**) toward *Escherichia coli B* was studied. All of the complexes show biological activity and evoke significant cell elongation. The obtained results confirm that methylation of Cp rings does not lead to a decrease in bioactivity. Most of the prepared amino acid complexes are slightly more efficient when compared with non-methylated analogs or the starting MeVDC. Effective concentrations for all complexes occurred in the range 0.4–3.2 mM.

The presence of a chelate bond in the molecule of amino acid vanadocene complexes stabilizes these compounds in aqueous solution at physiological pH while keeping their biological activity. Therefore the studied substances should be interesting drugs for *in vitro* and *in vivo* tests on human tumors.

EXPERIMENTAL SECTION

Methods and materials

All operations were performed under argon using conventional Schlenk-line techniques. The solvents were purified and deoxygenated by standard methods. Water was deionized, double-distilled and saturated with argon. Carbonate-free sodium hydroxide was prepared by slow dissolution of sodium in water under argon atmosphere. Glycine, L-alanine, L-valine, L-leucine, L-isoleucine, L-phenylalanine, L-histidine and L-trptophane were obtained commercially (Fluka) and used without further purification. Vanadocene dichloride (**1**), 1,1′dimethylvanadocene dichloride (**2**), and unsubstituted complexes $[Cp_2V(\mathbf{aa})]Cl$ (**14–21**) were prepared using the published methods.[25,28,29] Elemental analysis, EPR, IR, Raman spectroscopy and mass spectrometry checked identity and purity of all prepared compounds.

The sterile M9 broth was prepared according to the modified published procedure (15.40 g $Na_2HPO_4 \cdot 12\ H_2O$, 3.00 g KH_2PO_4, 5.00 g NaCl, 1.00 g NH_4Cl, 0.25 g $MgSO_4 \cdot 7H_2O$, 0.11 g $CaCl_2$, 8.00 g glucose and 10.00 g casamino acids as carbon and amino source per litre of distilled water; pH = 7.4).[30]

Measurements

IR spectra were recorded in the 4000–350 cm^{-1} region on a Perkin-Elmer 684 as KBr pellets. Raman spectra of solid samples were recorded on a Bruker IFS 55s with extension FRA 106 at 50–3500 cm^{-1} in quartz capillaries.

EPR spectra were measured on ERS 221 (ZWG Berlin) apparatus in microwave X-band (~9.5 GHz). The apparatus was calibrated with DPPH value $g = 2.0036 \pm 2$. Solution samples were measured in flat quartz cuvettes (width 0.3 mm) at 300 K.

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Positive-ion electrospray ionisation (ESI) mass spectra were measured on an Esquire 3000 ion trap analyzer (Bruker Daltonics, Bremen, Germany) in the range m/z 50–1000. The samples were dissolved in methanol and analyzed by direct infusion at a flow rate of 1 µl/ min. The selected precursor ions were further analyzed by MS/MS analyses under the following conditions: isolation width $m/z = 4$, collision amplitude 0.9 V, ion source temperature 300 °C, flow rate and pressure of nitrogen 4 l/min and 10 psi, respectively.

Antimicrobial studies

Escherichia coli strain B was used for all microbiological experiments. The bacteria were grown aerobically on a blood agar at 37 °C/24 h. From this culture the bacterial suspension according to McFarland (second degree) in an M9 broth was prepared and subsequently incubated for 18 h at 37 °C.

The 12.8 mM solution of bactericide was prepared by dissolving appropriate amounts of **1–10** and **14–21** in 4 ml M9. Subsequently, this solution was diluted with sterile medium M9 to a concentration of 12.8–0.1 mM). The each tube of concentration line were inoculated with 100 µl of bacterial suspension and incubated (37 °C/24 h/mixing). After incubation, native preparations from each tube were prepared and phase-contrast microscoped at a magnification of 1000.

The relative percentage of filamentous cells and maximal observed cell elongation was determined. The relative length of the filaments was estimated by comparison with a control sample of normal size cells.

Syntheses of compounds

Complexes (**3–10**) were prepared in the following way: 1,1′-dimethylvanadocene dichloride (0.49 g, 1.75 mmol) was dissolved in 20 ml water. After dissolving, an equimolar amount of appropriate amino acid (glycine, L-alanine, L-valine, L-leucine, L-isoleucine, L-phenylalanine, L-histidine or L-tryptophane) was gradually added. The solution was stirred for 10 min and neutralized with 3.5 ml NaOH ($c = 0.5$ mol l^{-1}). After evaporation of the solvent in vacuum, a green solid residue was crystallized from the acetone-methanol mixture.

