THIN-LAYER AND PAPER CHROMATOGRAPHY OF SOME
1,2-DIOXIMES AND THEIR MONOXIMES

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(Received November 18th, 1970)

SUMMARY

Both ascending thin-layer chromatography on Silica Gel G with developers containing ethers and ascending paper chromatography with dialkyl ethers on papers impregnated with formamide were successful in the separation of the monoximes and dioximes (including their possible stereoisomers) of furil, benzil, biacetyl and cyclohexane-1,2-dione. Selectivities of various separation and visualisation methods as well as the detection limits for some individual oximes were examined and evaluated. Spraying of the developed chromatograms with ammoniacal solutions of Ni(II) and Cu(II) salts followed by UV irradiation for the isomeric oximes (γ, β) and/or by oximation of the monoximes gave the best results in checking the purity and isomeric composition of the oxime reagents tested.

INTRODUCTION

The use of 1,2-dioximes and their monoximes in inorganic analysis[1] as well as in studying metal chelate equilibria[2,3] is very extensive, but appropriate attention is not always paid to the purity of these reagents. It refers namely to the oxime derivatives of the 1,2-diketones R-CO-CO-R', if at least one of the substituents R, R' represents an aryl group or its heterocyclic analogue. In agreement with the theory[4-7], these oximes can form relatively more stable stereoisomers which differ from each other in their physical and chemical properties. Among three known stereoisomers of the dioximes with both substituents R and R' aromatic, the α-isomer only (having the configuration *anti-* in MEISENHEIMER's nomenclature[4,5]) gives typical metal chelates. The γ-isomer (possessing the *amphi*-configuration) reacts nonselectively forming 1 metal : 1 dioxime salts in basic media. The β-isomer (with the *syn*-configuration) does not react with metal ions at all. The monoximes with both substituents R and R' aromatic can exist as two stereoisomers, *i.e.* in the chelating α-form and the inactive β-form with *anti* and *syn* configurations, respectively, of the oxime group with respect to the carbonyl[4,5]. In the case of the dioximes and monoximes having merely aliphatic or alicyclic substituents, the existence of stereoisomers was not observed; commercial preparations of these reagents, however, often contain monoxime impurities in the dioximes and *vice versa*.

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Although it is evident that chromatographic methods are able to play an important role in the analysis of the oxime reagents, only a few communications have been published in this field. The components found in commercial preparations of furil dioxime[8-10] were separated by paper chromatography (PC) using formamide and diethyl ether as stationary and mobile phases, respectively[9], as well as by thin-layer chromatography (TLC) on Silica Gel G with a carbon tetrachloride–petroleum ether–diethyl ether–ethanol (12:8:4:1.5) mixture[10]. In the latter paper, the chromatographically separated components were not identified. The individuality of alicyclic dioximes having rings with 5 to 12 carbon atoms was checked by TLC on Silica Gel G with a benzene–carbon tetrachloride–chloroform (80:15:15) solvent mixture[11].

In the present paper, the migration behaviour of some analytically valuable oxime reagents and their usual admixtures was examined by ascending chromatography with various solvents on thin layers of Silica Gel G as well as on papers impregnated with formamide in order to find simple qualitative tests of purity for these reagents.

EXPERIMENTAL

Chemicals and reagents

The list of the substances chromatographed is given in Table II, together with their symbols and the preparative methods used. The oximes were either synthesised according to the original methods or used as commercially supplied products (Lachema). Many commercial as well as freshly synthesised preparations were not individual substances (see also Table V) but owing to the detection selectivity, most chromatographically separated components could be identified even without laborious isolation and application as standards. In general, α- and γ-dioximes as the natural products of the oximation of aromatic 1,2-diketones were isolated by fractional crystallisation from ethanol. β-Monoximes were prepared by heating the corresponding α-monoximes with charcoal in benzene[12] and β-dioximes by heating the α- or γ-dioximes in aniline[13] or *n*-amyl alcohol[8] under reflux for several hours. Cyclohexane-1,2-dione monoxime was obtained by hydrolysis of its dioxime in 1 N sulphuric acid and 50 % (v/v) ethanol at room temperature. After 24 h, the products of the hydrolysis were extracted into diethyl ether. In addition to the dioxime and monoxime, other more slowly migrating species were present in the extract but among them the monoxime only provided the dioxime by subsequent oximation.

All other chemicals and solvents used were of pure or *pro analysi* quality from Lachema N.E., Brno. The 95 % (v/v) ethanol contained an addition of 5 % of methanol, chloroform contained 1 % of ethanol. All solvents were distilled freshly before use. Ethylene glycol (pure, Lachema) and formamide (pure, Reakhim, U.S.S.R.) were used directly without any preliminary purification.

Thin-layer chromatography

The glass plates 20 × 20 cm were coated with MN-Silica Gel G using the apparatus and the procedure described by Stahl[15]; the layer thickness was nominally 0.25 mm. The same results, differing only a little in R_F value, were also achieved with Silica Gel CH-X (Lachema, grain size 5–40 μ) containing 13 % of plaster. The Silica Gel G layers were activated by heating for 90 min at 105° and deactivated again

before use by standing in air (at room temperature 20 ± 2° and 50–60 % relative humidity) for about 5–24 h.

The samples to be analysed were applied (as solutions in ethanol, benzene, acetone, or diethyl ether) to the start in appropriate amounts with regard to the sensitivity of their detection (between 1 to 50 μg of each oxime to one starting point). Solutions of some oxime derivatives of furil and benzil were stored in amber glass bottles to prevent isomerisation changes caused by daylight irradiation (see also Fig. 2).

