Liquid Chromatographic and Spectral Analysis of the Stereoisomers of Dimethylaminorex

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The individual enantiomers of *cis*- and *trans*-3,4-dimethylaminorex were prepared by treating ephedrines or pseudoephedrines with cyanogen bromide. These compounds represent potential designer drug modifications of aminorex and 4-methylaminorex, which have appeared recently in the clandestine drug market. The UV spectra for these compounds are typical of phenethylamine-type compounds, and FTIR spectra allow for differentiation of *cis*- and *trans*-isomers. The mass spectra for the dimethylaminorex stereoisomers show characteristic fragments at *m/z* 57, 118, and 190. The *cis*- and *trans*-isomers were separated in a reversed-phase liquid chromatographic system on a C18 stationary phase, with the *cis*-isomer displaying the higher capacity factor.

The pharmacological properties and abuse potential of the various derivatives of 2-amino-5-aryl-2-oxazolines have received considerable attention in recent years (1–3). Early reports (4, 5) described the anorectic activity of a large series of these compounds as potential substitutes for the amphetamine-type anorectics. The original reports on 2-amino-5-phenyl-2-oxazoline (aminorex) described it as a potent anorectic with interesting central nervous system (CNS) stimulating properties (4). Its anorectic properties were initially examined in rats and showed it to be equipotent with *d*-amphetamine. Substitution of halogens, particularly fluorine and chlorine at the *para* position of the aromatic ring, yielded anorectic activity up to 4 times that of aminorex. Electron donating alkoxy groups on the phenyl ring reduced activity, as did complete aromatization of the heterocyclic system to yield the oxazole.

The methyl derivative of aminorex, 4-methylaminorex or 2-amino-4-methyl-5-phenyl-2-oxazoline, was shown (4) to possess considerable anorectic activity in rats, having a slightly higher ED_{50} than aminorex and activity comparable to racemic amphetamine. The addition of a methyl group at the 4-position introduces geometric (*cis-trans*) isomerism into these compounds. The anorectic properties of the racemic *cis*- and racemic *trans*-methylaminorex, as well as the (+)-*trans*-methylaminorex, were essentially equipotent.

The anorectic activity of various aminorex derivatives has been substantiated in humans, and early animal studies revealed that these compounds also possessed CNS stimulant and cardiovascular effects similar to those of amphetamine.

In recent years, racemic *cis*-methylaminorex has appeared among the growing number of designer drugs available on the clandestine market. This compound was also recently classified as a Schedule I substance. Recently, Glennon and Misenheimer (6) reported the stimulus-generalization properties of the 4 individual stereoisomers of 4-methylaminorex compared to (*S*)-(+)-amphetamine. These studies showed the *trans*-

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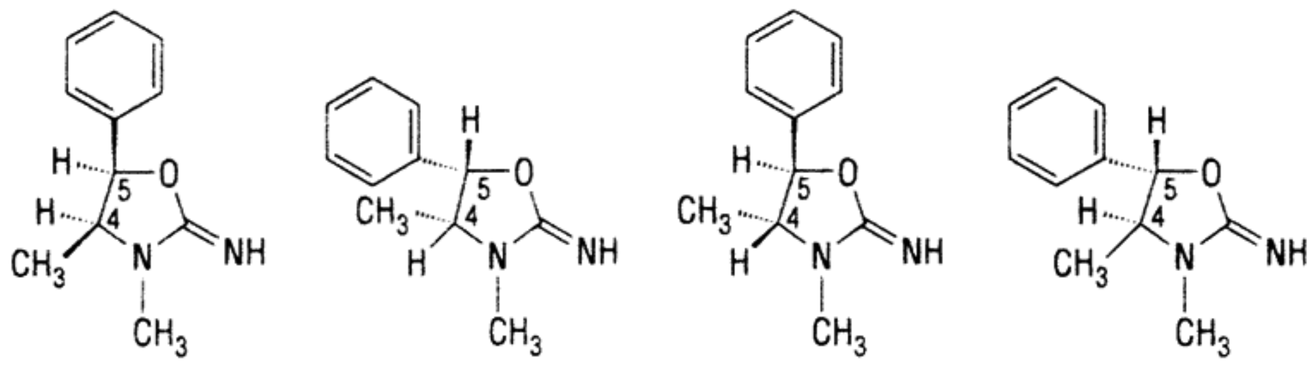


cis–(4S, 5R) cis–(4R, 5S) trans–(4R, 5S) trans–(4R, 5S)

