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STOICHIOMETRY OF THE OXIDATION OF ARYLHYDRAZINES WITH FERRICYANIDE

QUANTITATIVE MEASUREMENTS OF ABSORPTION SPECTRA OF ARYLDIAZENES

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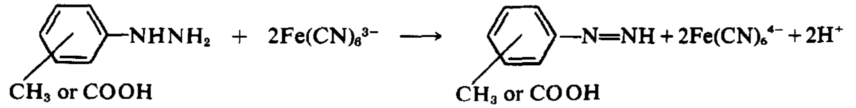
Abstract—A novel method for generating aryldiazenes quantitatively by the oxidation of arylhydrazines with exactly two equivalents of ferricyanide has been confirmed with *o*-, *m*-, and *p*-tolylhydrazine and with *o*- and *p*-hydrazinobenzoic acid. The rapidity and stoichiometry of the reaction permit accurate measurements of UV and visible absorption spectra in aqueous solution. Although oxidation of acetylphenylhydrazine by ferricyanide was slower, the product was shown to be acetylphenyldiazene. Acetylphenyldiazene is partially hydrogenated during mass spectrometric analysis. The source of hydrogen was shown to be H_2O in the spectrometer. This phenomenon, previously reported only in quinones, was also observed in the mass spectrum of methyl phenyldiazenecarboxylate.

Aryldiazenes (Ar-N═NH) have long been postulated as unstable intermediates in the course of oxidation of arylhydrazines,[1] reduction of diazonium salts,[2] and fragmentation of certain azo compounds.[3] However, formation of an aryldiazene was not confirmed directly until Kosower *et al*[4] published visible and UV absorption data of phenyldiazene generated by the decarboxylation of phenyldiazenecarboxylic acid. Subsequently these workers reported spectral properties of several substituted phenyldiazenes.[5] Although formation and identification of phenyldiazene by various methods have been observed in recent years,[6-8] the only other study directed to its spectral properties is that of Itano[9] on the product of oxidation of phenylhydrazine with ferricyanide. Since arylhydrazines are more readily available from commercial sources than aryldiazenecarboxylic acid esters, the ferricyanide method may be more generally useful for the formation of a variety of aryldiazenes. In the present study the stoichiometry of the reaction of ferricyanide with arylhydrazine to produce aryldiazene is confirmed, and quantitative determinations of spectral properties of several substituted aryldiazenes are reported.

RESULTS

Stoichiometry. The assumption that the oxidation of arylhydrazine by ferricyanide takes place according to



was confirmed by titration of H^+ ions formed by this reaction. As shown in Fig 1, the amount of H^+ ion formed during the oxidation of *o*-tolylhydrazine was equivalent to that of $K_3Fe(CN)_6$ added to the reaction mixture. The reaction was instantaneous. After the ratio of $K_3Fe(CN)_6$ to *o*-tolylhydrazine reached 2 : 1, the reaction mixture continued to consume ferricyanide ion. However, the rate of consumption became quite low, and the yellow color of ferricyanide ion remained much longer. The same result was obtained for the oxidation of other tolylhydrazines (*m*, *p*) and of hydrazinobenzoic acids (*o*, *p*).

UV and visible absorption spectra of aryldiazenes. Although the oxidation proceeds according to the above equation in the presence of excess arylhydrazine, some other reaction or reactions occur as the concentration ratio of $K_3Fe(CN)_6$ to arylhydrazine approaches 2. Therefore the reaction was always carried out at a concentration ratio lower than 1. In the present experiments the concentration of arylhydrazine in the blank cell was half that in the sample cell, and ferrocyanide formed by reduction of ferricyanide in the sample cell was matched by the addition of an equal concentration of $K_4Fe(CN)_6$ added to the blank cell. The spectrum of aryldiazene was obtained when the ratio of concentrations of $K_3Fe(CN)_6$ to arylhydrazine in the sample cell was 1, and spectra of mixtures of arylhydrazine with corresponding

