perbenzoic acid in 10 ml of THF. The solution was stirred at 0–10° for 30 min. EtOH (2 ml) saturated with dry HCl was added. After crystallization occurred, 30 ml of Et_2O was added and the mixture was filtered to give 1.7 g of the HCl salt of 5,6,7,8,9,10,11,12-octahydro-3-methoxy-*N*,*N*,5α-trimethyl-5,11-methanobenzocyclodecen-13β-amine *N*-oxide (**17**), mp 170–173° dec. Anal. Calcd for $C_{19}H_{30}NO_2Cl$: C, 67.13; H, 8.90; N, 4.13. Found: C, 66.61; H, 9.04; N, 3.98.

Acknowledgment. The authors are indebted to Mr. J. Malis and Mrs. E. Hernadi of the Wyeth Laboratories Pharmacology Department for their work in the determination of the biological activity of the compounds reported.

References and Notes

(1) The "butenyl-bridged" compounds are named as benzocyclononenes and the "benzo-bridged" compounds as dibenzo[*a*,*e*]cyclononenes and decenes in the Experimental Section in accordance with Chemical Abstracts recommendations.
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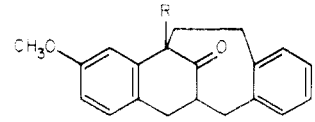
(8) We considered the alternate structure



to be improbable since we have found that 1-alkyl-β-tetralones are preferentially alkylated at the 1 position. This, plus the well-established fact that benzyl halides are far more reactive than the corresponding phenethyl halides, indicates that the intermediate first formed is 1-methyl-1-[*o*-(2-bromoethyl)]benzyl-2-tetralone, which on cyclization gives **10a** (n = 2) as indicated in Scheme II.
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Azaparacyclophanes. A Novel Class of Conformationally Rigid Analogs of Phenylethylamine and Phenylpropylamine[1]

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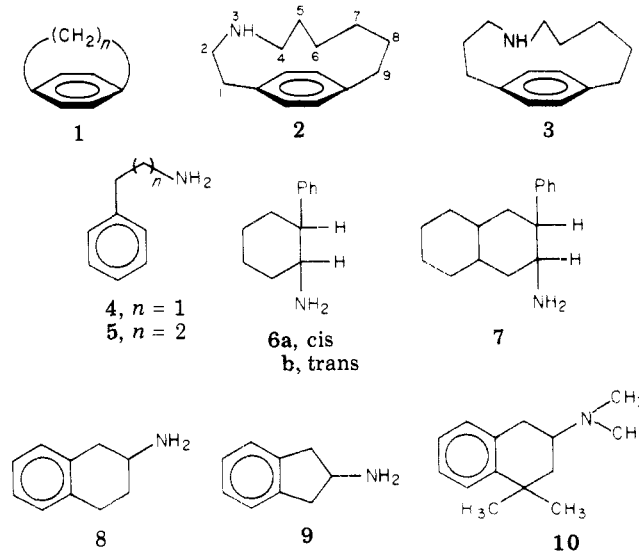
Aza[9]paracyclophanes **2** and **3**, analogs of phenylethylamine and phenylpropylamine, have been prepared and tested for pharmacological activity. Compound **3** was found to induce mydriasis without signs of sympathomimetic or anticholinergic activity.

The [*n*]paracyclophanes (**1**) represent a class of aromatic compounds in which the para positions of a benzene ring are incorporated into a bridging ring system.[3,4] The inclusion of a biologically active moiety into the conformationally rigid paracyclophane framework might lead not only to a unique class of drugs but also, because of its rigidity, could provide information concerning the stereochemical requirements of the biological receptor for that drug.

Most paracyclophanes (**1**) with values of n between 7 and 16 have been reported.[5] As the value of n decreases below 10, the benzene ring is increasingly distorted into a boat shape. For example, when n = 7, the para aromatic carbons are almost 20° above the usual benzene plane,[5] and this is reflected in the red shift of the ultraviolet spectrum of the benzene absorption.[3,5] Examination of CPK molecular models shows that rotation of the bridge about the aromatic nucleus is restricted at $n \approx 10$. This is consistent with the observed rotational barrier for an analogous group of bridged compounds, the [*m*,*n*]paracyclophanes.[4] Thus, at $n < 10$ the bridge is in a fixed conformation above the aromatic ring and cannot become coplanar with it.

