Collision induced dissociation (CID) to probe the outer sphere coordination chemistry of bis-salicylaldoximate complexes

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Synthesis of Ligands and complexes

3-X-Substituted Ligands

The synthesis of the 3-X-substituted ligands (X=H, Me, OMe, Cl, and ^{t}Bu) and their corresponding copper complexes are described by Forgan *et al.*[S1]

3-Bromo-5-*tert*-butyl-2-hydroxybenzaldehyde oxime

2-Bromo-4-*tert*-butylphenol (1). 4-*tert*-Butylphenol (5.140 g, 34 mmol) and tetra-*n*-butylammonium tribromide (16.50 g, 34 mmol) were dissolved in dichloromethane (100 ml) and methanol (60 ml) and stirred for 30 min. The solvent was removed *in vacuo* and the resulting orange oil partitioned between diethyl ether (100 ml) and water (100 ml). The organic phase was separated, washed with saturated brine (2 x 50 ml) and water (50 ml), dried over $MgSO_4$ and the solvent removed to give a slightly pink oil which was purified by silica-60 wet flash column chromatography (2% ethyl acetate in hexane eluent) to give a colourless oil (5.82 g, 75%). ^{1}H NMR (250 MHz, $CDCl_3$): δ_H (ppm) 1.13 (s, 9H, C(C*H*$_3$)$_3$), 6.83 (d, 1H, Ar*H*), 7.05 (dd, 1H, Ar*H*), 7.32 (d, 1H, Ar*H*); ^{13}C NMR (63 MHz, $CDCl_3$): δ_C (ppm) 31.9 (3C, C(**C**H$_3$)$_3$), 34.6 (1C, C(**C**H$_3$)$_3$), 110.4 (1C, aromatic **C**), 116.2 (1C, aromatic **C**H), 126.6 (1C, aromatic **C**H), 129.3 (1C, aromatic **C**H), 145.5 (1C, aromatic **C**), 150.4 (1C, aromatic **C**); FABMS *m/z* 230 (MH^+).

3-Bromo-5-*tert*-butyl-2-hydroxybenzaldehyde (2). A mixture of hexamethylenetetramine (9.1 g, 65 mmol) and **(1)** (3.00 g, 13.1 mmol) was heated in trifluoroacetic acid (50 ml) to 90˚C for 16 h under reflux. The reaction mixture was poured still hot into 2 M HCl (150 ml) and stirred for 8 h. An off-white precipitate was filtered, washed with water (50 ml) and vacuum dried over P_2O_5 to yield an off white powder (3.039 g, 90%). (Anal. Calc. for

$C_{11}H_{13}BrO_2.0.5H_2O$: C, 49.6; H, 5.3. Found: C, 50.0 H, 5.6%); ^{1}H NMR (250 MHz, $CDCl_3$): δ_H (ppm) 1.15 (s, 9H, C(C*H*$_3$)$_3$), 7.33 (d, 1H, Ar*H*), 7.63 (s, 1H, Ar*H*), 9.68 (s, 1H, C*H*O), 11.2 (br, 1H, O*H*); ^{13}C NMR (63 MHz, $CDCl_3$): δ_C (ppm) 31.6 (3C, C(**C**H$_3$)$_3$), 34.7, (1C, **C**(CH$_3$)$_3$), 111.2 (1C, aromatic **C**), 121.0 (1C, aromatic **C**), 129.8 (1C, aromatic **C**H), 138.0 (1C, aromatic **C**H), 144.6 (1C, aromatic **C**), 156.2 (1C, aromatic **C**), 196.7 (1C, Ar**C**HO); FABMS *m/z* 258 (MH^+).

Oximation General Procedure. *c*1.2 equivalents of KOH and $NH_2OH.HCl$ were dissolved separately in EtOH, mixed thoroughly and a white KCl precipitate removed by filtration. The filtrate was added to the precursor aldehyde, refluxed for 3 hr and the solvent removed *in vacuo*. The residue was redissolved in $CHCl_3$, washed with water 3 times, dried over $MgSO_4$ and the solvent removed *in vacuo* to yield the crude product.