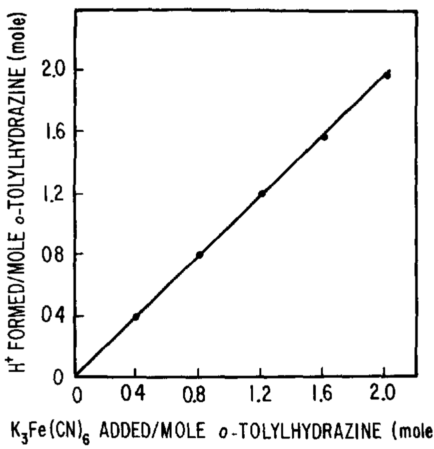


Fig 1. Stoichiometry of oxidation of *o*-tolylhydrazine by $K_3Fe(CN)_6$. Protons formed by the successive addition of 20 μl of 0·2 M $K_3Fe(CN)_6$ to 4 ml of $2{\cdot}5\times10^{-3}$ M *o*-tolylhydrazine were titrated with 0·04 N NaOH. Nitrogen was passed through the reaction mixture. Initial pH of the medium was 6·8.

aryldiazene were obtained when the ratio was less than 1. In Fig 2, the UV absorption spectrum of $5{\cdot}0\times10^{-5}$ M *o*-tolyldiazene in phosphate buffer, pH 6·8, is shown (curve a). Since the spectrum of *o*-tolylhydrazine in aqueous solution has not yet been reported, this is also shown (curve c). Curve b represents the absorption spectrum of a mixture of $2{\cdot}5\times10^{-5}$ M *o*-tolyldiazene and $2{\cdot}5\times10^{-5}$ M *o*-tolylhydrazine. UV absorption spectra of *o*-diazenobenzoic acid and *o*-hydrazinobenzoic acid are shown in Fig 3. Three isosbestic points appeared in the UV spectrum of each mixture of arylhydrazine and aryldiazene. Visible absorption spectra of *o*-tolyldiazene and *o*-diazenobenzoic acid are indicated in Fig 4. Similar results were obtained by the oxidation of *m*- and *p*-tolylhydrazine and of *p*-hydrazinobenzoic acid with ferricyanide ion. Absorption maxima (λ_{max}) and molar extinction coefficients at λ_{max} (ϵ_{max}) of these aryldiazenes and arylhydrazines and isosbestic points of mixture of corresponding pairs are shown in Table 1. The aryldiazenes were unstable even under nitrogen atmosphere, so that slow spectral changes were observed. The visible spectrum of *p*-diazenobenzoic acid changed rapidly; therefore the λ_{max} in this region was determined by taking the absorption spectrum of the compound at intervals and extrapolating the apparent λ_{max} to zero time.

Acetylphenyldiazene. Although the oxidation of arylhydrazine by ferricyanide ion was instantaneous, the oxidation of acetylphenylhydrazine proceeded very slowly. Occurrence of side reactions was indicated by the formation of about 2·5 moles of H$^+$ ion per 2 moles of $K_3Fe(CN)_6$ added. In order to determine whether acetylphenyldiazene (C_6H_5—N═N—$COCH_3$) is generated by the oxidation of acetylphenylhydrazine by $K_3Fe(CN)_6$, the reaction was carried out on a preparative scale. After reaction in aqueous solution, the product was extracted according to the method of Bock *et al*,[10] and purified with the use of a short path distillation apparatus

Table 1. Absorption maxima (λ_{max}) and molar extinction coefficients at λ_{max} (ϵ_{max}) or aryldiazenes and arylhydrazines in phosphate buffer, pH 6·7–6·8

Compound	λ_{max} (nm)	ϵ_{max}	Compound	λ_{max} (nm)	ϵ_{max}	Isosbestic points (nm)[a]
Tolyldiazene			Tolylhydrazine			
ortho	275·0	5,520	*ortho*	231·1	7,420	213
	—	—		278·5	1,490	220
	404	188		—	—	248
meta	274·0	6,820	*meta*	234·6	7,720	213
	—	—		281·0	1,350	227
	399	160		—	—	249–251
para	221·0	8,150	*para*	—	—	211
	283·0	8,970		233·7	9,250	226
	—	—		284·0	1,390	251
	397	207		—	—	—
Diazenobenzoic acid			Hydrazinobenzoic acid			
ortho	—	—	*ortho*	210·3	25,600	256
	271·0	4,980		243·4	7,060	297
	—	—		312·0	2,600	370–373
	405	146		—	—	—
para	216·4	9,970	*para*	—	—	214
	274·4	10,500		268·4	14,700	243
	404·5	210		—	—	297

[a] Isosbestic points in the absorption spectra of phenylhydrazine and phenyldiazene are 207, 221, and 246–7 nm.

