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Applications of Ion-Molecule Reactions for Distinguishing Organic Isomers in a Tandem Quadrupole Mass Spectrometer

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APPLICATIONS OF ION-MOLECULE REACTIONS FOR DISTINGUISHING ORGANIC ISOMERS IN A TANDEM QUADRUPOLE MASS SPECTROMETER

By

Siu H. Stephen Chan

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ABSTRACT

APPLICATIONS OF ION-MOLECULE REACTIONS FOR DISTINGUISHING ORGANIC ISOMERS IN A TANDEM QUADRUPOLE MASS SPECTROMETER

By

Siu H. Stephen Chan

Many chemical reactions are isomer-specific whereas simple ionic fragmentation, upon which the specificity of normal mass spectrometry is based, is much less so. The unique location of the collision chamber (Q2) of a tandem quadrupole mass spectrometer (TQMS) provides enhanced selectivity for mechanistic studies and analytical applications of ion-molecule reactions. This can be exploited to develop new analytical techniques for differentiating organic isomers.

Low-energy collisions between the $[M-1]^-$ ions of dichlorobenzene isomers and D_2O or ND_3 are found to produce distinct hydrogen/deuterium (H/D) exchange product patterns that allow the isomers to be clearly differentiated when the traditional mass spectrometric techniques fail. The predominant products observed for o-, m- and p-dichlorobenzene are anions substituted with 3, 2, and 1 deuteriums, respectively. The reactivity of D_2O is about three times that of ND_3 for these reactions. A mechanism involving the formation of a five-membered-ring intermediate is proposed for the exchange reaction and is found to fully explain all the experimental results. The results of computer simulations based on the proposed mechanism are consistent with the observed results.

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Ion-molecule reactions are also explored for analyzing isomers of tetrachlorobenzo-p-dioxin (TCDD). For reactions between D_2O and the $[M-1]^-$ ions of TCDD isomers, 2,3,7,8-TCDD is distinguished from 1,2,3,4-, 1,3,6,8-, 1,2,7,8- and 1,3,7,8-TCDD by its unique formation of a single product having only one H/D exchange. When alcohol molecules are reacted with the $[M-1]^-$ ions from isomeric TCDDs, 2,3,7,8-TCDD is distinguished from the other less toxic isomers by its lower reactivity for the formation of the $[M-1+alcohol]^-$ adduct products.

While ions formed by low-energy ion-molecule reactions in Q2 provide useful information for isomeric differentiation, these ions will not be detected if their mobilities are too low to reach the detector. The detection of these ions can be improved if a gentle longitudinal potential gradient is created along the ion path of Q2. Theoretically, this can be accomplished by using Q2 made of BeO rods on which are winded high-resistance wires used for the generation of the transverse field. The nearly constant potential gradient will provide the ions with a continuous force towards the detector.

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ACKNOWLEDGEMENTS

I would like to thank my research advisor, Dr. Christie Enke, for his help and guidance that make the completion of my Ph.D a reality. Through his scientific insight, I have learned a lot about being a better scientist. I also would like to thank the other members of my guidance committee, especially Dr. William Reusch who agreed to take on the duty of a second reader for my dissertation just two weeks before my oral defense.

During my graduate school career at Michigan State University, the experience of teaching has provided me a great deal of personal satisfaction. Thanks to all my students who allowed me to enjoy myself as a teacher. Especially during the rough years of my research, seeing my classes was something that I was looking forward to every week.

The most rewarding experience of my life comes from the year when I was a secondary school teacher many years ago. Thanks to the tremendous friendship and help offered by all of my colleagues who accepted me just the way I was. I would also like to extend a special thank to all the kids of my classes who gave me so much fun for being a teacher. I will never forget the hiking trips with all the kids. It certainly was the toughest job I have ever loved.

I also want to express my gratitude to two of my undergraduate teachers, Drs. Paul Merritt and Nick Zevos for their kindness, help and confidence in me that stirred me into the field of chemistry. My undergraduate career at Potsdam, New York, would be less enjoyable if not for the warm hospitality of Mr. & Mrs Sellers during my final year there. Thanks for all the gifts, camping trips and the party in honor of my graduation. You guys have made the cold winter of Potsdam so warm to live in.

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Over the past many years as a graduate student, I am grateful to have learned many things beside chemistry. Now I understand more about the importance of humanity for the development a mature decent human being. Through appropriate education starting from very young, perhaps, one day we may live in the "dream world" of Dr. Martin L. King Jr..

Although the road to the completion my Ph.D have been a long and tough one, I would like to express my greatest thank to my parents, sisters and brother for their love, moral support and understanding. Finally, I can say with enthusiasm that I am still alive.

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Introduction

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CHAPTER 1

INTRODUCTION

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Solvent molecules often play an important role in ionic reactions ırring in solution. The effects of solvation can be eliminated if the ctions are carried out in the gas phase. A mass spectrometer can be a ıplete laboratory for studying ion-molecule reactions. The gas phase ınique requires only a small fraction of the sample amount normally uired for similar studies in the condensed phase.

J. J. Thomson first reported the products of ion-molecule reactions in 3 (1). The vacuum technology in Thomson's day is considered primitive oday's standard. As a result of poor vacuum in the mass spectrometer, nerous ion-molecule reactions often took place; and the reactions were erally treated as a nuisance. With the advent of better vacuum and s spectrometric technology, a systematic study of ion-molecule tions was begun in 1952 with the work of Tal'roze and Ljubimova (2). rest in ion-molecule reactions has been widespread since the early s. Over the past three decades, numerous techniques have been found ul for carrying out ion-molecule reactions. These techniques will be ly described in the latter part of this chapter.

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The main objective of this dissertation is the application of ion-ecule reactions for distinguishing among isomers of organic ıpounds. A tandem mass spectrometric system (MS/MS) allows ection of a specific mass ion from the ion source for reaction with ected reagent molecules in an isolated reaction chamber. Although ny MS/MS systems exist today, the unique features of a triple adrupole mass spectrometer are well suited for analytical applications of -molecule reactions. In this thesis, these features are compared to those other techniques with emphasis on the applicability of ion-molecule ıctions as analytical tools.

llision Processes in Ion-Molecule Reactions

The first step in an ion-molecule reaction is a collision between an ı and a neutral molecule. The collision causes energy or momentum to change between the interacting species. The results of the collision pend very much on the collision energy involved. Although an exact atment of collision processes is quantum mechanical (3), most collision ults can be sufficiently explained by models based on classical chanics. The classical ion-molecule collision theory has been discussed detail by Su and Bower (4). In general, a fraction of the relative nslational energy prior to collision is converted into internal energy of collision partners. The internal energy gained may promote both the and molecule to some new excited electronic, vibrational and rotational es. Depending on the energy within the interacting systems, the tations can cause fragmentation, ion-molecule reactions and even

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oton emission. For vibrationally excited ions, collisions at thermal ergy can also result in the reduction of internal energy.

Most studies of ion-molecule collisions have been performed in high-ergy regimes (over 1 KeV) or low-energy regimes (below 100 eV). Electric d magnetic sector instruments have been used almost exclusively for the lisions at high energy (> 1 KeV). The products of these collisions are ically monitored in mass-analyzed ion kinetic energy spectrometry, or KES, for structural elucidation of ionic species (5). In this high-energy ime, momentum transfer is minimal. The direct electronic excitation uced by the collisions often causes unimolecular decay (6). Because of short interacting time for the collision species, ion-molecule reactions often not observed during high energy collisions. However, product s resulting from dissociation, charge stripping, charge inversion and rge exchange may be formed.

In contrast to the high-energy collisions, the energy involved in low-rgy collisions can not efficiently promote electronic excitation. These energy " billiard ball " collisions involve mostly vibrational excitation by nentum transfer, and produce large scattering angles compared to e in the high collision energy regime. During these low energy esses, the energy conversion efficiency from translational to internal is emely high. In many cases, efficiencies close to 100% are observed (7). use the efficiency of collisionally activated dissociation (CAD) increases increased internal energy, low energy collisions are an efficient ns to promote CAD. When collisions occur at very low energies (< 3 eV e interaction time between ions and neutral molecules can be several

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ational periods long. During this long interaction time, ion-molecule ctions involving the formation of new chemical bonds may occur. isions at thermal or near thermal energies are typically used for lies of ion-molecule reactions; thus, these reactions are not easily lied in sector instruments. Although hybrid instruments involving the of a combination of sector (magnetic and electric) and quadrupole ces can also be used for low-energy ion-molecule reactions, most of e reactions have been studied in quadrupole, ion trap, flowing afterglow ion cyclotron resonance instruments.

In most experiments, a well-defined collision cell or region is gnated for ion-molecule reactions. In the case of a quadrupole rument, neutral molecules are injected into the collision cell under mal energy. The collision energy for ion-molecule reactions is due to inetic energy of the ions entering the collision cell. The kinetic energy ergy in laboratory frame (E_{lab}) is derived from the potential difference een the ion source and the collision cell. However, conservation of y and momentum in a collision requires that only a fraction of E_{lab} is able for excitation of the interacting species (8). This fraction is the ive collision energy (E_{cm}) in a coordinate system moving with the r of mass of the collision partners. As such, E_{cm} is defined as E_{lab} the ratio of the mass of the neutral molecule (M_n) and the sum of the s of the neutral molecule and the reactant ion (M_i). It is described by uation as:

$$E_{cm} = E_{lab} [M_n / (M_n + M_i)]$$

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This equation implies that for a collision with a specific ion, the effective energy can be varied by either changing the potential of the collision cell relative to the ion source or changing the mass of the neutral molecules. A heavier neutral molecule provides a more efficient means to convert E_{lab} into internal energy. In the case of electron impact, the large mass ratio of aneutral molecule to an electron makes E_{lab} nearly equal to E_{cm}.

The average rate of collisions can be increased by raising the pressure of neutral collision gas in the collision cell. Generally, under multiple collision conditions, the average internal energy of ions often ncreases over that of single collision conditions (9,10). This increase in nternal energy may improve the yield of CAD products. For reactive ion-molecule collisions, multiple collision conditions result in a higher frequency of ion-molecule interactions, which in turn increase the product yield from such reactions. When collisions occur at thermal energies, multiple collision conditions may also have a stabilizing effect on reaction ntermediates which are often needed for the formation of final ion products. Clearly, for reaction products arising from second or higher rder sequential reactions, multiple collisions are necessary.

Most ion-molecule reactions occur at thermal or near thermal nergies. Although the majority of these reactions are exothermic or hermoneutral, some endothermic ion-molecule reactions have also been tudied (12-15). The dependence of product yield on translational energy iffers for endothermic and exothermic reactions (16). Average dipole rientation (ADO) theory predicts that ion-molecule reactions proceed via rmation of a long-lived collision complex (17). Accordingly, product ion

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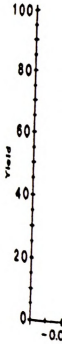
eld of an exothermic reaction is greatest for ion kinetic energy
proaching zero and decreases with increasing ion kinetic energy. This
ay be explained assumming that products are formed only if the collision
mplex lives long enough. As kinetic energy increases, the life-time of the
mplex becomes too short to form products. The formation of ion-molecule
duct ions is particularly energy sensitive (18,19). The optimum kinetic
ergy for adduct ion formation is often barely positive; the energy range
ailable for these reactions is extremely narrow (see Figure 1-1).
wever, there are many instances where adduct ions formed through
thermic reactions may not be observed as final products at all. In such
ses, the excess energy released is large enough to cause rupture of
emical bonds.

Ion fragmentation derived from ion-molecule collisions provides
uable structural information regarding the original molecule. The yield
l efficiency of the fragmentation depend largely on the amount of excess
rational energy in the chemical bonds. For collisions at the very low end
he low-energy regime, the total internal energy deposition to an ion
eases somewhat linearly with an increase in collision energy. This
ease levels off as the collision energy increases up to a certain level.
a large molecule, the internal degrees-of-freedom is a large number
to the large number of chemical bonds. The fragmentation efficiency
a large ion is extremely low, even at very high collision energies,
use each chemical bond shares only a small fraction of the total
rnal energy gained (20,21). As an alternative, to collision-induced
mentation, ion-molecule reactions may be explored for structural



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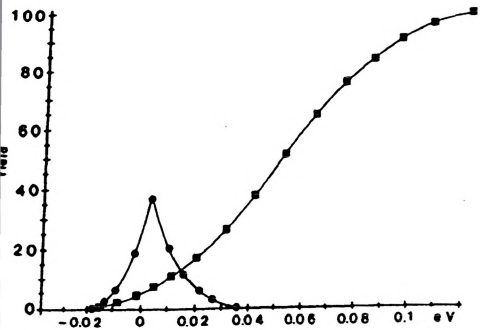

center-of-mass collision energy

'igure 1-1 Center-of-mass axial kinetic energy spectrum of protonated α-hydroxyisobutyrate ethyl ester. The yield of adduct ions, m/z 150 (●), and the most intense fragment, m/z 115 (■), were recorded against the experimental center-of-mass energy of the protonated molecular ion (m/z 133) (Adapted from Ref. 18)

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formation that is often much more structure-specific than the result of nple ionic fragmentation (22,23).

stinguishing Organic Isomers in Tandem Mass Spectrometry

Mass spectrometry (MS) has been the method of choice for analyzing nples that demand low detection limits and structural information. ny examples may be found where a single stage mass spectrometer uipped with electron ionization (EI) or chemical ionization (CI) is used to oduce sufficiently distinct mass spectra to allow the differentiation of tain organic isomers. Still many isomers are found to have identical or y similar mass spectra when the traditional MS techniques are used. In ticular, the differentiation of many positional isomers of aromatic npounds are known to be extremely difficult. These isomers include orious compound types such as polychlorinated biphenyls (PCBs) and ychlorodibenzo-p-dioxins (PCDDs). Over the past few decades, a number different approaches have been adapted in order to increase the mass ctral differences of some isomers. These are primarily within the nework of MS/MS, which provides an additional mass spectrometric ension to allow extra processes for the ions. These extra processes may ude metastable decomposition, collisionally activated dissociation D), photodissociation and ion-molecule reactions. All have been onstrated to be useful in differentiating organic isomers. In icular, the specificity of ion-molecule reactions has been useful when fragmentation techniques failed (24).

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MS/MS in Sector Instruments for Isomeric Differentiation

The application of MS/MS for isomeric differentiation was first ribed by Shannon and McLafferty (25) for analyzing isomers of $C_2H_5O^+$. o-sector instrument was used to monitor ions derived from metastable mpositions in the field-free region between sectors. The differences in extent of the competing decomposition pathways were used to rentiate the isomers of $C_2H_5O^+$. Since this beginning, the application of MS for distinguishing isomers has been extended to different types of uments. Different processes have also been applied to each of the rent instruments for differentiating isomers. The features and rtical applications of non-sector instruments will be discussed in the section.

The application of metastable decomposition was further extended by fferty et al. (26) to differentiate isomers of $C_2H_6N^+$ and $C_3H_8N^+$ using ame instrumental settings as described before. Larger molecules such ereoisomers of 2-acetamido-2-deoxyhexose were also successfully entiated on the basis of differences in the relative abundances of the table ions (27). Despite these successes, metastable decompositions do not produce useful information for isomeric differentiation.

The yield of ionic fragmentation can be improved if ions are allowed e collisions with neutral molecules in the field-free region of a sector ment. Such collisions increase the internal energy of the ions and CAD to occur. In order to reduce the effect of scattering due to tic collisions, neutral molecules with low molecular weight such as

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re introduced into the field-free region to induce CAD. Kiremire et al. used CAD to produce diagnostic fragments that allow the rentiation of pyranococumarin isomers (28). Recently, McLafferty et ave employed a combination of both ion-molecule reactions and CAD to rentiate $C_4H_4^+$ isomers (29). In this case, NH_3 was introduced into the source under EI conditions to produce distinctive products of ion-cule reactions for three different $C_4H_4^+$ structures originated from different hydrocarbons. The ion products at m/z 69, presumably the $_7N^+$ adduct, were subjected to CAD with He in the field-free region of a sector instrument. The distinctive spectra thus obtained allow the ers to be distinguished. The CAD spectra of unreacted $C_4H_4^+$ isomers lmost indistinguishable.

The internal energy of an ion can also be increased by interaction photons. The process of photodissociation was applied in the field-free n of a reversed geometry double focusing mass spectrometer to entiate isomers of xylene and ethylnitrobenzene (30,31). In the case of e, the difference in translational energy released by the $[M]^+$ ions as a ion of photon energy was used to differentiate the isomers. For the rs of ethylnitrobenzene, $[M\text{-}OH]^+$ ions were selected to interact with ns. Differences in the relative photoabsorption cross-sections of the $H]^+$ ions were used for isomeric differentiation.

Although the energetic processes in sector instruments do not allow olecule reactions involving the formation of new chemical bonds, stripping (32,33) and charge exchange reactions (34,35) do occur. s et al. (36) have used the charge stripping reactions to analyze

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ners of $C_2H_5O^+$ ions. Stripping of an electron from $C_2H_5O^+$ by energetic sion with He generates a doubly charged ion. The resulting mass tra permit the clear identification of four distinct $C_2H_5O^+$ species. The ge exchange reactions were explored by Guilhaus et al. (37) to erentiate isomers of $[C_6H_6]^{2+}$ ions. The doubly charged ion can ange a charge with Xe atoms in a high-energy collision to form two ly charged ions. These processes can be represented as :

$$[C_6H_6]^{2+} + Xe \text{ -------> } [C_6H_6]^{+} + Xe^{+}$$

isomers of $[C_6H_6]^{2+}$ were distinguished primarily by the relative dances of $[C_6H_6]^{+}$ ions formed by the charge exchange.

omeric Differentiation in MS/MS Instruments Suited for Ion-Molecule eactions

Ion-molecule reactions can be performed on instruments that allow olecule collisions at low energies (a few eV or less). A technique that ll suited for ion-molecule reactions can still allow CAD, because some entation occurs even at very low energies. Thus, by adjustment of the ion energies, either ion-molecule reactions or CAD products can be ed. With the advent of laser technology, photodissociation can also place in instruments that are primarily used for ion-molecule ons. With this in mind, applications of CAD, photodissociation and olecule reactions will be discussed in this section. The emphasis will ion-molecule reactions and their potential in analytical applications.

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Currently, there are four different MS/MS techniques that are ›mmonly used to perform ion-molecule reactions. They are tandem ›wing afterglow-selected ion flow tube (SIFT)-drift (or tandem FA-SIFT) 8), fourier transform ion cyclotron resonance (or FTMS) (39-41), ıadrupole ion trap mass spectrometry (ITMS) (42-45) and triple ıadrupole mass spectrometry (TQMS) (46-48). The unique features, which ll be described later, of TQMS are utilized in this dissertation to explore ıalytical applications of ion-molecule reactions for distinguishing organic ›mers.

Tandem Flowing Afterglow-SIFT-Drift

Tandem FA-SIFT is an extension of the flowing afterglow technique veloped in 1963 by Ferguson et al. (49). In this technique, the flowing erglow source is used to generate the reactant ions. These ions are then ss-selected by the selected ion flow tube (SIFT) to enter the flow-drift e, where neutral reagents are added to carry out ion-molecule reactions. lium buffer gas is used in the flow-drift tube to thermalize the ions and ry them to the reaction regions. The ionic reactants and products are ected by a quadrupole mass filter coupled with an electron multiplier. fact, this instrumental configuration is very similar to that of a triple ıdrupole mass spectrometer. The major differences of the two are in the ign and configuration of the ion source and the collision region, So far, FA-SIFT technique has been limited to fundamental studies of the nomena of ion-molecule reactions. Only a few laboratories are equipped ı such instruments, and they are not commercially available. Perhaps,

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popularity of this technique is overshadowed by the higher versatility greater sensitivity of the TQMS for analytical applications.

adrupole Ion Trap

Although the basic concept of a quadrupole ion trap was first ibed by Paul and Steinwedel in the 1950s (50), the use of a quadrupole rap (ITMS) as an MS/MS instrument was first reported by Louris et al. 87 (42). This three-dimensional quadrupole device consists of two end-lectrodes and a central ring electrode to which direct current and frequency voltages can be applied. The radiofrequency voltage mines the mass-to-charge ratios of ions which can be trapped within eld and can be used to selectively eject ions from the traps. A high round pressure of helium (10^{-3} torr) is normally used in the trap to the motion of stored ions for optimum operation. Chemical tion (CI) with a variety of CI reagents can also be carried out at ures orders of magnitude less than those employed in traditional CI). Photodissociation (54), CAD (55-57) and ion-molecule reactions (53,) have been performed in the MS/MS mode of an ITMS. Over the past ears, the popularity of ITMS has grown tremendously. The technique ticularly well suited for ion-molecule reactions. Its versatility will, in ture, undoubtedly allow for the development of many analytical ations in a wide range of areas.

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ourier Transform Ion Cyclotron Resonance

Fourier transform ion cyclotron resonance is also known as FTMS. an extension of ion cyclotron resonance (ICR) that was first described :. O. Lawrence in 1930 (59). The introduction of fourier transformation CR by Comisarow and Marshall (39) has made FTMS the best technique ion-molecule experiments requiring ultra-high mass resolution. In a cal FTMS experiment, ions are generated by electron impart in a cubic at a total pressure of 1 to 100 μtorr. These ions are trapped in the cell to the effects of magnetic and electric fields and follow a circular ectory. After a certain time, referred to as trapping time, during which tions with neutral molecules can take place, the ions are then excited to er orbits by a fast frequency-swept radiofrequency pulse. The coherent ion of all excited ions induces image currents in a circuit shunting two iver plates. These currents are amplified and transmitted to a puter for fourier transformation to generate an interpretable mass trum (60,61). The basic features of FTMS are quite similar to ITMS, pt for a difference in resolving power and operational pressure. A lving power of over a million is achievable with a modern FTMS rument. The technique is particularly well suited for fundamental ies of ion-molecule reactions.

Although some of the first papers about ICR were related to CAD esses (62,63), the technique was mostly used for ion-molecule reactions. feasibility of using ion-molecule reactions to differentiate organic ers has been demonstrated by numerous reports (64-76). These ers include $C_2H_5O^+$ (68,71), C_5H_{10} (69), $C_6H_6O^+$ (70), trans- and cis-4-

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t-butylcyclohexyl acetate (75), C_8H_{10} (72), C_7H_8O (72), trans- and cis-α-calone (73), $C_3H_3^+$ (76) and tautomers of $C_3H_8N^+$ (74). Most of the studies re motivated by the fact that the majority of these isomers do not give ficient mass spectrometric information for isomeric differentiation. The ccess of these examples demonstrates the usefulness of the high ecificity of ion-molecule reactions compared to the other processes. In dition to ion-molecule reactions, photodissociation has also been used in MS to differentiate isomers. Under irradiation by a laser beam, isomers $C_4H_9O_2^+$ (77,78) and $C_5H_6^+$ produce characteristic fragment patterns that y be used for their differentiation.

Triple Quadrupole Mass Spectrometry

The development of a triple quadrupole mass spectrometer (TQMS) by t and Enke (46,47) for general MS/MS applications has stimulated espread interest in MS/MS techniques. The instrumentation of TQMS olves three quadrupole devices positioned in series. The first quadrupole) and the third quadrupole (Q3) are used as mass-filters to which a bination of direct-current (DC) and radiofrequency (RF) potentials are lied. Mass selection and mass scanning are accomplished by sweeping DC and RF potentials applied to the four poles of Q1 and and Q3. The avior of ions traveling through a quadrupole is described by the Mathieu erential equations (79,80) that describe the stable trajectory of ions racting with the fields inside the quadrupoles. The second quadrupole is used as a non-mass-selective collision chamber where only RF ntial is applied. The processes of ion-molecule interactions occur in Q2 ng the MS/MS operation. The main function of the RF potential applied

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2 is to focus ions scattered by the collision processes. The transmission erties and the ion containment efficiency of an RF-only quadrupole filter have been discussed in detail by Miller et al. (81) and Dawson et 82). The versatility of a TQMS instrument allows a variety of ational modes, which provide different types of MS and MS/MS mation. All the possible modes of a TQMS have been reviewed and ssed in depth by others, including colleagues and students of Dr. C. G. (83). Only the mode used for the experiments involving ion-molecule ons will be discussed.

All the experimental work described in this dissertation was rmed on either a Finnigan TSQ-70B or a Finnigan TSQ-700 TQMS ument. Except for some of the work described in Chapter 3, all the ments were performed on the TSQ-70B model. Both Finnigan models the design of a curved collision chamber that is different from the ional linear design. It is believed that the curved design reduces noise the formation of neutral products that cannot be transmitted beyond urved chamber (84). In addition, there is also a fundamental nce in the design of Q2 between the TSQ-70B and TSQ-700 models. In echnically the TSQ-700 model should not be considered as a triple upole system because it uses an octopole design for the collision er. Theoretically, the octopole design can reduce the radial energy ed by the RF potential and allow a higher ion transmission efficiency The operational mode used for all MS/MS experiments is depicted in 1-2. Briefly, ions created in the source are mass-selected by Q1 to he collision chamber (Q2) where neutral reagents are introduced for

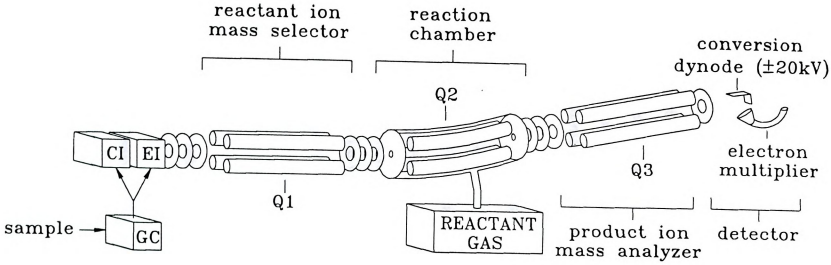

Figure 1-2 Schematic of a triple quadrupole mass spectrometer based on the Finnigan TSQ-70B model

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action with the ions. Product ions emerging from Q2 are then mass-
zed by Q3 for product mass spectra.

The versatility of this TQMS allows for very wide range of ionization
niques. They include, but are not limited to, CI, EI, fast atom
bardment, atmospheric pressure ionization and electrospray. In
ast, only limited ionization techniques are allowed in Tandem-FA-
, ITMS and FTMS. The capacity of a TQMS to provide independently
mass resolution to both mass filtering stages results in an instrument
high selectivity. An even higher selectivity can be achieved when a
S is coupled with gas or liquid chromatography. This capacity is
rtant for trace analysis of environmental samples. Perhaps, the two
est advantages of a TQMS over the ion trapping techniques are the
ie location of its collision chamber and the independent and
taneous of the operation of the three quadrupoles. In a TQMS, ion-
ule reactions take place in a region where the ions and the molecules
atially separated from the ion source by a mass filtering device. Such
ing ensures that only ions with a specific m/z value can enter the
on chamber. Since ion selection, ion-molecule reaction and mass
ng all occur stimultaneously in different regions, analytical samples
introduced continuously into the system without any interruptions.
trast, ITMS and FTMS require the ionization, ion-molecule reaction
ass scanning to be processed in the same region at different times.
an ion trapping instrument is in its trapping mode, it loses its
n as a mass analyzer. Finally, a TQMS has the advantage of
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ability of controlling collision energy is important for energy-resolved lies of both endothermic and exothermic reactions.

Despite all the advantages described above, there are some dvantages in using a TQMS to study ion-molecule reactions. First of the residence time of the reactant ions in the collision chamber is very t compared to those of ITMS and FTMS. This short residence time may ibit the formation of some ion products which require long-lived rmediates and have slow kinetics. Second, an ion can also gain an etermined amount of radial kinetic energy due to the RF potential ied to the collision chamber. The octopole design of the TSQ-700 model elieved to minimize this problem. Finally, because the product mass tra obtained from the TQMS represent only the ions transmitted into they are not necessarily truly representative of the product distribution e collision chamber. This could be a problem when sampling product derived from ion-molecule reactions conducted under thermal energy high collision pressure conditions. This particular problem will be ssed in greater detail in Chapter 5.

The primary ion-molecule interaction used in most tandem lrupole systems is CAD. The CAD efficiency in a TQMS is often ficantly higher than that of a multisector instrument. For some ers, information provided by CAD in a TQMS is selective enough to it their differentiation (86-88). Isomers of dimethoxyindole (87), ral morphinan (86) and o/p bisphenol-A (88) have been successfully nguished by CAD. However, when CAD fails to produce useful

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rmation for isomeric differentiation, the specificity of ion-molecule tions may be useful.

The use of the TQMS for ion-molecule reactions is relatively new. et al. (89) first explored the use of ion-molecule reactions in a TQMS to rentiate isomeric structures. Since then, several laboratories have rted successes in using ion-molecule reactions to differentiate isomers. ey et al. used ammonia as the neutral reagent to differentiate isomers 2-cyclopententanediol (90) $C_2H_5O^+$ (91) and hexachlorinated biphenyl The same group also explored the use of endothermic proton transfer ions and trimethysilyl ions to differentiate isomers of $C_6H_6^+$ (93) and yclopententanediol (94), respectively. Buta-1,3-diene and benzene were by Jalonen (95) to differentiate the structures of $C_2H_5O^+$ and $C_2H_3O^+$ ers. Kostianinen et al. (96,97) have explored the use of oxygen to rentiate several isomers of tetrachlorodibenzo-p-dioxin. Recently, h et al. (98) have reported success in using dimethyl ether to entiate $C_7H_7^+$ isomers.

