Mechanism for Semiquinone Formation in the Oxygen Negative Chemical Ionization Mass Spectrometry of 2,3,7,8-Tetrachlorodibenzo-*p*dioxin

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The high toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin (2,3,7,8-TCDD) has led to the development of selective and highly sensitive quantitative methods for mass spectral analysis. Analytical selectivity has been demonstrated for the oxygen negative chemical ionization (ONCI) reaction of TCDD in an oxygen-rich plasma. While the reaction product (4,5-dichloro-1,2-benzoquinone anion) of 2,3,7,8-TCDD and oxygen is well known, the mechanism of its formation has not been studied thoroughly. In this report the results of a study involving the reaction of $^{18}O_2$ with 2,3,7,8-TCDD under high pressure and low pressure ONCI conditions are reported. A mechanism is proposed which is compatible with the observation that one ^{18}O atom is incorporated into the product anion.

INTRODUCTION

The high toxicity of 2,3,7,8-tetrachlorodibenzo-pdioxin (2,3,7,8-TCDD) has led to the development of quantitative methodology for its determination at the part-per-trillion (pg/g) level in environmental samples.¹⁻³ Several mass spectrometric and gas chromatographic/mass spectrometric (GC/MS) methods have been developed for this analysis utilizing electron impact,⁴⁻⁶ low pressure negative chemical ionization,⁷ metastable ion monitoring⁸ and atmospheric pressure ionization (API) mass spectrometry.⁹⁻¹¹

One methodology that has been shown to be highly selective for 2,3,7,8-TCDD is the oxygen negative chemical ionization (ONCI) technique first reported by Hunt *et al.*¹² further expanded by Hass *et al.*¹³ and by Mitchum *et al.*^{10,11} The formation of the 4,5-dichloro-1,2-benzoquinone anion via an ion-molecule reaction involving 2,3,7,8-TCDD and O₂ has led to the postulation of a mechanistic pathway.¹³

In this report, the results of a study involving the reaction of ${}^{18}O_2$ with 2,3,7,8-TCDD under ONCI/MS and ONCI/APIMS conditions are reported. A mechanism is presented which is both compatible with the experimental results and incorporates both low and high pressure mechanistic pathways.

EXPERIMENTAL

ONCI/MS. The ONCI/MS experiments were performed with a Finnigan-MAT 4000 mass spectrometer system incorporating the standard PPNCI (Pulse Positive Negative Chemical Ionization) electronics and EI/CI ion source. A Townsend discharge¹⁴ was used to generate the oxygen negative ion plasma by operating the electron trap in the discharge mode. This was accomplished by isolating the trap and applying -800 V to initiate and sustain the Townsend discharge. The discharge was operated at 0.5 Torr oxygen while maintaining the ion source temperature at 200 °C. Spectra of 2,3,7,8-TCDD were recorded while ballistically heating the direct probe. Oxygen-18 experiments were performed at a source pressure of 0.5 Torr ¹⁸O₂ (95.2% enriched).

ONCI/APIMS. The analytical system consisted of a combined gas chromatograph/mass spectrometer (GC/MS) system. The GC was a modified Finnigan-MAT 9500 gas chromatograph incorporating a heated mass spectral interface. The capillary GC column was a 50 m OV-17 fused silica column (Alltech, FSOT-5) with helium as the carrier gas set at a head pressure of 40 psi. The GC injector consisted of a modified commercial splitless capillary injector. Typical GC conditions were as follows: injector, 250 °C; mass spectral interface, 260 °C; column, 120 °C and programmed to 250 °C at 15 °C/min after injection of $1-2 \mu 1$ of sample.

The mass spectrometric system included a recently developed atmospheric pressure ionization (API) source which was designed to operate on a Finnigan-MAT 4000 mass spectrometer system. This source has been described in detail elsewhere.¹⁵ The source utilizes a ⁶³Ni foil as the primary source of electrons, and its operating principles are similar to those described by Carroll *et al.*¹⁶ for a source operating at earth potential. The ionization occurs at atmospheric pressure and the ions, entrained in the carrier gas,

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enter the mass spectrometric system through a 50 μ m orifice. For this experiment, the API source was used in the GC mode in which the fused silica GC column was coupled directly to the API source. The mass resolution, $m/\Delta m$, was set to c. 750, the ion source temperature was 170 °C, and the mass scan parameters were as follows: (i) ¹⁸O₂ experiments: $32\pm0.25\mu$ for 0.05 s; $36\pm0.25\mu$ for 0.05 s; $170-190\mu$ for 2.2 s; (ii) ¹⁶O₂ experiments: the same except that m/z 35 (Cl⁻) was monitored instead of m/z 36.

