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Utilization of a Crown Ether/Amine-Type Rotaxane as a Probe for the Versatile Detection of Anions and Acids by Thin-Layer Chromatography

Shinobu Miyagawa,[a] Masaki Kimura,[a] Shin Kagami,[a] Tsuneomi Kawasaki,[b] Yuji Tokunaga*[a,c]

[a] Dr. S. Miyagawa, Dr. M. Kimura, S. Kagami, Prof. Dr. Y. Tokunaga
Department of Materials Science and Engineering, Faculty of Engineering, University of Fukui, Bunkyo, Fukui 910-8507 (Japan)
E-mail: tokunaga@u-fukui.ac.jp
[b] Prof. Dr. T. Kawasaki
Department of Applied Chemistry
Tokyo University of Science
Kagurazaka, Shinjuku-ku, Tokyo, 162-8601 (Japan)
[c] Prof. Dr. Y. Tokunaga
Research and Education Center for Regional Environment, University of Fukui, Bunkyo, Fukui 910-8507 (Japan)

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Abstract: In this study, a crown ether/amine-type [2]rotaxane was synthesized and utilized as a probe for the detection of acids and anions. The addition of acids to the amine-type [2]rotaxane solution generated corresponding crown ether/ammonium-type [2]rotaxanes, which were purified by silica gel column chromatography as ammonium salts. The isolated yields of the [2]rotaxanes, possessing a variety of anions, depended on the acidity and polarity of the counter anions. The behaviours of the ammonium-type [2]rotaxanes on thin-layer chromatography (TLC) silica gel reflected the properties of the counter anions. The treatment of the amine-type [2]rotaxane with acids afforded the corresponding ammonium-type [2]rotaxanes bearing several different anions. The ammonium-type [2]rotaxanes behaved similarly to the purified [2]rotaxanes on the TLC silica gel. Furthermore, we succeeded in the analysis of anions using mixtures of the amine-type [2]rotaxane and salts in an appropriate solvent. We demonstrated the detection of anions by the combination of TLC and the utilization of the [2]rotaxane probe.

Introduction

Considering the critical role of anions in living organisms, the environment, and medicine, anion receptors based on small molecules have been developed for specific and/or selective anion sensing.[1] Many supramolecular structures have been synthesized to achieve strong anion binding through different kinds of noncovalent interactions.[2,3] Firstly, the size and shape of the binding sites, as well as the functionalities of anion receptors, have been designed specifically for the effective binding of target anions.[4−12] Secondly, the changes in the optical (absorbance or fluorescence)[1c,2a,3,4,5,6,8,9a,11,12a,13] and electrochemical properties[1c,2a,3,4,6b,11,12a] of receptors complexed with anions have been investigated for anion sensing.

Ion chromatography[14] and capillary electrophoresis[15] are powerful tools for the qualitative and quantitative analyses of many anions simultaneously. However, a simple and inexpensive analytical method for anions, which can be operated by any person, is still required.

Generally, ammonium salts are easily and rapidly neutralized in the presence of bases to produce deprotonated amines. Stoddart's group discovered the crown ether/*sec*-ammonium-type rotaxane,[16] and they experienced difficulty in neutralizing the ammonium salt because of the unprecedented strong hydrogen bonds between the crown ether and the ammonium ion.[17] Takata's group investigated the kinetics and the thermodynamic stability of the ammonium moiety in crown ether/ammonium-type rotaxanes.[18,19] Further, we regulated the acidity of the ammonium protons by changing the size of the crowns in the rotaxanes, and developed a five-state molecular shuttling system comprising a pair of [2]rotaxanes.[20]

In 2013, Loeb's group directly utilized a crown-encircled amine as a frustrated Lewis base.[21] They treated the deprotonated amine rotaxane, consisting of aniline and a crown ether, with hydrogen gas (4 atm) in the presence of $B(C_6F_5)_3$. The rotaxane acted as a Brønsted–Lowry base to produce a crown/ammonium rotaxane bearing $HB(C_6F_5)_3^-$ as a counter ion without the corresponding amine-$B(C_6F_5)_3$ complex. Although the strong basicity of the encircled amine has been fully elucidated, the straightforward utilization of this property has not been sufficiently investigated, except for Loeb's example.