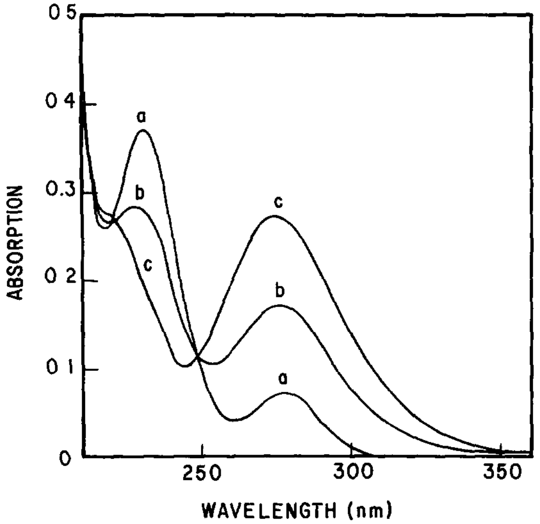


Fig 2. UV absorption spectra of *o*-tolyldiazene and *o*-tolylhydrazine. Curve a: $5{\cdot}0\times10^{-5}$ M *o*-tolyldiazene; curve b: $2{\cdot}5\times10^{-5}$ M *o*-tolyldiazene and $2{\cdot}5\times10^{-5}$ M *o*-tolylhydrazine; curve c; $5{\cdot}0\times10^{-5}$ M *o*-tolylhydrazine. Reaction was carried out in phosphate buffer, pH 6·8 (μ = 0·200). Light path, 1 cm. The spectra were taken within 3 min after *o*-tolyldiazene was generated.

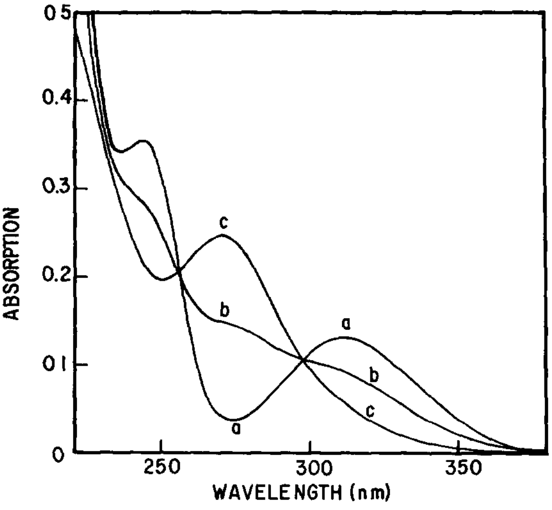


Fig 3. UV spectra of *o*-diazenobenzoic acid and *o*-hydrazinobenzoic acid. Reaction conditions are the same as in Fig 2.

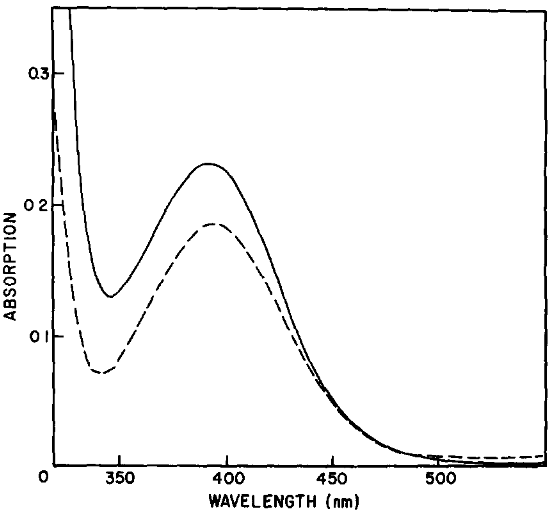


Fig 4. Visible absorption spectra of *o*-tolyldiazene (——) and *o*-diazenobenzoic acid (----) in phosphate buffer, pH 6·7. $2{\cdot}5\times10^{-3}$ M Arylhydrazine was reacted with $2{\cdot}5\times10^{-3}$ M $K_3Fe(CN)_6$ in the sample cell. $1{\cdot}25\times10^{-3}$ M Arylhydrazine and $2{\cdot}5\times10^{-3}$ M $K_4Fe(CN)_6$ were in the blank cell. The spectra were recorded within 4 min after reagent solutions were mixed.

