Electronic Supplementary Information (ESI)

Water Exchange Reaction of a Manganese Catalase Mimic: Oxygen-17 NMR Relaxometry Study on (aqua)manganese(III) in Salen Scaffold and its reactions in mildly basic medium

V. Rao Mundlapati[a,b], Priyambada Jena[c], Achyut N. Acharya[c], Akshaya K Kar[d], Anadi C. Dash[d]* and Himansu S. Biswal[a,b]*

[a]*School of Chemical Sciences, National Institute of Science Education and Research (NISER), Institute of Physics Campus, Bhubaneswar 751 005, India. E-mail: himansu@niser.ac.in*

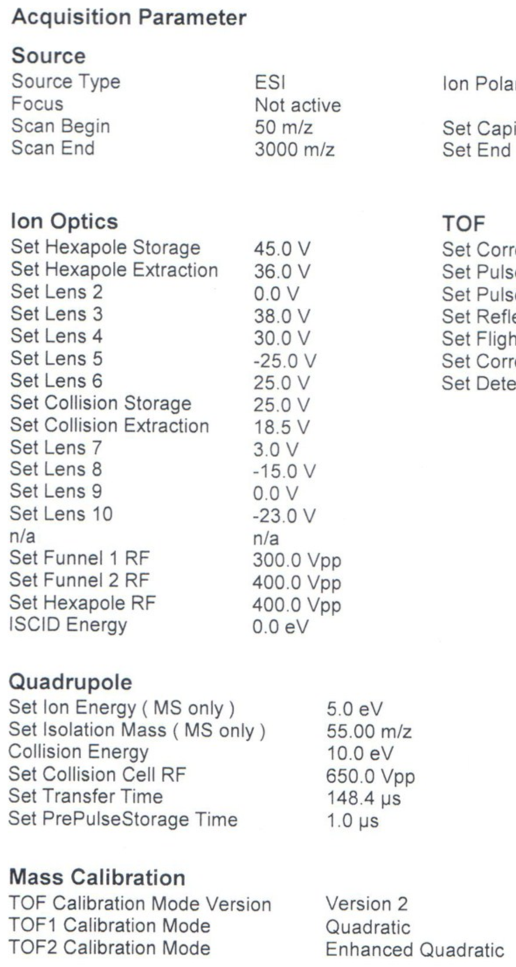
[b]*Homi Bhabha National Institute, Training School Complex, Anushakti Nagar, Mumbai 400094, India*

[c]*Department of Chemistry, College of Engineering and Technology, Bhubaneswar 751003, India. E-mail: aacharya@cet.edu.in*

[d]*Department of Chemistry, Utkal University, Bhubaneswar 751004, India.*

E-mail: acdash41@gmail.com

ESI-MS Acquisition Parameters



Acquisition Parameter

Source

Source Type	ESI	Ion Polarity	Positive	Set Nebulizer	0.4 Bar
Focus	Not active			Set Dry Heater	180 °C
Scan Begin	50 m/z	Set Capillary	4500 V	Set Dry Gas	4.0 l/min
Scan End	3000 m/z	Set End Plate Offset	-500 V	Set Divert Valve	Waste

Ion Optics

Set Hexapole Storage	45.0 V
Set Hexapole Extraction	36.0 V
Set Lens 2	0.0 V
Set Lens 3	38.0 V
Set Lens 4	30.0 V
Set Lens 5	-25.0 V
Set Lens 6	25.0 V
Set Collision Storage	25.0 V
Set Collision Extraction	18.5 V
Set Lens 7	3.0 V
Set Lens 8	-15.0 V
Set Lens 9	0.0 V
Set Lens 10	-23.0 V
n/a	n/a
Set Funnel 1 RF	300.0 Vpp
Set Funnel 2 RF	400.0 Vpp
Set Hexapole RF	400.0 Vpp
ISCID Energy	0.0 eV

TOF

Set Corrector Fill	63.0 V
Set Pulser Pull	780.0 V
Set Pulser Push	810.0 V
Set Reflector	1700.0 V
Set Flight Tube	8600.0 V
Set Corrector Extract	662.0 V
Set Detector TOF	2020.0 V

Processing

Summation	5000 x
Guessed Noise	200
Peak Width	5 pts
Average Noise	1
Guessed Average	100

Quadrupole

Set Ion Energy (MS only)	5.0 eV
Set Isolation Mass (MS only)	55.00 m/z
Collision Energy	10.0 eV
Set Collision Cell RF	650.0 Vpp
Set Transfer Time	148.4 µs
Set PrePulseStorage Time	1.0 µs

Mass Calibration

TOF Calibration Mode Version	Version 2
TOF1 Calibration Mode	Quadratic
TOF2 Calibration Mode	Enhanced Quadratic

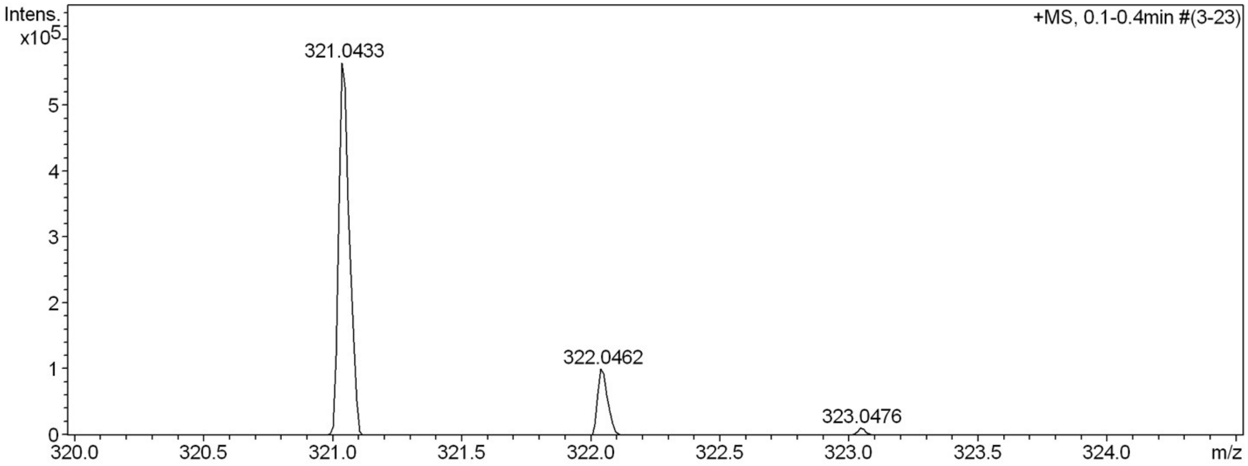


Figure **S1** –ESI-MS of [Mn^{III}(salen)(OH$_2$)$_2$]ClO$_4$ in aqueous medium.