Generally, in the detection of primary, secondary, and tertiary ammonium salts by thin-layer chromatography (TLC) on silica gel, the ammonium salt is deprotonated (the acid of the ammonium salt is absorbed by the silica gel), and a spot corresponding to the resulting amine is detected on the TLC plate. In contrast, since the encapsulated ammonium in the crown ether/ammonium-type rotaxane is extremely stable owing to the strong hydrogen bonds between both components, as described above, the rotaxane, accompanied by a counter anion, might exist as an ammonium salt under the TLC-on-silica-gel conditions. Therefore, the R_f values of the rotaxanes could be affected by the counter anion (Figure 1). Herein, we report the versatile detection of acids and anions by the combination of a

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crown ether/amine-type rotaxane as a probe and TLC on silica gel.

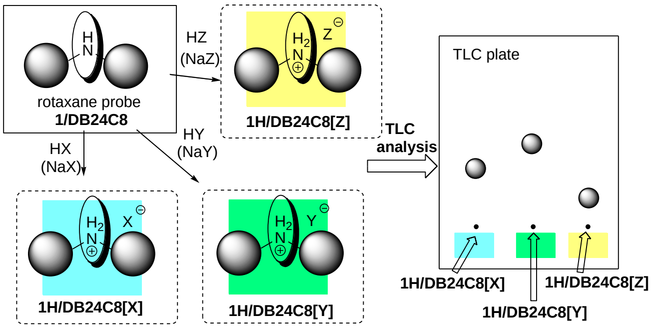


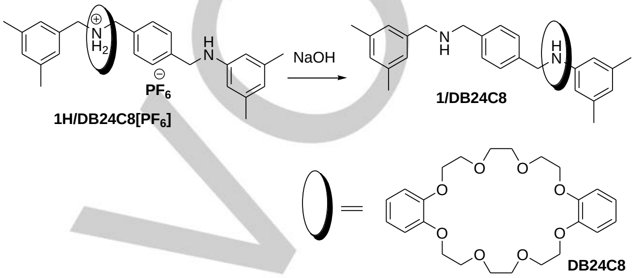
Figure 1. Concept of detection of acids and anions by the combination of a crown ether/amine-type rotaxane as a probe and thin-layer chromatography (TLC).

Results and Discussion

Isolation of the deprotonated rotaxane (1/DB24C8)

For this investigation, the rotaxane structure of **1H/DB24C8[X]** (deprotonated form: **1/DB24C8**) is shown in Scheme 1. Dialkylammonium and aniline stations are present in the dumbbell-like axle component for dibenzo[24]crown-8 as the macrocyclic component.[22] The rotaxane bearing two stations in the axle component might be suitable for isolation as a crown ether/amine-type rotaxane, because the second station holding DB24C8 can stabilize the deprotonated form. First, we attempted to isolate **1/DB24C8** by treating a solution of **1H/DB24C8[X]** in dichloromethane with a basic aqueous solution (aq. NaOH), and the organic phase was concentrated to afford the rotaxane mainly in its deprotonated form, **1/DB24C8**. However, **1/DB24C8** was not reproducibly isolated. Although the treatment of a solution of **1H/DB24C8[X]** with potassium *tert*-butoxide and Bu_4NF,[19] using several solvents, afforded **1/DB24C8** in situ, we could not completely isolate the rotaxane in its deprotonated form (a mixture of the deprotonated and the protonated rotaxanes was isolated) after workup. Takata et al. reported that the stability of the ammonium group in the crown/ammonium-type rotaxane is dependent on the counter ion and that PF_6^- is the most suitable counter ion for stabilizing the protonated rotaxane.[19] Finally, we succeeded in completely isolating **1/DB24C8**. A suspension of **1H/DB24C8[X]** in ether and an excess amount of aqueous NaOH were vigorously stirred for 54 h, after which the suspension transformed into a two-phase solution. Separation of the organic phase followed by concentration gave the deprotonated rotaxane **1/DB24C8**. Since the counter anion, PF_6^-, is required to stabilize the ammonium-type rotaxane, the removal of the PF_6 species is important for isolating **1/DB24C8**. Since both PF_6 salts (**1H/DB24C8[PF$_6$]** and $NaPF_6$) are insoluble in ether, PF_6^- completely migrates to the aqueous phase as $NaPF_6$ during the treatment of **1H/DB24C8[X]** in the presence of aqueous NaOH. The reaction time was reduced when toluene was used as the organic solvent. The presence of **1/DB24C8** was confirmed by nuclear magnetic resonance (NMR) spectroscopy (Figures 2a and S10).