3-Bromo-5-*tert*-butyl-2-hydroxybenzaldehyde oxime. Hydroxylamine hydrochloride (0.709 g, 10.2 mmol), potassium hydroxide (0.674 g, 10.2 mmol) and **(2)** (2.00 g, 7.8 mmol) were reacted according to the general procedure to yield a white solid (2.00 g, 94%). Crystals suitable for XRD analysis were grown by slow evaporation of a hexane/chloroform solution. (Anal. Calc. for $C_{11}H_{14}BrNO_2$: C, 48.6; H, 5.2; N, 5.2. Found: C, 48.4; H, 5.0; N, 5.3%); ν_{max}/cm^{-1} ($CHCl_3$) 3569 (free NOH), 3437br (H-bonded NOH), 3160br (PhOH), 2968 (C-H), 1625 (C=N); ^{1}H NMR (250 MHz, $CDCl_3$): δ_H (ppm) 1.20 (s, 9H, C(C*H*$_3$)$_3$), 7.06 (d, 1H, Ar*H*), 7.48 (d, 1H, Ar*H*), 8.13 (s, 1H, C*H*N); ^{13}C NMR (63 MHz, $CDCl_3$): δ_C (ppm) 31.7 (3C, C(**C**H$_3$)$_3$), 34.6, (1C, **C**(CH$_3$)$_3$), 110.6 (1C, aromatic **C**), 117.3 (1C, aromatic **C**), 127.2 (1C, aromatic **C**H), 132.2 (1C, aromatic **C**H), 144.3 (1C, aromatic **C**), 151.7 (1C, aromatic **C**), 152.6 (1C, Ar**C**HN); FABMS *m/z* 273 (MH^+).

5-*tert*-Butyl-2-hydroxy-3-nitrobenzaldehyde oxime

5-*tert*-Butyl-2-hydroxybenzaldehyde (3). Magnesium turnings (20 g, 800 mmol), methanol (373 ml), toluene (160 ml) and magnesium methoxide (a few drops of 8% w/w methanol solution) were refluxed until all magnesium was dissolved and H_2 evolution stopped. 4-*tert*-Butylphenol (200 g, 1.3 mol) was added and refluxed for 1 hr. Toluene (333 ml) was added and the mixture distilled under vacuum to remove the methanol/toluene azeotrope. A slurry of paraformaldehyde (120 g, 4 mol) in toluene (200 ml) was added slowly with continuous distillation, and heated for a further 2 hrs. After cooling to room temperature, H_2SO_4 (20%,

800 ml) was added slowly with stirring and heated to 50 °C to dissolve all solids. The product was extracted with toluene (2 x 400 ml), washed with H_2SO_4 (10%, 2 x 150 ml) and water (150 ml), dried over $MgSO_4$ and the solvent removed *in vacuo*. Purification by vacuum distillation (1 mm Hg, 120 °C) and silica-60 wet flash column chromatography (2% ethyl acetate in hexane eluent) yielded a bright yellow oil (144.2 g, 62.3%). (Anal. Calc. for $C_{11}H_{14}O_2$: C, 74.1; H, 7.9. Found: C, 73.4; H, 8.3%); ^{1}H NMR (250 MHz, $CDCl_3$): δ_H (ppm) 1.25 (s, 9H, C(C*H*$_3$)$_3$), 6.87 (d, 1H, Ar*H*), 7.43 (d, 1H, Ar*H*), 7.52 (dd, 1H, Ar*H*), 9.81 (s, 1H, C*H*O), 10.80 (s, 1H, O*H*); ^{13}C NMR (63 MHz, $CDCl_3$): δ_C (ppm) 31.5 (3C, C(**C**H$_3$)$_3$), 34.0 (1C, **C**(CH$_3$)$_3$), 117.0 (1C, aromatic **C**H), 120.0 (1C, aromatic **C**), 129.5 (1C, aromatic **C**H), 134.5 (1C, aromatic **C**H), 142.5 (1C, aromatic **C**), 159.5 (1C, aromatic **C**), 197.0 (1C, Ar**C**HO); FABMS *m/z* 179 $(MH)^+$.