(0·1 Torr, 20–22°). The yield was low. Acetylphenyldiazene was also synthesized by the method of Bock *et al*[10] in good yield. Both products showed the same IR and NMR spectra. The IR spectrum showed a strong carbonyl stretching band at 1750 cm^{-1}. NMR spectra in $CDCl_3$ showed the signals of acetyl protons at δ 2·4 and of two groups of phenyl protons at δ 7·35–7·65 and 7·80–8·00. The peak ratio of acetyl proton *vs* phenyl proton was shown to be correct. UV and visible absorption spectra[11] of acetylphenyldiazene in CH_2Cl_2 showed absorption maxima at 290·5 nm (π–π^* transition, ϵ 10,200) and 445·5 nm (n–π^* transition, ϵ 88). Slowness of its spectral changes indicated that this compound is fairly stable even under aerobic conditions. The mass spectrum of the compound, with distinctive peaks at *m/e* 43 (CH_3CO^+) (base peak), *m/e* 105 (M-43) (C_6H_5—N═N$^+$), and *m/e* 77 ($C_6H_5^+$), was in accord with the structure of acetylphenyldiazene. During the analysis an interesting phenomenon was observed, namely, the M^+ peak (*m/e* 148) was small and was exceeded by a peak at *m/e* 150. The cause of the M+2 peak was investigated by injection of D_2O into the mass spectrometer (Table 2). Diminution of the M+2 peak with enhancement of M+3 and M+4 peaks resulted. After D_2O was pumped out and H_2O was injected, a high M+2 peak reappeared. The mass spectrum of methyl phenyldiazenecarboxylate (C_6H_5—N═N—$COOCH_3$), similar structurally to acetylphenyldiazene, also has a higher peak at M+2 than at M^+.

DISCUSSION

Stoichiometric study of the reaction showed that the most likely position to be oxidized in arylhydrazines is the —NH—NH— group. Formation of the —N═N— bond is further suggested by the appearance of a low intensity band of the n–π^* transition at around 400 nm and a shift of the absorption band of the π–π^* transition to a longer wavelength. Although formation of tetraphenyltetrazene by the oxidation of diphenylhydrazine has been observed,[12] formation of 1,4-diaryltetrazenes by oxi-

Table 2. Changes in M + 1, M + 2, M + 3, and M + 4 peaks of acetylphenyldiazene during the mass spectral analysis under H_2O or D_2O vapor

Conditions[a]	Relative peak height[b] M + 1(149)	M + 2(150)	M + 3(151)	M + 4(152)
1	22	700	188	50
2	9	199	336	228
3	0	168	367	348
4	0	646	260	72

[a] Condition 1: Normal condition (~ 1×10^{-6} Torr).
Condition 2: After equilibration of the apparatus with D_2O (~ 1×10^{-6} Torr).
Condition 3: After injection of 10 μl of D_2O into the analyzer.
Condition 4: After pumping out of D_2O vapor and injection of 10 μl of H_2O.
[b] Peak height of M^+ ion was taken as 100%.

dation of arylhydrazines has not. Since the hydrazine group is electron donating and the carboxyl group is electron withdrawing, the π–π^* transition band of *p*-hydrazinobenzoic acid is at an especially long wavelength according to the disubstituent rule.[13] Although the azo group of azobenzene has been reported to be electron releasing,[14] it is not known whether or not the diazeno group of aryldiazene is electron releasing. However, the fact that the π–π^* transition band of *p*-tolyldiazene is at a longer wavelength than that of the other substituted phenyldiazenes (Table 1) might suggest that diazeno group is weakly electron attractive.

Replacement of the N-proton of phenylhydrazine with an acetyl group results in a more slowly oxidized compound, and side reactions[15] during the prolonged time of reaction may account for the low yield of acetylphenyldiazene after the oxidation of acetylphenylhydrazine with ferricyanide. According to the data of Table 2, H_2O in the mass spectrometer is the source of H atoms in the M + 2 ion in the spectrum of acetylphenyldiazene. Only certain quinone derivatives were previously known to react in this manner.[16] The extent of the reaction must be minor because the mass spectrum of acetylphenylhydrazine differs radically from that of acetylphenyldiazene.