The chromatograms were developed by the ascending technique in a standard large volume unsaturated N-chamber (Desaga, Heidelberg, G.F.R.) up to a solvent front distance of 15 cm from the start. The development was started immediately after the introduction of the solvent. After removing the mobile phase in a stream of hot air, the developed chromatograms were sprayed with the detection reagents described in Table I.

Paper chromatography

Impregnation. Whatman No. 1 chromatographic papers in a rectangular form 47 × 18.5 cm were immersed twice into an ethanolic solution containing a chosen volume percentage of formamide as the impregnant. In order to remove excess impregnating solution from the paper, a procedure recommended by CHURÁČEK[16] was used.

Application of samples. After the impregnated paper had been dried for 15–30 min in air, sample solutions in ethanol or diethyl ether were applied to the starting line placed at a distance of 2.5 cm from the longer side of the chromatogram. About 5–50 μg of the oxime was placed on each starting point according to the sensitivity of the detection. Distances between the neighbouring starting points were 2.5 cm.

TABLE I

DETECTION LIMITS OF SOME OXIMES CHROMATOGRAPHED ON THE SILICA GEL G THIN LAYERS

Evaluated visually 60 min after spraying (except the detection with KMnO$_4$). Spot coloration given in parenthesis: R = red, Y = yellow, G = green, Br = brown, O = orange, Bl = blue, P = pink. Spray solutions (water solution, volume ratios given): (D 1) 0.05 M NiSO$_4$ + 10 % NH$_3$ (1 : 1); (D 2) 0.05 M CuSO$_4$ + 10 % NH$_3$ (1 : 1); (D 3)[a] 0.05 M CuSO$_4$ + concentrated NH$_3$ (1 : 1); (D 4)[a] 0.05 M Fe(NH$_4$)$_2$(SO$_4$)$_2$ + gaseous NH$_3$; (D 5) 0.1 N KMnO$_4$.

Oxime	Detection limits (in μg) for the reagents				
	D 1	D 2	D 3	D 4	D 5
Biacetyl monoxime	1 (P)	20 (G)	2 (O)	30 (Bl)	2 (YG)
α-Furil monoxime	0.5 (YG)	1 (YG	0.1 (O)	5 (Bl → Y)	0.1 (YG)
α-Benzil monoxime	2 (YG)	2 (YG)	0.2 (O)	5 (Bl)	0.5 (YG)
Dimethylglyoxime	0.1 (R)	0.5 (YBr)	0.5 (YBr)	0.5 (RBr)	0.2 (YG)
Cyclohexane-1,2-dione dioxime	0.2 (R)	0.5 (YBr)	0.5 (YBr)	1 (R)	0.2 (YG)
α-Furil dioxime	0.2 (R)	0.2 (Br)	0.5 (YBr)	1 (R)	0.1 (YG)
γ-Furil dioxime	1 (YBr)	0.5 (Br)	0.5 (YBr)	2 (O)	0.1 (YG)
α-Benzil dioxime	0.1 (R)	0.5 (YG)	1 (YBr)	30 (RBr)	0.5 (YG)
β-Benzil dioxime	0.2 (O)	30 (YG)	30 (YBr)	30 (P)	0.5 (YG)

[a] Prepared freshly before use.

Development. The chromatograms were developed by an ascending method described by WILLIAMS AND KIRBY[17] to a distance of 15 cm between the solvent front and the start. As a chamber, a cylindrical desiccator with a tightly fitting and loaded cover was employed, the inner walls of which were lined with filter paper. Thorough chamber saturation as well as the cover loading proved to be necessary especially when using diethyl ether as the mobile phase.

Detection. As a rule, the paper chromatograms were sprayed first with an ammoniacal solution of a Ni(II) or Cu(II) salt. After the detection results had been noted, the γ- and β-oximes were converted into the α-isomers (giving more sensitive reactions) by irradiating them with a mercury lamp for about 1 h. Using an additional spray with 5 % hydroxylamine hydrochloride in 2 N sodium hydroxide, a very sensitive test for α-monoximes was obtained.

RESULTS AND DISCUSSION

Sensitivity and selectivity of the detection

Detection limits for some oxime derivatives together with the composition of the spray reagents used here (for TLC) are given in Table I. α-Dioximes and α-monoximes react immediately with an ammoniacal nickel salt solution to give characteristically coloured chelates. Coloured spots of γ-dioximes also appear in a few minutes (or hours for very small amounts of oxime), while β-dioximes and β-monoximes can be detected in this way only if the sprayed chromatograms have been ex-

TABLE II

R_F VALUES OF THE OXIME DERIVATIVES OF SOME 1,2-DIKETONES ON SILICA GEL G THIN LAYERS

Preparation of samples: A, commercial preparation purchased from Lachema (trade name in parenthesis); the other samples were obtained by: B, fraction crystallisation of oximation products; C, boiling α- or γ-isomers in amyl alcohol or aniline under reflux; D, acid hydrolysis of dioxime; E, boiling α-monoxime in benzene with charcoal under reflux.

