

Automotive Clearcoat Damage Due to Oviposition of Dragonflies

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ABSTRACT: Automotive industries are increasingly interested in learning how to prolong the clearcoat resin lifetime and avoid its thermal, photochemical, and chemical degradation. While chemical degradation by acid rain has been well known since the beginning of the decade and the subject of many studies, chemical degradation of the automotive clearcoat by living organisms (except by bird droppings) is a newly recognized problem. In this work, we report the chemical degradation of the automotive clearcoat caused by oviposition of dragonflies. These insects, very common in Brazil, are attracted by the reflecting surface of cars exposed in the sun and lay eggs on them. We observed that the eggs, at the high temperatures (50–92°C) of the car surface, can cause damage similar to that of acid rain. In experiments on resin-coated plates, we excluded the involvement of H₂O₂—or hydroquinone—derived radicals, of enzyme-catalyzed hydrolysis, and of photosensitizer-induced damage. The damage was very similar, however, to that produced by the sulfur-containing amino acids, cysteine and cystine, at high temperature. Due to this similarity, and because the eggs are rich in sulfur amino acids, we propose a mechanism involving cysteine and cystine residues in the clearcoat damage. © 2000 John Wiley & Sons, Inc. *J Appl Polym Sci* 75: 1632–1639, 2000

Key words: dragonflies; oviposition; clearcoat; automotive resin degradation; acid rain

INTRODUCTION

Recently, several articles concerning the acid rain resistance of automotive clearcoat resins have been published.¹ Investigations of damage caused by acid rain have been reported since the beginning of this decade.² In recent studies, Lamers et al.³ reported that exposure of automobiles to sun-

light improves the acid etch resistance of acrylic/melamine clearcoats, while Wernstahl⁴ tried to correlate the loss of gloss with the breakage of a crosslinked structure. Resins used for clearcoat formulation are composed mainly of melamine moieties crosslinked with an acrylic polymer, which are susceptible to acid-catalyzed hydrolysis.⁵ White et al.⁶ performed experiments using an environmental smog chamber and reported ring-shaped fractured blisters on the topcoat surface in the spots that had been exposed to acidic chamber dews under heating.

Interestingly, quite similar damage has been claimed by car dealers and owners in Brazil to occur upon oviposition of dragonflies on a car sur-

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face during sunny days in the autumn. Like other aquatic insects, dragonflies lay eggs on any surface reflecting sunlight and, therefore, mimicking a water mirror, including cars parked in sunlit yards. These voracious insects often fly several kilometers to search for alate preys and therefore extend their damage to cars parked over long distances. The eggs may be laid singly or in masses and are embedded in a protein gel (oothecae), which provides mechanical protection, moisture, and food for the embryos.⁷ Hardening of the egg shell or chorion, called sclerotization, is initiated by an NAD(P)H-dependent "respiratory burst," which provides hydrogen peroxide as a cosubstrate for a phenol oxidase activity, which catalyzes the polymerization of tyrosine residues into a highly crosslinked macromolecular structure.

In this work, we studied the chemical nature of the damage caused by dragonfly eggs deposited on cars reflecting sunlight and, hence, heated to high temperatures (50–92°C), during the period of dragonfly oviposition (March–May) in Brazil. To resolve the possible mechanisms of automotive clearcoat resin degradation, we examine here the following working hypotheses: photochemical degradation, acid and enzymatic hydrolysis, and free-radical or microorganism attack. We then compare the damage caused by chemical agents and eggs on sample clearcoat plates both visually and by electron scanning microscopy or profilometry.