As previously reported,[22] the DB24C8 unit predominantly encircled the aniline moiety under the deprotonation conditions because the hydrogen bond between the aniline and crown ether is stronger than that between the amine and the crown ether, owing to the high acidity of aniline-NH (Scheme 1). In the ^1H NMR spectrum (500 MHz, $CDCl_3$) of **1/DB24C8** (Figure 2a), the signals indicative of the benzylic protons, H_d and H_e, significantly shifted upfield, relatively to those indicative of **1H/DB24C8[X]** (Figure 2h), which is consistent with the deprotonation of the dialkylammonium group and the loss of the deshielding effects of DB24C8. In contrast, the characteristic signal of another benzylic proton, H_h, shifted to the low field region, due to the deshielding effects of DB24C8.



Scheme 1. Synthesis of crown/amine-type rotaxane **1/DB24C8**.

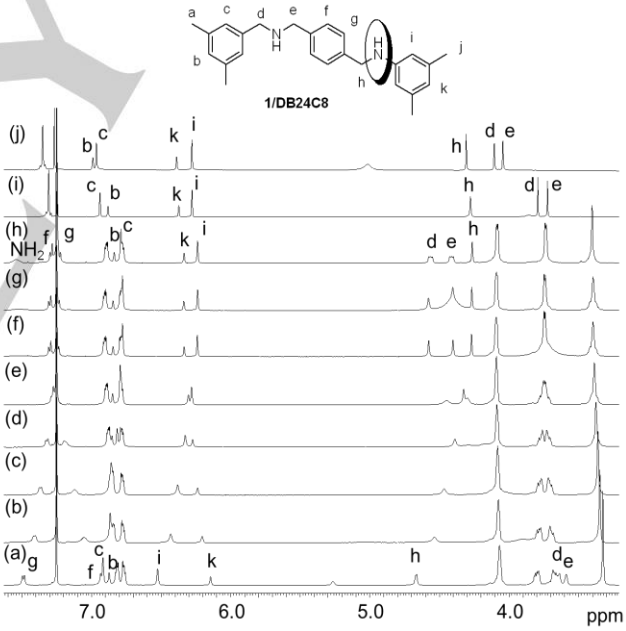


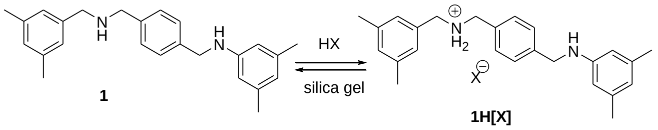
Figure 2. ^1H NMR spectra (500 MHz, $CDCl_3$) of the deprotonated rotaxane **1/DB24C8**, mixtures of **1/DB24C8** (10 mM) and acetic acid (0–6 eq), **1H/DB24C8[PF$_6$]**, and axle molecule **1**. (a) **1/DB24C8**, (b) **1/DB24C8** and AcOH (0.5 eq), (c) **1/DB24C8** and AcOH (1.0 eq), (d) **1/DB24C8** and AcOH (1.5 eq), (e) **1/DB24C8** and AcOH (2.0 eq), (f) **1/DB24C8** and AcOH (4.0 eq), (g) **1/DB24C8** and AcOH (6.0 eq), (h) **1H/DB24C8[PF$_6$]**, (i) **1**, and (j) **1** and AcOH (97 eq).

As similar deprotonated rotaxane bearing dialkylamine and aniline moieties in its axle component exists as a mixture of translational isomers in solution,[23] VT NMR experiments (CD_2Cl_2) were performed to reveal the translational isomerism of

1/DB24C8 (Figure S3). Although significant signals based on the isomers were not observed above –90 °C, small signals (ca 3%), which can be assigned to the benzyl protons in translational isomer were detected at 4.16, 4.35, and 4.50 ppm at –50 °C. At –70 °C, H$_i$ and H$_j$ signals broadened, and H$_j$ signals were split in two at –90 °C. This observation suggests slow rotation of DB24C8 around aniline moiety in the axle component.

Purification of the axle molecule (2H$_0$X$_0$) using preparative TLC on silica gel after treatment with acids

First, after the axle molecule **1** was treated with hydrochloric acid and toluenesulfonic acid (TsOH), separately (Scheme 2). The generated protonated axle molecules (**1H$_{1-2}$[Cl]$_{1-2}$** and **1H$_{1-2}$ [OTs]$_{1-2}$**) were subjected to preparative TLC (CHCl$_3$/MeOH, 10:1 and toluene/THF, 3:2) to afford the deprotonated axle molecule, **1**, in all cases. All the products were validated by ^{1}H NMR spectroscopy (Figures S1 and S2). The results show that the hydrochloric acid and TsOH were completely removed (absorbed) by the silica gel.



Scheme 2. Interconversion between the amine **1** and its ammonium salt **1H[X]**.

