Food Chemistry **26** (1987) 271–282

ESR Study of a 2,6-Diterbutyl Hydroxytoluene Analog Incorporated into Rigid PVC: Effects of the Immersion in Various Aqueous Media

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(Received 17 December 1986; revised version received 18 February 1987; accepted 10 March 1987)

ABSTRACT

Study by electron spin resonance (ESR) of N (3,5-diterbutyl-4 hydroxybenzyl) 4 amino 2,2,6,6-tetramethylpiperidinyl oxy: 'BHT-amino TEMPO', a paramagnetic analog of BHT, incorporated into the usual compound of rigid polyvinyl chloride (PVC) revealed that after contact of the labelled polymer with aqueous media (water, 3% acetic acid, 20% ethanol) the BHT-amino TEMPO migrated and that the structure of the polymer was altered after 10 days of immersion at 50°C. This modification was assigned to the penetration of the aqueous media into the PVC. The ESR spectrum of the modified polymer consisted of two superimposed signals, the first one corresponding to BHT-amino TEMPO highly immobilized in the plastic material and the second to the same molecule moving more freely in spaces probably occupied by the solvent. Estimates of BHT-amino TEMPO levels in the immersion media seemed to indicate that they were of the same order of magnitude as the losses of the freer markers in the PVC after 10 days of immersion at 50°C.

INTRODUCTION

Plastics are currently used for packaging food. Besides their specific qualities, these containers have facilitated food distribution and, in many cases, have prevented breakage and pollution due to handling. Plastic materials used for food packaging should not cause any unacceptable

Food Chemistry 0308-8146/87/$03·50

alterations of the food. However, the contact between a container and its contents may result in the reciprocal migration of some of the products present (Botrel, 1982). Most of the chemical monitors used to estimate the inertia of plastic container materials employed gas-phase (Yu & Inoco, 1984; Kontominas, 1985) and liquid-phase (Schabron & Fenska, 1980; Perlstein, 1983; Vargo & Inoco, 1984) chromatographies or radioactive tracers (Bieber & Freytag, 1984; Haesen & Le Goff, 1984). These techniques are not always easy to apply and the migrating components of some foods are difficult to evaluate. The foods must therefore be replaced by conventional simulators (e.g. water, alcoholic water, acidulated water, oil) depending on the composition of the foodstuff (Directive 82/711/EEC, J.O. No. L 297 of 23 October, 1982).

We have studied the migration of 2,6-diterbutyl hydroxytoluene (BHT) in water and then in two simulators of aqueous foodstuffs (3% acetic acid and 20% ethanol) using the spin-labelling technique. It is important to determine the migrating ability of this stabilizer in order to have an idea of its long-term behaviour (Birley, 1982). The spin-labelling technique, which is based on ESR spectroscopy, is of current use in biology to study the behaviour of natural components which are made artificially paramagnetic (Berliner, 1976). When applied to migratory processes, this method should allow the specific detection of small amounts of chemical species in complex media without having to use simulators. From a practical viewpoint, the label nitroxide radical was bound to the adjuvant, incorporated into the usual composition of the PVC, and then analyzed by ESR spectroscopy.

The shape of the nitroxide ESR signal: intensity, width, band spacing, is directly related to the mobility of the molecule and thus reflects the nature of the medium. The paramagnetic molecule can thus provide information on the adjuvant behaviour when the plastic container is put into contact with different media.

MATERIAL AND METHODS

Material

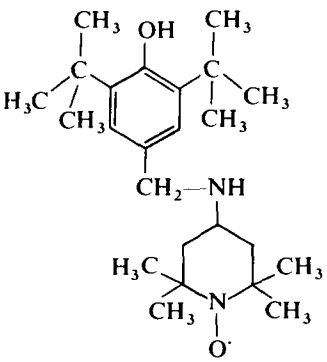
The ESR spectra were measured on a Bruker ER 100D spectrometer equipped with an X-band klystron (9·2 to 10 GHz) and the nuclear magnetic resonance (NMR) spectra on a Perkin–Elmer R32 (90 MHz) spectrometer; the mass spectra were measured by desorption and NH_3 chemical ionization on a ZAB 2 F (VGLD) spectrometer.