***cis*-(4*S*,5*R*); *cis*-(4*R*,5*S*); *trans*-(4*R*,5*R*); *trans*-(4*S*,5*S*)**

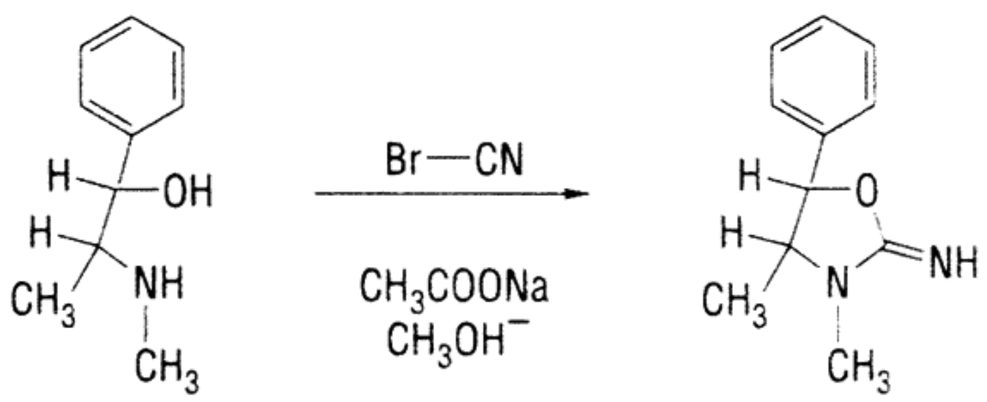
(4*S*,5*S*)-isomer to be more potent than either *cis*-isomer [the (4*S*,5*R*)- and (4*R*,5*S*)-isomers], which, in turn, were more potent than the *trans*-(4*R*,5*R*)-isomer. The more potent *trans*-(4*S*,5*S*)-isomer was found to be similar in potency to (*S*)-amphetamine. These stimulant and euphoriant effects, as well as blood pressure elevation, are likely the result of a sympathomimetic mechanism similar to amphetamine.

The stereoisomers of 4-methylaminorex have the potential to become significant problems in the clandestine drug market. These compounds can be prepared in a 1-step synthesis from readily available starting materials, norephedrine, norpseudoephedrine, and cyanogen bromide. Aminorex is prepared by an analogous synthesis from commercially available 2-amino-1-phenylethanol. Aminorex and 4-methylaminorex have already appeared on the clandestine street market (1, 3). The potential exists for the 3,4-dimethylaminorex isomers to appear in street samples as further designer modifications of the aminorex molecule. The dimethylaminorex isomers could be prepared via the same synthetic route described above from commercially available ephedrine and pseudoephedrine starting materials. In this study, we report the synthesis and analytical profiles of the 4 isomers of dimethylaminorex as "designer drug" analogues of aminorex and 4-methylaminorex.

Experimental

Instrumentation

The liquid chromatograph consisted of a Laboratory Data Control Constametric 3000 pump, 3100 spectromonitor UV detector operated at 220 nm, CI 4100 integrator, Rheodyne 7125 injector, and Waters Associates 30 cm × 3.9 mm μBondapak C18 column. Infrared spectra were recorded on a Perkin-Elmer Model 1710 Fourier transform infrared (FTIR) spectrophotometer. Ultraviolet spectra were recorded on a Shimadzu Instruments Model UV-160 spectrophotometer. Nuclear magnetic resonance spectra (^{1}H) were determined on a Varian EM-360 60 ^{1}H MHz spectrometer.



Scheme 1

Scheme 1. Synthesis of the stereoisomers of 3,4-dimethylaminorex.

Synthesis of cis- and trans-3,4-Dimethyl-5-phenyl-4,5-dihydro-2-amino-2-oxazolines (cis- and trans-3,4-Dimethylaminorex)

