MALDI TOF Post-Source Decay Investigation of Alkali Metal Adducts of Apolar Polypentylresorcinol Dendrimers

Hendrik Neubert,*,[†] Kevin A. Knights,[‡] Yolanda R. de Miguel,[‡] and David A. Cowan[†]

Drug Control Centre, King's College London, London, UK, and Department of Chemistry, King's College London, London, UK

ABSTRACT: Polypentylresorcinol dendrimers of generation two and three with varying focal groups and terminal tetrahydropyranyl (THP) ethers could be efficiently ionized intact as their alkali metal ion adducts by matrix-assisted laser desorption/ionization time-of-flight (MALDI TOF) mass spectrometry using α-cyano-4-hydroxycinnamic acid matrix. Post-source decay (PSD) MALDI TOF experiments were carried out to confirm the dendrimer structures. Li and Na ion adducts consistently produced far more intense fragmentation, unlike K and Cs ion adducts which did not produce any noteworthy fragmentation. Fragment structures and pathways were proposed on the basis of these findings. A major route of decay is the sequential loss of terminal THP ethers which proved particularly useful in structure confirmation. Li was shown to be the cation of choice to aid the analysis of THP containing polypentylresorcinol dendrimers.

Introduction

The chemistry of dendrimers has developed tremendously in recent years.[1] Because of the size, varied polarity, and symmetry of the branching units, sophisticated tools are required for the structural analysis of these compounds and their synthetic intermediates. Mass spectrometric data often provide the best evidence for a dendritic structure,[2] and since the advent of soft ionization mass spectrometry, the mass analysis of dendrimers is mainly achieved by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI TOF MS),[3−9] electrospray ionization MS (ESI MS),[10−13] laser desorption/ionization Fourier transform ion cyclotron resonance (LDI FTICR) MS,[14] and time-of-flight secondary ion mass spectrometry (TOF SIMS).[15] MALDI TOF MS has proven to be particularly valuable for investigating dendrimers, which are often not amenable to any other mass spectrometric method, and has been successfully applied to study aryl ester[3] and aryl ether[5] dendrimers, phosphorus-containing dendrimers,[4,9] poly(amidoamine) dendrimers,[6] pyridine-based dendrimers,[7] and carbosilane-based dendrimers[8] and to characterize dendritic linear-block copolymers.[5,16] MALDI usually generates protonated dendrimers, but those lacking sufficiently basic sites can still be ionized by adduct formation with metal cations to yield a pseudo-molecular ion. Like many synthetic polymers, apolar dendrimers often form adducts with monovalent metal ions such as Ag^+ and with alkali metal ions if heteroatoms are present in the structure. For example, sodium ion adducts may be formed solely by the presence of sodium as an impurity. The technique of delayed extraction is routinely employed to increase mass resolution of MALDI spectra.[17,18] This is also advantageous as it may allow more time for cation adduct formation of dendrimers and synthetic polymers and hence result in an increased signal intensity.[19] However, the contribution of preformed cation adducts to the total ion signal is unknown in the majority of cases.

Tandem MS experiments, in particular the methods of collision-induced dissociation (CID) and post-source decay (PSD), have increased the utility of MALDI in polymer and dendrimer analysis to aid structural characterization.[20] Whereas CID experiments are only possible with more sophisticated MALDI instruments, PSD can be done on many commercial reflectron TOF instruments.[20] Ions produced by MALDI may undergo fragmentation in the field-free region of the drift tube, and this process is called PSD.[21] The TOF reflector will separate precursor and metastable ions by their difference in kinetic energy. MALDI PSD is routinely being used to characterize biopolymers[22] and synthetic polymers,[23−26] but reports on dendrimer characterization are still infrequent.[27,28]

This report describes the deliberate addition of the alkali cations Li, Na, K, and Cs to a novel family of aryl ether-like polypentylresorcinol dendrimers for MALDI TOF and MALDI PSD TOF investigation. The value of this tool to facilitate confirmation of dendrimer structure is illustrated.

Experimental Section

Materials. A range of second- and third-generation polypentylresorcinol dendrimers with terminal tetrahydropyranyl (THP) protecting groups (see Scheme 1) were synthesized by the repetitive Wittig reaction of the THP functionalized benzaldehydes with a di(triphenylphosphonium bromide) salt, followed by hydrogenation of the resultant styrene functionality.[29] The dendrimers were purified by silica column chromatography. The MALDI matrix α-cyano-4-hydroxycinnamic acid (CHCA) was of mass spectrometry grade and obtained from Aldrich (Poole, UK). The chloride salts of lithium, sodium, potassium, and cesium were of analytical grade and were purchased from Fluka (Poole, UK).

Sample Preparation. The solvent used for both dendrimers and CHCA MALDI matrix was tetrahydrofuran (THF). The

* Corresponding author: Fax +44 20 7848 4980; e-mail hendrik.neubert@kcl.ac.uk.
† Drug Control Centre.
‡ Department of Chemistry.