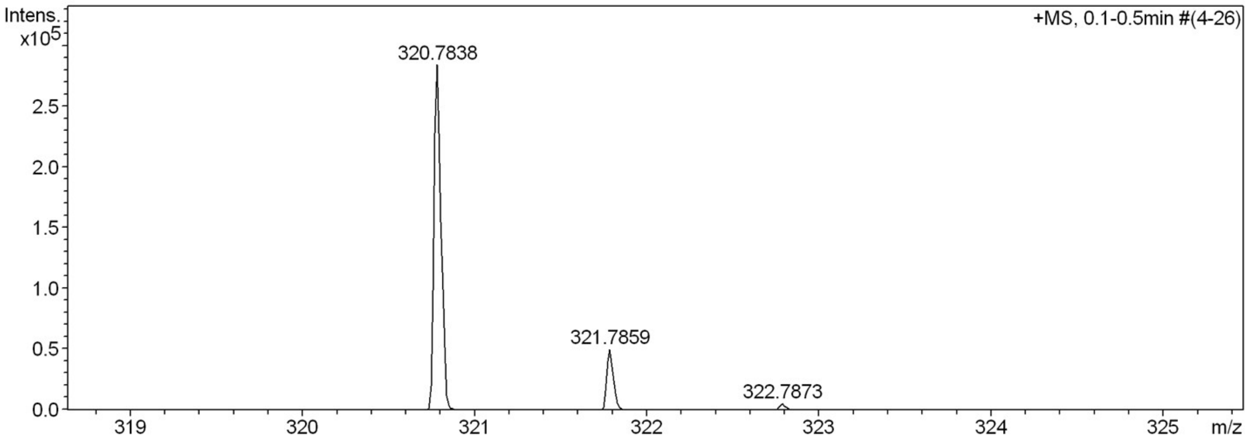


Figure **S2(a)**-ESI-Ms for[$Mn^{III}(salen)(OH_2)_2](ClO_4)$ in acetonitrile solvent

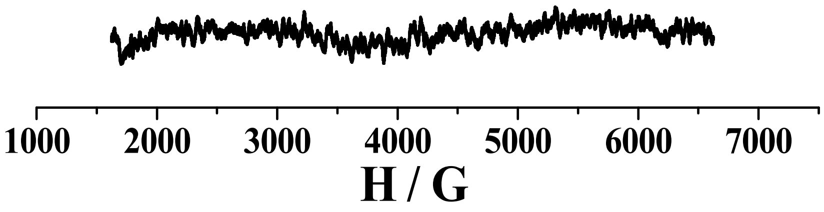


Figure **S2(b)**- A solution of $Mn^{III}(salen)(OH_2)_2^+$ at pH =10 was equilibrated at 65°C for 100 minutes and then set aside to cool and EPR spectrum (X-band) was monitored at room temperature. No evidence of Mn^{II} and Mn^{IV}.

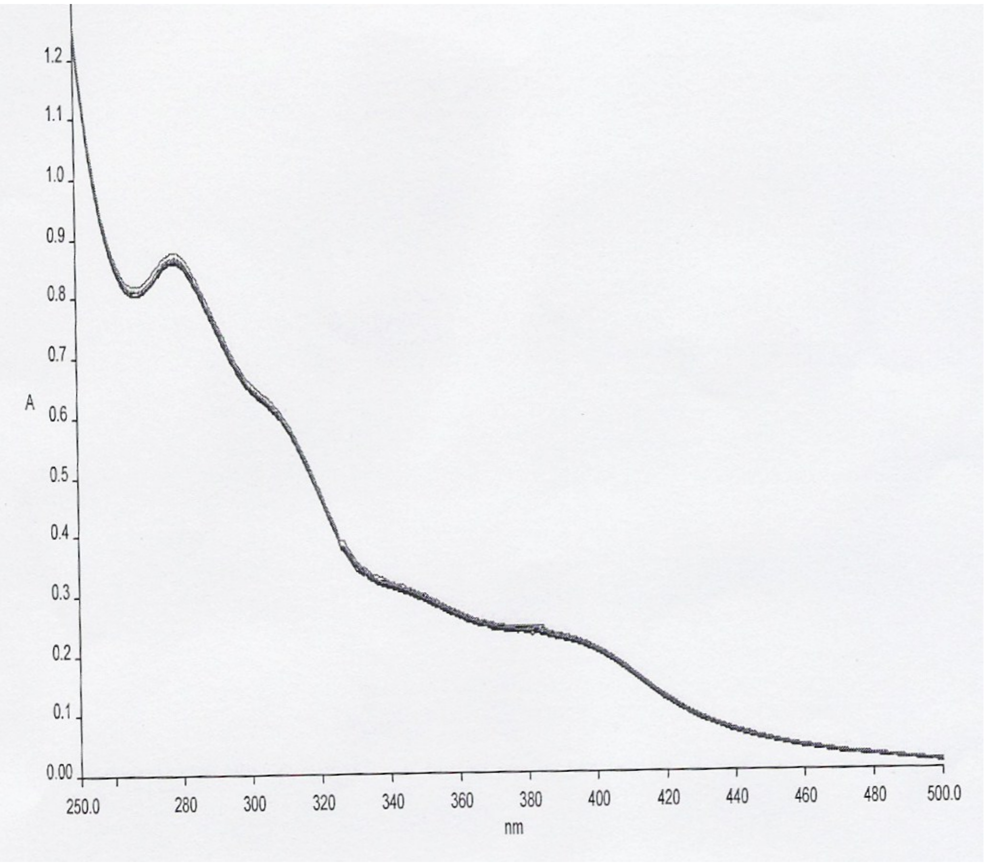


Figure **S3**- Time dependent repeat scan (overlapping) spectra of *trans*-Mn^{III}(salen)(OH$_2$)$_2^+$ at pH =4.2, 50^{o}C, [complex]$_T$ = 6.033 x 10^{-5} mol dm^{-3} , no. of curves 9 (time 1-174 min) , final pH 4.3.

