Nucleophilicities of amino acids and peptides†

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The kinetics of the reactions of amino acids with stabilized diarylcarbenium ions (Ar_2CH^+) have been studied photometrically in aqueous solution at variable pH. In the range of $10.5 < pH < 12$, the amino acids react much faster than the competing nucleophiles hydroxide and water. Though the pK_{aH} values of the amino acids vary by almost four units, the nucleophilic reactivities of all primary amino groups differ by less than a factor of 4. The secondary amino group of proline is 10^2 times more reactive, and the thiolate site in cysteine exceeds the reactivities of the primary amino groups by a factor of 10^4. Nucleophilicity parameters N as defined by the correlation $\log k_{20^\circ C} = s(N + E)$ have been determined in order to include amino acids into the most comprehensive nucleophilicity scales presently available, which provide a direct comparison of n-, π-, and σ-nucleophiles.

Introduction

Nucleophilic reactivities of amino acids and their derivatives have been the topic of numerous kinetic investigations, including nucleophilic aromatic[1–3] and aliphatic substitutions,[4] additions to carbonyl groups[5] and Michael acceptors[6–9] as well as reactions with heteroelectrophiles, *e.g.* nitrosonium ions.[10] Amino acid esters form adducts with cationic transition metal–π-complexes,[11] and the kinetics of these reactions have been used by Sweigart, Kane-Maguire and Honig to develop a nucleophilicity scale for amino acid derivatives.[12] While the latter study includes also numerous other nucleophiles,[12] most previous kinetic investigations provide a good comparison of nucleophilic reactivities within the group of amino acids and small peptides, but do not allocate these compounds in a general scheme of nucleophilicity.

The most comprehensive nucleophilicity scale presently available is based on reactions with benzhydrylium ions of different reactivity, and it has become possible to directly compare n-nucleophiles (amines, alcohols, phosphanes), π-nucleophiles (alkenes, arenes, organometallics), and σ-nucleophiles (hydride donors) with each other.[13–17] The rate constants have been correlated on the basis of eqn (1), where nucleophilicity N is defined as the negative value of the intercepts on the E-axis of $\log k$ *vs.* E correlations.

$$\log k_{20^\circ C} = s(N + E) \qquad (1)$$

k = second-order rate constant in $M^{-1}\ s^{-1}$

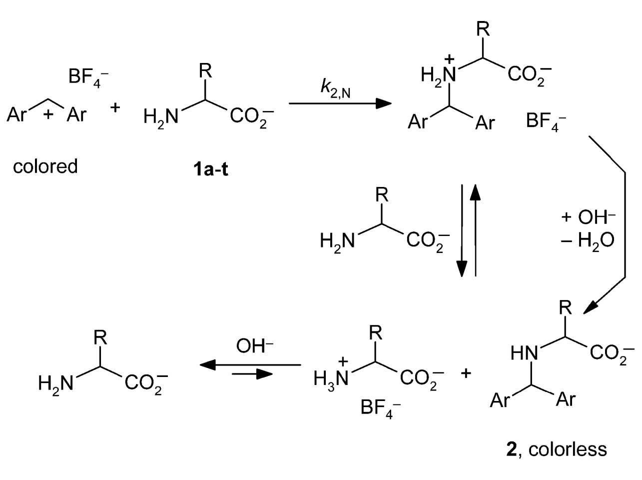
s = nucleophile-specific slope parameter

N = nucleophilicity parameter

E = electrophilicity parameter

It has been shown that the nucleophilicity scales derived from reactions with benzhydrylium ions also hold for reactions with electron deficient arenes[18] and ordinary Michael systems[19] as well as for S_N2 reactions, though in the latter case, eqn (1) has to be extended by an additional, electrophile-specific slope parameter.[20]

We have now started a program to investigate the nucleophilic and electrophilic reactivities of biomolecules, which is not only intended as a guide for their use in synthesis but also for predicting rates of individual steps in biotransformations in the absence of an enzyme. Because such rate constants are often too small for direct measurements, the comparison of calculated rate constants with the rates of the enzymatic processes shall be used to predict the absolute magnitudes of enzyme activities. In this article, we will report on the determination of the reactivity parameters N and s (eqn 1) for a variety of amino acids and their derivatives, and we will include these compounds in our comprehensive nucleophilicity scale, which puts the nucleophilic reactivities of amino acids in relation to numerous other nucleophiles of different structures. For that purpose we have investigated the kinetics of the reactions of amino acids and peptides with the reference electrophiles listed in Table 1 (Scheme 1).



