Molecular products from the pyrolysis and oxidative pyrolysis of tyrosine

Joshua K. Kibet, Lavrent Khachatryan, Barry Dellinger

Department of Chemistry, Louisiana State University, Baton Rouge, LA 70803, USA

Abstract

The thermal degradation of tyrosine at a residence time of 0.2 s was conducted in a tubular flow reactor in flowing N₂ and 4% O₂ in N₂ for a total pyrolysis time of 3 min. The fractional pyrolysis technique, in which the same sample was heated continuously at each pyrolysis temperature, was applied. Thermal decomposition of tyrosine between 350 and 550 °C yielded predominantly phenolic compounds (phenol, p-cresol, and p-tyramine), while decomposition between 550 and 800 °C yielded hydrocarbons such as benzene, toluene, and ethyl benzene as the major reaction products. For the first time, the identification of p-tyramine, a precursor for the formation of p-tyramine and its degradation to phenol and p-cresol, and toxicological discussion of some of the harmful reaction products is also presented.

1. Introduction

Amino acids are the fundamental building blocks of proteins and have applications in the food, agricultural, cosmetics, and pharmaceutical industries. Pyrolytic processes are commonly used in their manufacture (Chiavari and Galletti, 1992; Wu et al., 2010). Consequently, pyrolysis studies of amino acids is critical because formation of mutagenic and carcinogenic products in pyrolyses of proteinaceous foods is a health concern in the fields of food processing, preservation, and safety (Djilas et al., 1994). The pyrolytic behavior of common amino acids has been investigated in detail but despite this effort, potentially diagnostic fragments bearing polar functional groups, e.g. COOH, NH, OH frequently escape detection because of thermal instability, low volatility, and high adsorptivity (Chiavari et al., 2001).

Tyrosine is a large amino acid found in substantial quantities in many animal and plant proteins (Mahoney et al., 2007) as well in tobacco (Sharma et al., 2006) and is the metabolic precursor for the synthesis of catecholamine neuron transmitters, dopamine and norepinephrine in the central and peripheral nervous system (Neri et al., 1995; Mahoney et al., 2007). Amino acids in the form of proteins are the main source of nitrogen in wood (Hansson et al., 2004). Most biomass materials such as bagasse, straw, wood, and tobacco contain nitrogen which can be converted to environmentally harmful products such as hydrogen cyanide, phenols, nitrosamines, furans, and aromatic compounds (Chiavari et al., 2001; Li et al., 2008). The thermodynamic end products of amino acids are simple inorganic compounds (CO₂, H₂O, NH₃, and CO); however, more complex chemicals are formed as by-products (amines, nitriles, amides, and hydrocarbons) (Chiavari et al., 2001; Li et al., 2006; Ren et al., 2011; Ren and Zhao, 2012). Compared to other bio-polymeric materials (i.e., cellulose, lignin, and pectin), tyrosine undergoes decomposition at fairly high temperatures (> 300 °C).

The health consequences resulting from consumption of tobacco products has been blamed on the production of toxic molecular products as well as free radicals during tobacco burning. Radicals...
produced during combustion of tobacco are very reactive and capable of causing oxidative stress, cancers, and reproductive health diseases. For example, tyrosyl radical has been reported from the fractional pyrolysis of bright tobacco (Maskos et al., 2008). Tyrosyl radical may originate either from the decomposition of protein-containing tyrosine residues or from free tyrosine molecules (Maskos et al., 2008).

Our investigations of the thermal degradation of tyrosine avoid many experimental pitfalls by using a continuous flow reactor system, with collection of the reactor effluent with an in-line GC–MS at the head of the GC column at −60 °C. Our efforts were directed toward identification of some intermediate products, e.g., the existence of p-tyramine as a product has been expected, but its actual formation is controversial (Li et al., 2008). A p-tyramine is reported to displace amines such as dopamine from its vesicles and is a false neuron transmitter as well as a metabolic product of catecholamine biosynthesis (Kitahama et al., 2005). It is one of the amines present in the human brain thought to influence behavior (Hiroi et al., 1998).

This study will demonstrate the formation of p-tyramine and its subsequent degradation to toxically important pollutants, such as phenol, and p-cresol. A mechanism of p-tyramine formation and degradation from the thermal decomposition of tyrosine is presented for the first time.