Oxime	Abbrev.	Preparation	R_F for solvent systems[a]			
			S1	S2	S3	S4
α-Furil monoxime	α-FMH	A, B, D,	0.51	0.11	0.23	0.15
β-Furil monoxime	β-FMH	B, E	0.43	0.16	0.18	0.12
α-Furil dioxime	α-FH$_2$	B, A (Neoniklon)			0.16	0.12
γ-Furil dioxime	γ-FH$_2$	B, A (Neoniklon)	0.6–	<0.1	0.13	0.08
β-Furil dioxime	β-FH$_2$	C	0.4		0.11	0.08
α-Benzil monoxime	α-BMH	B, D,	0.78	0.30	0.54	0.47
β-Benzil monoxime	β-BMH	B, E	0.71	0.40	0.47	0.41
α-Benzil dioxime	α-BH$_2$	B, A (Niklon)	0.61	<0.1	0.23	0.15
γ-Benzil dioxime	γ-BH$_2$	B, A (Niklon)	0.61	<0.1	0.23	0.15
β-Benzil dioxime	β-BH$_2$	C	0.61	<0.1	0.23	0.15
Biacetyl monoxime	DMH	A, D	0.79	0.33	0.60	0.49
Dimethylglyoxime	DH$_2$	A	0.79	0.07	0.33	0.23
Cyclohexane-1,2-dione						
monoxime	CyMH	D	0.16	0.05	0.16	0.15
dioxime	CyH$_2$	A (Nioxime)	0.13	0	0.11	0.11
Benzoylmethylglyoxime		A (Palladon)	0.71	0.06	0.27	0.13
			0.61	0.05	0.14	0.07
			0.50	0.05		

[a] For composition of solvents see Table III.

posed to diffused daylight for several days, or to sunlight or UV radiation for a shorter time. The coloration of the β-monoxime and dioxime spots is then similar to that of the α-isomers and the sensitivity of their detection increases as well. Among other reasons, these isomerisation changes caused by irradiation make the preparation of an isomerically pure oxime difficult particularly in the case of the benzil dioximes; this observation is in a good agreement with the literature[14].

The sensitivities for the detection of monoximes (giving less intensely coloured nickel chelates) can also be raised to the dioxime detection level by an additional oximation with 5 % hydroxylamine hydrochloride solution in 2 N sodium hydroxide solution.

When solutions of Cu(II), Co(II), and Fe(II) salts and ammonia are used for the detection, similarly coloured spots of α- and γ-dioximes appear immediately while β-dioximes react distinctly more slowly. The coloration of the chelates of Cu(II) and Co(III) with α-monoximes is similar to that of the corresponding dioximes. α-Monoximes give a less sensitive "iron blue" reaction; the blue monoxime spots change slowly into the reddish brown ones of the dioxime; the rate being proportional to the excess of Fe(II) and the alkalinity of the medium.

All the oximes considered including β-monoximes and β-dioximes can also be detected nonselectively with a neutral permanganate solution giving yellow-green spots on a pink-brown background; unfortunately the chromatograms obtained are not too stable.

Combining the above detection methods on the same chromatogram, all oxime

$S\ 5$	$S\ 6$	$S\ 7$	$S\ 8$	$S\ 9$	$S\ 10$	$S\ 11$	$S\ 12$	$S\ 13$
0.55	0.83	0.40	0.60	0.51	0.53	0.30	0.24	0.71
0.51	0.76	0.33	0.55	0.49	0.49	0.26	0.19	0.64
						0.25	0.22	
0.5–	0.5–	<0.1	0.5–	0.4–	0.4–	0.20	0.16	0.2–
0.3	0.3		0.3	0.3	0.2	0.17	0.10	0.0
0.77	0.97	0.75	0.95	0.81	0.80	0.56	0.53	0.93
0.77	0.97	0.71	0.91	0.81	0.78	0.53	0.49	0.91
0.47	0.84	0.30	0.57	0.45	0.41	0.31	0.28	0.59
0.47	0.69	0.17	0.48	0.45	0.41	0.28	0.27	0.37
0.52	0.74	0.24	0.51	0.50	0.47	0.28	0.27	0.47
0.75	0.93	0.74	0.87	0.75	0.71	0.69	0.75	0.91
0.62	0.73	0.39	0.59	0.61	0.50	0.45	0.63	0.64
0.12	0.33	0.23	0.55	0.12	0.11	0.25	0.27	0.13
0.08	0.21	0.12	0.44	0.09	0.08	0.17	0.21	0.08
0.67	0.82	0.36	0.62	0.65	0.60	0.26	0.32	0.68
0.53	0.68	0.25	0.38	0.42	0.24	0.13	0.19	0.49
0.31	0.35	0.06						0.15

and even non-oxime reducing species can be easily localised and identified. Detection with an ammoniacal solution of Ni(II) (followed by UV irradiation for 1–2 h) and with permanganate gave the most reliable results in our experience.

Thin-layer chromatography of oximes

In general, chromatographic separations of monoximes from dioximes on thin layers of Silica Gel G are quite efficient and depend very little on the nature of the developing solvents. However, a successful separation of isomeric monoximes or dioximes of the same diketone requires the presence of an ether in the developing solvent.

Some of the solvent systems examined with the corresponding R_F values are given in Tables II and III; the tables include only data for the mobile phases giving well defined circular or ellipse-shaped spots, free of tailing. Of course, the R_F values demonstrate above all the relative differences in the migration of the individual oximes and can vary to some degree on various chromatograms. In some cases, we were successful in limiting the R_F changes, tailing, and edge effects by using a longer activation time (90 instead of the usual 30 min), deactivation, and chromatography in an unsaturated chamber. A longer deactivation period resulted in insufficient resolution of the spots, a shorter one in lower R_F values. The ascending development to a solvent front distance of 15 cm from the start took 50–70 min on average.

In most cases, monoximes migrate more rapidly than the dioximes in the solvent systems 1–13, but none of these mobile phases is able to provide complete separation of all the oximes examined of the various 1,2-diketones. Therefore the suitability of each solvent for individual oxime groups is evaluated and discussed separately (see Table III).