Scheme 1. Representation of the Structures of All Investigated Dendrimers[a]

Dendrimer	n	R	x*
G2-PP	1		-
G2-BPB	1		-
G3-BPB	2		-
G3-MEsU	2		✓
G3-MEs	2		-
G3-Alc	2		-
G3-Ald	2		-

* double bond in position x (in n=1 only)

[a] PP = 4-pentyloxyphenyl, BPB = 1-benzyloxy-4-pent-4-enyloxybenzene, MEs = methyl ester, MEsU = methyl ester (unsaturated sidearms), Alc = benzyl alcohol, Ald = benzaldehyde, THP = tetrahydropyran.

Table 1. MALDI TOF Mass Measurement of Intact Polypentylresorcinol Dendrimer−Alkali Metal Ion Adducts Using α-Cyano-4-hydroxycinnamic Acid Matrix

cationizing agent		G2-PP	G2-BPB	G3-BPB	G3-MEs	G3-MEsU	G3-Alc	G3-Ald
LiCl	*m/z*	1275.87	1363.81	2700.65	2492.58	2488.50	2464.54	2462.52
	ppm[a]	+45	−20	−20	+4	−20	−17	−17
NaCl	*m/z*	1291.79	1379.79	2716.60	2508.38	2504.45	2480.38	2478.62
	ppm[a]	+4	−19	−26	−66	−27	−68	+32
KCl	*m/z*	1307.77	1395.80	2732.63	2524.53	2520.42	2496.46	2494.53
	ppm[a]	+7	+6	−5	+3	−27	−26	+6
CsCl	*m/z*	1401.73	1489.84	2826.58	2618.38	2614.50	2590.25	2588.49
	ppm[a]	+21	+70	−2	−32	+28	−84	+15

[a] Error in ppm (parts per million = difference from calculated mass/calculated mass × 10^6). Usually a mass accuracy of less than ±100 ppm is achieved with external calibration. Two decimal places are provided merely for convenience of comparison with Figure 1.

dendrimers were prepared at 0.5 mg/mL in THF and mixed with an equal volume of 20 mg/mL α-cyano-4-hydroxycinnamic acid in THF. This solution (1 μL) was codeposited with 1 μL of an aqueous solution of either LiCl, NaCl, KCl, or CsCl (10 mg/mL) on a 384 Microtiter plate MALDI target having a polished stainless steel surface. The droplet was dried before inserting into the MALDI TOF mass spectrometer (dried-droplet method).[30]

MALDI TOF Mass Spectrometry. All MALDI TOF MS experiments were performed on a Bruker Autoflex equipped with a nitrogen laser emitting at 337 nm. Spectra of intact dendrimer alkali metal ion adducts were acquired in the positive reflectron mode with an acceleration voltage of 20 kV. A delayed extraction time of 40−120 ns was selected to optimize spectral resolution. The detector voltage was 1.75 kV, and in order to avoid detector saturation low mass material was deflected. Usually, 200 laser shots were averaged to obtain a mass spectrum. External calibration was employed using various peptides which bracketed the mass range of the investigated dendrimer adducts.

For MALDI post-source decay analysis, the pulsed ion deflector was set to allow the selection of the precursor ion in a ±10 Da mass window. Pulsed ion extraction of 80 ns was employed for enhanced spectral resolution. The reflector voltage was stepped from a maximum of 20 kV depending on the mass of the precursor ion down to 0.59 kV in 18−16 segments in order to focus metastable fragments on the detector. The spectra obtained at each reflector voltage setting were pasted together in XMASS employing a calibration file from the PSD fragments of the ACTH clip (18−39). The mass assignment of fragments was performed in XMASS using an annotation macro for PSD spectra. The observed mass accuracy in PSD mode was usually within 1 Da.

Results and Discussion

The dendrimers investigated in this study were designed and synthesized to develop high-loaded supports for solid-phase chemistry. Their aryl ether-like structures are schematically presented in Scheme 1. All dendrimers contain the same molecular skeleton but differ in number of generations and type of focal group. All terminal alcohols are protected as their tetrahydropyranyl (THP) ethers.