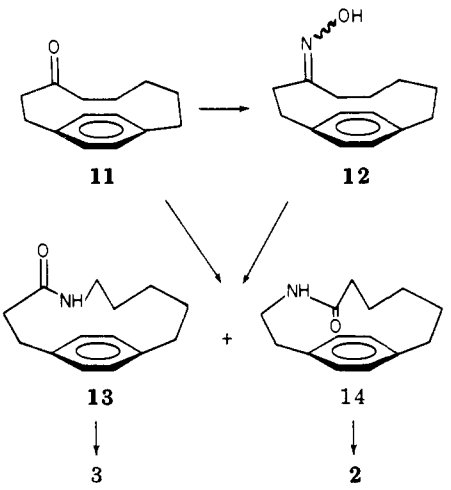
We have prepared the paracyclophane analogs, 3-aza-[9]paracyclophane (**2**) and 4-aza[9]paracyclophane (**3**), of phenylethylamine (**4**) and phenylpropylamine (**5**), respectively. The amine-containing bridge in **2** and **3** is restricted to conformations which place it above and in close proximity to the aromatic ring. Such restrictions would be expected to modify the spectrum of biological activity displayed by the corresponding open-chain analog



by limiting interaction with receptor sites that require simultaneous binding of the amine group and aromatic system. A further effect could arise from pK_a changes induced by the proximity of the aromatic system and the amine group. The conformations imposed by the paracyclophane structure are very different from those indicated to be important in structure–activity theories based on molecular orbital,[6] solid state,[7] and solution[8] studies.

The biological properties of various conformationally restrained analogs of phenylethylamine (**4**) have been

Scheme I



described. For example, in the semirigid amphetamine analogs, *dl-cis-* and *dl-trans*-2-phenylcyclohexylamine **6a,b**, the cis isomer **6a** shows a stronger amphetamine action than the trans isomer **6b**.[9] While the four possible rigid isomers of **7**, *dl*-2-amino-3-phenyl-*trans*-decalin, show amphetamine activity, the stereochemical preference of **6a** is not carried over.[10] Marked amphetamine effects, as well as hallucinogenic activity, also are noted for 2-aminotetralin (**8**).[11] Both 2-aminoindan (**9**)[12] and the 2-aminotetralin derivative **10**[13] show potent analgesic action. Compound **9**, in addition, is a bronchodilator.

Chemistry. For the synthesis of azaparacyclophanes **2** and **3** (Scheme I), the ketone **11**[14] was converted into the oxime **12** and rearranged (Beckmann reaction) to an approximately equimolar mixture of amides **13** and **14**. Alternatively, the ketone **11** was treated with hydrazoic acid (Schmidt reaction), forming a similar mixture of **13** and **14**.

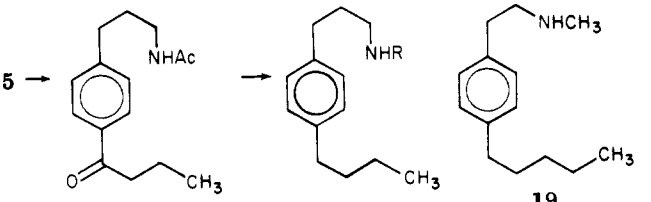
The structural assignments to the amide isomers **13** and **14** are based on examination of their NMR spectra. Those methylene protons situated over the benzene ring will be strongly shielded and show a large upfield shift relative to the chemical shift value in the open-chain compound, while those methylene groups of **13** and **14** located α or β to the benzene ring will not be unusually shielded and would be expected to show normal chemical shifts.[3,5,15]

In compound **14** the C_2 methylene appears as a quartet at δ 3.45 (D_2O exchange of the amide proton at δ 4.65 reduces the multiplicity to the expected triplet), but the analogous C_5 methylene of **13** appears at δ 2.60 (D_2O exchange of the amide proton at δ 4.37 also changes the multiplicity of this absorption), an upfield shift of 0.7 ppm relative to the normal value of δ 3.3.[16]

Reduction of amides **14** and **13** with lithium aluminum hydride gave the corresponding amines **2** and **3**. The isomeric amines are readily distinguished by their mass spectra; the phenylethylamine **2** (base peak *m/e* 70) forms the more stable benzylic ions by cleavage α to the nitrogen and thus undergoes fragmentation more readily than the phenylpropylamine isomer **3** (base peak *m/e* 203, M^+). Similarly, a peak at *m/e* 160 in the spectrum of **2** which is ascribed to the formal ion $^+CH_2C_6H_4(CH_2)_4CH_2\cdot$ is not found in the spectrum of **3**.