EXPERIMENTAL

Adults of *Miathyria* sp., *Tauriphila* sp., and *Erythemis* sp. (Odonata) were caught with large nets at noon at the margins of the Tietê River in the municipality of Novo Horizonte (State of São Paulo) during April and May. Most female dragonflies began oviposition expontaneously soon after being caught or were forced to lay eggs by holding their wings, which simulates a flicking motion that females use when they lay eggs onto water. The eggs, yellow or green, were collected and stored in deionized water at –20°C, conditions under which they do not undergo biological changes for years according to Kawasaki et al.⁸ Tests with either dragonfly eggs or chemicals were carried out with sample pieces made of aluminum plates covered with a complete painted system or just the clearcoat resin. Plates covered

by a 50- μ m layer of resin were prepared in the laboratories of Renner DuPont (São Paulo).

H₂SO₄ (95–97%), HCl (37%), HNO₃ (65%), formic acid (98–100%), acetic acid (100%), propionic acid (100%), and H₂O₂ (30%) were purchased from Merck (Darmstadt, Germany). Trichloroacetic acid (99%), L-tyrosine, L-cystine, L-cysteine, L-methionine, L-alanine, albumin, and uric acid, of the highest purity available, were obtained from Sigma (St. Louis, MO) and methanesulfonic acid was from Eastman (Rochester, NY).

The experiments were conducted at room temperature or in a thermostated oven WTB Binder FD 115. Amino acid analysis of the dragonfly eggs was performed with a Beckman 7300 high-performance chromatograph. Profilometry of the resin layer was done with a Dektak 3030 apparatus. Scanning Electron microscopy (SEM) micrographs of the plate surface were obtained using a Phillips SEM 515 instrument. The samples were covered with a thin layer of gold and 30 kV was used in the light source. Capillary electrophoresis analysis of the anions present in acid rain was performed with an Applied Biosystems/Perkin-Elmer equipment, with an UV/vis detector operated at 254 nm and 10 kV. The capillary column was of silica with a 0.75- μ m internal diameter and 70-cm length. To determine organic anions, an aqueous solution of sodium 3,5-dinitrobenzoate (10 mmol/L) with cetyltrimethylammonium bromide as a support electrolyte (0.20 mmol/L), pH 5.9, and 10 s of injection time was used. To determine inorganic anions, an aqueous solution of sodium chromate (10 mmol/L) with cetyltrimethylammonium bromide as a support electrolyte (0.10 mmol/L), pH 8.75, and 0.50 s of injection time was used.

Unless otherwise stated, a 500- μ L aliquot of acid or a H₂O₂ solution or 50 mg eggs were deposited on the surface of the plates, heated at a given temperature in the oven, and the chemical damage on the plate surface analyzed 20 min (80°C) or 24 h (25°C) thereafter. Plates with dragonfly eggs were also exposed directly to the sunlight. The plates were then allowed to reach room temperature, rinsed with plentiful water, and buffed with a chamois. Damage on the resin could be visually detected and then evaluated by profilometry and SEM. To determine the amino acid composition, yellow and green eggs were homogenized in a Potter-Elvehjem homogenizer with deionized water and hydrolyzed with HCl (6 mol/L) at 110°C for 24 h.

RESULTS AND DISCUSSION

Our first experimental approach to study the damage caused to the clearcoat by dragonfly eggs was to test, on sample plates, the effect of hydrogen peroxide (an expected intermediate of egg sclerotization)⁷ plus ferrous ions, the so-called Fenton reagent. This mixture combination is a source of hydroxyl radicals^{9,10} able to attack the methylene bridges between the O and N atoms of the resin crosslink structure. Surprisingly, peroxide-induced damage to the polymeric film was not observed, even when running the experiment at high temperature (80°C), either in the presence of surfactants to overcome the low resin wettability or when adding ascorbate to recycle the ferrous ions.¹¹ Another oxyradical source, 6-hydroxydopamine,¹² was tested on the clearcoat in order to verify if quinone and semiquinone intermediates, formed in the phenol oxidase reaction, could attack the resin with negative results.¹³ Tests with the tyrosine/tyrosine oxidase system also produced negative results. Noteworthy in this report is that the resin studied contains 9.7% nitrogen derivatives bearing methylene bridges —N—CH₂—N— and —N—CH₂—O—, which can transfer hydrogen atoms to strong oxidants such as oxyradicals and so are susceptible to cleavage with consequent weakening of the resin crosslinked structure. To circumvent this problem, the studied resin also contains 1% of a hindered amine light stabilizer (HALS), an antioxidant which protects the polymer against radical attack. This may explain the lack of effectiveness of the Fenton reagent and semiquinone in damaging the clearcoat by free-radical pathways.

