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Journal of Chromatography A, 978 (2002) 177–183

JOURNAL OF CHROMATOGRAPHY A

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Gas chromatographic analysis of the thermally unstable dimethyl methylphosphonate carbanion via trimethylsilyl derivatization

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Received 16 April 2002; received in revised form 12 August 2002; accepted 19 August 2002

Abstract

A sensitive gas chromatographic method was developed to monitor the reaction of lithium diisopropylamide (LDA) with dimethyl methylphosphonate (DMMP) to generate the phosphonate carbanion (DMMPA). Analysis of the DMMPA was complicated due to its thermal instability and lack of a chromophore. To overcome these problems, samples were derivatized with trimethylsilylchloride (TMSCl) to form DMMPA–TMS which was sufficiently volatile and thermally stable for GC analysis. Work-up of the derivatized solution with 10 vol.% 2-propanol in hexanes was necessary to quench residual TMSCl prior to GC analysis. The presence of DMMPA–TMS and other sample components was confirmed by GC–MS analysis. This method was utilized to profile the synthesis of DMMPA as DMMP was added to LDA and then aged at −78 °C. Method precision for DMMPA–TMS of less than 0.2% RSD was achieved for repeat injections after normalization of the response with *n*-dodecane contained in the sample. Due to the thermal instability of the DMMPA, subambient derivatization temperatures were essential to the stability, and consequently, accurate quantification. Under optimized conditions, this derivatization was successfully utilized as a process monitoring tool.

Keywords: Derivatization, GC; Dimethyl methylphosphonate; Phosphonate carbanion

1. Introduction

Carbanion species have long been used as valuable and versatile reactants in synthetic organic reactions. It is known that carbanions are more stabilized by elements in the second row of the periodic table due to their greater electropositive character and polarizability compared with first row elements [1,2]. These characteristics make the phosphonate carbanion a useful tool in organic synthesis. One of the most prominent applications is the Horner–Emmons–Wadsworth reaction to provide alkene compounds in high yield under mild conditions with high (*E*) selectivity [3,4]. Recently, a highly (*Z*) selective reaction has been reported by modification of phosphate esters [5]. Chiral phosphonate carbanion groups are used as chiral templates [2] in the Wittig rearrangement [6] and in the Claisen rearrangement [7]. Due to the versatility and applicability of the phosphonate carbanion in reactions [8–10], analyti-

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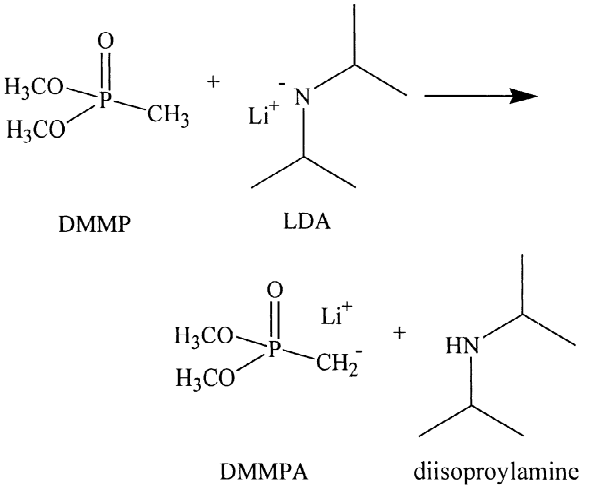
0021-9673/02/$ – see front matter
PII: S0021-9673(02)01305-5

cal methods to characterize these carbanions are essential.

Direct characterization of phosphonate carbanions is difficult due to their reactive nature and thermal instability. For this reason, most reactions of this type are performed at subambient temperatures. To the best of our knowledge, analytical methods to analyze phosphonate carbanions have not been reported in the literature. Here we report an analytical method developed to characterize these unstable carbanion intermediates.