The rigid PVC, in the form of sheets, was of the quality used for bottling mineral water (Dorlyl Laboratory, Le Havre).

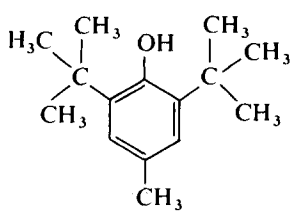
Methods

Synthesis of *N* (3,5-diterbutyl-4-hydroxybenzyl) 4-amino 2,2,6,6-tetramethyl-piperidinyloxy: BHT-amino TEMPO (Fig. 1)

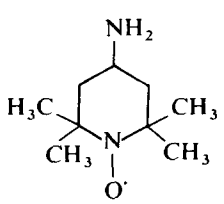
BHT was labelled paramagnetically by binding to it a nitroxide radical (4-amino 2,2,6,6-tetramethylpiperidinyloxy) often used because of its low reactivity (product of JANSEN). 600 mg ($2{\cdot}2 \times 10^{-3}$ moles) of 4-amino 2,2,6,6-tetramethylpiperidinyloxy (Amino-TEMPO) and 300 mg of 3,5-diterbutyl-4-hydroxybenzaldehyde were dissolved in 25 ml of anhydrous



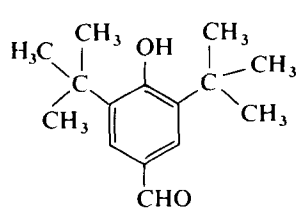
N(3,5-diterbutyl-4 hydroxybenzyl) 4-amino 2,2,6,6-tetramethylpiperidinyloxy 'BHT-amino TEMPO'



2,6-diterbutyl hydroxytoluene 'BHT'



4-amino 2,2,6,6-tetramethylpiperidinyloxy 'Amino-TEMPO'



3,5-diterbutyl-4 hydroxybenzaldehyde

Fig. 1. Chemical formulae of each molecule described in the text.

methanol and then stirred in the presence of molecular sieves. The mixture was allowed to react for 1 h at 30°C. 75 mg ($1{\cdot}19 \times 10^{-3}$ moles) of sodium cyanoborohydride were then added. The pH was adjusted to 7 with glacial acetic acid. The mixture was left to react for 3 h at 30°C; the reaction was stopped by adding 20 ml of a solution of water and acetic acid, pH 5. The molecular sieve was separated by filtration. The filtrate was concentrated under vacuum to half of the initial volume, then extracted three times with 30 ml of methylene chloride. The CH_2Cl_2 fraction containing the BHT-amino TEMPO was washed with water to neutrality, dried over sodium sulfate and evaporated. 327 mg of BHT-amino TEMPO were recovered and crystallized in a benzene–hexane mixture (1/1). A 65% yield was obtained. (The unreacted Amino-TEMPO could be recovered by extraction from the aqueous phase.)

Mass spectrum: $m/e = 390$ ($M^+ + 1$), 389 (M^+), 219 ($M\text{-}C_9H_8N_2O$), 172 ($M\text{-}C_{15}H_{21}O$).

NMR spectrum: $CDCl_3$ in the presence of 0·03M phenylhydrazine (Lee and Keana, 1975). (ppm) = 1·1 (s, 6H, $2CH_3$ TEMPO); 1·7 (s, 6H, 2 CH_3 TEMPO); 1·35 and 1·77 (m, 4H, CH_2 TEMPO); 1·45 (s, 18H, tert-butyl); 3·7 (s, 2H, CH_2-N); 3·05 (m, 1H, CH- N TEMPO).

Analysis: $C_{24}H_{41}N_2O_2$. Calc % C:74·03 H:10·53 N:7·19; $C_{24}H_{41}N_2O$, H_2O, C:70·76 H:10·56 N:6·88; Exp % C:71·69 H:10·19 N:7·59.

Preparation of rigid PVC samples and migration test

92 ppm of BHT-amino TEMPO and 76 ppm of Amino-TEMPO (0·23 and 0·44 mole/g of PVC) were incorporated separately into the compounds of rigid PVC; these amounts corresponded to about 3-fold the amount of BHT usually employed. In order to study the long-term behaviour of the paramagnetic molecules, the plastic materials were kept in an oven at 50°C for one month.