Considering the presence of amide and ester moieties in the resin structure, tests were per-

Table I Composition of Rain Collected in a Municipality of the State of São Paulo on November 25, 1997, and Analyzed by Capillary Electrophoresis

Anions	[Anions] (ppm)
SO ₄ ²⁻	93.6
NO ₃ ⁻	5.1
Cl ⁻	6.1
HCOO ⁻	1.6
H ₃ CCOO ⁻	1.1
H ₃ CH ₂ CCOO ⁻	1.0

Table II Visual Evaluation of Damage to the Clearcoat Induced by Acid Exposure

Acid	[Acid] (%)	25°C ^a	80°C ^a
Sulfuric	0.10	+	++
	1.0	++	+++
	10	+++	++++
Methanesulfonic	0.10	+	++
	1.0	++	+++
	10	+++	++++
Trichloroacetic	0.10	+	++
	1.0	++	+++
	10	+++	++++
Hydrochloric	0.10	—	—
	1.0	—	+
	10	+	++
Nitric	0.10	+	++
	1.0	++	+++
	10	+++	++++
Uric	0.10	—	—
	1.0	—	—
	10	—	—
Formic	0.10	—	—
	1.0	—	—
	10	—	++
Acetic	0.10	—	—
	1.0	—	—
	10	—	++
Propionic	0.10	—	—
	1.0	—	—
	10	—	++

^a (—) no damage; (+) damage.

formed with catalytic (micromolar) amounts of model enzymes such as urease, peptidase, and esterase with the aim of revealing any possible hydrolytic role of dragonfly egg-contained enzymes. No damaging was observed on the clearcoat by addition of enzyme solutions buffered at their optimum pH and temperature.

Exposure of the clearcoat plates to dragonfly eggs under sun irradiation at room temperature (20–25°C), for 12 h, in order to test for any photodynamic action on the resin of possible photosensitizer dyes present in the eggs, also did not cause any detectable damage. In summary, these experiments ruled out the involvement of free-

radical attack, enzyme-catalyzed hydrolysis, and photosensitization in the chemical damage promoted by dragonfly eggs to the car surface.

The devastating effect of an episodic acid rain (pH 3.0) on cars parked in the yard of an automobile industry in the State of São Paulo led us to investigate the effect of acids on the car clearcoat, assuming that the protein gel in which the eggs are embedded might contain strong acid activity. Relevant in this respect are observations of Fornaro et al.¹⁴ that the average pH of rain in São Paulo during 1988–1990 was 4.5, with values ranging from 3.8 through 7.1. Anions of several inorganic and organic acids were found by capillary electrophoresis in the rain (Table I), among them anions of strong nonvolatile acids (e.g., H_2SO_4), strong volatile acids (e.g., HCl and HNO_3), and weak volatile acids (e.g., formic, acetic and propionic), with SO_4^{2-} and Cl^- the predominant anions. Also relevant to mention is that temperature measurements of the car surfaces, performed with a thermistor, revealed that at noon, during a sunny day, the temperature of cars is higher the darker the car is, ranging from 52°C (white) through 93°C (black).