As to the group of possible oxime derivatives of furil, the solvent systems 3, 4, 11, and 12 are most suitable for the separation of α-, γ-, β-FH$_2$, while for the separation of α- and β-FMH, solvents 1,3–8, and 11–13 are preferable (Fig. 1). The systems 4, 11, and 12 were formed by the replacement of diethyl ether by its less volatile higher homologues in an assumed solvent mixture[10]. The last two solvents guarantee outstanding reproducibility and large differences in R_F values for neighbouring spots of isomeric furil oximes. A certain disadvantage of these solvents, however, *viz.* the close vicinity, up to coincidence, of the β-FMH and α-FH$_2$ should be mentioned. Therefore in the presence of α-FH$_2$, both isomeric monoximes should be chromatographed with solvents 7, 8, and 13, where the distances between β-FMH and α-FH$_2$ are sufficient. If, however, α-FH$_2$ is absent, solvents 11 and 12 exhibit the optimum resolution efficiency for α- and β-FMH, only comparable with that of solvent 13.

All three isomers of BH$_2$ can be separated with R_F values decreasing in sequence $\alpha > \beta > \gamma$, if solvents 6, 7, and 13 are used (Fig. 2). With solvents 5, 9, and 10, this sequence changes, so that β-BH$_2$ migrates more rapidly than the other two isomers whose spots almost coincide. With respect to this fact, the latter solvents cannot be used for the analytically valuable separation and detection of γ-BH$_2$ in preparations of α-BH$_2$. In contrast to furil derivatives, both benzil monoximes travel far ahead of benzil dioximes in most developing systems. The separation of α- and β-BMH is possible with solvents 1, 3, 4, 8, and 12.

Although the solvent systems 1–13, above, have all been devised for the chromatography of isomeric oximes of furil and benzil, they also give efficient separations

TABLE III

SOLVENT SYSTEMS USED FOR TLC SEPARATION OF OXIME DERIVATIVES

Solvent	Composition (v/v)	Suitable for separation of the oximes named between two adjoining semicolons
S 1	Diethyl ether–dibutyl ether (1:1)	CyMH, CyH₂; FMH (α, β); BMH (α, β); Palladon
S 2	Chloroform (with 1% of ethanol)	Monoximes from dioximes
S 3	Petroleum ether–carbon tetrachloride–diethyl ether–ethanol (8:12:4:1.5)	All oxime groups except BH₂ (α, γ, β)
S 4	Cyclohexane–carbon tetrachloride–dibutyl ether–ethanol (10:10:4:2.5)	All oxime groups except BH₂ (α, γ, β)
S 5	Benzene–chloroform–tetrahydrofuran (16:1:3)	All oxime groups except BH₂ (α, γ, β).
S 6	Chloroform–dioxane (5:1)	BMH (α, β) and larger amounts of FH₂ (α, γ, β)
S 7	Cyclohexane–chloroform–dioxane (4:5:1)	All groups except FH₂ (α, γ, β) and BMH (α, β)
S 8	Cyclohexane–chloroform–dioxane–ethanol (4:5:1:1)	All groups except FH₂ (α, γ, β)
S 9	Cyclohexane–benzene–chloroform–tetrahydrofuran (2:16:1:3)	All groups except FH₂ (α, γ, β)
S 10	Benzene–chloroform–tetrahydrofuran (24:1:3)	DMH, DH₂; CyMH, CyH₂; BH₂ (γ, β); Palladon
S 11	Petroleum ether–carbon tetrachloride–diisopropyl ether–ethanol (14:13:4:2)	DMH, DH₂; CyMH, CyH₂; BH₂ (γ, β); Palladon
S 12	Petroleum ether–diisopropyl ether–ethanol (17:5:5:2.5)	All groups except BH₂ (α, γ, β)
S 13	Chloroform–dioxane (9:1)	All groups except BH₂ (α, γ, β); FMH (α, β); BH₂ (α, γ, β); Palladon; DMH, DH₂; CyMH, CyH₂

TABLE IV

R_F VALUES OF OXIME DERIVATIVES OF SOME 1,2-DIKETONES ON WHATMAN No. 1 PAPER IMPREGNATED WITH FORMAMIDE USING DIALKYL ETHERS AS THE DEVELOPERS

Developing solvents: A, diethyl ether; B, diisopropyl ether; C, di-n-butyl ether; D = B + C (45:5); E = B + C (30:20) (the volume ratios are given).