2. Experimental section

2.1. Materials

Tyrosine (> 99%, purity) used in this study was purchased from sigma Aldrich (USA) and used without further treatment.

2.2. Fractional pyrolysis and the System for Thermal Diagnostic Studies (STDS)

The fractional pyrolysis technique was applied in this study and is an experimental procedure in which the same sample is continuously pyrolyzed at each pyrolysis temperature (Agblevor et al., 2010). The fractional pyrolysis technique offers some advantages in comparison with conventional pyrolysis. First, only one loading of sample is used and can be heated multiple times. Secondly, it provides partial accumulation of any fraction and analysis for products in the gas phase as well as in the charred material. Thirdly, the intermediate neutral, but unstable products may be collected before they disappear in secondary processes.

The System for Thermal Diagnostic Studies (STDS) is a well-established technique for analysis of a wide range of organic materials including biomass materials (Rubey and Grant, 1988). The STDS contains various units: the reactor compartment which houses a high temperature furnace and the reactor, the temperature control console which controls the pyrolysis temperature within a temperature gradient of ±5 °C, the injection port, the cryogenic trap, and the in-line detection system. The experimental details for STDS are given elsewhere (Rubey and Carnes, 1985; Rubey and Grant, 1988).

2.3. Pyrolysis and oxidative pyrolysis

The thermal degradation of tyrosine was investigated in a tubular flow reactor over the temperature range of 200–800 °C at atmospheric pressure, typically in 50 °C increments under two reaction regimes (pyrolysis in N2 and oxidative pyrolysis in 4% O2 in N2) using the System for Thermal Diagnostic Studies (STDSs) (Rubey and Grant, 1988). The concentration of oxygen in the oxidative pyrolysis experiments was kept low to optimize the formation of oxidative products (Sharma et al., 2004a,b). The gas flow rate was designed to maintain a constant residence time of 0.2 s. 30 ± 0.2 mg of tyrosine were loaded into the tubular quartz reactor (0.3 cm i.d. × 17.7 cm, volume 1.25 mL) and held in place by quartz wool to avoid being swept by carrier gas flowing through the reactor. The reactor containing the sample was then placed inside an electrically heated furnace at a heating rate of 10 °C s⁻¹ for a total pyrolysis time of 3 min. The furnace was then turned off and the sample cooled with flowing N2 while exposing the reactor to a cooling fan. This method of thermolysis of tyrosine closely resembles the TGA technique wherein a sample boat is used to hold the sample in the reactor. The benefits of this technique are two-fold: (1) the sample is held intact in the reactor, and (2) the carrier gas flows uniformly through the sample during the entire analysis, resulting in highly reproducible analyses. Due to high flow rates, the contact time with charred material is short (0.2 s) to minimize secondary reactions.

2.4. GC–MS characterization of molecular products

The gas chromatography–mass spectroscopy (GC/MS) analysis of the pyrolysate was conducted with an Agilent 6890N gas chromatograph equipped with a 5973N mass selective detector (MSD) (Kibet et al., 2012) with an ion source in the electron impact (EI) mode at 24 eV to minimize extensive fragmentation (Sharma et al., 2004a,b). A detailed description of product characterization has been described elsewhere (Kibet et al., 2012). The compounds were identified using a NIST library (standard mass spectral data base developed by NIST) and confirmed by enhanced data library (mass spectral data base developed by Agilent technologies). This was to ensure the mass fragmentation pattern was well matched and enhance the confidence of compound identification. Furthermore, each of the compounds identified was thoroughly checked against literature data to ensure the correct reaction product was reported. In addition, pure compounds were used to compare chromatographic retention time as well as mass spectral patterns with those of the analyte. Accordingly, critical emphasis has been given to those products which can easily be correlated with the structure of the starting material (tyrosine). Representative GC–MS spectra from pyrolysis and oxidative pyrolysis of tyrosine are provided as supporting information. The experimental results were averaged from two replicates.