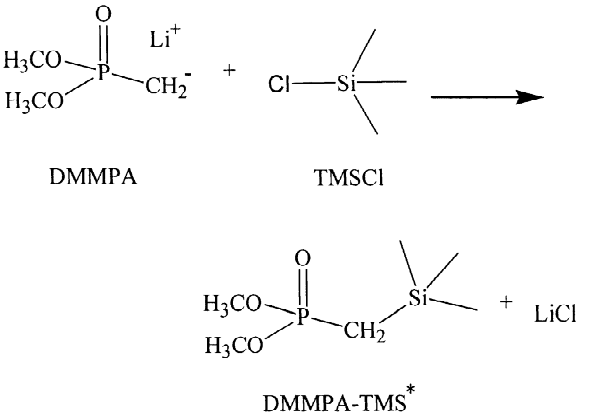
Our approach utilizes a trimethylsilylchloride (TMSCl) derivatization procedure to trap phosphonate carbanions prior to gas chromatographic analysis. TMSCl has been utilized in synthetic organic reactions as a trapping reagent for such compounds as the dianion of 4,4′-dimethyl-2,2′-bipyridine [11], sulfonyl anions [12], and other alkenylsulfenic acid derivatives [13]. Silylation with TMSCl is also a common analytical derivatization method for alcohols, carboxylic acids, amines and amides [14,15]. In addition, a TMSCl derivatization for gas chromatographic analysis has been previously reported as a simple and robust method to monitor unstable lithiated intermediates produced in the synthesis of (tributylstannyl)methanol [16].

This work will present and characterize a TMSCl derivatization procedure to monitor the formation of dimethyl methylphosphonate carbanion (DMMPA) generated from the reaction of DMMP with lithium diisopropylamide (LDA) shown in Eq. (1)



(1)

DMMPA is non-volatile, thermally unstable and rapidly converted back to DMMP in the presence of a protic solvent, complicating chromatographic analysis. However, by introducing TMSCl to this system, DMMPA is converted to DMMPA–TMS (Eq. (2)). DMMPA–TMS has a boiling point of ~120 °C at 22 Torr and is sufficiently volatile to permit GC–FID analysis [17,18]



*Note, structure of DMMPA–TMS indicates C-silylation, however, O-silylation is also possible (2)

DMMPA is derivatized by TMSCl while any residual neutral DMMP remains unchanged. GC analysis of the derivatized solution allows separation of DMMPA–TMS from residual DMMP and other reaction components. This derivatization procedure combined with GC–FID analysis serves as a valuable process monitoring tool for both carbanion formation and consumption reactions.

2. Experimental

2.1. Materials used

Trimethylsilylchloride (TMSCl), *n*-hexyllithium (HexLi), diisopropylamine (DIPA), and dimethyl methylphosphonate (DMMP) were all purchased from Aldrich (Milwaukee, WI, USA). Hexanes and THF were purchased from Fisher Scientific (Fair Lawn, NJ, USA) and 2-propanol (IPA) from EM Science (Gibbstown, NJ, USA).

2.2. Lab scale synthesis of DMMPA profiled by TMSCl derivatization

Into a three-neck flask was charged tetrahydrofuran (THF, 320 ml) and DIPA (20 ml, 1.04 eq). The solution was cooled to −30 °C, and then hexyllithium (HexLi, 55 ml, 0.96 eq) was added slowly with stirring to maintain the temperature < -20 °C. After the addition of HexLi was finished and the solution aged for 30 min, the LDA solution was cooled to −75 °C and then 20 ml of neat DMMP (1.0 eq) was added slowly over 20 min. The reaction was then aged between −75 and −78 °C. Conversion of DMMP to DMMPA was monitored by the TMSCl derivatization procedure for GC analysis described in Section 2.3.

2.3. TMSCl derivatization

Sealed, nitrogen purged scintillation vials were prepared with derivatization solution (10 ml THF, 5 ml TMSCl) and chilled on dry ice. One ml of the reaction solution described in Section 2.2 was removed and immediately added to the chilled TMSCl derivatization solution (~90× molar excess of TMSCl) and stored on dry ice until analysis. One ml of this derivatized sample was added to 5 ml of a 10 vol.% IPA in hexanes solution, to quench residual TMSCl. Residual salts (LiCl) precipitated and the supernatant was directly injected into the GC for analysis.

The reaction described in Section 2.2 was sampled after every 4 ml DMMP had been added (20% increments of the total DMMP charge). Subsequently, reaction samples were derivatized every 5 min for the next hour and then intermittently over the next 4 h. A final sample was taken ~24 h later. All derivatized samples were kept on dry ice (−78 °C) prior to work-up and GC analysis.

2.4. GC–FID conditions

A GC method was developed on a Hewlett-Packard 6890 GC system (Piscataway, NJ, USA) equipped with an RTX-1 column (100% polysiloxane, 15 m×0.32 mm×1.0 μm) from Restek (Bellefonte, PA, USA). Two different temperature programs with initial oven temperatures of 50 and 75 °C were utilized, as noted. After a 5-min hold at the initial temperature, the oven temperature was ramped to 150 °C at 20 °C/min, and held at 150 °C for 5 min. Either a 0.1 or 2 μl injection volume was used, as noted. The inlet temperature was 180 °C and the FID detector was at 250 °C. The helium carrier gas pressure was set at 10 psi, constant pressure mode, with a 50:1 split. The range was set at 0 for maximum sensitivity of the reaction components.

