Electronic Supplementary Material (ESI) for Organic & Biomolecular Chemistry.

ELECTRONIC SUPPLEMENTARY INFORMATION

Mechanistic insights on the amidolysis of a phosphate triester: the antagonistic role of water

Amanda K. Jacobs,[a] Leociley R. A. Menezes,[b] Juliano M. Fernandes,[a] Guilherme L. Kosteczka,[a] Guilherme Sassaki[b] and Renan B. Campos[a]*

[a] *Academic Department of Chemistry and Biology. Universidade Tecnológica Federal do Paraná (UTFPR). Rua Deputado Heitor Alencar Furtado, 5000, 81.280-340, Curitiba, Paraná, Brazil.*
[b] *Department of Biochemistry and Molecular Biology. Universidade Federal do Paraná. Av. Cel. Franscisco H. dos Santos, 100, 81.531-980, Curitiba, Paraná, Brazil.*
**renan@utfpr.edu.br*

SUMMARY

1. KINETIC DATA

1.1. Activation parameters

The activation parameters ($\Delta G^{\ddagger}$, $\Delta H^{\ddagger}$ e $\Delta S^{\ddagger}$) were determined using Equations S1, S2 and S3 being that the Equation S1 served as a basis for the Eyring plot (Figure S7).

$$\ln\left(\frac{k_{obs}}{t}\right) = \left[\ln\left(\frac{k_b}{h}\right) + \frac{\Delta S^{\ddagger}}{R}\right] - \frac{\Delta H^{\ddagger}}{RT} \qquad \text{(Equation S1)}$$

$$\Delta G^{\ddagger} = -\ln\left(\frac{k_{obs}h}{k_b t}\right)RT \qquad \text{(Equation S2)}$$

$$\Delta G^{\ddagger} = \Delta H^{\ddagger} - T\Delta S^{\ddagger} \qquad \text{(Equation S3)}$$

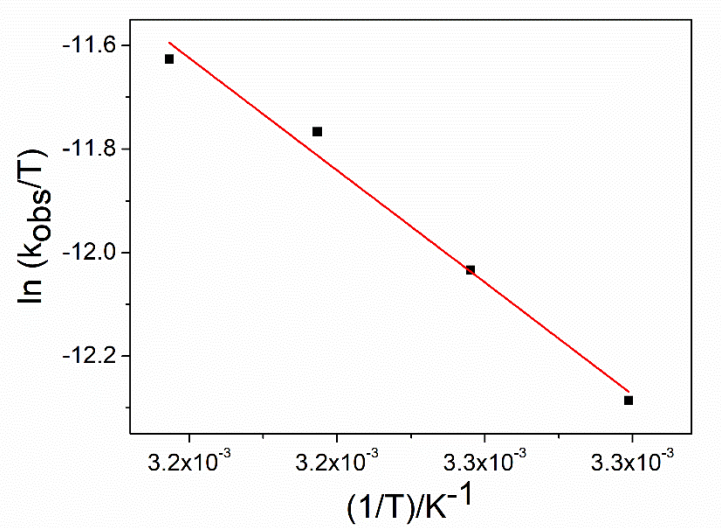

Figure S1. Eyring plot for DEDNPP solvolysis in FMD (97% v/v).

Table S1. Thermodynamic parameters for the solvolysis of DEDNPP in FMD at 25 °C.

Parameter	Value / cal·mol^{-1}
$\Delta G^{\ddagger}$	2.1 x 10^4
$\Delta H^{\ddagger}$	8.6 x 10^3
$\Delta S^{\ddagger}$	-41.5

