Tetrahedron Vol 41, No 21, pp 4983 to 4986, 1985
Printed in Great Britain.

0040-4020/85 $3.00 + .00

DECOMPOSITION IN APROTIC SOLVENTS OF 2,4-DIHYDROXY-7-METHOXY-1,4-BENZOXAZIN-3-ONE, A HYDROXAMIC ACID FROM CEREALS

HÉCTOR R. BRAVO and HERMANN M. NIEMEYER*
Facultad de Ciencias, Universidad de Chile, Casilla 653, Santiago, Chile

(*Received in UK 4 October* 1984)

Abstract—The decomposition of the title compound (DIMBOA, 1) in aprotic solvents was analysed in terms of linear solvation energy relationships using donor numbers. The results indicate rate-limiting cyclic hemiacetal opening in low donor number solvents and rate-limiting isocyanate formation in high donor number solvents. The addition of H_2O to DIMBOA decomposing in high donor number solvents had no effect upon the reaction rate, allowing one of the two proposed mechanisms to be rejected.

Extracts of certain Gramineae such as rye, wheat and maize contain hydroxamic acids[1] which inhibit growth and development of plant pathogens[2,3] and are involved in cereal resistance to various insects.[4,5] Knowledge of the reactivity of these compounds in solution is essential for the molecular interpretation of their widespread toxicity.[6,7]

Two mechanisms for the decomposition of these acids in aqueous solutions have been proposed. Both coincide in proposing the fast opening of the hemiacetal as the first step. In one mechanism,[8] an isocyanate intermediate (3) would be formed in the rate-limiting step of the reaction by attack of the hydroxamic oxygen atom on the aldehyde function of the α-ketoaldehyde (2) (Scheme 1, path A). In the other,[9] cyclisation of 2 would form the 5-membered hemiketal 5, water would then add in the rate-limiting step to the aldehyde function of 5 converting it to a better electrophilic leaving group, and finally formic acid and water would be formed from this electrophilic residue and the hydroxamic hydroxyl group, leaving a compound which tautomerizes to the corresponding benzoxazolinone (4). The participation of water may be assessed by studying the effect of added water on the decomposition reaction in aprotic solvents. In this paper, we describe the decomposition of 2,4-dihydroxy-7-methoxy-1,4-benzoxazin-3-one (DIMBOA, 1), the main hydroxamic acid in maize extracts,[10] in aprotic solvents. A preliminary account of this work has been published.[11]

EXPERIMENTAL

Isolation of DIMBOA. DIMBOA was isolated from ethereal extracts of 7-day old seedlings of *Zea mays* L. cv LH Rinconada grown under continuous light in a greenhouse at $28 \pm 3°$.[5] Identification was made by comparison of UV, IR and NMR spectra with reported spectra.[12–14]

Solvents. Solvents were purified and dried by described methods.[15] In addition, they were left 5 hr over 0.3 nm molecular sieves, passed through a 0.4 m column packed with molecular sieves and finally distilled under inert atmosphere.

Kinetics. Decompositions of DIMBOA (0.06 M) were carried out in vessels fitted with teflon-coated silicon septa. Kinetics were followed by treating aliquots of the reaction mixture with $FeCl_3$ reagent (50 g $FeCl_3 \cdot 6\,H_2O$, 500 ml 95% EtOH and 5 ml 10 N HCl) and measuring the absorbance at 590 nm. Absorbances of blanks made with aliquots treated with the solvent of the $FeCl_3$ reagent were subtracted from the absorbance of each sample. Reactions followed first order kinetics for the least 3 half-lives. Standard errors of the rate constants were lower than 4%. Activation parameters were determined from measurements at 3 or 4 different temperatures.