Formation of 1H/DB24C8[X]: NMR titration experiments of rotaxane 1/DB24C8 with acids

To investigate the formation of the ammonium rotaxanes (**1H/DB24C8[X]**), **1/DB24C8** was treated with acids, including acetic acid (AcOH) (p*K*a: 4.8), chloroacetic acid (p*K*a: 2.9), trifluoroacetic acid (TFA) (p*K*a: 0.23), and methanesulfonic acid (MsOH) (p*K*a: −1.9). Figure 2 displays the ^{1}H NMR spectra of **1/DB24C8** (10 mM) in the presence of AcOH (0–6.0 equiv) in CDCl$_3$. Upon increasing the amount of AcOH, the H$_h$ signal (0 equiv of AcOH: 4.67 ppm) moved to the high field region, and the H$_d$ and H$_e$ signals (0 equiv of AcOH: 3.65 and 3.60 ppm) shifted down field. These shifts were observed until the addition of 4.0 equiv of AcOH (Figure 2f). The results revealed that the formation of the mono-ammonium salt (**1H/DB24C8[X]**) was gradual and that the process was completed after 4.0 equiv of AcOH was added.

In the cases of chloroacetic acid (Figure S4), TFA (Figure S5), and MsOH (Figure S6), ca 1.5, 1.0–1.5 and 1.0 equiv of the acids, respectively, were enough for the mono-ammonium salt formation under these conditions. The strong acid [p*K*a: < 0.2 (TFA)] was effective for the quantitative mono-protonation of **1/DB24C8**. Furthermore, the addition of excess amounts of strong acids (TFA and MsOH) caused the protonation of the aniline moiety of [2]rotaxane.[24]

Purification of rotaxanes 1H/DB24C8[X] using silica gel column chromatography after the treatment of 1/DB24C8 with acids and TLC analysis of the ammonium rotaxanes 1H/DB24C8[X]

The purification of **1H/DB24C8[X]** was performed using silica gel column chromatography. Similar to the case of the protonated axle molecule (**1H$_{1-2}$[X]$_{1-2}$**), if silica gel absorbs the acid from the protonated rotaxane (**1H$_{1-2}$/DB24C8[X]$_{1-2}$**), the rotaxane will be isolated in its deprotonated form after purification. After the treatment of **1/DB24C8** with several acids (2.0 equiv), we employed chromatography to isolate the rotaxanes as mono-ammonium salts, **1H/DB24C8[X]**, (Table 1). When AcOH was employed, we could not isolate the rotaxane, **1H/DB24C8[CH$_3$CO$_2$]**. As described above, **1H/DB24C8[CH$_3$CO$_2$]** is not sufficiently stable as a salt under the chromatographic conditions because of the low acidity of AcOH. For chloroacetic acid and dichloroacetic acid (weak acids), the yields of the corresponding **1H/DB24C8[ClCH$_2$CO$_2$]** and **1H/DB24C8[Cl$_2$CHCO$_2$]** were moderate (49% and 57%), and these rotaxanes included 1.5 and 1.0 equiv of the corresponding acids, respectively (runs 1 and 2), as these organic acids could not be completely absorbed under the chromatographic conditions. In contrast, the utilization of strong acids afforded the corresponding mono-protonated rotaxanes (**1H/DB24C8[X]**) in good yields (runs 3, 5, 6, 8 and 9). However, the isolated yield of **1H/DB24C8[HSO$_4$]** was quite low, presumably owing to the high polarity of H$_2$SO$_4$ and/or **1H/DB24C8[HSO$_4$]** (run 7), as high polar acid can be absorbed by silica gel easily. The potential stability (p*K*a value) and the affinity for silica gel might be related to the isolated yield. A strong diacid (ethane disulfonic acid) was employed (run 10), and a 2:1 salt (**1H/DB24C8[(CH$_2$SO$_3$)$_2$]$_{1/2}$**) was isolated in 60% yield after treatment with 1 equiv of ethane disulfonic acid and chromatographic purification. At first, a 1:1 salt was mainly formed in the presence of 1 equiv of the diacid. In consideration of the yield and basicity of aniline, silica gel promotes the deprotonation of anilinium and captures the highly polar disulfonic acid. Finally, a 2:1 salt of **1H/DB24C8[(CH$_2$SO$_3$)$_2$]$_{1/2}$** was isolated. These isolated rotaxanes (**1H/DB24C8[X]**) were validated by ^{1}H NMR spectroscopy (Figure S11).