For pharmacological comparison, the open-chain analog of **3**, *N*-methyl-3-(4'-butylphenyl)propylamine (**18**), was prepared as follows (Scheme II). Phenylpropylamine (**5**) was converted to the acetamide and acylated with butanoyl chloride (Friedel–Crafts reaction) to yield the ketoamide **15**, which was reduced (Wolff–Kishner reaction) to the

Scheme II



15 **16**, R = H; **17**, R = CHO; **18**, R = CH_3 **19**

amine **16**. After formylation of **16** to produce **17**, reduction with lithium aluminum hydride gave the desired secondary amine **18**. That **18** was exclusively the para isomer was shown by GLC examination of the product from potassium permanganate oxidation of **18** and subsequent methyl esterification. Only dimethyl terephthalate, and no trace of the isophthalate or phthalate esters, was detected.

The open chain analog of **2**, *N*-methyl-2-(4'-pentylphenyl)ethylamine (**19**), was prepared, in a manner analogous to the synthesis of **18**, from phenylethylamine (**4**).

Biological Results. The compounds were evaluated in a mouse behavior screen similar to that described by Irwin[17] at doses of 10, 30, 100, and 300 mg/kg ip. The 4-aza[9]paracyclophane (**3**) as the hydrochloride salt was observed to elicit mydriasis at 30 mg/kg ip and subsequently at 25 mg/kg. Mouse papillary diameter was measured by the method of Pulewka,[18] i.e., with the use of a binocular microscope containing an ocular micrometer. Illumination of the eye always results in maximal miosis in normal mice. In an endeavor to identify the nature of this mydriatic effect, **3** as the hydrochloride salt was administered to a cat anesthetized with sodium pentobarbital and monitored for blood pressure and heart rate. Cumulative intravenous doses of 1, 3, and 10 mg/kg of compound **3** hydrochloride salt did not antagonize the normal heart rate and blood pressure changes to a standard dose of 1 μg/kg of acetylcholine. This infers a lack of anticholinergic activity. This compound also failed to increase the heart rate or blood pressure upon intravenous administration indicating a lack of direct agonistic or indirect, e.g., ganglionic stimulant sympathomimetic activity. In addition, in the mouse behavior screen, signs of exophthalmia and piloerection, which might be indicative of sympathomimetic activity, were not detected.

The corresponding open-chain analog **18** as the hydrochloride salt also elicited mydriasis in the mouse behavior screen at 100 mg/kg intraperitoneally. At this dose, this compound also elicited exophthalmia, a sign of sympathomimetic activity. The **2** and **19** analogs, as maleate salts, were tested for mydriatic activity in the mouse behavior screen. Compound **2** (maleate salt) was devoid of mydriatic activity at doses through 30 mg/kg ip and compound **19** (maleate salt) did not elicit mydriasis at doses through 100 mg/kg ip. The next dose of each drug was lethal.

Compounds **3** and **18** as hydrochloride salts and **2** and **19** as maleate salts were evaluated for anorexigenic activity by the method of Kettler, Braun, and Kandel.[19] All four compounds were inactive at 30 mg/kg po. The compounds were also evaluated for central dopaminergic activity utilizing rats with 6-hydroxydopamine lesions of the nigrostriatal pathway as described by Ungerstedt.[20] In this assay agents releasing endogenous dopamine elicit ipsilateral rotation and agents directly stimulating the dopamine receptor induce contralateral rotation. All four compounds were evaluated at 30 mg/kg intraperitoneally

and were determined to be devoid of dopaminergic stimulant activity.

The remarkable and unique finding in this study is that mydriasis is induced by the hydrochloride salt of 4-aza-[9]paracyclophane (3) without signs of sympathomimetic or anticholinergic activity.