3. Results

3.1. Pyrolysis

This investigation revealed the principal products of tyrosine pyrolysis in an N2 atmosphere were phenolic compounds (phenol, p-cresol, and o-cresol), acetonitrile, benzaldehyde, ethylbenzene, and toluene. The maximum release of phenolic compounds and nitrogen containing compounds of low molecular weight occurred between 350 and 450 °C, while the maximum concentration of aromatic hydrocarbons and nitrogen containing compounds of high molecular weight occurred between 550 and 650 °C. Phenol and p-cresol reached maximum concentrations at about ~450 °C, Fig. 1A. Acetonitrile and propionitrile achieved a maximum concentration at about ~400 °C, (cf. Fig. 1B). Hydrogen cyanide was formed in significant amounts throughout the entire pyrolysis temperature range, (cf. Fig. 1B). The major hydrocarbon products: ethyl benzene, toluene, and benzene, peaked between ~550 and 650 °C (cf. Fig. 1C).

The hydrocarbon products are believed to be the result of thermal cracking and concerted rupture of the C–C chain followed by molecular growth to form aromatic species (Li et al., 2008). Gener-
ally, product profile concentrations first increased with increase in pyrolysis temperature, before falling off at high temperatures due to decomposition. Low molecular weight hydrocarbons (propene and 1-butene) yields were lowest (cf. Fig. 1D).

3.2. Oxidative pyrolysis

The principal products in this experiment were p-tyramine and phenol with a combined yield of over 80%. The formation of p-tyramine, with a maximum yield between 350 °C and 400 °C, (cf. Fig. 2A) was a very important observation. This compound has been known to have a low volatility and is not easily transported for detection. Li et al. (2008) concedes that the low volatility behavior of p-tyramine/4-(2-aminoethyl)phenol was responsible for eluding detection in their experiments. A p-tyramine should be an important signature of tyrosine pyrolysis formed from decarboxylation reactions. In our study, p-tyramine was observed in high concentration under oxidative pyrolysis conditions and low concentrations from pyrolysis (cf. Figs. 1A and 2A, respectively). Oxidative pyrolysis also formed compounds of biological interest: hydroquinone, benzofuran, dibenzofuran, and dibenzo-p-dioxin, as well as phenolic compounds (phenol, p-cresol, and o-cresol). The maximum release of hydroquinone, benzofuran, dibenzo-p-dioxin, phenol, p-cresol, benzonitrile, and benzaldoxime was generally between 400 and 450 °C (cf. Fig. 2A–C). Hydrogen cyanide was formed in low amounts throughout the entire temperature range, (cf. Fig. 2B). Benzene was the dominant product among the aromatic compounds, with a maximum concentration being observed at ~550 °C. Ethyl benzene, which was one of the main products in pyrolysis experiments, was formed in nearly trace amounts under oxidative pyrolysis conditions and exhibited a maximum yield at about 450 °C (cf. Fig. 2D). Oxidative pyrolysis of tyrosine did not result in significant formation of hydrocarbons.

The majority of products from the thermal degradation of tyrosine pass through a maximum. This observation is characteristic of the nature of fractional pyrolysis or oxidative pyrolysis. Three reasons are explored to explain this phenomenon: (1) exhaustion of reaction products from the sample, (2) degradation of some intermediates, for instance p-tyramine to other major reaction products such as p-cresol and phenol or (3) oxidation of products to CO, CO₂, and H₂O, etc. during oxidative pyrolysis.

4. Discussion

4.1. Decomposition/oxidation pathways for tyrosine

4.1.1. Initial decomposition

The mechanisms of thermal degradation of amino acids have been extensively studied (Simmonds et al., 1972; Ratcliff et al., 1974; Chiavari and Galletti, 1992; Basiuk, 1998; Basiuk and Douda, 1999, 2000; Li et al., 2008). Consequently, this investigation will focus primarily on the mechanistic pathways of new, major products from oxidative pyrolysis of tyrosine (p-tyramine, phenol, and p-cresol, Fig. 2A).