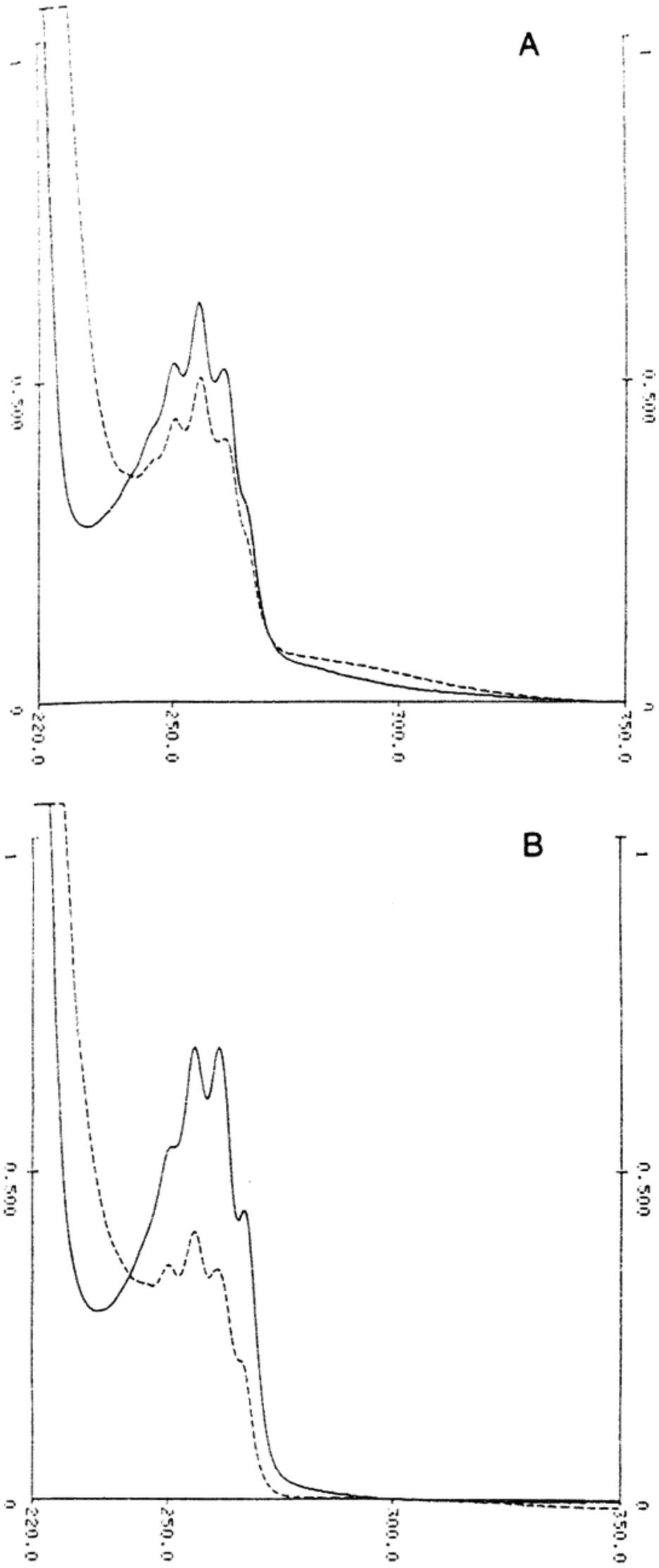
A solution of 1.6 g cyanogen bromide (15 mmol) in 10 mL methanol was added over a 10 min period to a cold (ice bath) stirred solution composed of 2.5 g of the appropriate ephedrine or pseudoephedrine (15 mmol) and 2.4 g sodium acetate (29 mmol) in 25 mL methanol. After the addition was complete, the mixture was stirred 1 h at room temperature and the solvent was evaporated under reduced pressure. The remaining oil was suspended in 25 mL water and made basic with 10% sodium hydroxide. The resulting oils were isolated by extraction with two 2 mL portions of chloroform and evaporation of the combined chloroform extracts. The product oils were crystallized from mixtures of carbon tetrachloride and ethyl acetate.

Liquid Chromatographic Procedures

The analytical column was 30 cm × 3.9 mm id packed with μBondapak C18 (Waters Associates). The analytical column was preceded by a 7 cm × 2.1 mm id guard column packed with CO:Pell ODS (Whatman). The derivatives were dissolved in LC-grade acetonitrile or methanol (1.0 mg/mL) and chromatographed with a mobile phase of pH 3.0 phosphate buffer and methanol (5 + 1). The phosphate buffer was prepared by dissolving 9.2 g monobasic sodium phosphate (NaH_2PO_4) in 1 L double-distilled water and adjusting the pH to 3.0 with H_3PO_4. The mobile phase flow rate was 1.5 mL/min, and the detector was operated at 0.2 AUFS. A 10 μL aliquot of sample solution was injected into the liquid chromatograph.