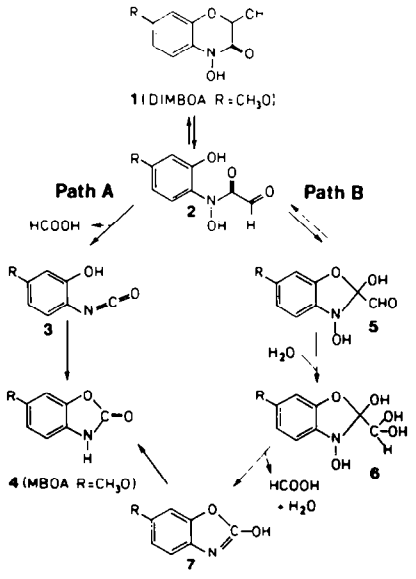
Solubility of DIMBOA. Solns of DIMBOA in equilibrium with solid DIMBOA were prepared in various solvents at 11°. Aliquots were filtered at different time intervals and analysed with $FeCl_3$ reagent. Saturation was assumed when the concentration of DIMBOA in the filtrate did not change with time. Experiments were carried out in triplicate. Standard deviations from the mean were less than 10%.

IR spectra. These were recorded in a Perkin–Elmer 621 spectrophotometer with 0.5 mm light path NaCl cells.

Products studies. The main decomposition product of DIMBOA was 6-methoxy-benzoxazolin-2-one (MBOA, 4), as judged by its melting point (152–153°) and comparison of UV, IR and NMR spectra with resported spectra.[16]

RESULTS

The main decomposition product of 1,4-benzoxazin-3-ones was the corresponding benzoxazolinone both in



Scheme 1.

* Author to whom correspondence should be addressed.

aqueous[16] and non-aqueous solutions,[11] suggesting that the decomposition mechanisms prevailing in both types of solvents are similar.

Although it is reasonable to accept that the opening of the hemiacetal ($1 \rightleftharpoons 2$) in aqueous media is fast compared with subsequent steps in the decomposition reaction, the situation might differ in aprotic solvents. Evidence of a change in the rate-limiting step of a reaction provoked by a change in reaction medium may be obtained through a study of changes in activation parameters with medium.[17]

Rate studies

Rate constants are collected in Table 1. Solvent effects on $\Delta G^{\neq}$ were comparatively small. Hence, values of $\Delta S^{\neq}$ and $\Delta H^{\neq}$ were obtained. They were examined using donor numbers of the solvents, a measure of their ability to donate an electron pair.[18] Two different ranges of $\Delta S^{\neq}$ and $\Delta H^{\neq}$ were obtained: one for low donor number solvents ($\Delta S^{\neq} = -250$ to -170 J mol^{-1} K^{-1}; $\Delta H^{\neq} = 33$ to 59 kJ mol^{-1}) and another for high donor number solvents ($\Delta S^{\neq} = -170$ to 13 J mol^{-1} K^{-1}; $\Delta H^{\neq} = 50$ to 120 kJ mol^{-1}). Within each range, activation parameters correlated linearly with donor number (Fig. 1). An isokinetic relationship was obtained (Fig. 2) with a value of β (298 K) falling within the range expected for solvent effects on reaction rates and equilibria.[17] The proximity of the experimental temperature to the isokinetic temperature presumably accounts for the insensitivity of $\Delta G^{\neq}$ to solvent.

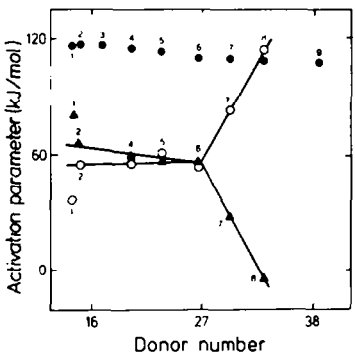


Fig. 1. Activation parameters at 326 K for the decomposition of DIMBOA in aprotic solvents. ●($\Delta G^{\neq}$), ○($\Delta H^{\neq}$), ▲ ($-T\Delta S^{\neq}$). Acetonitrile (1), dioxane (2), acetone (3), tetrahydrofuran (4), trimethylphosphate (5), N,N-dimethylformamide (6), dimethylsulfoxide (7), pyridine (8), hexamethylphosphoramide (9).