The effect of several acids on the surface of the plates was then investigated at room temperature (20–25°C, 24 h exposure) or under controlled heating (80°C for 20 min). In both conditions, gradual evaporation of water, which increases the

acid concentration, can be observed. The higher the temperature and the acid concentration, the more dramatic was the damage to the clearcoat, as evaluated visually. The type of damage observed differed with the acid used: While formic, acetic, propionic (only at 80°C), nitric, and trichloroacetic acid immediately destroyed the resin film, H_2SO_4 , HCl , and methanesulfonic acids only bleached the surface initially and destroyed the resin film after a couple of hours. High temperature is crucial for damage to the resin film. The degree of chemical attack decreases in the order $\text{HNO}_3 > \text{CH}_3\text{SO}_3\text{H}$, $\text{Cl}_3\text{CCO}_2\text{H} > \text{H}_2\text{SO}_4 > \text{HCl} > \text{formic acid} > \text{acetic acid} \sim \text{propionic acid}$ (Table II). The damage increases with time if the attacked area is not immediately washed with water, which is in agreement with the literature.⁶

The apparent similarity of the damage caused by sulfuric acid and dragonfly eggs led us to suppose a common mechanism for both agents, that is, the damage caused by eggs might also be attributed to acid hydrolysis, promoted by strongly acidic sulfonic groups of proteoglycans possibly present in the protein gel. However, eggs freshly collected into deionized water give a pH between 6 and 7, which should exclude the hypothesis of general acid hydrolysis. Nevertheless, application of egg mass samples onto the plates at temperatures above 70°C resulted in considerable damage to the resin film; no damage is observed at room

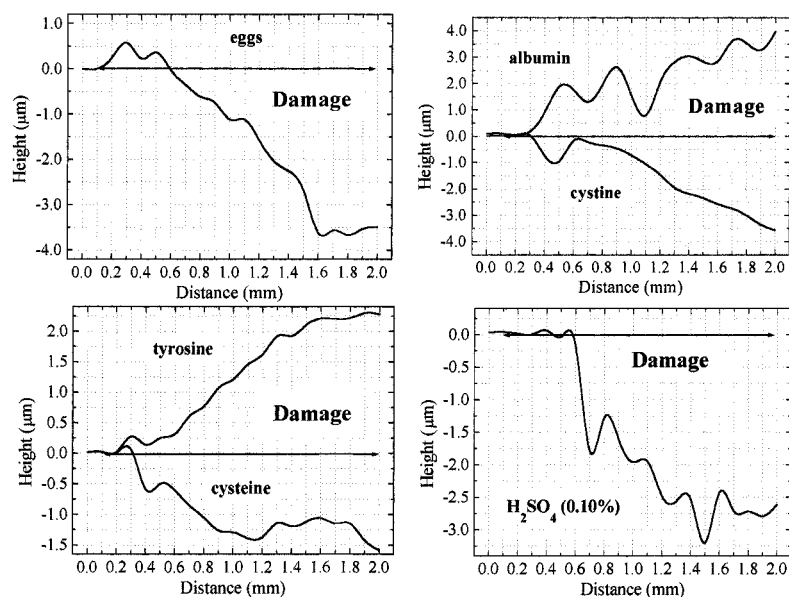


Figure 1 Profilometry of clearcoat attacked by dragonfly eggs and chemicals at 90°C for 3 h. The initial 100 μm scanned are due to the normal surface.