2.5. GC–MS conditions

Method conditions for the GC 6890/MS 5973 (Hewlett-Packard) were similar to the GC–FID conditions. The GC effluent was ionized using electron impact ionization (EI) and the m/z scanned from 10 to 500 using a single quadrupole mass analyzer. Helium carrier gas pressure was decreased to 5 psi to compensate for the vacuum pull of the mass spectrometer. Samples were prepared in the same manner as those analyzed by GC–FID.

3. Results and discussion

Derivatization of DMMPA with TMSCl results in formation of DMMPA–TMS while neutral DMMP remains unchanged. Unlike DMMPA, both DMMPA–TMS and DMMP are sufficiently volatile and thermally stable to permit GC analysis. Additionally, the ratio of DMMPA–TMS to DMMP in the GC chromatogram can be used to measure the extent of DMMPA formation. A typical chromatogram is shown in Fig. 1 of the DMMPA after TMSCl derivatization. Both DMMP and DMMPA–TMS are well resolved from one another and other reaction components, demonstrating method specificity. The broad peak at ~4 min is related to DIPA in the reaction samples and did not interfere in the determination of DMMP or DMMPA–TMS.

3.1. GC–MS analysis

The lack of a DMMPA–TMS authentic sample hindered positive identification of DMMPA–TMS in the GC chromatogram. Therefore, GC–MS was utilized to confirm the formation of DMMPA–TMS after TMSCl derivatization. Similar conditions for

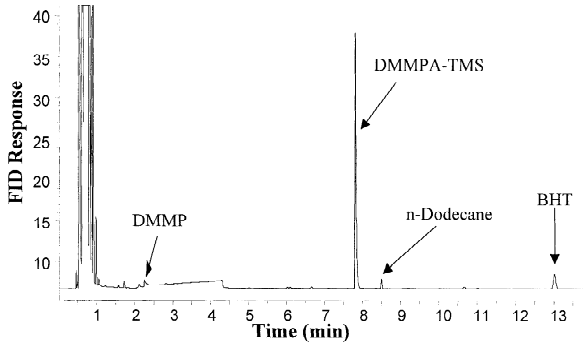


Fig. 1. Typical chromatogram of carbanion formation at an initial 75 °C hold for 5 min. Separation conditions: RTX-1 column (15 m×0.32 mm×1.0 μm); temperature program of a 75 °C hold for 5 min, then ramp to 150 °C and hold for 5 min; 0.1 μl injection (50:1 split at 180 °C); FID at 250 °C; He carrier gas at 10 psi constant pressure. Note, BHT (2,6-di-*tert*.-butyl-4-methylphenol) is stabilizer in THF. Other components identified in text.

GC–MS and GC–FID facilitated identification of DMMPA–TMS by retention time and EI-mass spectrum (see Fig. 2). A small molecular ion peak was observed at an m/z of 196, consistent with DMMPA–TMS. The base peak at 181 m/z resulted from the loss of a methyl group, common for TMS derivatives. Peaks were also observed at 166 and 151 m/z due to loss of two and three methyl groups, respectively. The peak at 123 m/z corresponded to loss of the entire TMS group which was found at 73 m/z.

3.2. Evaluation of derivatization conditions

The effects of both temperature and TMSCl concentration on the derivatization of DMMPA were evaluated. As expected, the temperature of the derivatization solution was critical. Derivatization at room temperature reduced the concentration of DMMPA–TMS by 26% compared with −78 °C (dry ice). This decrease in DMMPA–TMS was likely due to thermal instability of DMMPA and underscores the need to keep the derivatization solution and sampling apparatus cold. In contrast, variations in TMSCl concentration had minimal effect on the extent of derivatization. Three aliquots of DMMPA were derivatized with 21 vol.%, 31 vol.%, or 42 vol.% TMSCl in THF which corresponded to a 60–120 times molar excess of TMSCl relative to DMMP. Within experimental error, the amount of DMMPA–TMS was identical for all three TMSCl concentrations. Therefore, 21 vol.% TMSCl is sufficient to derivatize all DMMPA present.

