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Amino Acid Degradations Produced by Lipid Oxidation Products

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ABSTRACT

Differently to amino acid degradations produced by carbohydrate-derived reactive carbonyls, amino acid degradations produced by lipid oxidation products are lesser known in spite of being lipid oxidation a major source of reactive carbonyls in food. This article analyzes the conversion of amino acids into Strecker aldehydes, α-keto acids, and amines produced by lipid-derived free radicals and carbonyl compounds, as well as the role of lipid oxidation products on the reactions suffered by these compounds: the formation of Strecker aldehydes and other aldehydes from α-keto acids; the formation of Strecker aldehydes and olefins from amines; the formation of shorter aldehydes from Strecker aldehydes; and the addition reactions suffered by the olefins produced from the amines. The relationships among all these reactions and the effect of reaction conditions on them are discussed. This knowledge should contribute to better control food processing in order to favor the formation of desirable beneficial compounds and to inhibit the production of compounds with deleterious properties.

Keywords acrylamide, carbonyl–amine reactions, flavor formation, lipid oxidation, Maillard reaction, Strecker aldehydes, styrene

INTRODUCTION

Amino acid catabolism by microorganisms in foods has long received a considerable attention mostly as a consequence of its repercussions on food quality and safety (Ardö, 2006; Fernández and Zúñiga, 2006). Thus, it was early known that both pleasant and unpleasant aroma compounds were produced as a consequence of amino acid degradations (Urbach, 1993). Moreover, amino acids are well known precursors of compounds such as biogenic amines that may compromise the health of consumers (Marcobal et al., 2012).

In addition to the amino acid catabolism produced by microorganisms, amino acids can also be degraded chemically as a consequence of food processing. These reactions play a major role in the formation of taste and flavor compounds as well as in the loss of essential amino acids and the formation of toxicological suspect compounds. Thus, nowadays is widely recognized that the formation of Strecker aldehydes from their parent amino acids is initiated by α-dicarbonyl compounds in the course of the Maillard reaction (Granvogl et al., 2012). Moreover, the formation of amines as a consequence of Maillard reaction has also been demonstrated (Granvogl et al, 2006), and this degradation seems to play a role in the formation of acrylamide as a consequence of the thermal processing in food products (Taeymans et al., 2004; Zhang et al., 2009).

Differently to amino acid degradations produced by carbohydrate-derived reactive carbonyls, amino acid degradations produced by lipid oxidation products are lesser known in spite of being lipid oxidation a recognized source of reactive carbonyls in foods (Cheng, 2010; Choe and Min, 2006). Furthermore, chemical reactivity of lipid-derived reactive carbonyls is very similar to that of carbohydrate-derived reactive

carbonyls, and both types of carbonyl compounds usually produce very similar nonenzymatic browning reactions (Zamora and Hidalgo, 2005).

In an attempt to update the diverse studies carried out in recent years in this field, this article reviews the amino acid degradations produced by oxidized lipids as well as the reactions in which the compounds produced in these degradations are involved.

STRECKER DEGRADATION OF AMINO ACIDS PRODUCED BY LIPID OXIDATION PRODUCTS

The Strecker degradation of amino acids is produced by a wide range of lipid oxidation products, including primary, secondary, and tertiary products of lipid oxidation (Zamora et al., 2008). However, the reaction is produced to different extents depending on the lipid oxidation product involved, which is likely a consequence of both the influence of the lipid-derived carbonyl on the reaction pathway and the existence of parallel reactions in which these oxidized lipids may be involved and that compete with the Strecker reaction.

The Strecker degradation of amino acids by oxidized lipids was firstly described in 2004 for the formation of phenylacetaldehyde by phenylalanine degradation in the presence of epoxyalkenals (Hidalgo and Zamora, 2004), and later extended to other lipid-derived reactive carbonyls (Hidalgo et al., 2005; Zamora et al., 2007). A general mechanism for the Strecker degradation of amino acids produced by lipid-derived reactive carbonyls is shown in Figure 1. The first step of the reaction is the formation of an imine (imine A) between the amino group of the amino acid and the carbonyl group of the lipid. This imine suffers then an electronic rearrangement as a consequence of the exit of the proton of the carboxylic group. This electronic rearrangement produces the

loss of carbon dioxide and the formation of a new imine (imine B), which is the origin of the the Strecker aldehyde after hydrolysis.

At the same time that the amino acid is degraded, the lipid-derived carbonyl is transformed into a new derivative in which the carbonyl group has been converted into an amino group (Figure 2). However, this compound is not usually stable and evolves either into polymers or into more stable heterocyclic structures. Thus, the formation of 2-alkylpyridines is usually observed in the reaction of epoxyalkenals and alkadienals with amino acids (Hidalgo and Zamora, 2004; Zamora et al., 2007). On the contrary, 2-alkylpyrroles were identified in the reaction of hydroxyalkenals and oxoalkenals with amino acids (Hidalgo et al., 2005; Zamora et al., unpublished).

Although short-chain volatile compounds have received much more attention than long-chain lipid oxidation products, these last compounds are usually produced to a much higher extent than short-chain derivatives. Thus, for example, the Fe(II)/Fe(III)-catalyzed linoleic acid hydroperoxide decomposition produced volatiles comprising less than 5 mol% of total products (Grosch, 1976). For that reason, the ability of long-chain lipid oxidation products, in which the carbonyl group is usually a ketone group, to convert amino acids into their corresponding aldehydes was also investigated. Two different lipid oxidation products were assayed: conjugated epoxyoxooctadecenoates (Zamora et al., 2005) and conjugated oxooctadecadienoates (Zamora et al. 2007) [general structures for these compounds are shown in Figure 2]. According to the obtained results, the ability of aldehydes and ketones for degrading amino acids was similar in most experiments and differences found might be more related to the different solubility of the lipid oxidation products assayed than to a different reactivity. In this case, the reaction also followed a reaction pathway similar to that shown in Figure 1. The only difference was that the carbonyl group was in the middle of a chain, and,

107 therefore, the heterocyclic derivative that was produced after the reaction was slightly
108 different. Thus, both epoxyoxooctadecenoates and oxooctadecadienoates were
109 converted in fatty acid chains containing a pyridine ring in the middle of the chain
110 (Figure 2). Consequently, this reaction provides a route for the formation of long chain
111 heterocyclic fatty acid derivatives in food products. Although heterocyclic fatty acid
112 derivatives are not usually determined, their formation (or their syntheses) has been
113 objective of diverse studies (see, for example, Fürmeier and Metzger, 2003; Gardner et
114 al., 2000; Hidalgo and Zamora, 1995).