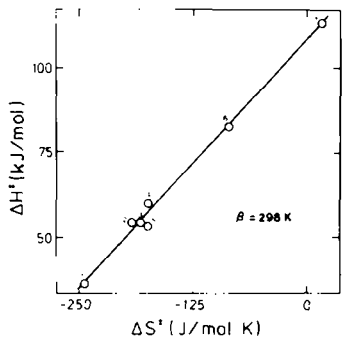


Fig. 2. Isokinetic relationship for the decomposition of DIMBOA in aprotic solvents. Acetonitrile (1), dioxane (2), tetrahydrofuran (3), trimethylphosphate (4), N,N-dimethylformamide (5), dimethylsulfoxide (6), pyridine (7). Correlation coefficient: 0.996.

Water was added to dry high donor number solvents until concentrations ranging from 0.01 to 0.4 M were obtained. This range covered the initial concentration of DIMBOA (0.06 M). The decomposition rates varied erratically within the limits of confidence of the measurements (Table 2).

Insight into the molecular interpretation of activation parameters was obtained through a study of the interaction of DIMBOA with solvent.

Interaction of DIMBOA with solvents

These interactions were assessed by measuring the solubilities and IR spectra of DIMBOA in various solvents.

The logarithms of solubilities correlated linearly with the donor number of the solvents (Fig. 3), pointing to the interaction of the solvent with electrophilic sites in DIMBOA. The nature of this interaction was probed by IR studies of the OH stretching region of solutions of

Table 2. Effect of addition of H_2O on the decomposition rates of 0.06 M DIMBOA in aprotic solvents at 53°

Solvent	H_2O added/M	$10^3\, k_{obs}$/min^{-1}
Dimethylsulfoxide	0.01–0.8	1.08 ± 0.21[a]
Dimethylformamide	0.09–0.34	0.83 ± 0.05[a]
Pyridine	0.05–0.36	1.43 ± 0.11[a]

[a] Average of 5, 3 and 4 points in dimethylsulfoxide, dimethylformamide and pyridine, respectively.

Table 1. Rates of decomposition of 0.06 M DIMBOA at different temperatures in various solvents

Solvent	Rate constants/10^4 min^{-1} (temperature/K) $k_1(T_1)$	$k_2(T_2)$	$k_3(T_3)$	$k_4(T_4)$
Acetonitrile	1.29 (326)	1.76 (326)	2.90 (346)	
Dioxane	1.10 (326)	1.85 (336)	3.36 (346)	
Dioxane + 0.23 M H_2O	1.61 (333)		7.25 (345)	21.7 (353)
Acetone	1.17 (326)			
Tetrahydrofuran	1.25 (316)	2.38 (326)	4.52 (336)	
Dimethylformamide	8.3 (326)	19.1 (336)	27.3 (346)	
Dimethylsulfoxide	10.8 (326)	26.2 (336)	54.5 (346)	163.0 (356)
Dimethylsulfoxide + 0.23 M H_2O		22.6 (336)	61.3 (346)	150.0 (356)
Pyridine	14.3 (326)	43.3 (336)	152.0 (346)	
Hexamethylphosphoramide	26.0 (326)			

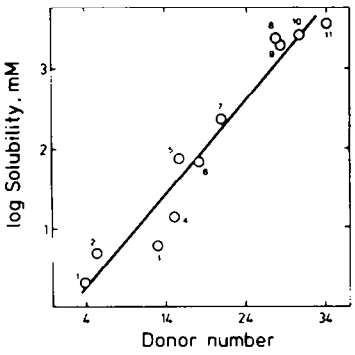


Fig. 3. Solubilities of DIMBOA in aprotic solvents. Nitromethane (1), nitrobenzene (2), benzonitrile (3), acetonitrile (4), dioxane (5), acetone (6), tetrahydrofuran (7), N,N-dimethylformamide (8), N-methyl-2-pyrrolidone (9), dimethylsulfoxide (10), pyridine (11). Correlation coefficient: 0.945.