Despite the applications described above, the use of ion-molecule ons in the TQMS is still limited to only a few laboratories. Their sses demonstrate the analytical potential of ion-molecule reactions in quadrupole mass spectrometry. By taking the advantages of the city of ion-molecule reactions and the sensitivity and selectivity of the , we have explored novel applications of ion-molecule reactions for guishing organic isomers, which will be described in this tation.

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CHAPTER 2

REACTIVITIES OF DEUTERATED REAGENTS TOWARDS THE [M-1]$^-$ IONS OF CHLORINATED BENZENES FOR H/D EXCHANGE REACTIONS

roduction

The analytical potential of hydrogen/deuterium (H/D) exchange ctions was first demonstrated by Hunt and co-workers (1) for erentiating primary, secondary and tertiary amines. This early work loyed chemical ionization (CI) using ND_3 as a reagent to produce ND_4^+ t would then undergo H/D exchange reactions with amine molecules. extent of the degree of H/D exchange is different for primary, secondary tertiary amines. Based on a comprehensive study, the same group r reported H/D exchange reactions on the [M-1]$^-$ ions of several aromatic pounds with deuterated reagents (2). Although protonated molecules n many aromatic compounds were also found to have H/D exchange tions with deuterated reagents (2,3), thus far only H/D exchange tions with [M-1]$^-$ ions were shown to produce useful mass trometric information for differentiating aromatic isomers (4). Such ytical potential encourages further studies on H/D exchange reactions [M-1]$^-$ ions. This chapter will describe the H/D exchange reactions een the [M-1]$^-$ ions of various chlorinated benzenes and deuterated ents . The focus of this chapter is a study of relative reactivities for rent deuterated reagents with different types of [M-1]$^-$ ions. The

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anistic details for the reactions of anions of dichlorobenzene isomers e discussed in Chapter 3.

rimental

A triple quadrupole mass spectrometer (Finnigan TSQ-70B model) ised to perform all the experiments. The operation and configuration triple quadrupole system have been described in Chapter 1. During xperiments, only $[M-1]^-$ ions were allowed by the first quadrupole (Q1) er the collision chamber for reactions with deuterated reagents. Unit resolution was maintained in the third quadrupole whereas Q1 was for slightly better than unit mass resolution. Samples are introduced he ion source by a direct insertion probe. The $[M-1]^-$ ions were ced by CI using NH_3 as a reagent. The temperature of the ion source he manifold of the spectrometer were set at 150 °C and 70 °C, tively. The pressure of the CI source and the collision chamber for periments were maintained at 1.5 torr and 4 mtorr, respectively. A on energy of 2.8 eV (according to the Q2 offset voltage reading of the ment) was used. All the experiments were performed twice to test for tency in the results. The replicates were compared for discrepancies ns of relative peak ratios and the absence or presence of stray peaks. ate runs consistently gave spectra that were approximately the The data shown come from one set of results rather than the e of the replicates.

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All the chlorinated benzenes were purchased from Aldrich Chemical npany, Inc., Milwaukee, WI. All the deuterated reagents were chased from Cambridge Isotopes Laboratory, Woburn, MA except ND_3 ch was obtained from MSD Isotopes, Montreal, Canada. All the micals were the best purity available and were used without further ification.

ults and Discussion

Characterization and Optimization of H/D Exchange Reactions Involving the $[M-1]^-$ of o-Dichlorobenzene

Many ion-molecule reactions have been found to occur only within narrow collision energy ranges (5,6). In a triple quadrupole system, ionic products derived from successful ion-molecule reactions may not bserved if the instrumental parameters of the system are not set at opriate values. Usually, the standard tuning program can do a factory job in optimizing many instrumental parameters. For a given molecule reaction system, the two major factors that control the ency of the reaction system are collision energy and collision pressure. e factors must be optimized in addition to the optimization by the ng program. Sometimes, compromises have to be made between the factors in order to achieve a maximum yield of a certain ion product. e H/D exchange reactions are the main theme of this research work, section is devoted to the characterization and optimization of the effects lision energy and collision pressure on the exchange reactions. The

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y is based on the H/D exchange reactions between the $[M-1]^-$ of o-lorobenzene and D_2O.

The kinetic energy of an ion entering the collision chamber is rolled by the offset voltage potential between the ion source and the sion chamber. Theoretically, no ion should be able to enter the collision nber if the offset voltage potential is set at zero or less. In reality, for a plex instrument such as the Finnigan TSQ-70B, the kinetic energy of on as it leaves the source, as well as other instrumental parameters, affect its transmission. The ion transmission efficiency as a function ffset potential for the $[M-1]^-$ of o-dichlorobenzene is shown in Figure 2- his figure demonstrates that some ions can still manage to enter the sion chamber even at an offset potential below zero. From the ative plot, the transmission window of the ions centers at around 1.8 V a width of about 3 V. Transmission of the ion reaches a maximum at 3.0 V. This information is important because it suggests that there d be little or no gain in ion transmission into the collision chamber if ffset potential is increased beyond 3.0 V. For ion-molecule reactions ring at high collision pressures, a higher collision energy would favor dissociation and increase the chance of ion scattering. If the offset tials are set at below 3.0 V, ion transmission into the collision ber could be reduced significantly. Optimum is balance between ion nission and reaction efficiency. It may be different for each reaction; will almost always be in the low energy region.

Ion-molecule reactions between the $[M-1]^-$ ions of o-dichlorobenzene $_2O$ produce H/D exchange ion products. All three hydrogens on the

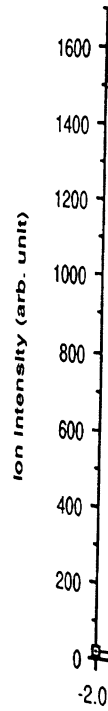
Ion Intensity (arb. unit)
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Figure 2-1 Io
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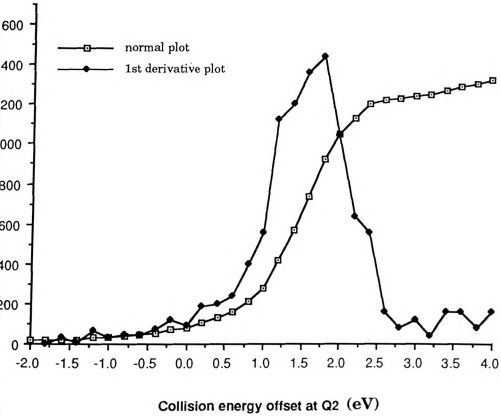

2-1 Ion intensity as a function of collision energy offset at Q2 for the $[M-1]^-$ of o-dichlorobenzene.

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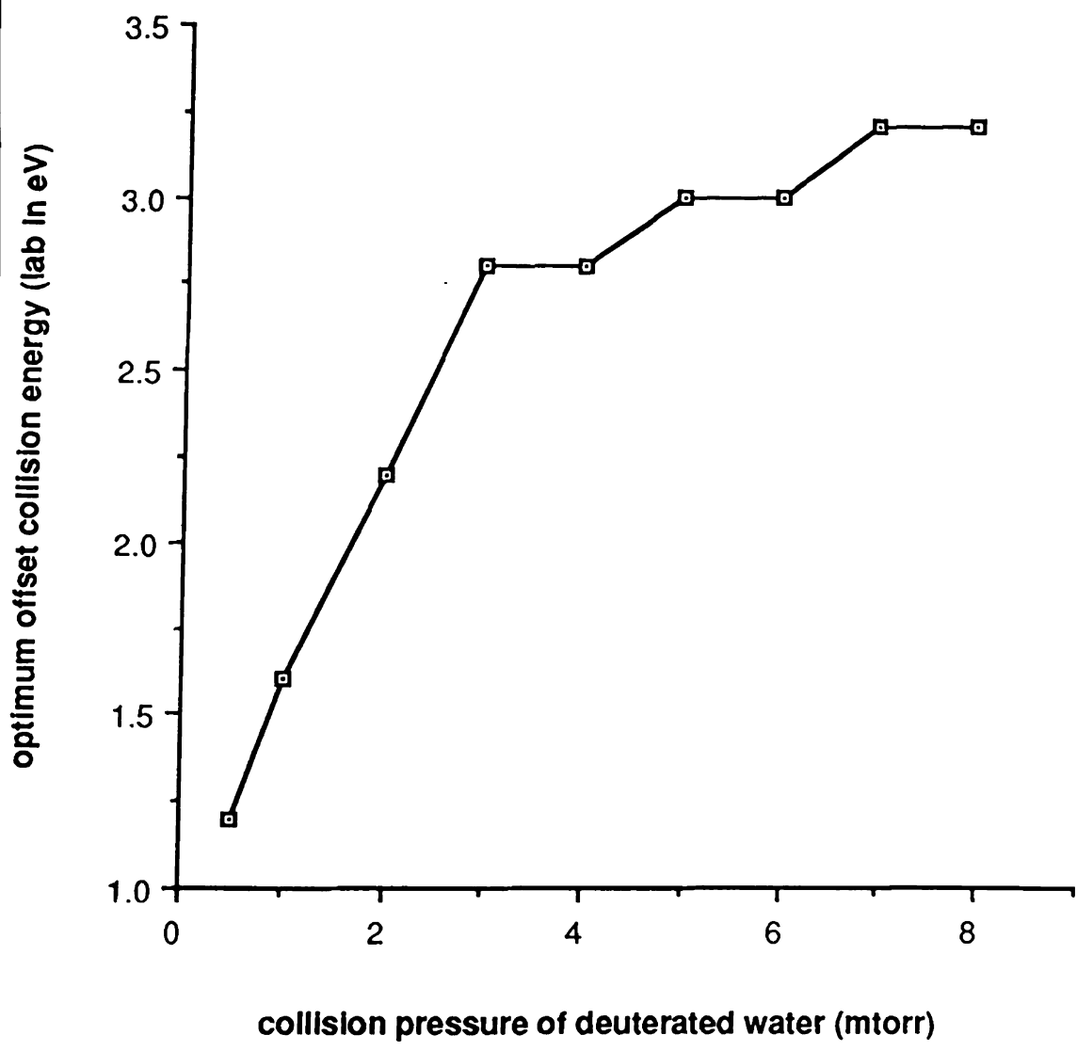
ns of o-dichlorobenzene are exchangeable, but the ion products ting from the exchange of all three hydrogens are detected only within ecific range of collision energy and collision pressure. The values of ion energy and collision pressure were obtained from the offset voltage tial and the pressure gauge reading of the instrument, respectively. llision energies above 5.0 eV or at collision pressure below 0.3 mtorr, D exchange products of any kind were observed. Figure 2-2 shows the um collision energies for the formation of the D_3-substituted product function of collision pressure. At a specific collision pressure, the um collision energy is determined by varying the collision energy the percentage distribution of the D_3-substituted product in the ct spectrum is optimized. The optimum collision energy increases as llision pressure is increased. An increase in collision pressure also s a higher relative yield of the D_3-substituted product up to around 4.0 as shown in Figure 2-3. The relative yield of the D_3-substituted ct reaches a maximum of about 85% when the collision pressure is sed to 4.0 mtorr. Further increases of the collision pressure only e the ion transmission efficiency (see Table 2-1). For example, an se of collision pressure from 4.0 to 5.0 mtorr would reduce the total ansmission by 60%. However, for a collision pressure increase from 4.0 mtorr, the percentage distribution of the D_3-substituted product ses from 65% to 85%, but the total ion transmission is reduced by 6%. The results suggest that for the H/D exchange reactions n the $[M-1]^-$ of o-dichlorobenzene and D_2O, a maximum of about 85% reactant ions passing through Q2 are substituted with three iums. The rest of the reactant ions are either nonreactive or uted with only one or two deuteriums. The optimum collision energy

optimum offset collision energy (lab in eV)

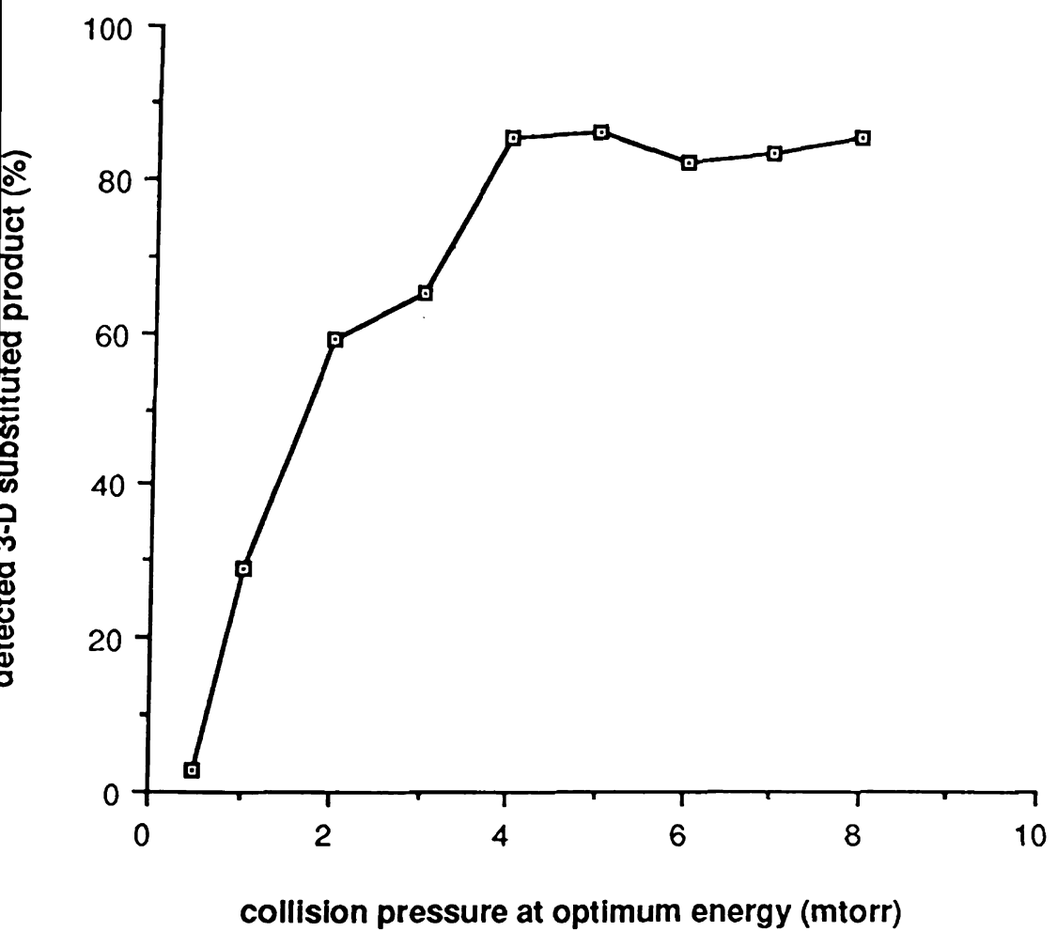
Figure 2.



re 2-2 A plot of optimum collision energy as a function of collision pressure of D_2O. At a specific pressure value, an optimum collision energy is obtained by varying the collision offset voltage of the instrument until the D_3-substituted product is optimized.

detected 3-D substituted product (%)

Figure



gure 2-3 Percent of detected ions that have exchanged with three deuteriums at different collision pressure settings. Offset energy is the optimum for each pressure. The percent detected D_3-substituted product is based on 100% for the total ion intensity of all detected ions.

Table 2-1 *Total ion intensity of all products and ion intensity for the* D_3*-substituted product at different collision pressure settings. The optimum collision offset energy is shown right*

[illegible] of all products and ion intensity for the D_3-substituted product at different collision pressure settings. The optimum collision offset energy is shown right next to each pressure.

collision pressure (mtorr) (optimum collision energy)	total ion intensity of all products	ion intensity for the D_3-substituted product
0.5 (1.2 eV)	47,400,000	1,400,000
1.0 (1.6 eV)	47,500,000	13,800,000
2.0 (2.2 eV)	22,600,000	13,300,000
3.0 (2.8 eV)	18,800,000	12,200,000
4.0 (2.8 eV)	17,600,000	14,900,000
5.0 (3.0 eV)	7,500,000	6,500,000
6.0 (3.0 eV)	4,100,000	3,300,000
7.0 (3.2 eV)	2,200,000	1,900,000
8.0 (3.2 eV)	1,000,000	800,000

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Figure 2-4.

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lision pressure for the exchange reactions are at 2.8 eV and 4.0 respectively.

Exchange Reactions between Deuterated Reagents and the $[M-1]^-$ of chlorobenzene

lthough D_2O has been widely used as a reagent for H/D exchange s, a number of other deuterated reagents have also been found to H/D exchange reactions with both anions and cations of organic nds (2,7). An H/D exchange reaction requires reagents of mately equal relative acidity or proton affinity (2,7). For large ce, the preferred reaction is proton transfer; thus, no single ted reagent is universally effective for all kinds of H/D exchange . For negative ions reacting with a deuterated reagent, H/D e is generally not observed if the difference in acidities is greater kcal (7). In this section, different deuterated reagents are used to th the $[M-1]^-$ of o-dichlorobenzene for H/D exchange reactions. The change reactivities and efficiencies are compared among the ed reagents employed.

e deuterated reagents chosen for this study are D_2O, ND_3, CH_3OD, , $CDCl_3$, CD_3COCD_3, CD_3CN, CD_3SOCD_3, CD_4, C_6D_6 and C_5D_5N e). Among these reagents, $CDCl_3$, CD_3COCD_3, CD_3CN and D_3 were found in this study to produce no H/D exchange products. gents that have produced H/D exchange products are shown in -4. All the spectra were obtained under optimized collision offset s and collision pressure settings except for ND_3 which could not be

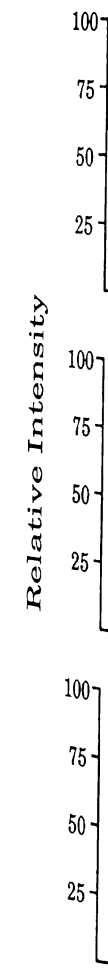
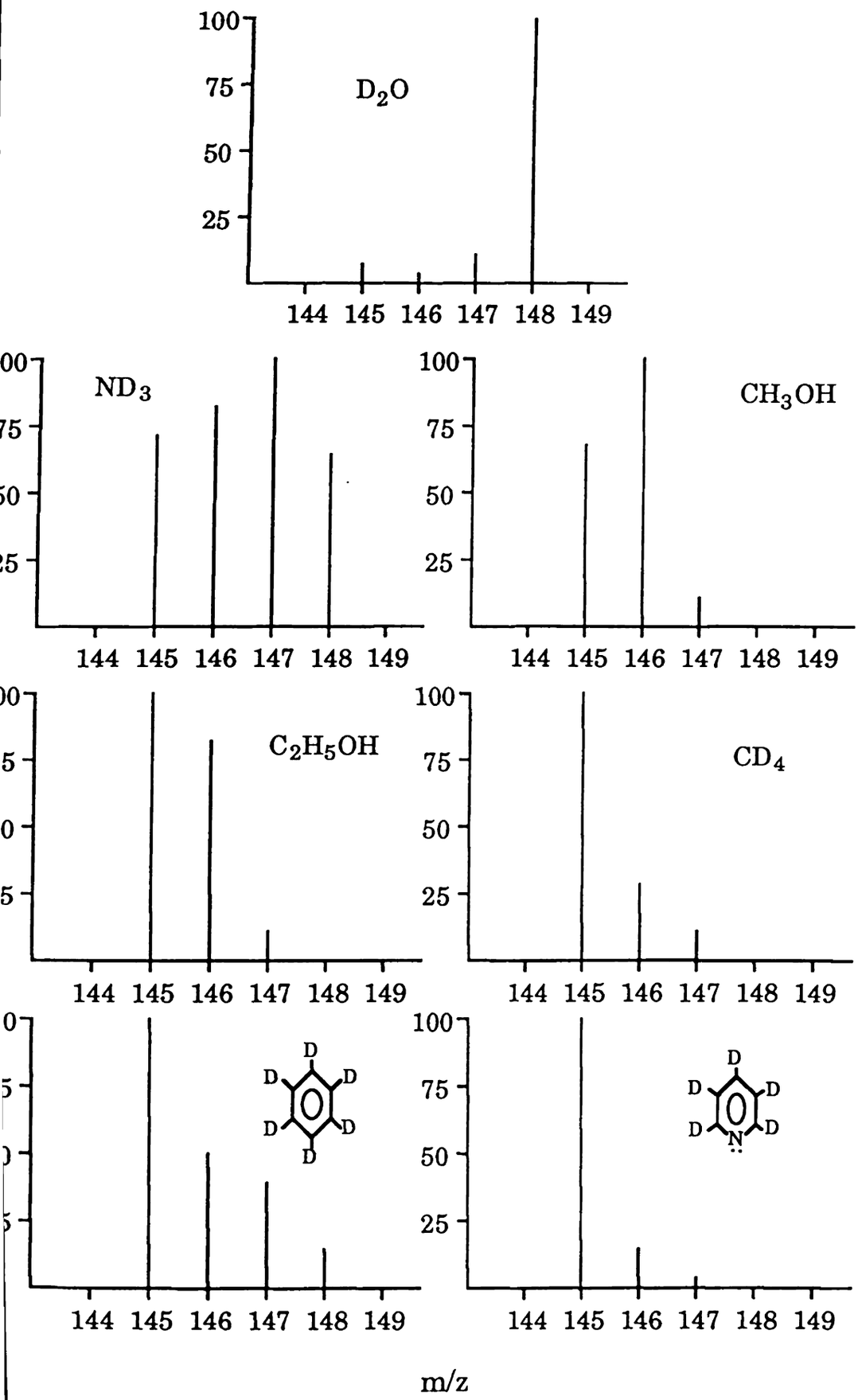


Figure 2-4



4 Product mass spectra obtained from H/D exchange reactions between the $[M-1]^-$ of o-dichlorobenzene (m/z 145) and different deuterated reagents. The reagents that do not produce any observable H/D exchange peaks are not shown in the figure.

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ized because the optimum collision pressure setting for the reagent is ted to be well beyond the highest collision pressure allowable by the ment. The results clearly show that D_2O is the most effective reagent e exchange reactions. Although the optimum collision offset setting the reagents is at about 3.0 V, the optimum collision pressure setting substantially depending on types of reagents used. From optimizing llision pressure settings of the different reagents, it is found that the optimum pressure setting for D_2O is at 4.0 mtorr, the optimum re settings for CH_3OD, C_2H_5OD, CD_4, C_6D_6 and C_5D_5N are at 2.5, 1.5, 5 and 2.0 mtorr, respectively. It is quite possible that these pressure g differences are due to biases in pressure calibrations for the ion from one compound to another. In other words, the real pressures e different readings of the different reagents may actually be closer to nother than indicated. In addition to D_2O and ND_3, C_6D_6 is the only reagent to allow the production of the D_3-substituted product despite newhat bulky structure. Since H/D exchanges between C_6D_6 and the of o-dichlorobenzene involve only aromatic deuteriums and aromatic gens, the chemical similarity between the two exchanging atoms may e the main contributing factor for the unusual reactivity of C_6D_6. The vities of the other reagents are too low to be useful for analytical ations.

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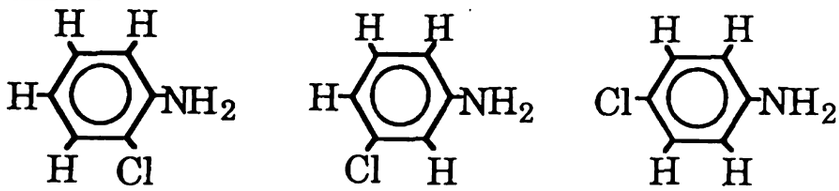
/D Exchange Reaction for the [M-1]$^-$ of Isomers of Substituted hlorobenzenes with D_2O

Deuterated water has been found to be a powerful reagent for anging deuterium for the hydrogen atoms of the [M-1]$^-$ ion from o-orobenzene. Successful applications of the exchange reactions for entiating isomers of chlorinated benzenes will be discussed in detail e next chapter. Because the characteristic chemistry of a specific ogen depends on the chemical environment of its parent molecule, ituting one of the two chlorine atoms with other chemical ionalities may alter the reactivity of its [M-1]$^-$ with D_2O. If the ionalities also contain hydrogen atoms, [M-1]$^-$ may be formed from tonation of one of these hydrogens. In this section, the [M-1]$^-$ ions of obenzene isomers substituted with different chemical functionalities xplored for H/D exchange reactions with D_2O.

The isomers selected for this study are listed in Figure 2-5. They chosen originally for their electron-withdrawing and electron-ing properties towards the benzene rings. Except for NO_2, all the ed substituents also contain one or more hydrogens. Because most of hydrogens are expected to be more acidic than the hydrogens on the atic ring, it is conceivable that for most of the isomers, the [M-1]$^-$ ant ions are formed by direct or indirect deprotonation of the tuent instead of the benzene ring. The results of H/D exchange ons between the [M-1]$^-$ ions and D_2O are shown in Figures 2-6 gh Figure 2-12. The product ions derived from successful H/D nges are represented by peaks having one or more m/z units above the

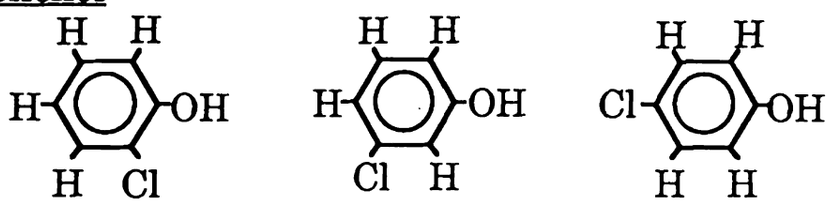
chloroaniline

(I)



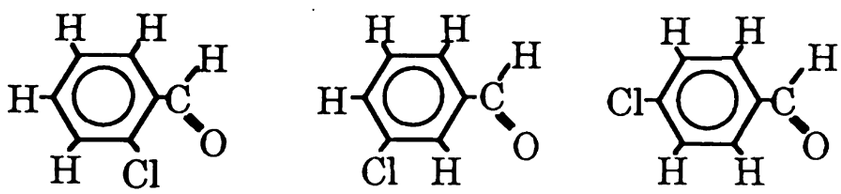
chlorophenol

II)



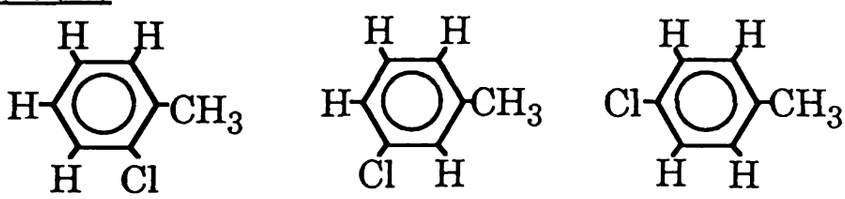
chlorobenzaldehyde

II)



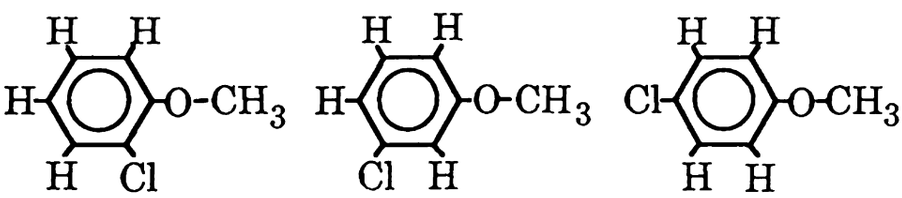
chlorotoluene

V)



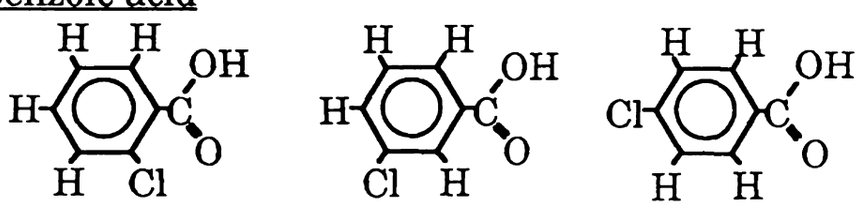
chloroanisole

7)



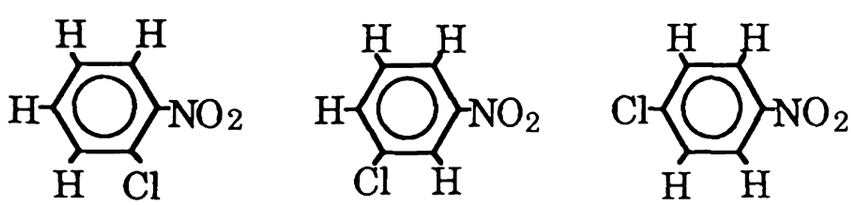
chlorobenzoic acid

I)