The first objective of this study was the efficient (reliable and relatively routine) ionization and mass measurement (less than ±100 ppm) of intact polypentylresorcinol dendrimers for quality control purposes. In a conventional MALDI TOF experiment without alkali metal doping, the investigated dendrimers formed adducts with ubiquitous sodium cations present as impurities. Adduct signals were relatively weak, and protonated dendrimers could not be observed due to the lack of basic sites in the dendrimer structure. To enhance the spectral ion current and hence the signal quality of the dendrimer adducts, the chloride salts of the alkali metals lithium, sodium, potassium, or cesium were added to the MALDI sample preparation of each dendrimer. The strategy of attaching one particular or various alkali metals to aid ionization in MALDI mass spectrometry is widely used for intact apolar polymers,[31−34] fullerenes,[35] and dendrimers.[3,9] All investigated molecular species were observed in the normal reflectron mode as either their Li, Na, K, or Cs ion adducts (Table 1). For example, Figure 1a shows the conventional MALDI mass spectrum of the sodium ion
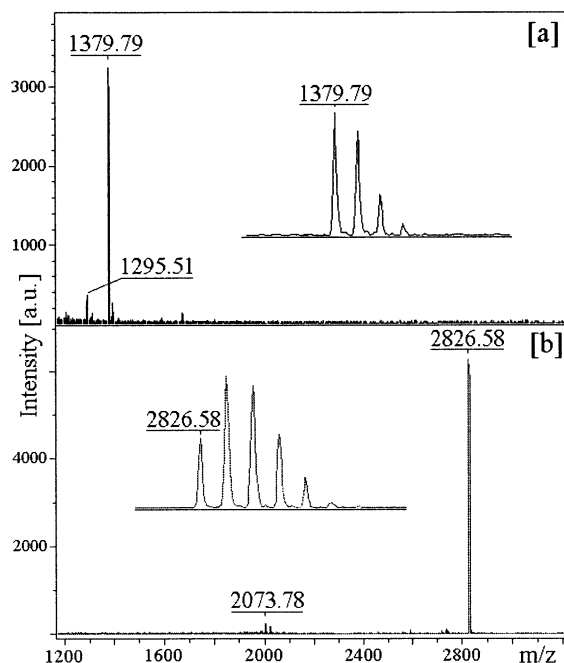
Figure 1. (a) MALDI mass spectrum of the Na$^+$ adduct of the second-generation dendrimer G2-BPB. A signal at *m/z* 1295.51 is likely to correspond to the Na$^+$ adduct of a dendrimer lacking one terminal tetrahydropyranyl group. (b) MALDI mass spectrum of the Cs$^+$ adduct of the third-generation dendrimer G3-BPB. The inserts shows the isotopic envelopes of the main signals.

adduct of the second-generation dendrimer G2-BPB at *m/z* 1379.79. A signal at *m/z* 1295.51 is likely to correspond to the sodium adduct of the dendrimer lacking one terminal tetrahydropyranyl group. This ether hydrolysis was occasionally observed for many of the analyzed dendrimers, likely being caused by the acidity of the CHCA MALDI matrix and not from incomplete synthesis. Figure 1b presents the MALDI mass spectrum of the cesium ion adduct of the third-generation dendrimer G3-BPB at *m/z* 2826.58, demonstrating the lack of imperfect moieties; i.e., no matrix-mediated ether hydrolysis or similar was observed.

Counterions have been shown to influence the selectivity and attachment efficiency of cations to the analyte as demonstrated for poly(methyl methacrylate) (PMMA);[36] therefore for our convenience we chose to use the same counterion for all alkali metals, i.e., chloride. The acquired adduct masses agree well with the calculated values; i.e., the achieved mass accuracy with external calibration was usually well below ±100 ppm. All dendrimers under investigation could be identified by mass using MALDI MS. As an adjunct, the mass measurement of Li, Na, K, and Cs ion adducts of each dendrimer increased the certainty of identification by mass.

The second and major objective of this study was to provide a straightforward means for structurally characterizing apolar polypentylresorcinol dendrimers by MALDI TOF post-source decay (PSD) mass spectrometry. The matrix CHCA was chosen for the PSD investigation because it promotes analyte fragmentation compared to other matrices.[37] All dendrimer alkali metal ion adducts that were observed in the normal reflectron mode were selected for PSD investigation, and the produced fragments were separated in time by varying the reflector voltage. Not all alkali metal ion
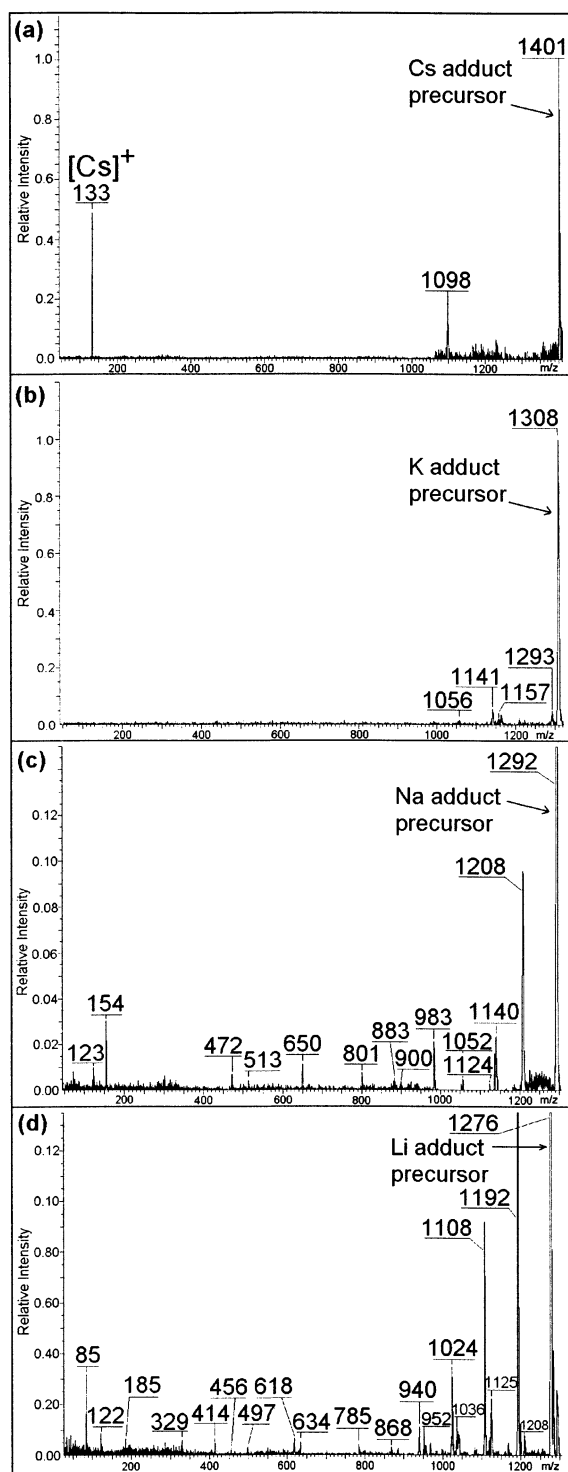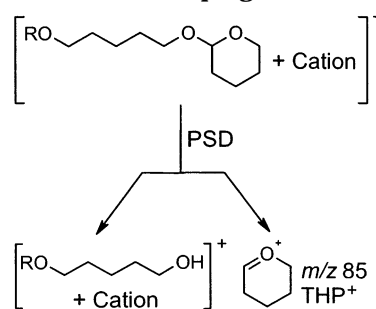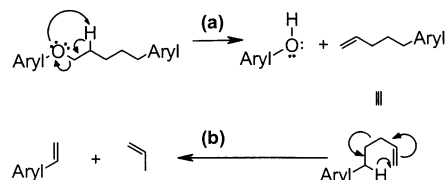

