

TABLE 28

 R_F VALUES OF SOME ORGANIC ACIDS AND THEIR DERIVATIVES(H. F. MUELLER, T. E. LARSON AND M. FERRETTI, *Anal. Chem.*, 32 (1960) 687)Solvents: S_1 = Ethanol-butan-1-ol-conc. NH_4OH (5:45:50). S_2 = Butan-1-ol-acetic acid-water (4:1:5). S_3 = Pentan-1-ol-5 M formic acid (1:1).

Paper: Whatman No. 1 (ascending).

Detection: 1% ethanolic FeCl_3 , containing 0.1% HCl (hydroxamates); 0.04% bromophenol blue (in 95% ethanol at pH 6.7); 0.1% neutral ethanolic mercurochrome solution.

Acid	R_F		
	S_1^a	S_2^b	S_3^c
Butyric	0.37	0.78	—
Crotonic	0.31	0.78	—
Propionic	0.28	0.66	—
Acetic	0.16	0.58	—
Pyruvic	—	0.55	0.06, 0.62
Formic	0.14	0.48	—
Alkylbenzenesulphonic (ABS, an anionic detergent)	0.77	—	0.72, 0.04
Lactic	0.14	0.49	0.55
Succinic	—	0.55, 0.52, 0.47 ^d	0.60
Gallic	0.09	—	0.48
Malonic	0.02	—	0.50
Oxalic	—	0.59, 0.55 ^d	—
Aconitic	—	0.53, 0.66 ^d	0.67
<i>l</i> -Malic	—	0.57, 0.52, 0.44, 0.49 ^d	0.32
Citric	0.23	0.52	0.23
Isocitric	—	0.59, 0.45 ^d	—
Tartaric	—	0.52	0.15

^a Sodium salt.^b Hydroxamate.^c Acid.^d Hydroxamates not satisfactory. Two or three values for a single acid given in order of intensity.

TABLE 29

 R_F VALUES OF SOME GUANIDINE BASES(J. P. BLASS, *Biochem. J.*, 77 (1960) 484)Solvents: S_1 = Butan-1-ol-formic acid-water (77:11:12, by vol.). S_2 = Propan-2-ol-acetic acid-water (15:1:4, by vol.).

Paper: Whatman No. 1 (descending).

Detection: Sakaguchi reagent; pentacyanoferriate reagent (I. SMITH, *Chromatographic Techniques*, William Heinemann Medical Books Ltd., London, 1958, p. 149).

Compound	R_F^*	
	S_1	S_2
Arginine	0.07	0.17
γ -Guanidobutyric acid	0.45	0.56
Glycocyamine	0.23	0.38
Taurocyamine	0.11	0.29
Creatine	0.35	0.33

* Figures represent the mean of at least four determinations; S.D. is about 10%.

TABLE 30

R_F VALUES OF SOME PHENOLIC GLYCOSIDES(J. D. ANDERSON, L. HOUGH AND J. B. PRIDHAM, *Biochem. J.*, 77 (1960) 564)Solvents: S₁ = Butan-1-ol-pyridine-water (6:4:3, by vol.).S₂ = Butan-1-ol-ethanol-water (40:11:19, by vol.).S₃ = Ethyl acetate-acetic acid-water (9:2:2, by vol.).

Paper: Whatman No. 1 (descending).

Detection: Diazotised *p*-nitraniline-Na₂CO₃ (T. SWAIN, *Biochem. J.*, 53 (1953) 200).

Compound	<i>R_F</i>		
	S ₁	S ₂	S ₃
Arbutin	0.75	0.40	0.51
<i>p</i> -Hydroxyphenyl β -gentiobioside	0.55	0.07	0.17
<i>p</i> -Hydroxyphenyl β -cellobioside	0.61	0.10	0.24
Salicin	—	0.56	0.48
<i>o</i> -Hydroxybenzyl β -glucoside	—	0.59	0.48

TABLE 31

ELECTROPHORETIC MOBILITIES OF SOME PHENOLIC GLYCOSIDES

(J. D. ANDERSON, L. HOUGH AND J. B. PRIDHAM, *Biochem. J.*, 77 (1960) 564)

Electrolyte: 0.1 M sodium borate buffer (pH 10).