Scheme 1

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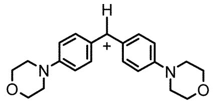
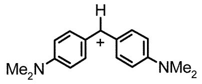
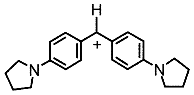
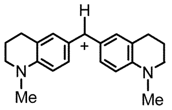
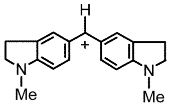
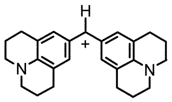
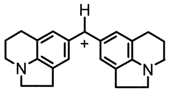
Table 1 Benzhydrylium ions (Ar_2CH^+) employed in this work

Electrophile		E^a
$(mor)_2CH^+$		−5.53
$(dma)_2CH^+$		−7.02
$(pyr)_2CH^+$		−7.69
$(thq)_2CH^+$		−8.22
$(ind)_2CH^+$		−8.76
$(jul)_2CH^+$		−9.45
$(lil)_2CH^+$		−10.04

[a] Electrophilicity parameters E from ref. 14.

Experimental procedures

Kinetics

The reactions of benzhydrylium ions with the anions of the amino acids were studied in aqueous solution at a pH where the α-amino group was deprotonated.

As the reactions of the colored benzhydrylium ions with amino acid anions gave rise to colorless products, the reactions could be followed by UV–vis spectroscopy. The rates of slow reactions ($\tau_{1/2} > 10$ s) were determined by using a J&M TIDAS diode array spectrophotometer, which was controlled by Labcontrol Spectacle software and connected to a Hellma 661.502-QX quartz Suprasil immersion probe (5 mm light path) *via* fiber optic cables and standard SMA connectors. The temperature of the solutions was kept constant (usually 20 ± 0.2 °C) during all kinetic studies by using a circulating bath thermostat and monitored with a thermocouple probe that was inserted into the reaction mixture.

Hi-Tech SF-61DX2 stopped-flow spectrophotometer systems (controlled by Hi-Tech KinetAsyst2 software) were used for the investigation of rapid reactions of benzhydrylium ions with the anions of the amino acids ($\tau_{1/2}$ <10 s at 20 °C).

Results

The rates of the reactions of the colored benzhydrylium tetrafluoroborates with amino acids were followed photometrically under pseudo-first-order conditions by using usually more than ten equivalents of the amino acid anions. Under such conditions, the concentrations of the amino acid anions **1a–t** were almost constant throughout the reactions and resulted in an exponential decay of the benzhydrylium absorbances, from which the pseudo-first-order rate constants k_{obs} were derived. As shown in Fig. 1 for the reaction of glycine with $(dma)_2CH^+$, k_{obs} depends on the pH value. At pH < 7, the observed rate constant k_{obs} corresponds to the previously reported rate constant k_W for the reaction of this carbocation with water.[21] As the pH value is increased by changing the phosphate buffer solution, the rate constant increases by a factor of 2×10^2 when a 0.905 mM glycine solution is used. Control experiments with the corresponding buffer solutions in the absence of the amino acid indicate a reaction rate which is similar to that previously reported for the reaction of $(dma)_2CH^+$ with OH^-.[22] Fig. 1 shows that at pH 10 to 11.5, the contribution of the reaction with water (k_W) and with hydroxide ($k_{1\Psi,OH}$) is negligible, and the slight increase of k_{obs} in this range is caused by the change of the zwitterion–anion ratio of the amino acid. The pH-rate-profiles for the reactions of **1b**, **1i**, **1j**, **1l**, **1m**, and **1q** with $(dma)_2CH^+$ and for the reaction of **1o** with $(ind)_2CH^+$ and $(lil)_2CH^+$ are similar and are given in the ESI.†

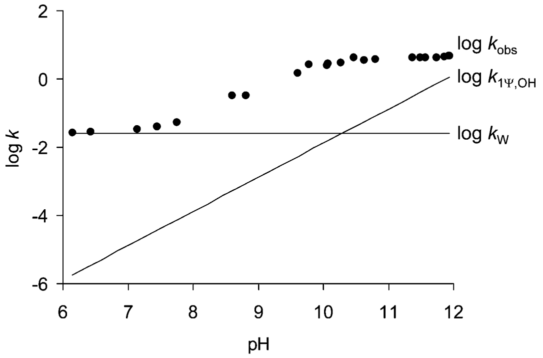


Fig. 1 Plot of the measured rate constants log k_{obs} for the reactions of glycine ($c_0 = 9.05 \times 10^{-4}$ M) with $(dma)_2CH^+$ ($c_0 = 3.36 \times 10^{-5}$ M) at 20 °C in aqueous phosphate buffer *vs.* pH compared with log k_W for the reaction with water and log $k_{1\Psi,OH}$ for the reaction with OH^- ($k_{1\Psi,OH} = k_{2,OH}[OH^-]$ was calculated from the measured pH and $k_{2,OH}$ from ref. 22; k_W from ref. 21).