Results and Discussion

The analytical profiles for the isomers of methylaminorex (1) and aminorex (2) and its 4-phenyl regioisomer (7) have been reported previously. These compounds are all available through the same synthetic methodology: cyanogen bromide cyclization of the requisite phenethanolamine to yield the 2-oxazoline. Thus, it is reasonable that continued designer drug interest in this series will lead to the use of ephedrine or



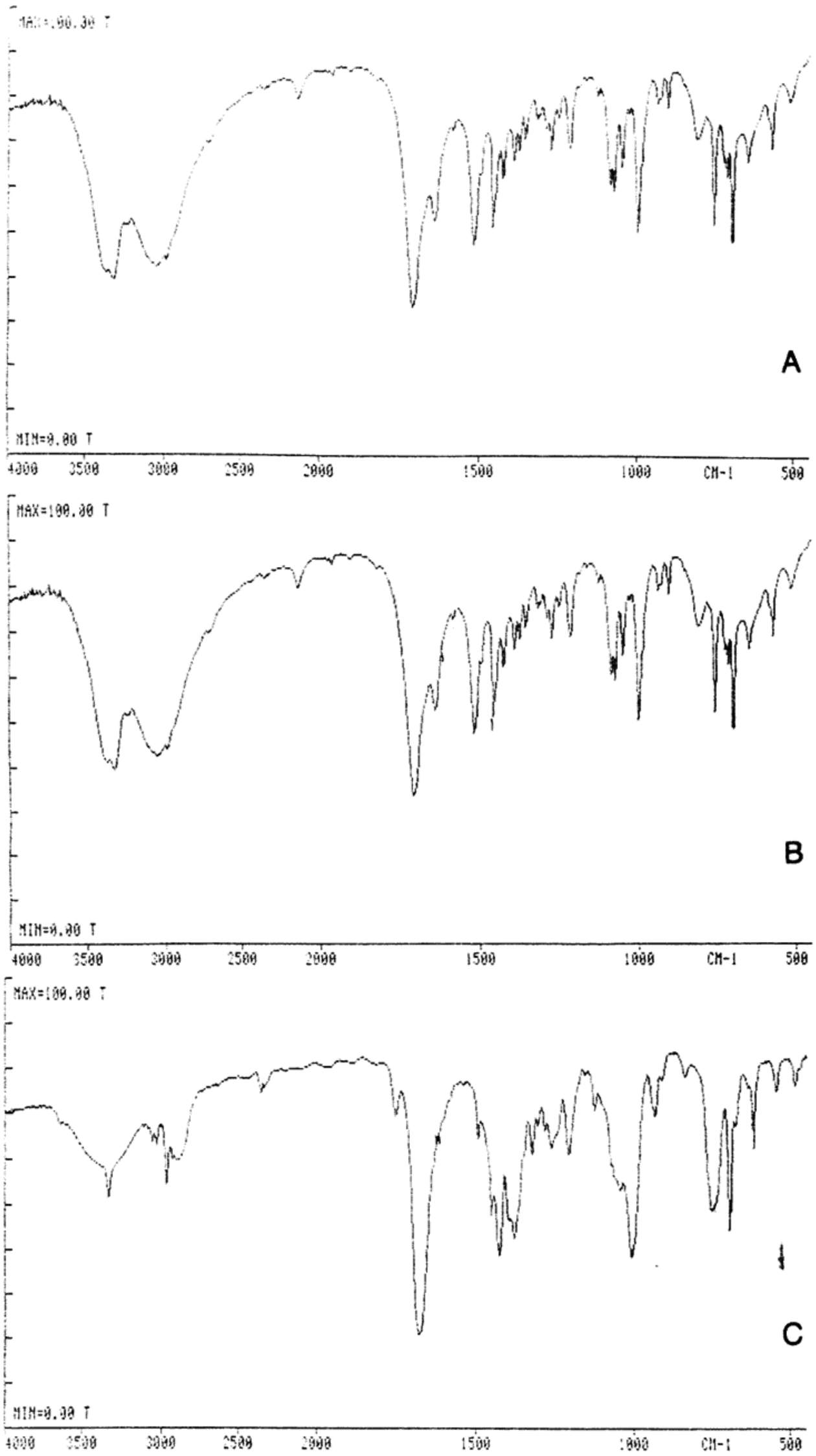


Figure 1. Ultraviolet absorption spectra of the isomers of 3,4-dimethylaminorex. A = *cis*-(*S,R*)-dimethylaminorex; B = *trans*-(*R,R*)-dimethylaminorex. Spectra represented by the solid line were determined in dilute sulfuric acid; spectra represented by the dashed line were determined in aqueous base.