115 Differently to secondary and tertiary lipid oxidation products, the primary products
116 of lipid oxidation (the hydroperoxides) do not have a carbonyl group. However, they are
117 easily decomposed to produce both free radicals and carbonyl compounds (Gardner,
118 1989). The carbonyl compounds produced in this decomposition are the secondary and
119 tertiary lipid oxidation products discussed above, which produce the Strecker
120 degradation of the amino acids by the mechanism indicated in Figure 1. However, this
121 cannot be the unique mechanism by which lipid hydroperoxides degrade amino acids
122 because carbonyl compounds are only a part of the secondary and tertiary lipid
123 oxidation products formed (Gardner, 1989; Reis and Spickett, 2012), and the yields
124 obtained for the formation of Strecker aldehydes by reaction with the hydroperoxides is
125 quite similar to the yields obtained with the lipid-derived reactive carbonyls (Zamora et
126 al., 2008). Therefore, hydroperoxides should have an alternative way to produce the
127 Strecker degradation of amino acids in addition to the reaction of the carbonyls
128 produced as a consequence of their decomposition. This alternative pathway is likely a
129 free radical degradation analogous to that collected in Figure 3. Lipid hydroperoxides
130 are decomposed by means of different agents, including heat and metal traces, to
131 produce the corresponding alkoxyl radicals (Girotti, 1998; Kasaikina et al., 2006; Pazos,

et al., 2008; Ueda et al., 1996). These radicals can then abstract a proton from the amino acid so that the radicals are converted into hydroxy acids (usual products in these reactions) at the same time that an amino acid radical is produced. This radical (a oxygen-centered radical has been drawn, but other radicals are also possible) suffers then an electronic rearrangement with the loss of carbon dioxide and the exit of a proton, which may react with a new hydroxyl radical to produce the corresponding hydroxy acid. The product of this electronic rearrangement is an imine that is the responsible for the phenylacetaldehyde formation after hydrolysis. A pathway similar to this has been proposed for the degradation of amino acids observed by pyrolysis-GC/MS in the absence of carbohydrates (Chu and Yaylayan, 2008; Yaylayan and Keyhani, 2001).

CONVERSION OF AMINO ACIDS INTO α-KETO ACIDS BY LIPID OXIDATION PRODUCTS

As observed in Figure 1, the trigger for the electronic rearrangement of imine A, which is the key step for the Strecker degradation, is the exit of the carboxylic proton under the acid conditions at which the reaction is produced. However, the exit of the proton at the α-carbon of the amino acid would produce the same result without the loss of carbon dioxide. Zamora et al. (2006a) showed that this proton can leave as a function of the electronic effects of the substituents at the α-carbon. Thus, long-chain saturated amines, in which the α-carbon is joined to an amino group and an alkyl chain, were converted into carbonyl compounds only to a very low extent. However, the yield for benzylamine, in which the α-carbon is joined to an amino group and an aromatic ring, increased to 4.3%. Furthermore, when this carbon was trisubstituted, such as in 2-

phenylglycine methyl ester, in which the α-carbon is joined to an amino group, an aromatic ring, and a methoxycarbonyl group, the reaction yield increased to 49%.

Amino acids are usually trisubstituted at the α-carbon: they have the amino group, the carboxylic group, and the chain, which usually starts with a methylene group. Therefore, the proton at the α-carbon should leave with a relative easiness. In fact, when phenylalanine was incubated overnight with 4,5-epoxy-2-decenal at 37 ºC, the corresponding α-keto acid was produced to a higher extent than the Strecker aldehyde (Zamora et al., 2006b). The reaction pathway for this reaction is shown in Figure 4. It is analogous to the reaction pathway for Strecker aldehyde formation shown in Figure 1. The only difference is the exit of the proton at the α-carbon in the place of the exit of the carboxylic proton. Therefore, the electronic rearrangement of imine A produces the imine C in the place of the imine B shown in Figure 1. The later hydrolysis of this imine C is the origin of the α-keto acid. At the same time that the α-keto acid is produced, the oxidized lipid is modified analogously to the observed for the Strecker aldehyde formation, and the formation of the corresponding pyridine and pyrrole derivatives shown in Figure 2 should also be expected as byproducts of the α-keto acid formation.

Although this degradation has been observed for different lipid-derived carbonyls and a similar degradation of methyl 2-phenylglycine by ribose has also been described (Zamora et al., 2006a), no reports have appeared to date regarding to the ability of lipid hydroperoxides to produce this degradation. In the case of hydroperoxides, the ratio between Strecker aldehydes and α-keto acids will likely depend on the different ability of alkoxyl radicals to convert amino acids into either heteroatom- or carbon-centered radicals, respectively.

CONVERSION OF AMINO ACIDS INTO SHORTER ALDEHYDES BY LIPID OXIDATION PRODUCTS

When phenylalanine is heated at high temperature in the presence of air and lipid-derived reactive carbonyls, the formation of benzaldehyde is usually observed in addition to the formation of phenylacetaldehyde [see, for example, Zamora et al. (2007)]. The formation of this aldehyde, having two carbons less than the original amino acid is likely to take place from the corresponding α-keto acid or Strecker aldehyde by a reaction pathway similar to that shown in Figure 5. In the presence of lipid-derived reactive carbonyls, amino acids are converted into either α-keto acids, which have the same number of carbons than the original amino acid, or Strecker aldehydes, which have one carbon less than the original amino acid. Because both of these compounds have a carbonyl carbon, they can be in equilibrium, to a certain extent, with their corresponding tautomeric forms. The oxidation of these forms would produce the corresponding peroxides which, after degradation, would be converted into the corresponding aldehyde having two carbons less than the initial α-amino acid. A proof for this mechanism, which was proposed by Chu and Yaylayan (2008) for the conversion of phenylacetaldehyde into benzaldehyde, was obtained by Smit et al. (2004) when they observed that the conversion of phenylpyruvic acid into benzaldehyde was accompanied by the formation of oxalic acid. The proposal of the oxidation of enols by single electron transfer to molecular oxygen as the key step in this process is based on the findings of Tokunaga et al. (2005) and Kaneda et al. (1982) that observed additions of molecular oxygen to enol ethers and enamines, respectively. Chu and Yaylayan (2008) proposed a different pathway for benzaldehyde formation from phenylpyruvic acid that did not require the presence of oxygen. However, the amount of benzaldehyde produced in the degradation of phenylalanine by lipid oxidation products

under nonoxidative conditions is very low in comparison to the benzaldehyde produced at high temperature and in the presence of air (Zamora et al. unpublished results).

Depending on the reaction conditions, phenylpyruvic acid can also be decarboxylated to phenylacetaldehyde (Zamora et al., 2006b), which can be later oxidized to benzaldehyde according to the reaction pathway collected in Figure 5. However, this mechanism should be easily distinguished from the direct conversion of phenylpyruvic acid into benzaldehyde described in Figure 5 because oxalic acid cannot be produced if the two carbons are not lost in one step.