Experimental Section

Melting points were obtained on a Mel-Temp melting point apparatus and are uncorrected. Infrared spectra were recorded on a Perkin-Elmer 137 spectrophotomer and nuclear magnetic resonance spectra on Varian A-60 and HA-100 spectrometers, with Me_4Si as an internal standard. Mass spectra were determined on a Varian CH-4 spectrometer. The NMR and ir spectra of all compounds were consistent with the assigned structures. Mass spectra were determined for all intermediates and products except 17 and 19, and all showed corresponding molecular ions and fragmentation patterns consistent with structures shown. Elemental analyses were performed by the Syntex Analytical Department; results for the elements indicated are within 0.4% of the theoretical values. GLC analyses were done using a Varian Series 1400 instrument with FI detector (150°, 0.03 in. × 200 ft, OV-225 + 25% Igepal CO-880).

Oxime (12) of 3-Keto[8]paracyclophane. A solution of 3-keto[8]paracyclophane (11)[14] (1.5 g, 7.4 mmol) and hydroxylamine hydrochloride (2.0 g, 29 mmol) in 20 ml of 50% pyridine in EtOH was heated at 80° for 3 h. Water was added to the cooled solution, and the precipitate was filtered yielding a white solid, 1.2 g (75%). An analytical sample, recrystallized from acetone–hexane, gave a mixture of crystalline forms, presumably the syn and anti isomers, mp 134–137°. Anal. ($C_{14}H_{19}NO$) C, H, N.

3-Keto-4-aza[9]paracyclophane (13) and 4-Keto-3-aza-[9]paracyclophane (14). Method A. Beckman Reaction. A suspension of 12 (0.96 g, 4.4 mmol) in 32 g of polyphosphoric acid was stirred manually and heated from room temperature to 120° during 40 min. The resulting solution was cooled and poured onto ice. The products were extracted into CH_2Cl_2, which was washed with $NaHCO_3$ solution and with brine. Evaporation of the solvent gave a yellow oil, which was chromatographed on silica gel. Gradient elution (hexane–Et_2O, then Et_2O–MeOH) gave a mixture of 13 and 14 (0.26 g), followed by almost pure 13 (0.24 g, 25%), which gave white crystals from Et_2O–MeOH: mp 125–126°; NMR ($CDCl_3$) δ 7.07 (s, 4 H, aromatic), 4.37 (b, 1 H, –NH–), 2.93 (t, 2 H, J = 6 Hz, C_2 methylene), 2.60 (m, 4 H, C_9 and C_5 methylenes), 2.20 (t, 2 H, J = 6 Hz, C_1 methylene), 1.47 (m, 2 H, C_8 methylene), 0.89 (m, 4 H, C_6 and C_7 methylenes). Anal. ($C_{14}H_{19}NO$) C, N, O.

Method B. Schmidt Reaction.[21] The ketone 11 (2.5 g, 12 mmol) was added to a stirred mixture of trichloroacetic acid (32 g) and sulfuric acid (2.5 g) at 65–70°. Sodium azide (1.25 g, 19 mmol) was then added. After 15 min, the mixture was cooled, diluted with H_2O, and treated with excess concentrated NH_3. The products were extracted into CH_2Cl_2, which was separated, washed with brine, and dried over $MgSO_4$, and the solvent was removed by evaporation. Chromatography of the residue on silica gel (gradient elution, 50% Et_2O–hexane to Et_2O) gave unchanged ketone 11 (0.2 g), followed by 14 (1.0 g, 38%). Recrystallization from benzene–hexane gave an analytical sample: mp 119–120°; NMR ($CDCl_3$) δ 7.05 (m, 4 H, aromatic), 4.65 (b, 1 H, –NH–), 3.45 (q, 2 H, J = 6 Hz, C_2 methylene), 2.80 (t, 2 H, J = 6 Hz, C_1 methylene), 2.56 (t, 2 H, J = 6 Hz, C_9 methylene), 0.75–1.70 (complex, ~8 H, C_5–C_8 methylenes). Anal. ($C_{14}H_{19}NO$) C, H, N.

Further elution of the column yielded 13 (0.72 g, 27%).

3-Aza[9]paracyclophane (2). The amide 14 (1.06 g, 4.9 mmol) in THF (25 ml) was added during 15 min to a stirred mixture of $LiAlH_4$ (0.49 g, 13 mmol) in THF (50 ml) under nitrogen. The mixture was refluxed for 15 h and then cooled and excess reagent decomposed by slow addition of 2 ml of H_2O in 25 ml of THF, followed by 0.5 ml of 15% NaOH solution. The mixture was filtered, and the filtrate was evaporated in vacuo. The residue was dissolved in Et_2O and dried over $MgSO_4$. Evaporation of the solvent gave a yellow oil, which was chromatographed on silica gel (gradient elution, 50% Et_2O–hexane to Et_2O to 10% MeOH–Et_2O). There was obtained 0.80 g (80%) of 2 as a yellow oil: MS (70 eV) m/e 203 (68, M^+), 160 (28), 118 (33), 117 (28), 105 (22), 104 (37), 91 (22), 84 (23), 70 (100), 57 (27), 44 (54), and 43 (23).