Figure 2. MALDI post-source decay spectra of adducts of the second-generation dendrimer G2-PP with (a) Cs, (b) K, (c) Na, and (d) Li ions. To show low-intensity fragments in (c) and (d), the relative intensity scale was expanded by approximately a factor of 8. No low-intensity fragments were observed with Cs and K other than those annotated in the spectra.

adducts showed heavy fragmentation; in fact, the observed levels of decay strongly depended on the cationizing agent. Generally, Li ion adducts showed very intense fragmentation, followed by Na ion adducts, whereas K and Cs ion adducts showed very little or no fragmentation at all. For example, Figure 2 presents the PSD spectra of the Li, Na, K, and Cs metal ion attachments of dendrimer G2-PP. The Cs ion dendrimer

Scheme 2. Post-Source Decay Fragmentation Pattern of the Second-Generation Dendrimer G2-PP[a]
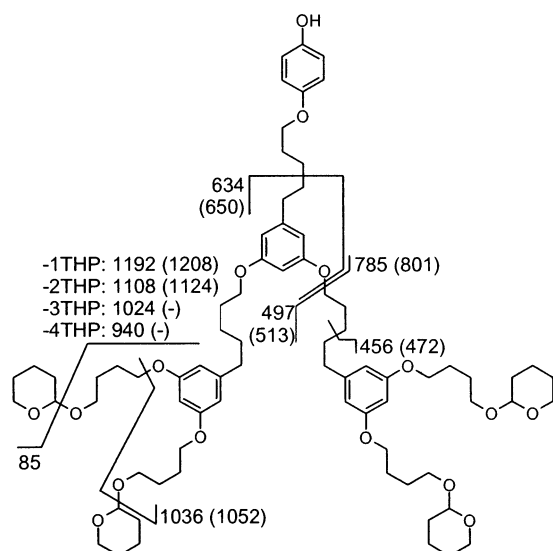
[a] Assigned fragments contain a Li ion (exception m/z 85 from terminal tetrahydropyranyl group). Observed Na ion adducts are given in parentheses.

Scheme 3. Post-Source Decay of Terminal Branching Units to Either Tetrahydropyranylium Ion or Free Hydroxyl Termini Containing the Cation in the R Grouping

Scheme 4. Proposed Breakdown Pathway for an Aromatic Ether Substructure of Polypentylresorcinol Dendrimers (a) by Elimination of a Phenolic Group and (b) by a Pericyclic-Type Rearrangement[a]


[a] The cation may be contained in either aryl grouping. The full structure of an aryl group may be inferred from Scheme 1.