When the benzhydrylium ions are combined with amino acids in water, competing reactions of the carbocations with hydroxide and water have to be considered. The observed pseudo-first-order rate constants k_{obs} reflect the sum of the reactions of the electrophile with the amino acid **1** ($k_{1\Psi,N}$), with OH^-($k_{1\Psi,OH}$), and with water (k_W).

$$k_{obs} = k_{1\Psi,N} + k_{1\Psi,OH} + k_W = k_{2,N}\,[\mathbf{1}] + k_{2,OH}\,[OH^-] + k_W \qquad (2)$$

Rearrangement of eqn (2) yields eqn (3), which defines $k_{1\Psi}$ as the overall rate constant minus the contribution of hydroxide.

$$k_{1\Psi} = k_{obs} - k_{2,OH}\,[OH^-] = k_{2,N}[\mathbf{1}] + k_W \qquad (3)$$

The concentrations of the amino acids [**1**] and of hydroxide $[OH^-]$ are calculated from pK_{aH} as described on page S2 of the

ESI.† With the already published values for $k_{2,\mathrm{OH}}$ and the calculated concentrations of hydroxide [OH^-],[22] the partial pseudo-first-order rate constants $k_{1\Psi,\mathrm{OH}}$ can be calculated. Control experiments showed that the consumption of the benzhydrylium ions in the phosphate buffers agreed with those calculated for the reactions with OH^- at the corresponding pH value within experimental error. The slopes of the plots of $k_{1\Psi}$ ($= k_{\mathrm{obs}} - k_{1\Psi,\mathrm{OH}}$) *versus* [**1**] correspond to the second-order rate constants $k_{2,\mathrm{N}}$, as shown in Fig. 2. The intercepts, which correspond to the reactions of the benzhydrylium ions with water (eqn 3), are generally negligible in agreement with the previously reported rate constants k_{W} for the reactions of benzhydrylium ions with water.[21]

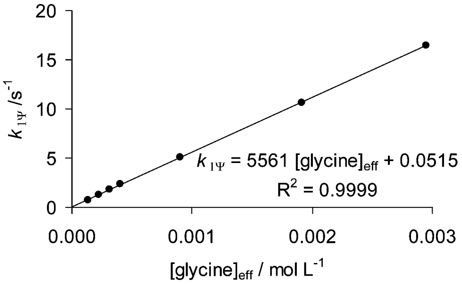


Fig. 2 Determination of the second-order rate constant $k_{2,\mathrm{N}} = 5.56 \times 10^3$ M^{-1} s^{-1} for the reaction of $(\mathrm{dma})_2\mathrm{CH}^+$ with glycine (**1a**) in water at pH 10.0 to 10.6 and 20 °C ($k_{\mathrm{W}} = 0.0206$ s^{-1}).

The linear dependence of $k_{1\Psi}$ on the concentration of amino acid anions, as depicted in Fig. 2, indicates rate-determining attack of the amino acid anion at the benzhydrylium ion (Scheme 1), because in the case of rate-determining proton transfer (second step in Scheme 1) a concave shape of this plot should result. The independence of $k_{1\Psi}$ of the pH value and the buffer concentration is also in accord with rate-determining attack of the amino acid anions at the benzhydrylium ions.

Table 2 collects all second-order rate constants determined in this investigation.

Structure nucleophilicity relationships

When log $k_{2,\mathrm{N}}$ for the reactions of the amino acid anions and peptide anions **1a–t** with benzhydrylium ions are plotted against their electrophilicity parameters E, linear correlations are obtained (Fig. 3) indicating that these reactions follow eqn (1). The slopes of these correlations yield the parameters s [$= s_{\mathrm{N}}$, if they are used for $\mathrm{S_N2}$ reactions],[20] and the intercepts on the abscissa correspond to the negative values of the nucleophilicity parameters N.

An important message comes from the reactivity data in Table 2: all N-parameters for the primary amino groups in α-amino acids are closely similar ($12.7 < N < 14.1$). The same information can also be derived directly from the rates of the reactions of these amino acid anions with the bis(dimethylamino)-substituted benzhydrylium ion $(\mathrm{dma})_2\mathrm{CH}^+$: the second-order rate constants for the reactions of primary amino groups in deprotonated α-amino acids vary by less than a factor of 4 ($1820 < k_{2,\mathrm{N}} < 6140$ M^{-1} s^{-1}). A significantly higher nucleophilic reactivity is found for proline, the only proteinogenic amino acid with a secondary amine structure ($N = 18.1$) which reacts approximately 100 times faster with benzhydrylium ions than all the primary amino groups of the amino acids investigated in this work.