| Oxime	Abbrev.	10% HCONH₂			20% HCONH₂					30% HCONH₂			40% HCONH₂		
		A	B	C	A	B	C	D	E	A	B	C	A	B	C
α-Furilmonoxime	α-FMH	0.90	0.81	0.46	0.95	0.85	0.42	0.60	0.40	0.87	0.68	0.21	0.36	0.31	0.11
β-Furil monoxime	β-FMH	0.90	0.80	0.34	0.95	0.75	0.29	0.49	0.26	0.85	0.61	0.13	0.25	0.24	0.11
α-Furil dioxime	α-FH₂	0.86	0.69	0.22	0.91	0.70	0.19	0.42	0.23	0.81	0.45	0.09	0.34	0.18	0.04
γ-Furil dioxime	γ-FH₂	0.80	0.51	0.14	0.83	0.51	0.12	0.29	0.15	0.60	0.28	0	0.20	0.10	0
β-Furildioxime	β-FH₂	0.69	0.28	0.07	0.72	0.29	0.06	0.15	0.08	0.41	0.13	0	0.11	0	0
α-Benzil monoxime	α-BMH	0.96	0.92	0.90	0.97	0.97	0.94	—	—	0.97	0.97	0.84	0.75	0.73	0.77
β-Benzil monoxime	β-BMH	0.96	0.92	0.93	0.96	0.98	0.98	—	—	0.97	0.97	0.85	—	0.73	0.75
α-Benzil dioxime	α-BH₂	0.90	0.83	0.65	0.95	0.89	0.69	—	—	0.83	0.79	0.48	0.37	0.35	0
γ-Benzil dioxime	γ-BH₂	0.92	0.84	0.65	0.95	0.89	0.69	—	—	0.83	0.89	0.48	0.47	0.50	0.39
β-Benzil dioxime	β-BH₂	0.92	0.87	0.67	0.95	0.94	0.69	—	—	0.83	0.87	0.48	0.49	0.50	—
Biacetyl monoxime	DMH	0.95	0.82	0.62	0.81	0.69	0.46	—	—	0.76	0.70	0.47	0.57	0.57	0.31
Dimethylglyoxime	DH₂	0.95	0.77	0.47	0.81	0.67	0.33	—	—	0.75	0.66	0.32	0.57	0.53	0.20
Cyclohexane-1,2-dione monoxime	CyMH	0.48	0.12	0.08	0.47	0.12	0.04	—	—	0.30	0.11	0	0.06	0	0
dioxime	CyH₂	0.48	0.13	0.03	0.46	0.12	0.04	—	—	0.30	0.05	0	0.06	0	0
Benzoylmethylglyoxime (Palladon)		0.92	0.92	0.45	0.92	0.89	0.45	—	—	0.77	0.73	0.25	0.49	0.35	0.11
		0.77	0.70	0.05	0.76	0.50	0.05	—	—	0.57	0.26	0	0.29	0.11	0
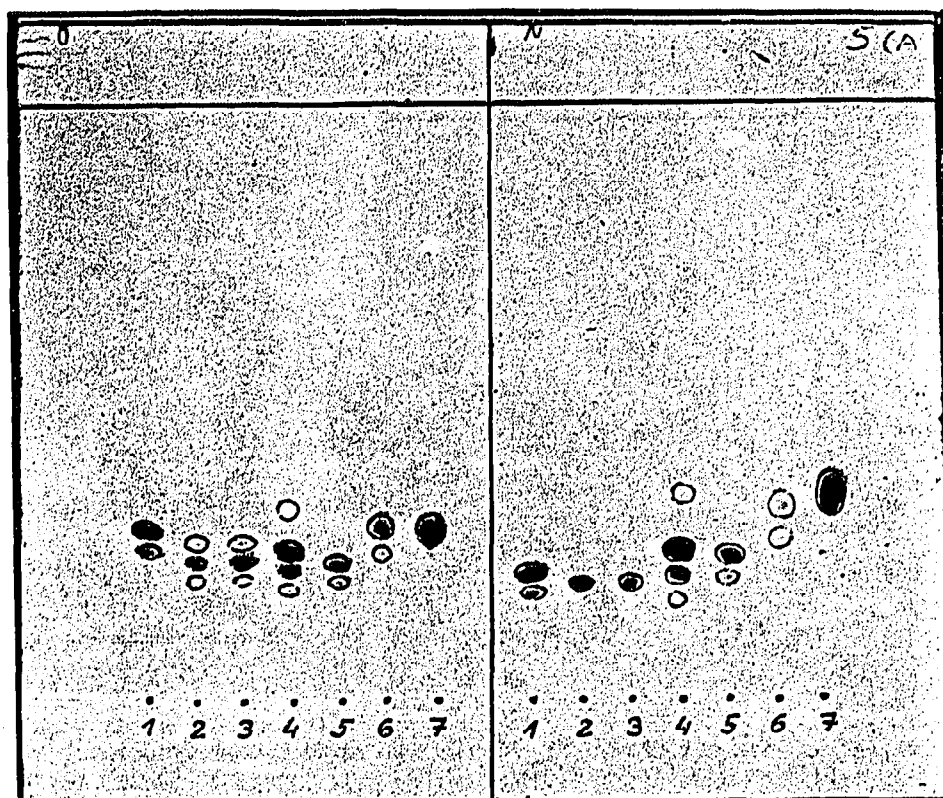
Fig. 1. Separation of oxime derivatives of furil on Silica Gel G thin layers using petroleum ether–carbon tetrachloride–diisopropyl ether–ethanol (14:13:4:2) developer (S11). The chromatographed samples contained (from top to bottom): (1) α-FH$_2$, γ-FH$_2$; (2 and 3) γ-FH$_2$; (4) α-FMH, α-FH$_2$, γ-FH$_2$, β-FH$_2$; (5) α-FH$_2$, γ-FH$_2$; (6) α-FMH, β-FMH; (7) α-FMH. Right-hand side: as described in the text; left-hand side: the sample spots applied to the start were irradiated with UV light for about 30 min before development. As a result of this irradiation, γ-FH$_2$ provided all three isomers of FH$_2$.

of dimethylglyoxime and cyclohexane-1,2-dione dioxime from their monoximes (see Tables II and III). Commercial preparations of benzoylmethylglyoxime (so-called Palladon) are also separated by these solvents into two or three, up to this time, unidentified components giving analogous colour reactions with the chelate forming detection reagents.

While the separation of monoximes from dioximes is only affected to a small extent by the amount of the samples to be chromatographed, some separations of similarly migrating isomeric oximes (especially in the case of the benzil derivatives) are of limited capacity. In such cases, several sample quantities ought to be chromatographed simultaneously with known amounts of standard mixtures on the same chromatogram.

Attempts were also made to determine oximes separated in this way by UV absorption spectrophotometry, but their incomplete desorption even by polar eluting solvents as well as the presence of very finely dispersed adsorbent in the eluates caused significant errors.