Confirmational study. As a confirmatory experiment, the ¹⁸O experiments were repeated on an Extranuclear APIMS instrument. For this work the Extranuclear APIMS was interfaced with the GC utilizing a 50 m SP2100 fused silica column. This GC/(Extranuclear) APIMS system has been described in detail elsewhere.^{10,11}

Gas mixtures. The oxygen-18 gas was obtained from Monsanto Research Corporation. The ¹⁸O₂ gas was stated to be 95.2% enriched, and was used as received. For the API experiment the ¹⁸O₂ gas was mixed with prepurified nitrogen to give a gas mixture of 0.1% ¹⁸O₂ in nitrogen (0.1% ¹⁸O₂/N₂). The 0.1% oxygen (natural abundance) in nitrogen gas mixture (0.1% O₂/N₂) was obtained from Linde (Union Carbide) and used as received. The gas mixtures were used as the make-up gas in the GC/MS system and passed through a 13X molecular sieve trap prior to use. In the low pressure ONCI/MS experiment pure ¹⁸O₂ was used.

TCDD. The 2,3,7,8-TCDD was obtained from ECO-Control, Inc. and was used as received. Typically, 5-10 ng of 2,3,7,8-TCDD was injected into the GC or used on the direct probe for each experiment.

RESULTS

The ONCI background spectrum consisted of reagent ions generated from a Townsend discharge operating at a source pressure of 0.5 Torr oxygen. At 200 °C the spectrum consisted of the reagent ions 0^{-1} (m/z 16) and O_2^{-1} (m/z 32). When ¹⁸O₂ was used as the reagent gas the corresponding reagent ions at m/z 18 and m/z36 were observed.

The ONCI spectrum obtained for 2,3,7,8-TCDD is shown in Fig. 1(a). The corresponding spectrum using ¹⁸O₂ as the reagent gas is shown in Fig. 1(b). The major product ion, m/z 176, is quantitatively shifted to m/z 178 and represents incorporation of one atom of ¹⁸O into the 4,5-dichloro-1,2-benzoquinone anion. Under these conditions, an anion corresponding to $[M-Cl+O]^-$ (m/z 301) is shifted 2u upon reaction with ¹⁸O₂ to m/z 303 representing incorporation of one atom of ¹⁸O into the product anion. The reaction of O⁻⁻ with 2,3,7,8-TCDD forms both $[M-H]^-$ and $[M-H+O]^{-.17}$ The anion at m/z 337 (Fig. 1(a)) represents the incorporation of one atom of oxygen with



Figure 1. Oxygen negative chemical ionization mass spectrum of 2,3,7,8-TCDD with (a) O_2 as the reagent gas, and (b) $^{18}O_2$ as the reagent gas.

concomitant loss of one hydrogen atom. This anion was shifted 2u upon reaction with ¹⁸O₂. The 2,3,7,8-TCDD molecular anion formation was not observed in both the ¹⁶O₂ and ¹⁸O₂ experiments; however the $[M-H]^-$ anion was formed via reaction with O⁻⁺.

In the high pressure limit (atmospheric), using an oxygen negative ion reagent plasma generated from ⁶³Ni irradiation, the background mass spectrum with O_2/N_2 as the make-up gas consisted of the major ions, m/z 32, m/z 35 and m/z 37, corresponding to O_2^{-1} , ³⁵Cl⁻ and ³⁷Cl⁻, respectively. When the make-up gas was ¹⁸O₂/N₂ the background mass spectrum consisted of m/z 35, m/z 36 and m/z 37, corresponding to O_2^{-1} , ³⁵Cl⁻, ¹⁸O₂⁻¹ and ³⁷Cl⁻, respectively. Cluster ions of O_2^{-1} with O_2 were not observed due to the source temperature and partial pressure of O_2 .

For this mechanistic study the limited range mass spectrum $(m/z \ 170-190)$ was obtained for 2,3,7,8-TCDD with O_2/N_2 as the make-up gas (Fig. 2(a)). The mass spectrum $(m/z \ 170-190)$ obtained for 2,3,7,8-TCDD with ${}^{18}O_2/N_2$ as the make-up gas is shown in Fig. 2(b). When ${}^{18}O_2/N_2$ was used as the reagent gas, the major product ion $(m/z \ 176)$ shifted by 2u $(m/z \ 178)$, representing the incorporation of one atom of ${}^{18}O$ into the 4,5-dichloro-1,2-benzoquinone anion.

The results of the confirmational study performed using an Extranuclear APIMS system were essentially identical to those obtained on the Finnigan-MAT APIMS system.