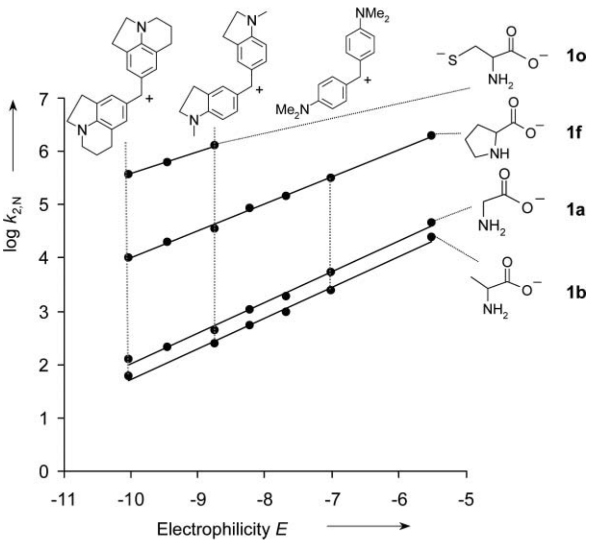
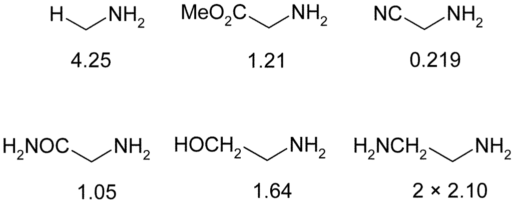


Fig. 3 Plots of the rate constants log $k_{2,\mathrm{N}}$ (in $\mathrm{H_2O}$, 20 °C) for the reactions of amino acid anions with benzhydrylium ions (from Table 2) towards the electrophilicity parameters E of $\mathrm{Ar_2CH^+}$ (from Table 1).

In previous work[23] we have shown that the introduction of neutral electron acceptor groups in amines causes a reduction of nucleophilic reactivity by factors of 2 to 20 (Scheme 2).



Scheme 2 Second-order rate constants $k_{2,\mathrm{N}}/10^3$ M^{-1} s^{-1} for the reactions of amines with $(\mathrm{dma})_2\mathrm{CH}^+$ ($\mathrm{H_2O}$, 20 °C, data from ref. 23).

In contrast, a $\mathrm{CO_2}^-$ group causes a slight increase of nucleophilic reactivity. As shown in Table 3, the anions of glycine, alanine, serine, aspartate (dianion) and glutamate (dianion) are 1.05 to 1.5 times more nucleophilic than the corresponding amines. This finding, though surprising that the Coulombic attraction between the negatively charged amino acid anions and the positively charged benzhydrylium ions does not cause a more significant rate enhancement, is in line with Hammett's substituent constants for $\mathrm{CO_2}^-$ ($\sigma_{\mathrm{p}} = 0.0$ and $\sigma_{\mathrm{m}} = -0.1$, ref. 24).

Pyrrolidine and proline are 10^2 times more reactive than the analogous compounds in Table 3, but the reactivity ratio $k_{\mathrm{proline}}/k_{\mathrm{pyrrolidine}} = 3$ again indicates that the $\mathrm{CO_2}^-$ substituent causes only a slight increase of nucleophilicity.

Comparison of the anions of glycine, β-alanine, and γ-aminobutyric acid with the corresponding amines shows that the introduction of $\mathrm{CO_2}^-$ groups in positions more remote from the amino group has a comparable small accelerating effect (Scheme 3).

According to its N parameter, the dianion of cysteine (**1o**) is by far the most nucleophilic amino acid investigated, indicating the much higher reactivity of sulfur. Because **1o** could only be

Table 2 Second-order rate constants for the reactions of benzhydrylium tetrafluoroborates **2** with amino acid anions and peptide anions **1** in water (20 °C)