Paper: Whatman No. 3.

Apparatus: J. B. PRIDHAM (*J. Chromatog.*, 2 (1959) 605).Units: *M_{SA}*, mobility relative to salicylic acid.Detection: Diazotised *p*-nitraniline-Na₂CO₃ (T. SWAIN, *Biochem. J.*, 53 (1953) 200).

Compound	<i>M_{SA}</i>
Arbutin	—
<i>p</i> -Hydroxyphenyl β -gentiobioside	0.28
<i>p</i> -Hydroxyphenyl β -cellobioside	0.22
Salicin	0.00
<i>o</i> -Hydroxybenzyl β -glucoside	0.15

TABLE 32

R_F VALUES OF SOME PHENOLIC COMPOUNDS(W. J. BURKE, A. D. POTTER AND R. M. PARKHURST, *Anal. Chem.*, 32 (1960) 727)Solvents: S₁ = Butan-1-ol-acetic acid-water (4:1:5).S₂ = Butan-1-ol-ethanol-formic acid-water (5:1:1:1).S₃ = 15% aq. acetic acid.S₄ = Chloroform-propan-2-ol-acetic acid-water (1:2:1:1).S₅ = Butan-2-ol saturated with water.

Paper: Whatman No. 1 (ascending).

Temperature of run: 26.7° ± 1° (in solvent-saturated atmosphere).

Detection: Neutral silver nitrate (saturated)-acetone reagent (50 ml:1 l; sufficient water to dissolve precipitate), dip or spray.

Compound	<i>R_F</i> × 10 ³					<i>AgNO₃</i> colour*	Reaction time** (h)
	S ₁	S ₂	S ₃	S ₄	S ₅		
<i>Benzoic acids</i>							
3-(OH)	90-95	92-96	68-74	90-94	92-95	lp	1-2
4-(OH)	90-95	88-93	68-77	86-92	92-95	p-t	1-2
2,4-(OH) ₂	90-95	91-96	61-71	88-94	35-50	p	1-2
3,4-(OH) ₂	73-81	85-92	64-70	74-80	94-95	db	2-3
2,3,4-(OH) ₃	75-83	83-89	50-61	65-72	0-45	g	i
3,4,5-(OH) ₃	61-68	63-70	56-61	55-60	—	b	i
4-(OH)-3-(OCH ₃)	87-92	89-94	65-75	93-95	91-95	p-o	1
4-(OCH ₃)	89-94	88-95	68-78	92-97	91-95	—	—
3,4-(OCH ₃) ₂	88-94	90-95	73-82	93-96	90-93	—	—
3,4-(OCH ₂ O)	89-94	91-95	63-73	93-96	91-95	lp	3-4
<i>Cinnamic acids</i>							
3-OH	89-93	90-95	60-70	89-93	94-96	p-t	3-4
4-(OH)	87-92	91-96	56-65	88-94	94-96	p-t	1-3
3-(OH)-4-(OCH ₃)	86-91	90-96	46-56	85-90	94-97	lb	1-2
4-(OH)-3-(OCH ₃)	86-93	90-95	50-63	91-95	94-97	b	3
3-(OCH ₃)	90-94	90-95	64-74	90-97	94-96	—	—
4-(OCH ₃)	89-94	89-95	50-63	92-96	94-97	—	—
3,4-(OCH ₃) ₂	90-95	93-96	54-64	95-97	95-97	—	—
3,4-(OCH ₂ O)	89-94	95-97	44-62	90-95	95-97	p-t	1-3
3,4,5-(OCH ₃) ₃	88-94	89-94	69-78	94-97	92-95	—	—
<i>Phenols</i>							
2-(OH)	88-92	90-95	75-83	88-94	94-97	gr	1.2-2.4
3-(OH)	90-94	89-93	73-79	90-94	95-97	b	2-3
4-(OH)	88-93	87-93	77-83	85-88	95-96	g-gr	1-2
2,3-(OH) ₂	70-80	80-95	68-74	70-76	90-94	g	i
3,5-(OH) ₂	72-83	73-77	64-71	61-66	89-94	r-b	1-3
2-(OH)-3-(OCH ₃)	83-88	84-90	70-77	89-93	95-97	g	1-2
2,6-(OCH ₃) ₂	89-94	—	77-83	94-97	95-97	lb	1-2
<i>Miscellaneous</i>							
Rufigallic acid	0	0	0	0	0-05	t	i
Hexahydroxydiphenyl	47-57	45-55	47-54	38-47	0-40	r-b	i
Hexahydroxybenzophenone	70-75	65-77	31-36	47-53	—	y-b	0-1
m-Digallic acid	58-64	65-73	51-57	51-57	0-25	lt	0-2
Flavellagic acid	0	15-30	—	0-20	0-06	b	i
Ellagic acid	23-35	30-48	0-08	0-35	0-13	lt	0-1
Galloflavin	33-41	25-40	02-22	26-36	0-15	br-b	i
Isogalloflavin	—	27-45	0-23	27-34	0-07	b	i
β-Methyltropolone	90-96	78-95	81-97	93-97	94-96	lg	1-2
Purpurogallin	80-87	80-90	0-31	75-82	0-50	p-b	i
Purpurogallin-carboxylic acid	62-71	64-75	—	47-62	0-08	lt	2-3