*[(MeCp)$_2$V(gly)]Cl (**3**)*

Yield: 0.48 g (1.5 mmol, 86%). Calculated for $C_{14}H_{18}ClNO_2V$ (MW 318.6): C, 52.8; H, 5.7; N, 4.4; Cl, 11.1. Anal. found: C, 52.7; H, 5.9; N, 4.4; Cl, 11.0. EPR (CH_3OH solution): $A_{iso} = 66.9$ G, $g_{iso} = 1.986$. IR (KBr, cm^{-1}): 3431s, 3243w, 3087s, 2964m, 1634vs, 1495m, 1454m, 1379m, 1349m, 1284m, 1261s, 1098vs, 1029s, 935w, 921w, 860m, 803s, 666w, 603w, 555m, 479w, 400m. Raman: 3131 (3), 3120(2), 3097 (3), 3071 (4), 2970 (2), 2930 (5), 2907 (1), 2077 (<1), 1505(1), 1410 (1), 1300 (1), 1243 (2), 1074 (2), 633 (<1), 441 (1), 418 (1), 396 (1), 373 (1), 337 (1), 298 (10), 265 (1), 183 (5). Positive-ion MS: m/z 601 [2M-Cl]$^+$; 283 [M-Cl]$^+$ (100%). Positive-ion MS/MS of 283: m/z 265; 226 [M-Cl-CO_2CH]; 208 [$MeCp_2V$]$^+$ (100%).

*[(MeCp)$_2$V(ala)]Cl (**4**)*

Yield: 0.53 g (1.59 mmol, 91%). Calculated for $C_{15}H_{19}ClNO_2V$ (MW 332.7): C, 54.1; H, 5.8; N, 4.2; Cl, 10.7. Anal. found: C, 54.1; H, 5.7; N, 4.3; Cl, 11.6. EPR (CH_3OH solution): $A_{iso} = 67.4$ G, $g_{iso} = 1.986$. IR (KBr, cm^{-1}): 3431vs, 3244w, 3087s, 2965m, 1634vs, 1496m, 1456m, 1378m, 1348m, 1284m, 1262s, 1098vs, 1029s, 983w, 934w, 921w, 859m, 802s, 666w, 604w, 555m, 478w, 399w. Raman: 3112 (7), 3102 (7), 3019 (4), 2960 (1), 2929 (5), 2902 (5), 2397 (5), 2244 (2), 1808 (7), 1403 (5), 606 (<1), 540 (<1), 289 (10), 222 (3). Positive-ion MS: m/z 629 [2M-Cl]$^+$; 297 [M-Cl]$^+$ (100%). Positive-ion MS/MS of 297: m/z 279 [M-H_2O]; 226 [$(MeCp)_2V + H_2O$]$^+$; 208 [$(MeCp)_2V$]$^+$ (100%).

*[(MeCp)$_2$V(val)]Cl (**5**)*

Yield: 0.52 g (1.45 mmol, 83%). Calculated for $C_{17}H_{24}ClNO_2V$ (MW 360.7): C, 56.6; H, 6.7; N, 3.9; Cl, 9.8. Anal. found: C, 56.5; H, 6.5; N, 4.1; Cl, 10.0. EPR (CH_3OH solution): $A_{iso} = 66.9$ G, $g_{iso} = 1.986$. IR (KBr, cm^{-1}): 3423vs, 3237w, 3099m, 3076w, 2963m, 2937w, 2630w, 2096w, 1645w, 1635vs, 1589w, 1538w, 1500s, 1463m, 1396m, 1381m, 1357w, 1329w, 1312m, 1261w, 1233w, 1190w, 1137vw, 1123vw, 1095w, 1034m, 981m, 926vw, 900w, 856s, 805m, 745vw, 716w, 666vw, 635vw, 580w, 546w, 472w, 431w, 362w. Raman: 3112 (4), 3096 (4), 3075 (2), 2924 (8), 2793 (5), 2179 (2), 436 (2), 362 (1), 337 (1), 296 (10), 121 (2). Positive-ion MS: m/z 685 [2M-Cl]$^+$; 325 [M-Cl]$^+$ (100%). Positive-ion MS/MS of 325: m/z 307 [M-H_2O]; 208 [$MeCp_2V$]$^+$ (100%).