Migration tests were carried out on 150-mg PVC samples immersed in hermetic pill boxes containing, respectively, 2 ml of water, acetic acid or ethanol at 3% and 20% in aqueous solution ($V/A = 0{\cdot}74$ ml/cm^2 where V/A is volume of aqueous media/area of PVC sample); these boxes were kept in an oven at 50°C for 10 days, the time interval recommended for simulating the long-term behaviour of foodstuffs at room temperature (Figge, 1980).

Measurements

ESR spectra of the PVC samples were measured before immersion ($t = 0$) and then at regular intervals afterwards. For the latter measurements, the PVC samples, extracted from the aqueous media using small tweezers, were

dried with filter paper and then analyzed before being immersed again. The immersion media were analyzed on the last day of the migration test.

The amounts of the markers and their variations during the different experiments were estimated:

By the weight of the area of the third depression (immobile markers).
By reference to a free marker grading scale (free markers).

RESULTS

ESR spectroscopy of BHT-amino TEMPO and Amino-TEMPO

Figure 2 shows the spectrum of the BHT-amino TEMPO molecule (a) in an aqueous medium at room temperature and (b) in a rigid medium (solution deep-frozen at 180 K in liquid nitrogen). In an aqueous medium the

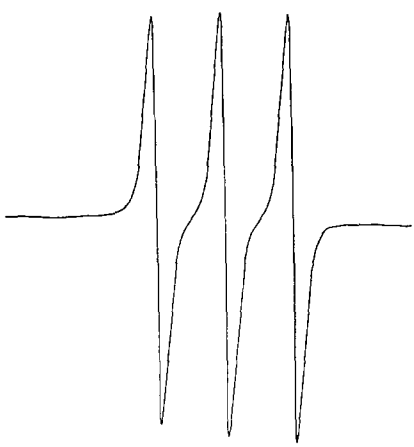


Fig. 2(a). Spectrum of the BHT-amino TEMPO in a fluid medium (CH_2Cl_2).

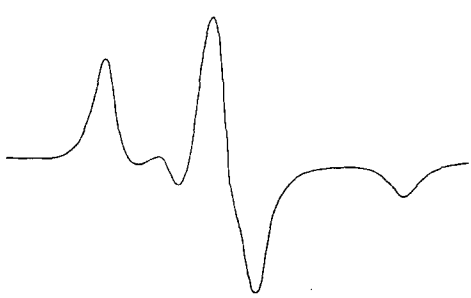


Fig. 2(b). Spectrum of the BHT-amino TEMPO in a rigid medium (solution deep-frozen at 180 K in liquid nitrogen).

nitroxide showed a spectrum (total width: 35 G) consisting of three thin, equidistant peaks of equal amplitude, characteristic of a freely moving species. At 180 K, a very asymmetrical, enlarged triplet (total width: 69 G) was observed characteristic of a highly immobile nitroxide in a very viscous medium (Berliner, 1979).

The aspect of the spectra of BHT-amino TEMPO and of Amino-TEMPO was identical in both the aqueous and rigid media which suggests that binding of the nitroxide radical to the BHT molecule does not modify the movements of the nitroxide.

ESR spectroscopy of rigid PVC samples

Long-term behaviour

The ESR spectra of PVC samples, containing the paramagnetic molecule, were similar to those of the same molecules at 180 K, showing that BHT-amino TEMPO and Amino-TEMPO retain their paramagnetic character during the transformation and extrusion of the polymer. However, after 30 days at 50°C, the overall intensity of the spectrum clearly decreased (Fig. 3).

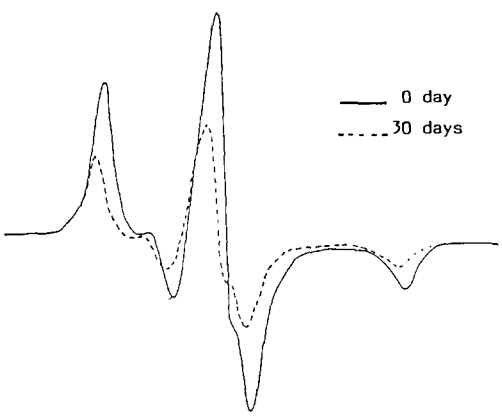


Fig. 3. Spectrum of the BHT-amino TEMPO incorporated into rigid PVC after 30 days at 50°C.