The main decomposition pathway for p-tyramine is likely a concerted, 4-center decarboxylation pathway. The decarboxylation of amino acids has been predicted using density functional theory, which found the decarboxylation channel for high molecular weight amino acids, including tyrosine, proceeds from the higher-energy anti carboxylic hydrogen conformer and involves the direct heterolytic loss of CO₂ accompanied by direct proton transfer (Li and Brill, 2003) (Fig. 3A). The calculated activation energy for direct decarboxylation in tyrosine was found to be 72 kcal mol⁻¹ in the absence of water (Li and Brill, 2003). While in the presence of water, the direct decarboxylation is catalyzed, and the calculated energy barrier drops to an average of 45 kcal mol⁻¹ (Li and Brill,
More recently, a statistical mechanical investigation (QM/MM) (Alexandrova and Jorgensen, 2011) indicated the most likely pathway for decomposition of amino acids in the presence of water occurs via direct decarboxylation, where CO₂ elimination is the first as well as the rate determining step (Alexandrova and Jorgensen, 2011). For instance, the computed free energy of activation for decarboxylation of glycine in the presence of water was found to be 45 kcal mol⁻¹, and the resultant rate constant was 10⁻¹² s⁻¹ at 25 °C (Alexandrova and Jorgensen, 2011), which was in agreement with experimental data (Li and Brill, 2003). The high activation energy and low pre-exponential factor for decarboxylation of amino acids results in a very slow process at room temperature which accelerates rapidly with increasing temperature.

\[ \text{p-cresol formation is probably initiated by the cleavage of bond # 5 with an estimated bond strength of} \ 72 \ \text{kcal mol}^{-1}. \] (Fig. 3B) (Davico et al., 1995; Wu et al., 1996; Lucarini et al., 2002; Blanksby and Ellison, 2003; Tumanov and Denisov, 2003; Gomes et al., 2004; Mulder et al., 2005). Cleavage of bond # 5 results in the formation of \( p \)-methylene phenolic radical, which forms \( p \)-cresol by donation of a hydrogen by a suitable organic donor, RH. (Rxs 1 and 2, 2003). More recently, a statistical mechanical investigation (QM/MM) (Alexandrova and Jorgensen, 2011) indicated the most likely pathway for decomposition of amino acids in the presence of water occurs via direct decarboxylation, where CO₂ elimination is the first as well as the rate determining step (Alexandrova and Jorgensen, 2011). For instance, the computed free energy of activation for decarboxylation of glycine in the presence of water was found to be 45 kcal mol⁻¹, and the resultant rate constant was 10⁻¹² s⁻¹ at 25 °C (Alexandrova and Jorgensen, 2011), which was in agreement with experimental data (Li and Brill, 2003). The high activation energy and low pre-exponential factor for decarboxylation of amino acids results in a very slow process at room temperature which accelerates rapidly with increasing temperature.

\[ \text{p-cresol formation is probably initiated by the cleavage of bond # 5 with an estimated bond strength of} \ 72 \ \text{kcal mol}^{-1}. \] (Fig. 3B) (Davico et al., 1995; Wu et al., 1996; Lucarini et al., 2002; Blanksby and Ellison, 2003; Tumanov and Denisov, 2003; Gomes et al., 2004; Mulder et al., 2005). Cleavage of bond # 5 results in the formation of \( p \)-methylene phenolic radical, which forms \( p \)-cresol by donation of a hydrogen by a suitable organic donor, RH. (Rxs 1 and 2, 2003). More recently, a statistical mechanical investigation (QM/MM) (Alexandrova and Jorgensen, 2011) indicated the most likely pathway for decomposition of amino acids in the presence of water occurs via direct decarboxylation, where CO₂ elimination is the first as well as the rate determining step (Alexandrova and Jorgensen, 2011). For instance, the computed free energy of activation for decarboxylation of glycine in the presence of water was found to be 45 kcal mol⁻¹, and the resultant rate constant was 10⁻¹² s⁻¹ at 25 °C (Alexandrova and Jorgensen, 2011), which was in agreement with experimental data (Li and Brill, 2003). The high activation energy and low pre-exponential factor for decarboxylation of amino acids results in a very slow process at room temperature which accelerates rapidly with increasing temperature.

\[ \text{p-cresol formation is probably initiated by the cleavage of bond # 5 with an estimated bond strength of} \ 72 \ \text{kcal mol}^{-1}. \] (Fig. 3B) (Davico et al., 1995; Wu et al., 1996; Lucarini et al., 2002; Blanksby and Ellison, 2003; Tumanov and Denisov, 2003; Gomes et al., 2004; Mulder et al., 2005). Cleavage of bond # 5 results in the formation of \( p \)-methylene phenolic radical, which forms \( p \)-cresol by donation of a hydrogen by a suitable organic donor, RH. (Rxs 1 and 2, 2003).
Typically, the activation energy for simple bond cleavage reactions, such as Rxn 1 is closely related to the enthalpy of reaction, 72 kcal mol$^{-1}$. This is close to the activation energy (72.6 kcal mol$^{-1}$) for decarboxylation of tyrosine which produces p-tyramine (Li and Brill, 2003).