Table 1. The isolated yields of mono-protonated rotaxanes after silica gel column chromatography.[a]

run	acid	p*K*a	Isolated yield (%)
1	ClCH$_2$CO$_2$H	2.86	49[b]
2	Cl$_2$CHCO$_2$H	1.29	57[c]
3	TFA	−0.25	96
4	MsOH	−2.6	52
5	TsOH	−2.8	94
6	HNO$_3$	−1.3	<100
7	H$_2$SO$_4$	−3.0	9
8	HCl	−8.0	<100
9	HClO$_4$	−10	97
10	(CH$_2$SO$_3$H)$_2$[d]	−1.46 −2.06	60[e]

[a] After treatment of deprotonated rotaxane **1/DB24C8** with 2 eq of acids, the salts were purified by silica gel column chromatography. [b] The product (**1H/DB24C8[ClCH$_2$CO$_2$]**) including 1.5 eq of chloroacetic acid. [c] The product (**1H/DB24C8[Cl$_2$CHCO$_2$]**) including 1.0 eq of dichloroacetic acid. [d] The rotaxane **1/DB24C8** was treated with 1.0 eq of (CH$_2$SO$_3$H)$_2$. [e] Isolated as a **1H/DB24C8[(CH$_2$SO$_3$)$_2$]$_{1/2}$**.

Next, we subjected these isolated salts, mono-ammonium rotaxanes (**1H/DB24C8[X]**) and the deprotonated rotaxane (**1/DB24C8**), to TLC analysis (Figure 3). As expected, all the ammonium rotaxanes (**1H/DB24C8[X]**) and the deprotonated rotaxane (**1/DB24C8**) exhibited different behaviours on the TLC plate (eluent: CH_2Cl_2/acetone/H_2O, 3:16:1).

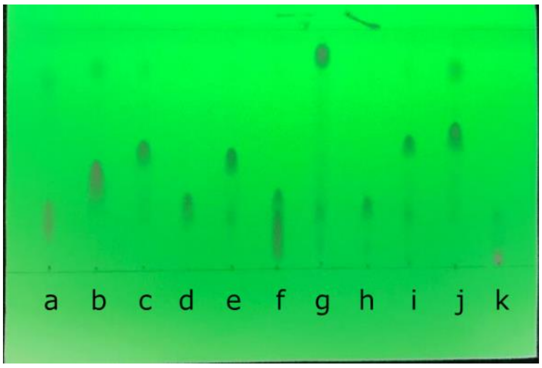


Figure 3. TLC analysis of the protonated rotaxanes **1H/DB24C8[X]** and deprotonated rotaxane **1/DB24C8** (eluent: CH_2Cl_2/acetone/H_2O, 3:16:1) under UV irradiation. (a) **1H/DB24C8[ClCH$_2$CO$_2$]**, (b) **1H/DB24C8[Cl$_2$CHCO$_2$]**, (c) **1H/DB24C8[CF$_3$CO$_2$]**, (d) **1H/DB24C8[MsO]**, (e) **1H/DB24C8[TsO]**, (f) **1H/DB24C8[(CH$_2$SO$_3$)$_2$]$_{1/2}$**, (g) **1H/DB24C8[ClO$_4$]**, (h) **1H/DB24C8[Cl]**, (i) **1H/DB24C8[HSO$_4$]**, (j) **1H/DB24C8[NO$_3$]**, and (k) **1/DB24C8**.

Detection of acids using the deprotonated rotaxane (1/DB24C8) as a probe

With the knowledge that the behaviours of each salt (**1H/DB24C8[X]**) on the TLC plate depend on the anion, we directly conducted TLC analysis of the mixtures of the deprotonated rotaxane (**1/DB24C8**) and several acids, separately. After mixing **1/DB24C8** (5 mM) in $CHCl_3$ (0.25 mL) and the acids (100 or 250 µM) in H_2O, $CHCl_3$, or THF (12.5 or 5 µL), a small amount of the organic layer was utilized for the TLC analysis using CH_2Cl_2/acetone/H_2O (3:16:1) as the eluent (Figure 4). The TLC behaviours of all the mixtures, except for the sulfuric acid mixture, were identical to those of the corresponding salts, which were purified by column chromatography on silica gel (Figure 3), individually. As described above, the deprotonation of the anilinium moiety proceeded under the TLC conditions even though **1/DB24C8** was treated with excess amounts of the acids. In the case of sulfuric acid (Figure 4i), excess amounts appear to disturb the predominant formation of the mono-protonated rotaxane, as silica gel cannot sufficiently absorb the sulfuric acid under these conditions. The detection of the acids is completed by the combination of the rotaxane probe and TLC analysis under the acidic conditions.