adduct showed hardly any fragmentation apart from the dissociation of the Cs cation at m/z 133 and an unidentified signal at m/z 1098 (Figure 2a). PSD of the K ion adduct of G2-PP also yielded only very few higher mass signals (Figure 2b). By analogy to the charged Cs adduct, the dissociation of K from the dendrimer K ion adduct is likely to be the major route of decay as evident from the lack of fragmentation (Figure 2b). However, isolated K ions could not be observed as they were outside the investigated PSD mass range. In contrast, the Li and Na ion adducts of G2-PP exhibited very intense post-source decay across the entire mass range with higher mass signals usually being more intense (Figure 2c,d). Thus, only the PSD spectra of Li and Na ion adducts could be exploited for structural characterization. With the exception of the PSD spectrum of the Na ion adduct of G2-PP, a signal at m/z 85 was commonly found in both the Li and Na PSD spectra of all investigated dendrimer species, suggesting a fragment without a cation, in this case the positively charged fragment of the terminal tetrahydropyranyl (THP) protecting group. However, many of the other fragments produced from the Li or Na ion adducts of the same dendrimer species were different in mass. The induction of different fragmentation pathways depending on the cation could be a plausible explanation; however, when comparing Li and Na PSD spectra, a systematic mass shift of 16 amu was observed for many of the fragments accounting for the mass difference between Li and Na. By using this approach, the mass of the alkali cation was subtracted from the observed m/z value, and a fragment structure could be proposed. Many PSD signals could be identified as being branching units or fragments of branching units mostly containing at least one terminal THP group (Scheme 2). With the exception of m/z 85, all assigned masses are inclusive of Li and if applicable inclusive of Na as annotated in parentheses. The high mass signals represent a series of sequential losses of terminal THP ether groups. In two instances, cleavage of the precursor ion at two or even three positions is proposed, which explains the formation of m/z 1036 and 634 in the Li

and m/z 1052 and 652 in the Na PSD spectrum, respectively. A more detailed discussion of the fragmentation mechanisms follows to support the proposed structures for many of the fragments, particularly those that arise from the seemingly unfavorable C−C cleavages (m/z 456 and 634 from the lithium ion adduct of G2-PP).

Fragmentation Mechanisms. Various fragmentation pathways can be established for the post-source decay of the Li and Na ion dendrimer adducts. The most abundant PSD fragment signals arise from losses of terminal THP ether groups, which resulted either in free hydroxylated branches or a tetrahydropyranylium ion at m/z 85 (Scheme 3). A similar elimination of THP from protonated tetrahydropyranyl derivatives was observed previously under CID conditions.[38] In another study, many protonated aliphatic acetal structures containing the THP ether, including a structurally similar *n*-pentyl derivative, were investigated by CID, and the base peak always appeared at m/z 85.[39] The fragmentation of our dendritic THP ethers resulted in tetrahydropyranylium ion signals of relatively low abundance compared to signals from hydroxylated counterparts without the protecting group and considering that there are four or eight THP ethers per molecule. This low abundance may be explained by the lack of ether activation by a proton. The proposed fragmentation pathway of the aromatic ether shown in Scheme 4a produces a phenol and a pent-4-enyl benzene group, either of which may contain the cation. A direct result of this decay is for example the Li-containing fragments of m/z 497 and 785. A pericyclic rearrangement as illustrated in Scheme 4b is probably the cause for the observation of the Li containing fragment at m/z 455. A result of both of the above-mentioned breakdown paths is m/z 634.

MALDI PSD of Related Dendrimer Species. The strategy of structurally analyzing dendrimers as their alkali metal ion adducts outlined above for G2-PP was extended to a range of related species with distinctive
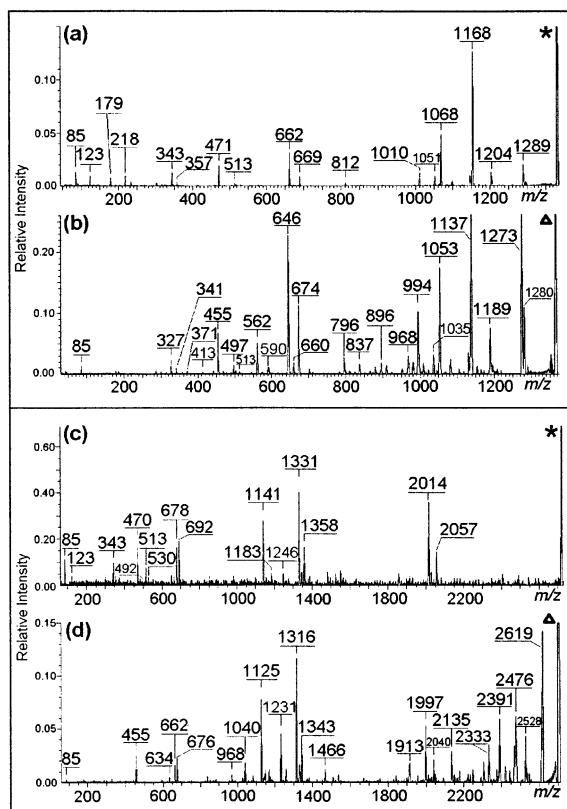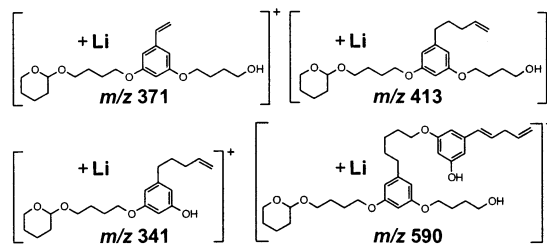
Figure 3. MALDI post-source decay spectra of the (a) Na and (b) Li ion adduct of the second-generation dendrimer G2-BPB and the (c) Na and (d) Li ion adduct of the third-generation dendrimer G3-BPB. The precursor ions of a Na ion adduct are annotated with a ★ and the precursor ions of a Li ion adduct with a △.