*[(MeCp)$_2$V(leu)]Cl (**6**)*

Yield: 0.57g (1.5 mmol, 86%). Calculated for $C_{18}H_{26}ClNO_2V$ (MW 376.7): C, 57.4; H, 7.0; N, 3.7; Cl, 9.4 Anal. found: C, 57.3; H, 7.0; N, 3.7; Cl, 9.6 EPR (CH_3OH solution): $A_{iso} = 66.6$ G, $g_{iso} = 1.983$. IR (KBr, cm^{-1}): 3423vs, 3236w, 3096s, 2958m, 2932w, 2871w, 1645m, 1635vs, 1521m, 1494m, 1454m, 1407m, 1386w, 1372w, 1339m, 1312w, 1277vw, 1260vw, 1240m, 1189w, 1170w, 1133m, 1081w, 1031m, 981m, 922m, 858s, 771w, 666w, 611w, 560s, 536w, 441m, 401w. Raman: not measured due to fluorescence. Positive-ion MS: m/z 713 [2M-Cl]$^+$; 339 [M-Cl]$^+$ (100%). Positive-ion MS/MS of 339: m/z 321[M-H_2O]; 261; 226 [$(MeCp)_2V + H_2O$]$^+$; 208 [$MeCp_2V$]$^+$ (100%).

*[(MeCp)$_2$V(ile)]Cl (**7**)*

Yield: 0.6 g (1.58 mmol, 90%). Calculated for $C_{18}H_{26}ClNO_2V$ (MW 376.7): C, 57.4; H, 7.0; N, 3.7; Cl, 9.4. Anal. found: C, 57.4; H, 7.4; N, 3.7; Cl, 9.2. EPR (CH_3OH solution): $A_{iso} = 66.9$ G, $g_{iso} = 1.990$. IR (KBr, cm^{-1}): 3430vs, 3243w, 3086vs, 2966s, 2931w, 2878w, 1652w, 1635vs, 1600w, 1516m, 1499s, 1456m, 1393vw, 1376m, 1344w, 1327w, 1297w, 1260m, 1194m, 1091m, 1031m, 990vw, 964vw, 936w, 862s, 802m, 667w, 569s, 478w, 438w, 367w. Raman: 3102 (3), 3088 (3), 2927 (8), 2915 (6), 1914 (1), 1501 (2), 1239 (3), 633 (2), 296 (10), 121 (2). Positive-ion MS: m/z 713 [2M-Cl]$^+$; 339 [M-Cl]$^+$ (100%). Positive-ion MS/MS of 339: m/z 321[M-H_2O]; 208 [$MeCp_2V$]$^+$ (100%).

*[(MeCp)$_2$V(phe)]Cl (**8**)*

Yield: 0.63g (1.54 mmol, 88%). Calculated for $C_{21}H_{24}ClNO_2V$ (MW 408.8): C, 61.7; H, 5.9; N, 3.4; Cl, 8.6. Anal. found:

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C, 61.5; H, 5.9; N, 3.4; Cl, 8.8. EPR (CH_3OH solution): $A_{iso} = 66.7$ G, $g_{iso} = 1.986$. IR (KBr, cm^{-1}): 3431vs, 3248w, 3092w, 2965m, 2958m, 2929m, 2375w, 2345w, 1645s, 1635vs, 1615w, 1496m, 1455m, 1437m, 1386m, 1262m, 1085m, 1029m, 983m, 860m, 803m, 753m, 704m, 666m, 601m, 485w, 446w. Raman: not measured due to fluorescence. Positive-ion MS: m/z 373 $[M\text{-}Cl]^+$ (100%). Positive-ion MS/MS of 373: m/z 355 $[M\text{-}H_2O]$; 295 $[M\text{-}C_6H_6]$; 265; 226 $[(MeCp)_2V + H_2O]^+$; 208 $[MeCp_2V]^+$ (100%).