* 1 = light; p = pink; t = tan; d = dark; b = brown; g = grey; o = orange; gr = green; r = red; br = brick.

** i = immediate.

TABLE 33
 R_F VALUES OF SOME PHENOLS AND RELATED COMPOUNDS
 (I. A. PEARL AND P. F. MCCOY, *Anal. Chem.*, 32 (1960) 1407)

Solvents: S_1 = Butanol saturated with 2% aqueous NH_4OH at 20° .

S_2 = Butanol-pyridine-water (10:3:3) at 20° .

Paper: Whatman No. 1 (descending).

Temperature of run: 20° .

Detection: D_1 = Fast Scarlet GB salt; Naphthalil Diazot Scarlet 2G (2,5-dichloroaniline).

D_2 = Fast Orange GG salt (*m*-dichloroaniline).

D_3 = Fast Blue VB salt (4-amino-4'-methoxy-diphenylamine).

D_4 = Fast Bordeaux GP Salt (2-amino-3-nitroanisole).

D_5 = Fast Black Salt K (4-amino-2,5-dimethoxy-4'-nitroazobenzene).

D_6 = Fast Bordeaux Salt BD (4-amino-2,5-dimethoxybenzonitrile).

D_7 = Fast Corinth Salt V (4-amino-2,4'-dimethoxybenzonitrile).

D_8 = Fast Red Salt GG (*p*-nitroaniline).

D_9 = Fast Red Salt RL (2-amino-3-nitrotoluene).

D_{10} = Fast Yellow Salt GC (*o*-chloroaniline).

(Note: These are commercial names of the stabilized diazo salt of the compound in parentheses). The solvent is allowed to evaporate, the spot exposed to NH_3 fumes and sprayed immediately with 0.05% aqueous diazo salt. The paper is then air dried, the colour noted, then sprayed with saturated aqueous Na_2CO_3 after 30 min and the colour again noted.