DIMBOA. The spectra showed absorptions corresponding to unassociated (3700–3400 cm^{-1}) and to associated (3300–3200 cm^{-1}) OH groups. At concentrations lower than 1 mM, hydroxyl groups of DIMBOA were essentially unassociated. Two bands were present in the free OH region of the spectra corresponding to the two hydroxyl groups of DIMBOA. As the donor number of the solvent increased the bands approached one another and were displaced towards the associated OH region. The absorption frequencies correlated with solvent donor number (Fig. 4), reflecting hydrogen bonding to solvent with the corresponding lengthening and weakening of the OH bond.[18,19]

DISCUSSION

The opening of the hemiacetal in DIMBOA requires the transfer of a proton from the hydroxyl group in C_2 to the ring oxygen atom. The rate of this transfer is related in aprotic solvents to the interaction of the proton with solvent through nucleophilic sites in the latter (Fig. 4). It is conceivable that depending on the strength of these interactions, this process may be rate-limiting.

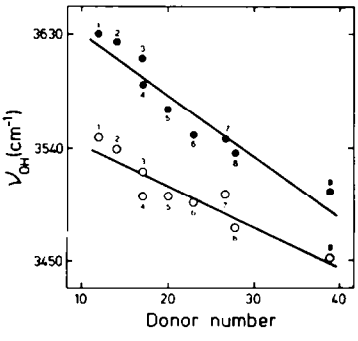


Fig. 4. Hydroxyl stretching frequencies of DIMBOA in aprotic solvents. Benzonitrile (1), acetonitrile (2), dioxane (3), ethylacetate (4), tetrahydrofuran (5), trimethylphosphate (6), N,N-dimethylformamide (7), N,N-dimethylacetamide (8), hexamethylphosphoramide (9). Correlation coefficients: ●, 0.953; ○, 0.911.

The inflection in the activation parameters (Fig. 1) may be attributed to a change in the rate-limiting step of the reaction. The range of $\Delta S^{\neq}$ in low donor number solvents points towards important differences in organization between the basal and transition states in the neighbourhood of the reaction centre. Highly negative $\Delta S^{\neq}$ values may be associated to the rate-limiting opening of the cyclic hemiacetal since a high degree of solvation of the transition state would be required for proton transfer from the hydroxyl group in C_2 to the ring oxygen atom. This difference of solvent organization would be particularly drastic in low donor number aprotic solvents.

In support of this proposition, $\Delta S^{\neq}$ and $\Delta H^{\neq}$ values for the decomposition of DIMBOA in dry dioxane, a low donor number solvent ($\Delta H^{\neq} = 54.3$ kJ mol^{-1}; $\Delta S^{\neq} = -191$ J mol^{-1} K^{-1}), were substantially different from those values in dioxane with H_2O added ($\Delta H^{\neq} = 116$ kJ mol^{-1}; $\Delta S^{\neq} = 7.5$ J mol^{-1} K^{-1}), in spite of the similarity between free energies of activation ($\Delta G^{\neq} = 117$ kJ mol^{-1} in dry dioxane and 114 kJ mol^{-1} in aqueous dioxane). In the first case, hemiacetal opening would be rate-limiting and the activation parameters fall on the line for other low donor number solvents. In the presence of H_2O, hemiacetal opening would become fast and the activation parameters now fall in the range for high donor number aprotic solvents. In contrast, activation parameters in dry dimethylsulfoxide, a high donor number solvent ($\Delta H^{\neq} = 88$ kJ mol^{-1}; $\Delta S^{\neq} = -69$ J mol^{-1} K^{-1}), were not substantially different from those in dimethylsulfoxide with H_2O added ($\Delta H^{\neq} = 91$ kJ mol^{-1}; $\Delta S^{\neq} = -60$ J mol^{-1} K^{-1}). The influence of added H_2O was negligible ($\Delta G^{\neq} = 110$ kJ mol^{-1} in dry dimethylsulfoxide, $\Delta G^{\neq} = 111$ kJ mol^{-1} with H_2O added).