CONVERSION OF AMINO ACIDS INTO AMINES BY LIPID OXIDATION PRODUCTS

In addition to the above described conversion of the α-amino group of amino acids into a carbonyl group, lipid-derived reactive carbonyls are also able to decarboxylate amino acids converting them into the corresponding amines, which are usually known as biogenic amines when they are produced as a consequence of the action of microorganisms (Shalaby et al., 1996). This reaction has lately received a considerable attention because of its potential implication in the formation of acrylamide as a consequence of asparagine degradation in the presence of lipid-derived reactive carbonyls (Capuano et al., 2010; Zamora and Hidalgo, 2008). It was first hypothesized to be produced as an intermediate step in the conversion of phenylalanine into styrene (Hidalgo and Zamora, 2007). Later, it was confirmed in the conversions of both asparagine into 3-aminopropionamide (Hidalgo et al., 2010a) and phenylalanine into β-phenylethylamine (Zamora et al., 2012a) by action of lipid oxidation products.

The reaction pathway proposed for this reaction is shown in Figure 6. As observed, the reaction is initiated, analogously to the reactions described above, with the

227 formation of the imine A between the amino group of the amino acid and the carbonyl
228 group of the lipid oxidation product. However, this time neither the exit of the hydrogen
229 at the carboxylic group (Figure 1) nor the exit of the hydrogen at the α-carbon (Figure
230 4) are produced. In this case, the electronic rearrangement of imine A produces a 5-
231 oxazolidinone intermediate, which was first identified in model systems of amino acids
232 and simple aldehydes (Aurelio et al., 2003; Tsuge at al., 1987) and, then, in mixtures
233 with aldehydes having a second functional group such as glycolaldehyde (Chu and
234 Yaylayan, 2009). The importance of the 5-oxazolidinone formation in this pathway lies
235 in its ability to decarboxylate and form a non-stabilized azomethine ylide, which is
236 prone to undergo a 1,2-prototropic shift and form two isomeric imines (imines B and D
237 in the figure). As can be observed, the formed imine B is identical to imine B produced
238 in the pathway shown in Figure 1. Therefore, its later hydrolysis produces the Strecker
239 aldehyde and the modified lipid oxidation product shown in Figure 2. However, imine
240 D is not produced by any of the mechanisms described previously. The difference
241 between this imine and the imines formed by previous mechanisms (Figures 1, 3, and 4)
242 is that the carbon-nitrogen double bond is not at the side of the amino acid residue but at
243 the other side. This structural difference is the responsible for the formation of the
244 amine of the amino acid after hydrolysis. At the same time that the amine is produced,
245 the initial lipid-derived reactive carbonyl is recovered. Therefore, by means of this
246 reaction pathway, amines and Strecker aldehydes of amino acids are produced
247 simultaneously, although the relative amounts at which both compounds are produced
248 are not necessarily the same and it will depend on the amino acid and the lipid-derived
249 reactive carbonyl involved, and the reaction conditions (Zamora et al., unpublished
250 results).

This mechanism was confirmed by using deuterated water in the reaction (Hidalgo et al, 2010a). The produced amines were mono- and di-deuterated at the α-carbon of the amino group. The deuteration of the monodeuterated amine occurred during the 1,2-prototropic shift in the azomethine ylide. The second deuterium in the dideuterated amine was introduced at the beginning of the reaction by keto-enol tautomerism in the original amino acid.

CONVERSION OF THE AMINES DERIVED FROM AMINO ACIDS INTO STRECKER ALDEHYDES BY LIPID OXIDATION PRODUCTS

The amines produced in the degradation of amino acids can suffer later reactions in the presence of lipid-derived reactive carbonyls. One of them is their conversion into the corresponding Strecker aldehydes. This reaction, which was observed in the conversion of β-phenylethylamine into phenylacetaldehyde in the presence of different lipid oxidation products (Zamora et al, 2012b), seems to takes place according to the reaction pathway shown in Figure 7. The initial step is the formation of the corresponding imine between the amino group of the amine and the carbonyl group of the oxidized lipid. This imine is the same imine D of Figure 6. The conjugated system of the lipid-derived carbonyl and the electronic effects of the substituents at the α-carbon of the amine favor an electronic rearrangement that convert imine D into imine B, which is the same imine B formed in Figures 1 and 6. The hydrolysis of imine B is the origin of the Strecker aldehyde. By means of this reaction, the lipid oxidation product has been transformed into an amino derivative that, in most cases, is the indicated in Figure 2 (Zamora et al., 2012b).

The existence of this reaction suggests that imine D in Figure 6 can be converted into imine B to some extent in the course of the reaction. Therefore, the amine/aldehyde

ratio produced by pathway collected in Figure 6 is not only influenced by the conversion of the azomethine ylide into one or other imine but also by the conversion of imine D into imine B. To our best knowledge, the relative contribution of both reactions to the amounts of amines and aldehydes formed by amino acid degradation in the presence of lipid oxidation products has not been investigated at present.

CONVERSION OF THE AMINES INTO VINYLOGOUS DERIVATIVES OF AMINO ACIDS BY LIPID OXIDATION PRODUCTS

In addition to their conversion into Strecker aldehydes, the amines can also suffer an elimination reaction to be converted into olefins: the corresponding vinylogous derivatives of the amino acids. The role of oxidized lipids in this reaction was firstly observed in the conversion of phenylalanine into styrene (Hidalgo and Zamora, 2007), and, then, described in detail for the elimination of 3-aminopropionamide to produce acrylamide (Zamora et al., 2009).

The reaction takes place as shown in Figure 8. Alkylamines are usually eliminated through their conversion into quaternary ammonium salts followed by Hofmann elimination (Saunders and Cockerill, 1973). However, in the presence of the lipid-derived reactive carbonyl, the amine produces firstly the corresponding imine (imine D). This imine may be then converted into an iminium ion that suffer a milder elimination (Katritzky and El-Mouafy, 1982). This conversion is favored by the lipid because a reactive carbon in the lipid chain may react with the nitrogen of the imine to produce the corresponding iminium ion. Thus, when the reaction was carried out in the presence of alkadienals, the lipid was converted into 2-alkylpyridine, which facilitated the elimination of the amine.

298 This reaction pathway was confirmed by determining the activation energies of the
299 elimination reaction of amines, amines in the presence of lipid-derived reactive
300 carbonyls, and *N*-substituted alkylamines. Thus, the elimination reaction of *N*-
301 substituted alkylamines, which took place through iminium ions, had a similar
302 activation energy that the elimination reaction of primary amines in the presence of
303 lipid-derived reactive carbonyls (Zamora et al., 2009). On the other hand, the
304 elimination reaction of primary amines, which can only take place through the
305 formation of ammonium salts, had much higher activation energy. In addition, this
306 mechanism was also in agreement with the reactivities exhibited by the different
307 carbonyl compounds, which were related to the conjugation of the iminium ion.

308 ***THE ROLE OF LIPID OXIDATION PRODUCTS ON THE ADDITION OF***
309 ***NUCLEOPHILES TO THE VINYLOGOUS DERIVATIVES OF AMINO ACIDS***

310 The vinylogous derivatives of amino acids described in the previous section are not
311 stable compounds and can suffer the addition of nucleophiles. This reaction has recently
312 received a considerable attention because of its possible use in the elimination of
313 acrylamide (Adams et al., 2010; Claeys et al., 2005; Kim et al., 2005; Rydberg et al.,
314 2003; Salazar et al., 2012; Zamora et al., 2011a). Although compounds having either
315 amino or sulfhydryl groups are added to these olefins, there are significant differences
316 between both kinds of compounds.