Paper chromatography of oximes

The oximes under discussion having little solubility in water and being compounds of medium polarity cannot be chromatographed on paper with only water
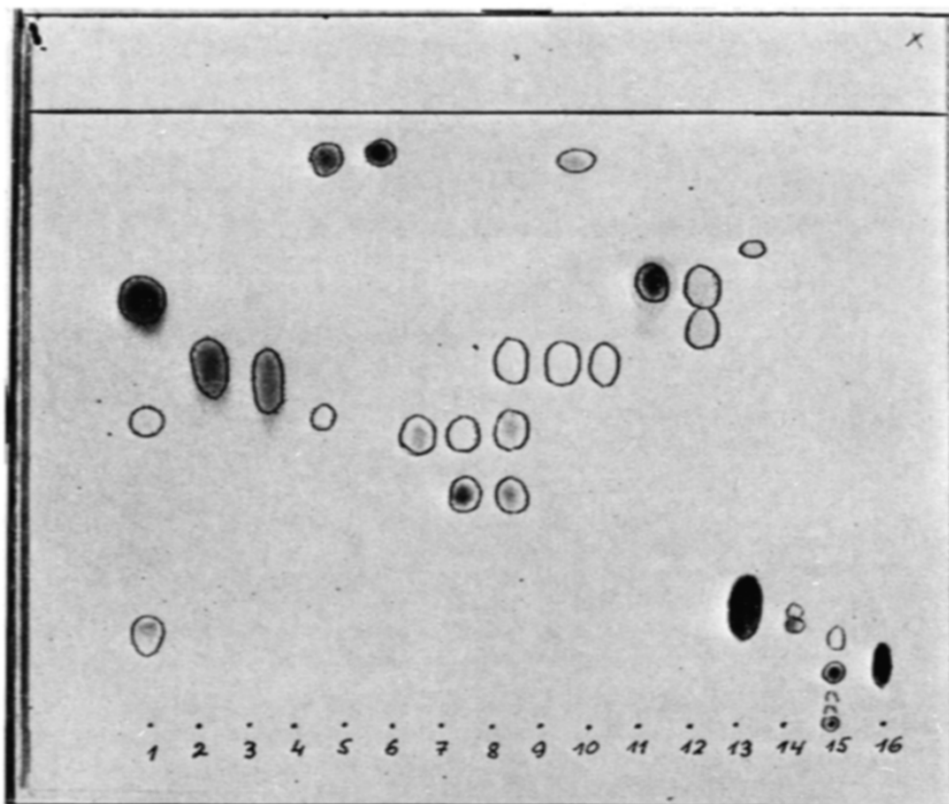
Fig. 2. Separation of isomeric furil monoximes and benzil dioximes on Silica Gel G layers using chloroform–dioxane (36:4) developer (S 13). Sample spots (from top to bottom): (1) Palladon (Lachema); (2) DMH; (3) DH₂; (4) α-BMH, β-BMH; (5) α-BMH; (6) β-BH₂; (7) β-BH₂, γ-BH₂; (8) α-BH₂, β-BH₂, γ-BH₂; (9) α-BMH, α-BH₂; (10) α-BH₂; (11) α-FMH; (12) α-FMH, β-FMH; (13) α-FMH, (α + γ) FH₂; (14) α-FH₂, γ-FH₂; (15) CyMH, CyH₂, their decomposition products; (16) Nioxime (Lachema).

as stationary phase. Among the polar organic solvents (ethylene glycol, dimethylformamide, formamide) tested in our preliminary experiments, formamide as the stationary phase and dialkyl ethers as the mobile phase were found to give the best results. Much better separations were also obtained by an ascending method than with a conventional descending one. Attempts to replace dialkyl ethers, at least in part, by other solvents (e.g. benzene, chloroform) did not yield a desirable effect.

The results obtained on Whatman No. 1 paper impregnated with formamide by ascending chromatography with dialkyl ethers or their mixtures are given in Table IV. In general the R_F values of most oximes decrease with the increasing molecular weight of the ether used as the mobile phase, and/or with the increasing fraction of higher ether in ether mixtures. Similarly the R_F values for the same ether decrease proportionally to the increasing formamide content in paper.