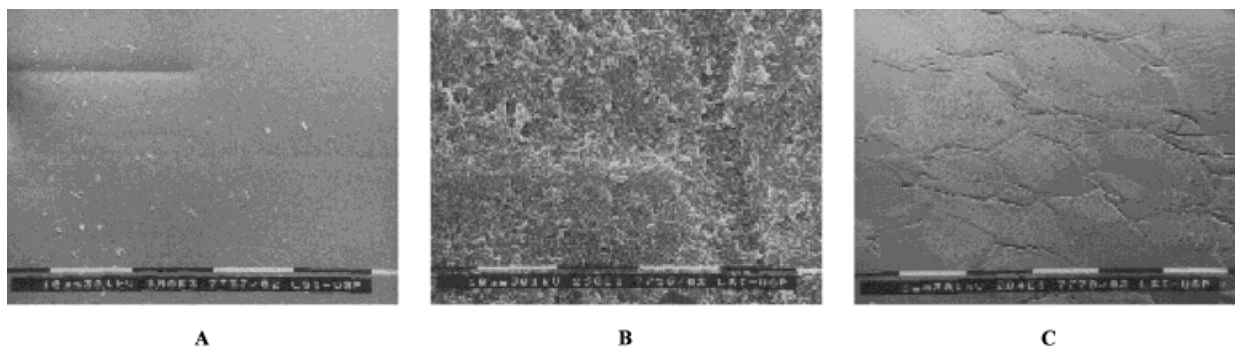


Figure 2 SEM of (A) the acrylic/melamine resin and after 3 h at 90°C in contact with (B) H₂SO₄ (0.10%) or (C) albumin. (A) and (B) are magnified 2500 times, and (C), 20.4 times.

temperature. At temperatures below 70°C, there appears to be damage, but it disappears if the surface is rinsed with water and heated again. We suppose that the damage “disappearance” is due to a reversible adsorption of water on the surface layer, which can cause changes in the refraction index and loss of gloss.

Considering that the sulfur content of the egg mass is relatively high (1.3%, determined by elemental analysis)—in agreement with the previous results of Kawasaki et al.,⁸ that the egg gel protein contains 1.3% cystine (determined in an automated amino acid analyzer) and 12 nmol of free SH groups per milligram of protein (determined by a literature procedure¹⁵), and that thiols are well-known efficient catalysts of ester hydrolysis, we challenged the clearcoat at 90°C with topical applications of cysteine (Cys-SH), cystine (Cys-SS-Cys), and, for comparison, methionine (Cys-S-CH₃) and alanine (a nonsulfur-containing amino acid). We also performed tests with ty-

rosine, the amino acid directly involved in the sclerotization process. Surprisingly, all amino acids were able to damage the clearcoat. Furthermore, bovine albumin was also found to damage the resin, but in a different way. As shown in Figure 1, the profiles of the attacked surface, using either eggs or chemicals, indicate deposition of material on the surface or corrosion of the resin. Profilometry analyses show that eggs, H₂SO₄, cysteine, and cystine corrode the resin film, leading to the formation of sulks of, respectively, 4.0-, 3.5-, 1.5-, and 3.5- μ m depth, while tyrosine and albumin adsorb on the surface, forming deposits of 2.5 and 4.0 μ m thickness, respectively (Fig. 1).

To visualize and better compare the damage caused by the chemicals, SEM analysis of the attacked region was performed. A normal resin has a smooth surface [Fig. 2(A)], which, in the presence of H₂SO₄ (0.10%), becomes rough and without gloss [Fig. 2(B)], probably due to break-