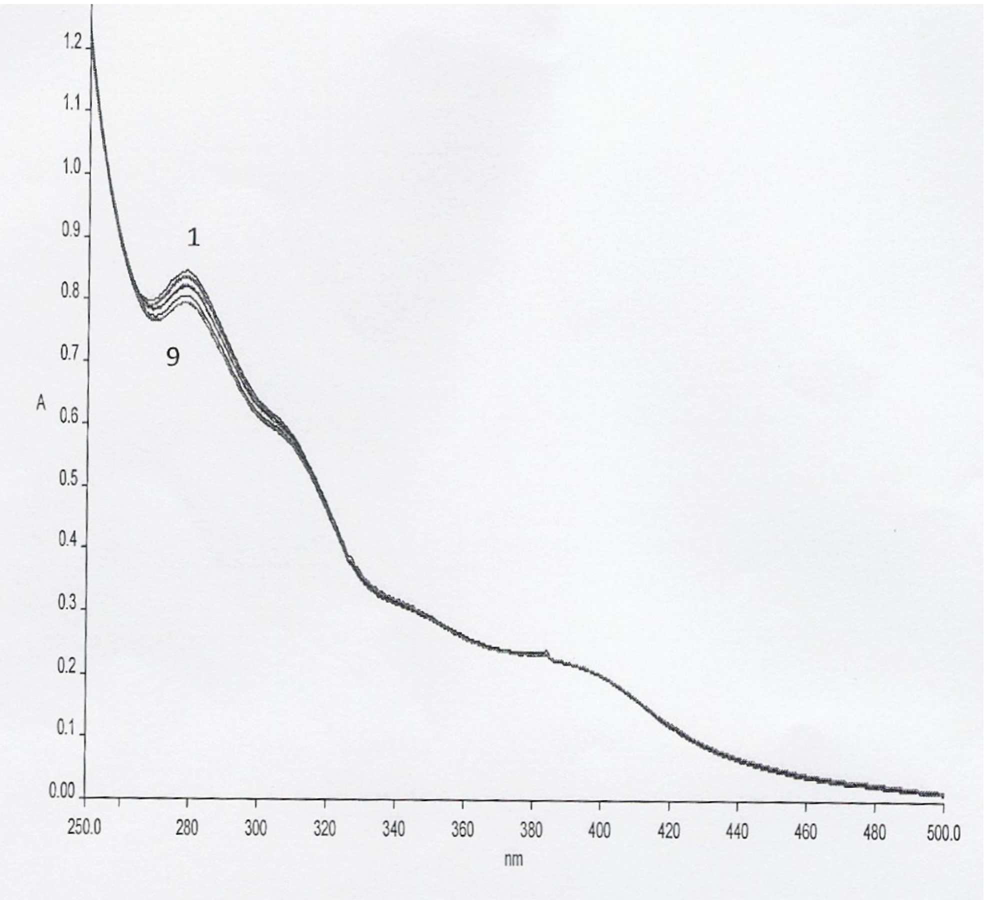


Figure **S4**- Time dependent repeat Scan Spectra of *trans*-$Mn^{III}(salen)(OH_2)_2^+$ at pH =8.05, 50^{o}C, $[complex]_T$ = 6.033 x 10^{-5} mol dm^{-3};Curve no. (time, min) : 1(1), 2(15), 3(31), 4955), 5(83), 6(104), 7(132), 8(153), 9(179).The final pH of the solution 7.12.