5-*tert*-Butyl-2-hydroxy-3-nitrobenzaldehyde (4). Nitric acid (70%, 7.0 ml, 160 mmol) was added dropwise to a solution of **(3)** (25.2 g, 140 mmol) in glacial acetic acid (25 ml) at 0 °C. The mixture was stirred at 55 °C for 16 h, cooled to room temperature and the thick mother liquors were decanted from the bright yellow solid, which was washed with 50:50 hexane: diethyl ether. Recrystallisation from hexane yielded a deep yellow product (10.4 g, 33%). (Anal. Calc. for $C_{11}H_{13}NO_4$: C, 59.2; H, 5.9; N, 6.3. Found: C, 58.7; H, 5.7; N, 6.4%); ^{1}H NMR (250 MHz, $CDCl_3$): δ_H (ppm) 1.33 (s, 9H, C(C*H*$_3$)$_3$), 8.12 (d, 1H, Ar*H*), 8.31 (d, 1H, Ar*H*), 10.38 (s, 1H, C*H*O), 11.22 (s, 1H, ArO*H*); ^{13}C NMR (63 MHz, $CDCl_3$): δ_C (ppm) 30.8 (3C, C(**C**H$_3$)$_3$), 34.4 (1C, **C**(CH$_3$)$_3$), 124.8 (1C, aromatic **C**), 127.8 (1C, aromatic **C**H), 134.2 (1C, aromatic **C**H), 134.6 (1C, aromatic **C**), 143.3 (1C, aromatic **C**), 154.4 (1C, aromatic **C**), 189.3 (1C, Ar**C**HO); FABMS *m/z* 223 (MH^+).

5-*tert*-Butyl-2-hydroxy-3-nitrobenzaldehyde oxime. Hydroxylamine hydrochloride (0.709 g, 10.2 mmol), potassium hydroxide (0.674 g, 10.2 mmol) and **(4)** (2.00 g, 9.00 mmol) were reacted according to the general procedure to yield a bright yellow solid (1.94 g, 90%). Crystals suitable for XRD analysis were grown by slow evaporation of a DCM solution. (Anal. Calc. for $C_{11}H_{14}N_2O_4$: C, 55.5; H, 5.9; N, 11.8. Found: C, 55.4; H, 5.7; N, 11.8%); ν_{max}/cm^{-1} ($CHCl_3$) 3575 (free NOH), 3402br (H-bonded NOH), 3230br (PhOH), 2968 (C-H), 1633 (C=N), 1537s (NO_2); ^{1}H NMR (250 MHz, $CDCl_3$): δ_H (ppm) 1.26 (s, 9H, C(C*H*$_3$)$_3$), 7.93 (d, 1H, Ar*H*), 8.05 (d, 1H, Ar*H*), 8.45 (s, 1H, ArC*H*N); ^{13}C NMR (63 MHz, $CDCl_3$): δ_C (ppm) 31.4 (3C, C(**C**H$_3$)$_3$), 35.0, (1C, C(**C**H$_3$)$_3$), 122.1 (1C, aromatic **C**), 123.6 (1C, aromatic

CH), 132.7 (1C, aromatic **C**H), 134.7 (1C, aromatic **C**), 143.7 (1C, aromatic **C**), 146.5 (1C, Ar**C**HN), 151.3 (1C, aromatic **C**); FABMS *m/z* 239 (MH^+).

3-X-Substituted copper complexes

Copper(II) complexes of the ligands were synthesised using the following general procedure. Stoichiometric amounts of the ligand and metal acetate (0.5 equivalents) were mixed in methanol (50 ml) for 24 h. Colour changes due to complex formation occurred immediately, along with precipitation. Complexes were isolated by filtration and dried under vacuum.

[Cu(X=NO$_2$)$_2$]. $Cu(OAc)_2.H_2O$ (0.460 g, 2.30 mmol) and 5-*tert*-butyl-2-hydroxy-3-nitrobenzaldehyde oxime (1.011 g, 4.43 mmol) yielded a bright green solid from the method above (0.903 g, 73%). (Anal. Calc. for $C_{22}H_{26}O_8N_4Cu$: C, 49.1; H, 4.9; N, 10.4. Found: C, 48.2; H, 3.3; N, 9.8%); ν_{max}/cm^{-1} (THF) 3257br (NOH), 2969 (C-H), 1631 (C=N), 1519s (NO_2); FABMS *m/z* 539 (MH^+).