317 The addition of amino compounds takes place as indicated in Figure 9. The
318 vinylogous derivative of the amino acid reacts very rapidly and easily with the amino
319 compound to produce the corresponding Michael adduct (adduct A). However, this
320 adduct still has a nucleophilic group and it can react with a new molecule of olefin to
321 produce a new adduct (adduct B). Nevertheless, adduct B is not usually produced under

usual reaction conditions because vinylogous derivatives of amino acids are produced to a much lower extent than the amount at which amino compounds are present (Zamora et al., 2010). Both additions are reversible and it is possible to recover the initial olefin just by heating the adduct (Zamora et al., 2010).

When lipid-derived reactive carbonyls are present, this Michael addition is inhibited, most likely as a consequence of the reaction of the carbonyl compound with the amino compound to produce the corresponding imine (imine E). This reaction avoids that the amino compound can react with the olefin. This competition was confirmed when cysteine and *N*-acetylcysteine were compared to determine their relative abilities to eliminate acrylamide. Although cysteine is much more effective than *N*-acetylcysteine, when a carbonyl compound was present, the effectiveness of both compounds was very similar (Zamora et al., 2011b).

Differently to the addition of amino compounds, the addition of compounds having a sulfhydryl group is not an equilibrium and the atmospheric oxygen plays a role. The reaction follows the reaction pathway shown in Figure 10.

In the absence of oxygen, thiols react very rapidly and efficiently with the olefin to produce the corresponding adduct (adduct C). This reaction has an activation energy that is lower than the determined for the addition of amino compounds (Hidalgo et al., 2010b). Therefore, it should be expected that the addition of thiols will occur much more easily than the addition of amino compounds.

When oxygen is present, thiols are also converted into the corresponding thiolyl radicals which can either polymerize or be added to the olefin to form a new radical that will continue the free radical chain. This alternative route was confirmed by inhibition of free radical reactions with antioxidants (Hidalgo et al., 2010b).

To our best knowledge, the effect of carbonyls in this reaction has not been studied so far. However, lipid-derived reactive carbonyls usually have a conjugated system that might suffer the addition of the thiol. The role that this alternative reaction may play in the removal of the olefins produced during amino acid degradation remains to be investigated.

COMPETITION AMONG THE DIFFERENT ROUTES OF AMINO ACID DEGRADATION PRODUCED BY LIPID OXIDATION PRODUCTS

As described above, amino acids can be degraded by a variety of pathways that produce many different products. However, all these routes are interconnected and the products of some reactions are frequently reactants of new reactions. Figure 11 shows schematically how all the routes discussed previously are interconnected. As can be observed, in the presence of lipid-derived reactive carbonyls, amino acids always produce the corresponding imine in the first step (imine A). This imine suffers then an electronic rearrangement to be converted into imines B, C, or D, depending on what is produced: the exit of the carboxylic proton, the exit of the proton at the α-carbon of the amino acid, or the formation of an intermediate 5-oxazolidinone, respectively. Nevertheless, the azomethine ylide formed by decarboxylation of the 5-oxazolidinone is also responsible for the formation of imine B, and imine D can be converted into imine B. The hydrolysis of imines B, C, and D produce the primary amino acid degradation products: Strecker aldehydes, α-keto acids, and amines, respectively. However, none of these primary amino acid degradation products is a final compound and they may suffer further reactions. Thus, α-keto acids can be both decarboxylated to produce the Strecker aldehyde and oxidized to the corresponding aldehyde having two carbons less than the initial amino acid. This oxidation product can also be produced by oxidation of the

Strecker aldehyde. Finally, the amine can be either converted into the Strecker aldehyde or suffer an elimination reaction to produce the corresponding vinylogous derivative of the amino acid. This last olefin is also not a final compound and may suffer the additions of nucleophiles. In the case of amino compounds, the main product that should be expected under usual reaction conditions is adduct A. In the case of thiols, both the formation of adduct C and, if the reaction is carried out under oxidative conditions, free radical reactions should be expected.

Although all these reactions can be produced simultaneously, some of them will be favored over others depending on the reaction conditions and the amino acids and lipid-derived reactive carbonyls involved. In addition, the relative proportions in which the different products are formed will play a major role on the quality of the food product because both food flavors and potentially toxic compounds are formed at the same time that essential amino acids are destroyed (Capuano and Fogliano, 2011; Jackson, 2009; van Boekel et al., 2010). Therefore, it is very important to know how some reactions could be favored over others in order to increase the amount of beneficial products formed and to reduce the amount of non-desirable products.

Activation Energy

Activation energy is defined as the minimum energy required to start a chemical reaction. Therefore, the comparison among the activation energies of the different reactions in Figure 11 may help to understand what reactions will be firstly produced. The activation energies determined for some of the reactions included in Figure 11 are collected in Table 1. As can be observed, both amino acids and carbonyls usually play a significant role in the activation energy of the reaction. For example, alkadienals are better than other lipid oxidation products for producing the conversion of phenylalanine

394 into phenylacetaldehyde. However, there is not any difference between alkadienals and
395 oxoalkenals for converting phenylethylamine into phenylacetaldehyde.

396 The role of the amino acid can be observed in the conversion of the amino acid into
397 the amine. As observed in Table 1 the activation energy for the conversion of
398 phenylalanine into phenylethylamine is much lower than the conversion of asparagine
399 into 3-aminopropionamide. Therefore, the decarboxylation of phenylalanine should be
400 expected to take place under softer reaction conditions than the decarboxylation of
401 asparagine, which is agreement with the strong heating conditions required for the
402 formation of acrylamide (Tareke et al., 2002).

403 In addition to the role of amino acids and lipid oxidation products, the different
404 routes shown in Figure 11 have different activation energies. Thus, the activation
405 energy of the conversion of phenylalanine into phenylacetaldehyde is slightly higher
406 than the conversion of phenylalanine into phenylpyruvic acid. However, the conversion
407 of phenylalanine into phenylethylamine seems to require a higher energy. This also
408 happens when two reactions are liked. Thus, higher activation energy is required to
409 convert asparagine into 3-aminopropionamide than to transform 3-aminopropionamide
410 into acrylamide. But the formation of the adduct between acrylamide and *N*-
411 acetylcysteine still has a lower activation energy. Therefore, from an energy activation
412 point of view, once the decarboxylation is produced, the reaction should finished in the
413 adduct if a nucleophile having a sulhydryl group is present. In practice, this is not
414 completely true, because reaction conditions also play a major role in these reactions.