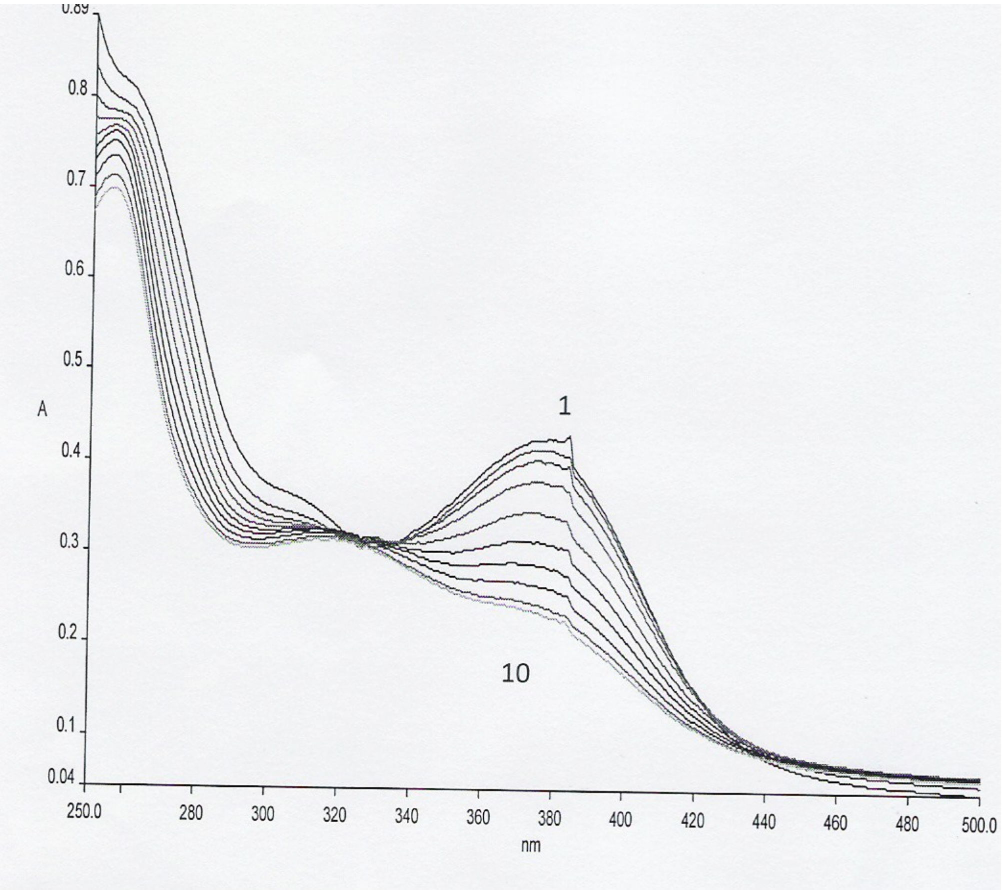


Figure **S5**- Time dependent spectral scans (absorbance reading downwards at 380 nm) of *trans*-$Mn^{III}(salen)(OH_2)(OH)$ at pH= 10.2, 50°C , $[complex]_T = 6.033 \times 10^{-5}$ mol dm^{-3}; Curve no. (time, min): 1(16),2(26),3(37),4(43),5(65),6(86),7(108), 8(133), 9(1620, 10(180).The final pH of the solution 9.2.

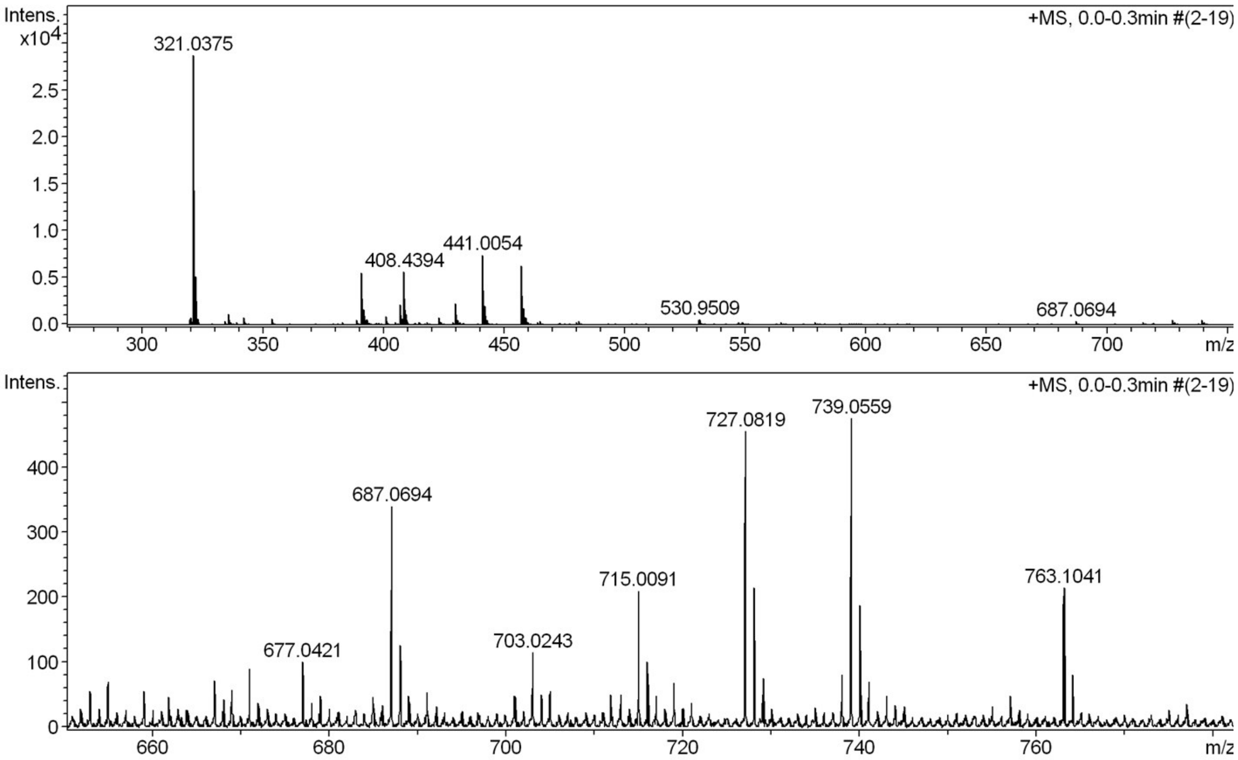


Figure **S6 -** A solution (1.095 x 10^{-3} mol dm^{-3}) of the complex, $[Mn^{III}(salen)(OH_2)_2]^+$ in water was adjusted to pH 10, heated at 65°C for 25 minutes, cooled to room temperature and then mass spectra was taken immediately.