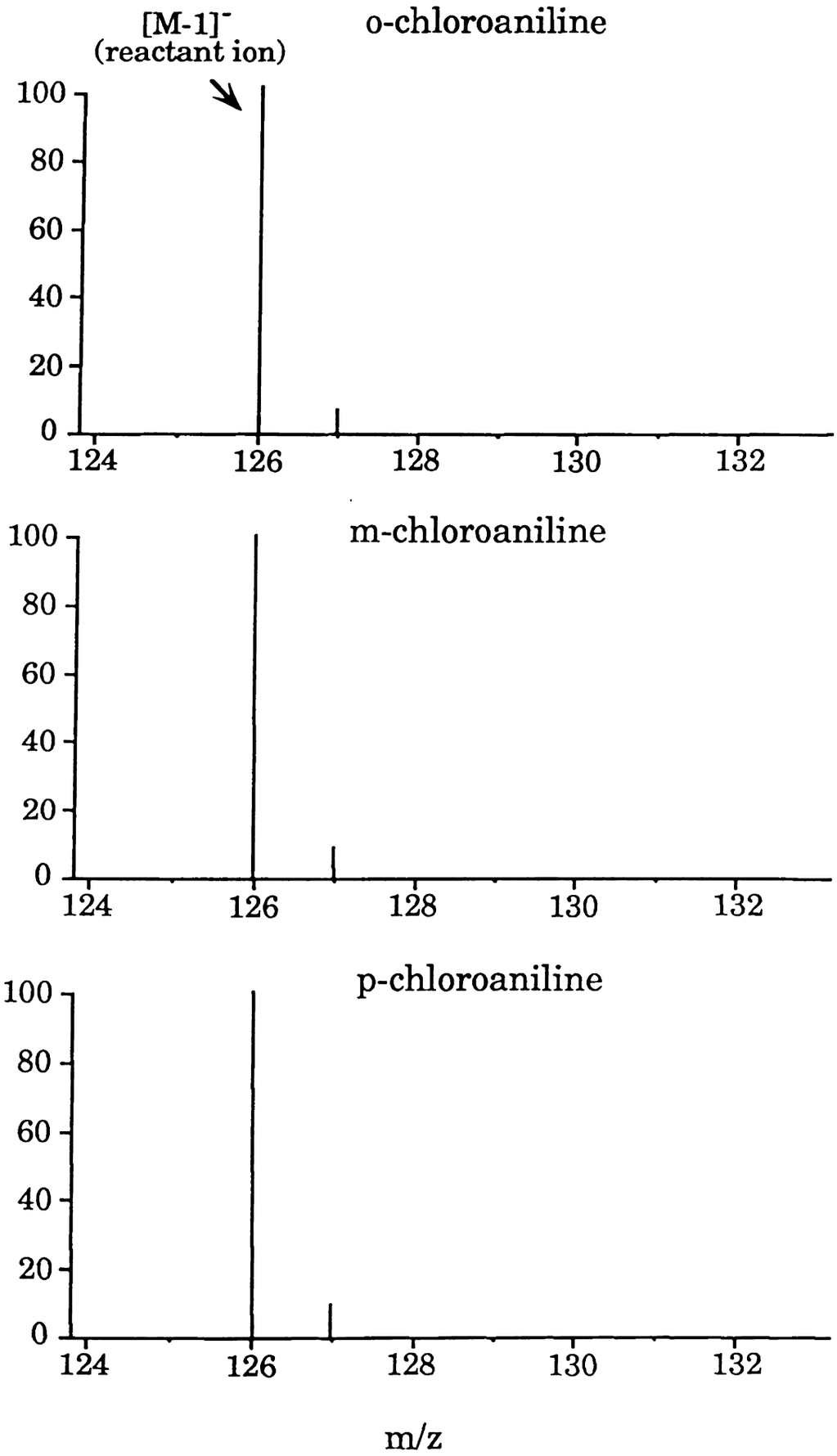
chloronitrobenzene

I)



ure 2-5 Selected isomers for H/D exchange reactions with D_2O

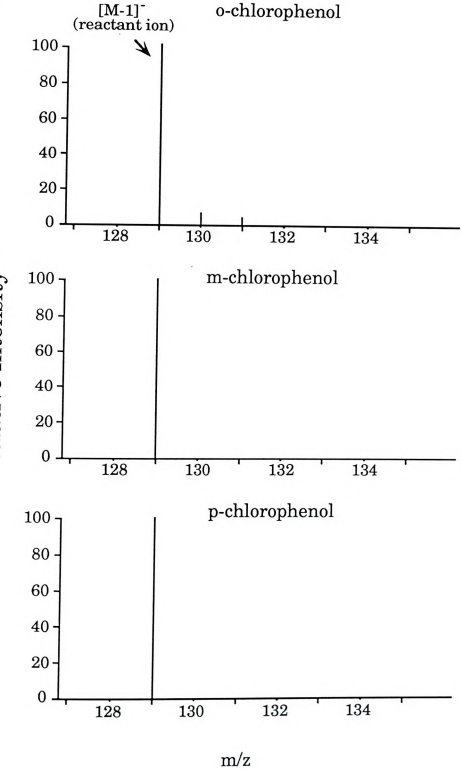
Figure 2-6



2-6 Product mass spectra for the $[M-1]^-$ ion of the isomers of chloroaniline upon H/D exchange reactions with D_2O.

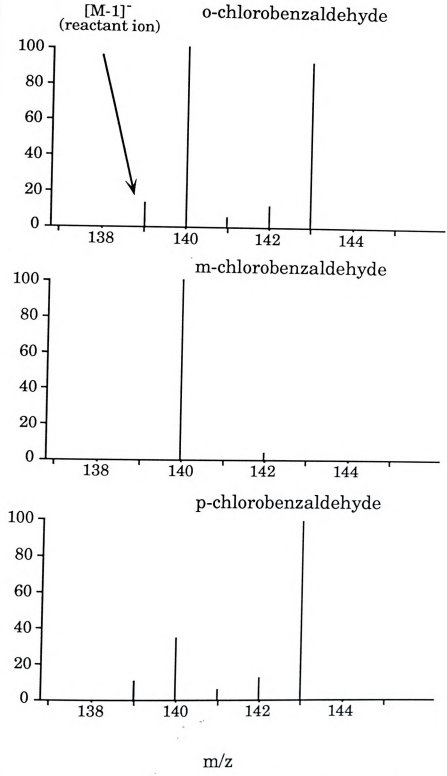
Relative Intensity

Figure



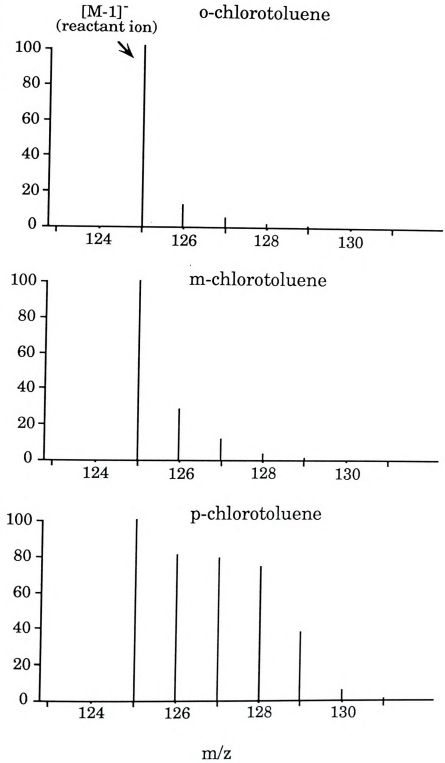
ire 2-7 Product mass spectra for the $[M-1]^-$ ion of the isomers of chlorophenol upon H/D exchange reactions with D_2O.

Figure 2



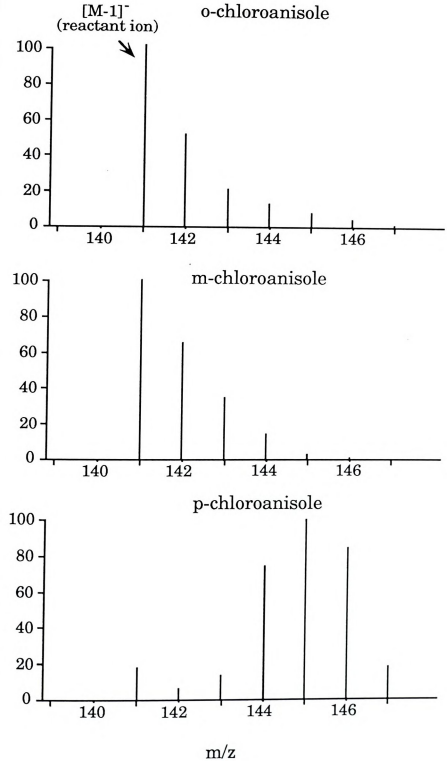
re 2-8 Product mass spectra for the $[M-1]^-$ ion of the isomers of chlorobenzaldehyde upon H/D exchange reactions with D_2O.

Figure 2-9



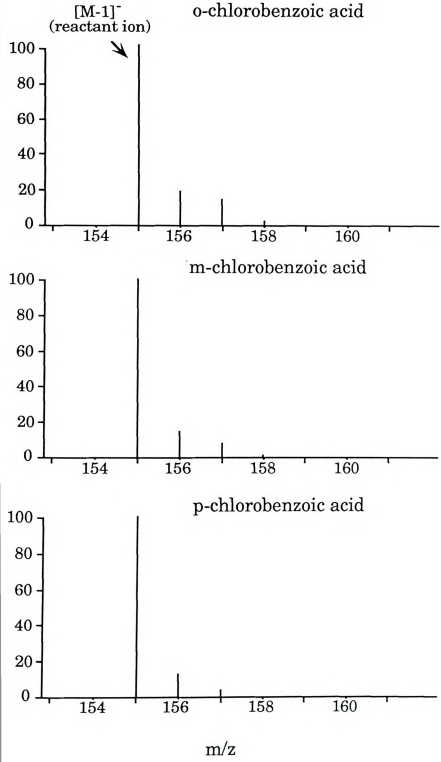
2-9 Product mass spectra for the $[M-1]^-$ ion of the isomers of chlorotoluene upon H/D exchange reactions with D_2O.

Figure 2-



e 2-10 Product mass spectra for the $[M-1]^-$ ion of the isomers of chloroanisole upon H/D exchange reactions with D_2O.

Figure 2-1



2-11 Product mass spectra for the $[M-1]^-$ ion of the isomers of chlorobenzoic acid upon H/D exchange reactions with D_2O.

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Relative Intensity

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Figure 2-

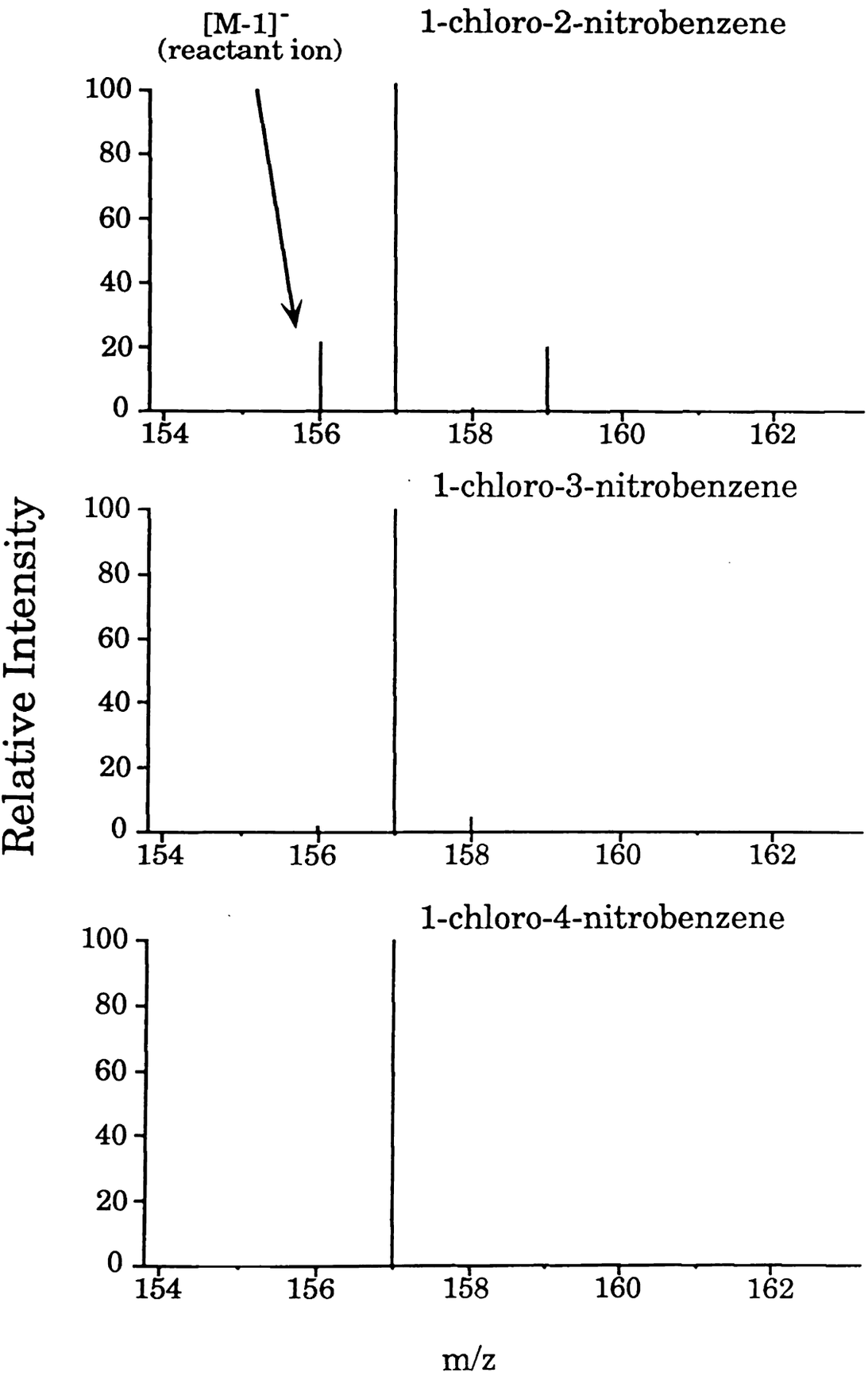

Figure 2-12 Product mass spectra for the $[M-1]^-$ ion of the isomers of chloronitrobenzene upon H/D exchange reactions with D_2O.

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m/z of the reactant. It is clear that substituting one of the two chlorine atoms with the selected substituents substantially reduces the reactivity of the $[M-1]^-$ ion with D_2O. Figure 2-6 shows that the product spectra of all three isomers of chloroaniline are essentially identical. Only one of the five hydrogens of $[M-1]^-$ from the chloroaniline isomers is exchangeable upon reactions with D_2O. Although the results suggest that there may be some interesting chemistry involved, the analytical value of the reactions is minimal.

For the other substituted chlorobenzenes, some spectral differences are observed for the different isomers. In Figure 2-7, the spectra indicate that while no H/D exchange product is observed for either p- or m-chlorophenol, o-chlorophenol gives product spectra indicative of two deuterium substitutions. For isomers of chlorobenzaldehyde, chlorotoluene and chloroanisole, the results presented in Figure 2-8 - Figure 2-10 clearly demonstrate that hydrogens on the substituents are also exchangeable, in addition to the aromatic hydrogens. The reactivities of the para isomers of these three substituted chlorobenzenes are consistently the highest compared to those of the meta and ortho isomers. Although the difference in reactivity for the ortho and meta isomers of chlorotoluene and chloroanisole is small, the reactivity of the meta isomer of chlorobenzaldehyde is very different from that of ortho isomer. While ions substituted with four deuteriums are major products for the anions of o-chlorobenzaldehyde, a small peak representing a maximum of only three deuterium substitutions is observed for the anions of m-chlorobenzaldehyde. Nevertheless, individually these isomers can still be differentiated on the basis of their differences in H/D exchange efficiencies

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with D_2O. For the isomers of chlorobenzoic acid, Figure 2-11 shows that the D_3-substituted products are observed for both the meta and ortho isomers. Product ions associated with a maximum of only two H/D exchanges are observed for the para isomers.

An important difference between chloronitrobenzene and the other substituted chlorobenzenes is the absence of non-aromatic hydrogens in the former. A chloronitrobenzene $[M-1]^-$ ion must be formed by direct or indirect deprotonation of one of the three aromatic hydrogen of a neutral molecule. This means that while the initial reaction site of a chloronitrobenzene $[M-1]^-$ ion is on the aromatic ring, the equivalent site of the $[M-1]^-$ of the other substituted chlorobenzenes may be on the substituents. The product spectra for the isomers of chloronitrobenzene are shown in Figure 2-12. Although ions having only one H/D exchange are the predominant products for all the isomers, the observed maximum H/D exchanges are different for different isomers. Only the ortho isomer is found to allow all three hydrogens of its anion to be exchanged; a maximum of two and one hydrogens exchanged is observed for the meta and para isomers, respectively. Compared to the same reactions with the $[M-1]^-$ of dichlorobenzene, the relative yields for the formation of the two and three H/D exchange products are significantly lower in the case of chloronitrobenzene. Clearly, substituting one of the two chlorines of a dichlorobenzene molecule with NO_2 substantially reduces the reactivity of its $[M-1]^-$ ion towards the formation of the products of higher order H/D exchanges. Nevertheless, the reactions are still useful for differentiating the three isomers.

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Conclusions

In the TSQ-70B triple quadrupole system, the efficiency of ransmitting the $[M-1]^-$ of o-dichlorobenzene from the ion source into the ollision chamber is a strong function of collision offset voltage, but is not ncreased beyond a collision offset potential voltage of around 3.0 V. The ptimum collision offset energy and collision pressure setting for the H/D xchange reactions between the $[M-1]^-$ of o-dichlorobenzene and D_2O are at .8 eV and 4.0 mtorr, respectively. No H/D exchange reaction products are bserved for measured collision pressures below 0.3 mtorr or collision nergies above 5.0 eV. Among the many deuterated reagents selected to eact with the $[M-1]^-$ of o-dichlorobenzene, D_2O is the most effective reagent r H/D exchange.

In the study of H/D exchange reactions between D_2O and the $[M-1]^-$ of lorobenzene isomers substituted with NH_2, OH, CHO, CH_3, OCH_3, COOH nd NO_2 functionalities, it was found that replacing a Cl of a chlorobenzene molecule with one of the functionalities substantially educed the reactivity of the $[M-1]^-$. While the para isomers of orobenzaldehyde, chlorotoluene and chloroanisole allow all hydrogens of e $[M-1]^-$ ions to be substituted with deuteriums, no more than two euterium substitutions are observed for the $[M-1]^-$ ions of the para isomers chloroaniline, chlorophenol, chlorobenzoic acid and chloronitrobenzene. r the $[M-1]^-$ ions of chlorobenzaldehyde, chlorotoluene and chloroanisole, euterium substitutions clearly occur on hydrogens connecting to the nctionalities as well as the aromatic hydrogens. The reactivity of the [M-

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1]$^-$ ions of chlorobenzoic acid, chlorophenol and chloroaniline are too low to produce useful information for isomeric differentiation. It is very likely that some other deuterated reagents may be more suitable than D_2O for exchanging hydrogens of substituted chlorobenzene [M-1]$^-$ ions.

References

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CHAPTER 3

MECHANISTIC STUDY OF H/D EXCHANGE BETWEEN [M-1]$^-$ IONS OF CHLORINATED BENZENES AND DEUTERATED WATER OR DEUTERATED AMMONIA

Introduction

Mass spectrometric studies on hydrogen-deuterium (H/D) exchange reactions in solution have been used for structure elucidation (1,2). Kinetic studies on isotopic methane-hydrogen and methane-deuterium systems have demonstrated the feasibility of H/D exchanges in the gas phase (3,4,5). Presumably, the reactant ions involved for the exchange reactions include H_3^+, D_3^+, CH_3^+, CD_3^+, CH_4^+, CD_4^+, CH_5^+ and CD_5^+. Hunt and co-workers (6,7) first reported H/D exchanges for organic compounds in the gas phase under chemical ionization (CI) conditions with deuterium oxide reagent. Active hydrogens in organic compounds were determined by H/D exchange reactions between neutral organic molecules and ionic deuterated reagents. Isotopic exchange for amines was observed when either ND_3 (7) or MeOD (8) was used as the CI reagent. Differences in the number of exchanges observed between ND_4^+ or $MeOD_2^+$ and amine molecules allowed the differentiation of primary, secondary and tertiary amines. Exchange of aromatic hydrogens was first reported by Beauchamp et al. using ion cyclotron resonance (ICR) spectroscopy for protonated benzene ions (9). This study suggested that ring protonation was a necessary condition for exchanging aromatic hydrogens of a cation. Further studies

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by Martinsen and Buttrill (10) on a variety of substituted benzenes confirmed that when protonation occurred on the substituent groups rather than on the aromatic rings, H/D exchange products were not observed.

Deuterium exchanges for the $[M-1]^-$ ions of a number of organic compounds were first shown by Stewart et al. under flowing afterglow conditions, using D_2O as the reagent (11). Exchange reactions were also observed for similar types of compounds under both CI and flowing afterglow conditions, using other deuterated reagents such as ND_3, MeOD, EtOD and CF_3CD_2OD (12,13). Based on the results obtained from a flowing afterglow instrument, Depuy and Bierbaum (14) have concluded that, as a general rule, H/D exchange can usually be observed between an anion and an exchange reagent which is as much as 20 kcal less acidic. The significance of using H/D exchange reactions for the differentiation of positional isomers of aromatic compounds was then demonstrated by Chakel (15).

In this work, we postulate a reaction mechanism for the H/D exchange reaction between the hydrogen on the $[M-1]^-$ ions of aromatic compounds and molecules of D_2O and ND_3. Isomers of chlorinated benzenes were the model compounds chosen for this study. The results confirm that the reaction is potentially useful for isomeric differentiation.

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Experimental

Two types of tandem quadrupole instruments were used for all experiments. In the cases where ND_3 was used as the deuterated reagent, the experiments were performed on a Finnigan TSQ-70B triple quadrupole mass spectrometer. Experiments involving the use of D_2O as a deuterated reagent were later performed on a Finnigan TSQ-700 tandem quadrupole instrument. The configuration and the components used for both the instruments are essentially identical, with the exception that the second quadrupole in the Finnigan TSQ-70B instrument is an octopole device in the Finnigan model TSQ-700.

For the experiments performed on the Finnigan TSQ-70B instrument, samples were introduced into the ion source by either a direct insertion probe or from an external glass reservoir connected to the ion source. For the samples in the external glass reservoir, the sample flow rates were controlled by two adjustable leak valves installed between the reservoir and the ion source. As far as possible, a steady-state sample flow rate was maintained during each individual experiment. For the experiments performed on the Finnigan TSQ-700 instrument, samples were introduced into the ion source through the Varian 3400 gas chromatograph (GC). A capillary column was used (30 m, 0.25 mm i.d., SE-54 phase with 0.25 μm film thickness) for component separation. The injection port of the GC was set at 250 °C under a splitless injection mode. A transfer line set at 275 °C directed the column efluent into the ion source. Helium was used as the carrier gas at a pressure of 15 psi. The temperature of the GC was programmed from 80 °C to 200 °C at 10 °C/min.

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The GC temperature program was set such that all the selected compounds emerging from the GC column were at least base-line separated. The isomers of o-dichlorobenzene were injected into the GC as mixtures. All the samples were dissolved in hexane to a concentration of about 10 ppm.

The $[M-1]^-$ ions of the test compounds were generated by CI in the ion source at 150 °C using NH_3 as a reagent. In general, the reactant ions extracted from the source were mass-selected by the first quadrupole of one of the tandem quadrupole instruments for reaction in the second quadrupole (Q2) or the octopole device (O2) where the deuterated reagents were introduced. The last quadrupole was set to scan for the spectra of the ion products of the reactions in Q2 or O2. A collision energy at 2.8 eV (according to the Q2 of O2 offset voltage reading of the instrument) was used throughout all the experiments. The pressure of the reagent gas in the ion source was maintained at 1.5 torr. The collision pressures for ND_3 and D_2O were maintained at approximately 7 and 4 mtorr, respectively, except when indicated otherwise. All the experiments were performed twice to test for consistency in the results. The replicates were compared for discrepancies in terms of relative peak ratios and the absence or presence of stray peaks. Duplicate runs consistently gave spectra that were approximately the same. The data shown come from one set of results rather than the average of the replicates.

All chlorinated isomers used in the experiments were purchased from Ultra Scientific, North Kingston, RI. All other halogenated compounds were purchased from Aldrich Chemical Company, Inc., Milwaukee, WI. Deuterated ammonia (99.5 atom %D) was obtained from

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MSD Isotopes, Montreal, Canada. Deuterated water (99+% grade) was purchased from Cambridge Isotopes Laboratory, Woburn, MA. All the chemicals were used without further purification.

Results and Discussion

A. Mechanistic Analysis

Low-energy collision between $[M-1]^-$ ions of all the isomers of dichlorobenzene and deuterated water or deuterated ammonia produce H/D exchange products. For each isomer, the same exchange products are observed for either D_2O or ND_3 reagent, though not in the same relative abundance. The product mass spectra of the three isomers of dichlorobenzene are shown in Figure 3-1 for both D_2O and ND_3. The selected reactant ions at m/z 145 contain only the ^{35}Cl isotope. Each substitution of a deuterium from an exchange reaction with the deuterated reagent is indicated by the gain of one mass unit from the mass of the reactant ions. Sequential H/D exchanges of the same ions are indicated by peaks at more than one m/z unit above the reactant ion mass, thus product ions with all three hydrogens substituted with deuteriums are represented by peaks at m/z 148. Although these isomers are known to produce nearly indistinguishable EI mass spectra, the pattern of H/D exchange products of the $[M-1]^-$ ion of each isomer is quite distinctive. For the anions of these isomers, only o-dichlorobenzene gives a predominant peak at m/z 148 which corresponds to ion products with all the three hydrogens exchanged with deuteriums; the predominant products for m-dichlorobenzene and p-

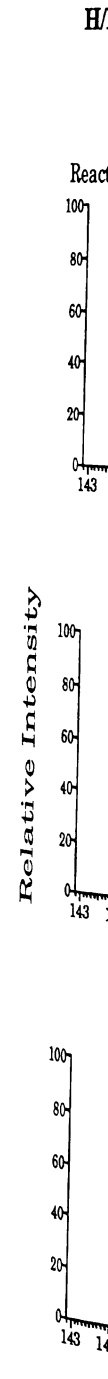
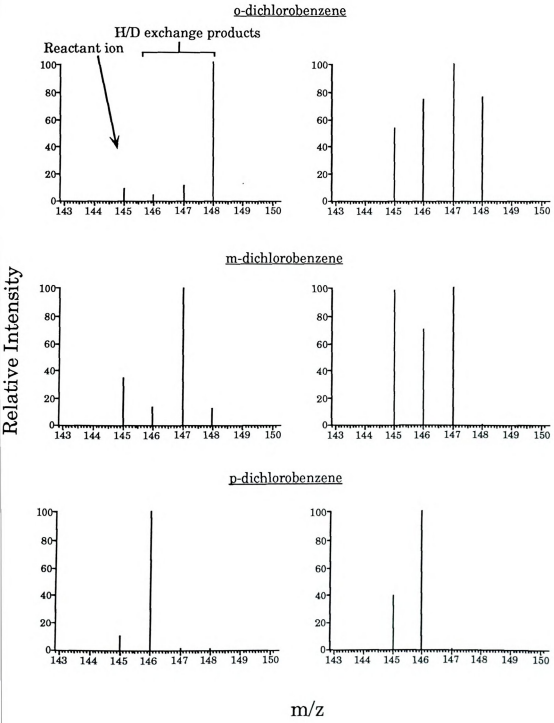


Figure 3-1 Product spectra obtained from the H/D exchange reactions between the $[M-1]^-$ ions of the individual isomers of dichlorobenzene and deuterated water and deuterated ammonia. The reactant ions are at m/z 145. Peaks at higher mass units are due to one or more H/D exchange reactions.

H/D Exchanges with D_2O **H/D Exchanges with ND_3**



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dichlorobenzene are ions substituted with two and one deuteriums, respectively. The dramatic differences in the product patterns observed indicate that the relative positions of the chlorine atoms on the aromatic ring affects the ability of certain ring hydrogens to undergo the H/D exchange reaction.

Since the reaction products observed with both D_2O and ND_3 reagents are essentially the same (though in differing relative abundances), we believe that the basic reaction mechanisms for the two species are essentially the same. We postulate the reaction mechanism shown in Scheme I for ortho dichlorobenzene $[M-1]^-$ ion and D_2O. In this mechanism, a five-membered-ring reaction intermediate is formed between the $[M-1]^-$ ion and the reagent molecule (either D_2O or ND_3). The formation of this intermediate is initiated by the attack of the deuterated molecule on the charge site (localized at the carbon atom with the missing hydrogen). An electron pair on the neutral reagent molecule reacts with a hydrogen atom adjacent to the charge site to form the intermediate complex. A successful H/D exchange occurs only if the decomplexation of the intermediate complex leaves a D atom at the initial charge site and removes the adjacent H atom. The charge site in the product ion is shifted one carbon atom around the ring from its position in the reactant ion. Further H/D exchange reactions can continue by the same mechanism until no more adjacent aromatic hydrogens remain. Products substituted with three deuteriums are the result of three successful H/D exchanges which, according to the proposed mechanism, requires that the initial reactant ion contain three aromatic hydrogens adjacent or sequentially adjacent to the initial charge site.