[Cu(X=Br)$_2$]. $Cu(OAc)_2.H_2O$ (0.100 g, 0.50 mmol) and 3-bromo-5-*tert*-butyl-2-hydroxybenzaldehyde oxime (0.270 g, 1.01 mmol) yielded a brown solid from the method above (0.284 g, 88%). (Anal. Calc. for $C_{22}H_{26}Br_2O_4N_2Cu$: C, 43.6; H, 4.3; N, 4.6. Found: C, 43.9; H, 4.1; N, 4.5%); ν_{max}/cm^{-1} ($CHCl_3$) 3216br (NOH), 2966 (C-H), 1602 (C=N); FABMS *m/z* 607 (MH^+).

5-R-Substituted Ligands

The synthesis of the 5-R-substituted ligands, where R = ^{t}Bu and ^{t}Oct and their corresponding copper complexes are described by Forgan *et al.*[S1] and those where R = H, and their corresponding copper complexes by Wood *et al.*[S2]

Experimental General Procedure

All samples were prepared using LCMS analytical grade acetonitrile received from Aldrich. 80 μM solutions of each complex were injected into Thermo-Fisher LCQ mass spectrometer. The selected precursor ion was subjected to increasing amounts of collision energy until fully dissociated. All studies were carried out under the same experimental conditions:

Sample flow rate:	2μL/min		
Sheath gas flow rate:	46 (Arb. Units)		
Aux. gas flow rate:	0 (Arb. Units)		
Capillary temperature	190°C		
RF amplitude	560V		
Spray Voltage	4.5kV	Capillary Voltage	-26.5V
Octapole 1 offset	6.8V	Lens Voltage	13V
Octapole 2 offset	10.5V	Tube lens offset	0V

The ratio between the intensity of the trapped precursor and the total number of fragment ions was plotted against the collision energy. A sigmoidal fit function from Origin 8.0 was used to fit each of the dissociation curves and the X_c value was determined using the sigmoidal fit equation (1) where A1 and A2 are determined minima and maxima for each curve.

$$Y = A2 + \frac{(A1 - A2)}{\left\{1 + \exp\left(\frac{(X - X_c)}{dX}\right)\right\}} \qquad (1)$$

Accurate Mass CID

A Waters Quadrupole-Time-of-Flight (Q-ToF) mass spectrometer was used in conjunction with an Aquity Sample Manager and Binary solvent Manager. All studies were carried out under the same experimental conditions:

Sample injection volume:	6μL		
Desolvation gas flow rate:	800L/h		
Aux. gas flow rate:	30L/h		
Source temperature	90°C		
Desolvation Temperature	350°C		
Spray Voltage	2.2kV	Extraction Cone	4.0
Sampling Cone	80	Ion Guide	2.0

The samples were sprayed using direct injection and 4 x 20 scans were taken and average masses were used. The data was interpreted using Waters MassLynx software and the fragment ion identities determined.

Fragment analysis

CID Fragments of 3-X-substituted $[Cu(L)(L\text{-}H)]^-$ series

In CID-MS the stability of a trapped species is determined by its chemical structure and for a series of closely related complexes the nature of substituents and the number of bonds they contain, as discussed in the main text. In order to be able to compare stabilities of a series of closely related trapped complexes it is important to ascertain whether their fragmentation occurs by identical pathways. A systematic study of the fragmentation of the $[Cu(\mathbf{L})(\mathbf{L}\text{-}H)]^-$ species was undertaken to confirm that this was the case for the copper(II) salicylaldoximato complexes reported in this paper. **Figure S1** shows the CID spectra for the $[Cu(\mathbf{L})(\mathbf{L}\text{-}H)]^-$ complex anions containing **L** with a ^{t}Bu-group in the 5-position and different 3-X-

subsituents, H, Me, ^{t}Bu, NO_2, Cl, Br and OMe, indicating the three major fragments found. The fragmentation patterns are virtually identical varying only in minor fragments. For certain species, *i.e.* X=Br, X=Cl, a small amount of XH loss is observed, these losses account for <2% of fragmentation.