1.2. Solvent effects: formamide/acetonitrile solutions.

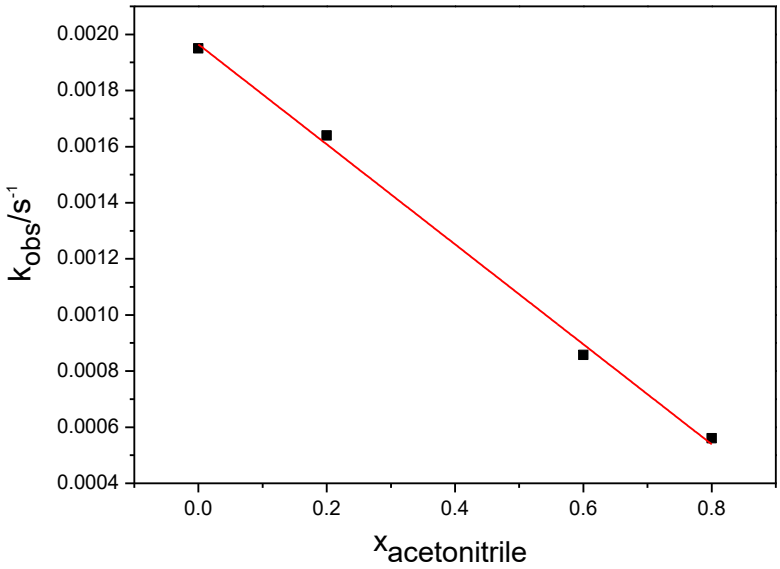

Figure S2. Linear profile obtained for the DEDNPP amidolysis in different acetonitrile molar fractions, indicating no significant cage effects are involved

1.3. Solvent effects: formamide/water solutions.

Table S2. Rate constants (k_{obs}) versus water fraction (x_{WATER}) for the amidolysis of DEDNPP at 25 °C.

k_{obs}/s^{-1}	X_{WATER}
1.96×10^{-3}	0.03
1.22×10^{-3}	0.15
8.45×10^{-4}	0.2
4.345×10^{-4}	0.4
2.7×10^{-4}	0.6
1.76×10^{-4}	0.8
$1{,}44 \times 10^{-5}$	10

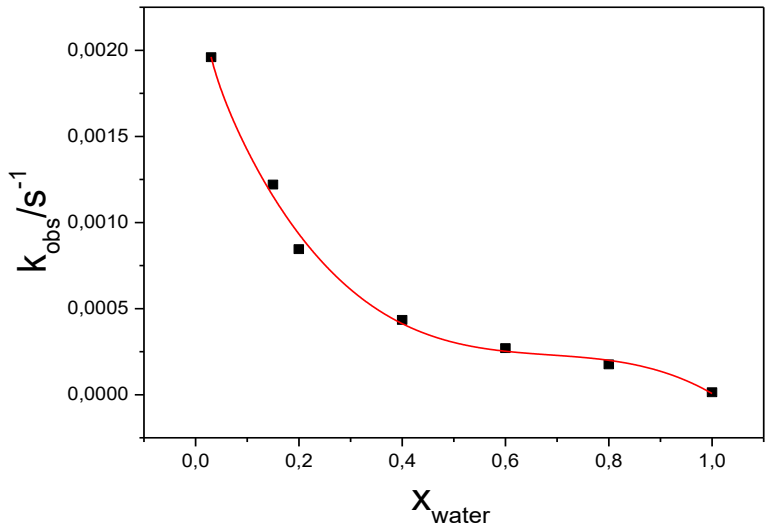

Figure S3. Second-best statistical fitting for k_{obs} of DEDNPP amidolysis versus molar fraction of water.