Migration test

PVC analysis. Figure 4 shows changes in the spectrum of the Amino-TEMPO incorporated into the PVC after 6 days of immersion in water. A complex signal composed of three broad, asymmetric bands (bands 1, 2, 3) and of two thin, well-defined bands (bands a, c) was observed. As shown in Fig. 5, this signal was the superposition of the nitroxide signals in two different media, one typical of the highly immobile molecule (bands 1, 2, 3)

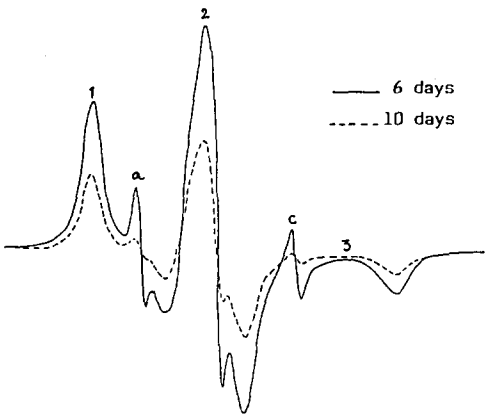


Fig. 4. Spectrum of the Amino-TEMPO incorporated into rigid PVC after 6 and 10 days of immersion in water.

and the other of a free marker (bands a, c; middle band b is masked by band 2).

Whatever the immersion medium, the aspect of the ESR spectra of the PVC samples incoporating Amino-TEMPO or BHT-amino TEMPO was the same. This indicates that the PVC contained an immobilized marker in its dense structure, and that water, acetic acid or aqueous ethanol penetrating the PVC disrupted some intramolecular bonds, creating empty areas or 'microspaces' (Kampouris & Regas, 1975; Messadi, 1982; Messadi & Taverdet, 1983) in which the marker can move more freely.

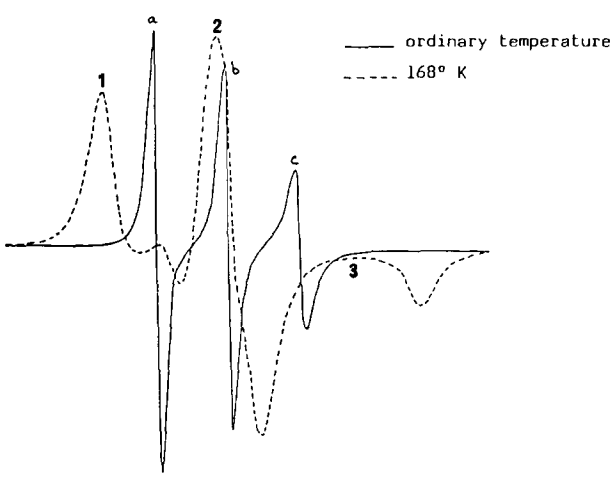


Fig. 5. Superposition of the Amino-TEMPO signals in oil at ordinary temperature and at 168 K.

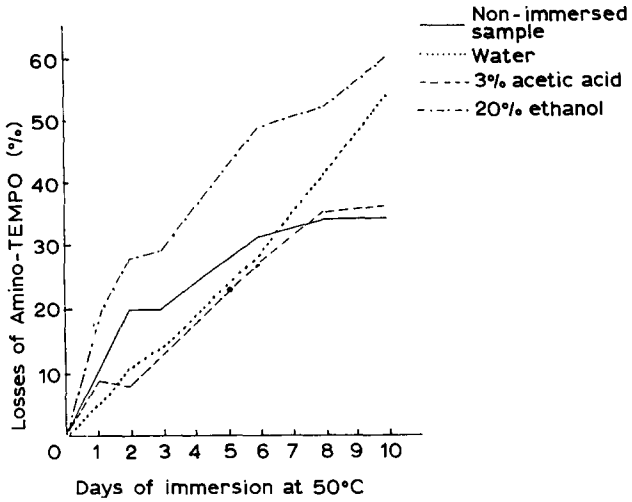


Fig. 6. Losses of Amino-TEMPO incorporated into rigid PVC after immersion in different aqueous media.