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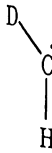
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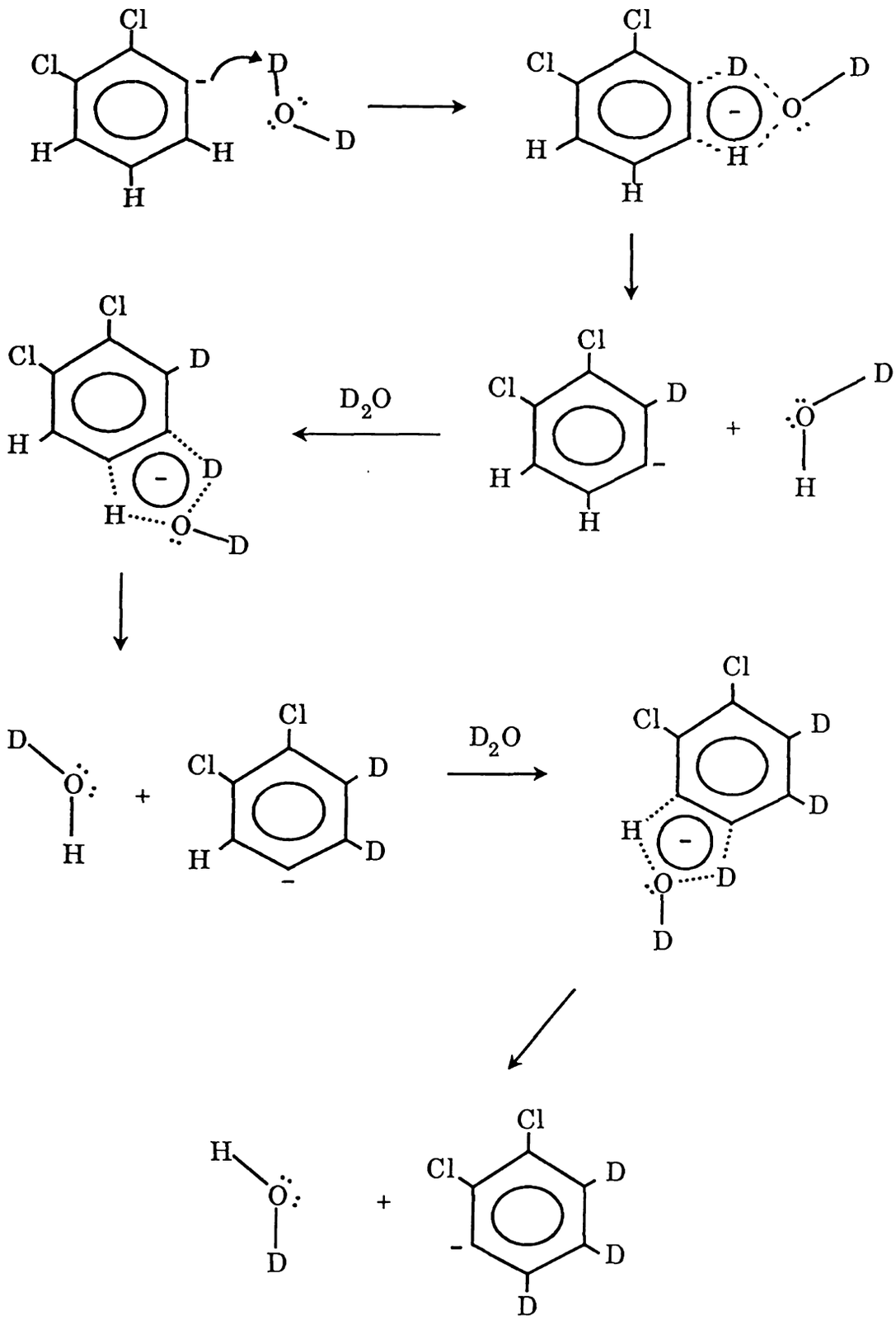
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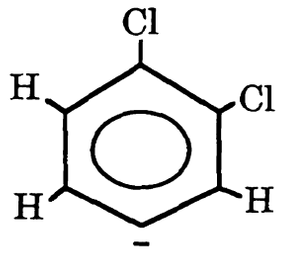
The number of adjacent (or sequentially adjacent) hydrogens in the initial reactant ion depends on the location of the charge and the relative positions of the chlorine atoms on the aromatic ring. All the possible charge sites for the three dichlorobenzene isomers are listed in Figure 3-2. The chlorine positions and the charge locations of the ortho-substituted anions allow sequential deuterium substitution, via the five-membered-ring intermediate, of all three hydrogens. For the meta-substituted anions, three different charge sites are possible. Both structure III and IV contain aromatic hydrogens adjacent to the charge site. These sites allow exchange of two aromatic hydrogens which are adjacent or sequentially adjacent to the charge sites. The third hydrogen located between the two chlorine atoms is separated from the charge sites and can not be exchanged through the five-membered-ring intermediate. Reactant ions substituted with two deuteriums are shown in the product spectra of m-dichlorobenzene as the predominant products. Structure V shows a meta-substituted anion formed with a charge site sandwiched between two chlorine atoms. Since the location of this charge site is remote from the three aromatic hydrogens, deuterium substitution through the five-membered-ring intermediate is not possible for this reactant ion. The relatively larger reactant peak in the product spectrum of m-dichlorobenzene compared to those of the other dichlorobenzene isomers suggests that some of the reactant ions have the structure V configuration. For the para-substituted anions, there is only one possible charge site because all four aromatic hydrogens in the original molecule are equivalent. Structure VI shows that the charge site is adjacent to only one hydrogen and separated from the other two hydrogens by a chlorine atom. By the proposed mechanism, such an ionic configuration allows only one hydrogen of the anion to be

Figure 3-2 The possible charge sites for the $[M-1]^-$ ions of dichlorobenzene isomers and the consequent location and number of exchangeable hydrogens according to the proposed reaction mechanism.

Ortho-substituted anions



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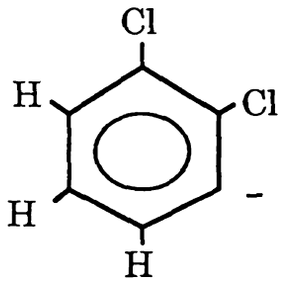

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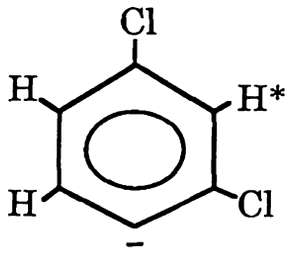
- Both structure I & II allow all three hydrogens to be exchanged

Meta-substituted anions



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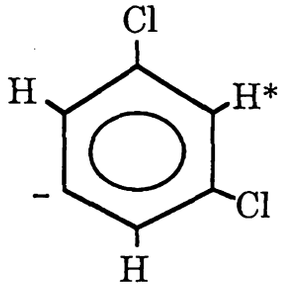

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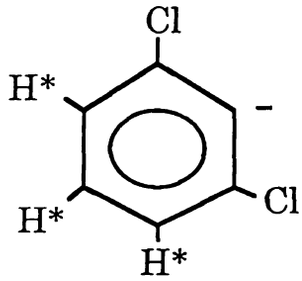

V

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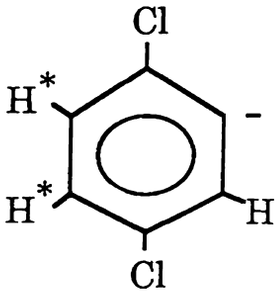
- Structure III and IV will exchange up to 2 H's
- Structure V will not react with D_2O, but may be isomerized to become II inside the ion source.

Para-substituted anion



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- Structure VI allows only one hydrogen to be exchanged

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exchanged. The predominant ions in the product spectrum of p-dichlorobenzene are substituted with only one deuterium.

Overall, the observed results correlate well with the proposed mechanism. The predominant deuterium exchange observed is one less than the maximum number of hydrogen atoms between the chlorine atoms in each dichlorobenzene isomer. However, a peak representing about 12% of a D_3-substituted product is found in the product spectrum of m-dichlorobenzene, in reactions with the D_2O reagent. For the $[M-1]^-$ of m-dichlorobenzene, the proposed mechanism would not allow the formation of any D_3-substituted product. Thus, either the proposed mechanism is wrong, or there is an additional process, specific to the m-dichlorobenzene, which is occurring. Two possible additional processes that could result in the D_3-substituted product are : a different substitution mechanism that can breach the intervening Cl atom located between the charge site and the remaining aromatic hydrogen, and a possible isomerization of structure V of m-dichlorobenzene to structure II of o-dichlorobenzene. Since the likelihood of the success of a Cl-breaching mechanism is expected to be about the same for both the anions of m- and p-dichlorobenzene, the substantially smaller D_3-substituted product peak observed for the p-dichlorobenzene favors the latter explanation. If the isomerization explanation is true, it appears that the degree of isomerization for the anions of p-dichlorobenzene is substantially lower than that of m-dichlorobenzene. Of all the anionic structures, the structure V anions of m-dichlorobenzene may be the most susceptible to isomerization.

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B. Collision Pressure Dependence on Products of H/D Exchanges

The number of collisions of an anion with a deuterated reagent while resident in Q2 is affected by the reagent gas pressure in Q2. A higher gas pressure allows a reactant ion to have a higher collision frequency with the deuterated reagent molecules. For ion products derived from a step-wise sequential process, increasing the total number of collisions can enhance the yield of the more highly substituted ion products. For the $[M-1]^-$ ion of o-dichlorobenzene, the formation of D_3-substituted products requires a minimum of three reactive collisions of the initial reactant ion with deuterated molecules. At low collision pressures, D_3-substituted product ions may not be observed because the reactant ions may leave Q2 before the required number of ion-molecule collisions can occur. For H/D exchanges with D_2O, the collision pressure study shown in Figure 3-3 supports the sequential process depicted in our proposed mechanism. Products with the highest degree of deuterium substitution are not observed until products with lower degrees of deuterium substitution are formed. The D_1-substituted ions are the predominant products at very low collision pressures. As the collision pressure increases, higher substituted ions replace the less substituted ions as predominant products. At a sufficiently high collision pressure, most the anions of o-dichlorobenzene are fully substituted before they leave Q2. These data support a mechanism that involves the substitution of only one deuterium per reactive collision.

For the reactant ions from m-dichlorobenzene, the pressure-dependence plot shown in Figure 3-3 indicates that following an increase in collision pressure, the relative intensity of the reactant ions decreases; but

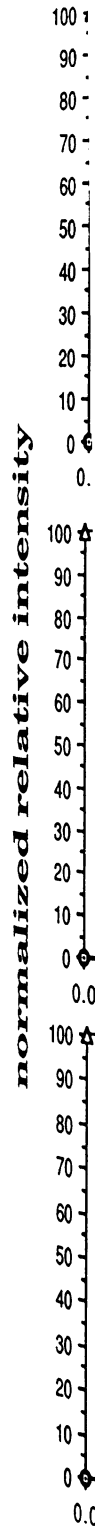
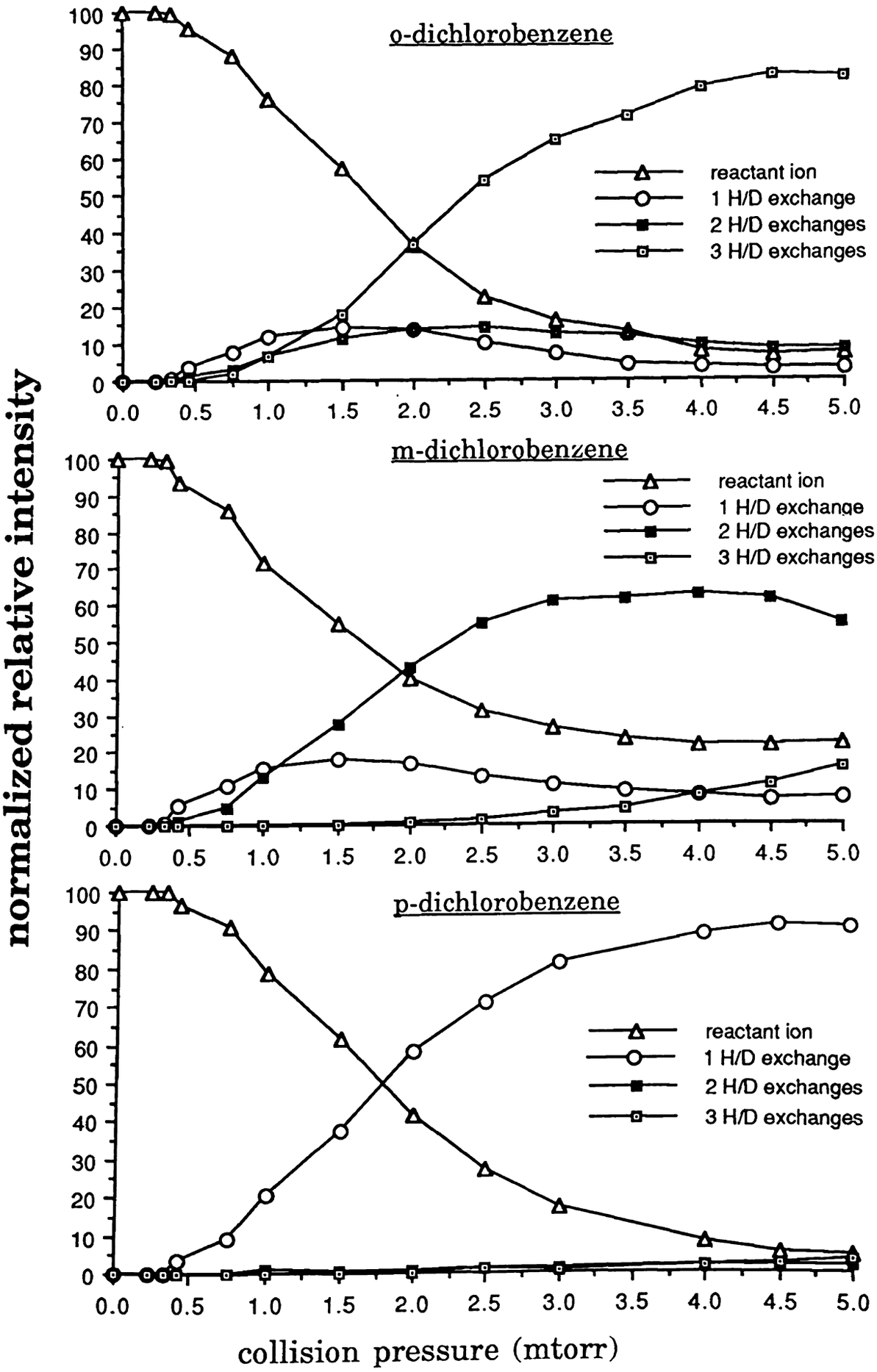


Figure 3-3 Plots for normalized relative product intensity vs collision pressure for H/D exchange reactions between the $[M-1]^-$ ions of dichlorobenzene isomers and D_2O molecules.

Exchanges with deuterated water



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the decrease of the reactant ions levels off beginning at about 3.5 mtorr. Thereafter, the contribution of the reactant ions remains at about 25% of the total ion intensity, despite further increases in collision pressure. This result suggests that in the case of m-dichlorobenzene, about 25% of all reactant ions are unreactive with D_2O molecules under any collision pressure conditions. This lack of reactivity can be accounted for within our proposed mechanism, if about 25% of the reactant ions have structure V, in which the charge site is not adjacent to any aromatic hydrogens. Since the contribution of the D_3-substituted products in the m-dichlorobenzene plot is about 20% at the collision pressure of 5.0 mtorr, we believe that at least 20% of all meta-substituted reactant ions undergo isomerization to reactant anions identical to those from o-dichlorobenzene. This isomerization most likely occurs in the source, possibly as part of the ionization process, though a slow isomerization of the ion, continuing through the ion lifetime in Q2 is also possible.

The pressure-dependence plot for p-dichlorobenzene (Figure 3-3) ndicates that an increase in collision pressure enhances primarily the ormation of the D_1-substituted product. Even at a high collision pressure, he D_1-substituted product remains predominant. This means that ions ubstituted with only one deuterium are generally the terminal products for eactions between D_2O and the p-dichlorobenzene $[M-1]^-$ ions. The elatively small contribution of the D_2- and D_3-substituted products in the p-ichlorobenzene plot suggests that the p-dichlorobenzene anions are less kely to undergo isomerization than are the anions of m-dichlorobenzene. t the collision pressure of 4.5 mtorr, over 90% of the p-dichlorobenzene

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ND_3 than f

the H/D ex

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exchange m

3-3 & 3-4 b

reactant ion

proposed me

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product. A

anions are converted into the expected predominant D_1-substituted products.

Collision pressure studies were also performed on reactions with ND_3 reagent. The pressure-dependence plots for H/D exchange reactions involving ND_3 reagent are shown in Figure 3-4. The trends for the formation of different H/D exchange products for each isomer are the same for both D_2O and ND_3 reagents. The major difference between the two reagents are their relative reactivities towards the anions of the dichlorobenzene isomers for H/D exchange reactions. To achieve a specific H/D exchange product yield, the required collision pressure is higher for ND_3 than for D_2O. A comparison between Figures 3-3 and 3-4 suggests that the H/D exchange efficiency of D_2O molecules is about three times better than that of ND_3 molecules. The similarity in the trends of H/D exchange product formation for the two deuterated reagents supports the notion that similar exchange mechanism are followed by both reagents.

Although H/D exchange products are observed upon collision between dichlorobenzene anions and D_2O or ND_3 reagent, reaction probably does not occur on every single collision. Based on our proposed H/D exchange mechanism, we have generated plots similar to those of Figures 3-3 & 3-4 by computer simulation of H/D exchange reactions initiated by reactant ions of all the structures shown in Figure 3-2 and following our proposed mechanism. In these simulations, the number of collisions times the percent of collisions that are reactive are used instead of the collision pressure as the parameter to calculate the relative intensities for each ion product. A simulated collision-dependence plot for o-dichlorobenzene

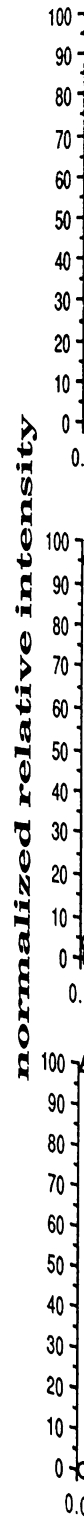

Figure 3-4 Plots for normalized relative product intensity vs collision pressure for H/D exchange reactions between the $[M-1]^-$ ions of dichlorobenzene isomers and ND_3 molecules.

Exchanges with deuterated ammonia

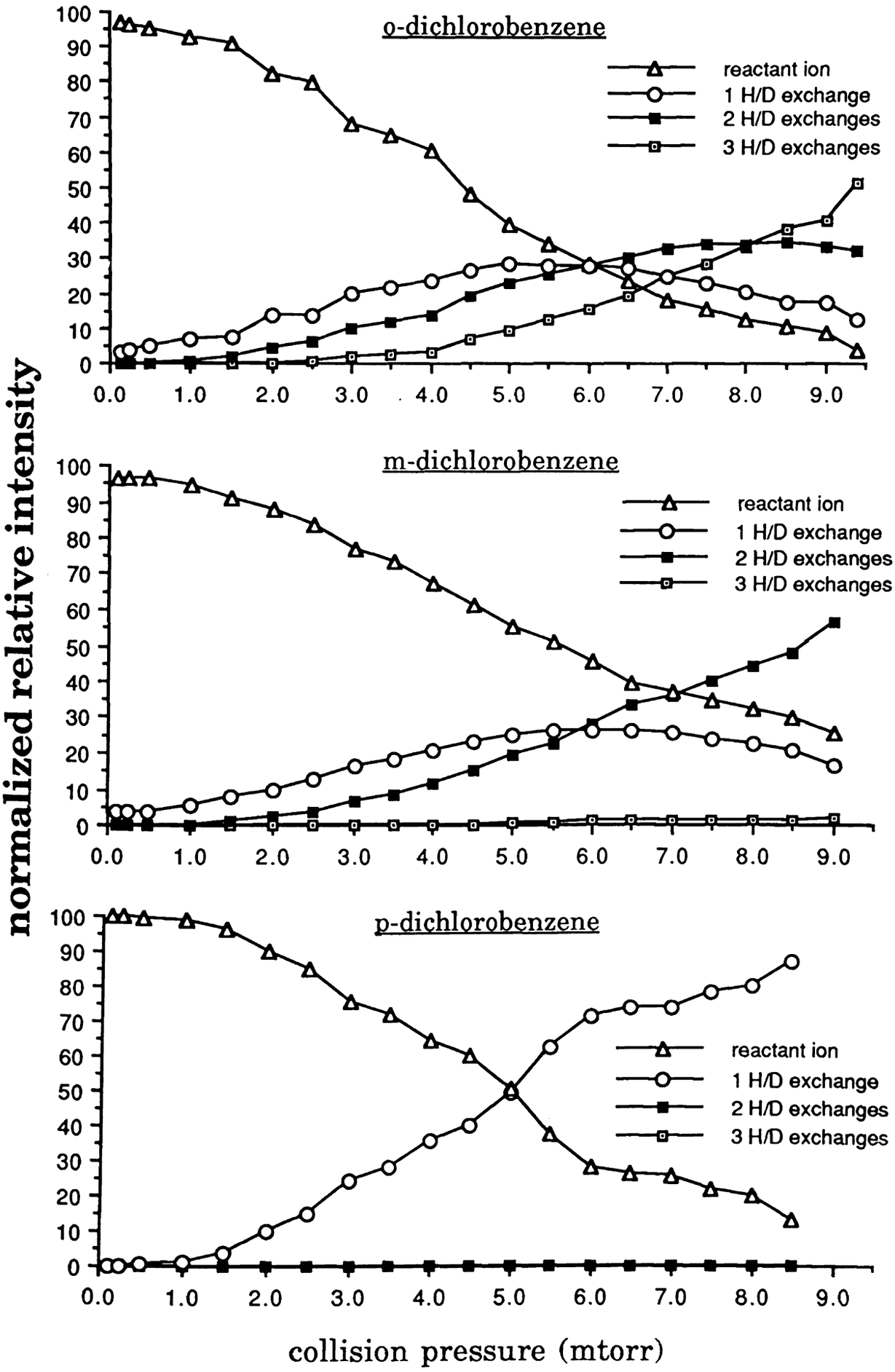

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Figure 3.
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reaction
number c
"average"

Fig
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reactive
pathway

anions having the structure II configuration is shown in Figure 3-5. The reaction pathway used for the simulation is shown in Figure 3-6 for the structure II reactant ions. In this study, a reactive collision is defined as either a successful H/D exchange or D/D exchange. In our calculations, we assume that there is an equal chance for an ion to undergo either a H/D exchange or a D/D exchange where both are possible. For example, for ion 1 shown in Figure 3-6, the chance of its conversion into either ion 1' or ion 2 is the same. The collision-dependence plot of Figure 3-5 shows the relative abundance of the reactant and various product ion masses as a function of the number of reactive collisions. In making this plot, it was assumed that no new reactant was added, nor products removed over the time shown. It is also assumed that the percent of collisions that are reactive is the same for all ion structures. A comparison of the collision dependence plot of Figure 3-5 with the experimental pressure-dependence plot of Figure 3-3 (top) reveals that the zero input and output flux assumption made in the product-ratio calculations is not valid under our experimental conditions. Continued influx of reactant ions extends its high abundance into the equivalent of higher collision numbers and reduces the relative abundance of the deuterium-exchanged product ions. Departure of ions from the reaction chamber before they have actually undergone the "average" number of collisions yields finite values for their abundance seen at high "average" collision numbers.

Figure 3-7 shows a simulated collision-dependence plot for structure I anions of o-dichlorobenzene. These reactant ions require at least four reactive collisions to form the D_3-substituted products. The reaction pathway used for calculating the relative product intensity of the structure

Figure 3-5 A simulated collision-dependence plot that represents a composite of individual collision-product plots for different percentages of reactive collision. The simulation is based on the proposed H/D exchange reaction mechanism between the structure II of o-dichlorobenzene and D_2O or ND_3 molecules.

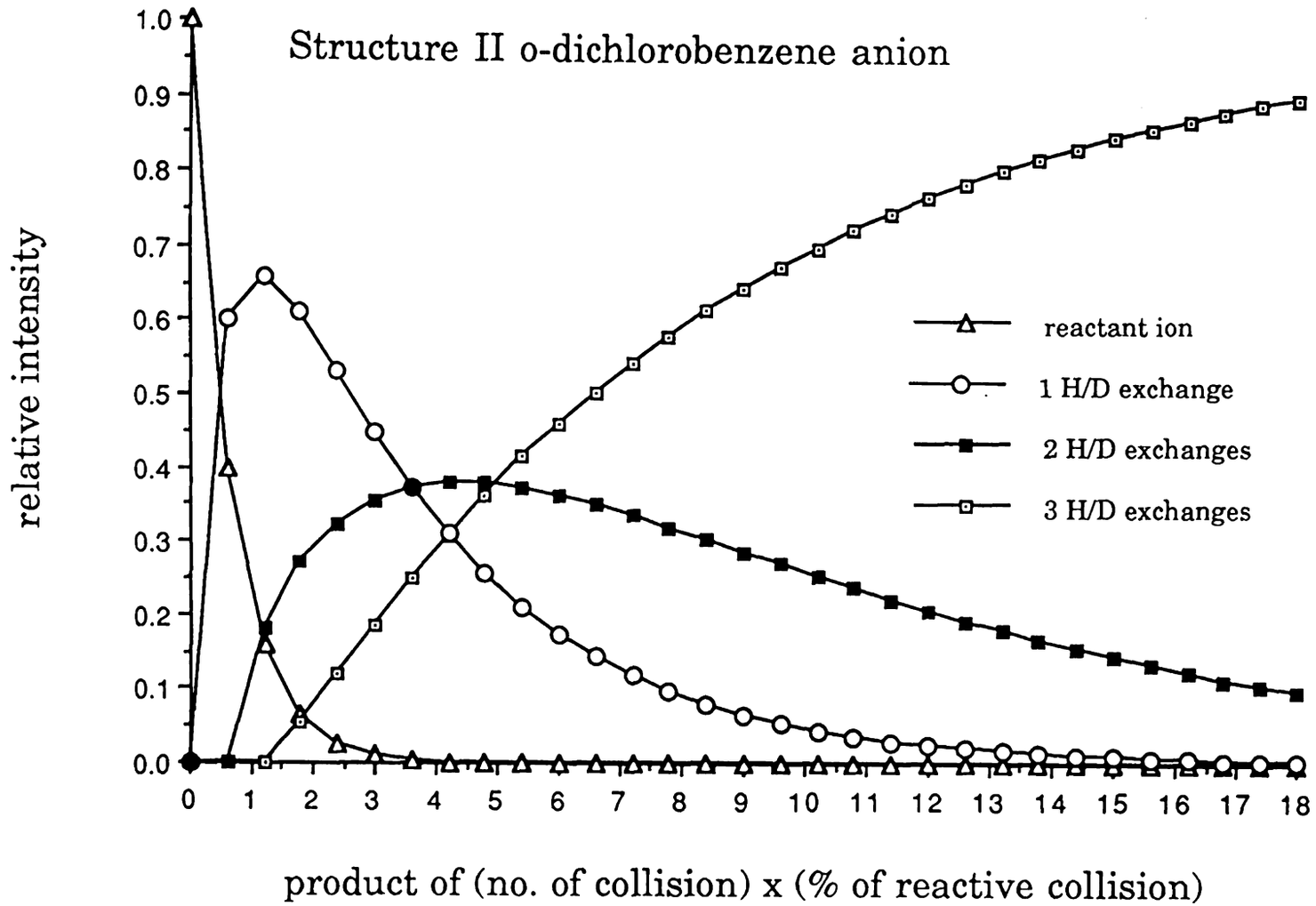

Structure II o-dichlorobenzene anion
relative intensity
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18
reactant ion
1 H/D exchange
2 H/D exchanges
3 H/D exchanges
product of (no. of collision) x (% of reactive collision)

Figure 3-6 The reaction pathway for the structure II anions of o-dichlorobenzene upon reactive collisions with D_2O or ND_3 molecules.