In contrast, when $CHCl_3$/MeOH (10:1) was used as the eluent in the TLC analysis (Figure S7), two spots were observed in the absence and presence of chloroacetic acid and TFA (Figure S7k, S7a, and S7c) on the TLC plate, and one R_f values of them were quite similar even without any acid. We suspected that excess MeOH might promote the protonation of **1/DB24C8** to produce **1H/DB24C8[MeO]**. ^{1}H NMR experiments of **1/DB24C8** in CD_3OD without an acid were performed. When mixtures of CD_3OD/$CDCl_3$ were used as solvents, **1H/DB24C8[MeO]** partially formed, as increasing the ratio of CD_3OD, the ratio of **1H/DB24C8[MeO]** was increased. Finally, the spectrum of **1/DB24C8** in only CD_3OD was quite similar to that of **1H/DB24C8[X]** (Figure S8; benzylic protons appeared at 4.23, 4.52, and 4.59 ppm).[25] We suspect that the two spots on the TLC plates correspond to **1H/DB24C8[X]** and **1H/DB24C8[MeO]** in the presence of the weak acids (HX).

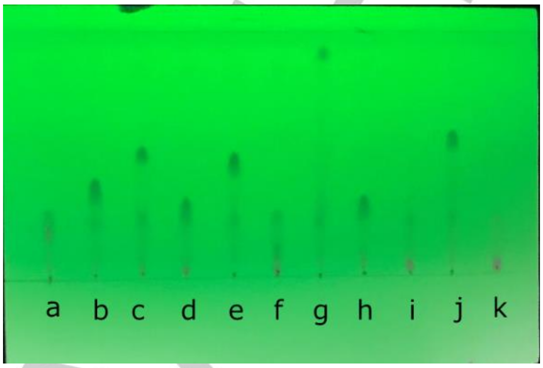


Figure 4. TLC analysis of the deprotonated rotaxane **1/DB24C8** in the presence of acids (eluent: CH_2Cl_2/acetone/H_2O, 3:16:1) under UV irradiation. Deprotonated rotaxane **1/DB24C8** in the presence of (a) $ClCH_2CO_2H$, (b) $Cl_2CH_2CO_2H$, (c) TFA, (d) MsOH, (e) TsOH, (f) $(CH_2SO_3H)_2$, (g) $HClO_4$, (h) HCl, (i) H_2SO_4, and (j) HNO_3. (k) Deprotonated rotaxane **1/DB24C8**.

Next, acid-competitive TLC analysis was performed. After treatment of a solution of the deprotonated rotaxane (**1/DB24C8**) in the presence of TsOH (2.0–0 equiv) and dichloroacetic acid (0–2.0 equiv), each solution was subjected to TLC analysis (Figure 5). Upon increasing the ratio of either of the two acids, a spot corresponding to **1H/DB24C8[X]** became noticeable. Even though the acidity of TsOH is higher than that of dichloroacetic acid, **1H/DB24C8[Cl$_2$CHCO$_2$]** could be detected by TLC.

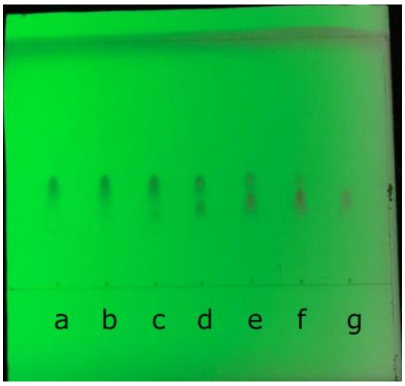


Figure 5. TLC analysis of the deprotonated rotaxane **1/DB24C8** in the presence of TsOH and $Cl_2CH_2CO_2H$ under UV irradiation. (a) **1/DB24C8** + TsOH (2 eq), (b) **1/DB24C8** + TsOH (1.75 eq) and $Cl_2CH_2CO_2H$ (0.25 eq), (c) **1/DB24C8** + TsOH (1.5 eq) and $Cl_2CH_2CO_2H$ (0.5 eq), (d) **1/DB24C8** + TsOH (1.0 eq) and $Cl_2CH_2CO_2H$ (1.0 eq), (e) **1/DB24C8** + TsOH (0.5 eq) and $Cl_2CH_2CO_2H$ (1.5 eq), (f) **1/DB24C8** + TsOH (0.25 eq), and $Cl_2CH_2CO_2H$ (1.75 eq), (g) **1/DB24C8** + $Cl_2CH_2CO_2H$ (2 eq).