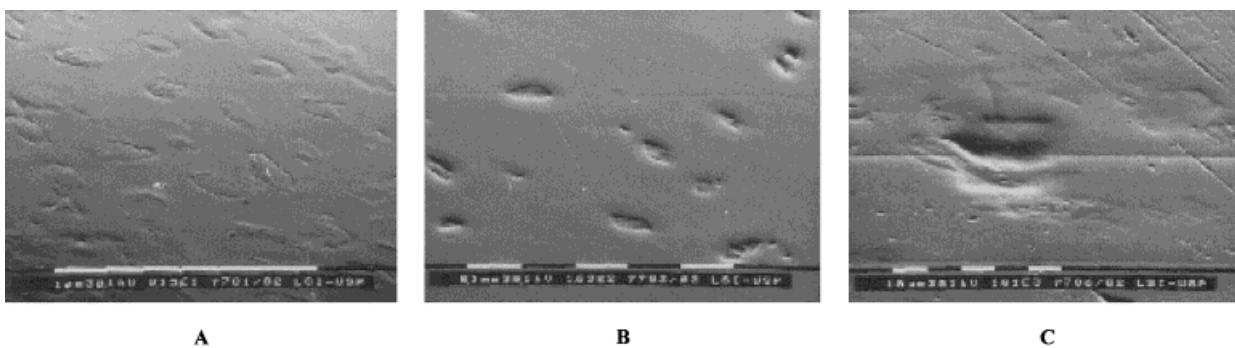
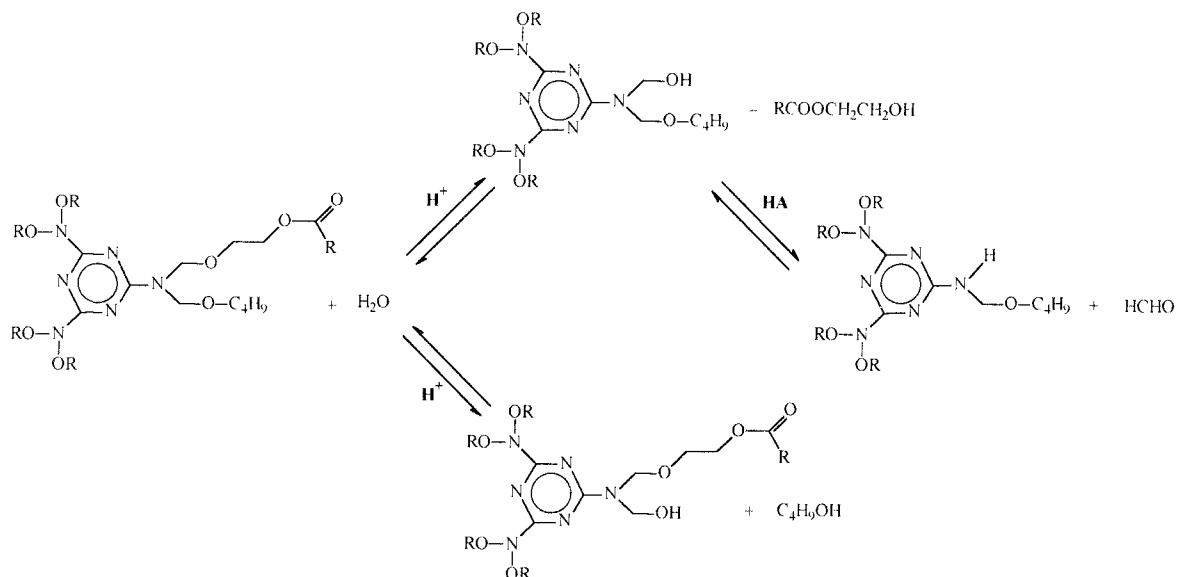


Figure 3 SEM of the acrylic/melamine resin after 3 h at 90°C in contact with (A) dragonfly eggs, (B) cysteine (10 mmol/L), and (C) cysteine (10 mmol/L). (A) and (B) are magnified 163 times, and (C), 101 times.



Scheme 1 Acid-catalyzed hydrolysis of an acrylic/melamine automotive resin proposed by Wernstahl.⁴

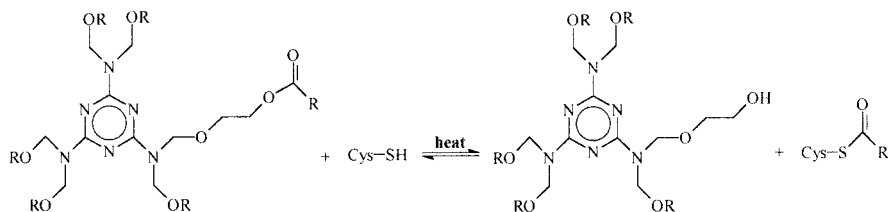
age of the methylene bridge between the O and N atoms in the crosslinked structure. In turn, bovine albumin adsorbs onto the surface, causing blisters as shown by the SEM study [Fig. 2(C)]. Eggs deposited manually or laid on the plate surface caused damage exactly in the place that they touched the surface. Egg-shaped boundaries on the resin surface can be clearly observed [Fig. 3(A)]. A quite similar effect is observed using a cysteine or cystine solution as the attack reagent [Fig. 3(B,C), respectively].