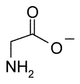
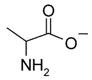
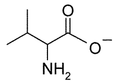
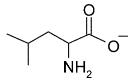
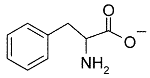
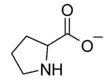
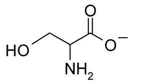
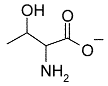
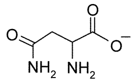
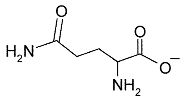
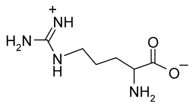
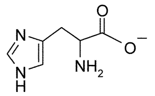
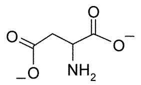
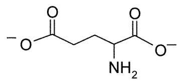
Amino acid		N	s	Ar_2CH^+	$k_{2,N}/M^{-1}s^{-1}$
	**1a**	13.51	0.58	$(mor)_2CH^+$	4.62×10^4
				$(dma)_2CH^+$	5.56×10^3
				$(pyr)_2CH^+$	1.96×10^3
				$(thq)_2CH^+$	1.11×10^3
				$(ind)_2CH^+$	4.55×10^2
				$(jul)_2CH^+$	2.19×10^2
				$(lil)_2CH^+$	1.26×10^2
	**1b**	13.01	0.58	$(mor)_2CH^+$	2.46×10^4
				$(dma)_2CH^+$	2.57×10^3
				$(pyr)_2CH^+$	1.00×10^3
				$(thq)_2CH^+$	5.53×10^2
				$(ind)_2CH^+$	2.56×10^2
				$(lil)_2CH^+$	6.27×10^1
	**1c**	13.65	0.57	$(dma)_2CH^+$	5.79×10^3
				$(pyr)_2CH^+$	2.20×10^3
				$(thq)_2CH^+$	1.21×10^3
				$(jul)_2CH^+$	2.38×10^2
	**1d**	14.01	0.52	$(dma)_2CH^+$	4.40×10^3
				$(pyr)_2CH^+$	1.66×10^3
				$(thq)_2CH^+$	1.05×10^3
				$(jul)_2CH^+$	2.31×10^2
	**1e**	14.12	0.53	$(dma)_2CH^+$	6.14×10^3
				$(pyr)_2CH^+$	2.64×10^3
				$(jul)_2CH^+$	3.10×10^2
	**1f**	18.08	0.50	$(dma)_2CH^+$	3.22×10^5
				$(pyr)_2CH^+$	1.50×10^5
				$(thq)_2CH^+$	8.80×10^4
				$(ind)_2CH^+$	3.64×10^4
				$(jul)_2CH^+$	2.02×10^4
				$(lil)_2CH^+$	1.05×10^4
	**1g**	13.16	0.55	$(dma)_2CH^+$	2.49×10^3
				$(pyr)_2CH^+$	9.07×10^2
				$(thq)_2CH^+$	5.05×10^2
				$(jul)_2CH^+$	9.53×10^1
				$(lil)_2CH^+$	5.68×10^1
	**1h**	12.69	0.60	$(dma)_2CH^+$	2.29×10^3
				$(pyr)_2CH^+$	9.73×10^2
				$(ind)_2CH^+$	2.45×10^2
				$(jul)_2CH^+$	7.86×10^1
	**1i**	13.03	0.53	$(dma)_2CH^+$	1.82×10^3
				$(pyr)_2CH^+$	6.47×10^2
				$(thq)_2CH^+$	3.81×10^2
				$(jul)_2CH^+$	7.36×10^1
				$(lil)_2CH^+$	4.47×10^1
	**1j**	13.45	0.54	$(dma)_2CH^+$	3.06×10^3
				$(pyr)_2CH^+$	1.13×10^3
				$(thq)_2CH^+$	6.50×10^2
				$(jul)_2CH^+$	1.30×10^2
				$(lil)_2CH^+$	7.31×10^1
	**1k**	12.96	0.57	$(dma)_2CH^+$	2.61×10^3
				$(pyr)_2CH^+$	9.85×10^2
				$(thq)_2CH^+$	5.53×10^2
				$(jul)_2CH^+$	1.09×10^2
				$(lil)_2CH^+$	4.55×10^1
	**1l**	13.83	0.54	$(dma)_2CH^+$	4.31×10^3
				$(pyr)_2CH^+$	2.32×10^3
				$(thq)_2CH^+$	1.28×10^3
				$(jul)_2CH^+$	2.16×10^2
	**1m**	13.81	0.53	$(dma)_2CH^+$	3.77×10^3
				$(pyr)_2CH^+$	1.69×10^3
				$(thq)_2CH^+$	8.96×10^2
				$(ind)_2CH^+$	4.54×10^2
	**1n**	13.96	0.54	$(dma)_2CH^+$	5.77×10^3
				$(pyr)_2CH^+$	2.09×10^3
				$(thq)_2CH^+$	1.17×10^3
				$(jul)_2CH^+$	2.37×10^2
				$(lil)_2CH^+$	1.40×10^2