chemical modifications, in particular varying generations and different focal groups. Figure 3 displays PSD spectra of the Na and Li ion adducts of a second- and third-generation polypentylresorcinol dendrimer with a 1-benzyloxy-4-pent-4-enyloxybenzene (BPB) focal group, G2-BPB and G3-BPB, respectively. Cs and K ion adducts did not show any noteworthy fragmentation; however, a mass shift of 16 amu as discussed above was again observed for many signals between the Li and Na PSD spectra within any one species. Many of the fragments from G2-BPB and G3-BPB could be readily assigned with the knowledge of possible fragmentation pathways, and representative examples are discussed below. Despite the similarity of the molecules, there were significant differences between the PSD spectra of the Li ion adducts. However, m/z 85 from the terminal THP ether and m/z 455 from a branching unit comprising of two THP groups were always observed. The two PSD signals at m/z 660 and 674 from the Li ion adduct of G2-BPB are likely to correspond to m/z 662 and 676 in the Li ion adduct PSD spectrum of G3-BPB. This technique has enabled us to assign subtle structural differences in these complex molecules, in this case the presence of a further double bond in the former fragments in addition to the double bond formed upon cleavage of a dipodal terminal branching unit. Characteristic lower mass fragments are depicted in Scheme 5. These fragments or molecular variants thereof were also found in PSD spectra from Li ion adducts of related dendrimers.

The study was further extended to the structural characterization of four generation 3 polypentylresor-

Scheme 5. Some Selected Characteristic Lower Mass Post-Source Decay Fragments of the Second Generation Dendrimer, G2-BPB[a]

[a] These fragments are also found in spectra from other polypentylresorcinol dendrimers that contain the same structural components.
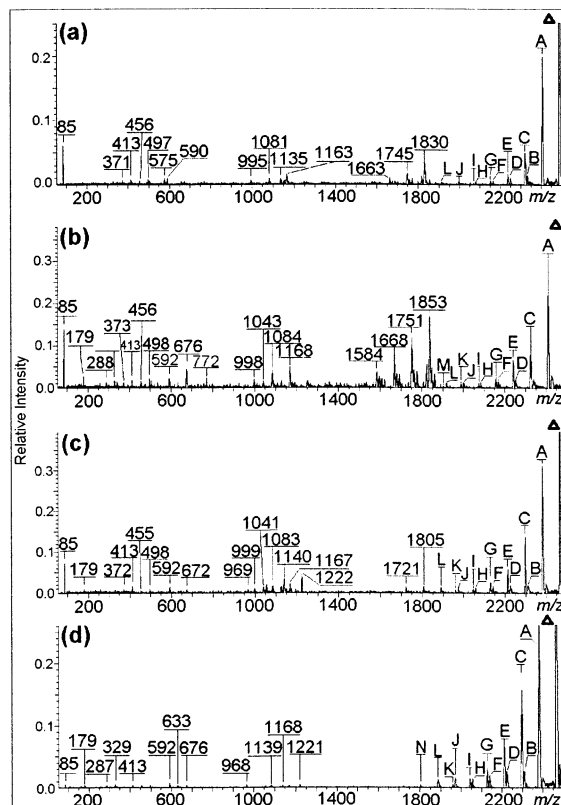
Figure 4. MALDI post-source decay spectra of the lithium ion adducts of the third-generation dendrimers (a) G3-MEsU, (b) G3-MEs, (c) G3-Alc, and (d) G3-Ald. A lithium ion adduct precursor ion is annotated with a △. Peaks labeled with a letter refer to up to seven sequential losses of tetrahydropyranyl (THP) groups and one or no loss of a pentenyloxytetrahydropyranyl group (pTHP) (see also Table 2).

cinol dendrimers differing in the type of focal group, ranging from benzoic acid methyl ester (G3-MEs), benzyl alcohol (G3-Alc), or benzaldehyde (G3-Ald). In addition, G3-MEsU features a double bond in each sidearm (Scheme 1, position x). Figure 4 shows the MALDI PSD spectra of the Li ion adducts of these dendrimers. Comparison of lithium and sodium adduct PSD spectra (Na adduct spectra provided in the Supporting Information) revealed again that the majority of higher mass signals still contained the Li cation as demonstrated for the other dendrimer species; thus, we were able to propose fragment structures as follows. Many lower mass fragments were already observed with other dendrimers investigated in this study (Schemes 2 and 5), namely m/z 85, 372, 413, 455, 497, and 590 (or 592 without the double bond). The high mass signals