The peaks highlighted in **Figure S1** are three of the major species, (i) the peaks highlighted in grey represent the trapped ions (the three fragment peaks seen next to these are the initial fragment losses and are detailed more fully in **Figure S3**), (ii) the peaks highlighted in red show the major $[Cu(\mathbf{L}\text{-}H)]^-$ species, and (iii) the peaks highlighted in blue indicate the rearranged cyano form of the deprotonated oxime. Under the experimental conditions used in **Figure S1** these peaks are minimal, but at higher NCE's the intensities of these peaks become significant. The analysis confirms that the fragmentation patterns are very similar in all cases and provides confidence that the X_c values (see main text) which relate to breakdown of the monoanions $[Cu(\mathbf{L})(\mathbf{L}\text{-}H)]^-$ can be used to judge their relative stabilities, given that they all dissociate by the same mechanism.

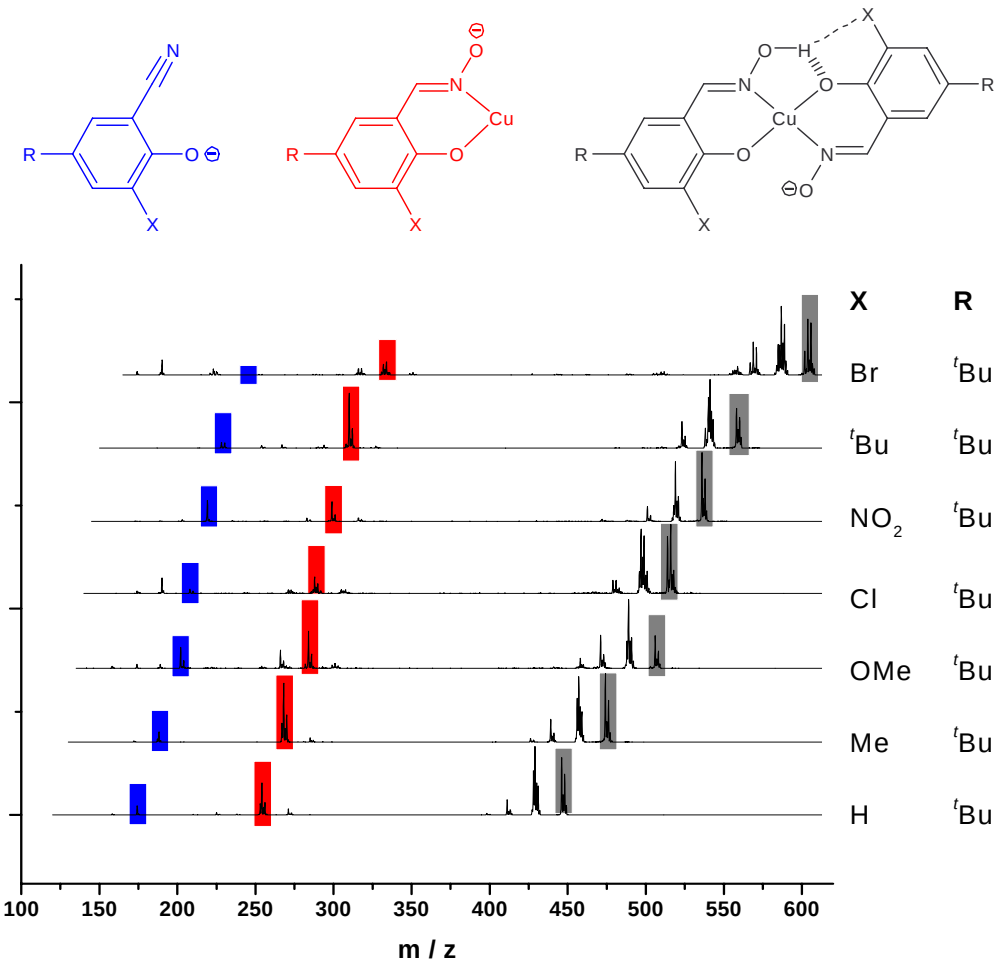

Figure S1: The CID mass spectrum for each 3-X-substituted species where NCE ≈ X_c. Structures of the trapped species (highlighted in grey) and two of the major fragments (highlighted in red and blue) are also indicated.