The developing times for the systems examined extend with increasing the degree of impregnation and increasing the viscosity of the mobile phase. If the ascending development with diethyl ether, diisopropyl ether, or di-n-butyl ether (up to a distance of 15 cm between the solvent front and the start) takes only 45, 50–60, or 150 min, respectively, on papers treated with 20 % formamide solution, the impregnation with 30 % formamide solution extends the respective developing times to 90, 120, and 210 min. The impregnation with 40 % formamide solution requires
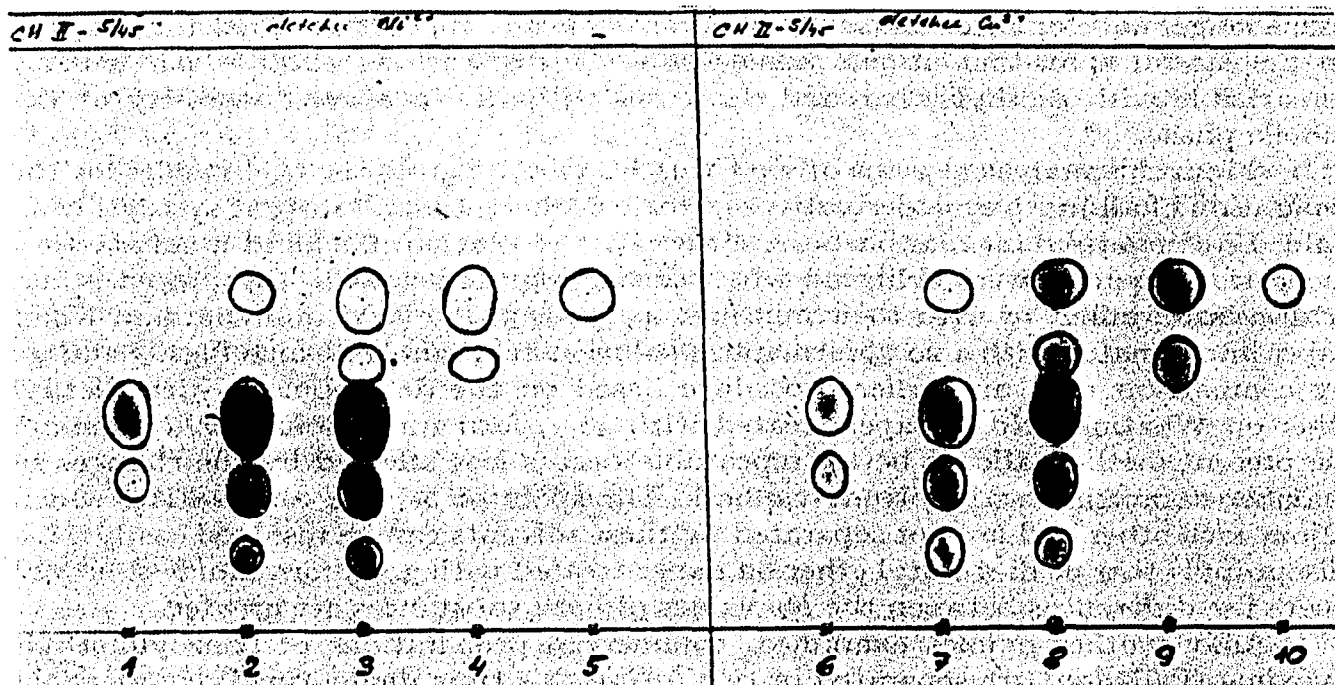
Fig. 3. Separation of all oxime derivatives of furil. Paper: Whatman No. 1 impregnated with 20 % (v/v) formamide solution in ethanol. Eluent: di-n-butyl ether–diethyl ether (5:45) (v/v). Spots (from top to bottom): (1) α-FH$_2$, γ-FH$_2$; (2) α-FMH, α-FH$_2$, γ-FH$_2$, β-FH$_2$; (3) α-FMH, β-FMH, α-FH$_2$, γ-FH$_2$, β-FH$_2$; (4) α-FMH, β-FMH; points 6–10 are points 1–5 repeated. Starting points 1–5 were detected by spraying with an ammoniacal nickel salt solution and UV irradiation, points 6–10 with an ammoniacal solution of a cupric salt.
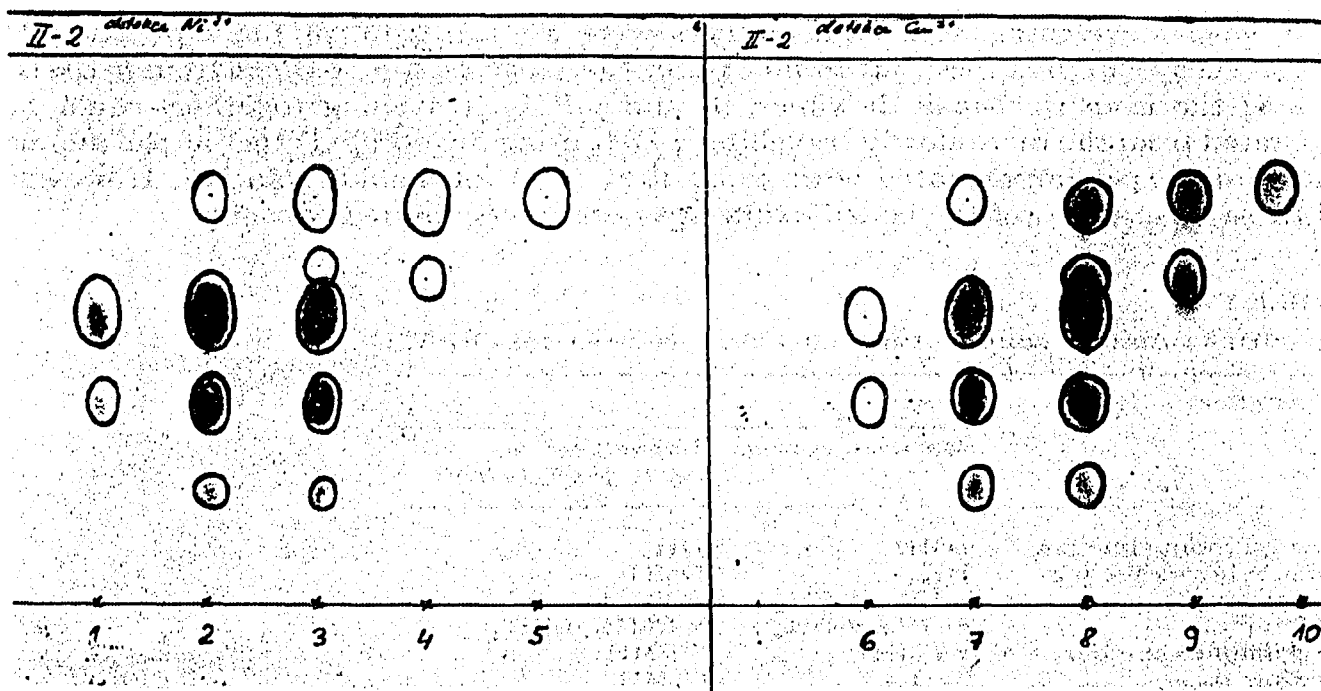

Fig. 4. Separation of isomeric furildioximes. Whatman No. 1 impregnated with 20 % formamide solution, developed with diisopropyl ether. Detection and sampling as in Fig. 3.

much longer developing times (several hours) and is not of practical importance owing to the lateral spreading of the oxime spots. This disturbing effect is particularly remarkable with diethyl ether and decreases with the increasing viscosity of the mobile phase.