Compound	<i>Color*</i>											
	R_F	S_1	S_2	D_1	D_3	D_4	D_5	D_6	D_7	D_8	D_9	D_{10}
Acetosyringone	0.38	0.76	y-bn	y-bn	y-bn	bn	y-bn	bn	y-bn	bn-gd	y-bn	
Acetovanillone	0.60	0.83	p	s	p	p	v	l-ph	pu	pu	o	
Caffeic acid	0.93	0.34	bn	bn-gn	bn	d-gn	g-bn	bn-gn	bn	d-bn	bn	b
<i>p</i> -Coumaric acid	0.18	0.57	v	c	pu	bn-re	v	v	p	b-g	bn-pu	p

		<i>y-gn</i>									
2,5-Dimuroxyacetophenone	0.03	—	—	—	—	—	—	—	—	—	—
2,5-Dihydroxybenzoic acid	0.13	0.30	g-gn	bn	y-bn	g-gn	bn-gd	d-gn	y-bn	bn	g-bn
3,5-Dihydroxybenzoic acid	0.06	0.44	gd	y	pu	d-o	b-v	o	pu	o	o
Ferulic acid	0.13	0.48	pu	p	v	p	g-pu	b	b-g	g-gn	bn
<i>p</i> -Hydroxybenzaldehyde	0.77	0.85	o	y-gd	o	l-t	bn	c	bn	re	bn-re
<i>p</i> -Hydroxybenzoic acid	0.10	0.56	o	gd	o	o	v	c	bn-o	re	pu
2-Hydroxy-5-methoxybenzoic acid	0.38	0.47	pu	re	l-bn	v	w**	bn-p	bn	w	bn-r
<i>p</i> -Hydroxyphenyl-acetic acid	0.09	0.62	r-o	c	p	re	bn	pu	b-g	v	pu
Protocatechuic acid	0.07	streak	bn	1-bn	gn	bn	bn-gn	bn	bn-pu	bn-pu	bn
Quinic acid	0.01	0.05	—	—	—	—	—	—	—	—	—
Salicyl alcohol	0.84	0.86	p	gd	pu	y-o	b	v	b-v	o	pu
Syringaldehyde	0.37	0.74	w	c	p	l-y	bn-o-e	pu	bn-gn	b	y
Syringic acid	0.05	0.31	c	o	pu	pu	b	v	b	v	s
Vanillic acid	0.10	0.41	o	o	r-bn	bn-o	g-b	v	b-v	v	pu
Vanillin	0.19	0.87	y-bn	y-gd	y-gd	gd	bn-gd	y-bn	bn-gd	y	y
Vanillin	0.44	0.87	c	bn	p	g-b	v	b-v	v	pu	o

* After satd. aqueous Na_2CO_3 ; b = blue; bn = brown; c = coral; d = dark; g = green; gd = gold; gn = green; l = light; o = orange; oe = olive; p = pink; ph = peach; pu = purple; r = red; re = rose; s = salmon; t = tan; v = violet; w = white; y = yellow.

** Colour due to sizing and resultant spray rejection.

CHROMATOGRAPHIC DATA

TABLE 34

R_F VALUES OF SOME HALOGENATED PHENOLS
 (H. S. CHOGUILL AND D. E. BISSING, *Anal. Chem.*, 32 (1960) 440)

Solvents: S₁ = Benzene-acetic acid-water (2:2:1, by vol.).

S₂ = Hydrochloric acid (20 %, w/v).

Paper: Whatman No. 1.

Temperature of run: 25°.

Detection: 1 % *p*-Nitrobenzenediazonium fluoroborate spray followed by 5 % KOH in methanol spray (*cf.* J. H. FREEMAN, *Anal. Chem.*, 24 (1952) 958).

Compound	<i>R_F</i> range		
	S ₁	S ₂	Colour*
<i>o</i> -Chlorophenol	0.67-0.70	0.72-0.83	a
<i>p</i> -Chlorophenol	0.07-0.16	0.70-0.83	a
<i>o</i> -Bromophenol	0.45-0.48	0.87-0.88	b
<i>p</i> -Bromophenol	0.31-0.32	0.80-0.82	b
<i>o</i> -Iodophenol	0.91-0.94	0.88-0.90	b
<i>p</i> -Iodophenol	0.90-0.94	0.54-0.57	b

* a = light red; b = dark red.