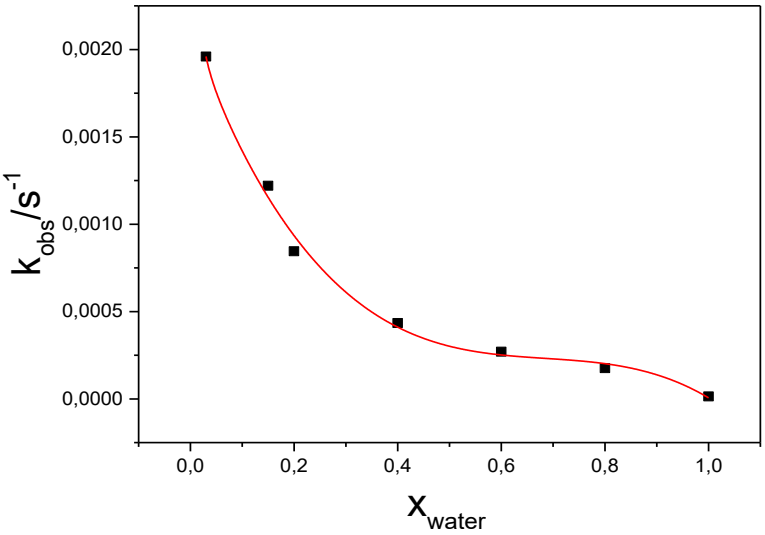

Figure S4. Third-best statistical fitting for k_{obs} of DEDNPP amidolysis *versus* molar fraction of water.

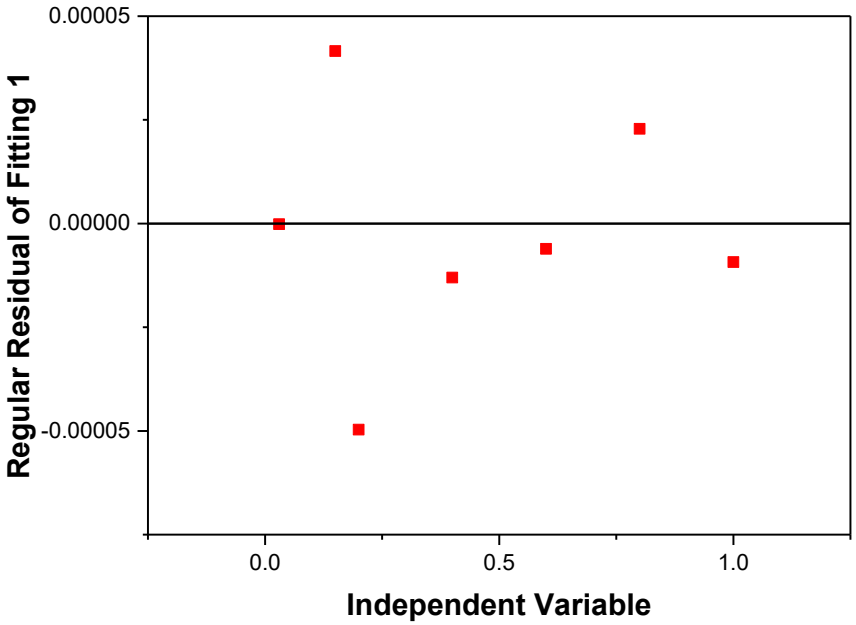

Figure 5S. Residual dispersion for the best statistical fitting for kobs of DEDNPP amidolysis versus molar fraction of water.

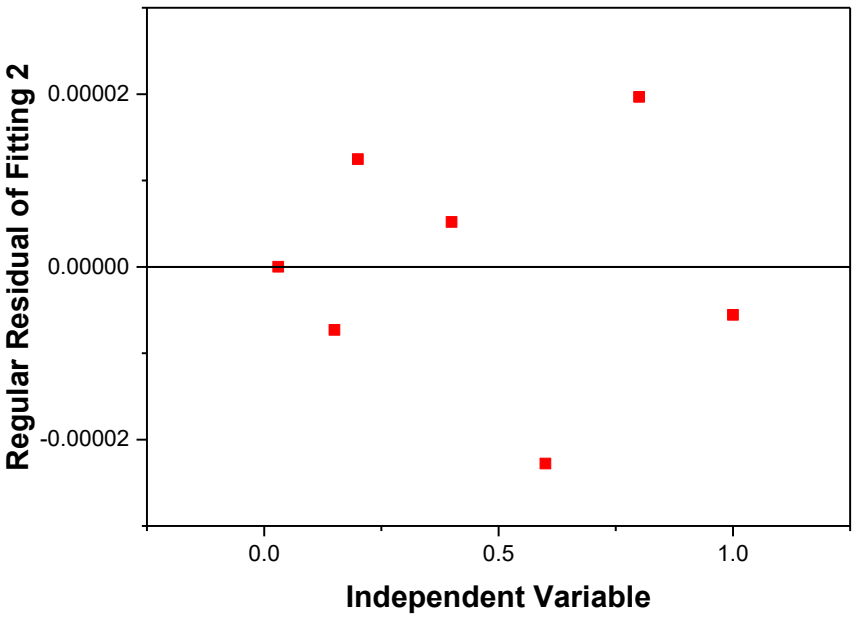

Figure 6S. Residual dispersion for the second-best statistical fitting for k_{obs} of DEDNPP amidolysis versus molar fraction of water.