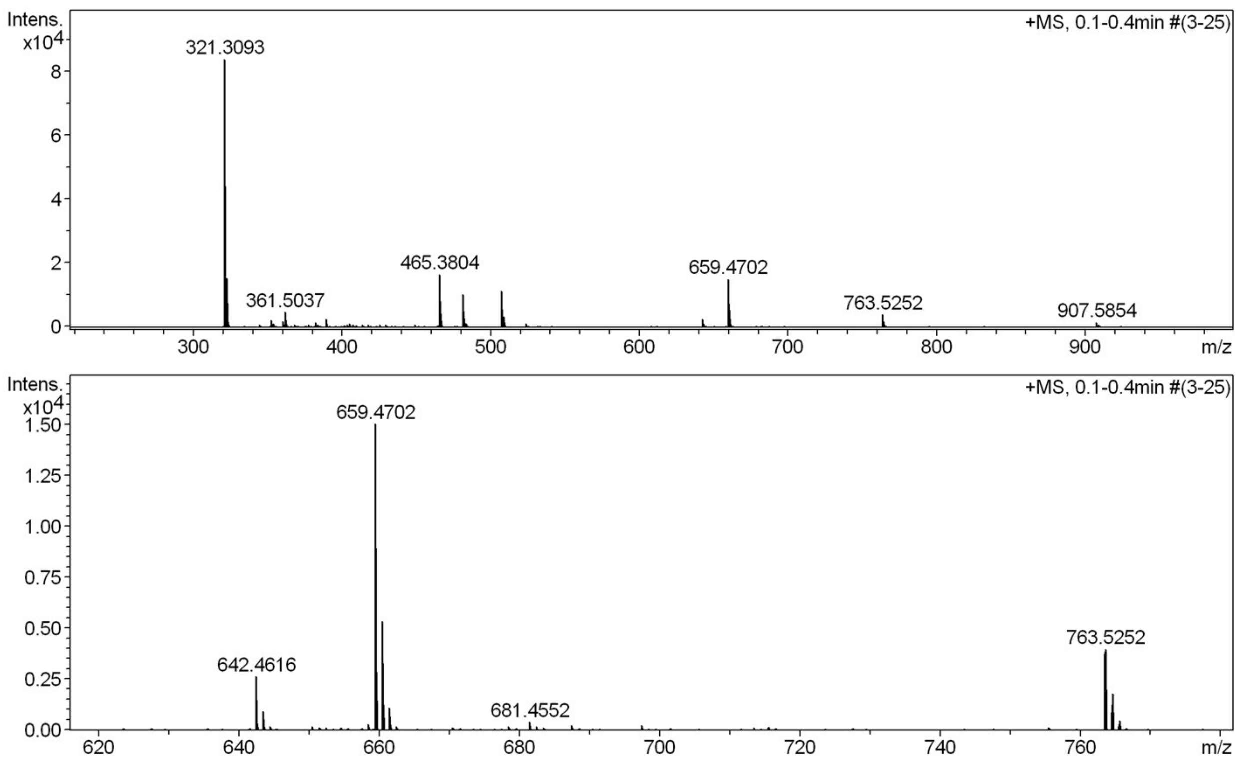
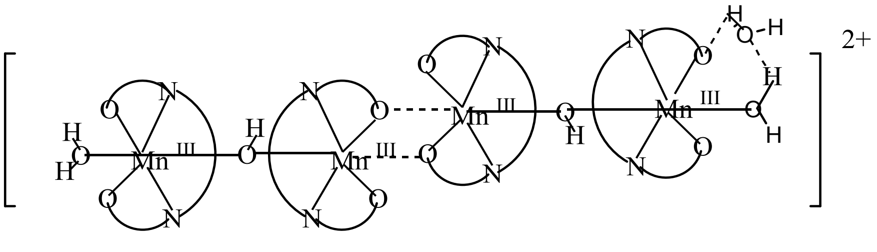


Figure **S7** – Conditions same as for S6; mass spectrum was taken after aging the solution at room temperature for one hr.

Scheme 1 : Interpretation of Mass spectra: m/z+ 687.07 (obs)

(a) $[Mn^{III}(salen)_4(OH)_2(H_2O)_3]^{2+}$: m/z^{2+} **686.5 (cal)**



C

(b) $\{[OMn^V(salen)(\text{-}OH\text{-})Mn^VO(salen)]^+ - 4H\}$: m/z^+ **687.4 (cal);**

$\{[OMn^V(salen)(\text{-}O\text{-})Mn^{VI}O(salen)]+ - 4H\}$ **: m/z+ 686.4 (cal)**

$\{[OMn^V(salen)(\text{-}O\text{-})Mn^{VI}O(salen)]+ - 3H\}$ **: m/z+ 687.4 (cal)**

$[Mn^{III}(salen)\text{-}(OH)\text{-}Mn^{III}(salen)]^+ + ClO_4^- \rightleftharpoons [O_3Cl\text{-}O\text{-}[Mn^{III}(salen)\text{-}(OH)\text{-}Mn^{III}(salen)]$

m/z+ 659.47

$\xrightarrow{-\ ClO_3^-}$

$[OMn^V(salen)(OH)\text{-}(salen)Mn^{III}]^+ + ClO_4^- \rightleftharpoons [OMn^V(salen)(OH)\text{-}(salen)Mn^{III}\text{-}O\text{-}ClO_3]$

m/z+ 675.5

$\xrightarrow{-\ ClO_3^-}$

$[OMn^V(salen)\text{-}(OH)\text{-}(salen)Mn^VO]^+ \xrightarrow{-\ H} [OMn^V(salen)\text{-}(O)\text{-}Mn^{VI}(salen)O]^+$

m/z+ 691..4 **m/z+ 690.4**

$\downarrow$ **- 4H** $\downarrow$ **- 2H**

$[OMn^V(salen\text{-}2H)(\text{-}OH\text{-})Mn^V(salen\text{-}2H)O]^+$

m/z+ 687.4

$[OMn^V(salen\text{-}2H)\text{-}(O\text{-})Mn^{VI}(salen)O]^+$

m/z+ 688.4

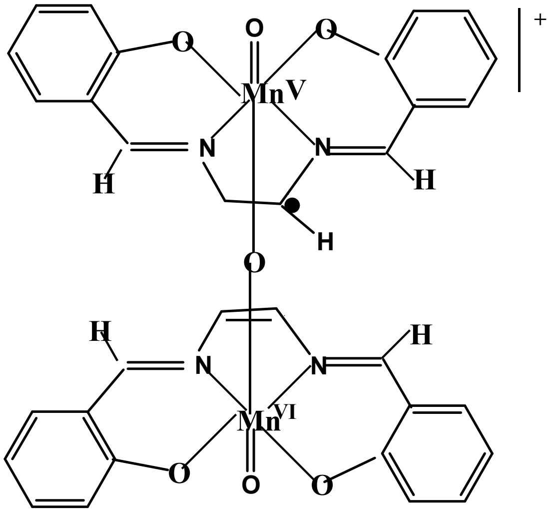
$\xrightarrow{-\ H}$ $\downarrow$ **- 2H**