From the analytical point of view this PC method gives the best results for the separation of all five oxime derivatives of furil. Although many solvent systems from Table IV can be used for that purpose, the greatest resolution effect and a satisfactory separation capacity were achieved when chromatographing with diisopropyl ether, or di-n-butyl ether, or with their mixtures 45:5, or 30:20 (v/v) on Whatman No. 1 paper impregnated with a 20 % formamide solution in ethanol (see also Figs. 3 and 4). By comparison with an earlier procedure based on the development with diethyl ether on Whatman No. 1 paper treated with 30 % formamide solution in ethanol[9], the present method affords better repeatable results less affected by the changes in chamber saturation with solvent vapours. The spots of β-FMH and α-FH$_2$, which adjoin each other and are not separated by these solvents can be resolved easily with diisopropyl ether or di-n-butyl ether on paper treated with 30 % formamide solution, where the differences between the R_F values of both substances are greater.

Among other oximes examined, commercial preparations of benzoylmethyl-glyoxime (Palladon) are separated in two (up to this time unidentified) components on using all the solvent systems and degrees of impregnation recommended here. The successful separation of DMH from DH$_2$ is only possible when developing with di-n-butyl ether; it is independent of the degree of impregnation. On the other hand, lower ethers and lower degrees of impregnation are more suitable for an effective separation of CyMH from CyH$_2$, otherwise both substances remain near the start.

The present method is not very suitable for oxime derivatives of benzil. Using diethyl ether, benzil monoximes as well as the dioximes migrate near the solvent front. Only when developing with higher ethers were we successful in the separation of monoximes from dioximes, but in no case in the separation of α-BMH from β-BMH. Among the isomeric benzil dioximes, β- and γ-BH$_2$ (travelling together) could be separated from the more slowly travelling α-BH$_2$ using diisopropyl ether as the mobile phase and paper impregnated with 30 or 40 % (v/v) formamide solution. However, all these separations of the benzil oximes possess a very limited capacity.

TABLE V

IMPURITIES FOUND IN SOME COMMERCIAL OXIME PREPARATIONS BY TLC

Preparation	Found	
	Macrocomponents	Impurities (up to 3–5 % (w/w))
Biacetyl monoxime p.a.	DMH	DH$_2$
Dimethylglyoxime p.a.	DH$_2$	DMH
Neoniklon p.a.	α-FH$_2$, γ-FH$_2$	α-FMH
Niklon p.a.	α-BH$_2$, γ-BH$_2$	α-BMH, β-BH$_2$
α-Furilmonoxime p.a.	α-FMH	β-FMH
Nioxime p.a.	CyH$_2$	CyMH
Palladon p.a.	three unidentified components giving colour reactions with Ni(NH$_3$)$_4{}^{2+}$	

By comparison with TLC, the spectrophotometric determination of the oximes after their elution from the paper chromatograms seems to be more promising because irregularities accompanying elutions from silica gel do not occur and the technique is much simpler.

The chromatographic procedures described here were successfully employed in checking the purity of most commercial oxime preparations (see also Table V) as well as in examining the course of oximation of the 1,2-diketones and their monoximes. For example, the isomerisation changes due to heating, UV irradiation, presence of specifically acting solvents or chelate-forming metal ions can be followed chromatographically much better than by other known methods.

REFERENCES

1 F. J. WELCHER, *Organic Analytical Reagents*, Vol. 3, Van Nostrand, New York, 1947, pp. 158, 272.
2 D. DYRSSEN, *Svensk. Kem. Tidskr.*, 75 (1963) 618.
3 J. TOUL, *Thesis*, J. E. Purkyně University, Brno, 1967.
4 J. MEISENHEIMER, *Ann. Chem.*, 444 (1925) 94; 446 (1926) 205; 468 (1929) 202; 469 (1929) 128.
5 J. MEISENHEIMER, *Ber.*, 54 (1921) 2206; 57 (1924) 289.
6 K. YAMASAKI AND CH. MATSUMOTO, *J. Chem. Soc. Jap., Pure Chem. Sect.*, 76 (1955) 569.
7 N. V. KHROMOV-BORISOV, *Zh. Obshch. Khim.*, 25 (1955) 136.
8 F. A. FRYER, D. J. GALLIFORD AND J. T. YARDLEY, *Analyst*, 88 (1963) 188.
9 J. TOUL AND A. OKÁČ, *Chem. Listy*, 59 (1965) 1468.
10 B. GLABIK, J. KASPRZYK AND W. WALCZYK, *Chem. Anal. (Warsaw)*, 12 (1967) 637.
11 J. BASSET, G. B. LETON AND A. I. VOGEL, *Analyst*, 92 (1967) 279.
12 T. W. TAYLOR AND M. S. MARKS, *J. Chem. Soc.*, (1930) 2302, 2305.
13 G. PONZIO, *Gazz. Chim. Ital.*, 60 (1930) 49, 78.
14 M. MILONE, *Gazz. Chim. Ital.*, 63 (1933) 744, 746.
15 E. STAHL (Editor), *Dünnschicht-Chromatographie: Ein Laboratoriumshandbuch*, Springer, Berlin, 1962.
16 J. CHURÁČEK, *J. Chromatogr.*, 33 (1968) 45.
17 R. J. WILLIAMS AND H. KIRBY, *Science*, 107 (1948) 727.