Detection of the salt anions using rotaxane 1/DB24C8 as a probe

After spotting solutions or suspensions of **1/DB24C8** (5 mM, 0.152 mL) and the salts (250 µM) in aqueous MeOH (12.5 µL), the TLC plate was dried in vacuo to remove water. Thereafter, the analysis was performed using the mixed solvent system (CH_2Cl_2/acetone/H_2O, 3:16:1) as an eluent. The results of the TLC analysis of the mixtures of **1/DB24C8** and several salts are shown in Figure 6. The behaviours of the salts on the TLC plate depend on the corresponding anions. These behaviours are similar to those of the mixtures of **1/DB24C8** and the corresponding conjugated acids. For examples, TFA (Figures 4c) and CF_3CO_2Na (Figure 6a), $HClO_4$ (Figure 4g) and $NaClO_4$ (Figure 6g), and HCl (Figure 4h) and NaCl (Figure 6h). However, the shapes of the spots were not partly identical. Probably, the excess salt, which was spotted at the same time, influenced the behaviours of each **1H/DB24C8[X]**, as some of the spots that were not detected in the acid detection experiments (Figure 4) appeared to push up the spots of **1H/DB24C8[X]**, in the presence of CF_3CO_2Na, NaBr, and $NaNO_3$ (Figure (B) 6a, 6c and 6j).

The protonation of **1/DB24C8** is not promoted in the presence of neutral salts in comparison with the experiments in the presence of acids (Figure 4). However, the spots of the corresponding salts were detected. As described above, MeOH promotes the protonation from the solvent, and the exchange of the counter ion afforded the protonated rotaxane with the removal of the solvent.

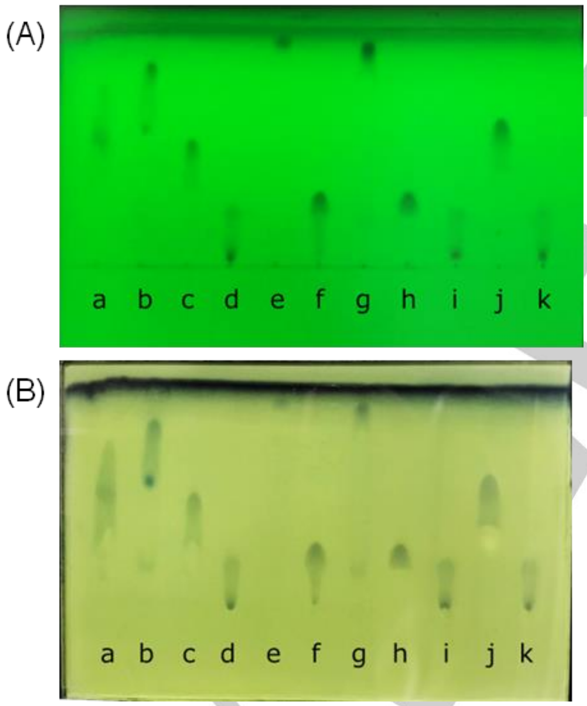


Figure 6. TLC analysis of the deprotonated rotaxane **1/DB24C8** in the presence of salts, (A) under UV irradiation and (B) after treatment of $12MoO_3{\cdot}H_3PO_4$ in EtOH. Deprotonated rotaxane **1/DB24C8** in the presence of (a) CF_3CO_2Na, (b) NaI, (c) NaBr, (d) NaH_2PO_4, (e) NH_4PF_6, (f) $C_2H_5SO_3Na$, (g) $NaClO_4$, (h) NaCl, (i) Na_2SO_4, and (j) $NaNO_3$. (k) Deprotonated rotaxane**1/DB24C8**.

Conclusion

We utilized the deprotonated rotaxane (**1/DB24C8**), which was prepared from the corresponding ammonium salt, **1H/DB24C8[PF₆]**, by deprotonation, as a probe for acid and anion detection, in cooperation with TLC. At first, mono-protonated rotaxanes (**1H/DB24C8[X]**) were isolated by silica gel column chromatography, and their R_f values reflected the corresponding counter anions. Secondly, we confirmed that the TLC behaviours of the mixtures of **1/DB24C8** and the acids coincided with those of the corresponding isolated mono-protonated rotaxanes **1H/DB24C8[X]**. In the presence of two acids, both protonated rotaxanes **1H/DB24C8[X]** could be detected by TLC. Finally, the treatment of **1/DB24C8** with salts produced the corresponding protonated rotaxanes (**1H/DB24C8[X]**), which were directly detected by TLC.