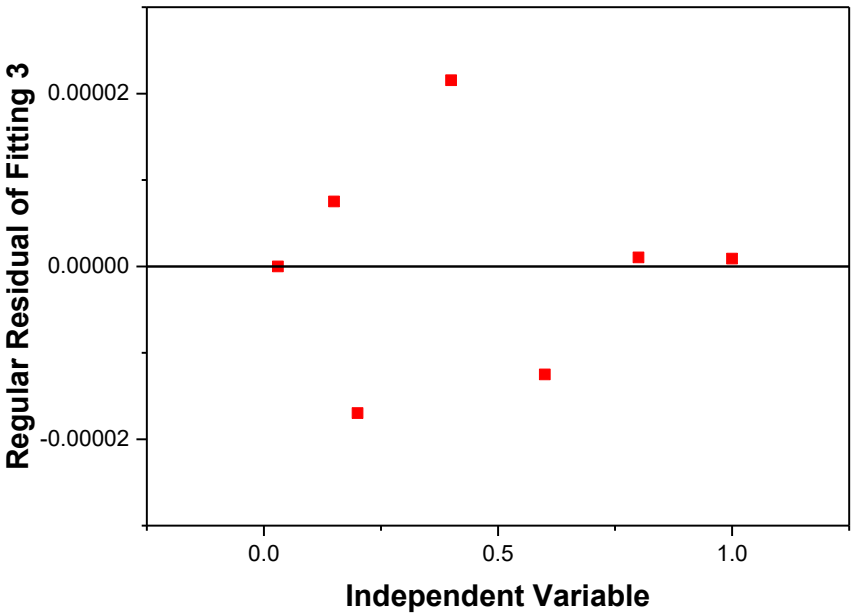

Figure 7S. Residual dispersion for the third-best statistical fitting for k_{obs} of DEDNPP amidolysis versus molar fraction of water.

Table S3. Five empirical scales of the investigated solvents: Dielectric Constant (ε), Dipolarity (SdP), Polarizability (SP), Basicity (SB) and Acidity (SA).

Solvent	ε	SdP[3]	SP[3]	SB[3]	SA[3]
Water	78.3[4*]	0.997	0.681	0.025	1.062
Formamide	109.5[5*]	1.006	0.814	0.414	0.549

3 J. Catalán, *J. Phys. Chem. B*, 2009, **113**, 5951–5960.
4 C. G. Malmberg and A. A. Maryott, *J. Res. Natl. Bur. Stand. (1934).*, 1956, **56**, 1.
5 G. R. Leader, *J. Chem. Soc.*, 1949, **73**, 856–857.
* Values obtained at 25 °C.