The pre-exponential factors for decarboxylation reactions of different amino acids span a wide range, from $10^{10}$ s$^{-1}$ for Met-amino acid (methionine amino acid) to $10^{16}$ s$^{-1}$ for $\alpha$-Aib ($\alpha$-amino isobutyric) amino acid (Li and Brill, 2003). The pre-exponential values for a complex bond scission reactions are $10^{15} - 10^{17}$ s$^{-1}$ (Hiatt and Benson, 1972). The generally higher pre-exponential factor for bond scission makes the cleavage of bond # 4 (Fig. 3B) favorable over decarboxylation reactions for simple amino acids. As a result, p-cresol is one of the major products in tyrosine pyrolysis, Fig. 1A.

Phenol has been proposed to form from further decomposition of p-cresol (Li et al., 2008). Because the concentration of phenol is a little higher than that of p-cresol for both pyrolysis and oxidation experiments (cf. Figs. 1A and 2A), it would appear there is an additional mechanistic channel for the formation of phenol. For instance it may be the result of cleavage of bond # 4 (cf. Fig. 3A), with an estimated bond energy of 100 kcal mol$^{-1}$, leading to the formation of p-hydroxylated phenyl radical (and latter to phenol by abstraction of hydrogen from RH) or displacement of the entire side-chain by H: This pathway may be feasible, if we compare it with one of the important channels, deamination of amino acids (Alexandrova and Jorgensen, 2011) which occurs by participation of the bond # 6 with exactly the same bond energy as bond # 4, 100 kcal mol$^{-1}$ (cf. Fig. 3A).

### 4.1.2. The main channels of oxidative pyrolysis

Whereas p-tyramine is the major product during oxidative pyrolysis of tyrosine, it is formed in low concentrations in pyrolysis (cf. Figs. 22A and 4A). This phenomenon can be understood if a more favorable, free radical mechanism is considered in presence of oxygen. For instance the initiation pathway presented in Rxn 3, Scheme 1 (assuming the activation energy is equal to the bond dissociation energy $\sim$86.5 kcal mol$^{-1}$), $k_3 = 3.2 \times 10^{15}$ exp ($-86500$ cal mol$^{-1}$/RT) S$^{-1}$ can be accelerated significantly in presence of oxygen. Rxn 4 (activation energy is around 40–42 kcal mol$^{-1}$), $k_4 = 10 \times 10^{12}$ (10$^{10}$) exp ($-41500$ cal mol$^{-1}$/RT) cm$^3$ molecule$^{-1}$ S$^{-1}$ (Khachatryan et al., 2003). The concentration of oxygen in the system was 4%, the equivalent of $4.50 \times 10^{17}$ mol cm$^{-3}$ at 673 K. Therefore the ratio of the rates $R_4/R_3$ can be computed and found to be in favor of Rxn 4 ($R_4/R_3 \sim 1.0 \times 10^5$) at 673 K.

Reactions 3 and 4 (cf. Scheme 2) form tyrosyl radical (Maskos et al., 2008). It is remarkable that the observable amounts of tyrosyl radical (Tyr) were produced at $<380^\circ$C from tobacco pyrolysis (Maskos et al., 2008), which matches well with the maximum yields of tyramine (370°C) from tyrosine pyrolysis in this study, Fig. 1A. Further decarboxylation of Tyr$^\cdot$ favors formation of p-tyraminyl radical, Rxn 5, and subsequent formation of p-tyramine via Rxn 6, (cf. Scheme 3).

Statistical mechanical investigations (QM/MM) indicated that in presence of water the decarboxylation of amino acids is more facile, and the activation energy drops from 72 kcal mol$^{-1}$ (without water) to 45 kcal mol$^{-1}$ (Li and Brill, 2003; Alexandrova and Jorgensen, 2011). Hydroxyl radicals (OH), which are the major reactive species in oxidation processes (Benson, 1976), may have a similar effect towards decarboxylation as water. Furthermore the processes of formation Tyr$^\cdot$ will be accelerated in Rxn 4 (cf. Scheme 2) when OH are the main chain carrier radical (abstraction of hydrogen from phenolic hydroxyl group).