$[OMn^V(salen\text{-}2H)\text{-}(O\text{-})Mn^{VI}(salen\text{-}2H)O]^+$

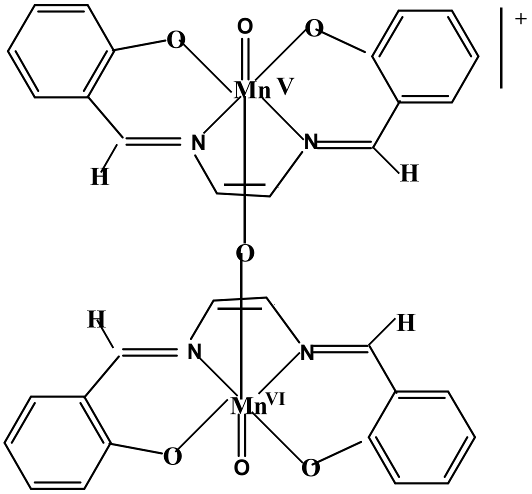
m/z+ 686.4

$[OMn^V(salen\text{-}2H)\text{-}(O\text{-})Mn^{VI}(salen\text{-}H)O]^+$

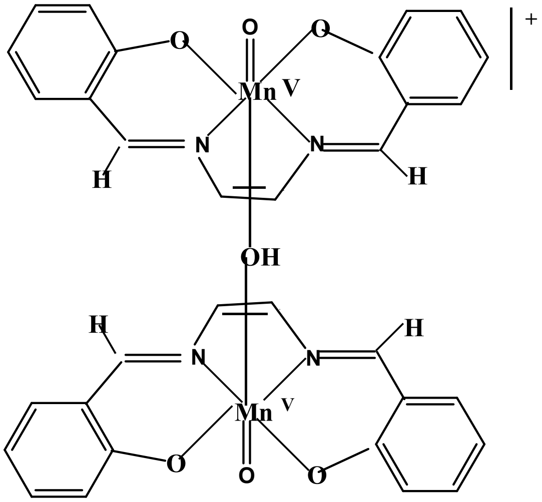
m/z+ 687.4



m/z+ 687.4



m/z+ 686.4



m/z+ 687.4

Table **S1**: Line width data forvariable temperature ^{17}O relaxation study of $Mn^{III}(salen)(OH_2)_2^+$ with $[complex]_{Total}$ = 0.00846 mol dm^{-3} .

$\boldsymbol{P_m}$ = 3.05 x 10 ^{4} (pH 4); $\boldsymbol{P_m}$ = 0.8 x 10 ^{4} + ***C*** (1/298.2   1/*T*) (pH 10). [a]

Temp/K	**1/*T* K^{-1}**	**$\Delta\nu_{obs}$ /Hz**	**$\Delta\nu_{solvent}$/Hz**	**10 $^{6}/T_{2r}$ (s^{-1})**	**$\Delta\nu_{obs}$ /Hz**	**$\Delta\nu_{solvent}$/Hz**	**10 $^{2}/T_{2p}$ (s^{-1})**
	pH = 4				pH = 10		
283.2	0.00353	269.1	71.8	2.03	270.5	73.2	6.20
288.2	0.00347	263.4	62.4	2.07	249.5	62.5	5.87
293.2	0.00341	255.5	57.0	2.04	210.4	55.1	4.88
298.2	0.00335	275.1	44.6	2.37	142.1	45.7	3.03
303.2	0.00330	279.3	39.4	2.47	125.8	40.1	2.69
308.2	0.00324	262.8	35.1	2.34	78.1	35.4	1.34
313.2	0.00319	247.4	31.5	2.22	71.4	31.6	1.25
318.2	0.00314	231.6	28.9	2.09	65.8	28.4	1.17
323.2	0.00309	221.1	26.3	2.01	55.8	26.0	0.936
328.2	0.00305	209.4	24.4	1.90	50.6	24.0	0.835
333.2	0.00300	197.8	22.7	1.80	46.4	22.6	0.748
338.2	0.00296	186.1	21.3	1.70	43.1	21.3	0.685
343.2	0.00291	174.4	20.4	1.59	39.9	20.7	0.603
348.2	0.00287	166.4	19.6	1.51	37.7	19.7	0.565
353.2	0.00283	159.3	18.8	1.45	35.0	19.4	0.490

[a] $\mathbf{P_m}$ = $2[complex]_{Total}/55.5$ (pH = 4). *calcd.* $\boldsymbol{P_m}$ = 0.8 x 10^{-4} at pH =10, 298.2 *K* based on $\{[Mn^{III}(salen)(OH)(OH_2]_{eq} + [Dimer]_{eq}\}/55.5$,   using $K_{eq(av.)}$ = 1.5 x 10^5 dm^3 mol dm^{-3} for Monomer $\rightleftharpoons$ Dimer ; dimer : $(OH_2)Mn^{III}(salen)$-OH-$Mn^{III}(salen)(OH)$.

Table **S2**. Calculated parameters for water exchange reaction of $[Mn^{III}(salen)(OH_2)_2]^+$.

1/T (K^{-1})	**pH = 4,** **P_m = 3.05 x 10^{-4}** [a]		**pH = 10,** **P_m = 0.8 x 10^{-4} + C x (1/298.2 1/T)** [a]	
	$10^{-6}/T_{2r}$ (s^{-1})	**$10^{-6}k_{ex}$ (s^{-1})**	**$10^{-2}/T_{2p}$ (s^{-1})**	**$10^{-7}k_{ex}$ (s^{-1})**
0.00353	2.03	2.4	6.20	1.1
0.00347	2.07	2.8	5.87	1.6
0.00341	2.04	3.3	4.88	2.4
0.00335	2.37	3.8	3.03	3.5
0.00330	2.47	4.3	2.69	5.1
0.00324	2.34	5.0	1.34	7.4
0.00319	2.22	5.7	1.25	10.5
0.00314	2.09	6.5	1.17	14.8
0.00309	2.01	7.3	0.936	20.7
0.00305	1.90	8.1	0.835	28.6
0.00300	1.80	9.2	0.748	39.0
0.00296	1.70	10.2	0.685	52.9
0.00291	1.59	11.6	0.603	71.0
0.00287	1.51	12.8	0.565	94.6
0.00283	1.45	14.2	0.490	125.0
	A_m/s^{-1}	5.24 x 10^{10} [b]		1.22 x 10^9 [b]
	E_m/kJ mol^{-1}	34.0 ± 1.5		19.0 ± 1.0
	Δ**$H^{\neq}$**/kJ mol^{-1}	18.3 ± 5.9		54.1 ± 4.3
	Δ**$S^{\neq}$**/J K^{-1} mol^{-1}	57.6 ± 20.2		+ 80.9 ± 17.0 .