2. NMR

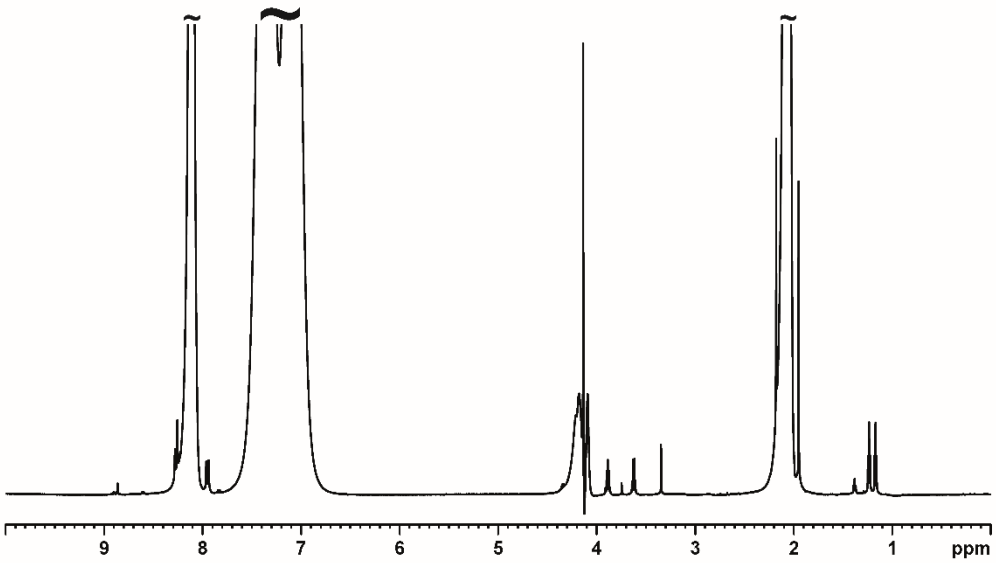

Figure S8. ^{1}H NMR spectrum for the reaction of DEDNPP (2.2 x 10^{-3} mol·L^{-1}) with FMD (2.3 x 10^{-2} mol·L^{-1}) in 10% of DMSO-d6, at pH 9.0 and 25 °C.

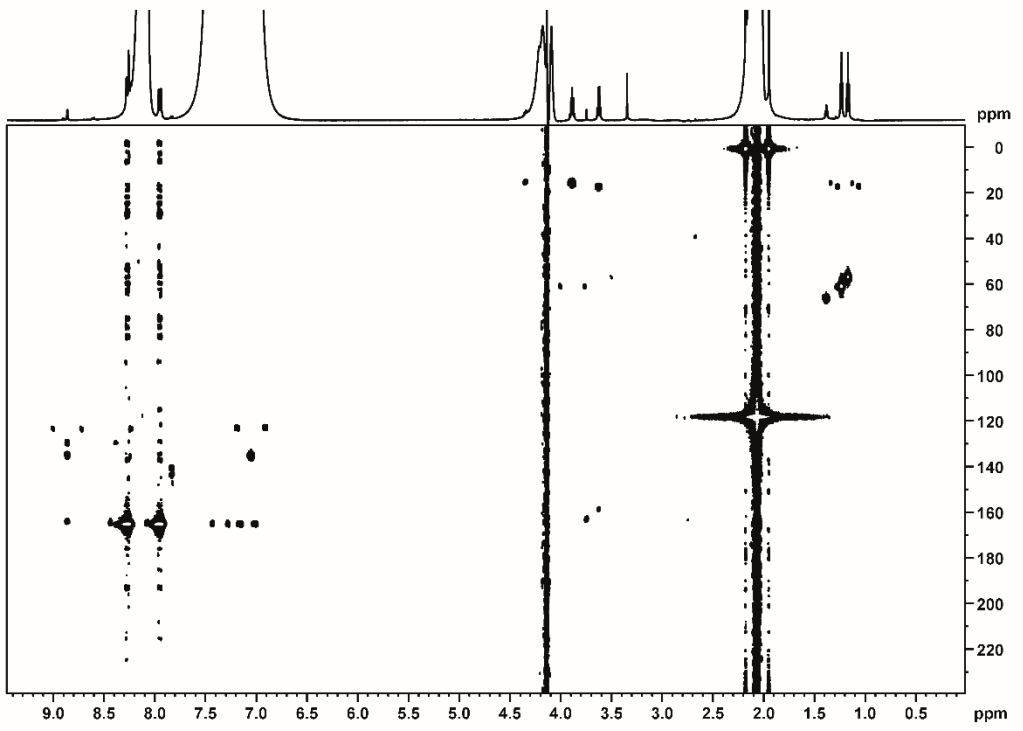

Figure S9. Full long-bond $^{1}H–^{13}C$ correlation map from HMBC NMR experiment at 600 and 151 MHz (Related to the reaction of 2.2 x 10^{-3} mol·L^{-1} DEDNPP with 2.3 x 10^{-2} mol·L^{-1} FMD).