Figure 2. Infrared spectra of isomers of 3,4-dimethylaminorex. A = *cis*-(*S,R*)-dimethylaminorex, B = *cis*-(*R,S*)-dimethylaminorex, C = *trans*-(*R,R*)-dimethylaminorex, and D = *trans*-(*S,S*)-dimethylaminorex.

pseudoephedrine as the phenethanolamine moiety in the cyanogen bromide cyclization reaction. The stereoisomers of dimethylaminorex were synthesized by this procedure as shown in Scheme 1, and these compounds were found to exist in the exocyclic double bond form because of the additional methyl group on the amine moiety of the ethanolamine fragment. The melting points for the dimethylaminorex isomers were determined in open capillary tubes and were quite low compared to the isomers of methylaminorex. The *cis*-(*S,R*)-isomer melted at 94–97°C and the *cis*-(*R,S*)-isomer melted slightly lower, at 90–93°C. The *trans*-(*S,S*)-isomer melted at 38–42°C and the *trans*-(*R,R*)-isomer at 39–44°C. The low melting points of these compounds may contribute to the necessity for organic solvent extraction in the synthetic work-up procedure. The higher-

Table 1. Proton NMR data (ppm) for the dimethylaminorex stereoisomers[a]

Proton	*cis*-(4*S*,5*R*)	*cis*-(4*R*,5*S*)	*trans*-(4*S*,5*S*)	*trans*-(4*R*,5*R*)
2-N-H	4.72 s	4.65 s	4.25 s	4.25 s
3-CH_3	2.84 s	2.85 s	2.82 s	2.82 s
4-H	3.92 p (*J* = 7 Hz)	3.90 p (*J* = 7 Hz)	3.40 m	3.38 m
4-CH_3	0.71 d (*J* = 7 Hz)	0.73 d (*J* = 7 Hz)	1.25 d (*J* = 7 Hz)	1.22 d (*J* = 7 Hz)
5-H	5.49 d (*J* = 8 Hz)	5.47 d (*J* = 8 Hz)	4.82 d (*J* = 9 Hz)	4.80 d (*J* = 9 Hz)
5-Ar-H	7.35 s	7.33 s	7.38 s	7.35 s

[a] All NMR spectra were determined in $CDCl_3$ with tetramethylsilane as an internal standard. Signal multiplicities are designated as follows: s = singlet, d = doublet, m = multiplet, and p = pentet.

melting methylaminorex isomers precipitate upon addition of base to the reaction mixture. The individual isomers of methylaminorex melt at temperatures above 177°C (1).

The ultraviolet absorption spectra of *cis*-(*S*,*R*)- and *trans*-(*R*,*R*,)-dimethylaminorex are shown in Figure 1. These spectra show the general absorption bands for phenethylamines in the 240–270 nm range. In Figure 1A, the spectrum for the *cis*-isomer shows 1 major absorption band with 2 slightly less intense bands in both acid and base solution. The spectrum for the *trans*-isomer in Figure 1B shows 2 bands of almost equal intensity in acid with a single major absorption of significantly lower intensity in base.

The infrared absorption spectra of the free base form of each isomer of dimethylaminorex are shown in Figure 2. These spectra were obtained from KBr disks on a Fourier transform infrared spectrophotometer. Because these compounds were synthesized as the individual stereoisomers, no spectra were obtained for racemic *cis*- or racemic *trans*-dimethylaminorex. The infrared spectra for the individual enantiomers of *cis*- can clearly be distinguished from the enantiomers of *trans*-dimethylaminorex. The spectra for the 2 *trans*-isomers (4*R*,5*R* and 4*S*,5*S*) appear identical in all respects, but slight differences in relative intensity exist between the individual stereoisomers of *cis*-dimethylaminorex.

The ^1H NMR data for the *cis*- and *trans*-dimethylaminorex isomers are shown in Table 1. The imino proton appears at a slightly higher field in the *trans*-isomers, while the *N*-methyl group does not appear to be influenced by the geometry of the 4,5-substituents. The methyl group at C-4 is upfield in the *cis*-isomer, appearing as a doublet centered at 0.7 ppm, whereas this signal in the *trans*-isomers occurs as a doublet at 1.2 ppm. This same trend was observed for the C-4 methyl group in the *cis*- and *trans*-isomers of 4-methylaminorex (1). The proton at C-4 occurs as a pentet centered at 3.9 ppm for *cis*-dimethylaminorex and as a multiplet centered at 3.4 ppm for the *trans*-isomer. The proton at C-5 occurs as a doublet in the spectrum of both isomers, with the signal for the *cis*-isomer slightly downfield compared to the *trans*-dimethylaminorex.