Additionally, activation parameters in acetonitrile deviated from the linear correlation (Fig. 1). The extra negative contribution to $T\Delta S^{\neq}$ in acetonitrile may be the reflection of the participation of this solvent not only as nucleophile but also as electrophile due to the higher electrophilic character of this solvent relative to the others studied. Indeed, the acceptor/donor number ratio is approximately double for acetonitrile (1.34) than for other low donor number solvents employed (dioxane = 0.73, tetrahydrofuran = 0.44, dimethylformamide 0.6).

Hence, the possibility of distinguishing the proposed decomposition mechanisms of DIMBOA based on the addition of H_2O is limited to those solvents where hemiacetal opening is fast, i.e. high donor number aprotic solvents, and those where additional solute–solvent interaction mechanisms are absent. Table 2 shows that added H_2O has no significant effect on the decomposition rate of DIMBOA in high donor number aprotic solvents, suggesting that path B (Scheme 1) is not the predominant mechanism for the decomposition of DIMBOA.

Acknowledgements—This work was supported by Agency for International Development, International Foundation for Science and Universidad de Chile.

REFERENCES

[1] J. I. Willard and D. Penner, *Residue Rev.* **64**, 67 (1976).
[2] L. J. Corcuera, M. D. Woodward, J. P. Helgeson, A. Kelman and C. D. Upper, *Plant Physiol.* **61**, 791 (1978).

[3] M. A. Elnaghy and P. Linko, *Physiol. Plant.* **15**, 764 (1962).
[4] J. A. Klun, C. L. Tipton and T. A. Brindley, *J. Econ. Entomol.* **60**, 1529 (1967).
[5] V. H. Argandoña, H. M. Niemeyer and L. J. Corcuera, *Phytochemistry* **20**, 673 (1981).
[6] C. B. Queirolo, C. S. Andreo, R. H. Vallejos, H. M. Niemeyer and L. J. Corcuera, *Plant Physiol.* **68**, 941 (1981).
[7] C. B. Queirolo, C. S. Andreo, H. M. Niemeyer and L. J. Corcuera, *Phytochemistry* **22**, 2455 (1983).
[8] J. B. Brendenberg, E. Honkanen and A. I. Virtanen, *Acta Chem. Scand.* **16**, 135 (1962).
[9] E. E. Smissman, M. D. Corbett, N. A. Jenny and O. Kristiansen, *J. Org. Chem.* **37**, 1700 (1972).
[10] M. D. Woodward, L. J. Corcuera, J. P. Helgeson, A. Kelman and C. D. Upper, *Plant Physiol.* **63**, 14 (1979).
[11] H. M. Niemeyer, H. R. Bravo, G. F. Peña and L. J. Corcuera, *Chemistry and Biology of Hydroxamic Acids* (Edited by H. Kehl), p. 22. Karger AG, Basel (1982).
[12] C. L. Tipton, J. A. Klun, R. R. Husted and M. D. Pierson, *Biochemistry* **6**, 2866 (1967).
[13] H. E. Gahagan and R. O. Mumma, *Phytochemistry* **6**, 1441 (1967).
[14] J. A. Klun, C. L. Tipton, J. F. Robinson, D. L. Ostrem and M. Beroza, *J. Agric. Food Chem.* **18**, 663 (1970).
[15] J. Riddick and W. B. Bunger, *Techniques of Chemistry* (Edited by A. Weissberger), Vol. II. Wiley-Interscience, New York (1970).
[16] M. D. Woodward, L. J. Corcuera, J. P. Helgeson and C. D. Upper, *Plant Physiol.* **61**, 796 (1978).
[17] J. E. Leffler and E. Grunwald, *Rates and Equilibria of Organic Reactions.* Wiley, New York (1963).
[18] V. Gutmann, *The Donor–Acceptor Approach to Molecular Interactions.* Plenum Press, New York (1978).
[19] T. Kagiya, Y. Sumida and T. Inoe, *Bull. Chem. Soc. Japan* **41**, 767 (1968).