CID Fragments of 5-R-substituted [Cu(L)(L-H)]$^-$ series

As for the 3-X-substituted series, the fragmentation profiles for the 5-R-substituted [Cu(**L**)(**L**-H)]$^-$ ions were fully investigated to ensure dissociation occurred in a similar manner. **Figure S2** shows the CID spectra for each species (analyte ion current/total ion current ≈ 5-10%). The highlighted fragments and their structures correspond to those highlighted in **Figure S2**. The fragments formed from each species are analogous and again allow us to conclude that the gas phase stability of the [Cu(**L**)(**L**-H)]$^-$ ions is dependent on variations of substituents (the length of the R group and the nature of the X group in this case) and not on differing fragmentation profiles. Evidence for the formulae and structures of the ions is presented in the next section.

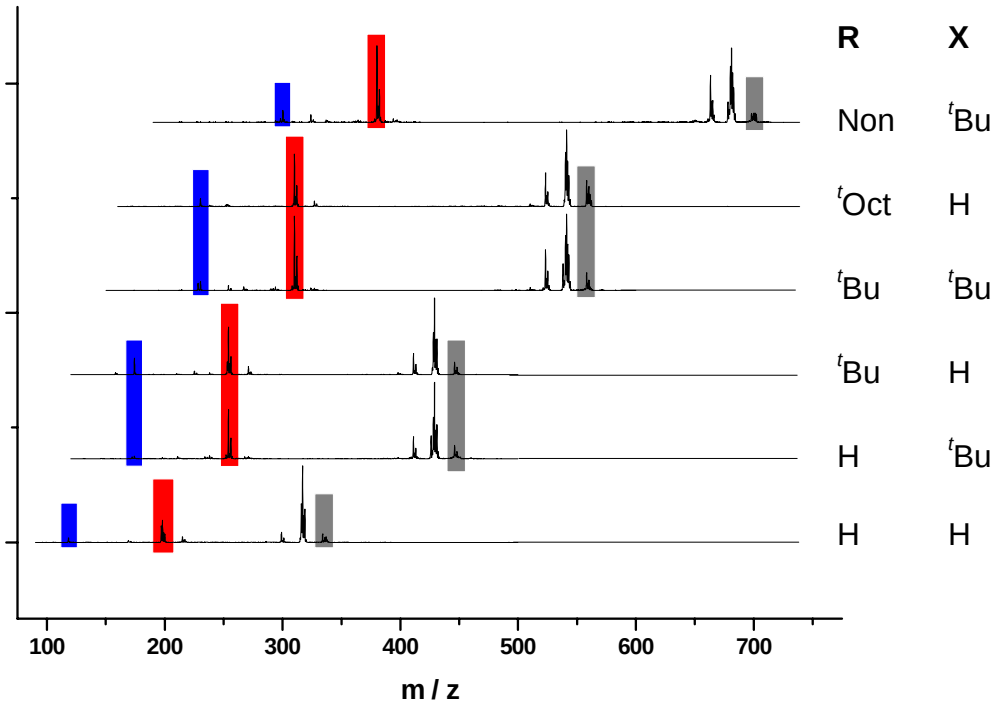

Figure S2: The CID mass spectrum for each 5-R-substituted species. Highlighted species are analogous to those in **Figure S1**.

Accurate CID Fragment masses for $[Cu(L1)(L1\text{-}H)]^-$

Accurate mass CID spectra were taken using a Waters Quadrupole-Time-of-Flight (Q-ToF) mass spectrometer. The accurate mass fragments were compared to the low resolution series to ensure fragmentation proceeded in the same manner within the collision cell as seen within the quadrupole ion trap. **Figure S3** details the observed accurate masses of fragments along with proposed structural formulae and possible dissociative mechanisms and pathways.