In this study, we used **1H/DB24C8**$^+$ as a counter ion for the anion detection. A quaternary ammonium cation could be employed in the same manner, since the differences seems to be negligible. The advantage of this method is that rotaxane **1/DB24C8** has no anion in the initial stage; therefore, **1H/DB24C8[X]** can be formed directly. As **1H/DB24C8[MeO]** was detected when MeOH was used as a solvent, protic solvents may form similar species. However removal of the solvent helps revive original deprotonated rotaxane. In contrast, the disadvantage is that the counter anion, i.e., the conjugate base of the weak acid, could not be adapted for this method.

Experimental Section

Materials and General Methods

The rotaxane **1H/DB24C8[PF₆]** was prepared using modified literature procedure.[22] All solvents and commercially available chemicals were used as received, except for dichloroethane, which was dried over 4Å molecular sieves. ^1H and ^{13}C NMR spectra were recorded using ECX-500II and ECA-600II spectrometers, with TMS as the internal standard. Mass spectra were recorded using JMS-700T (FAB) spectrometer, respectively. Infrared spectra were recorded using a Shimadzu FTIR-8600PC spectrometer. All reactions were performed under a positive atmosphere of dry N_2. All solvents were removed through rotary evaporation under reduced pressure. Thin-layer chromatography was performed using Merck Kieselgel $60PF_{254}$. Silica gel column chromatography was performed using Kanto Chemical silica gel 60N.

Rotaxane 1/DB24C8

A suspension of rotaxane **1H/DB24C8[PF₆]** (509 mg, 0.534 mmol) in toluene (50 mL) and 10% NaOH aq. (50 mL) was vigorously stirred for 24 h at room temperature. The organic phase was separated, washed with H_2O and sat NaCl aq., and dried (Na_2SO_4). After evaporation of the solvent, the residue was washed with hexane to afford the rotaxane **1/DB24C8** (0.350 g, 81%) as a white powder. IR ν max (NaCl) cm^{-1}: 3402, 2922, 2868, 1601, 1506, 1252, 1215, 1125, 1055, 953. ^1H-NMR (500 MHz, $CDCl_3$) δ: 7.54–7.46 (m, 2H), 6.97–6.90 (m, 4H), 6.88 (s, 1H), 6.86-6.75 (m, 8H), 6.54 (s, 2H), 6.15 (s, 1H), 5.26 (br s, 1H), 4.67 (s, 2H), 4.13–4.04 (m, 8H), 3.87–3.77 (m, 4H), 3.73–3.63 (m, 4H), 3.64 (s, 2H), 3.60 (s, 2H), 3.34 (br s, 8H), 2.30 (s, 6H), 2.05 (s, 6H). ^{13}C NMR (125 MHz, $CDCl_3$) δ: 149.8, 148.3, 140.5, 139.6, 137.8, 137.5, 137.4, 128.8, 128.4, 127.1, 126.0, 120.4, 116.6, 111.6, 69.9, 69.3, 67.8, 53.0, 52.9, 46.6, 21.4, 21.3. HRMS (FAB): m/z calcd. for $C_{49}H_{63}N_2O_8^+$ $[M+H]^+$ 807.4584, found 807.4573.

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Keywords: anion • thin-layer chromatography • rotaxane • ammonium salt

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[24] The anilinium moiety can act as a station for DB24C8. The addition of excess strong acids to the [2]rotaxane **1H/DB24C8[X]** generated the corresponding bis-ammonium salt **$1H_2$/DB24C8$[X]_2$**, which exists as two translational isomers (for examples, Figures S5 and S6). The assignment details are shown in Figure S6. These observations were already discussed previously, see ref 22 and 23.

[25] After exchange of CD_3OD for $CDCl_3$, only the deprotonated rotaxane (**1/DB24C8**) could be detected by NMR spectroscopy.

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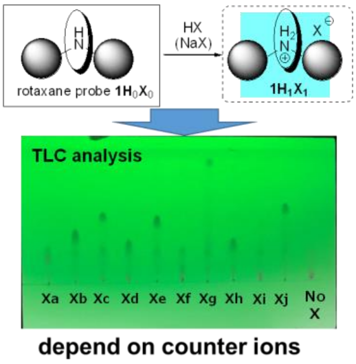

A crown ether/amine-type rotaxane was utilized as a probe for the detection of acids and anions. The treatment of the ‘amine-type’ [2]rotaxane with acids or salts afforded the corresponding ‘ammonium-type’ [2]rotaxanes bearing the corresponding anions. The behaviours of the ammonium-type [2]rotaxanes on thin-layer chromatography (TLC) silica gel reflected the properties of the counter anions.

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