415 ***Effect of pH***

416 Different studies have shown that formation of Strecker aldehydes, α-keto acids,
417 amines, and the conversion of amines into Strecker aldehydes are favored at acid pHs

(Hidalgo et al., 2005; 2010a; Zamora et al., 2006b; 2012a; 2012b). On the contrary, the conversion of 3-aminopropionamide into acrylamide or the addition of nucleophiles to acrylamide takes place better at neutral or slightly basic pHs (Hidalgo et al., 2010b; Zamora et al., 2009).

Effect of water activity

Water activity is also important. In particular, the elimination reaction to produce olefins from amines is very sensitive to water (Zamora et al., 2009). For that reason, the effect of water activity on the formation of acrylamide from asparagine (Hidalgo et al., 2009) is a compromise between the water activities required for the decarboxylation reaction (Hidalgo et al., 2010a) and for the elimination reaction (Zamora et al., 2009).

Effect of oxygen

The amount of oxygen also plays a major role on the products formed because free radical reactions are favored. For that reason, Strecker degradations are favored (Hidalgo and Zamora 2007). In addition, the presence of oxygen also favors the conversion of amines into Strecker aldehydes (Zamora et al., 2012b) or the formation of shorter aldehydes from Strecker aldehydes or α-keto acids (Zamora et al., unpublished). On the contrary, the formation of olefins is inhibited in the presence of oxygen (Hidalgo and Zamora, 2007; Hidalgo et al., 2009).

CONCLUSIONS

The above results show that amino acids are easily degraded by lipid oxidation products in a similar way to carbohydrates. According to the data published to date, this degradation produces three kinds of compounds in a first step: Strecker aldehydes, α-keto acids, and amines. However, they are not final compounds and these compounds

441 are involved in further reactions. Thus, α-keto acids are transformed into Strecker
442 aldehydes and other aldehydes having two carbons less than the original amino acid; the
443 amines are transformed into Strecker aldehydes and olefins; and the Strecker aldehydes
444 can be oxidized to shorter aldehydes. Finally, olefins are susceptible to suffer addition
445 of nucleophilic compounds. Although all these reactions are interconnected, it is
446 possible to modify the ratio among the different formed products as a function of the
447 oxidized lipid and the amino acid involved, and the reaction conditions (particularly,
448 pH, water activity, presence of oxygen, time, and temperature). By playing with these
449 tools it is possible to favor the formation of the desirable compounds produced in these
450 reactions and to decrease the amount of the undesired products that are also formed.
451 Further studies on how benefit/risk balance of amino acid degradations by lipid
452 oxidation products can be improved upon processing on actual food systems are needed.

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621

FIGURE LEGENDS

Figure 1. Proposed pathway for the conversion of amino acids into Strecker aldehydes in the presence of lipid-derived reactive carbonyls.

Figure 2. Transformations suffered by lipid-derived reactive carbonyls in the Strecker degradation of amino acids.

Figure 3. Proposed pathway for the free radical degradation of amino acids.

Figure 4. Proposed pathway for the conversion of amino acids into α-keto acids in the presence of lipid-derived reactive carbonyls.

Figure 5. Proposed pathways for the conversion of α-keto acids and Strecker aldehydes into shorter aldehydes under oxidative conditions.

Figure 6. Proposed pathway for the conversion of amino acids into amines and Strecker aldehydes in the presence of lipid-derived reactive carbonyls.

Figure 7. Proposed pathway for the conversion of amines into Strecker aldehydes in the presence of lipid-derived reactive carbonyls.

Figure 8. Proposed pathway for the conversion of amines into olefins in the presence of lipid-derived reactive carbonyls.

Figure 9. Reaction of olefins with amino compounds and role of lipid-derived reactive carbonyls in these reactions.

Figure 10. Reactions of olefins with thiols.

Figure 11. General scheme for amino acid degradations induced by lipid-derived reactive carbonyls.

Table 1 Activation energies of amino acid degradation reactions produced by lipid-derived reactive carbonyls[a]

	Lipid-derived reactive carbonyls					
reaction	none	Alkadienals	Epoxyalkenals	Oxoalkenals	Hydroxyalkenals	reference
Phe→PAC		28–38	58–59	55–64	67	Zamora et al., unpublished
Phe→PP			48	49–51		Zamora et al., unpublished
Phe→PEA		54				Zamora et al. 2012a
Asn→APA		81				Hidalgo et al., 2010a
PEA→PAC		55		54		Zamora et al., 2012b
APA→AA	100–110	41–61				Zamora et al., 2009
AA→AAGly	52					Zamora et al., 2010
AA→AAAC	30					Hidalgo et al., 2010b

[a]Values are given in kJ/mol. Abbreviations: AA, acrylamide; AAAC, adduct between acrylamide and *N*-acetylcysteine; AAGly, adduct between acrylamide and glycine; APA, 3-aminopropionamide; Asn, asparagine; PAC, phenylacetaldehyde; PEA, phenylethylamine; Phe, phenylalanine; PP, phenylpyruvic acid.