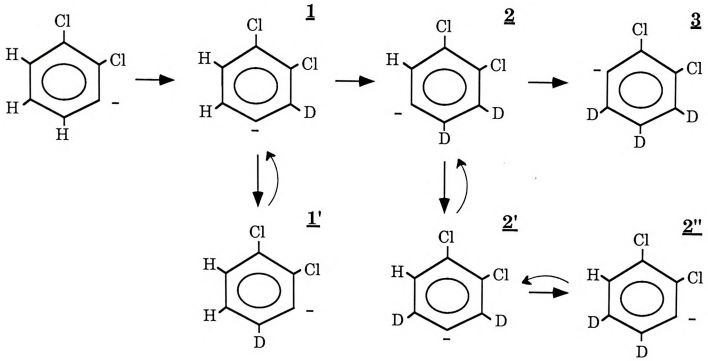
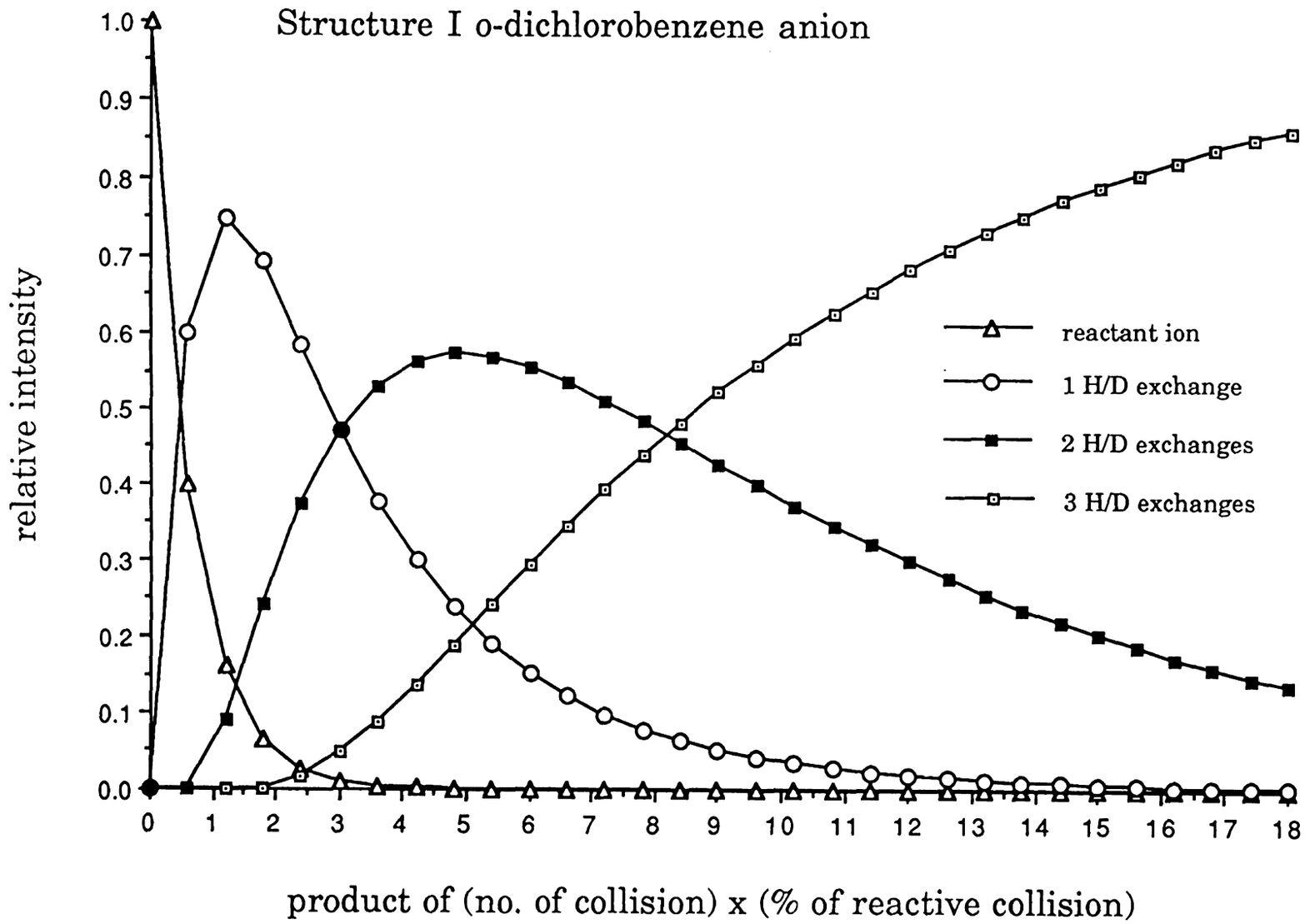

1
2
3
1'
2'
2''

Figure 3-7 A simulated collision-dependence plot that represents a composite of individual collision-product plots for different percentages of reactive collision. The simulation is based on the proposed H/D exchange reaction mechanism between the structure I anions of o-dichlorobenzene and D_2O or ND_3 molecules.


Structure I o-dichlorobenzene anion
relative intensity
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18
reactant ion
1 H/D exchange
2 H/D exchanges
3 H/D exchanges
product of (no. of collision) x (% of reactive collision)

I anion
structu
differen
the coll
and II
pressu
betwee
differen
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F
the th
contrib
V. Th
structu
match
ions wi
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I anions is shown in Figure 3-8. Comparisons between plots obtained from structure I reactant ions and those with structure II show marked differences in the relative abundances of product ions at many points along the collision number axis. Ideally, one could choose between structures I and II based on the comparison of the collision-dependence plots with the pressure-dependence plot. Unfortunately, the differences in conditions between the experimental and simulated data are greater than the differences between the structure I and II simulations thus precluding a clear preference for either one.

For the anions of m-dichlorobenzene, our simulations assume that of the three meta-substituted ionic structures shown in Figure 3-2, the contribution of structure III is insignificant compared to structures IV and V. The reaction pathways for the formation of various products for both structure III and structure VI anions are shown in Figure 3-9. In order to match the experimental results, we further assume that the number of ions with structures IV and V are initially formed in the ion source in a 50/50 ratio. Before the anions enter the collision chamber for H/D exchange reactions, 50% of the initially formed structure V anions are also assumed to isomerize to become structure II anions. As such, the collisional simulations of the anions of m-dichlorobenzene are based on collisions initiated by a mixture of anions with three different structures (50% structure IV, 25% structure V and 25% structure II). The results are shown in Figure 3-10 as a collision-dependence plot. The product formation trends represented in this plot compare well with the similar trends shown in the experimental pressure-dependence plot of Figure 3-3 (middle). This further supports the isomerization explanation and the existence of the

Figure 3-8 The reaction pathway for the structure I anions of o-dichlorobenzene upon reactive collision with D_2O or ND_3 molecules.

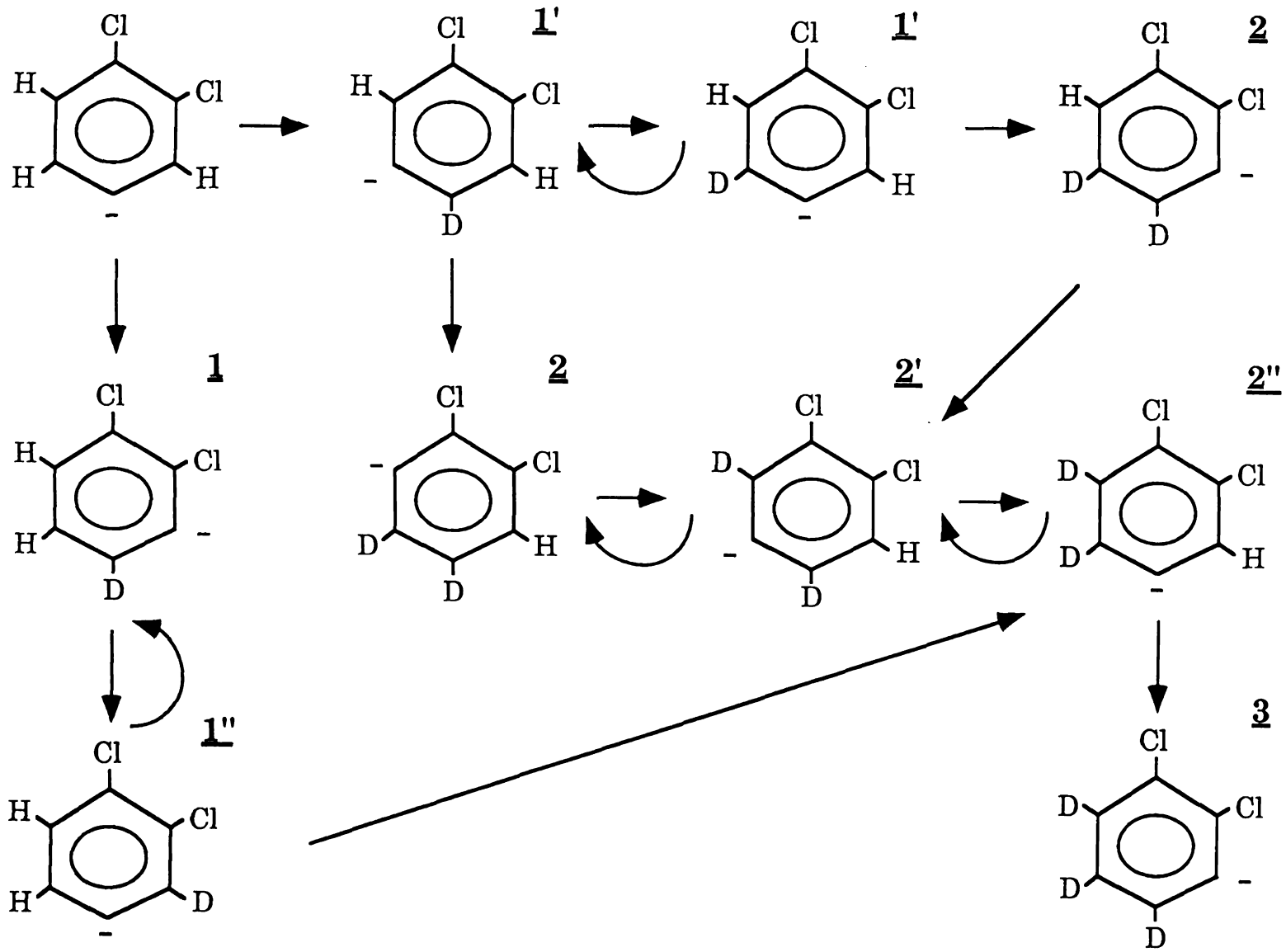
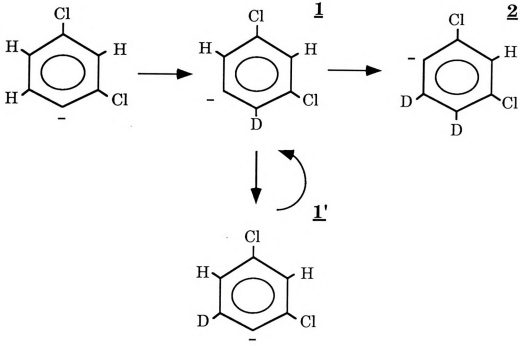

1
1'
1''
2
2'
2''
3

Figure 3-9 The reaction pathway for the structure III (top) and structure IV (bottom) anions of o-dichlorobenzene upon reactive collisions with D_2O or ND_3 molecules.


1
2
1'

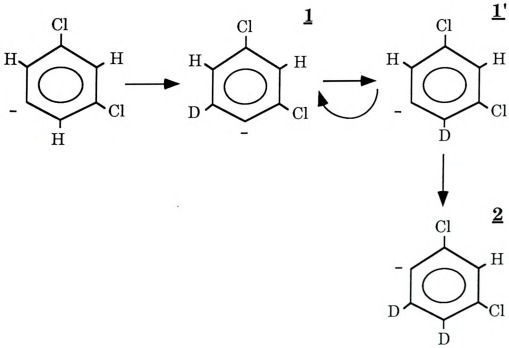
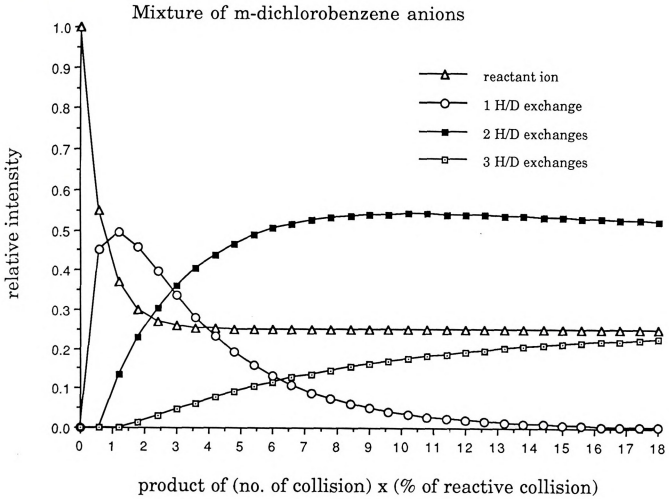

1
1'
2

Figure 3-10 A simulated collision-dependence plot that represents a composite of individual collision-product plots for different percentages of reactive collision. The simulation is based on the proposed H/D exchange reaction mechanism between a mixture of different m-dichlorobenzene anions and D_2O or ND_3 molecules. The mixture of anions is assumed to contain 50% structure IV, 25% structure V and 25% structure II.


Mixture of m-dichlorobenzene anions
relative intensity
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18
product of (no. of collision) x (% of reactive collision)
reactant ion
1 H/D exchange
2 H/D exchanges
3 H/D exchanges

unreacti

individu

dichlorob

Th

relatively

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collision-

reactant

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unreactive structure for the m-dichlorobenzene anions. (Simulated individual collision-product plots for the different ionic structures of dichlorobenzenes are shown in the Appendix.)

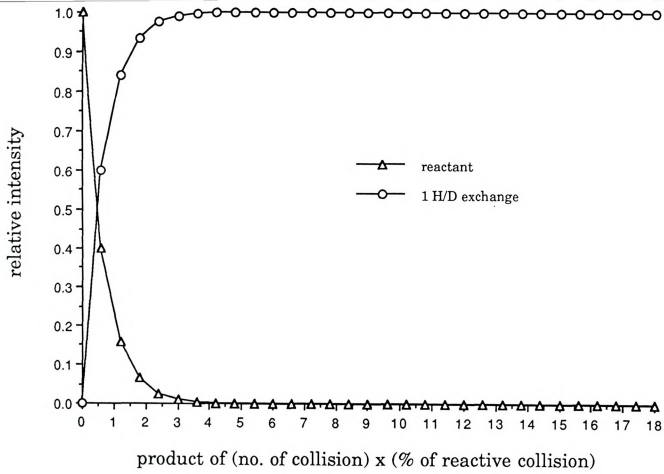
The collision simulations of the anions of p-dichlorobenzene are relatively simple because of the simplicity of the reaction pathway for the formation of the expected predominant products. Figure 3-11 shows the collision-dependence plot for the anions of p-dichlorobenzene, based on reactant ions with structure VI configuration. In this case, the trends for the formation of the predicted product shown in the simulated plot are comparable to those represented in the experimental pressure-dependence plot of Figure 3-3 (bottom). Overall, although the relative product abundances represented in all the simulated collision-dependence plots are different from those shown in the experimental pressure-dependence plots, the general trends for the formation of various H/D exchange products obtained from the simulated results are consistent with the experimental data .

C. Reactions with Other Aromatic Isomers

We have also used isomers of other chlorinated benzenes to test the proposed mechanism. According to the mechanism, in order for a $[M-1]^-$ anion to undergo an H/D exchange, there must be at least one aromatic hydrogen adjacent to the charge site on the anion. For the $[M-1]^-$ ions derived from 1,3,5-trichlorobenzene, 1,2,4,5-tetrachlorobenzene and 1,2,3,5-tetrachlorobenzene, none of the possible charge sites have any adjacent

Figure 3-11 A simulated collision-dependence plot that represents a composite of individual collision-product plots for different percentages of reactive collision. The simulation is based on the proposed H/D exchange reaction mechanism between p-dichlorobenzene anions (structure VI) and D_2O or ND_3 molecules.

Structure VI p-dichlorobenzene anion


relative intensity
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18
reactant
1 H/D exchange
product of (no. of collision) x (% of reactive collision)

arom
reac
1,2,
tetr
pred
to th
shou
neut
chlo
1,2,
the
with
mec
1,2,
desc
with
mos
diff
reac
be c
dem
ND
exce
of t
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aromatic hydrogens and no products resulting from the H/D exchange reactions are observed. The product spectra derived from the reaction of 1,2,3-trichlorobenzene, 1,2,4-trichlorobenzene and 1,2,3,4-tetrachlorobenzene anions with D_2O, shown in Figure 3-12, exhibit predominant exchanges of 2, 1 and 1 deuteriums, respectively. According to the proposed mechanism, reactant ions substituted with two deuteriums should be the predominant products for 1,2,3-trichlorobenzene because its neutral configuration contains three sequential hydrogens between the chlorine atoms. The original molecules of both 1,2,4-trichlorobenzene and 1,2,3,4-tetrachlorobenzene contain only two sequential hydrogens between the chlorine atoms, the predominant products should be ions substituted with only one deuterium. All the results are consistent with the proposed mechanism. The small D_2-substituted peak in the product spectrum of 1,2,4-trichlorobenzene could be due to isomerization of the reactant ions as described earlier. Similar results are also observed for the same reactions with ND_3 reagent. The dramatic differences in the product patterns for most of the isomers studied suggest that these reactions are useful for differentiating isomers of chlorinated benzenes. The results of the reactions are summarized in Table 3-1. Since the number of chlorines can be determined from the masses of different $[M-1]^-$ ions, our studies clearly demonstrate that tandem MS along with H/D exchange reactions using ND_3 or D_2O as a reagent can positively identify all the chlorinated benzenes except two of the tetrachlorinated isomers. Product spectra similar to those of the dichlorobenzene isomers are also obtained for the isomers of difluorobenzene and bromochlorobenzene. This strongly suggests that the same mechanism is operating on all halogenated benzenes.

Figure 3-12 Product spectra for the tetra and trichlorobenzene isomers that produce H/D exchange ion products. The H/D exchange products are due to reactions between the $[M-1]^-$ ions of the isomers indicated and D_2O molecules. No H/D exchange products are observed for the other tetra and trichlorobenzene isomers.

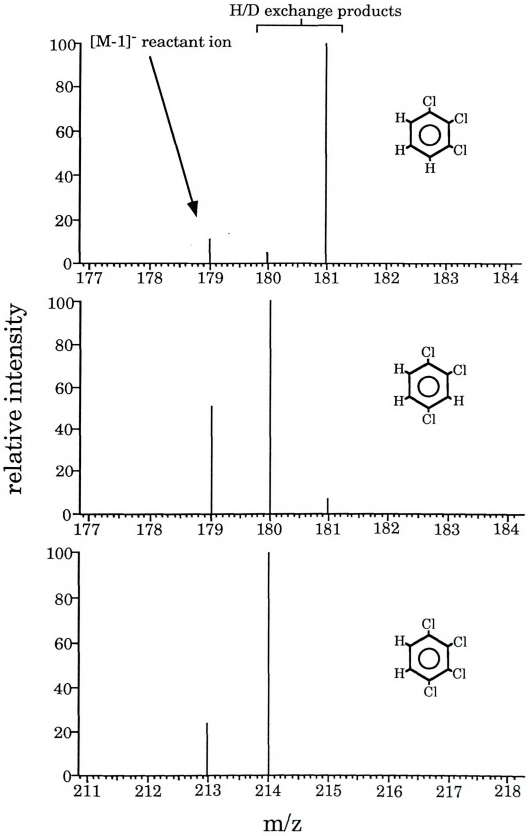

H/D exchange products
[M-1]⁻ reactant ion
relative intensity
m/z
100
80
60
40
20
0
177
178
179
180
181
182
183
184
211
212
213
214
215
216
217
218
Cl
H

Table 3-1. Maximum number of H/D exchanges for the $[M-1]^-$ ions of isomers of chlorinated benzenes (in cases of no isomerization).

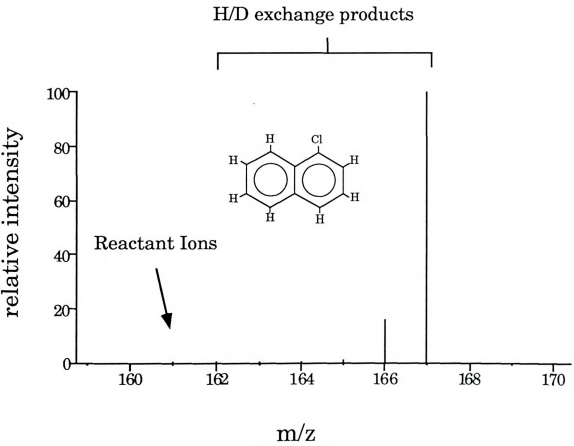
type of Cl substitution	position of Cl on an aromatic ring	max. # of D substitution
mono	(no isomers)	
di	1,2	3
	1,3	2
	1,4	1
tri	1,2,3	2
	1,2,4	1
	1,3,5	0
tetra	1,2,3,4	1
	1,2,3,5	0
	1,2,4,5	0
penta	(no isomers)	

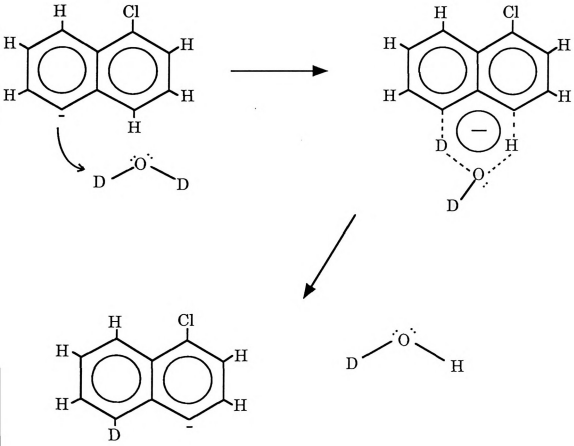
We have begun studies on larger chlorinated aromatic compounds. For a molecule made up of fused benzene rings, the initial negative charge site of an $[M-1]^-$ ion may be on a different ring than the exchanging aromatic hydrogens. For example, sequential H/D exchanges for aromatic hydrogens on different rings of a chlorinated naphthalene molecule require the formation of a six-membered-ring intermediate for relocation of the charge site from one ring to another assuming a similar mechanism is also operating. The product spectrum of 1-chloronaphthalene shown in Figure 3-13 indicates that reactant ions with all five hydrogens substituted with deuteriums are the predominant products . For the $[M-1]^-$ anion of 1-chloronaphthalene, substituting more than three hydrogens requires a mechanism of relocating the charge site from one ring to the other. In this case, the mechanism should involve the formation of a six-membered-ring intermediate, as shown in Scheme II. It is quite possible that the same mechanism may also apply to chlorinated compounds with more than two adjacent rings.

Conclusions

Using ND_3 or D_2O as a reagent, H/D exchange reactions are found to occur between the reagents and the $[M-1]^-$ ions of halogenated benzenes originating from molecules with two or more sequential aromatic hydrogens. The reactivity of D_2O is about three times that of ND_3 in the exchange reactions. A mechanism involving the formation of a five-membered-ring intermediate is consistent with the results derived from the

Figure 3-13 A product spectrum for H/D exchange reactions between the $[M-1]^-$ ions of 1-chloronaphthalene and D_2O molecules. Reactant ions having five hydrogens substituted with deuteriums are the predominant products.


H/D exchange products
relative intensity
100
80
60
40
20
0
Reactant Ions
H
Cl
H
H
H
H
H
H
160
162
164
166
168
170
m/z



Scheme II

isomers of all the chlorinated benzenes in this study. The results obtained from the isomers of florinated and brominated benzenes strongly suggest that the same mechanism is also applicable to all halogenated benzenes. The number of deuterium substitutions for the predominant H/D exchange products is one less than the number of sequential aromatic hydrogens in the original molecules of chlorobenzene. It is possible that a mechanism involving a six-membered-ring intermediate is used for exchanging hydrogens across two or more fused benzene rings.

Computer simulations based on the proposed mechanism are also consistent with the product formation trends derived from the experimental results. The anions of m-dichlorobenzene are more susceptible to isomerization than the anions of o- and p-dichlorobenzene. A comparison among the experimental pressure-dependence plots suggests the existence of an unreactive structure for the anions of m-dichlorobenzene which is predicted by our proposed mechanism. For most chlorinated benzenes, we have demonstrated the potential of using H/D exchange reactions for isomeric differentiation.

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CHAPTER 4

EXPLORATION OF ION-MOLECULE REACTIONS IN TANDEM MASS SPECTROMETRY FOR DISTINGUISHING ISOMERS OF POLYCHLORODIBENZO-P-DIOXINS

Introduction

Polychlorodibenzo-p-dioxins (PCDDs) are well known for the great concern over their occurrence in the environment. With the exception of the octachloro-congeners, all the very toxic PCDD congeners have chlorine substitution at the 2, 3, 7, and 8 positions (1). In particular, the extreme toxicity of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) has attracted much attention. For many years, GC/MS has been a standard method for analyzing TCDDs in environmental samples. However, isomers of TCDD often give identical or similar spectra when traditional mass spectrometric techniques are used. Although the use of oxygen negative chemical ionization (NCI) has been shown to provide distinct mass spectra for 1,2,3,4-TCDD and 2,3,7,8-TCDD (2), there is no experimental evidence that the technique is capable of differentiating among 2:2 substituted TCDD isomers (3).

Chemical interferences are the major concerns in analyzing environmental samples for 2,3,7,8-TCDD. Prior to the mass spectrometric analysis of a sample, extensive chemical cleanup and high resolution gas chromatography (HRGC) are necessary in order to reduce interferences to

a reasonable level. Even so, high resolution mass spectrometry (HRMS) with resolving power of over 10,000 is needed to resolve TCDDs from many common chemical interferences (4,5). However, for modern double-focusing mass spectrometers, achievement of the requisite resolving power results in a substantial loss of instrument sensitivity. Tandem mass spectrometry (MS/MS) was first described by Chess and Gross (6) as a rapid-screening method for analyzing TCDD. The loss of COCL by the metastable decomposition of the molecular ion of TCDD was monitored for the analysis. Despite the fact that this technique offers speed and reduced interferences from PCBs, its overall specificity and sensitivity are significantly worse than GC/HRMS. Since then, a number of workers have explored the use of MS/MS coupled with GC or HRGC for analyzing TCDD (7-20). In most cases, the increased specificity of MS/MS was found to greatly reduce chemical interferences, which were seen in HRMS when real samples with complex matrices were analyzed (5,9,10,14,15,17,19,20).

Since the MS/MS techniques mentioned above monitor only the loss of COCL by collisionally induced dissociation (CID) of $[M]^+$, they are not isomer-specific for 2,3,7,8-TCDD. From analyzing fly ash samples, Buser and Rapper (21) found a complex isomeric mixture of at least 17 TCDDs. It was estimated that 2,3,7,8-TCDD was present at only 1% of the total TCDD amount in the samples. Although isomer-specific chromatographic methods have been developed for analyzing 2,3,7,8-TCDD (22-24), lengthy separations are still required. Kostianinen and Auriola (25,26) have recently employed the reaction between the $[M]^-$ of TCDDs and O_2 in the collision cell of a triple quadrupole system to distinguish 1,2,3,4-, 1,2,3,6/1,2,3,7,- and 2,3,7,8-TCDD isomers.

In this work, we have further explored the use of reactions between the $[M-1]^-$ ionsof PCDDs and different neutral molecules for distinguishing the more toxic PCDDs from the less toxic ones.

Experimental

All the experiments were performed on the Finnigan TSQ-70B triple quadrupole mass spectrometer (TQMS). Samples were introduced into the Varian 3400 gas chromatograph (GC) which is directly interfaced with the TQMS. A capillary column was used (30 m, 0.25 mm i.d., SE-54 phase with 0.25 μm film thickness) to ensure the high purity of the PCDD samples entering the TQMS. The injection port of the GC was set at 250 °C under a splitless injection mode. The temperature was programmed from 150 °C to 300 °C at 15 °C/min. A transfer line directs the column into the ion source of the TQMS and was set at 275 °C. Helium was used as the carrier gas at a pressure of 15 psi. Each injection contained 1-2 μL of sample. The ion source temperature was held at 150 °C, the electron ionization at 70 eV, and the electron current at 200 μA. During ammonia chemical ionization (CI), a pressure of 1.5 torr was maintained throughout the experiments. During the MS/MS operations, ions extracted from the source were selected by the first quadrupole (Q1) of the TQMS to enter the second quadrupole (Q2) where neutral molecules were introduced for ion-molecule reactions. Product ions emerging from Q3 were then mass scanned to obtain the product mass spectra. A somewhat better than unit resolution was tuned for Q1 in order to ensure the purity of the selected parent ions, whereas Q3 was maintained at unit resolution. A collision energy of 3 eV (laboratory

energy as determined by the Q2 offset voltage) was used for all the experiments involving ion-molecule reactions. All the experiments were performed twice to test for consistency in the results. The replicates were compared for discrepancies in terms of relative peak ratios and the absence or presence of stray peaks. Duplicate runs consistently gave spectra that were approximately the same. The data shown come from one set of results rather than the average of the replicates.

All the PCDD congeners were purchased from AccuStandard, New Haven, CT, except for 1,3,6,8-TCDD and congeners with six chlorines or more, which were purchased from UltraScientific, North Kingstown, RI, and Cambridge Isotope Laboratories (CIL), Woburn, MA, respectively. All the congeners were obtained in toluene solution. All alcohols were obtained from Aldrich Chemical Company Inc., Milwaukee, WI. Deuterated water (99.5+% grade) was also purchased from CIL. Deuterated ammonia (99.5 atom %D) was obtained from MSD Isotopes, Montreal, Canada. All the chemicals were used without further purification.

Results and Discussion

A. Generation of Reactant Ions

A mixture of CH_4/N_2O was used by Oehme and Kirschmer as a reagent to obtain the negative chemical ionization (NCI) mass spectra of TCDD isomers (27). Most of the isomers were found to form predominantly the $[M-1]^-$ ion due to direct or indirect deprotonation by OH^-. In our

experiments, NH_3 was used as the NCI reagent and is found to produce a higher $[M-1]^-$ yield than the previous study with CH_4/N_2O. The abundance of the $[M-1]^-$ relative to $[M]^-$ is related to the acidity of the $[M]^-$ ion which, in turn is affected by the positions of the chlorines. The NCI spectra obtained from the five TCDD isomers (Figure 4-1) in this study indicate that 2,3,7,8-TCDD has the highest acidity and therefore the highest relative abundance of $[M-1]^-$. The acidity of 1,2,3,4-TCDD is shown to be the lowest among the isomers by having only a very small peak at m/z 319. This is primarily due to the lack of electron-withdrawing Cl adjacent to H in 1,2,3,4-TCDD. For the rest of the TCDD isomers, the acidity is directly related to the number of hydrogens at the 1, 4, 6 and 9 positions. The lower acidity of 1,3,6,8-TCDD compared to 1,2,7,8-TCDD and 1,3,7,8-TCDD is the result of having only two hydrogens at the 1, 4, 6 and 9 positions instead of three. The trends observed in our study (summarized in Table 4-1) matches well with the results obtained by Oehme et al. (27) using CH_4/N_2O as reagents. A similar trend is also observed for the lower chlorinated PCDD congeners. However, congeners containing five chlorines or more do not produce any $[M-1]^-$ ions under our CI conditions because of their relatively low acidity. For these higher chlorinated congeners, the predominant NCI product is $[M]^-$ (see examples in Figure 4-2), which is mainly due to the capture of a thermal electron inside the CI source.