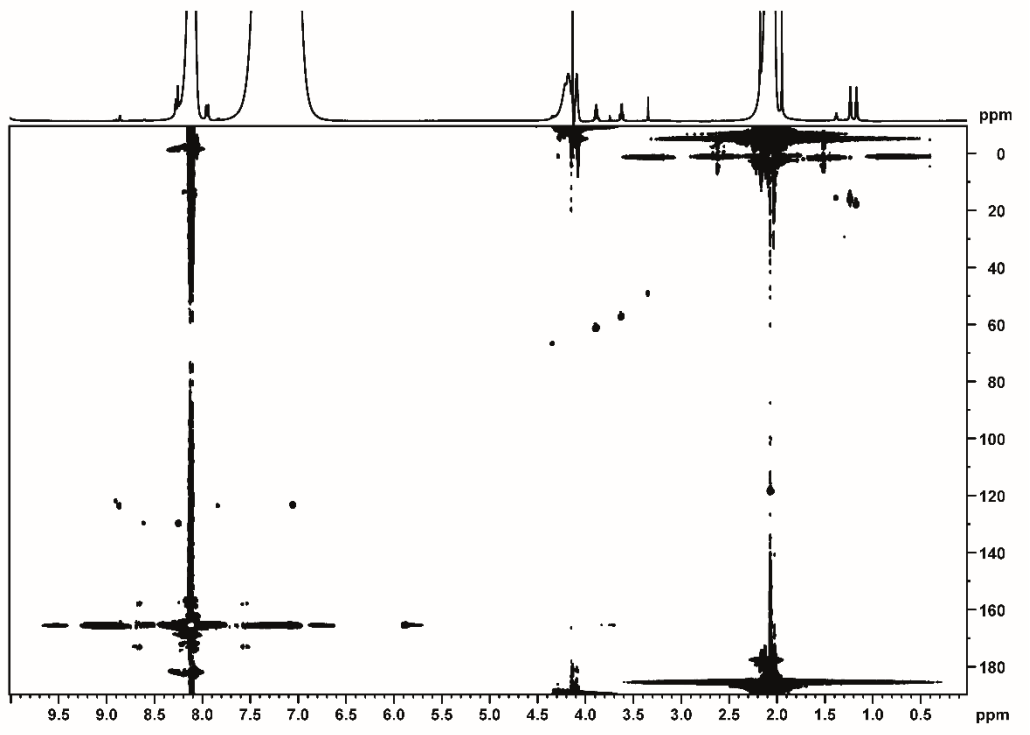

Figure S10. Full one-bond $^{1}H–^{13}C$ correlation map from HSQC NMR experiment at 600 and 151 MHz (Related to the reaction of 2.2 x 10^{-3} mol·L^{-1} DEDNPP with 2.3 x 10^{-2} mol·L^{-1} FMD).