TABLE 35

R_F VALUES OF SOME QUINONES AND PHENOLS

(A. T. DIPLOCK, J. GREEN, E. E. EDWIN AND J. BUNYAN, *Biochem. J.*, 76 (1960) 563)

Solvents: S₁ = Cyclohexane-benzene*.

S₂ = Benzene.

Paper: Not given (60 cm wide).

Impregnation: ZnCO₃.

Time of run: S₁, 1 h; S₂, not given.

Detection: U.V. light.

Compound	<i>R_F</i>	
	S ₁	S ₂
α -Tocopherol	0.71	
Ubiquinone 50	0.77	
RC-ubiquinone	0.73	
Ubiquinol 50	0.73	
<i>o</i> -Cresol		0.63
<i>m</i> -Cresol		0.44
<i>p</i> -Cresol		0.44
2-Methoxyphenol		0.26
3-Methoxyphenol		0.33
4-Methoxyphenol		0.33
γ -Tocopherol	0.45	

* J. BOUMAN AND E. C. SLATER, *Biochim. Biophys. Acta*, 26 (1957) 624.

TABLE 36

R_F VALUES OF SOME AMINOPHENOLS AND DERIVATIVES
(D. V. PARKE, *Biochem. J.*, 77 (1960) 493)

Solvents: S₁ = Diisopropyl ether saturated with water (washed with satd. aq. KMnO₄, satd. aq. FeSO₄, aq. 2 N Na₂CO₃, then water; dried over Na₂SO₄; distilled, b.p. 68°).

S₂ = CHCl₃-formic acid-methanol-water (1000:4:100:96, by vol.), (L. REIO, *J. Chromatog.*, 1 (1958) 338).

S₃ = Benzene-methyl ethyl ketone-formic acid-water (900:100:2:98, by vol.), (L. REIO, *J. Chromatog.*, 1 (1958) 338).

S₄ = Ethanol-butan-1-ol-aqueous 3 N NH₄OH/3 N ammonium carbonate buffer (11:40:19, by vol.) (M. E. FEWSTER AND D. A. HALL, *Nature*, 168 (1951) 78).

Paper: Whatman No. 1 (descending).

Time of run: 6 h (S₁, S₃); 8 h (S₂); 16 h (S₄).

Detection: D₁ = Gibb's reagent (ethanolic 2% 2,6-dichloroquinone-chloroimide, then 2 N Na₂CO₃).

D₂ = Diazo reagent, acid (1% NaNO₂ in 0.5 N HCl followed by 1% ethanolic dimethyl- α -naphthylamine).

D₃ = D₂ followed by 2 N Na₂CO₃.

D₄ = aq. 0.5% FeCl₃ followed by saturated NaHCO₃ (pH values in parentheses).

D₅ = aq. 0.2% Brentamine Fast Blue 2B salt followed by aq. 2 N NH₄OH.

D₆ = aq. 0.2% Brentamine Fast Garnet GBC salt followed by aq. 2 N NH₄OH.

D₇ = 1% naphthoresorcinol in aq. 20% trichloroacetic acid followed by heating (100°, 20–30 min).

D₈ = aq. 0.1% BaCl₂ followed by aq. 0.1% rhodizonic acid (sodium salt).