The mass spectra for these dimethylaminorex isomers are identical and an example spectrum (EI) is shown in Figure 3. The molecular ion is presented at *m/z* 190 as well as a peak for *m/z* 175 (M-15) most likely resulting from the loss of a methyl group. Because 4-methylaminorex shows an analogous loss of 15 mass units, the M-15 peak likely arises from the loss of the C-4 methyl group. The *m/z* 118 ion in the dimethylaminorex

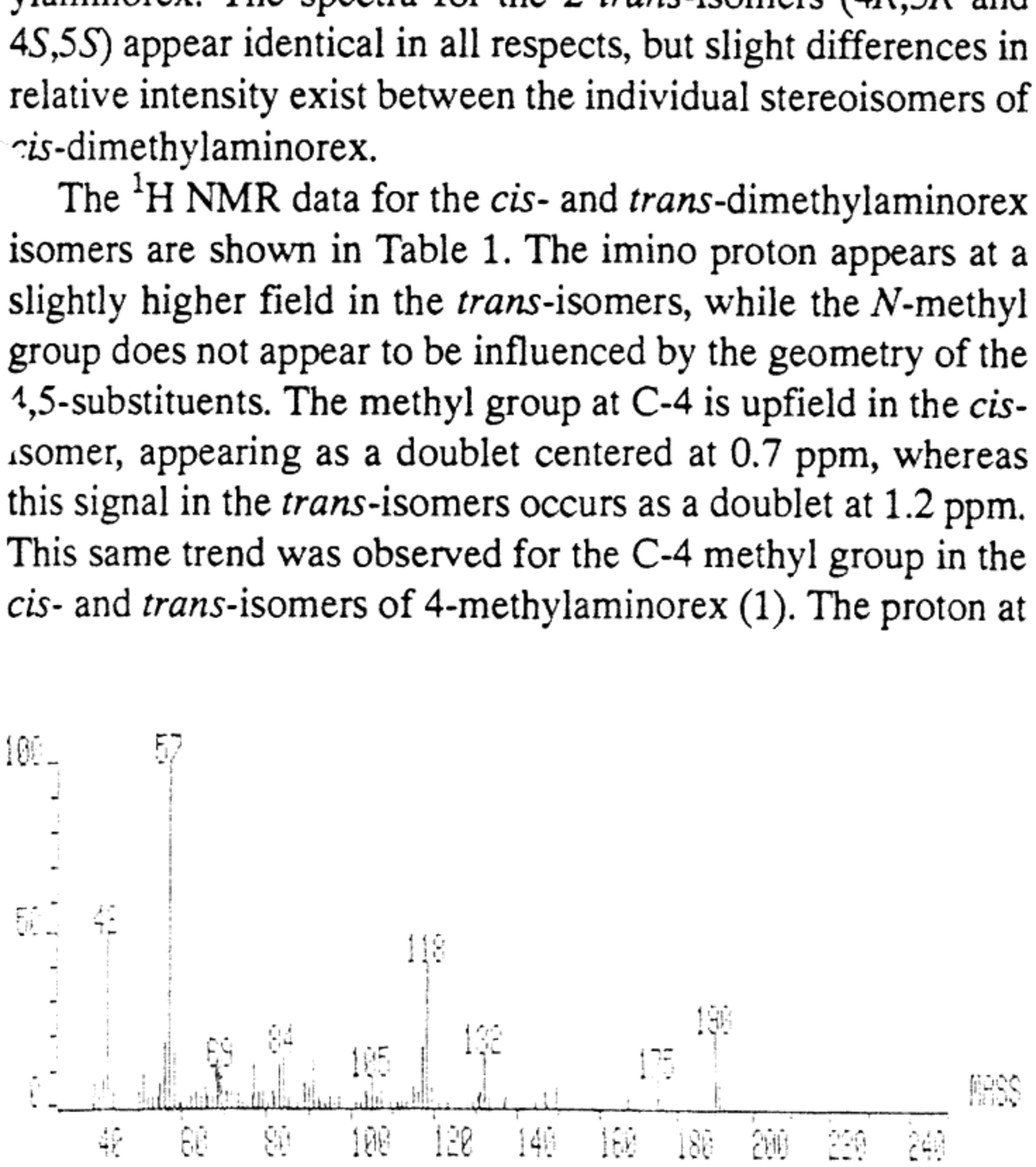


Figure 3. Mass spectrum of *cis*-(*S*,*R*)-dimethylaminorex.