Aryldiazenes are of interest both theoretically and experimentally; however, accurate measurements of physical properties are necessary in order to develop valid theories and interpretations. Preparation of aryldiazenes for absorption measurements by oxidation of arylhydrazines by ferricyanide has several advantages. The reaction is rapid and quantitative, and it can be carried out in the absorption cell; therefore a spectrum can be measured immediately after mixing of the reactants and before appreciable decomposition of the product takes place. Purity of the starting material is the determinant of the accuracy of the spectrum, and the starting material, the solid salt of an arylhydrazine, is not difficult to obtain and purify. As long as the arylhydrazine is pure, accuracy of results is not affected by its presence in both reference and sample cells. Release of two moles of proton per mole of arylhydrazine oxidized and presence of isosbestic points in the spectrum of each reaction mixture at successive stages of partial oxidation can be used as criteria for both the stoichiometry of the reaction and the presence of only one absorbing product in each new preparation of an aryldiazene.

Previous quantitative absorption data on aryldiazenes are those of Kosower *et el*[4,5] and of Itano.[9] In the latter study the spectra in aqueous solution of phenyldiazene generated by oxidation of phenylhydrazine by ferricyanide and by decarboxylation of phenyldiazenecarboxylic acid were compared. The wavelengths of absorption maxima were identical, and the molar extinction coefficients at these maxima agreed to within 2–3%. These results differed markedly, however, from those reported by Huang and Kosower.[4b] These workers reduced phenyldiazene with diazene and cited the appearance of isosbestic points in the UV spectrum of the reaction mixture as evidence for a direct reaction; however, byproduct that absorbed strongly at low wavelengths also appeared and had to be removed before a reasonable absorption spectrum of the major product could be obtained. The corrected spectrum was taken to be that of phenylhydrazine but, in fact, differed from the latter both in wavelengths and relative magnitudes of absorption maxima and minima. The molar extinction coefficient of phenyldiazene was then calculated from the known coefficients of phenylhydrazine with the assumption of quantitative yield, which was impossible because of the instability of phenyldiazene and the obvious impurity of the product assumed to be phenylhydrazine. Published spectra of phenyldiazene generated from phenyldiazenecarboxylic acid have higher absorbances at absorption minima around 240 and 350 nm than phenyldiazene produced by oxidation of phenylhydrazine with ferricyanide.[4b,9] Moreover, changes in spectra with time are more rapid with the former product.[4c,9] These observations suggest that the product obtained from phenyldiazenecarboxylic acid contains more contaminants.

The molar extinction coefficients of several substituted phenyldiazenes in MeCN were determined by Kosower *et al*[5] from weighed amounts of methyl aryldiazenecarboxylates. The procedure required saponification of the ester with KOH in MeCN, addition of water, evaporation to dryness, decarboxylation with a proton source in MeCN, and removal of insoluble salts by filtration. Calculation of aryldiazene yield was based upon determination of amount of unreacted aryldiazenecarboxylic acid. No mention was made of reaction stoichiometry, loss during transfers, and correction for decomposition of aryldiazene.

EXPERIMENTAL

o,m, and *p*-Tolylhydrazine-HCl (K&K Laboratories) were recrystallized from EtOH [m.p. *ortho*-, 194° (lit. 193·5–195·5°); *meta*-, 173–174° (lit. 168–170°); *para*-, 217–219° (lit. 222–234°)]. *o*- and *p*-Hydrazinobenzoic acid-HCl (Eastman Organic Chemicals) were recrystallized from water [m.p. *ortho*-, 186–188° (lit. 189–190°); *para*-, 237° (lit. 235–236°)]. 1-Acetyl-2-phenylhydrazine (Eastman Organic Chemicals) was recrystallized from water [m.p. 128–129° (lit. 128·5°), mol wt from mass spectrum: 150 (theory 150)]. Potassium ferricyanide (Mallinckrodt AR) and methyl phenyldiazenecarboxylate (Calbiochem, B Grade) were used as obtained. Potassium ferrocyanide ($K_4Fe(CN)_6 \cdot 3H_2O$, Mallinckrodt, AR) was dried to constant weight.

Generation of aryldiazenes. The method previously reported by Itano[9] was used for the generation and the spectroscopic measurement of aryldiazenes. O_2 was excluded by passing 99·998% N_2 through a soln of vanadous sulfate.[17] All of the reagent solns and solns in the reaction vessels were bubbled with O_2-free N_2 through long needles. Concentration or dilution of the solns was not observed during the passage of N_2. More detailed descriptions of the reaction vessel and experimental procedures have been published.[9]

Stoichiometry of arylhydrazine oxidation by $K_3Fe(CN)_6$. Four ml of $2 \cdot 5 \times 10^{-3}$ M arylhydrazine aqueous soln were bubbled with O_2-free N_2 for 10 min in a titration vessel (Radiometer, Copenhagen). After the pH of the soln was adjusted to 6·8, 0·2 M $K_3Fe(CN)_6$ was added in 20 μl increments, and 0·04 N NaOH was added with the Radiometer titrator TTT 11 to neutralize protons formed in the reaction.