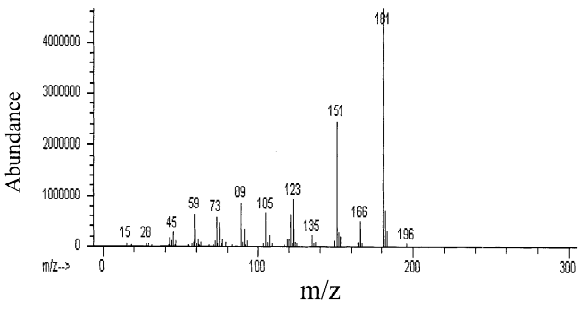


Fig. 2. Mass spectrum of DMMPA–TMS from GC–MS analysis. TMSCl derivative after 20 ml DMMP addition.

3.3. Solution stability of DMMPA–TMS

Solution stability of DMMPA–TMS in the TMSCl/THF derivatization solution and after work-up with 10:90 (v/v%) IPA–hexanes was evaluated. DMMPA–TMS concentration decreased and three major impurities increased after 2 days at room temperature in 10 vol.% IPA in hexanes (see Table 1). Lower levels of these same three impurities were also observed after 2 days storage in TMSCl/THF at room temperature and 6 °C. In contrast, these degradates were not detected after 2 days storage at −16 °C. Further analysis of DMMPA–TMS stability in TMSCl/THF was conducted at 2 and 4 weeks (see Table 2). Only in freezer conditions (−16 °C) was the DMMPA–TMS stable for 4 weeks. In the refrigerator, DMMPA–TMS decreased by almost 50% after 4 weeks, and near complete degradation was observed at room temperature with the appearance of several new impurities. Although the stability of the TMSCl-derivatized solution is superior to that of DMMPA, derivatized solutions should be analyzed immediately or held at −16 °C.

3.4. Profiling carbanion formation reaction: evaluation of the procedure with real samples

This TMSCl derivatization was utilized to monitor a lab scale reaction of DMMP with LDA. Initially, a GC method with an initial oven temperature of 50 °C

Table 1
Stability summary for DMMP derivatized solutions after 2 days. Separation conditions: 50 °C, 0.1 μl injection. Other conditions as in Fig. 1

Component	RRT	$t=0$	In 10 vol.% IPA/Hex $t=2$ days at room temp.	In TMSCl $t=2$ days at room temp.	In TMSCl $t=2$ days at 6 °C	In TMSCl $t=2$ days at −16 °C
Unknown	0.66	0.98	34	2	0.89	–
Unknown	0.87	0.64	10	2	0.76	–
DMMPA–TMS	1.00	26	19	27	30	32
Unknown	1.12	–	3	3	–	–

Peak area normalized to dodecane. RRT, retention time relative to DMMPA–TMS.

and a 0.1 μl injection was used to determine the level of DMMPA–TMS, which is proportional to the amount of DMMPA formed. The peak area of DMMPA–TMS was normalized to dodecane, an impurity found in the hexyllithium starting material. Dodecane was well resolved from other reaction components and similarly retained as DMMPA–TMS. Dividing the DMMPA–TMS peak area by that of dodecane minimized scatter associated with variations in sample volume derivatized and GC injector imprecision. As a result of normalization, less than 0.2% difference in the relative amount of DMMPA–TMS was achieved for replicate injections.

As described in Section 2, aliquots of the DMMPA reaction solution were removed and immediately derivatized with TMSCl over the course of DMMP addition and then periodically during a 4-h age at −75 to −78 °C. During DMMP addition, the level of DMMPA–TMS increased in a linear fashion ($r^2=0.976$), supporting the accuracy of this TMSCl derivatization across a range of DMMPA concentrations. Upon complete DMMP addition, the amount of DMMPA–TMS increased slightly, leveled off, and then degraded slowly during the age at −75 to −78 °C (see Fig. 3). From 4 to 24 h the DMMPA reaction solution warmed to room temperature. A final sample after 24 h was derivatized with TMSCl. No DMMPA–TMS was detected, indicating complete degradation of DMMPA had occurred. This decrease in DMMPA–TMS demonstrates the thermal instability of the carbanion and the importance of subambient temperatures.

Although the GC method with a 50 °C hold and 0.1 μl injection allowed profiling DMMPA–TMS during DMMPA synthesis, detection of trace unreacted DMMP proved difficult. Increasing the injection volume to 2 μl and the initial GC oven temperature to 75 °C increased DMMP sensitivity, in part due to improved DMMP peak shape at higher temperature. Using these modified GC conditions, DMMP was not detected after addition of 4 and 8 ml DMMP, suggesting carbanion formation occurred instantaneously (see Table 3). A small amount of residual DMMP was detected after 12 ml of DMMP were added. Residual DMMP increased as the addition continued, indicating conversion of DMMP to DMMPA slowed over the course of time. Upon

Table 2
Stability summary for DMMPA–TMS in derivatization solution over 4 weeks. Separation conditions as in Fig. 1

Conditions	DMMPA–TMS peak area[a] $t=2$ weeks	DMMPA–TMS peak area[a] $t=4$ weeks
Freezer (−16 °C)	33	33
Fridge (6 °C)	26	16
Room temp.	8	0.5

[a] Peak area normalized to *n*-dodecane.