Moreover, as noted by Kampouris & Regas (1975) with plastified PVC, we observed that samples of rigid PVC lost their transparency after several days of contact with the extractant and became opaque, then whitish; this would also suggest that water had penetrated the plastic. After several days of contact at 50°C, there was a clear decrease in the signal intensity of the multiplet spectrum.

Figures 6 and 7 show that the overall losses of Amino-TEMPO and BHT-amino TEMPO in rigid PVC samples immersed in different aqueous media

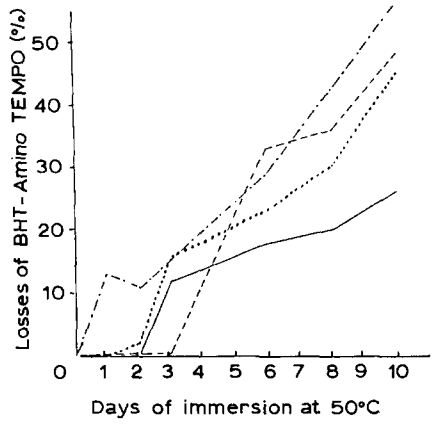


Fig. 7. Losses of BHT-amino TEMPO incorporated into rigid PVC after immersion in different aqueous media.

TABLE 1
Additional Losses of Amino-TEMPO and BHT-amino TEMPO[a] in Rigid PVC Samples After 10 Days of Immersion in Different Aqueous Media (Results in μg/g PVC)

Spin label	*Immersion media*		
	Water	*3% Acetic acid*	*20% Ethanol*
Amino-TEMPO	14·2	16·3	22·3
BHT-amino TEMPO	18·4	(1·5)	23·4

[a] Calculated by the difference in ESR signal intensity between immersed and non-immersed samples.

followed a progression similar to that of the non-immersed samples, but that after several days the losses became significantly higher. Additional losses of Amino-TEMPO and BHT-amino TEMPO, calculated by the difference in ESR signal intensity between immersed and non-immersed samples, were greater in ethanol than in water after 10 days of immersion but in the same media were of the same order of magnitude (Table 1). The proportions of

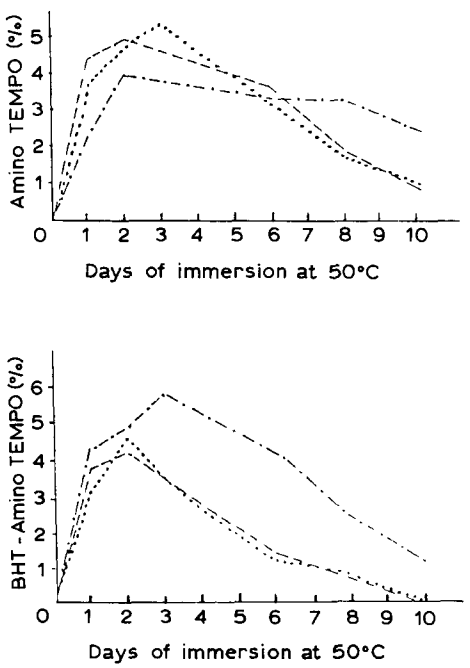


Fig. 8. Percentage of markers moving more freely in the PVC after immersion in different aqueous media.

TABLE 2
Losses of Amino-TEMPO and BHT-amino TEMPO in PVC Microspaces after 10 Days of Immersion in Different Aqueous Media (Results in μg/g PVC)

Spin label	*Immersion media*		
	Water	*3% acetic acid*	*20% ethanol*
Amino-TEMPO	3·1	3·4	1·8
BHT-amino TEMPO	3·8	3·5	3·3

immobile and freer markers were measured in rigid PVC samples by ESR spectroscopy.