B. <u>Reactions with D_2O</u>

Hydrogen/deuterium (H/D) exchange reactions between the $[M-1]^-$ ions of chlorinated benzenes and D_2O were found to be distinctive for the three dichlorinated isomers as well as the others (28,29). Upon low energy

Figure 4-1 Mass spectra of TCDD isomers derived from negative chemical ionization (NCI) using ammonia as a reagent.

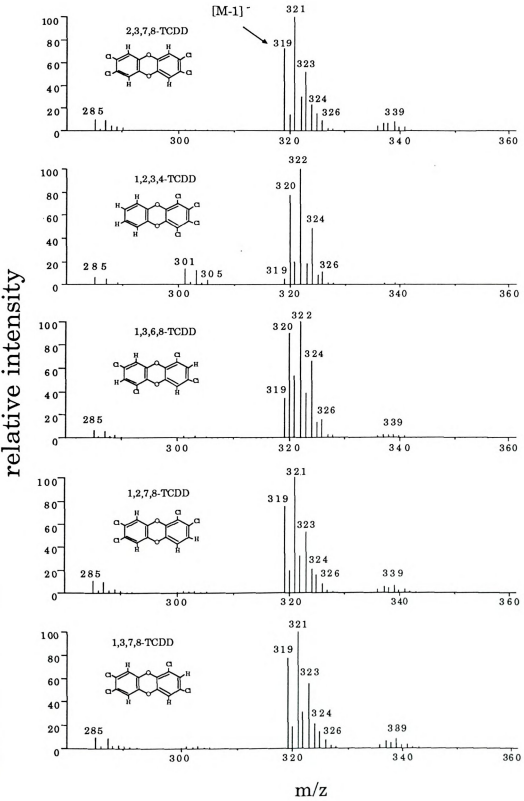
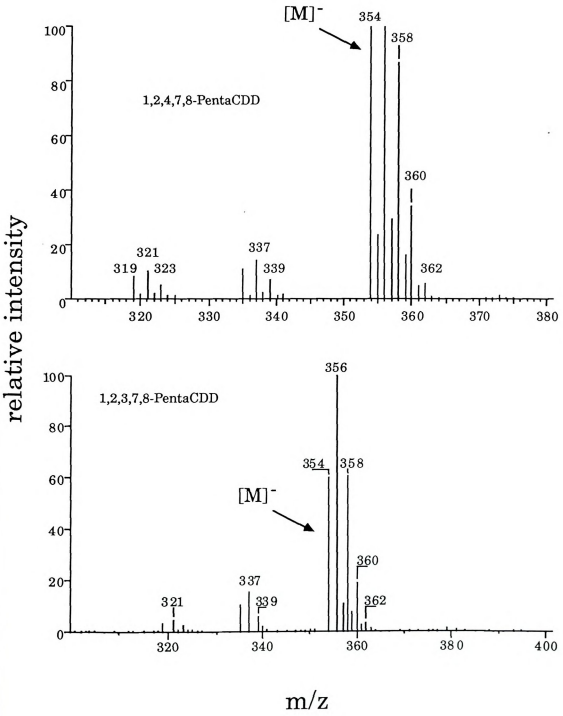

[M-1]⁻
2,3,7,8-TCDD
1,2,3,4-TCDD
1,3,6,8-TCDD
1,2,7,8-TCDD
1,3,7,8-TCDD
relative intensity
m/z
100
80
60
40
20
0
300
320
340
360
285
301
305
319
320
321
322
323
324
326
339

Table 4-1 Ion intensity ratios for m/z 319 and m/z 320 derived from the ammonia NCI mass spectra of the selected TCDD isomers.

TCDD isomers	319 / 320	# of Hs at the 1,4,6,9 positions
2378	100 / 20	4
1278	100 / 27	3
1378	100 / 25	3
1368	100 / 210	2
1234	100 / 1700	2

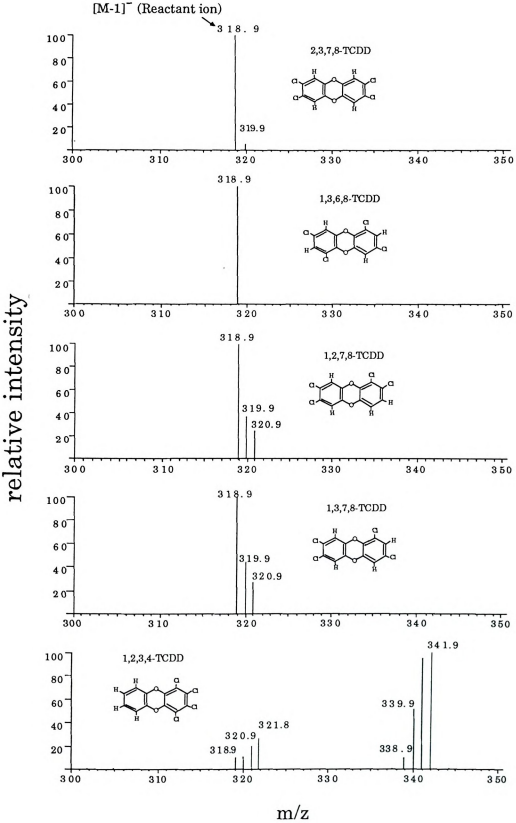
Figure 4-2 Mass spectra of 1,2,4,7,8- and 1,2,3,7,8-pentachlorodibenzo-p-dioxin derived from negative chemical ionization (NCI) using ammonia as a reagent.


[M]⁻
354
358
1,2,4,7,8-PentaCDD
360
321
319
323
337
339
362
100
80
60
40
20
0
320
330
340
350
360
370
380
relative intensity
356
1,2,3,7,8-PentaCDD
354
358
[M]⁻
360
337
321
339
362
100
80
60
40
20
0
320
340
360
380
400
m/z

collisions with D_2O the $[M-1]^-$ ions of most PCDD congeners undergo H/D exchange reactions. The product mass spectra for the five TCDD isomers selected for this study are shown in Figure 4-3. The exact mass for TCDD was used to select the $[M-1]^-$ reactant ions by Q1. The $[M-1]^-$ ions selected contain only the ^{35}Cl and ^{12}C isotopes that corresponds to m/z 318.9 in the mass spectra. Product ions with deuterium substitutions are shown by peaks with one or more m/z units above those for the reactant ions. Only product ions indicative of one deuterium exchange are observed for 2,3,7,8-TCDD. This product pattern is unique among the five TCDD isomers. For both 1,2,7,8- and 1,3,7,8-TCDD, up to two hydrogens are found to be exchanged. No H/D exchange product was observed for 1,3,6,8-TCDD. However, product ions associated with exchange of three hydrogens in the anions of 1,2,3,4-TCDD are found.

In this study, the reactivity of the 1,2,3,4-TCDD anions is found to be significantly higher than the anions of the other selected isomers. In addition, there is also a strong tendency for 1,2,3,4-TCDD to form a $[M-1+D_2O]^-$ adduct which is completely absent from the spectra of the other isomers. Although the mechanism of the exchange reactions is not known at this point, we propose that the H/D exchange reaction is initiated by attack of the negative charge site on the $[M-1]^-$ ions of TCDD isomers to one of the two deuteriums of a D_2O molecule. Multiple deuterium substitutions on the reactant ion are then formed by sequential H/D exchange reactions with different D_2O molecules. Clearly, the number of exchangeable hydrogens of a TCDD $[M-1]^-$ ion depends on the positions of the chlorines. The formation of $[M-1+D_2O]^-$ and the relatively high H/D exchange reactivity for the anions of 1,2,3,4-TCDD are probably due to the absence of

Figure 4-3 Product mass spectra of $[M-1]^-$ ions derived from TCDD isomers upon hydrogen/deuterium (H/D) exchange reactions with D_2O. The exchange products are indicated by peaks at one or more m/z units above the reactant ions.


[M-1]⁻ (Reactant ion)
relative intensity
m/z
2,3,7,8-TCDD
318.9
319.9
1,3,6,8-TCDD
318.9
1,2,7,8-TCDD
318.9
319.9
320.9
1,3,7,8-TCDD
318.9
319.9
320.9
1,2,3,4-TCDD
341.9
339.9
321.8
320.9
318.9
338.9
300
310
320
330
340
350
20
40
60
80
100

hindrance of Cls on the benzene ring where the charge is located. A complete understanding of the specificity of the H/D exchange reactions may not be known until a more comprehensive study involving all possible isomers is done. Nevertheless, this preliminary study has demonstrated that the H/D exchange reactions allow 2,3,7,8-TCDD to be differentiated from 1,2,7,8-, 1,3,7,8-, 1,3,6,8- and 1,2,3,4-TCDD. An attempt to observe H/D exchange reactions between D_2O and the $[M]^-$ ions of PCDD's was unsuccessful; no exchange products were observed. Since the $[M]^-$ ion is an odd-electron species, we conclude that the lone-pair electrons on the $[M-1]^-$ ions are critical for the initiation of a fruitful H/D exchange reaction.

C. Reactions with Alcohols

When the $[M-1]^-$ ions of PCDDs react with simple alcohols, ion-molecule adduct complexes, $[M-1+alcohol]^-$, are formed. Under our experimental conditions, the reactivities of the TCDD isomers are found to be dependent on the positions of the Cls. The alcohols selected for this study, in ascending order of acidity, are methanol, ethanol, isopropanol and isobutanol. The abundances of the adducts formed for various combinations of alcohol and TCDD isomers are tabulated in Table 4-2.

The reactivity of 2,3,7,8-TCDD is shown to be consistently the lowest among the isomers studied. According to our NCI experiment and others (27), the acidity of 2,3,7,8-TCDD is the highest among all the TCDDs. This means that the $[M-1]^-$ of 2,3,7,8-TCDD is the weakest base of all the anions of TCDDs. For 2,3,7,8-TCDD, there is only one possible site for the negative charge of the $[M-1]^-$ ion because the positions of all four hydrogens on the

Table 4-2 Percentages of the $[M-1+alcohol]^-$ adducts based on 100% of the $[M-1]^-$ reactants.

TCDD isomers	methanol	ethanol	isopropanol	isobutanol
2378	7	17	42	0.5
1368	20	29	50	2.0
1278	34	55	73	2.5
1378	46	74	100	2.7
1234	410	450	6600	3.0

molecule are equivalent. It is conceivable that adduct complexes are formed by interactions between the lone-pair electrons on the charge site and the H of the OH functional group of the alcohol molecules. This explanation is supported by a significant isotopic effect observed when alcohols with an OD functionality are used. The charge site on the $[M-1]^-$ of 2,3,7,8-TCDD is less likely to attack a proton because of its weaker basicity. The reactions with alcohols were also extended to the $[M-1]^-$ ions of 2,3- and 2,7-dichlorodibenzo-p-dioxin (DCDD). Overall, the reactivities of 2,3,-DCDD are also consistently lower than those of 2,7,DCDD. In the case of reactions with methanol, the ratio of $[M-1+alcohol]^-$: $[M-1]^-$ for 2,7-DCDD is found to be about 10 times that of 2,3-TCDD. The differences in reactivities for the two isomers coincide with the fact that while the structure of 2,7-DCDD allows four hydrogens to be adjacent to the chlorines, only the two hydrogens at the 1 and 4 positions of 2,3,DCDD are adjacent to the chlorines. This means that most of the $[M-1]^-$ ions of 2,3-DCDD would have a charge site at the location identical to that of a 2,3,7,8-TCDD $[M-1]^-$ ion. The results from these two DCDD isomers appear to substantiate the above explanation on the differences in TCDD reactivities with the alcohol molecules.

For the alcohol reagents, an increase in reactivity is observed when the acidity of the alcohol is increased from that of methanol to that of isopropanol. This is exactly what is expected for an acid/base reaction. The dramatic decrease in reactivity for isobutanol is probably due to the steric hindrance of its structure. For the higher chlorinated PCDD congeners, reactions between the $[M]^-$ ions and alcohol molecules do not produce any

products. The lack of the lone-pair electrons on the $[M]^-$ ions apparently do not allow the formation of the complexes.

D. Reactions with O_2

Oxygen molecules were also used as a reagent to react with the $[M]^-$ ions of PCDDs. At a collision pressure of 0.8 mtorr and collision offset energy of 2.8 eV, reactions between the $[M]^-$ of 1,2,3,4-TCDD and O_2 produce $[M\text{-}Cl+O]^-$ ions from oxygen/chlorine (O/Cl) exchanges. Although the ratio of $[M\text{-}Cl+O]^-$: $[M]^-$ for 1,2,3,4-TCDD is found to be about 3:1, the O/Cl exchange product is not observed for the $[M]^-$ ions from 2,3,7,8-, 1,3,6,8-, 1,2,7,8- and 1,3,7,8-TCDD under our experimental conditions. It is necessary to point out that the $[M]^-$ ions selected for the 2:2 substituted TCDD isomers also include the ^{13}C isotope of the $[M\text{-}1]^-$ ions, which could not be separated by the first quadrupole mass filter. However, this should have little or no effect on the formation of any product ions resulted from the $[M]^-$ ions because no reaction was observed when the $[M\text{-}1]^-$ ions of PCDDs were reacted with O_2. The $[M\text{-}Cl+O]^-$ ions are obtained from the $[M]^-$ ions of the penta-, hexa- and hepta-chlorinated PCDD congeners. Although others (25,26) have demonstrated the potential of using O_2 as a reagent to differentiate 2,3,7,8-TCDD from 1:3 substituted TCDD isomers, we could not produce experimental evidence to support the potential usefulness of using O_2 to differentiate 2,3,7,8-TCDD from the other 2:2 substituted TCDD isomers. More experiments are needed in order to determine the mechanism of the O/Cl exchange on the $[M]^-$ ions of PCDDs, and thus the reason for the apparent lack of reactivity of the 2:2 substituted TCDDs.

Conclusions

Ion-molecule reactions occurring in the center stage of a triple quadrupole mass spectrometer are feasible options for distinguishing the more toxic isomers of PCDDs from the less toxic ones. For reactions involving the formation of $[M-1+alcohol]^-$ adducts, the reactivity of the $[M-1]^-$ of 2,3,7,8-TCDD is consistently the lowest among the five TCDD isomers studied. Of the four simple alcohols selected for these reactions, methanol appears to allow 2,3,7,8-TCDD to be differentiated to a greater degree from the other less toxic isomers. The H/D exchange product patterns obtained from reactions between the $[M-1]^-$ ions of TCDD isomers and D_2O permit 2,3,7,8-TCDD to be differentiated from 1,3,6,8-, 1,2,7,8-, 1,3,7,8- and 1,2,3,4-TCDD. At this stage, it is not known whether the H/D exchange reactions are useful to differentiate 2,3,7,8-TCDD from all other TCDD isomers. A more comprehensive study involving all possible TCDD isomers is needed in order to gain additional insight into the mechanism of these reactions, and verify their applicability as analytical techniques.

The unique negative charge location on the $[M-1]^-$ of 2,3,7,8-TCDD may be further utilized for a contiuous search of ion-molecule reactions which are truly specific for 2,3,7,8-TCDD. With the selectivity of MS/MS, the added specificity provided by ion-molecule reactions may reduce sample cleanup and shorten GC separation time for 2,3,7,8-TCDD analysis.

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CHAPTER 5

IMPROVEMENT TO THE DETECTION OF LOW-ENERGY ION PRODUCTS IN A TRIPLE QUADRUPOLE MASS SPECTROMETER - A THEORETICAL CONSIDERATION

Introduction

Ion-molecule reactions occurring at thermal or near thermal energies are often very sensitive to structural differences. Ion products formed by these reactions possess little or no kinetic energy. Unfortunately, the lack of kinetic energy of the ion products creates a challenge for their detection. In a triple quadrupole mass spectrometer (TQMS), the detection of these ions in the second quadrupole collision chamber (Q2) depends on the ability of the ions to reach the detector. Without a guiding field, many of these ions would eventually be lost from the reaction chamber in directions other than towards the detector. In order to increase the detection efficiency of these ions, a standard technique uses a withdrawing potential applied between Q2 and the third quadrupole (Q3) to extract the low-energy and thermalized ions from Q2 for mass analysis by Q3. However, for thermalized ions, ion currents are limited by the rate of diffusion along the axis of Q2 from the point where their axial kinetic energy becomes zero until they reach the drawout potential field that extends only a short distance into the reaction chamber. This chapter will describe novel designs of the second quadrupole for facilitating the movement of low-energy ion products towards the detector.

Where the study of the thermochemistry of ion-molecule reactions is not the main goal, relatively high collision pressures (≈ 4 mtorr) of neutral molecules can be introduced into Q2 in order to assure an abundance of neutral molecules for reactions. Under high pressure conditions, ions transmitted into the reaction chamber may have multiple collisions with the neutral molecules before reaching the exit of Q2. Such reaction conditions increase the rate of ion-molecule reactions and thus the yield of products. Furthermore, multiple collision conditions have a stabilization effect on products and intermediates that would otherwise be unstable and undetectable under single collision conditions (1). A novel ion trapping technique (2,3) has been developed at Michigan State University by Dr. Watson's and Dr. Enke's research groups for enhancing multiple collision conditions even at low collision pressures. The detection of reaction products is improved essentially by a net gain of product signals due to the presence of more product ions.

The novel ion trapping technique requires no hardware modification to a traditional TQMS instrument. The so-called trap-and-pulse mode of this technique involves the use of an extraction lens (EL) that is placed between Q2 and Q3 as an ion trapping and pulsing device. Usually, +100V is applied to the extraction lens for trapping positive ions and -100V for negative ions. In this method, parent ions of a selected mass are continuously transmitted into Q2 for ion-molecule reactions while the trapping potential is applied to EL. After a certain length of time, a pulsing potential, which is opposite in polarity to the trapping potential, is applied to extract a fraction of the trapped ions from Q2 for detection. Ion trapping time can be varied to maximize ion signals. Normally, 90% of the

maximum intensity can be obtained within 100 ms of trapping time. This method is necessarily limited to collecting ions of a single nominal mass-to-charge value per pulse because the millisecond duration of the ion pulse is too short for full range Q3 mass scanning. Nevertheless, significant improvements in signal-to-noise ratios have been obtained over those in the conventional data collection method. The greater residence time of ions in Q2 made possible by extending the trap time revealed some reaction products not observed by continuous product extraction.

Designs for the Second Quadrupole Collision Chamber of a TQMS Instrument

The extraction lens used in both the standard and the trap-and-pulse methods can only extract ions that are very near the exit of Q2. A high percentage of the thermalized ions still stay in Q2 without being detected as products. Figure 5-1 shows a potential contour diagram for the standard ion extraction design based on calculations using SIMION (4), a computer program for ion optics calculations. This diagram indicates that the 100 V extraction potential applied to the lens at the end of the collision chamber can only penetrate a very short distance into Q2. Most ion products formed in Q2 are not directly affected by the extraction potential. An idea to assist the thermalized ions to move towards the exit of Q2 is to create a longitudinal electric field along the path of the ions towards that region. In this way, ions can experience a continuous pushing force in the direction of the Q2 exit. This is particularly important for ions that are heavily surrounded by neutral molecules under high collision pressure conditions.

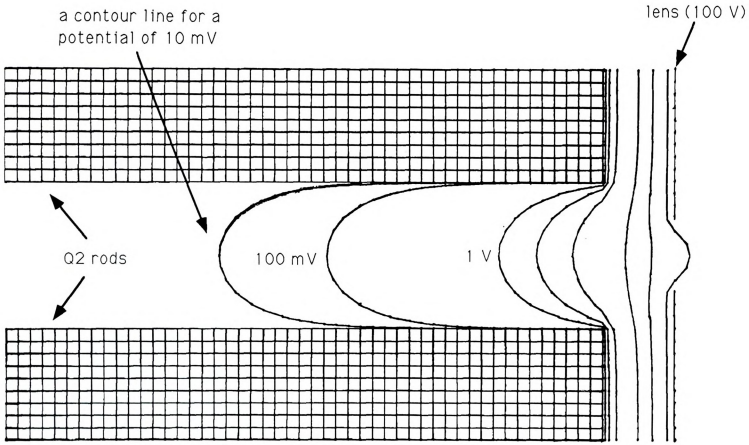


Figure 5-1 A potential contour diagram for the extraction lens design.

The first idea for creating such a longitudinal electric field is to modify the design of the Q2 rods in such a way that the rods are made of stainless steel segments which are separated by thin layers of non-conductive Teflon (Figure 5-2). Resistors are connected to each segment to divide the total voltage along the rod into equal steps for each segment. If a potential difference is applied to the ends of each rod, a potential field gradient is created in space along the ion path of Q2. Theoretically, for a true smooth continuous potential field gradient, an infinite number of infinitesimally thin segments are needed in each rod. If the rods are made of too few segments, the gradient created along the Q2 ion path would be in a step-jump fashion. Using SIMION calculations, the potential contour diagrams for the designs of 6-, 11-, 21-, 26- and 35-segmented Q2 rods have been generated. The configurations used for the calculations are shown in Figure 5-3 through Figure 5-7. In this case, all the calculations are based on a 5-V potential difference applied to the ends of a 20-cm Q2 assembly. Based on data extracted from the potential contour diagrams, plots of longitudinal potential as a function of spatial distance into Q2 for the 6-, 11-, 21-, 26- and 35-segmented designs are created and shown in Figure 5-8. The 6- and 11-segmented designs give distinct potential plateaus in the space along the ion path of Q2. At the region of a potential plateau, ions would experience only a very slight potential gradient. These ions may still stay in the collision chamber if the forward momenta of the ions are too small to allow their arrival to the next potential gradient regions. Since the potential gradient along the ion path of Q2 is related to the change of potential as a function of changing spatial distance inside Q2, plots that represent the 1st derivative of the results of Figure 5-8 would give a direct representation for the smoothness of the potential gradients for each

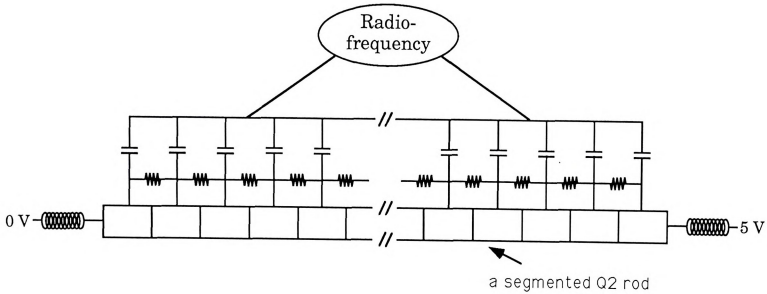


Figure 5-2 A design of segmenting the second quadrupole rods

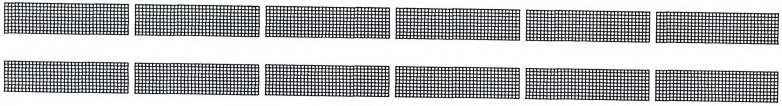


Figure 5-3 The configuration of a potential contour diagram for a Q2 made of 6-segmented rods with a 5-V potential applied.

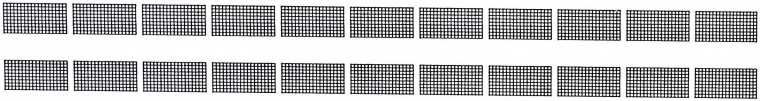


Figure 5-4 The configuration of a potential contour diagram for a Q2 made of 11-segmented rods with a 5-V potential applied.

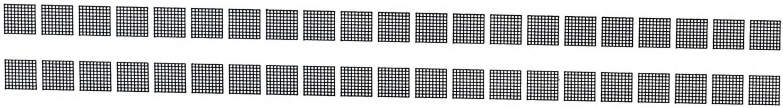


Figure 5-5 The configuration of a potential contour diagram for a Q2 made of 21-segmented rods with a 5-V potential applied.

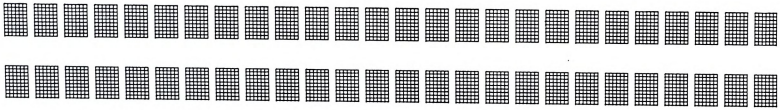


Figure 5-6 The configuration of a potential contour diagram for a Q2 made of 26-segmented rods with a 5-V potential applied.

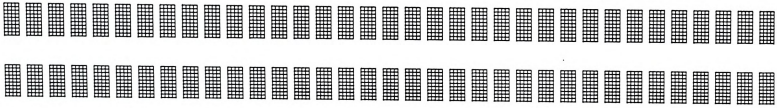
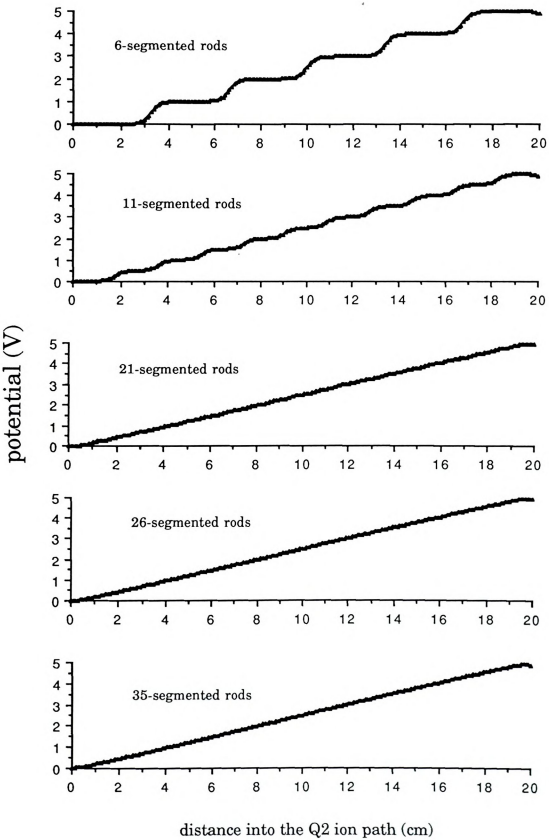


Figure 5-7 The configuration of a potential contour diagram for a Q2 made of 35-segmented rods with a 5-V potential applied.

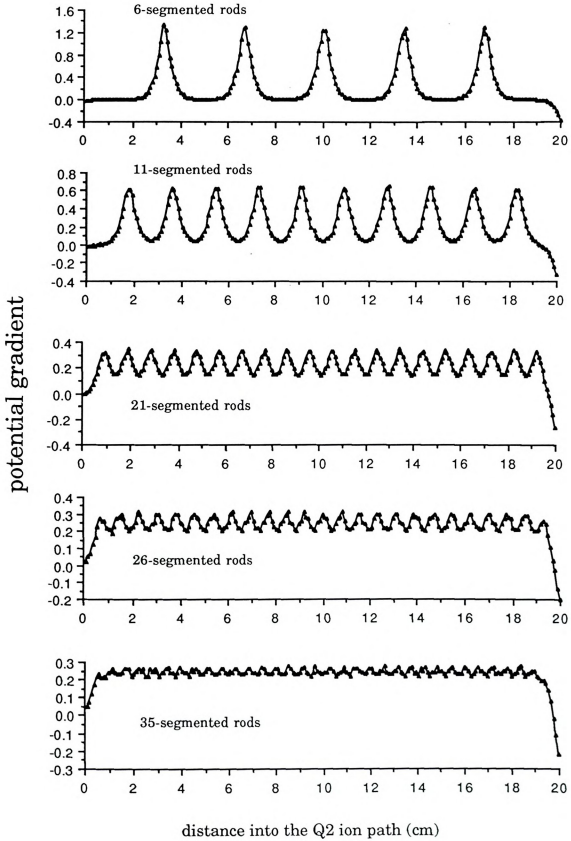
Figure 5-8 Potentials along the ion path for the designs of Q2 made of different numbers of segments when a potential difference of 5-V is applied.


6-segmented rods
11-segmented rods
21-segmented rods
26-segmented rods
35-segmented rods
potential (V)
distance into the Q2 ion path (cm)

design. The potential gradient plots shown in Figure 5-9 indicate that increasing the number of segments for the Q2 rods allows a smoother potential gradient. With a 5-V potential applied to the 20-cm Q2 rods, an idea potential gradient throughout the entire Q2 ion path would be 0.25 V/cm. According to the potential gradient plots, the average potential gradient variations for the 21-, 26- and 35-segmented designs are approximately 40%, 20% and 5%, respectively from the idea 0.25 V/cm. For this application, an average variation in potential gradients should not be more than 10%. This means that at least 30 segments are needed for creating a reasonably smooth potential gradient along a 20-cm Q2 ion path. An extraction lens is still needed for the segmented designs because the calculations suggest that there is a negative potential gradient near the exit region of Q2. Nevertheless, the smoothness of the field gradient created is clearly a function of the number of segments used to make the Q2 rods. Mechanically, there is a limitation to the maximum number of segments possible for rods with a specific length. If the four rods of Q2 are each made of 50 segments, the fabrication of the device would be very labor-consuming and mechanically tedious.

Although the idea of a segmented Q2 quadrupole is theoretically feasible, the making of the device is very difficult. In the summer of 1988, after discussions with Dr. E. Strangas in the electrical engineering department and Dr. C. G. Enke, a new idea emerged for creating a smooth field gradient within Q2 without physically segmenting the quadrupole. In this novel idea, a resistance wire is wounded around each Q2 rod to create the effect of a voltage divider. High resistance wires with resistivity up to 800 ohm/cmf (or 1.38×10^{-6} ohm·meter) can be purchased from several

Figure 5-9 The 1st derivative plots representing potential gradient vs spatial distance into the Q2 ion path.