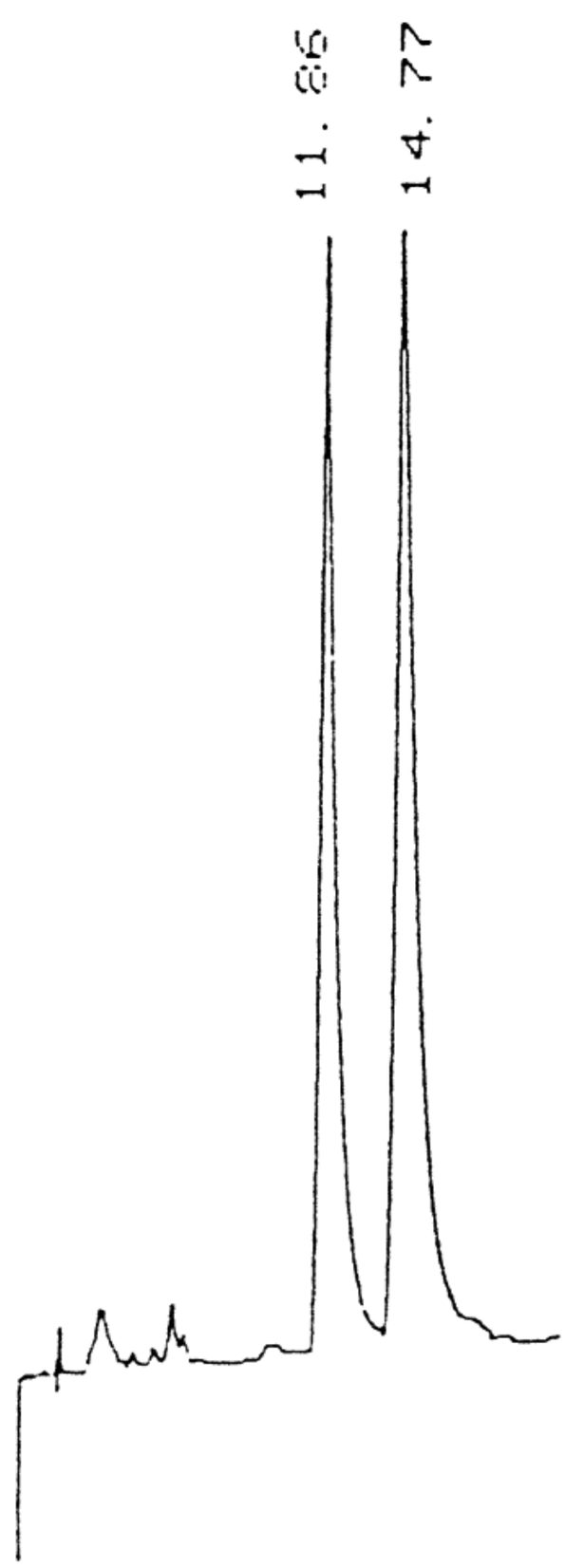
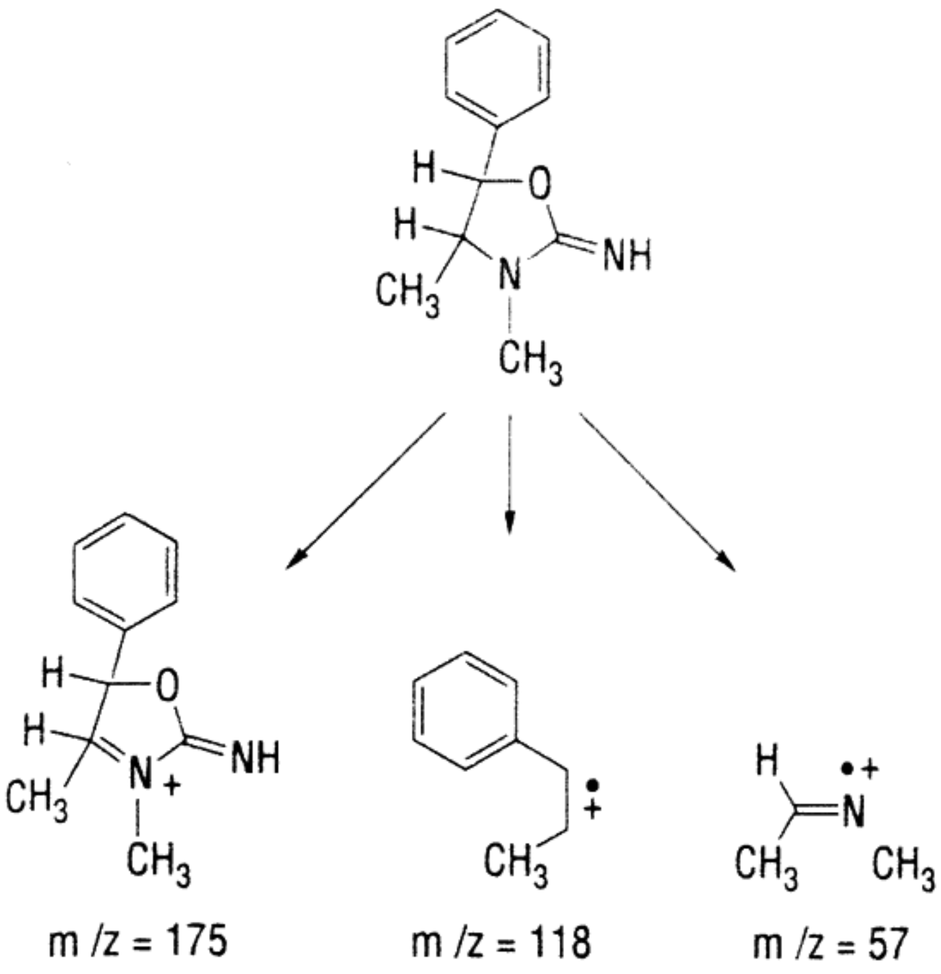