	β/Hz C / K^{-1}	(1.43 ± 0.13) x 10^9		(3.69 ± 0.52) x 10^9 0.1033 ± 0.0276
	X^2/R^2 [c]	0.00651/0.9479		0.03882/0.99308

[a] see foot note *a* of Table **S1**. [b] held fixed at the least squares best value for calculating errors of other parameters. [c] 10 $^{6}/T_{2r}$ (s $^{-1}$), 10 $^{2}/T_{2p}$(s^{-1}) values were fitted.

`Appendix –*I*

(1) First phase :

$$2\text{A} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \text{Dim (Dimer)} \qquad (1)$$

$$1/2(-d[\text{A}]_t/dt = k_1[\text{A}]_t^2 - k_{-1}[\text{Dim}]_t \qquad (2)$$

$$k_{-1} = \text{k}_1[\text{A}]_e^2 /[\text{Dim}]_e$$

where $[\text{A}]_e$ and $[\text{Dim}]_e$ denote the concentrations of A ($\text{Mn}^{\text{III}}(\text{salen})(\text{OH}_2)(\text{OH})$) and its Dimer respectively at equilibrium, and $[\text{A}]_t$ and $[\text{Dim}]_t$ are their respective concentrations at time 't'. With $[\text{Dim}]_0 = 0$ and $[\text{A}]_0$ as the zero time concentration of A, $[\text{Dim}]_{t\,(e)} = ([\text{A}]_0-[\text{A}]_{t(e)})/2$ and $\boldsymbol{k^{/} = 2k_1}$, equation (2) can be transformed to eq.(3)

$$-d[\text{A}]_t/\{([\text{A}]_t - [\text{A}]_e)([\text{A}]_0\,[\text{A}]_t + [\text{A}]_0[\text{A}]_e - [\text{A}]_e[\text{A}]_t)\} = -\,(\boldsymbol{k^{/}}/([\text{A}]_0 - [\text{A}]_e))dt \qquad (3)$$

Equ. (3) is solved to get the integrated expression :

$$\ln\left[\frac{([\text{A}]_t - [\text{A}]_e)\,([\text{A}]_0 + \text{C}_\text{I})}{([\text{A}]_t + \text{C}_\text{I}])\,([\text{A}]_0 - [\text{A}]_e)}\right] = -\,k^{/}([\text{A}]_e + \text{C}_\text{I})\ t \qquad (4)$$

where

$$\mathbf{C_I} = [\text{A}]_0\,[\text{A}]_e/\,(\text{A}]_0 - [\text{A}]_e).$$

Eq. (4) yields eq. (5)

$$([\text{A}]_t - [\text{A}]_e)/([\text{A}]_t + \text{C}_\text{I})$$

$$= [([A]_0 - [A]_e)/([A]_0 + C_I)] \exp(-k^{/}([A]_e + \mathbf{C_I})t) \qquad (5)$$

which can be recast as eq (6)

$$[A]_t = - C_{\mathbf{I}} + \frac{[A]_e + C_{\mathbf{I}}}{1-\{([A]_0 - [A]_e)/([A]_0 + C_{\mathbf{I}})\}\exp(- k^{/}([A]_e+C_{\mathbf{I}})t)} \qquad (6)$$

In terms of absorbance at time t (D_t), and molar extinction coefficients of the complex (ε_A) and the dimer (ε_{Dim}):

$$D_t = \boldsymbol{\varepsilon}_A [A]_t + \boldsymbol{\varepsilon}_{Dim} [Dim]_t = \boldsymbol{\varepsilon}_A [A]_t + \boldsymbol{\varepsilon}_{Dim} ([A]_0 - [A]_t)/2 \qquad (7)$$

Setting $\varepsilon^{/} = \varepsilon_{Dim}/2$, $D^{/} = \varepsilon^{/} [A]_0$, $D_0 = \varepsilon_A [A]_0$, $D_e = \varepsilon_A [A]_e + \varepsilon^{/} ([A]_0 - [A]_e)$ eq. (7) yields ,

$$\mathbf{A_t = \frac{(D^{/} - D_t)(A_0 - A_e)}{(D_e - D_0)}} \qquad \mathbf{(8)}$$

Using equs (5 - 8) , the expression for $C_{\mathbf{I}}$, and rearranging, it is possible to get:

$$\mathbf{D_t = P_1 - \frac{P_2}{1 - P_3 \exp(-P_4 \boldsymbol{t})}} \qquad \mathbf{(9)}$$

where,

$$\mathbf{P_1 = D^{/} + \frac{(D_e - D_0)[A]_0 [A]_e}{([A]_0 - [A]_e)^2}}$$

$$P_2 = \frac{(D_e - D_0)\,(2[A]_0[A]_e - [A]_e^2)}{([A]_0 - [A]_e)^2}$$

$$P_3 = \left[\frac{[A]_0 - [A]_e}{[A]_0}\right]^2$$

$$P_4 = k^{/}\left[\frac{2[A]_0[A]_e - [A]_e^2}{[A]_0 - [A]_e}\right]$$

$$[A]_e = [A]_0(1 - P_3^{1/2}); \qquad [\mathrm{Dim}]_e = ([A]_0 - [A]_e)/2$$

$$K_{eq} = [\mathrm{Dim}]_e/[A]_e^2$$

$$k^{/} = P_4P_3^{1/2}/\{[A]_0(1-P_3)\}$$

$$D^{/} = P_1 \quad [P_2/(1+P_3^{1/2})]; \;\; \varepsilon_{\mathrm{Dim}} = 2D^{/}/[A]_0$$

$$D_0 = P_1 - [P_2/(1-P_3)]; \quad \varepsilon_A = D_0/[A]_0 \,.$$

$$D_e = D_0 + [P_2P_3/(1-P_3)]$$

(2) Second phase (a rate process preceded by equilibrium):

$$2A \overset{K_e}{\rightleftharpoons} \mathrm{Dim} \xrightarrow{k_2} P$$

where $K_e = k_1/k_{-1}$ and k_2 denotes the pseudo first order rate constant for the transformation of the dimer (Dim) to the product(s) . The linear decrease of absorbance (D_t) with time (t) indicates that the transformation of the Dimer to Product is very slow. The absorbance – time data after the equilibrium for the first phase was analysed by the method of initial rate :