Table 2 *(Contd.)*

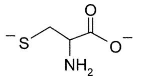
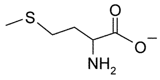
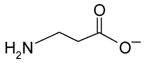
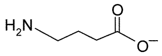
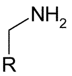
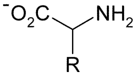
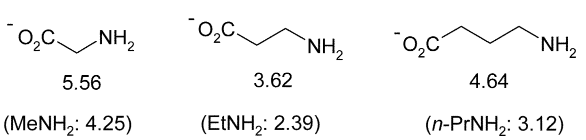
Amino acid		N	s	Ar_2CH^+	$k_{2,N}/M^{-1}s^{-1}$
	**1o**	23.43	0.42	$(ind)_2CH^+$	1.29×10^6
				$(jul)_2CH^+$	6.41×10^5
				$(lil)_2CH^+$	3.79×10^5
	**1p**	13.16	0.58	$(mor)_2CH^+$	3.45×10^4
				$(dma)_2CH^+$	3.11×10^3
				$(pyr)_2CH^+$	1.33×10^3
				$(thq)_2CH^+$	7.85×10^2
				$(ind)_2CH^+$	2.76×10^2
				$(jul)_2CH^+$	1.51×10^2
				$(lil)_2CH^+$	8.22×10^1
	**1q**	13.26	0.58	$(mor)_2CH^+$	3.62×10^4
				$(dma)_2CH^+$	3.62×10^3
				$(pyr)_2CH^+$	1.45×10^3
				$(thq)_2CH^+$	6.40×10^2
				$(lil)_2CH^+$	9.10×10^1
	**1r**	13.55	0.56	$(dma)_2CH^+$	4.64×10^3
				$(pyr)_2CH^+$	1.97×10^3
				$(thq)_2CH^+$	1.03×10^3
				$(ind)_2CH^+$	4.81×10^2
gly-gly⁻	**1s**	12.91	0.59	$(mor)_2CH^+$	2.55×10^4
				$(dma)_2CH^+$	2.72×10^3
				$(pyr)_2CH^+$	9.44×10^2
				$(ind)_2CH^+$	1.88×10^2
				$(lil)_2CH^+$	6.72×10^1
gly-gly-gly⁻	**1t**	12.26	0.63	$(mor)_2CH^+$	1.76×10^4
				$(dma)_2CH^+$	1.90×10^3
				$(pyr)_2CH^+$	7.33×10^2
				$(ind)_2CH^+$	1.64×10^2

Table 3 Rate constants $k_{2,N}/10^3$ M^{-1} s^{-1} for the reactions of amines and amino acid anions with $(dma)_2CH^+$ (H_2O, 20 °C)

R		
H	4.25[a]	5.56 (Gly⁻)
CH_3	2.39[a]	2.57 (Ala⁻)
CH_2OH	1.64[a]	2.49 (Ser⁻)
$CH_2CO_2^-$	3.62	3.77 (Asp^{2-})
$CH_2CH_2CO_2^-$	4.64	5.77 (Glu^{2-})

[a] From ref. 23.



Scheme 3 Second-order rate constants $k_{2,N}/10^3$ $M^{-1}s^{-1}$ for the reactions of amino acid anions and amines with $(dma)_2CH^+$ (H_2O, 20 °C, data from Table 2 and ref. 22,23).

investigated with three benzhydrylium ions which do not differ very much in electrophilicity, the separation of log $k_{2,N}$ into N and s is associated with some uncertainty. However, from the rate constants towards $(lil)_2CH^+$ one can derive that the sulfur of **1o** is approximately 6000 times more reactive than the corresponding amino group (from ratio **1o**–**1b**). This finding is in line with previous work of Thompson and co-workers,[7] who showed that in reactions of amino acids with quinone methides the primary site of alkylation is sulfur for cysteine and the α-amino groups for glycine (**1a**), serine (**1g**) and histidine (**1l**).

Fig. 4 shows that all amino acid anions except that of proline have reactivities of the α-amino group within one order of magnitude, despite basicities which vary by almost four orders of magnitude. This finding is in line with our previous report about the poor correlation between reactivities of amines in water and their corresponding basicities.[23]

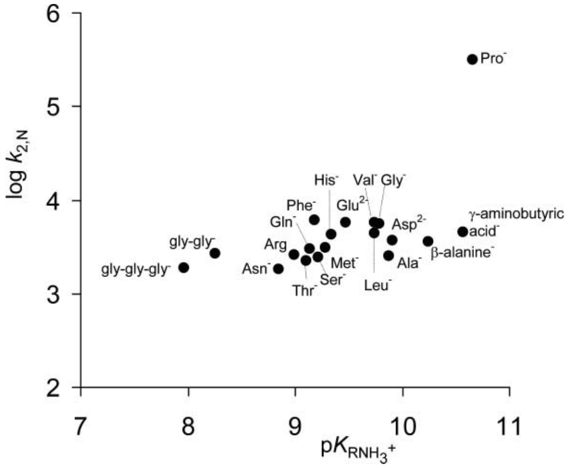
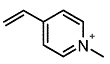
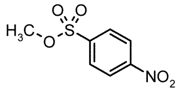


Fig. 4 Plots of the rate constants log $k_{2,N}$ (from Table 2) for the reactions of amino acid anions with $(dma)_2CH^+$ BF_4^- *vs.* pK_{aH}.

In order to explain this discrepancy one may assume that the nucleophilic reactivities reflect the intrinsic properties of the partially desolvated amino groups, while differential solvation of protonated and deprotonated forms of the amino acids accounts for the differences in pK_{aH}.