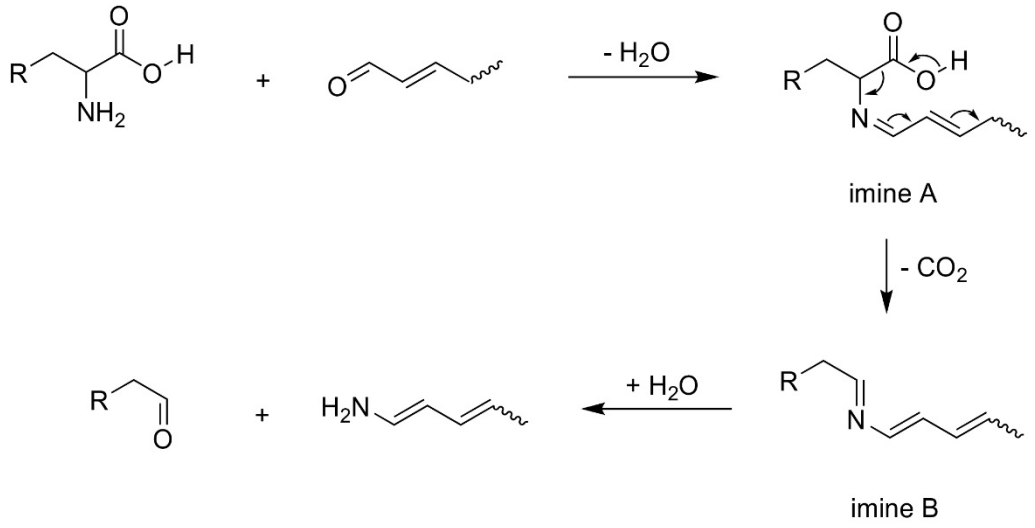

Figure 1

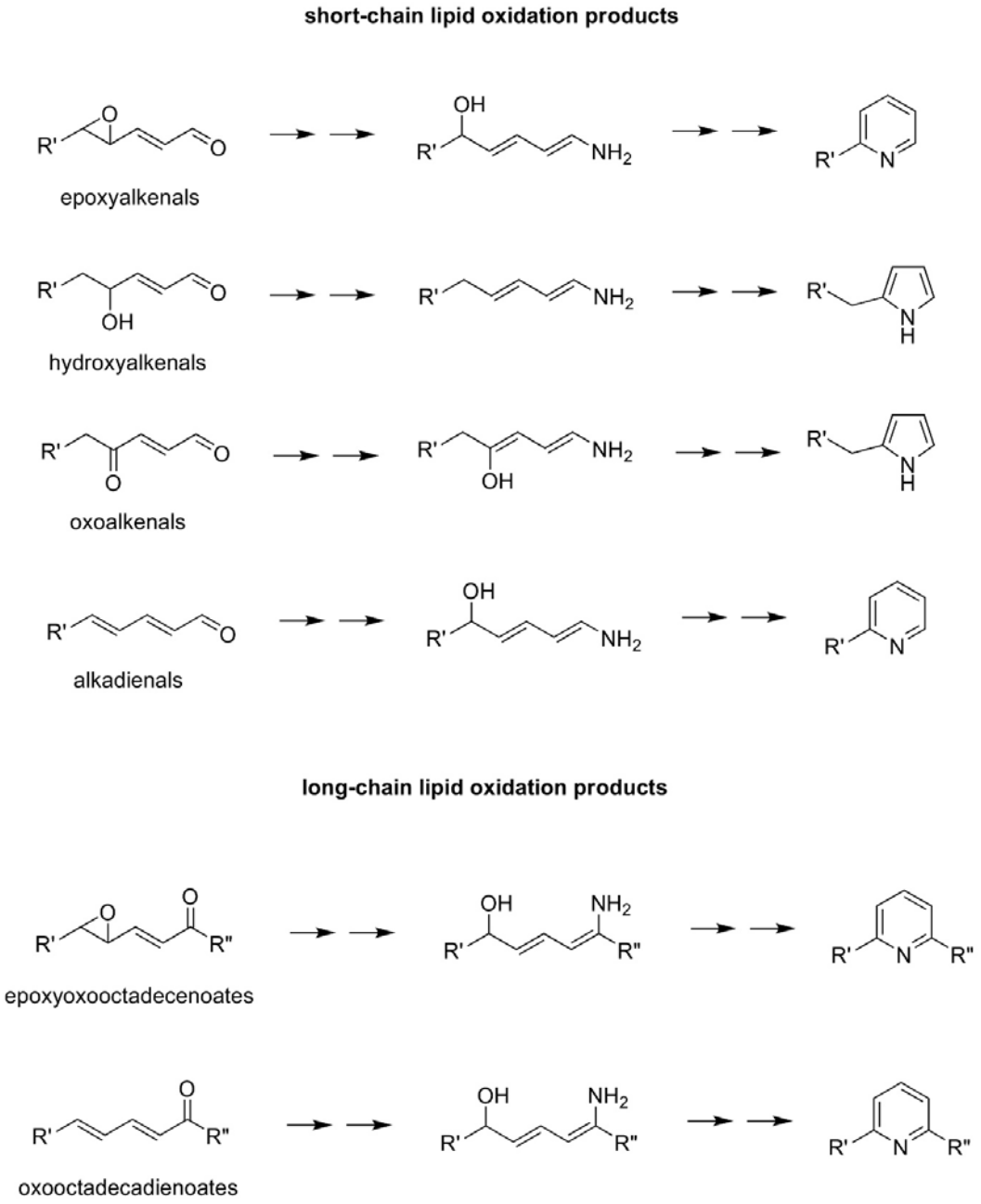
short-chain lipid oxidation products
epoxyalkenals
hydroxyalkenals
oxoalkenals
alkadienals
long-chain lipid oxidation products
epoxyoxooctadecenoates
oxooctadecadienoates