6-segmented rods
11-segmented rods
21-segmented rods
26-segmented rods
35-segmented rods
potential gradient
distance into the Q2 ion path (cm)

manufacturers. Winding the resistance wire by the conventional helical coil method will create an inductance that will make each rod an electro-magnet. It is certain that the magnetic field will have unwanted interactions with ions in Q2. The problem of inductance can be eliminated if wires are winded onto the rods by a non-circular zig-zag winding technique (Figure 5-10). In this winding technique, wire direction around the rod is reversed at a series of hooks installed on the back of each rod. Looking out from the inside of the quadrupole, this wire-winding design gives essentially the same physical features as the conventional circular coiling method except for the absence of inductance when an RF voltage is applied. Each winding turn of wires is equivalent to one segment of resistance. The maximum number of turns depends on the number of hooks on the rods. Clearly, winding 50 turns of wires on the rods of Q2 is a lot easier and more economical than making a Q2 quadrupole with 50 different segments. For a wire-winding design that gives an equivalence of 50 segments in each rod, ions entering Q2 should experience a very gentle longitudinal field gradient.

Because the purpose of using resistance wires for the Q2 modification is to create the effect of a voltage divider, rods that support these wires must not be conductors. Technically, a perfect insulator would be ideal for making the Q2 rods. Common metallic materials such as stainless steel and copper cannot be used to make rods in this case because of their high electrical conductivity. In order to evaluate the feasibility of this design, it is also necessary to obtain information about the resistance of the wires winded on Q2 as it is related to the amount of heat generated when a

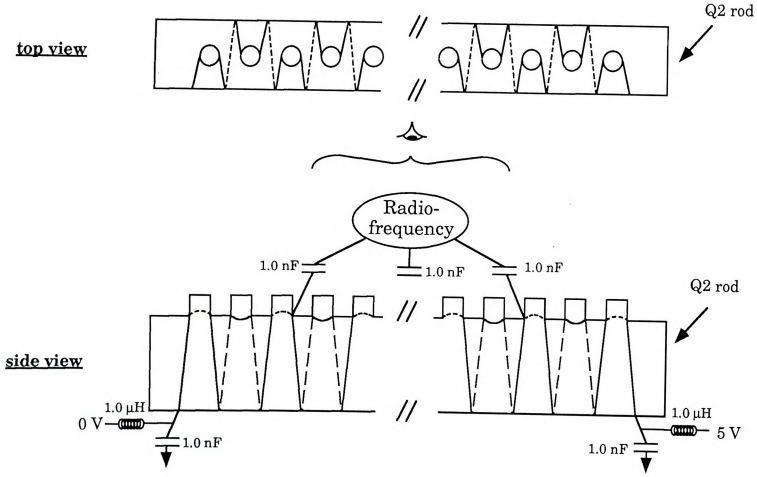


Figure 5-10 A design of modifying the second quadrupole using the non-circular zig-zag wire-winding technique. Dash lines represent wires winded on the back side of the rod.

potential gradient is applied. Generally, this can be calculated according to the following equation:

$$\text{resistance (R)} = \frac{\rho L}{A}$$

where ρ is the resistivity of the wire, L is the length of the wire, and A is the cross-section area of the wire. For a design that uses wires with resistivity of 800 ohm/cmf and a cross-section diameter of 0.5 mm, the resistance of a 50-turn wire winded on a 0.78 cm^2 x 20 cm rod is calculated to be about 11 ohm. This resistance will generate about 2.2 W of power when a 5-V potential difference is applied. The whole quadrupole winded with the resistance wires will generate close to 9 W of heat. For a system operated at high vacuum conditions, without the means of conduction, dissipation of such large amount of heat through radiation alone is too inefficient. The only efficient means for dissipating the heat would be by conduction through the rods. This imposes another restriction for the type of materials that can be used to make these rods. For this application, the four rods of Q2 must be made of a material that is both a good heat conductor and a good electrical insulator. This restriction excludes most available materials in the market. Upon an extensive library search, a ceramic material called beryllium oxide (BeO) (5) is found to be both an excellent heat conductor and electrical insulator. Figure 5-11 shows the room temperature thermal conductivity of BeO compared to various materials. In fact, the thermal conductivity of BeO is higher than most metallic materials. The Q2 design made of BeO can be fabricated by manufacturers for under $1000.

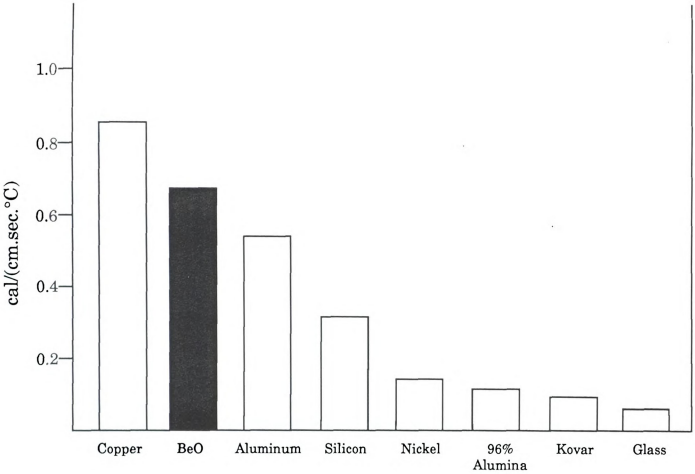


Figure 5-11 Room temperature thermal conductivity of BeO compared to various materials. (adapted from literature data compiled by Ceradyne, Inc., Costa Mesa, CA).

For the improvement of the trap-and-pulse technique described before, the wire-winding Q2 modification can be further refined. Instead of an extraction lens, the last 10 turns of the resistance wire of each rod can be used for a 10-step longitudinal field gradient either for trapping or pulsing of ions. This could provide an environment for a less abrupt potential field change at the Q2 exit region. Similarly, another 10-step longitudinal field gradient can be implemented near the entrance region of Q2 for a so-called inject-trap-and-pulse operation in which a large potential field applied at this region functions as an ion gate to allow only a packet of ions to enter the collision chamber for trapping and pulsing. The longitudinal potential field will be in "boat" and "chair" forms during ion trapping and pulsing, respectively.

Conclusions

The idea of facilitating the movement of low-energy ions in Q2 towards the detector could be accomplished by creating a longitudinal electric field gradient along the ion path inside Q2. This could be created by making Q2 rods behave as voltage dividers. This can be accomplished by either using segmented rods connected with resistors or rods winded with high resistance wires. Because the smoothness of a potential field gradient created within Q2 depends on the number of voltage-dividing segments on each rod, for creating rods with a high number of voltage-dividing segments, the design of using rods winded with resistance wires is mechanically more feasible than the other design. However, the wire-winding design requires the use of rods made of a material that is both a

good electrical insulator and a good heat conductor. Beryllium oxide would appear to be an ideal material for the application.

A smooth longitudinal potential field gradient created inside Q2 would allow the detector to sample low-energy product ions formed in every region of Q2. Product spectra obtained through the proposed method described in this chapter should represent the ion distribution in Q2 more closely than the traditional method.

References

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2. Dolnikowski, G. G.; Kristo, M. J.; Enke, C. G.; Watson, J. T., *Int. J. Mass Spectrom. Ion Phys.* **1988**, 82, 1.

3. Kristo, M. J., Ph.D Dissertation, Michigan State University, 1987.

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5. National Beryllia Corporation: National Beryllia Technical Bulletin on Berlox Beo.

CHAPTER 6

CONCLUSIONS AND SUGGESTED FUTURE EXTENSIONS

The specificity of ion-molecule reactions has been well-demonstrated in this dissertation as a mean for distinguishing isomers of halogenated aromatic compounds. The unique features of a TQMS instrument offer enhanced selectivity for analytical applications of ion-molecule reactions. In particular, H/D exchange reactions are found to produce very distinct product spectra for the $[M-1]^-$ of halogenated benzene isomers. These allow the isomers to be differentiated when traditional mass spectrometric techniques fail. A series of studies of H/D exchange reactions using various deuterated reagents suggests that D_2O is among the most effective reagents for exchanging aromatic hydrogens of the $[M-1]^-$ ions of chlorobenzene isomers. When D_2O is used to react with the $[M-1]^-$ ions of tetrachlorodibenzo-p-dioxin (TCDD) isomers, 2,3,7,8-TCDD gives an unique H/D exchange product pattern which allows its differentiation from the other selected TCDD isomers. In addition, alcohol reagents are also found useful for differentiating 2,3,7,8-TCDD from the other less toxic TCDD isomers. The results described in this dissertation strongly demonstrate the great potential of applying ion-molecule reactions as analytical tools for differentiating organic isomers. A tandem quadrupole mass spectrometer is particularly well-suited for such applications.

Although studies of ion-molecule reactions have been extensive over the past three decades, the use of ion-molecule reactions for analytical

applications is largely unexplored. This dissertation has merely explored a very small aspect of the enormous potential of the reactions. Certainly, there is much work still to be done. The following are some suggestions.

I. Substituting one of the two chlorines of a dichlorobenzene molecule with other functionalities such as NH_2, OH, CHO, CH_3, OCH_3, COOH or NO_2 is found to substantially reduce the reactivity of the $[M-1]^-$ ion for H/D exchange reactions with D_2O. The observed reduction in reactivity suggests that while D_2O is an excellent reagent for exchanging hydrogens of dichlorobenzene $[M-1]^-$ ions, different deuterated reagents are needed for effectively exchanging the hydrogens of the substituted chlorobenzene $[M-1]^-$ ions. The study of H/D exchange reactions on the substituted chlorobenzene $[M-1]^-$ ions should be continued by exploring the use of different deuterated reagents such as ND_3, CD_4, CH_3OD, CH_3COOD, etc.. The results of such a study may be important in establishing the effectiveness of using ion-molecule reactions for differentiating isomers of the substituted chlorobenzenes.

II. The proposed H/D exchange mechanism involving the formation of a five-membered-ring intermediate for reactions between the $[M-1]^-$ of chlorobenzene and D_2O is consistent with the results derived from all the isomers of chlorobenzenes. It is quite possible that a similar mechanism is also applicable to the $[M-1]^-$ of aromatic compounds containing two or more fused benzene rings. Our mechanistic study may be extended to compounds with multiple benzene rings. Isomers of chloronaphthalene are ideal candidates for initial studies. With a better understanding of the two-ring system, the investigation might then be continued on compounds

with three or more fused benzene rings. Information obtained from the mechanistic studies may be useful for developing better analytical strategies.

III. Although the ion-molecule reactions described in Chapter 4 are found useful to differentiate 2,3,7,8-TCDD from other less toxic isomers, the study was limited to only a few isomers. In order to investigate the usefulness of these reactions as analytical techniques, future studies should include all 22 TCDD isomers. Since the structures of polychlorodibenzofurans (PCDFs) are very similar to those of polychlorodibenzo-p-dioxins (PCDDs), the study of PCDDs may also be extended to include isomers of PCDF.

IV. Finally, the collision cell design described in Chapter 5 is expected to provide improvement in the detection of low-energy product ions formed inside the collision cell; the implementation of the proposed design would be an interesting project to explore.

APPENDIX

Appendix

Computer-simulated individual collision-product plots based on different percentages of reactive collision for the H/D exchange reactions derived from different structures of the $[M-1]^-$ ions of dichlorobenzene isomers. The simulations are based on the proposed H/D exchange reaction mechanism described in Chapter 3.

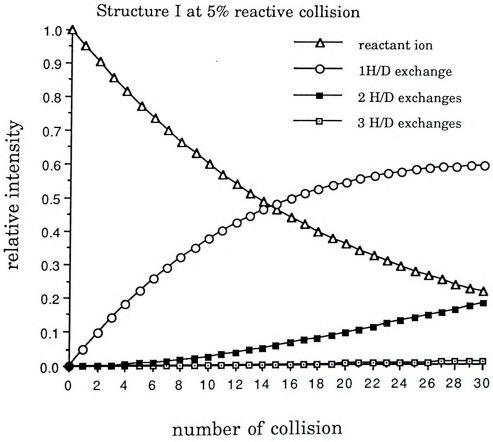

Figure A-1 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 5% reactive collision.

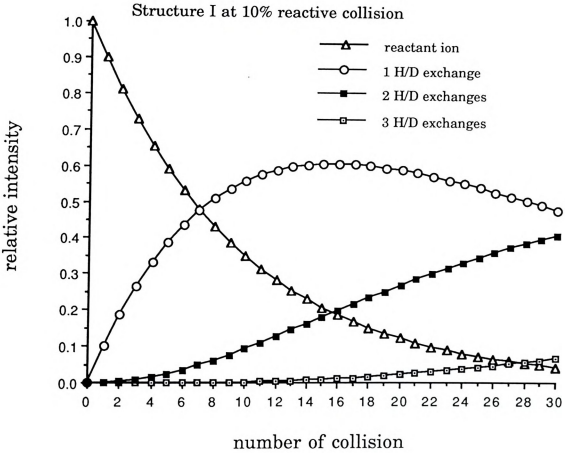

Figure A-2 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 10% reactive collision.

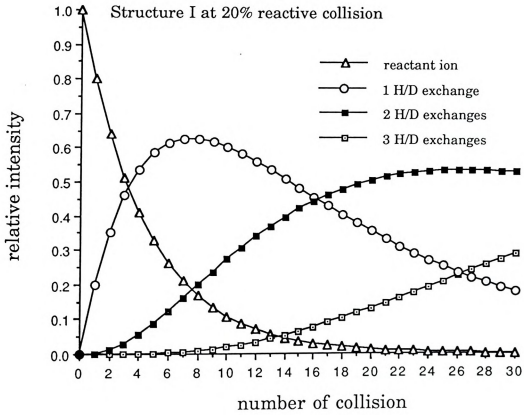

Figure A-3 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 20% reactive collision.

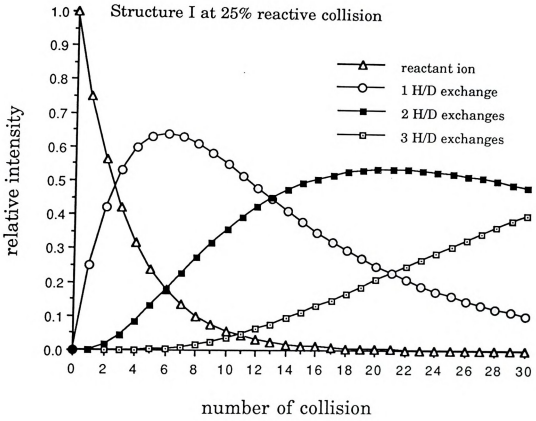

Figure A-4 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 25% reactive collision.

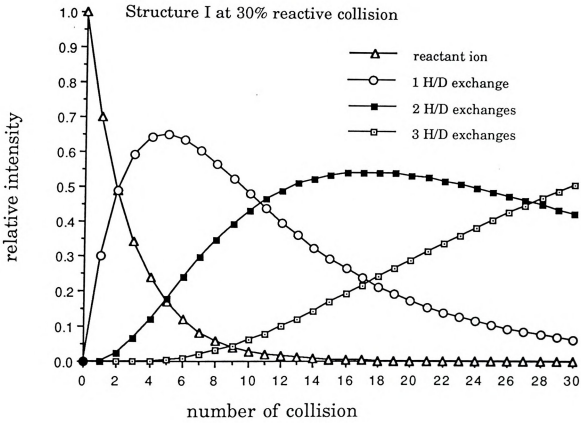

Figure A-5 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 30% reactive collision.

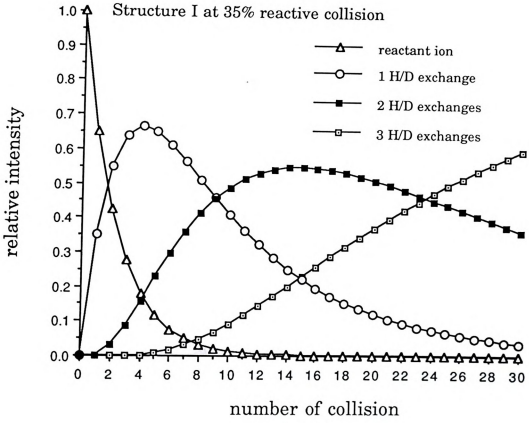

Figure A-6 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 35% reactive collision.

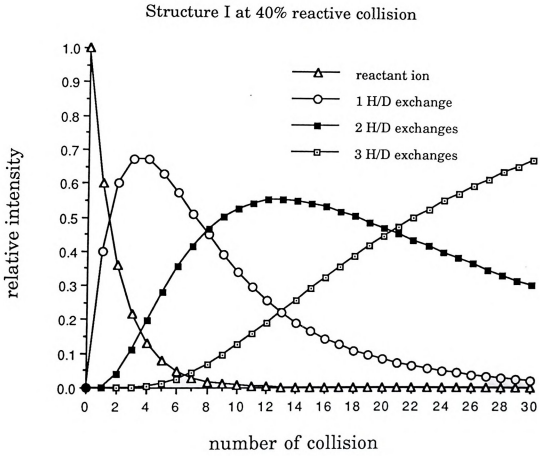

Figure A-7 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 40% reactive collision.

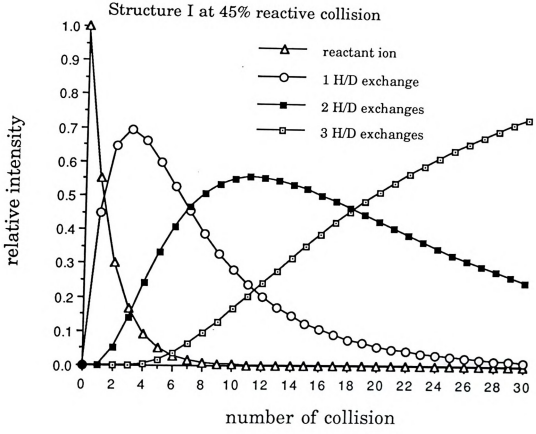

Figure A-8 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 45% reactive collision.

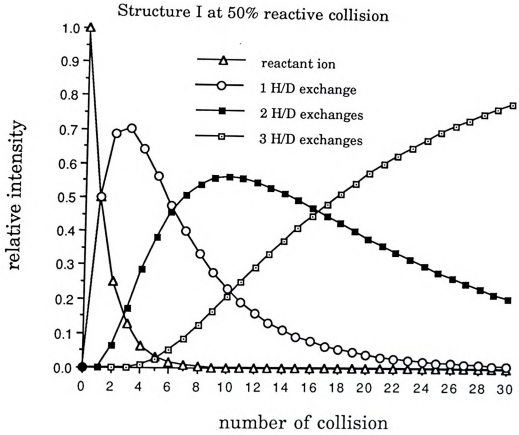

Figure A-9 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 50% reactive collision.

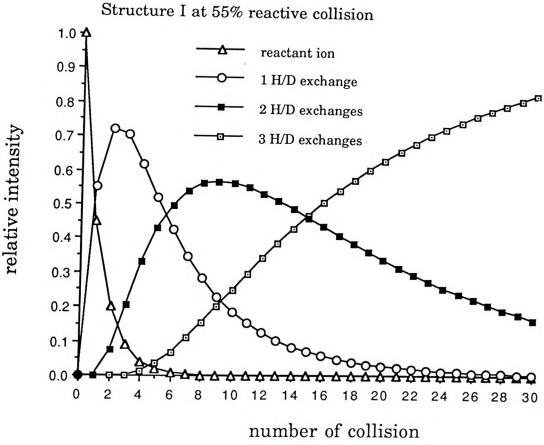

Figure A-10 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 55% reactive collision.

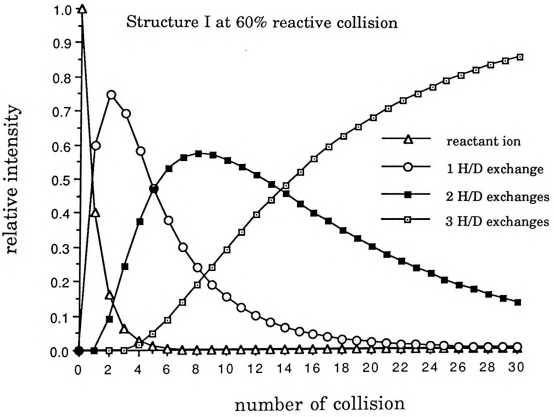

Figure A-11 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 60% reactive collision.

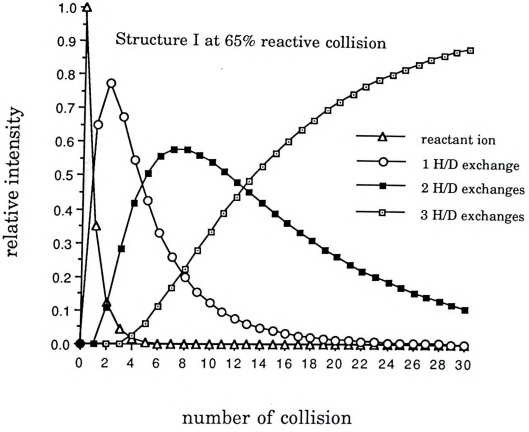

Figure A-12 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 65% reactive collision.

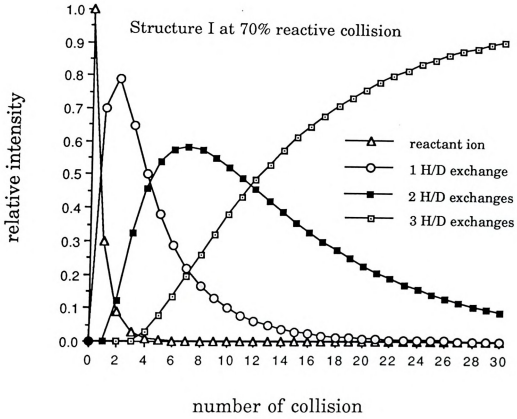

Figure A-13 A simulated collision-product plot for the structure I of o-dichlorobenzene based on an H/D exchange reactivity of 70% reactive collision.

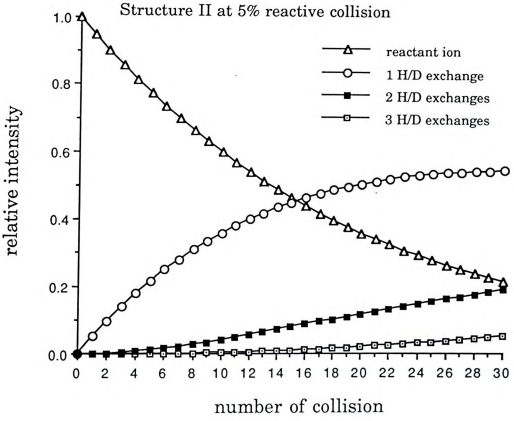

Figure A-14 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 5% reactive collision.

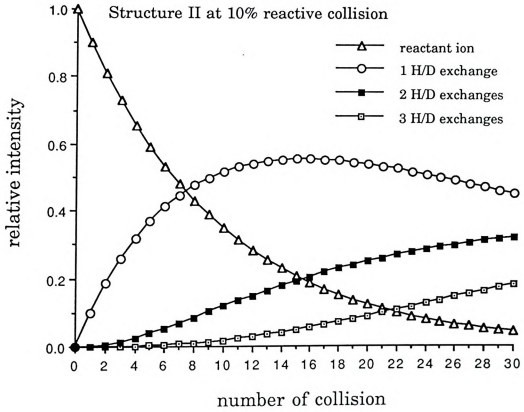

Figure A-15 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 10% reactive collision.

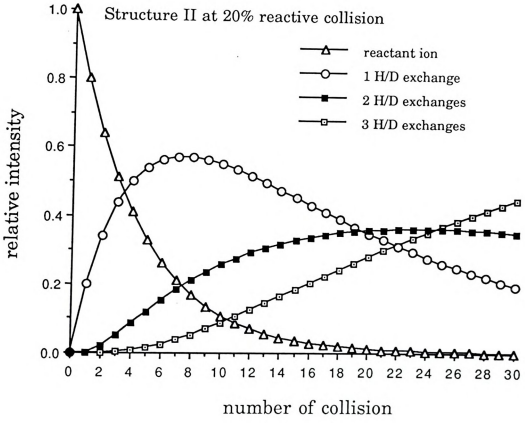

Figure A-16 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 20% reactive collision.

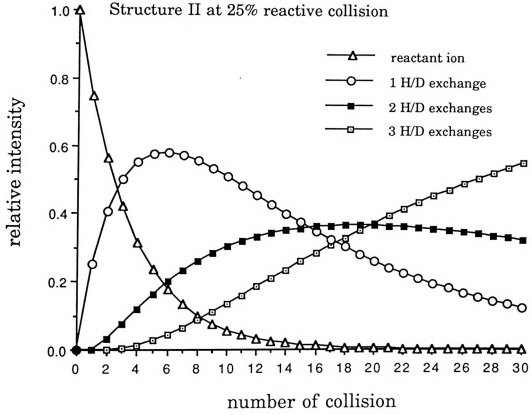

Figure A-17 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 25% reactive collision.

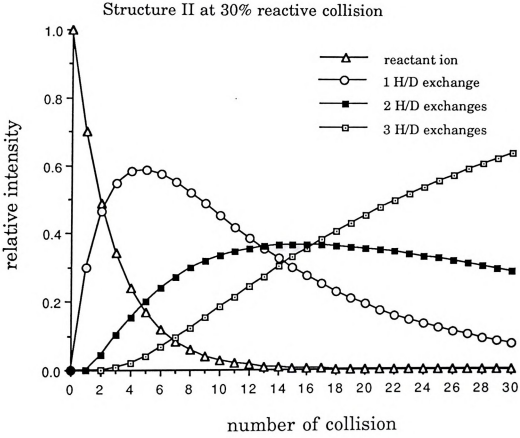

Figure A-18 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 30% reactive collision.

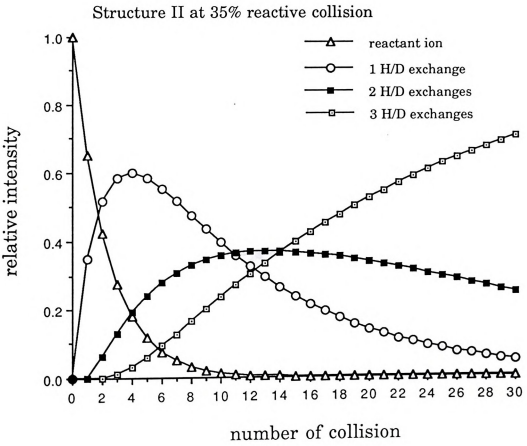

Figure A-19 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 35% reactive collision.

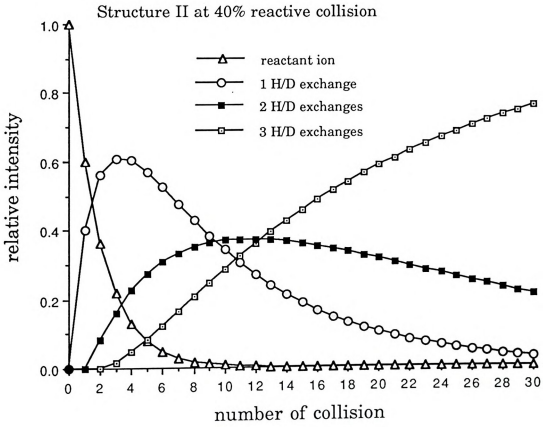

Figure A-20 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 40% reactive collision.

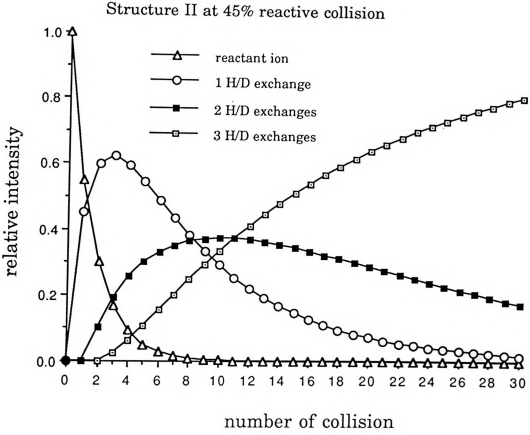

Figure A-21 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 45% reactive collision.

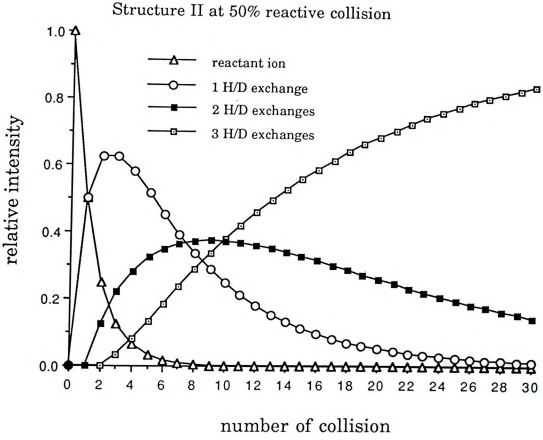

Figure A-22 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 50% reactive collision.