Table 4 Comparison of calculated (20 °C) and observed second-order rate constants for the reactions of amino acid anions with 1-methyl-4-vinylpyridinium ion and methyl 4-nitrobenzenesulfonate in H_2O

				
	k_{calcd}/M^{-1} s^{-1a}	k_{exp}/M^{-1} s^{-1b}	k_{calcd}/M^{-1} s^{-1c}	k_{exp}/M^{-1} s^{-1b}
1a	7.15×10^{-2}	1.51×10^{-1}	2.98×10^{-2}	5.38×10^{-2}
1b	4.30×10^{-2}	6.00×10^{-2}	2.42×10^{-2}	2.06×10^{-2}
1f	5.67	1.58	—	—
1g	5.85×10^{-2}	4.76×10^{-2}	3.11×10^{-2}	1.99×10^{-2}
1i	5.74×10^{-2}	2.44×10^{-2}	3.36×10^{-2}	1.72×10^{-2}
1j	4.32×10^{-2}	4.9×10^{-2}	2.53×10^{-2}	2.09×10^{-2}
1q	5.54×10^{-2}	8.1×10^{-2}	2.69×10^{-2}	4.05×10^{-2}
1r	8.14×10^{-2}	1.14×10^{-1}	3.42×10^{-2}	4.78×10^{-2}
1s	3.67×10^{-2}	3.37×10^{-2}	2.18×10^{-2}	1.74×10^{-2}
1t	1.43×10^{-2}	2.88×10^{-2}	1.25×10^{-2}	1.42×10^{-2}

[a] Calculated by correlation $\log k_{2,N} = s(0.762N - 12.27)$ from ref. 23 and N and s for the amino acid anions from Table 2. [b] $I = 0.1$ mol L^{-1}, at 25 °C, ref. 4,9. [c] Calculated by eqn (4) from ref. 23 and N and s for the amino acid anions from Table 2.

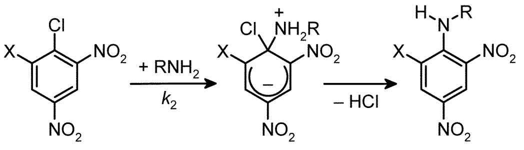
Reactions of amino acids with other electrophiles

In previous work, we have shown that the N and s parameters for amines which are derived from reactions with benzhydrylium ions can also be used to derive reactivities toward the 1-methyl-4-vinylpyridinium ion.[23] It has also been demonstrated that the rates of the reactions of amines with methyl 4-nitrobenzenesulfonate, typical S_N2 reactions, can be expressed by eqn (4).[23]

$$\log k = s(0.313\ N - 6.85) \tag{4}$$

Substitution of the N and s values of the amino acids given in Table 2 into eqn (4) yields rate constants, which agree within a standard deviation of factor 2.9 with Bunting's experimental data (Table 4). The applicability of the reactivity parameters N and s given in Table 2 for S_N2 type reactions has thus been demonstrated.

Gandler investigated the reactivity of different O-, S- and N-nucleophiles towards 2,4-dinitro-chlorobenzene **3** and picryl chloride **4** (Scheme 4).[2]



3, X = H
4, X = NO_2

Scheme 4

Fig. 5 shows a linear correlation between (log k_2)/s and N, where k_2 refers to the reactions of the nucleophiles with **3** (open circles) and with **4** (filled circles), and the nucleophile-specific parameters N and s are derived from the reactions of these nucleophiles with benzhydrylium ions (Table 2 and ref. 22,23). Because the slopes of these correlation lines (0.767 and 0.781) are deviating from unity, the rate constants for the nucleophilic additions to **3** and **4** are not properly reproduced by eqn (1), and it has to be examined whether the consideration of an additional, electrophile-specific slope parameter, as previously described for S_N2 reactions,[20] should generally be employed for nucleophilic aromatic substitutions of **3** and **4**.

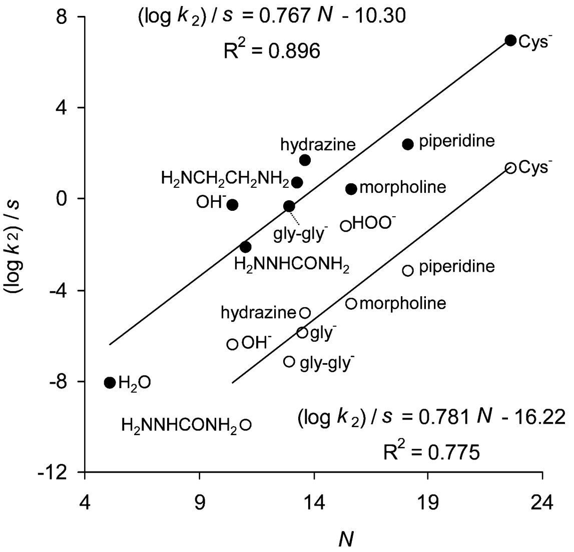
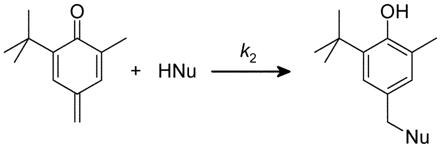


Fig. 5 Correlation between the rate constants k_2 for the reactions of O-, S- and N-nucleophiles with the chloronitrobenzenes **3** (open circles) and **4** (filled circles) in water at 25 °C (k_2 from ref. 2) with the N and s parameters given in Table 2 and in ref. 22,23. Data for hydrazine, which will be reported later, have also been included.