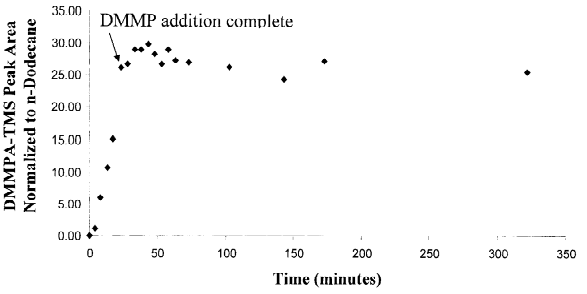


Fig. 3. Reaction profile of lab scale DMMPA formation during addition of DMMP to LDA and subsequent age at −75 °C. Samples derivatized with TMSCl and analyzed by GC–FID. Separation conditions: 50 °C initial oven temperature and 0.1 μl injection. Other conditions as in Fig. 1.

Table 3
Ratio of DMMPA–TMS to DMMP during and after DMMP addition to LDA. Separation conditions as in Fig. 1

Sample	% DMMP converted to DMMPA[a]
4 ml DMMP	No carbanion detected
8 ml DMMP	No carbanion detected
12 ml DMMP	99.1%
16 ml DMMP	90.2%
20 ml DMMP	92.6%
20 ml DMMP + 5 min age	93.6%

[a] Based on the assumption that FID response was proportional to the number of carbon atoms.

complete DMMP addition, the ratio of DMMPA–TMS to DMMP increased slightly after a 5-min age.

3.5. Application to monitoring DMMPA intermediate at pilot plant scale

DMMPA was a key intermediate used in the pilot plant production of a Merck drug development candidate. As such, methods were required to monitor the formation of DMMPA and its subsequent reaction. The TMSCl derivatization procedure presented in this work was applied to monitor the pilot plant scale synthesis and reaction of DMMPA. Due to the thermal instability of DMMPA, samples were immediately derivatized with TMSCl in THF. DMMPA formation was ~86% complete after a 1.5-h age, assuming the FID response for DMMPA–TMS was twice that of DMMP (see Fig. 4A). As shown in Fig. 4B, DMMPA–TMS was not detected after subsequent addition of the other reactant, indicating DMMPA was consumed in the reaction. The level of neutral DMMP increased during the reaction, due, in part, to the water content of the other reactant which quenched a portion of the DMMPA.

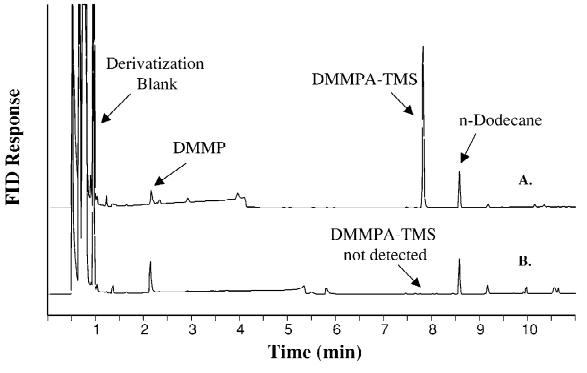


Fig. 4. Pilot plant scale reaction. (A) DMMPA formation and (B) reaction of DMMPA with other reactant. Both samples derivatized with TMSCl prior to GC–FID analysis. Separation conditions: as in Fig. 1.

4. Conclusions

The analysis of unstable carbanion intermediates, such as DMMPA, presented an analytical challenge. The TMSCl derivatization procedure developed in this work addressed this problem by converting DMMPA to the more stable DMMPA–TMS. Formation of DMMPA–TMS was confirmed by GC–MS. When combined with GC–FID analysis, this procedure was demonstrated to be an effective tool to monitor reactions with DMMPA. The derivatization procedure was robust with respect to TMSCl concentration, however, cold temperatures were essential to prevent decomposition of DMMPA and DMMPA–TMS. This procedure allowed a lab scale DMMPA formation reaction to be profiled and a linear increase in DMMPA–TMS was observed during DMMP addition. Application of this procedure to the pilot plant scale synthesis and reaction of DMMPA demonstrated the utility of this procedure to monitor both carbanion formation and consumption reactions.

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