$D_t = \varepsilon_A [A]_t + \varepsilon_{Dim} [Dim]_e \exp(-k_2 t) \approx \varepsilon_A [A]_t + \varepsilon_{Dim} [Dim]_e (1- k_2 t)$ (10)

$\mathbf{D_e} = \varepsilon_A [A]_e + \varepsilon_{Dim} [Dim]_e$ (11)

From equs (10) and (11),

$D_t - \mathbf{D_e} = \varepsilon_A ([A]_t - [A]_e) - \varepsilon_{Dim} [Dim]_e k_2 t$

$D_t = \mathbf{D_e} - \varepsilon_A([A]_e - [A]_t) - \varepsilon_{Dim} [Dim]_e k_2 t = D_e[1- \{\varepsilon_A([A]_e - [A]_t)/D_e\}] - \varepsilon_{Dim} [Dim]_e k_2 t$ (12)

As $\varepsilon_A([A]_e - [A]_t)/D_e << 1$, eq(12) reduces to eq(13)

$D_t = \mathbf{D_e} - \varepsilon_{Dim} [Dim]_e k_2 t$ (13)

and $k_2 = |\boldsymbol{G}|/ (\varepsilon_{Dim} [Dim]_e)$, where $|G|$ is gradient of the D_t versus t plot.

Appendix –II

Treatment of line width data and evaluation of the parameters defining $\boldsymbol{T_{2m}}^{-1}$, $\boldsymbol{\tau_m}^{-1}$ (k_{ex}), and $\Delta \boldsymbol{w_m}$

$(\boldsymbol{\Delta} v_{obs} - \boldsymbol{\Delta} v_{solvent})\, \pi/\boldsymbol{P_m} =$

$$\boldsymbol{T_{2r}^{-1} = \tau_m^{-1}(T_{2m}^{-2} + (T_{2m}\tau_m)^{-1} + \Delta\omega_m^2) / [(T_{2m}^{-1} + \tau_m^{-1})^2 + \Delta\omega_m^2] + T_{2os}^{-1}} \quad \text{(3a)}$$

The contribution of $1/\boldsymbol{T}_{2os}$ was neglected and

$\boldsymbol{T_{2m}^{-1}} = \boldsymbol{A}_m \, \mathrm{Exp}(-\boldsymbol{E_m}/\boldsymbol{RT})$;

$\boldsymbol{\tau_m}^{-1} = k_{ex} = (k_b\boldsymbol{T}/h) \, \mathrm{Exp}[\Delta\boldsymbol{S}^{\neq}/\boldsymbol{R} - \Delta\boldsymbol{H}^{\neq}/\boldsymbol{RT}]$;

$\Delta\boldsymbol{w_m} = \boldsymbol{\beta}/\mathbf{T}$

were substituted in equation (3a) and simplified to :

$\boldsymbol{10^{-6}/T_{2r} = 10^{-6}}$**Up/Down** **(S1)**

where

$\mathrm{Up} = \mathrm{Up}_1 + \mathrm{Up}_2 + \mathrm{Up}_3$

$\mathrm{Down} = \mathrm{DD}_1 + \mathrm{DD}_2 + \mathrm{DD}_3 + \mathrm{DD}_4$

The Up_{is} and DD_{is} are given by the following expressions.

$\mathrm{Up}_1 = \boldsymbol{A_m}^2(k_b\boldsymbol{T}/h) \, \mathrm{Exp}[\Delta\boldsymbol{S}^{\neq}/\boldsymbol{R} - (\Delta\boldsymbol{H}^{\neq} + 2\boldsymbol{E_m})/\boldsymbol{RT}]$

$\mathrm{Up}_2 = \boldsymbol{A}_m(k_b\boldsymbol{T}/h)^2 \, \mathrm{Exp}[2\Delta\boldsymbol{S}^{\neq}/\mathbf{R} - (2\Delta\boldsymbol{H}^{\neq} + \boldsymbol{E_m})/\boldsymbol{RT}]$

$\mathrm{Up3} = [\boldsymbol{\beta}^2 k_b/(h\boldsymbol{T})] \, \mathrm{Exp}[\Delta\boldsymbol{S}^{\neq}/\boldsymbol{R} - \Delta\boldsymbol{H}^{\neq}/\boldsymbol{RT}]$

$\mathrm{DD}_1 = \boldsymbol{A_m^2} \, \mathrm{Exp}(-\boldsymbol{2E_m}/\boldsymbol{RT})$

$\mathrm{DD}_2 = 2(\boldsymbol{A_m}k_b\boldsymbol{T}/h) \, \mathrm{Exp}[\Delta\boldsymbol{S}^{\neq}/R - (\Delta\boldsymbol{H}^{\neq} + \boldsymbol{E_m})/\boldsymbol{RT}]$

$\mathrm{DD}_3 = (k_b\boldsymbol{T}/h)^2 \, \mathrm{Exp}[2\Delta\boldsymbol{S}^{\neq}/\boldsymbol{R} - 2\Delta\boldsymbol{H}^{\neq}/\boldsymbol{RT}]$

$\mathrm{DD}_4 = (\boldsymbol{\beta}/\boldsymbol{T})^2$

The parameters for **equation S1**($Y = 10^{-6}/T_{2r}$; $X = 1/\boldsymbol{T}$) to minimize the residuals were set as :

$P_1 = \boldsymbol{A_m}$, $P_2 = \boldsymbol{E_m/R}$, $P_3 = \Delta\boldsymbol{S^{\neq}/R}$, $P_4 = \Delta\boldsymbol{H^{\neq}/R}$ and $P_5 = \boldsymbol{\beta}$.

$k_b = 1.380658 \times 10^{-23}$ J K^{-1}, $h = 6.6260755 \times 10^{-34}$ J s were used.