McCracken, Bolton and Thatcher determined the rate constants for the nucleophilic additions of amino acids and other nucleophiles towards the quinone methide **5** (Scheme 5).[6] Fig. 6 shows a good correlation between reactivities of nucleophiles toward **5** and the nucleophilicity parameters N and s, and the unexpected slope of this correlation ($\ll$1) suggests to explore the reactivity of this electrophile towards a larger variety of nucleophiles.



5

Scheme 5

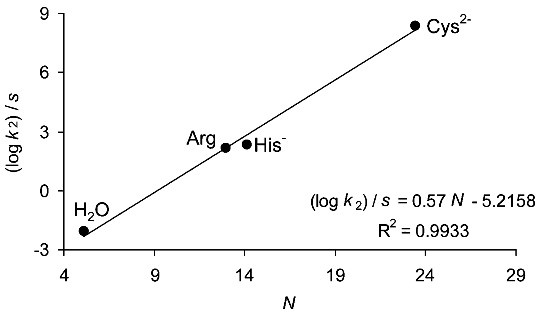


Fig. 6 Relationship between the rate constants for the reactions of nucleophiles with quinone methide **5** in water with the N and s parameters for amino acids given in Table 2 (N and s parameters for water taken from ref. 22).

Conclusions

Primary amino groups in amino acids and small peptides have closely similar nucleophilicities in water, significantly higher than that of hydroxide (Fig. 7). While the pK_{aH} value of proline is comparable to that of β-alanine and γ-aminobutyric acid, its nucleophilic reactivity exceeds that of all other amino acids by several orders of magnitude. Only cysteine, where thiolate is the reactive site, is even more nucleophilic.

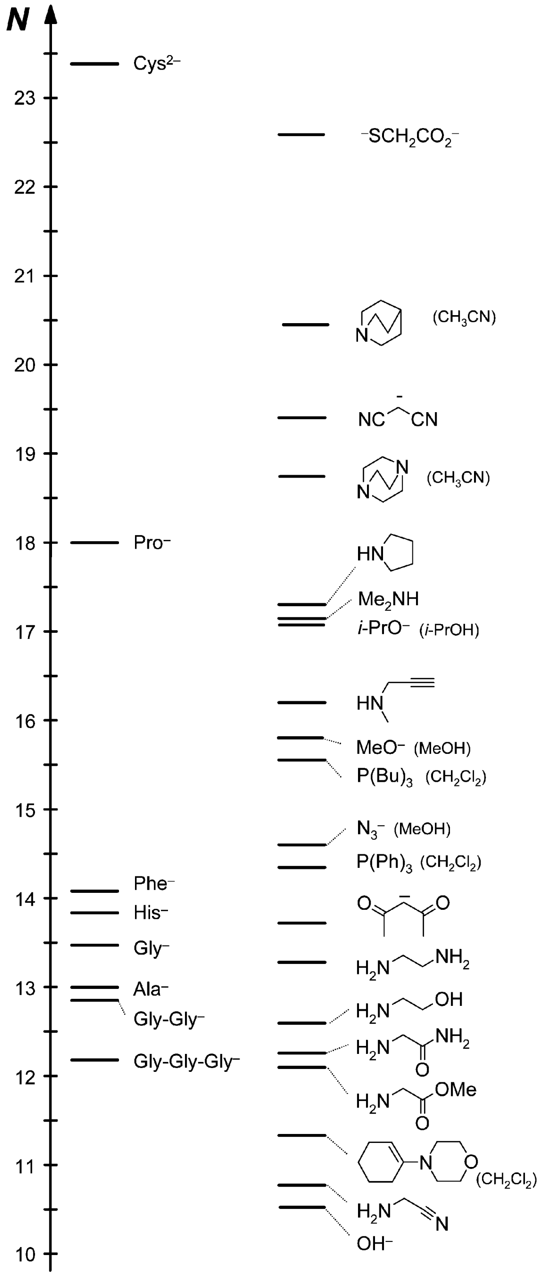


Fig. 7 Comparison of nucleophilicities of amino acid anions with other C-, N-, P-, O- and S-nucleophiles in water (data referring to other solvents are marked, nucleophilicity parameters are listed in ref. 25).

Because the N parameters derived from the reactions with benzhydrylium ions are known also to hold for reactions with ordinary Michael acceptors and alkyl esters, it has become possible to predict absolute rate constants for the reactions of amino acids with a large variety of electrophiles.

Acknowledgements

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25 For a compilation of published E, N, and s parameters, see: www.cup.uni-muenchen.de/oc/mayr/DBintro.html.