The picrate derivative had mp 174–176°. Anal. ($C_{20}H_{24}N_4O_7$) C, H, N.

The maleate salt was an oil.

4-Aza[9]paracyclophane (3). The above procedure for conversion of the amide 14 to the amine 2 was used to convert 13 (1.0 g, 4.6 mmol) to the oily amine 3 (0.78 g, 84%): MS (70 eV) m/e 203 (100, M^+), 172 (24), 117 (27), 104 (20), and 91 (23).

The solid hydrochloride derivative had mp 172° dec. Anal. ($C_{14}H_{22}NCl$) C, H, N.

***N*-Acetyl-3-(4'-butanoylphenyl)propylamine (15).** A solution of butanoyl chloride (27 g, 0.25 mol) in 1,2-dichloroethane (25 ml) was added slowly to a suspension of $AlCl_3$ (56 g, 0.42 mol) in 500 ml of the same solvent. *N*-Acetylphenylpropylamine (29 g, 0.16 mol, prepared by acetylation of 5) in 1,2-dichloroethane (100 ml) was added during 1 h, and the resulting mixture was stirred for 18 h. After pouring into an ice and water mixture, the organic phase was separated and washed successively with 10% HCl, H_2O, saturated $NaHCO_3$ solution, and brine. The solution was dried over $MgSO_4$ and then evaporated to give 40.7 g (100%) of 15, mp 52–55°. A sample recrystallized from Et_2O at 0° had mp 64–65°. Anal. ($C_{15}H_{21}NO_2$) C, H, N.

3-(4'-Butylphenyl)propylamine (16). The amide 15 (40 g, 0.16 mol) was added to a solution of KOH (55 g, 0.98 mol) in ethylene glycol (300 ml). Hydrazine (70 ml of 95%, 2.1 mol) was added, and the mixture was distilled until 100 ml of distillate had been collected. After an additional 6 h of refluxing, the solution was cooled and 600 ml of brine added. The product was extracted into benzene, which was washed with brine and evaporated. Distillation of the residue gave the amine 16 (22.5 g, 75%) as a brownish oil: bp 100–105° (0.5 mm); MS (70 eV) m/e 191 (M^+).

For analysis the amine was formylated with acetic–formic anhydride forming 17, mp 54–55° from Et_2O–benzene. Anal. ($C_{14}H_{21}NO$) C, H, N.

***N*-Methyl-3-(4'-butylphenyl)propylamine (18).** A solution of 17 (6.7 g, 31 mmol) in 15 ml of THF was added slowly to a stirred suspension of $LiAlH_4$ (2.2 g, 58 mmol) in 150 ml of THF, and the mixture was refluxed for 7 h. The mixture was cooled, and a solution of 2.2 ml of H_2O in 20 ml of THF was slowly added. A 15% NaOH solution (6 ml) was then added, and the resulting mixture was filtered. The filtrate was evaporated and the residue taken up in Et_2O and dried over $MgSO_4$. Evaporation of the solvent and distillation of the oily residue gave the amine 18 (5.4 g, 86%) as a colorless oil: bp 106° (0.7 mm).

The solid hydrochloride was prepared: mp 183–185°. Anal. ($C_{14}H_{24}NCl$) C, H, N.

To show that 18 was in fact the para isomer, a small sample was oxidized by refluxing overnight with excess $KMnO_4$ in *t*-BuOH–H_2O (2:1). The reaction mixture was cooled and filtered through Celite, and the filtrate was evaporated to dryness. The residue was triturated with MeOH, and a portion of this MeOH solution was treated with diazomethane and then examined by GLC. Of the three possible benzenedicarboxylic dimethyl esters, only dimethyl terephthalate was found, showing that 18 was the para isomer.