These reactions (4–6) are the main pathways that promote the formation of p-tyramine, which is the major product during oxidative pyrolysis. Therefore the rate of decomposition of tyrosine is enhanced under oxidative pyrolysis because the process occurs in a reactive regime, in the presence of O$_2$ and OH. For this reason, decarboxylation reaction will also proceed via a free radical mechanism in addition to a molecular process under pyrolysis. This explains why the concentration of p-tyramine for oxidative pyrolysis experiments is much higher than that from pyrolysis.

Mechanisms presented here as well some additional reactions for formation of major products from tyrosine pyrolysis/oxidative pyrolysis, based also on literature (Li et al., 2008), are summarized in Scheme 3.

There are two additional pathways for the formation of p-tyramine from the oxidative pyrolysis of tyrosine, Scheme 3 (parallel to the initiation reactions by molecular oxygen): (1) abstraction of H from the hydroxyl group of tyrosine by hydroxyl radical to form tyrosyl radical followed by CO$_2$ elimination to form tyraminyl radical and ultimately the formation of p-tyramine (2) abstraction of H from acidic group of tyrosine by hydroxyl radical to form intermediate I followed by CO$_2$ elimination leading to intermediate II and eventually p-tyramine.

As discussed above, p-tyramine is formed in relatively high yields in oxidative pyrolysis compared to a pyrolysis (cf. Figs. 1A and 2A). Consequently, p-tyramine is an important precursor for formation of many other reaction products identified in this study. Two pathways for the formation of phenol and p-cresol through p-tyramine and tyrosine are included in Scheme 3. The first pathway involves the scission of bond # 5 (cf. Fig. 3A) to form p-hydroxy methylene radical before forming p-cresol and eventually phenol (ref. Scheme 1, Rxn 2). The second channel proceeds from the scis-

![Scheme 1](image1.png)

**Scheme 1.** Chain reaction leading to the formation of p-methylene phenolic radical and ultimate formation of p-cresol.
sion of an amino group (cf. Scheme 3) to form \( p \)-hydroxy methylen radical and finally \( p \)-cresol and phenol as shown in Scheme 3.

The formation of other major products (by decreasing yields after \( p \)-tyramine, phenols and cresols) such as benzonitrile and acetonitrile are probably the result of dipeptide or polypeptide decomposition reactions. Dipeptide forming reactions occur readily because they are simple dehydration reactions which are usually enhanced by increase in temperature (Simmonds et al., 1972; Ratcliff et al., 1974; Sharma et al., 2004a,b; Li et al., 2008). Although the concentration of dipeptide is considered low, it is believed to play a critical role in the formation of many observed products of amino acid pyrolysis (Chiavari and Galletti, 1992; Sharma et al., 2004a,b). For instance the formation of acetonitrile from pyrolysis of tyrosine may proceed via the decomposition of cyclic dipeptides (Sharma et al., 2004a,b). This channel involves a molecular process, and a free radical mechanism in which acetonitrile is eventually formed from dehydration of acetamide.

4.1.3. New classes of compounds not reported in literature

Oxidative pyrolysis of tyrosine yielded other important compounds of biological interest: hydroquinone, \( p \)-benzoquinone, benzofuran, dibenzofuran, and dibenzo-\( p \)-dioxin. The main precursor for formation of hydroquinone and ultimately \( p \)-benzoquinone is \( p \)-cresol (cf. Scheme 4). An OH radical displaces the methyl in \( p \)-cresol, yielding hydroquinone. Subsequently, \( p \)-benzoquinone formation is initiated via endothermic dissociation of a phenoxyl-hydrogen (\( \Delta H_{\text{rxn}} = 81.3 \text{ kcal mol}^{-1} \)) or \( H^+ \) abstraction by ‘OH to form \( p \)-semiquinone radical (Lucarini et al., 2002; McFerrin et al., 2009). Subsequent loss of phenoxyl-hydrogen by unimolecular decomposition (\( \Delta H_{\text{rxn}} = 87 \text{ kcal mol}^{-1} \)) (Mulder et al., 2005) or abstraction (\( \Delta H_{\text{rxn}} = 40 \text{ kcal} \)) (Wiater et al., 2000) by \( \text{OH} \) results in the formation of \( p \)-benzoquinone.

