

PHOTOCHEMICAL AND RADIOCHEMICAL ALKYLATION
OF CAFFEINE BY ALKYL AMINES

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Photochemical and radiochemical reactions of purines may serve as model systems of radiation damage to DNA in living organisms. Elad and co-workers (1) have shown that reactions involving alkylation of the purine bases in nucleic acids are closely paralleled by reactions of caffeine. Although purines are relatively stable compared to pyrimidines in interaction with U. V. light (2), several reactions are reported concerning them. Linschitz and Conolly (3) reported the photochemical addition of alcohols to the 5, 6 double bond of purine. Elad and co-workers (4) have reported the substitution at the 8-position of a number of purines by alcohols. Jerumanis and Martel observed the photosensitized reaction of ethers with caffeine to form a similar product (5). Recently, Yang and co-workers (6) have described the alkylation at the 6 position of purine by n-propylamine. In our laboratory we have been studying the photochemical and radiochemical substitution at the 8-position of caffeine by a series of alkyl amines. A solution of caffeine (4.24 g) in 200 ml of 5 M aqueous amine was degassed by bubbling nitrogen gas through the warmed solution. The solution was then irradiated at ambient temperature with either cobalt-60 gamma rays for 48 hours in a glass stoppered pyrex bottle, or photolyzed with a medium pressure Hg-arc (Hanovia 550 w) in a quartz immersion well. After removal of solvent on a rotary evaporator, the reaction mixture was separated on a silica gel column (Ligroin:acetone [4:1]). Analysis by thin layer chromatography, using Baker-flex^(R) silica gel 1B-F prepared plates eluted with chloroform:acetone:ethyl acetate (2:1:1), was used as the guide for combining suitable fractions in the column chromatography.

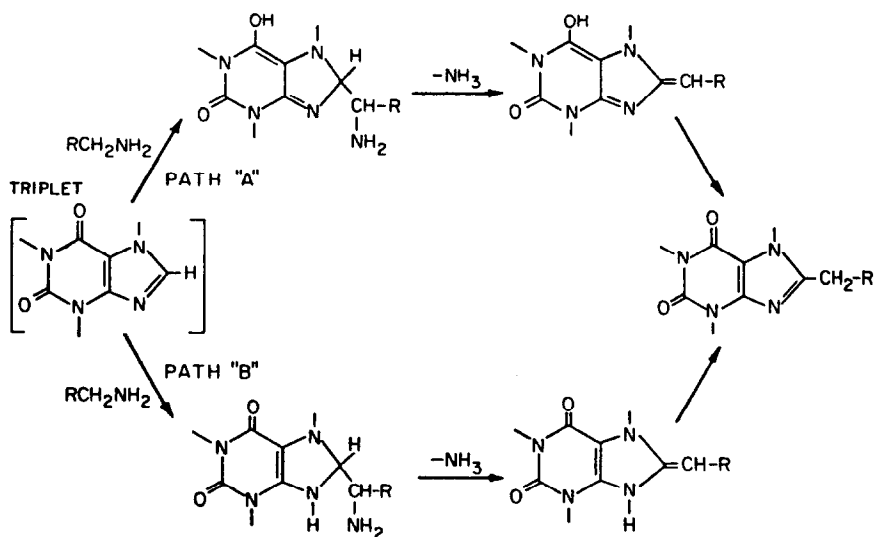
Product identification was based on NMR, mass spectrometry, U. V. spectra, and comparison of the melting point with that reported in the literature. The NMR spectra had the characteristic peaks of the N-methyl groups of caffeine at 3.9, 3.6, and 3.4 δ . In alkylated products, the peak corresponding to the 8-position hydrogen at 7.6 δ was missing and peaks corresponding to the alkyl group added appeared in the appropriate places. Yields were of the order of 5 to 10% of the total caffeine available. The method of work up discriminated against isolation of volatile products.

Table 1 lists the amine used and the product observed. In order to further characterize the reaction, several studies involving the radiolytic reaction were carried out. The paramagnetic ions Cu^{++} and Ni^{++} both show efficient quenching of the reaction leading to 8-alkyl caffeine. While diamagnetic ions such as Mg^{++} and Zn^{++} had no observable effect. This is assumed to indicate a triplet state intermediate (7). Since the overall reaction proceeds under the influence of UV light as well as ionizing radiation, processes involving ionization or fragmentation of the amine as an initial step in the mechanism are not indicated. The unreactivity of t-butylamine implies that the presence of a hydrogen atom on the carbon α to the amino group is necessary. A possible mechanism in accord with the observed data is presented below. The major difference between paths A and B is the site on the caffeine which abstracts the hydrogen atom from the amine. Both carbonyl oxygen and imine nitrogen are known to be able to abstract hydrogen atoms (8), but since the resulting intermediates are tautomeric the question may be moot and the true mechanism may well be a combination of the two paths.

The abundance of naturally occurring amines in biological systems and the sensitivity of genetic information to chemical reactions of DNA moieties suggest reactions, such as those reported here, may have some relevance to radiation damage in living systems.

TABLE 1

<u>AMINE</u>	<u>PRODUCT</u>
$\text{CH}_3\text{CH}_2\text{NH}_2$	8-ethyl caffeine
$(\text{CH}_3\text{CH}_2)_2\text{NH}$	8-ethyl caffeine
$(\text{CH}_3\text{CH}_2)_3\text{N}$	8 ethyl Caffeine
$\text{CH}_3\text{CH}_2\text{CH}_2\text{NH}_2$	8-n-propyl caffeine
$(\text{CH}_3)_2\text{CHNH}_2$	8-iso propyl caffeine
$(\text{CH}_3)_3\text{CNH}_2$	N.R.



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