Absorption spectra of aryldiazenes. Buffer soln was made by dissolving 6·805 g KH_2PO_4 and 13·40 g $Na_2HPO_4 \cdot 7H_2O$ in water to make 1,000 ml of the soln (μ = 0·200, pH = 6·80). Reagents were dissolved in this buffer solution. pH of the mixture changed slightly to lower pH (0·09–0·14 unit) when a high concentration of arylhydrazine was oxidized (Fig 4). The spectra were recorded on a Cary model 17 spectrophotometer. For UV absorption spectra, concentrations of arylhydrazine in the sample and blank cells were adjusted to $1 \cdot 0 \times 10^{-4}$ M and $5 \cdot 0 \times 10^{-5}$ M, respectively. Arylhydrazine in the sample cell was oxidized by $1 \cdot 0 \times 10^{-4}$ M or less $K_3Fe(CN)_6$. The concentration of $K_4Fe(CN)_6$ in the blank cell was the same as the concentration of $K_3Fe(CN)_6$ added to the sample cell. For visible absorption spectra, concentrations of arylhydrazine, $K_3Fe(CN)_6$, and $K_4Fe(CN)_6$ were increased 25 fold. NMR spectra were taken with a Varian T-60 NMR spectrometer with TMS as internal standard. IR spectra were measured with a Perkin-Elmer model A257 spectrometer. Mass spectra were taken by Dr. John Wright in a LKB Type 9000 analyzer at ionization voltage of 70 eV. Hydrogenation of the sample during mass analysis was confirmed with the use of D_2O. The analyzer was equilibrated with D_2O by repeatedly injecting and pumping out D_2O or by injecting 10 μl of D_2O just before the analysis.

Acetylphenyldiazene. Three grams of acetylphenylhydrazine (20 mmole) were suspended in a mixture of 156 ml of H_2O and 4 ml of pyridine. The suspension was cooled to 2° and N_2 was passed through it. Then 40 ml of aqueous 1 M $K_3Fe(CN)_6$ (40 mmole) were added to the soln at one time. The soln was stirred vigorously for 15 min at 2° and then for 20 min at 20°. Acetylphenyldiazene, which separated as a red oil, was extracted with CH_2Cl_2, and the solution was washed successively with dil. $Na_2S_2O_3$, 2 N HCl, dil $KHCO_3$, and H_2O according to the method of Bock *et al.*[10] After the solvent was distilled off, acetylphenyldiazene was purified by molecular distillation at 20–22° (0·1 Torr). SGM Scientific Model JM 7335 was used for this purpose. Acetylphenyldiazene was also prepared according to the method of Bock *et al*[10] IR spectrum (film) cm^{-1}: 3050, 1750, 1579, 1496, 1448, 1416, 1352, 1309, 1207, 1177, 1146, 769, 689. Mass spectrum *m/e* (intensity); 150(M+2) (2), 148 (M^+) (0·2), 108 (4), 107 (2), 106 (3), 105 (42), 94 (3), 93 (4), 78 (8), 77 (41), 76 (2), 66 (2), 65 (2), 64 (3), 63 (2), 52 (3), 51 (16), 50 (6), 44 (3), 43 (100), 42 (2), 41 (6), 40 (3), 39 (5), 38 (3), 32 (2), 28 (15), 27 (3), 18 (7), 17 (2), 15 (15), 14 (2).

Mass spectrum of acetylphenylhydrazine. Mass number *m/e* (intensity): 151 (M+1) (5), 150 (M^+) (49), 149 (6), 109 (8), 108 (100), 107 (32), 93 (6), 92 (21), 91 (10), 80 (5), 79 (6), 78 (9), 77 (29), 66 (2), 65 (11), 64 (3), 63 (3), 53 (2), 52 (4), 51 (12), 50 (4), 43 (23), 41 (2), 39 (7), 38 (2), 29 (4), 28 (3), 27 (2), 18 (2), 15 (4).

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