The percentage of markers in the microspaces caused by the migration of the extractant reached maximal values after 3 or 4 days of immersion (this might coincide with maximal solvent penetration of the polymer) and then decreased gradually until day 10 (Fig. 8). These results are comparable to those obtained by Messadi on the diffusion of benzylic alcohol (Messadi & Vergnaud, 1981) and of aqueous ethanol (Messadi, 1982) in samples of plastified PVC immersed for 200 h at 50°C. Whatever the extractant considered, after 10 days of contact the losses of Amino-TEMPO and BHT-amino TEMPO in PVC microspaces were of the same order of magnitude (Table 2).

Analysis of immersion media. On day 10 of the migration test, the immersion media showed signals characteristic of freely moving nitroxide molecules, indicating that Amino-TEMPO and BHT-amino TEMPO migrated from the rigid PVC towards the various aqueous media.

The amounts detected (Table 3) were comparable to the losses of free marker in PVC.

TABLE 3
Amounts of Amino-TEMPO and BHT-amino TEMPO in Different Aqueous Media (Results in μg/g PVC)

Spin label	*Immersion media*		
	Water	*3% acetic acid*	*20% ethanol*
Amino-TEMPO		2·7	2·2
BHT-amino TEMPO	4·8	4·0	4·6

DISCUSSION

Nitroxide lifetimes are known to be short at temperatures between 200 and 250°C (Ramos & Catoire, 1979; Faucitano & Buttafava, 1983). However, the effect of an intermediate temperature (50°C) on nitroxides does not seem to induce significant degradation of the nitroxide. Therefore, the decrease of the Amino-TEMPO and BHT-amino TEMPO signals in samples of non-immersed PVC maintained for several days at 50°C can be attributed to a thermal degradation of the polymer. Under low O_2 partial pressure, nitroxides are known to react with free radicals (Scott, 1985) and may even be used as secondary antioxidants to protect polymers (Chevassus, 1977). The variations in the losses noted in control samples over 10 days at 50°C (Figs 6 and 7) could indicate a heterogeneous distribution of the markers in the polymer mass.

The decrease in the overall intensity of multiplet signals of immersed PVC samples (Fig. 4) is due not only to losses of Amino-TEMPO and BHT-amino TEMPO concomitant to polymer degradation, but also to their migration into the aqueous media so that the difference in marker loss between immersed and non-immersed PVC samples approximately corresponds to the migration rate (Table 1). However, the values obtained are clearly higher than the amounts of markers estimated in the aqueous media (Table 3). Such discrepancies are probably best explained by marker loss due to polymer degradation; this loss was quite likely much higher in immersed PVC samples during solvent penetration than in those recorded in the control samples. Moreover, the results obtained by measuring the difference in signal intensity between two PVC samples not having the same marker distribution certainly are probably not very accurate.

The losses of freely moving marker in the PVC after 10 days of immersion were similar to the quantities found in the different solvents; this would suggest that the proportions of Amino-TEMPO or BHT-amino TEMPO that migrated depended on the proportions solubilized in the PVC by the solvent.

In any case, nitroxide binding on a BHT molecule did not seem to change the amount of marker which was migrating.

These results should be considered carefully since the stability and chemical inertia of nitroxides vary significantly with the type of medium in which they are found (air, N_2, aqueous solvent, oil) and with the temperature at which they are studied. It will thus be necessary (1) to compare the data of this study, which was carried out for 10 days at 50°C, with those of an equivalent test for 3 months at ordinary temperature and (2) to use other methods of chemical monitoring: (radioactive tracers).

CONCLUSION

ESR spectroscopy analysis of the paramagnetic analogs of adjuvants used in the manufacture of plastic material, provides additional information compared to the techniques usually employed to study molecular transfer between a plastic container and its contents. With this method, we quantified the relative proportions of a paramagnetic analog of BHT immobilized in a dense PVC structure or freely moving in the microspaces created by solvent penetration.

The results of this experiment indicate that the amount of BHT-amino TEMPO migrating depends on the amount solubilized by the extractant.

This preliminary work needs to be examined in more detail and extended. Many points remain to be elucidated, particularly the stability and inertia of nitroxides in the various experimental conditions used in this study.

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