Collision induced dissociation (CID) to probe the outer sphere coordination chemistry of bis-salicylaldoximate complexes†

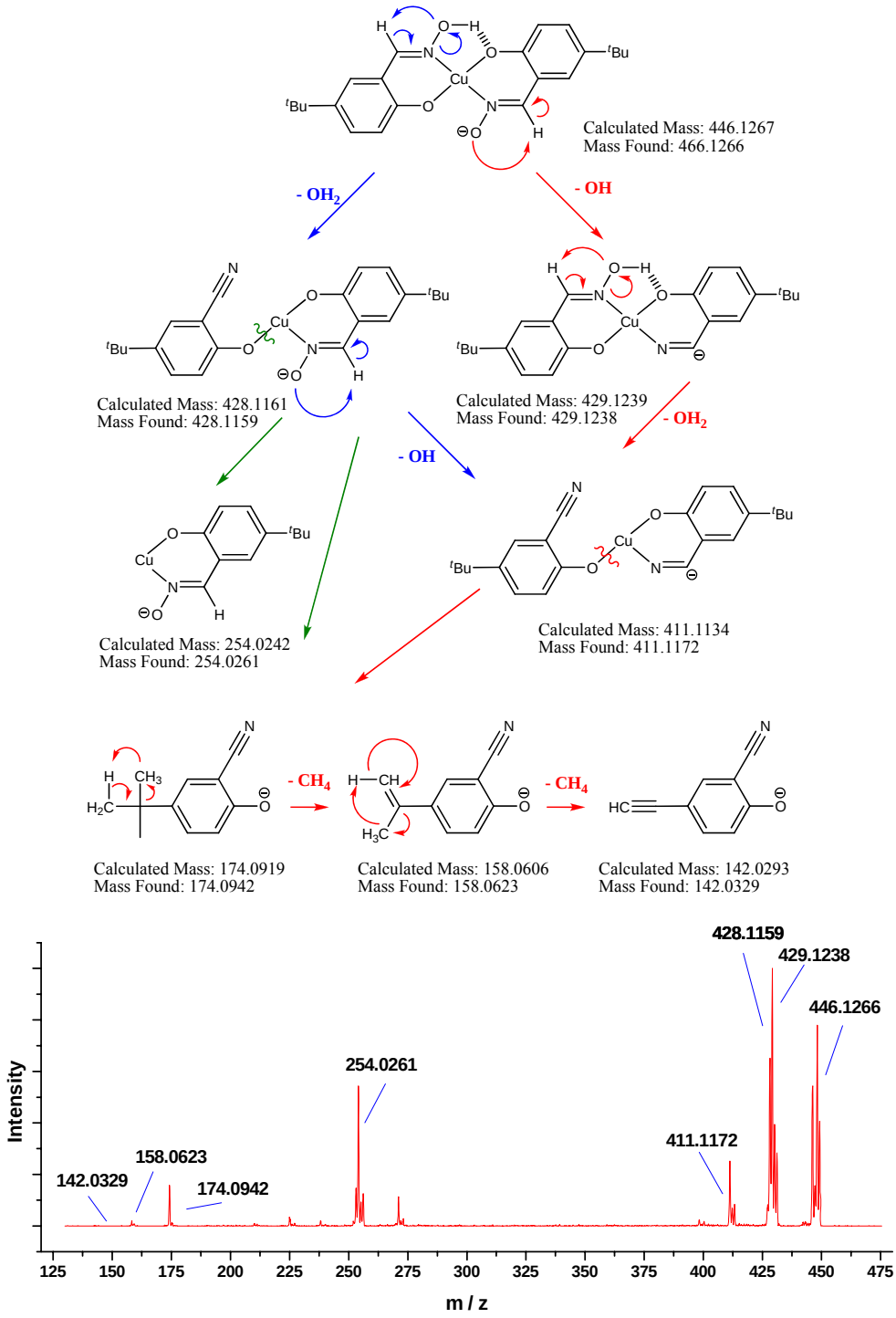

Figure S3: High resolution CID mass spectrum (below) and proposed fragment structures and fragmentation pathways for $[Cu(\mathbf{L})(\mathbf{L}\text{-H})]^-$, where X = H and R = ^{t}Bu. (top)

References

1) R. S. Forgan, P. A. Wood, J. Campbell, D. K. Henderson, F. E. McAllister, S. Parsons, E. Pidcock, R. M. Swart and P. A. Tasker, *Chem. Commun*., 2007, 4940.
2) P. A. Wood, R. S. Forgan, A. R. Lennie, S. Parsons, E. Pidcock, P. A. Tasker, and J. E. Warren, *CrystEngComm*, 2008, **10**, 239.