[(MeCp)$_2$V(his)]Cl (**9**)

Yield: 0.6 g (1.5 mmol, 86%). Calculated for $C_{18}H_{22}ClN_3O_2V$ (MW 398.7): C, 54.2; H, 5.6; N, 10.5; Cl, 8.9. Anal. found: C, 54.2; H, 5.6; N, 10.5; Cl, 8.7. EPR (CH_3OH solution): $A_{iso} = 67.4$ G, $g_{iso} = 1.983$. IR (KBr, cm^{-1}): 3434vs, 3241w, 3125w, 2967w, 2930w, 1634vs, 1495m, 1456w, 1436m, 1394w, 1387m, 1352vw, 1321m, 1265m, 1111m, 1025m, 965s, 851m, 820s, 707w, 656w, 621m, 580w, 553vw, 479m, 382w. Raman: not measured due to fluorescence. Positive-ion MS: m/z 761 $[2M\text{-}Cl]^+$; 363 $[M\text{-}Cl]^+$ (100%). Positive-ion MS/MS of 363: m/z 345$[M\text{-}H_2O]$; 285 (100%); 339; 208 $[MeCp_2V]^+$.

[(MeCp)$_2$V(trp)]Cl (**10**)

Yield: 0.68 g (1.52 mmol, 87%). Calculated for $C_{21}H_{19}ClN_2O_2V$ (MW 447.8): C, 56.3; H, 4.3; N, 6.3; Cl, 7.9. Anal. found: C, 56.1; H, 4.5; N, 6.4; Cl, 8.0. EPR (CH_3OH solution): $A_{iso} = 66.9$ G, $g_{iso} = 1.986$. IR (KBr, cm^{-1}): 3413vs, 3267w, 3099m, 3061w, 2964w, 2928w, 1658w, 1645w, 1634vs, 1495m, 1455m, 1436m, 1378m, 1343m, 1322vw, 1262s, 1233w, 1206w, 1166w, 1100s, 1031m, 978w, 854m, 806m, 747s, 658w, 603w, 560m, 429m, 394w. Raman: 3112 (1), 3094 (<1), 3087 (<1), 3078 (<1), 3058 (3), 2923 (9), 2851 (<1), 1545 (3), 1503 (1), 1360 (1), 1343 (1), 1238 (2), 1055 (<1), 1013 (4), 758 (5), 440 (1), 289 (10), 200 (2). Positive-ion MS: m/z 829 $[2M\text{-}Cl]^+$; 412 $[M\text{-}Cl]^+$ (100%). Positive-ion MS/MS of 412: m/z 394$[M\text{-}H_2O]$; 334; 288; 226 $[(MeCp)_2V + H_2O]^+$; 208 $[MeCp_2V]^+$ (100%); 159.

[(MeCp)$_2$V(ala)]BPh$_4$·CH$_3$OH (**11**)

Complex **4** (0.2 g, 0.6 mmol) was dissolved in 2 ml of methanol, 1 ml of saturated methanolic solution of $NaBPh_4$ was added and the mixture was stirred for 10 min. Green precipitate was filtered on the frit, washed with 2 ml of cold methanol and dried in dynamic vacuum. Crystals suitable for X-ray diffraction analysis were obtained after slow cooling of saturated MeOH solution from 20 to −15 °C. Yield: 0.19 g

Table 4. Crystal data of **11–13**, measurement and refinement details[a]

Compound	11	12	13
Moiety formula	$C_{15}H_{20}NO_2V$, $C_{24}H_{20}B$, CH_4O	$C_{18}H_{26}NO_2V$, F_6P	$C_{18}H_{26}NO_2V$, F_6P
Crystal system	Orthorhombic	Monoclinic	Trigonal
Space group	$P2_1$ (No. 4)	$P2_1$ (No. 4)	$P3_1$
a, Å	9.77100(10)	8.98400(10)	10.5290(2)
b, Å	11.38200(10)	20.3530(2)	10.5290(2)
c, Å	29.8180(3)	11.85600(10)	16.3220(4)
β,°	107.2860(19)	104.4380(7)	90.00
Z	8	4	3
V, $Å^3$	3316.16(6)	2099.42(4)	1567.03(6)
D_c, g cm^{-3}	1.299	1.532	1.540
Crystal size, mm	0.5 × 0.4 × 0.2	0.6 × 0.3 × 0.25	0.55 × 0.17 × 0.12
Color	Green	Green	Green
Shape	plate	bar	bar
μ, mm^{-1}	0.339	0.615	0.618
h range	−12, 12	−11, 11	−13, 13
k range	−14, 14	−26, 26	−13, 13
l range	−38, 38	−15, 15	−21, 21
Reflections measured	40 751	60 820	22 123
independent (R_{int}[a])	7611 (0.045)	9583 (0.031)	4616 (0.0362)
observed [$I > 2\sigma(I)$]	6814	9259	4489
No. of parameters	431	547	274
GOF[b]	1.059	1.036	1.053
$R(F)$[c], $wR(F^2)$[c]	0.0334, 0.0781	0.0296, 0.0786	0.0252, 0.066
$\Delta\rho$, $eÅ^{-3}$	0.204, −0.245	0.469, −0.393	0.268, −0.190