Figure 4. Reversed-phase liquid chromatographic separation of *trans*-dimethylaminorex (11.86 min) and *cis*-dimethylaminorex (14.77 min).



Scheme 2

Scheme 2. Mass spectral fragmentation scheme for the stereoisomers of 3,4-dimethylaminorex.

isomers is not observed in the spectrum of methylaminorex and likely is the phenylpropane skeleton, C_9H_{10} (Scheme 2). The base peak in the dimethylaminorex spectra occurs at *m/z* 57 and likely results from a retro-Diels-Alder-type fragmentation to yield C_3H_7N by splitting out the C–N, rings positions 4 and 3, respectively, and the accompanying methyl substituents. According to data from Klein et al. (1), the most abundant ion at *m/z* 43 in the EI-MS of methylaminorex has the elemental composition C_2H_5N. This ion possibly originates from the 2-imino tautomer by the loss of the N-3 and C-4 with its methyl substituent (CH_3–CH=NH). The analogous reaction with dimethylaminorex, which is locked in the 2-imino tautomeric form, would yield the *m/z* 57 ion due to the additional methyl group present on N-3.

The liquid chromatographic separation of the *cis*- and *trans*-isomers of dimethylaminorex is shown in Figure 4. This separation was achieved in the reversed-phase mode with a C18 stationary phase and a mobile phase of pH 3 phosphate buffer and methanol (5 + 1). The peak eluting first corresponds to the *trans*-isomer (11.86 min) and the *cis*-isomer elutes approximately 3 min later (14.77 min). The chromatographic system used for this separation is commonly used in our laboratory for the analysis of other basic drugs. However, further refinements of the system will be necessary to adequately resolve the isomers of both dimethylaminorex and methylaminorex in a single isocratic system.

In summary, the 3,4-dimethylaminorex isomers can be prepared from cyanogen bromide cyclization of ephedrine or pseudoephedrine. These compounds represent potential designer drug modifications of the 2-amino-5-phenyl-2-oxazoline (aminorex) system. These dimethyl derivatives contain the exocyclic imino (C=NH) double bond due to the additional *N*-methyl substituent at the 3-position of the oxazoline ring. These compounds are low-melting solids that show UV absorption properties characteristic of phenethylamine-type compounds. The *cis*- and *trans*-isomers can be separated by liquid chromatography with a reversed-phase system, and the *trans*-isomer displays the lower capacity factor. The *cis*- and *trans*-isomers can be differentiated on the basis of their infrared absorption spectra, and the EI-MS for all isomers show characteristic fragments at *m/z* 57 (base peak), 118, and 190 (molecular ion).

References

(1) Klein, R.F.X., Sperling, A.R., Cooper, D.A., & Kram, T.C. (1989) *J. Forensic Sci.* **34**, 962–979

(2) Ivaba, D., & Brewer, L.M., (1987) *Microgram* **20**, 55–61

(3) Klein, R.F.X., & Morello, D.R. (1990) *Microgram* **23**, 119–127

(4) Poos, G.I., Carson, J.R., Rosenau, J.D., Roszkowski, A.P., Kelley, N.M., & McGowin, J. (1963) *J. Med. Chem.* **5**, 266–272

(5) Yelnosky, J., & Katz, R., (1963) *J. Pharmacol. Exp. Ther.* **141**, 180–184

(6) Glennon, R.A., & Misenheimer, B., (1990) *Pharmacol. Biochem. Behav.* **35**, 517–521

(7) Noggle, F.T., Jr, Clark, C.R., & DeRuiter, J. (1991) *Microgram* **23**, 281–288