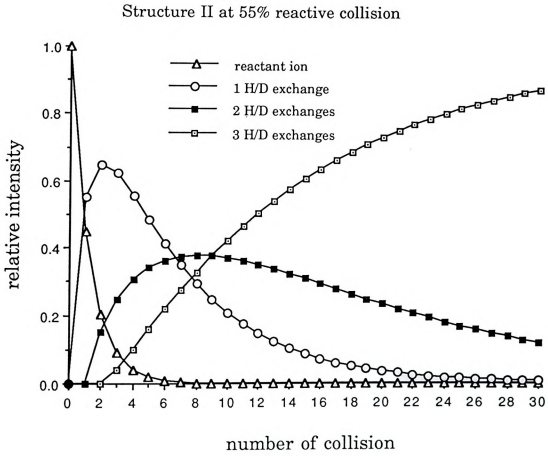

Figure A-23 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 55% reactive collision.

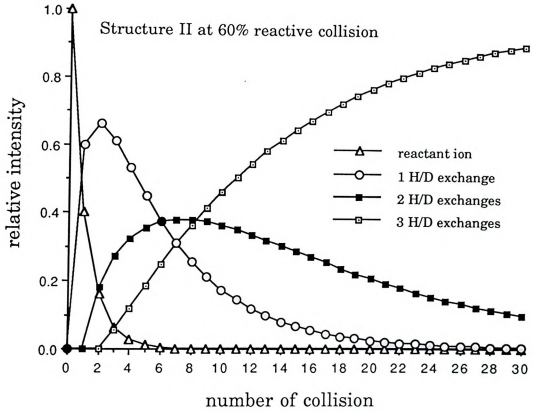

Figure A-24 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 60% reactive collision.

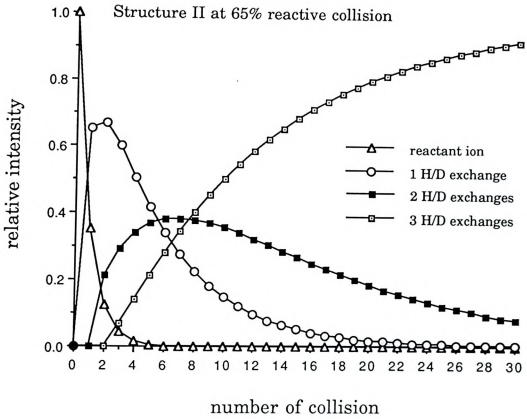

Figure A-25 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 65% reactive collision.

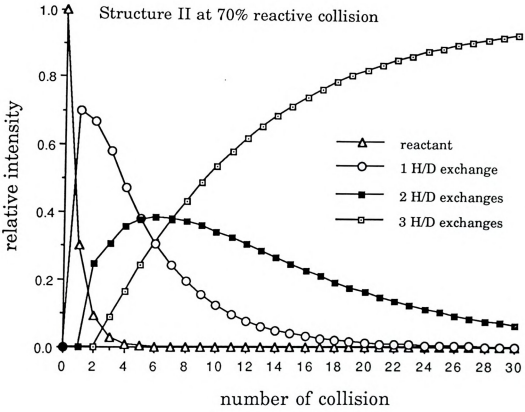

Figure A-26 A simulated collision-product plot for the structure II of o-dichlorobenzene based on an H/D exchange reactivity of 70% reactive collision.

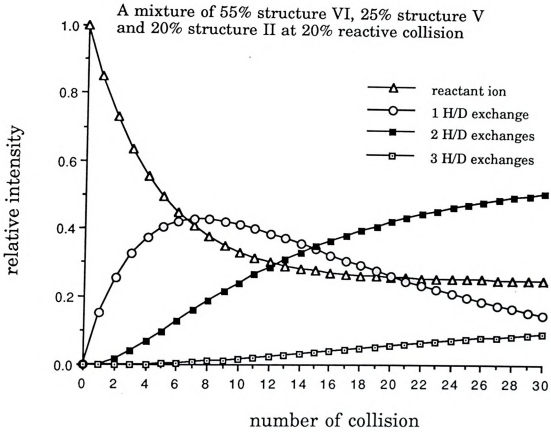

Figure A-27 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 20% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

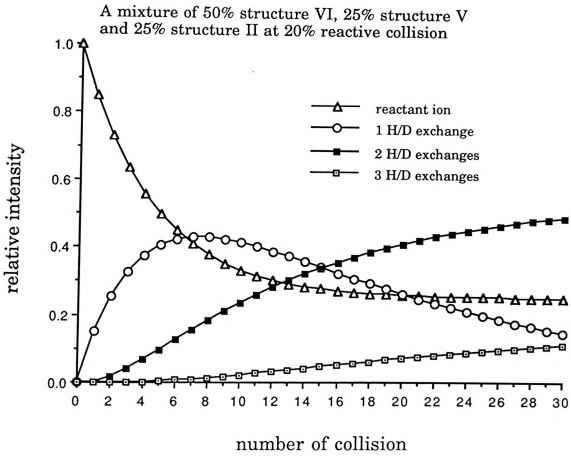

Figure A-28 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 20% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

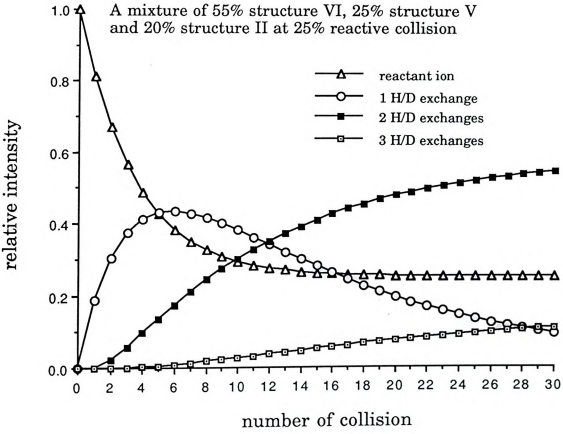

Figure A-29 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 25% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

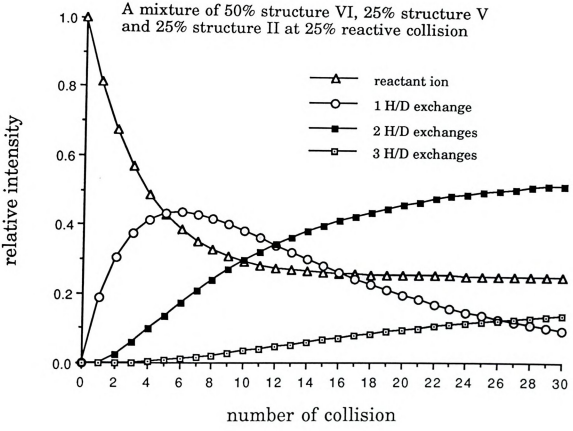

Figure A-30 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 25% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

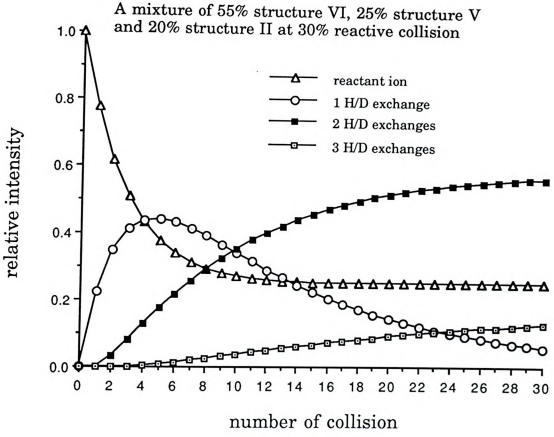

Figure A-31 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 30% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

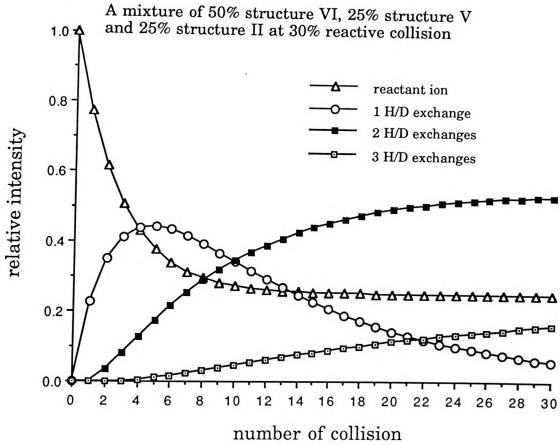

Figure A-32 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 30% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

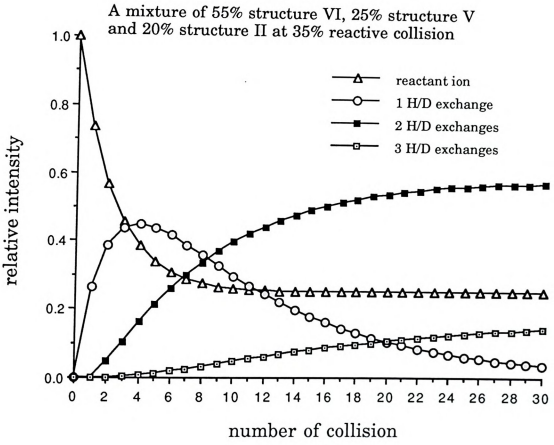

Figure A-33 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 35% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

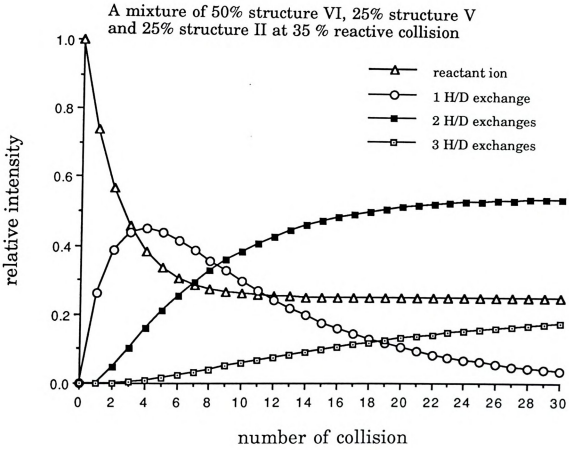

Figure A-34 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 35% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

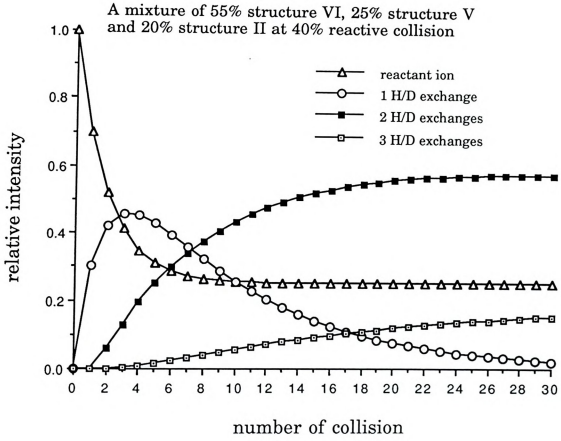

Figure A-35 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 40% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

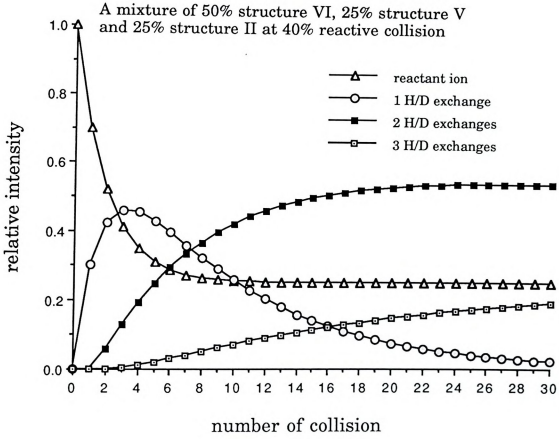

Figure A-36 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 40% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

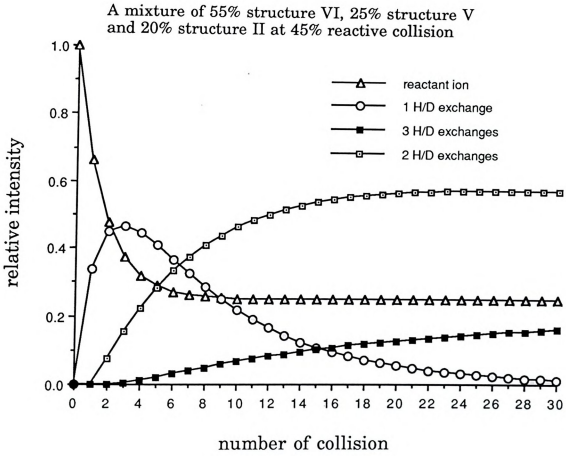

Figure A-37 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 45% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

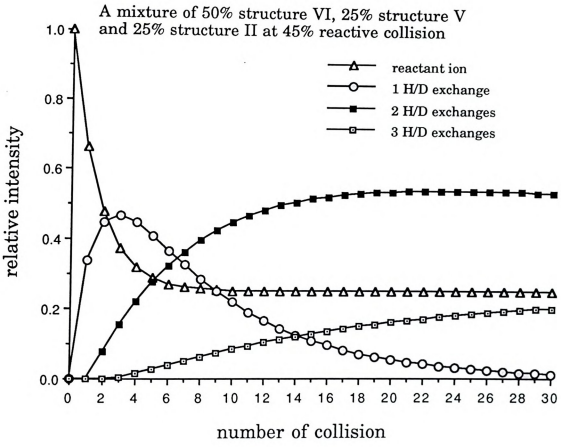

Figure A-38 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 45% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

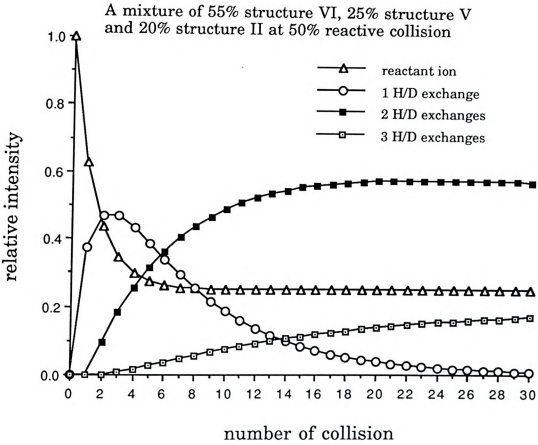

Figure A-39 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 50% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

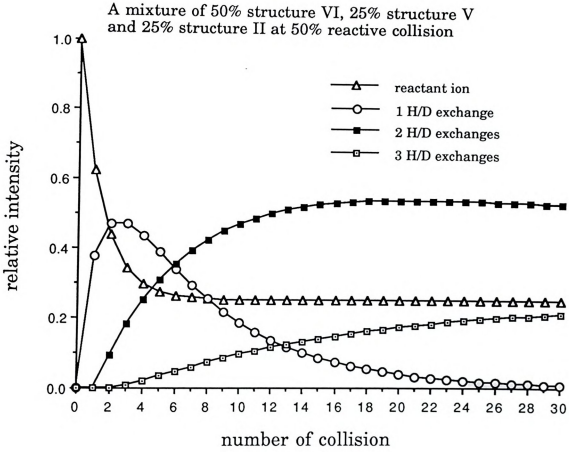

Figure A-40 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 50% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

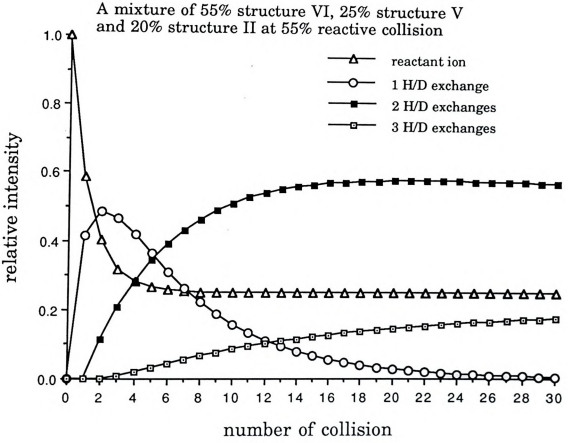

Figure A-41 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 55% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

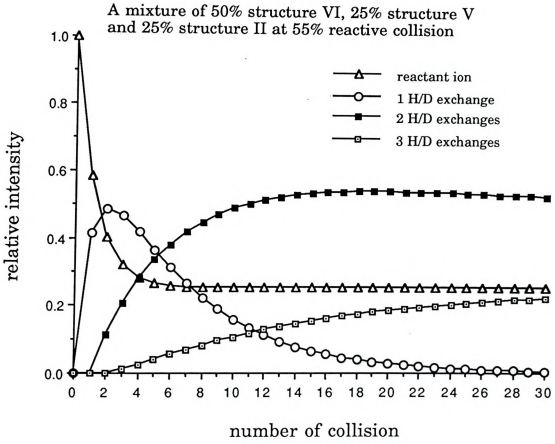

Figure A-42 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 55% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

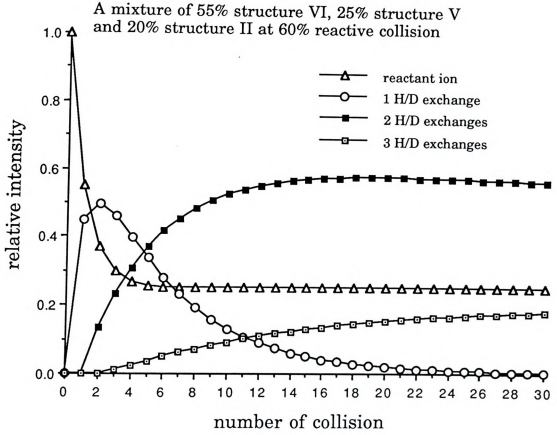

Figure A-43 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 60% reactive collision. The mixture contains anions of 55% structure VI, 25% structure V and 20% structure II.

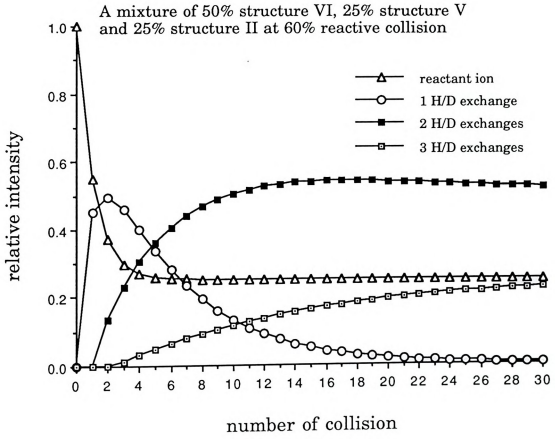

Figure A-44 A simulated collision-product plot for a mixture of dichlorobenzene anions originated from m-dichlorobenzene. The calculations are based on an H/D exchange reactivity of 60% reactive collision. The mixture contains anions of 50% structure VI, 25% structure V and 25% structure II.

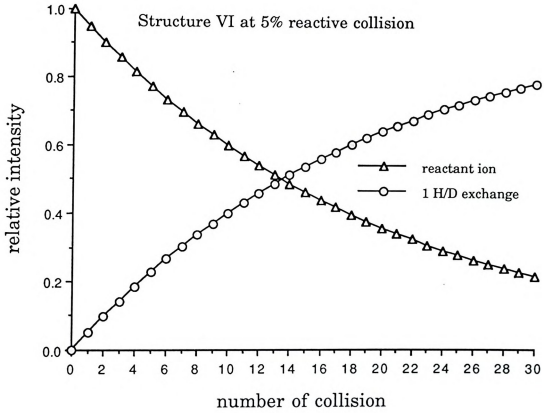

Figure A-45 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 5% reactive collision.

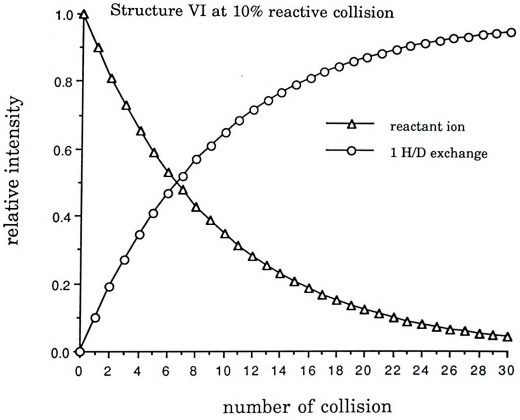

Figure A-46 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 10% reactive collision.

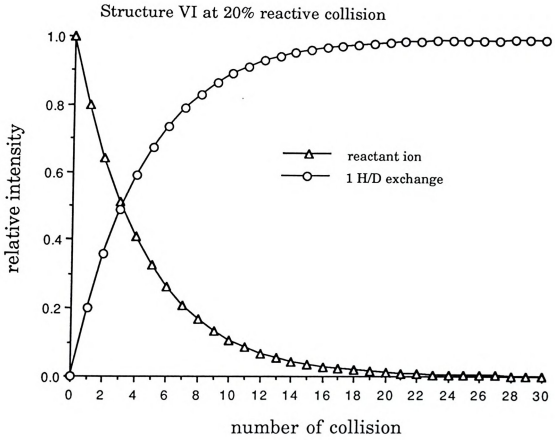

Figure A-47 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 20% reactive collision.

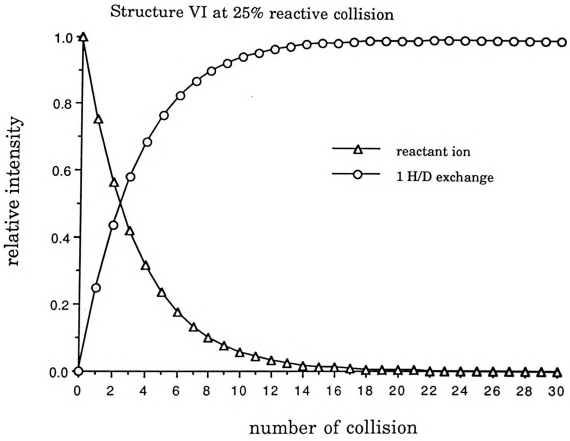

Figure A-48 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 25% reactive collision.

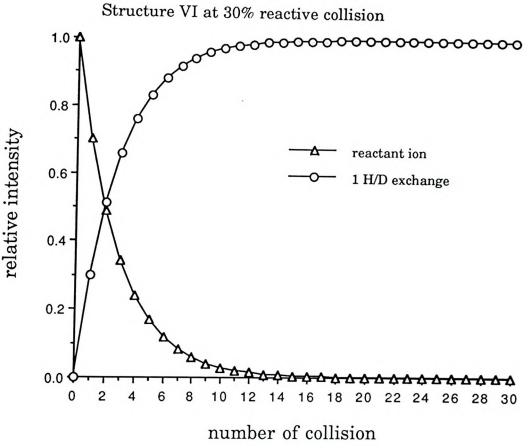

Figure A-49 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 30% reactive collision.

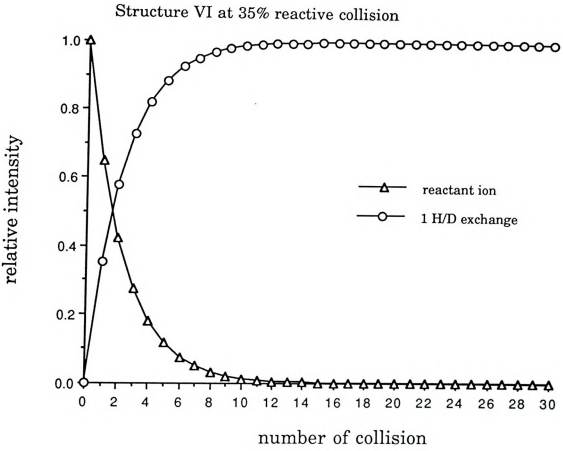

Figure A-50 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 35% reactive collision.

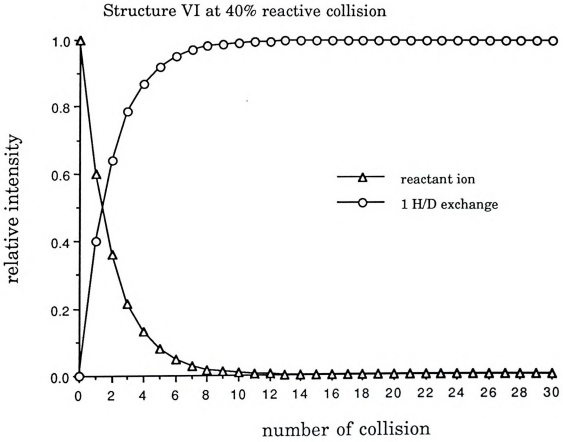

Figure A-51 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 40% reactive collision.

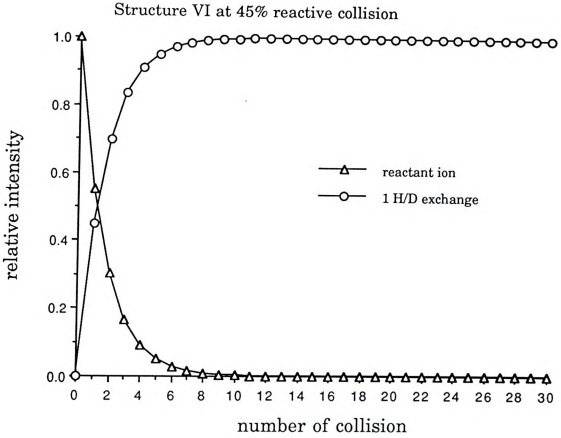

Figure A-52 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 45% reactive collision.

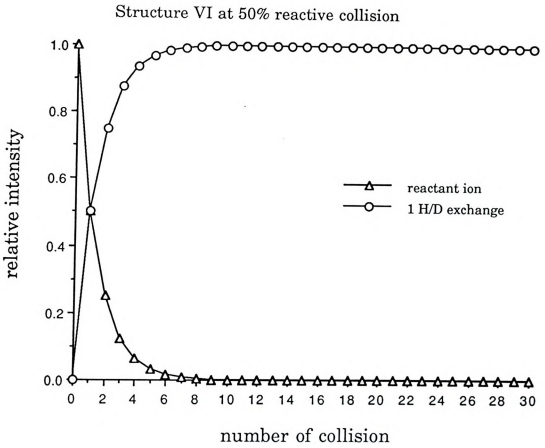

Figure A-53 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 50% reactive collision.

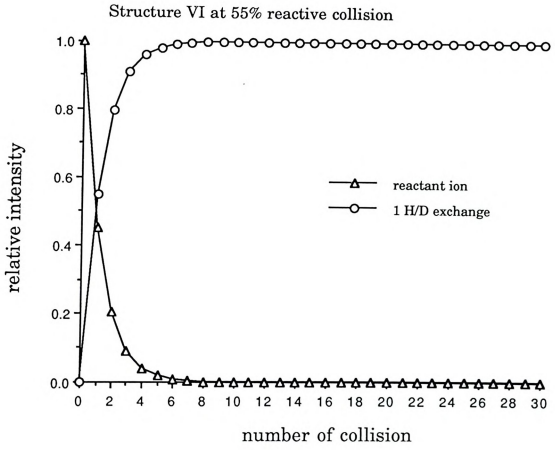

Figure A-54 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 55% reactive collision.

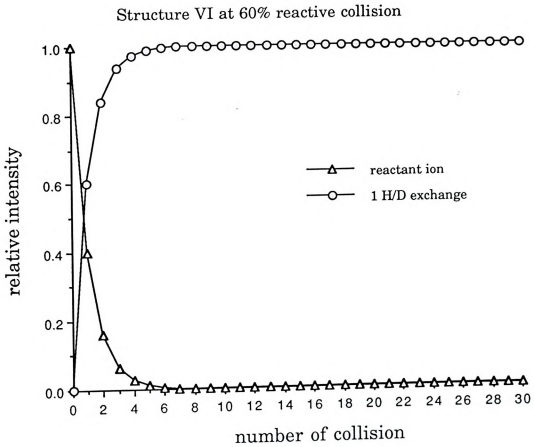

Figure A-55 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 60% reactive collision.

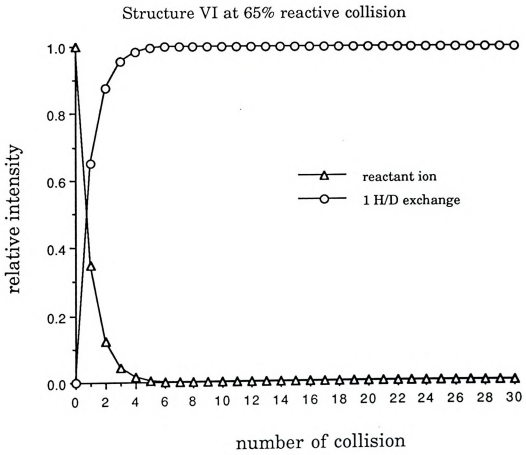

Figure A-56 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 65% reactive collision.

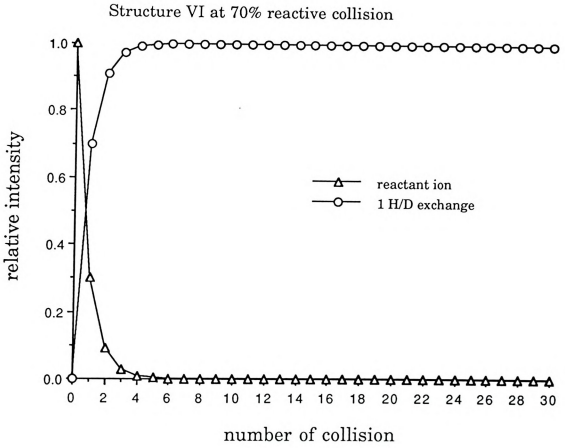

Figure A-57 A simulated collision-product plot for the structure VI of p-dichlorobenzene based on an H/D exchange reactivity of 70% reactive collision.