Table 2. Mass Assignment for High Mass Post-Source Decay Signals from Lithium Ion Adducts of the Third-Generation Dendrimers G3-MEsU, G3-MEs, G3-Alc, and G3-Ald[a]

	fragment		fragment mass (*m/z*)			
label	-THP	-pTHP	G3-MEsU	G3-MEs	G3-Alc	G3-Ald
precursor	0	0	2488	2493	2465	2463
A	1	0	2403	2408	2380	2379
B	0	1	2331		2312	2308
C	2	0	2320	2325	2296	2295
D	1	1	2248	2253	2224	2223
E	3	0	2236	2240	2211	2210
F	2	1	2164	2168	2141	2138
G	4	0	2151	2156	2128	2127
H	3	1	2080	2084	2056	2055
I	5	0	2067	2073	2044	2042
J	4	1	1997	2000	1973	1972
K	6	0		1988	1960	1959
L	5	1	1912	1918	1888	1887
M	7	0		1904		
N	6	1			1805	1803

[a] The fragments are labeled with letters A to M as annotated in Figure 4. THP refers to a tetrahydropyranyl and pTHP to a pentenyloxytetrahydropyranyl group. For comparison, the masses of the Li ion adduct precursor ions are quoted in the first row. The observed mass tolerance was usually within 1 Da.

of each of the four species followed a pattern that could be explained by sequential losses of THP groups and pentenyloxytetrahydropyranyl (pTHP) groups, as detailed in Table 2. Up to seven subsequent losses of THP groups were usually observed, with the difference between the fragment signals being 84 ±1 Da, i.e., [M + Li-*n*THP + *n*H]$^+$ (*n* ≤ 7, THP = 85 amu). The loss of the eighth THP group could not be detected. Signal intensities usually decreased exponentially from higher to lower mass fragments, accounting for THP and pTHP losses and illustrating the decreased probability and higher energy needed for cleavage at multiple positions. The loss of pTHP groups is likely to follow a pathway as explained in Scheme 4a. Interestingly, the high mass fragments resulting from THP and pTHP losses were absent with Na as the cation. All high mass signals that contained the focal group exhibited a mass shift according to the mass difference between the focal groups as shown in Table 2. The uniformity in mass difference between fragments from different dendrimer species increases the certainty of identification not only of the fragment but also of the entire molecule.

The abundance of the fragment ions containing the cation indicated that the affinity for the smaller alkali metals is greater than that for larger alkali metals; i.e., Cs and K are so loosely coordinated that they dissociated easily under PSD conditions. At the same time, all PSD signals of reasonable abundance contained at least one THP ether in the fragment structure. The cyclic acetal group of the THP ether is proposed as the site for cation attachment, sometimes with the participation of a second acetal group on an adjacent dendritic arm.

Summary and Conclusion

We employed MALDI TOF and MALDI PSD TOF mass spectrometry to analyze a number of polypentylresorcinol dendrimers as their alkali metal ion adducts. The PSD spectra of lithiated and sodiated dendrimers were more informative than the PSD spectra from the corresponding K and Cs ion adducts. Many of the fragments from the Li and Na ion adducts could be assigned and fragmentation pathways were proposed.

This facilitated the confirmation of dendrimer structure. The fragmentation patterns provide a fingerprint spectrum for a particular species. As an adjunct, the tetrahydropyranyl ether is a commonly employed protecting group for alcohols that is stable to bases and other reagents. The fact that the investigated dendrimers contained those THP ethers not only enabled alkali metal ion adduct formation and efficient ionization but also provided potential cleavage sites under PSD conditions which proved to be extremely useful for end-group determination. Thus, this technique should aid the analysis of dendrimers and other polymers that contain THP or similar ethers.