Figure 2. Negative ion APIMS mass spectrum of 2,3,7,8-TCDD with (a) 0.1% O_2 in nitrogen as the make-up gas, and (b) 0.1% $^{18}O_2$ in nitrogen as the make-up gas.

DISCUSSION

The reaction of 2,3,7,8-TCDD with oxygen in an electron-rich plasma was first investigated by Hunt *et al.*¹² and later by Hass *et al.*¹³ using the conventional low pressure (1-2 Torr) negative ion mass spectrometric sources. These workers proposed a reaction scheme in which the molecular radical anion of 2,3,7,8-TCDD reacted with O₂ to give the observed 4,5-dichloro-1,2-benzoquinone radical anion $(m/z \ 176)$ plus the neutral 4,5-dichloro-1,2-benzoquinone. Mitchum *et al.*^{10,11} have also studied the reaction of O₂ with 2,3,7,8-TCDD at atmospheric pressure and observed the same semiquinone product.

In order to further elucidate the mechanism for semiquinone production, the ${}^{18}O_2/N_2$ experiment described in this report was performed. The results of this experiment clearly show (compare Fig. 1(a) with 1(b) and 2(a) with 2(b)) that the product ion (4,5-dichloro-1,2-benzoquinone) incorporates one atom of ${}^{18}O$.

The mechanism proposed by Hass *et al.*¹³ predicts that 2,3,7,8-TCDD would react with ${}^{18}O_2$ to give the



m/z 176 product ion with no ¹⁸O incorporation. Since the present results are not in agreement with this reaction scheme, we propose a similar reaction mechanism which is consistent with the experimental observation. Under ONCI conditions it has been proposed that semiquinone formation proceeds via molecular anion addition to neutral O_2 .^{13,14} In con-trast, due to the high concentration of O_2^{--} relative to thermal electron population under ONCI/API conditions nucleophilic addition of O_2^{-1} to neutral 2,3,7,8-TCDD was expected.^{10,11} The reaction mechanism proposed for 2,3,7,8-TCDD reacting in an oxygenrich plasma is shown in Scheme 1 (where *O is ¹⁸O). In this mechanism, the reaction of ${}^{18}O_2$ with 2,3,7,8-TCDD can be bimodal and proceed through a sixmembered transition state which can decompose to give a neutral and an anionic product each of which contains one atom of ¹⁸O. The six-membered intermediate is sterically more favored than the fourcentred intermediate proposed by Hass for formation of the semiguinone.

The scheme accounts for what would appear to be a mechanistic disparity in the mode of formation of intermediate c under ONCI and ONCI/APIMS. Resonance electron capture occurs under low pressure conditions (~1 Torr) in a Townsend discharge initiated oxygen plasma where there is an abundance of electrons with a broad energy distribution. Subsequent radical anion addition to molecular O₂ results in the peroxy intermediate radical anion c which was initially proposed by Hunt.¹⁴ In the high pressure limit, atmospheric pressure, molecular anion formation via resonance electron capture is not observed for 2,3,7,8-TCDD and the predominant process appears to be dissociative electron capture to Cl^{-.11} The absence of a molecular anion under rigorous oxygen-free conditions would suggest the reaction of the radical anion of 2,3,7,8-TCDD with molecular oxygen to be unimportant. However, at atmospheric pressure the formation of the molecular anion is observed for other isomeric tetrachlorodibenzo-p-dioxins¹¹ which suggests that in general the same reactant radical anion is produced in both the low and high pressure regime. Another pathway which must be considered for 2,3,7,8-TCDD and other isomeric TCDDs at atmospheric pressure involves the nucleophilic attack by the radical anion O₂⁻⁻ on neutral 2,3,7,8-TCDD. The nucleophilicity of

 O_2^{-1} is well documented^{18,19} and its reaction with 2,3,7,8-TCDD would also lead to the intermediate peroxy radical anion, c.

The present mechanism does not offer a rationale for production of the $[M-Cl+O]^-$ ($[M-19]^-$) anion of 2,3,7,8-TCDD. The formation of an $[M-19]^{-1}$ anion via nucleophilic substitution in p-chloronitrobenzene, polychlorinated benzenes and biphenyls at atmospheric pressure has been noted.¹⁹ The formation of the $[M-19]^-$ anion for all positional TCDD isomers at atmospheric pressure¹¹ and the formation of the

 $[M-19]^{-}$ anion of 2,3,7,8-TCDD under low pressure ONCI conditions would suggest a mechanism involving simple nucleophilic attack as well as formation via reaction intermediate c as proposed by Hass.¹

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