The initial studies with acids were actually done to elucidate the mechanism of acid rain degradation of automotive resins, but then realized to be similar to the mechanism of damage caused by dragonfly eggs. While the pH of egg-containing deionized water is neutral, this does not exclude elevated acidity on the contact area between eggs and resin. The egg proteins contain many acid and basic amino acids, which may act as catalysts

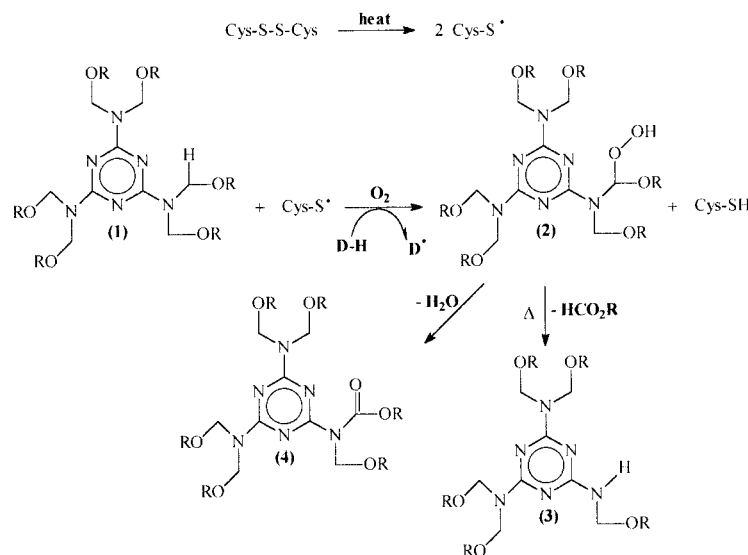
of acrylic/melamine resin hydrolysis⁴ (Scheme 1). Meanwhile, cysteine can promote nucleophilic attack on the ester linkages, leading to cleavage and thioester fragments (Scheme 2).

Hydrogen abstraction by a cysteinyl radical is another possible competitive decomposition mechanism (Scheme 3), since it is well known that disulfides are sensitive to S—S homolysis by heating.¹⁶ As this radical lives longer than do hydroxyl radicals, the probability of resin damage is greater. Eggs deposited on a hot resin surface in the presence of molecular oxygen could oxidize the melamine moiety (**1**), leading to hydroperoxide (**2**) formation (Scheme 3). Intermediate **2** could undergo further decomposition by a homolysis pathway, leading to amine **3** formation (preferential), or lose water to give an urethane structure (**4**) (side reaction).³

Another mechanism that could be formulated is acid hydrolysis by H₂S or sulfenic/sulfonic ac-



Scheme 2 Nucleophilic attack of an acrylic/melamine automotive resin by cysteine.



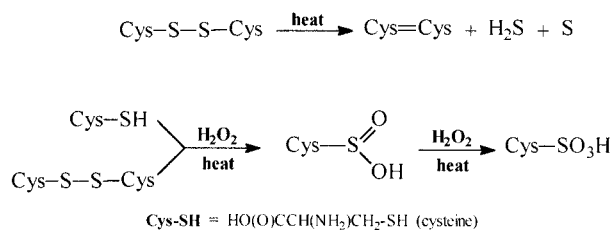
Scheme 3 Radical chain degradation mechanism of an acrylic/melamine automotive resin in the presence of cystine. After hydroperoxide (2) formation, the proposed mechanism follows Lamers et al.³

ids.¹⁶ The former can be produced by thermal degradation, because of the relative weakness of the disulfide bond (present in cystine residues), and the latter, by cysteine or cystine oxidation in the presence of hydrogen peroxide (from the respiratory burst) (Scheme 4).