Figure 2

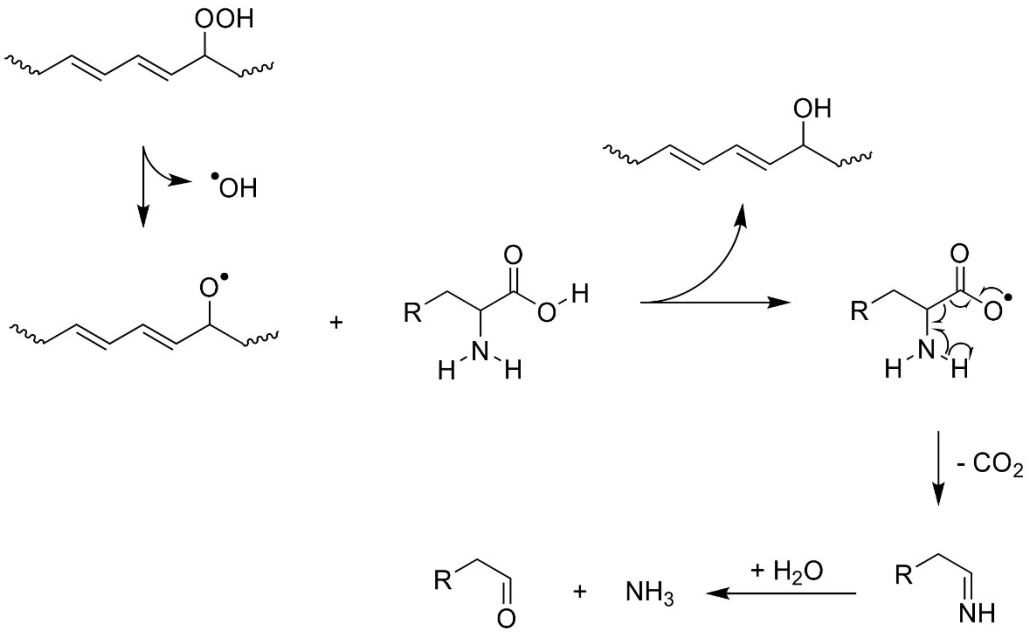

Figure 3

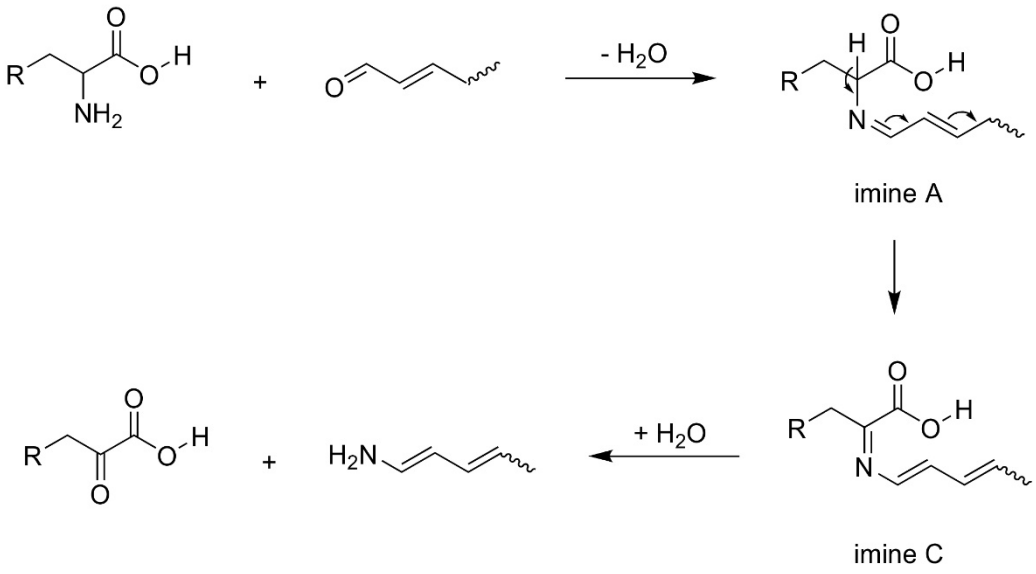

Figure 4

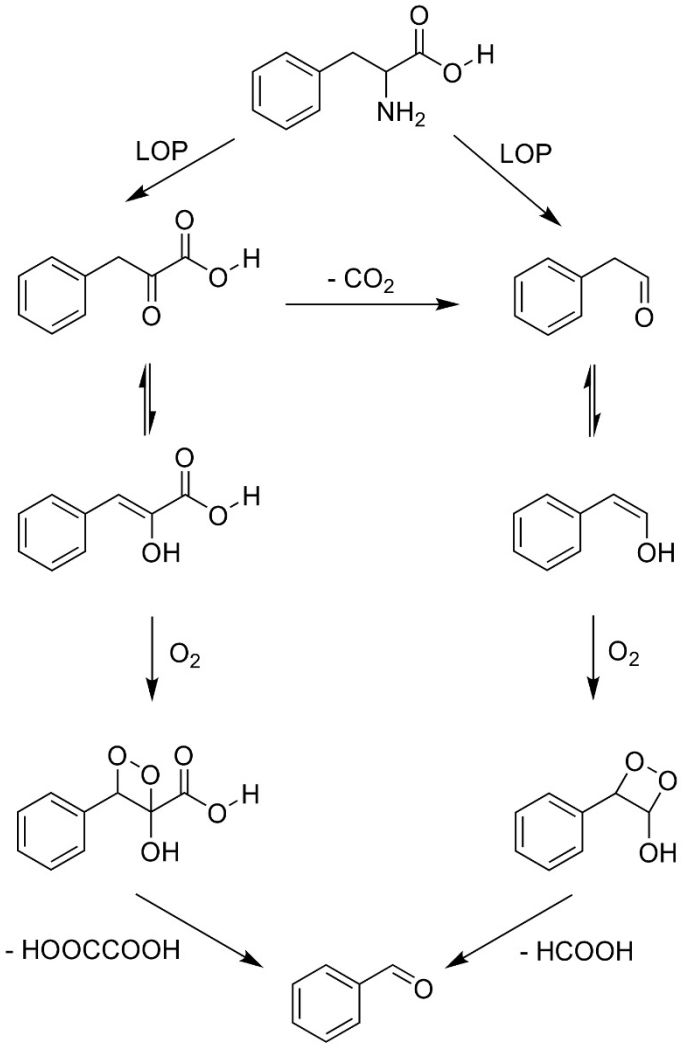

Figure 5

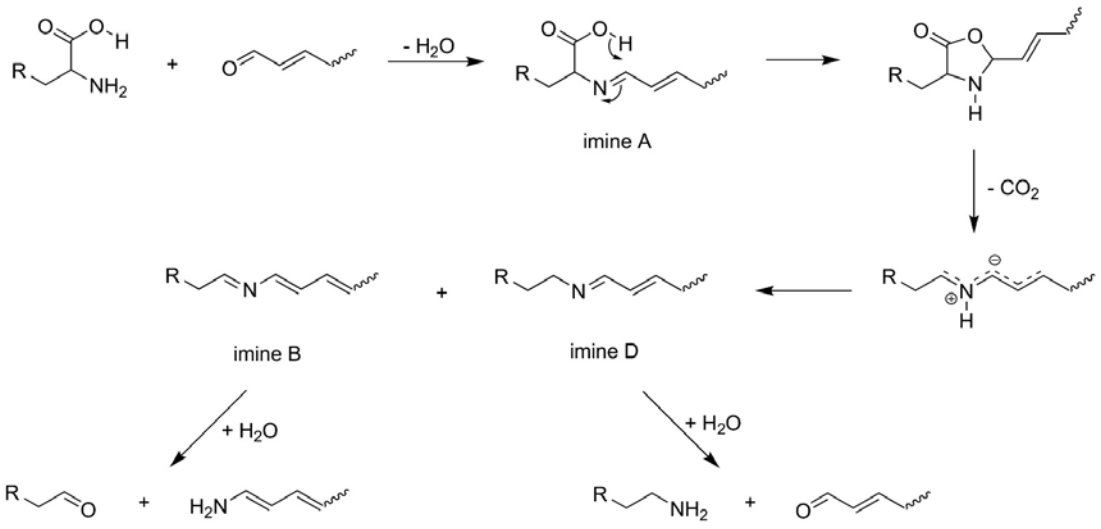

Figure 6

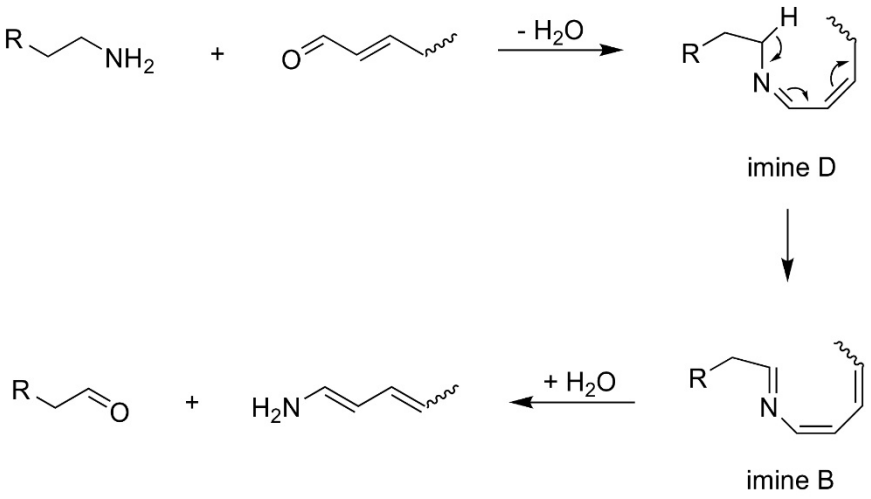

Figure 7

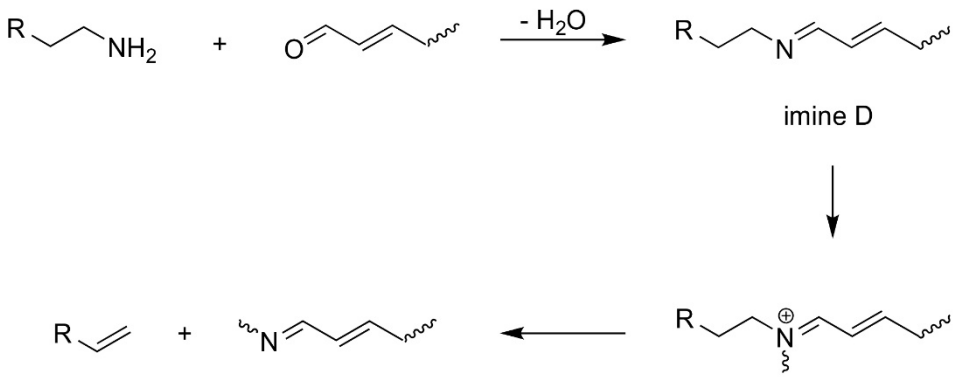

Figure 8