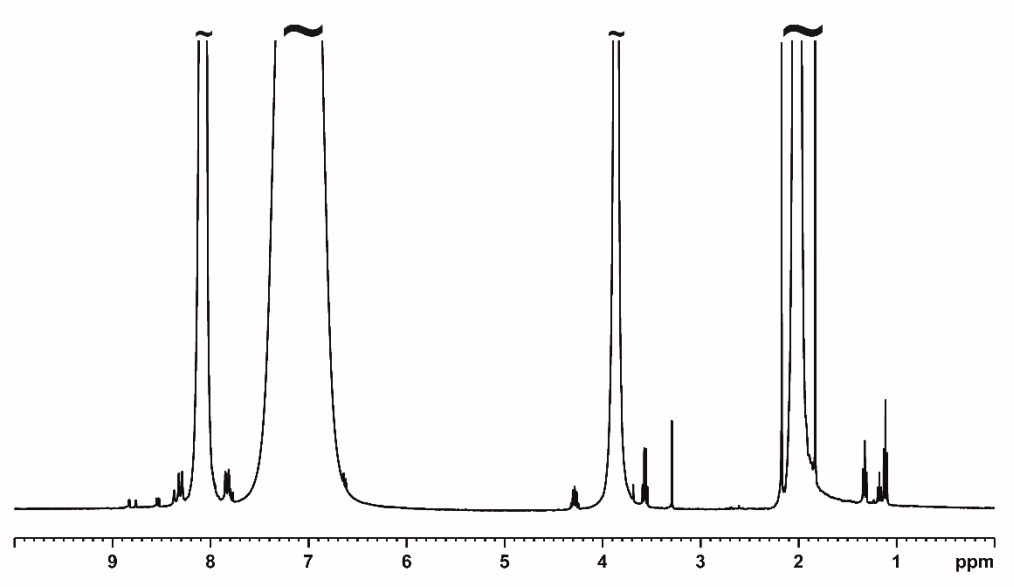

Figure S11. ^{1}H NMR spectrum for the reaction of DEDNPP (2.2 x 10^{-3} mol·L^{-1}) with FMD (2.1 x 10^{-2} mol·L^{-1}) contaning 2.4 x 10^{-3} mol·L^{-1} H_2O and 10% of DMSO-d_6, at pH 9.0 and 25 °C.

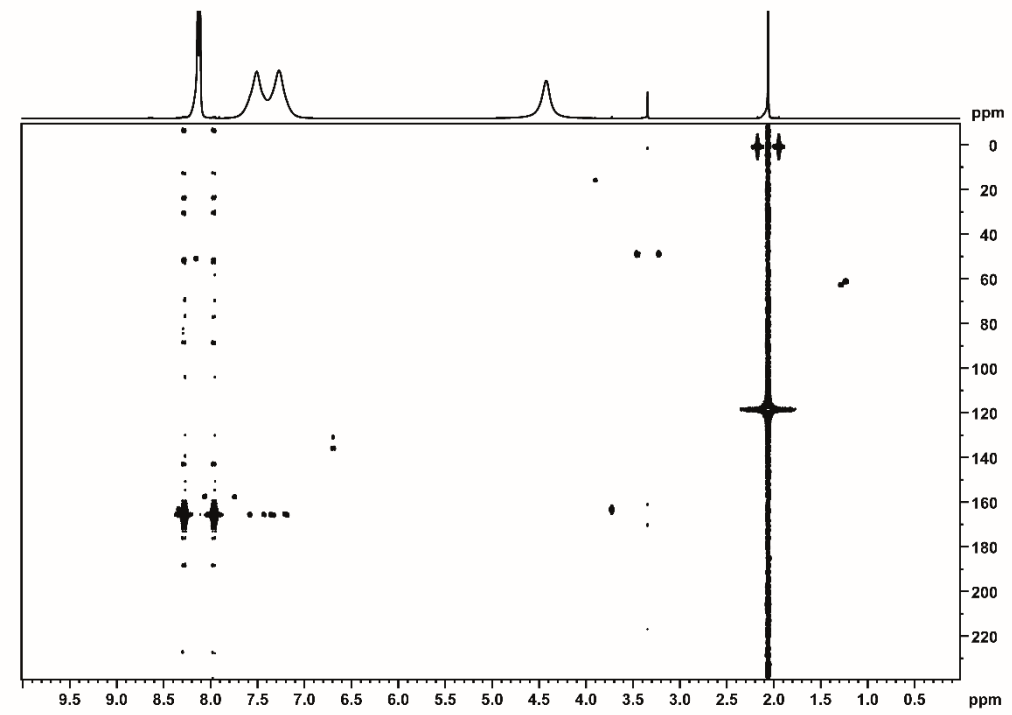

Figure S12. Full long-bond $^{1}H–^{13}C$ correlation map from HMBC NMR experiment at 600 and 151 MHz (Related to the reaction of 2.2 x 10^{-3} mol·L^{-1} DEDNPP with 2.1 x 10^{-2} mol·L^{-1} FMD).