CONCLUSIONS

In conclusion, independently of the proposed degradation mechanism, only a superficial layer of resin can be damaged by acids and dragonfly eggs under heating. However, once the outer layer is destroyed, the inner ones become more susceptible to further attack. It was found here that a resin undergoes degradation only at tempera-

tures above 70°C and after 3 h in the presence of the eggs, which reproduces the damage to cars observed in parking areas under the sun. SEM and profilometry were useful techniques to study the damage caused by the eggs and, while they do not characterize it at the molecular level, revealed that the damage caused by eggs is quite similar of that produced by cysteine and cystine (Fig. 3). This result, allied with the fact that the eggs contain a relatively high concentration of SH and S—S groups, make us believe that nucleophilic attack by cysteine residues (Scheme 2), attack by cysteinyl radicals (Scheme 3), and/or acid hydrolysis by H₂S and sulfinic/sulfonic acids (Scheme 4) are the main pathways leading to decomposition of the acrylic/melamine automotive resin under conditions of heat produced by sunlight.



Scheme 4 Formation of H₂S and sulfonic acids by thermal degradation of cystine and by hydrogen peroxide oxidation of cystine and cysteine.¹⁴

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REFERENCES

1. Ito, S.; Matoba, T.; Tominaga, A. *Prog Org Coat* 1997, 32, 97. Azuma, I.; Kosaka, N.; Iwamura, G.; Marutani, Y.; Uemura, H. *Prog Org Coat* 1997, 32, 1. Nakane, Y.; Ishidoya, M. *Prog Org Coat* 1997, 31, 113. Wernstahl, K. M.; Carlsson, B. *J Coat Technol* 1997, 69, 69. Lamers, P. H.; Johnston, B. K.; Tyger, W. H. *Polym Degrad Stab* 1997, 55, 309. Schmidt, H.; Fink, D. *JOCCA—Surf Coat Int* 1996, 79, 66. Arnold, J. C.; Li, J.; Isaac, D. H. *J Mater Process Technol* 1996, 56, 126.
2. Wolff, G. T.; Rodgers, W. R.; Collins, D. C.; Verma, M. H.; Wong, C. A. *J Air Waste Mgmt Assoc* 1990, 40, 1638.
3. Lamers, P. H.; Johnston, B. K.; Tyger, W. H. *Polym Degrad Stab* 1997, 55, 309.
4. Wernstahl, K. M. *Polym Degrad Stab* 1996, 54, 57.
5. Bauer, D. R. *J Appl Polym Sci* 1982, 27, 3651.
6. White, D. F.; Fornes, R. E.; Gilbert, R. D.; Speer, J. A. *J Appl Polym Sci* 1993, 50, 541.
7. Needham, J. G.; Westfall, M. J., Jr. *Dragonflies of North America*; University of California: Los Angeles, 1955.
8. Kawasaki, H.; Sato, H.; Suzuki, M. *Insect Biochem* 1974, 4, 99.
9. Walling, C. *J Am Chem Soc* 1975, 8, 125.
10. Halliwell, B.; Gutteridge, J. M. C. *Free Radicals in Biology and Medicine*, 2nd ed.; Clarendon: Oxford, 1989; p 543.
11. Quinlan, G. J.; Halliwell, B.; Moorhouse, C. P.; Gutteridge, J. M. C. *Biochem Biophys Acta* 1988, 962, 196.
12. Monteiro, H. P.; Winterbourn, C. C. *Biochem Pharmacol* 1989, 38, 4177.
13. Aeschbach, R.; Amadò, R.; Neukon, H. *Biochem Biophys Acta* 1976, 439, 292.
14. Fornaro, A.; Isolani, P. C.; Gutz, I. G. R. *Atmos Environ* 1993, 27, 307.
15. Murphy, M. E.; Kehrer, J. P. *Biochem J* 1989, 260, 359.
16. Cremllyn, R. J. *An Introduction to Organosulfur Chemistry*; Wiley: Chichester, UK, 1996.