The formation of dibenzo-\( p \)-dioxin and dibenzofuran from oxidative pyrolysis of tyrosine has captured our attention because of the health impacts of the chlorinated analogues of these compounds (Dellinger et al., 2008; Zheng et al., 2008). Although these compounds are reported extensively in literature, never before have they been documented during the combustion of amino acids. Hydroxyl radical is believed to play a critical role during oxidative pyrolysis of tyrosine and influences the reaction products observed. The precursor for these compounds is phenol.

When subjected to heat, phenol forms both dibenzofuran (Wiater et al., 2000) and dibenzo-\( p \)-dioxin (Evans and Dellinger, 2003a,b; Khachatryan et al., 2003; Evans and Dellinger, 2005a,b). The formation pathway for dibenzo-\( p \)-dioxin/dibenzofuran proceeds via free radical mechanisms either through radical–molecule or radical–radical pathways (Louw and Ahonkhai, 2002; Evans and Dellinger, 2003a,b; Khachatryan et al., 2003; Asatryan et al., 2005; Evans and
Dellinger, 2005a,b). In the radical–molecule pathway, the enol form of the phenoxy radical displaces a ring hydrogen of the phenol molecule to form a hydroxyl biphenyl ether intermediate, Rxn 7 (Scheme 4), followed by ring closure and ultimately the formation of dibenzo-

\[ \text{Scheme 3. Mechanistic pathways for formation of major phenolic compounds from decomposition of tyrosine.} \]

4.1.4. Toxicological concerns for major reaction products from the thermal degradation of tyrosine

The toxicology of molecular products from pyrolysis of tyrosine is a subject of environmental concern (Kibet et al., 2012). Phenol and cresol can be oxidized by DNA to produce resonance stabilized persistent free radicals (PFRs) with long lifetimes, which can cause extensive cellular damage, oxidative stress, tumors, and cancer (Dellinger et al., 2000, 2007; Smith and Hansch, 2000; Dellinger et al., 2001). The phenolic compounds found in biomass burning and cigarette smoke as well as in this work are also considered toxic and include phenol, substituted phenols (p-cresol, and o-cresol), and hydroquinone (Selassie et al., 1999; Smith and Hansch, 2000). Phenol affects liver enzymes, lungs, kidneys, cardiovascular system, and may attack the nervous system (Talhout et al., 2011). Following H atom abstraction from the phenol hydroxyl group, the resultant phenoxy radical exhibits some electron-deficient character (Dellinger et al., 2007) which would be stabilized by electron-donating substituents such as amino, methoxy, and methyl groups, imparting longer life times (Smith and Hansch, 2000). Substituted phenoxy radicals (e.g. p-cresol, o-cresol, p-tyramine, etc.) bearing electron-donating substituents would therefore be expected to be more toxic because they are more stable and have longer lifetimes (Dellinger et al., 2000, 2007; Smith and Hansch, 2000; Dellinger...
et al., 2001). Additionally, phenoxy radicals are precursors for formation of dibenzo-p-dioxin/dibenzo-furan, which are easily chlorinated in the presence of a redox-active transition metal such as copper or iron to form polychlorinated dibenzo-p-dioxin/dibenzo-furan (PCDD/F) (Louw and Ahonkhai, 2002; Evans and Dellinger, 2003a,b; Khachatryan et al., 2003; Kibet et al., 2012). Clearly, the formation of compounds of biological interest including, hydroquinone, p-benzoquinone, benzofuran, dibenzofuran and dibenzo-p-dioxin suggests the thermal degradation of tyrosine is an important contributor to degradation in air quality when biomass is burned.

5. Conclusion

A detailed kinetic analysis for the formation of p-tyramine as a principal product in the thermal degradation of tyrosine has thoroughly been discussed. For the first time, compounds of biological interest including dibenzo-p-dioxin, dibenzofuran, hydroquinone and p-benzoquinone are reported from oxidative pyrolysis of tyrosine. We believe these toxic compounds are formed during heat treatment of proteinaceous foods, tobacco burning and biomass burning, and are precursors for oxidative stress and cancers.

Acknowledgements

The authors appreciate financial support from Reynolds’ Tobacco Company.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.chemosphere.2013.01.071.

References