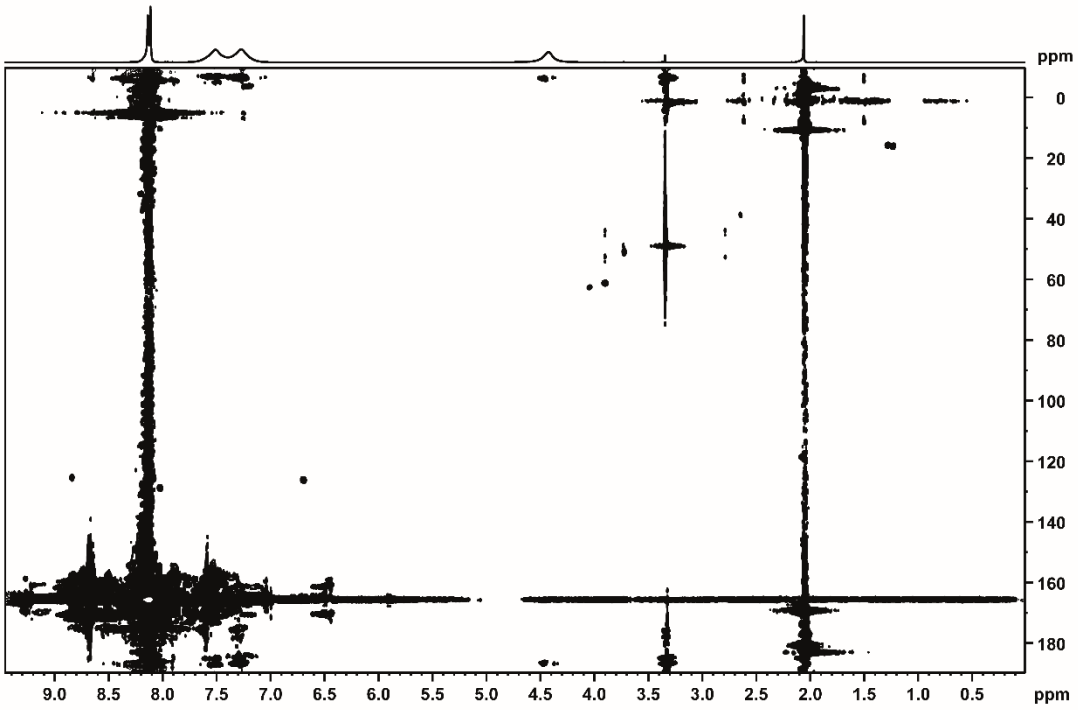

Figure S13. Full one-bond $^{1}H–^{13}C$ correlation map from HSQC NMR experiment at 600 and 151 MHz (Related to the reaction of 2.2 x 10^{-3} mol·L^{-1} DEDNPP with 2.1 x 10^{-2} mol·L^{-1} FMD).

Table S4. NMR (600 MHz, H_2O/DMSO-d_6) data for the species detected in the Reaction of DEDNPP with FMD.

Compound	δ_H	δ_C	δ_P
FMD	8.11 (d 13.7 Hz, 1H)	165.5	
DEDNPP	8.89 (dd 2.7 and 0.8 Hz, 1H), 8.60 (dd 9.1 and 2.7 Hz, 1H), 7.83 (dd 9.2 and 0.8 Hz, 1H), 4.34 (dq, 7.4 and 7.1 Hz, 4H), and 1.38 (td 7.1 and 1.1 Hz, 6H)	15.3, 66.5, 123.5, 126.0, 132.3, 142.7, 148.7, and 150.3	-7.5
INT	7.90 (s, 1H), 4.04 (m, 4H), and 1.28 (t 7.0 Hz, 6H)	157.7, 62.8, and 15.7	12.5
DNP	8.86 (d 3.1 Hz, 1H), 8.02 (dd 9.7 and 3.1 Hz, 1H), 6.69 (d 9.7 Hz, 1H)	123.1, 123.5, 129.6, 136.0, 137.1, and 164.7	
DEP	3.90 (*dq* 7.1 and 7.0 Hz, 4H) and 1.23 (*td* 7.0 Hz, 6H)	16.0 and 61.0	0.8