[a] $R_{int} = \Sigma|F_o{}^2 - F_{o,mean}{}^2|/\Sigma F_o{}^2$.

[b] $GOF = \{\Sigma[w(F_o{}^2 - F_c{}^2)^2]/(N_{diffrs} - N_{params})\}^{1/2}$.

[c] $R(F) = \Sigma\|F_o| - |F_c\|/\Sigma|F_o|$, $wR(F^2) = \{\Sigma[w(F_o{}^2 - F_c{}^2)^2]/(\Sigma w(F_o{}^2)^2]\}^{1/2}$.

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(0.29 mmol, 48%). Calculated for $C_{40}H_{44}BNO_3V$ (MW 648.5): C, 74.0; H, 6.8; N, 2.2. Anal. found: C, 74.3; H, 6.9; N, 2.2. EPR (CH_3OH solution): $A_{iso} = 67.1$ G, $g_{iso} = 1.986$.

*[(MeCp)$_2$V(leu)]PF$_6$ (**12**)*

After dissolving complex **6** (0.2 g, 0.53 mmol) in 2 ml of water, 1 ml of saturated KPF_6 solution was added and mixture was stirred for 10 min. The precipitate was filtered on the frit, washed with 2 ml of cold water and dried in dynamic vacuum. Single crystals suitable for X-ray diffraction analysis were obtained after slow cooling of saturated aqueous solution of **12** from 20 to 4 °C. Yield: 0.1 g (0.21 mmol, 40%). Calculated for $C_{18}H_{26}$ F_6NO_2PV (MW 484.3): C, 44.6; H, 5.4; N, 2.9. Anal. found: C, 44.5; H, 5.5; N, 3.0. EPR (CH_3OH solution): $A_{iso} = 67.1$ G, $g_{iso} = 1.987$.

*[(MeCp)$_2$V(ile)]PF$_6$ (**13**)*

The complex **13** was prepared according to procedure described for **12**. Yield: 0.11 g (0.22 mmol, 43%). Calculated for $C_{18}H_{26}F_6NO_2PV$ (MW 484.3): 44.6; H, 5.4; N, 2.9. Anal. found: C, 44.6; H, 5.2; N, 2.8. EPR (CH_3OH solution): $A_{iso} = 66.7$ G, $g_{iso} = 1.988$.

X-ray crystallography

The X-ray data for **11–13** were obtained at 150 K using Oxford Cryostream low-temperature device on a Nonius KappaCCD diffractometer with Mo Kα radiation ($\lambda = 0.71073$ Å), a graphite monochromator, and the φ and ω scan mode. Data reductions were performed with DENZO-SMN.[31] The absorption was neglected. Structures were solved by direct methods (Sir92)[32] and refined by full matrix least-square based on F^2 (SHELXL97).[33] The hydrogen atoms on nitrogen were found on a difference Fourier map and refined isotropically; those on carbon atoms were fixed into idealized positions (riding model) and assigned temperature factors, either $H_{iso}(H) = 1.2\ U_{eq}$(pivot atom) or $H_{iso}(H) = 1.5\ U_{eq}$(pivot atom) for methyl moiety. Crystal data are summarized in Table 4.

Full crystallographic data for the structures of reported in this paper have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication no. CCDC 603 088, 603 089 and 603 090 for **11, 12** and **13**, respectively. Copies of the data can be obtained free of charge on application to CCDC, 12 Union Road, Cambridge CB2 1EZ, UK [fax: (+44) 1223-336-033; e-mail: deposit@ccdc.cam.ac.uk].

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