Supporting Information Available: PSD spectra of sodium ion adducts of G3-MEsU, G3-MEs, G3-Alc, and G3-Ald. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Fischer, M.; Vögtle, F. *Angew. Chem., Int. Ed.* **1999**, *38*, 884–905.
(2) Matthews, O. A.; Shipway, A. N.; Stoddart, J. F. *Prog. Polym. Sci.* **1998**, *23*, 1–56.
(3) Mowat, I. A.; Donovan, R. J.; Bruce, M.; Feast, W. J.; Stainton, N. M. *Eur. Mass Spectrom.* **1998**, *4*, 451–458.
(4) Blais, J. C.; Turrin, C. O.; Caminade, A. M.; Majoral, J. P. *Anal. Chem.* **2000**, *72*, 5097–5105.
(5) Yu, D.; Vladimirov, N.; Frechet, J. M. J. *Macromolecules* **1999**, *32*, 5186–5192.
(6) Zhou, L.; Russell, D. H.; Zhao, M. Q.; Crooks, R. M. *Macromolecules* **2001**, *34*, 3567–3573.
(7) Chessa, G.; Scrivanti, A.; Seraglia, R.; Traldi, P. *Rapid Commun. Mass Spectrom.* **1998**, *12*, 1533–1537.
(8) Wu, Z. C.; Biemann, K. *Int. J. Mass Spectrom. Ion Process.* **1997**, *165*, 349–361.
(9) Blais, J. C.; Turrin, C. O.; Caminade, A. M.; Majoral, J. P. *Proc. 48th ASMS Annual Conference on Mass Spectrometry and Allied Topics*, Long Beach, 2000.
(10) Tolic, L. P.; Anderson, G. A.; Smith, R. D.; Brothers, H. M.; Spindler, R.; Tomalia, D. A. *Int. J. Mass Spectrom.* **1997**, *165*, 405–418.
(11) Schwartz, B. L.; Rockwood, A. L.; Smith, R. D.; Tomalia, D. A.; Spindler, R. *Rapid Commun. Mass Spectrom.* **1995**, *9*, 1552–1555.
(12) Weener, J. W.; van Dongen, J. L. J.; Meijer, E. W. *J. Am. Chem. Soc.* **1999**, *121*, 10346–10355.
(13) Neubert, H.; Kicman, A. T.; Cowan, D. A.; Bansal, S. S. *Tetrahedron Lett.* **2002**, *43*, 6723–6727.
(14) Walker, K. L.; Kahr, M. S.; Wilkins, C. L.; Xu, Z. F.; Moore, J. S. *J. Am. Soc. Mass Spectrom.* **1994**, *5*, 731–739.
(15) Coullerez, G.; Lundmark, S.; Malkoch, M.; Magnusson, H.; Malmstrom, E.; Hult, A.; Mathieu, H. J. *Appl. Surf. Sci.* **2003**, *203*, 620–624.
(16) Choi, J. S.; Lee, E. J.; Choi, Y. H.; Jeong, Y. J.; Park, J. S. *Bioconjug. Chem.* **1999**, *10*, 62–65.
(17) Brown, R. S.; Lennon, J. J. *Anal. Chem.* **1995**, *67*, 1998–2003.
(18) Vestal, M. L.; Juhasz, P.; Martin, S. A. *Rapid Commun. Mass Spectrom.* **1995**, *9*, 1044–1050.
(19) Mowat, I. A.; Donovan, R. J.; Maier, R. R. J. *Rapid Commun. Mass Spectrom.* **1997**, *11*, 89–98.
(20) Hanton, S. D. *Chem. Rev.* **2001**, *101*, 527–569.
(21) Spengler, B. *J. Mass Spectrom.* **1997**, *32*, 1019–1036.
(22) Chaurand, P.; Luetzenkirchen, F.; Spengler, B. *J. Am. Soc. Mass Spectrom.* **1999**, *10*, 91–103.
(23) Przybilla, L.; Francke, V.; Rader, H. J.; Mullen, K. *Macromolecules* **2001**, *34*, 4401–4405.
(24) Przybilla, L.; Rader, H. J.; Mullen, K. *Eur. Mass Spectrom.* **1999**, *5*, 133–143.
(25) Trimpin, S.; Eichhorn, P.; Rader, H. J.; Mullen, K.; Knepper, T. P. *J. Chromatogr. A* **2001**, *938*, 67–77.
(26) Goldschmidt, R. J.; Wetzel, S. J.; Blair, W. R.; Guttman, C. M. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 1095–1106.

(27) Adhiya, A.; Wesdemiotis, C. *Int. J. Mass Spectrom.* **2002**, *214*, 75−88.

(28) McCarley, T. D.; Noble, C. O. *Proc. 49th ASMS Annual Conference on Mass Spectrometry and Allied Topics*, Chicago, 2001.

(29) Knights, K. A. Ph.D. Thesis, University of London, 2003.

(30) Karas, M.; Hillenkamp, F. *Anal. Chem.* **1988**, *60*, 2299−2301.

(31) Keki, S.; Szilagyi, L. S.; Deak, G.; Zsuga, M. *J. Mass Spectrom.* **2002**, *37*, 1074−1080.

(32) Wang, Y. Q.; Rashidzadeh, H.; Guo, B. C. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 639−643.

(33) Jackson, A. T.; Yates, H. T.; MacDonald, W. A.; Scrivens, J. H.; Critchley, G.; Brown, J.; Deery, M. J.; Jennings, K. R.; Brookes, C. *J. Am. Soc. Mass Spectrom.* **1997**, *8*, 132−139.

(34) Wyttenbach, T.; von Helden, G.; Bowers, M. T. *Int. J. Mass Spectrom.* **1997**, *165*, 377−390.

(35) Fati, D.; Leeman, V.; Vasil'ev, Y. V.; Drewello, T.; Leyh, B.; Hungerbuhler, H. *J. Am. Soc. Mass Spectrom.* **2002**, *13*, 1448−1458.

(36) Hoberg, A. M.; Haddleton, D. M.; Derrick, P. J.; Jackson, A. T.; Scrivens, J. H. *Eur. Mass Spectrom.* **1998**, *4*, 435−440.

(37) Kaufmann, R.; Chaurand, P.; Kirsch, D.; Spengler, B. *Rapid Commun. Mass Spectrom.* **1996**, *10*, 1199−1208.

(38) Edelson-Averbukh, M.; Mandelbaum, A. *Int. J. Mass Spectrom.* **2001**, *210*, 545−556.

(39) Vainiotalo, P.; Malkonen, P. J.; Vainiotalo, A. *Org. Mass Spectrom.* **1989**, *24*, 415−420.