Compound	<i>R_F</i>				Colour*							
	S ₁	S ₂	S ₃	S ₄	D ₁	D ₂	D ₃	D ₄	D ₅	D ₆	D ₇	D ₈
<i>o</i> -Aminophenol	0.79	0.93	0.96	—	bl	y	gn	b	y	y		
<i>m</i> -Aminophenol	0.60	0.81	0.89	—	v	r	y	neg.	o	r		
<i>p</i> -Aminophenol	0.38	0.85	0.98	—	bl-b** v	gn	neg.	v	v			
3-Aminocatechol	—	0.90	0.96	—	bl		v(7)	y	y			
							v(9)					
4-Aminocatechol	—	0.33	0.46	—	bl		gn(7)	v	v			
							v(9)					
2-Aminoresorcinol	—	0.87	0.87	—	v		pi(7)	r	r			
						v	bu(9)					
4-Aminoresorcinol	—	0.44	0.80	—	v		o(7)	v	f			
							v(9)					
5-Aminoresorcinol	—	0.01	0.02	—	neg.			neg.	v	r		
Aminoquinol	—	—	—	—	neg.			neg.	neg.	neg.		
Aniline-N-glucuronide	—	—	—	0.09	v						bl*** neg.	
<i>p</i> -Aminophenyl-glucuronide	—	—	—	0.02	b						b *** neg.	
Glucuronic acid	—	—	—	0.02	neg.						bl*** neg.	
Phenylsulphamic acid	—	—	—	0.26	v						neg.*** w	
Acetanilide	—	—	—	0.83	v						neg.*** neg.	
Aniline†	—	—	—	0.87	v						neg.*** neg.	

* bl = blue; b = brown; gn = green; r = red; v = violet; y = yellow; o = orange; w = white on red background; pi = pink; bu = buff; neg. = negative.

** Also reported as negative.

*** Detection reagent applied after hydrolysis by spraying with N HCl and heating (100° for 10 min).

† Volatile, therefore only amounts of 200 µg or more can be detected.

TABLE 37

R_F VALUES OF *m*-DINITROBENZENE AND ITS METABOLITES
(D. V. PARKE, *Biochem. J.*, 78 (1961) 262)

Solvents: S₁ = Benzene-acetic acid-water (1:1:2, by vol.).

S₂ = Diisopropyl ether saturated with water (D. V. PARKE, *Biochem. J.*, 77 (1960) 493).

Paper: Whatman No. 1 (descending).

Time of run: 6 h (S₁).

Detection: D₁ = 0.1 N NaOH.

D₂ = 1% aq. FeCl₃.

D₃ = Gibbs's reagent (0.2% ethanolic 2,6-dichloroquinone-chloroimide followed by 2 N Na₂CO₃).

D₄ = 0.02% ethanolic *p*-dimethylaminocinnamaldehyde containing 10% (v/v) of 2 N H₂SO₄.

D₅ = Reducing reagent (D₄ reagent followed by 1.5% (w/v) Ti₂(SO₄)₃ in N H₂SO₄).

Compound	<i>R_F</i>		Colour*				
	S ₁	S ₂	D ₁	D ₂	D ₃	D ₄	D ₅
<i>m</i> -Dinitrobenzene	0.95	0.93	—	—	—	n	v
<i>m</i> -Nitrosonitrobenzene	0.96	0.93	—	—	—	pv	v
<i>m</i> -Nitrophenylhydroxylamine	0.97	0.92	—	—	—	pv	v
3,3'-Dinitroazoxybenzene	0.97	0.92	—	—	—	n	v
<i>m</i> -Nitroaniline	0.89	0.91	o	—	—	v	v
<i>m</i> -Phenylenediamine	0.05	0.20	—	—	—	v	v
2,4-Dinitrophenol	0.93	0.73	y	n	y	n	v
2,6-Dinitrophenol	0.91	0.71	y	n	y	n	v
3,5-Dinitrophenol	0.58	0.85	y	n	y	n	v
2-Amino-4-nitrophenol	0.08	0.75	o	bn	o	v	v
2-Amino-6-nitrophenol	0.86	0.83	o	bn	v	v	v
3-Amino-5-nitrophenol	0.07	0.74	y	bu	v	v	v
4-Amino-2-nitrophenol	0.56	0.82	r	bn	v	v	v
2,4-Diaminophenol	0.00	0.01	b	r	—	v	v
2,6-Diaminophenol	0.00	0.03	bn	bn	—	v	v
3,5-Diaminophenol	0.00	0.00	bn	bn	—	v	v

* o = orange; y = yellow; r = red; b = blue; bn = brown; bu = buff; n = none; v = violet; p = pale.