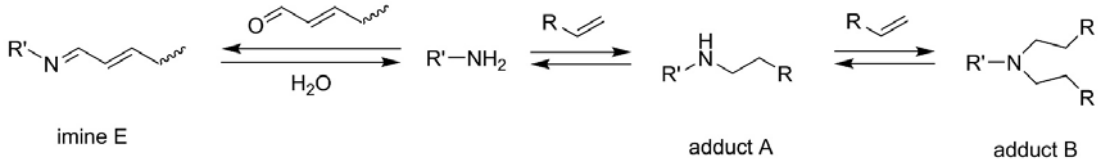

Figure 9

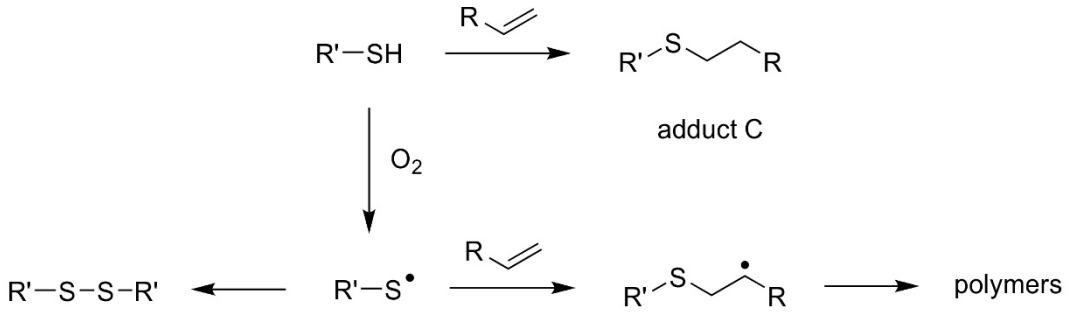

Figure 10

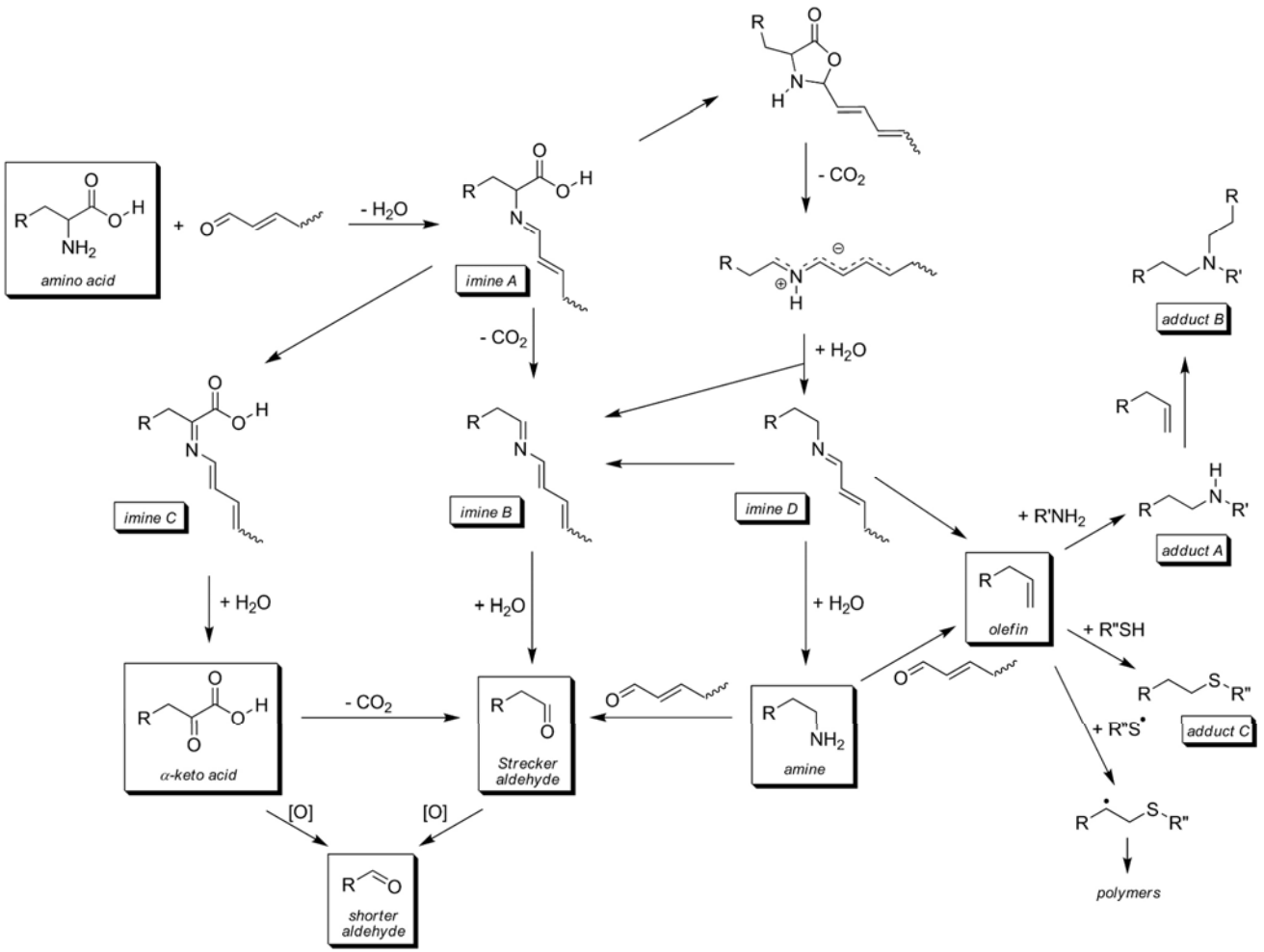
amino acid
- H2O
imine A
- CO2
imine B
imine C
imine D
+ H2O
α-keto acid
Strecker aldehyde
amine
olefin
shorter aldehyde
[O]
[O]
+ R'NH2
adduct A
adduct B
+ R"SH
adduct C
+ R"S•
polymers

Figure 11