***N*-Methyl-2-(4'-pentylphenyl)ethylamine (19).** Compound 19 was prepared from 4 in 19% yield by methods analogous to the preparation of 18 from 5. Recrystallization of the maleate salt from *n*-PrOH–Et_2O gave white crystals, mp 111–113°. Anal. ($C_{18}H_{27}NO_4$) C, H, N.

Oxidation and subsequent methyl esterification of 19 (cf. compound 18) showed only dimethyl terephthalate when examined by GLC, showing that 19 was the para isomer.

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Mode of Action and Quantitative Structure–Activity Correlations of Tuberculostatic Drugs of the Isonicotinic Acid Hydrazide Type[1]

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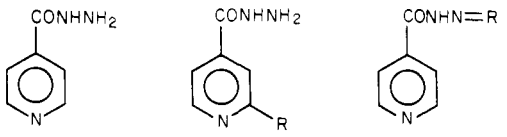
Quantitative structure–activity studies have been performed for a series of 2-substituted isonicotinic acid hydrazides by correlating electronic, steric, and lipophilic properties of the substituents with the biological activity data (MIC) from serial dilution tests with *Mycobacterium tuberculosis* (strain H 37 Rv). The reaction rates for the quaternization of 2-substituted pyridines with methyl iodide were also determined. The rate constants show a similar dependence on the steric and electronic effects of the substituents as the antibacterial activities of the corresponding pyridine-4-carboxylic acid hydrazides. The obtained correlations give evidence that the reactivity of the pyridine nitrogen atom is essential for the biological activity of 2-substituted isonicotinic acid hydrazides and seem to support the hypothesis that isonicotinic acid derivatives are incorporated into an NAD analogue.

Isonicotinic acid hydrazide (I) (INH, isoniazid) has been one of the most effective agents in tuberculosis therapy since 1952, when its action against *Mycobacterium tuberculosis* (*M. tbc*) was first discovered.[2] Although many derivatives of INH have been synthesized, none have demonstrated antibacterial activity greater than INH itself. Most derivatives involve ring substitution at position 2 and 6 of the pyridine moiety (II) or modification of the hydrazine moiety as in the hydrazone structure (III).



I II III

Isoniazid derivatives develop the typical high activity against *M. tbc* only if they can be transformed to INH (I) or to a derivative of type II. Substitution of electron-donating or -withdrawing groups at the position ortho to the ring nitrogen yields compounds with decreased antibacterial action. The hydrazones (III), on the other hand, have activities comparable to INH itself.

The mode of action of INH and its derivatives has not yet been established. The present state of knowledge has been summarized in several recent review articles.[3,4] Among the several proposed mechanisms of action, such as the formation of yellow pigments[5] and inhibition of mycolate synthesis,[6,7] only one hypothesis derives a detailed mechanism that may be tested in a quantitative structure–activity analysis. This is the "isonicotinic acid" hypothesis of Krüger-Thiemer[8–12] (Figure 1).

According to this hypothesis, isonicotinic acid (INA) is responsible for the inhibitory activity of INH against mycobacteria. INH, via passive diffusion, rapidly permeates the bacterial cell membrane, with the hydrazide function serving only as a carrier group. Once inside the cell, the INH is oxidized enzymatically to INA. At the predominant intracellular pH the INA is nearly completely ionized ($pK_a = 4.84$) and therefore cannot leave the cell.[13] This results in the accumulation of INA within the bacterial cell and accounts for the low MIC of INH (1 μM); special binding capacities on the part of the bacteria are therefore unnecessary. Subsequently the accumulated INA, instead of the natural metabolite nicotinic acid (NA), is quaternized and incorporated into an NAD analogue (or a precursor). The NAD analogue thusly produced can no longer function as the natural coenzyme. Disturbance of the normal metabolism (especially lipid metabolism) causes degeneration of bacterial cells (loss of acid fastness) and cell death.[14,15] The following observations, from this and other laboratories, strongly support the INA hypothesis.

1. The oxidative degradation of INH to INA can be performed in vitro by horseradish peroxidase and H_2O_2.[16,17] This is compatible with the observation that INH-resistant strains of *M. tbc* possess only small amounts of peroxidase (and catalase) as compared with INH-sensitive strains.[18,19]

2. Carboxylic acid hydrazides such as benzoic acid hydrazide, nicotinic acid hydrazide (NH), or picolinic acid hydrazide develop an antibacterial activity against *M. tbc* which is at least 200 times smaller than that of INH.[10]