quantities enhances the staining of enzyme bands. In addition, excess amounts of PMS results in (1) the adherence of fine formazan deposits produced close to the surface of the gel and resulting in a diffuse staining of the gels, and (2) the production of negative bands[15].

Résumé. L'effet de la phenazine methosulfate (PMS) sur l'activité de plusieurs enzymes oxydatifs a été étudié. En tant que transporteur d'électron, la PMS a un rôle important sur la coloration des isoenzymes séparés par électrophorèse en gel d'amidon. En ce qui concerne la déshydrogénase lactique, un excès de PMS provoque une coloration diffuse, alors que dans le cas de la déshydrogénase succinique on observe l'apparition de «bandes négatives» qui n'ont pas encore été décrites.

S. L. MANOCHA, G. H. BOURNE, and M. SANDLER

Yerkes Regional Primate Research Center, Emory University, Atlanta (Georgia 30322, USA), July 4, 1966.

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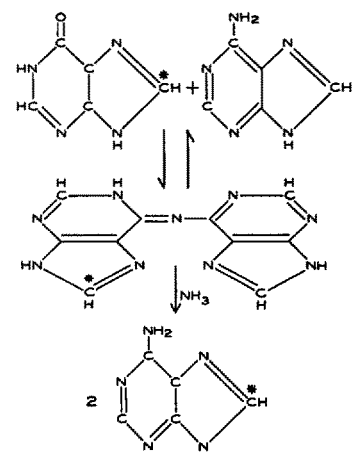
On the Interaction of Hypoxanthine with Adenine[1]

A non-enzymatic conversion of hypoxanthine to adenine was proposed on the basis of spectrophotometric observations obtained from solution mixtures containing adenine and hypoxanthine in 1–5 *M* ammonium phosphate[2]. The syntheses of the Schiff bases of 2-amino-4-methylpyrimidine were said[3] to support the reaction involving a condensation of the keto form of hypoxanthine and adenine through a Schiff base intermediate and its subsequent cleavage by the addition of ammonia to give 2 molecules of adenine. The results of these experiments have been presented to explain a mechanism that could account for the aberration of tetrads observed in nucleic acids.

Certain considerations should be taken into account, however. The concentration of the reactants (about 10^{-3} *M*) makes this reaction highly improbable. In the event that a certain amount of intermediate is formed, the low pH of the reaction mixture is certainly an unfavorable condition for the cleavage of the Schiff base by nucleophylic ammonia molecules (NH_3), which are non-existent at pH 6.

This communication is to verify whether the spectrophotometric measurements described by BOWNE actually represent a transformation of hypoxanthine to adenine. Under the conditions used by BOWNE[2], radioactive C^{14}-hypoxanthine was incubated with non-radioactive adenine in ammonium phosphate, pH 6. If the reaction of hypoxanthine and adenine was to take place, the presence of radioactive adenine would be expected at the end of the reaction (Figure). However, the results of several experiments showed the absence of radioactive adenine in the incubated reaction mixture. No trace of radioactive adenine could be detected by a combined paper chromatographic and X-ray procedure nor could any substantial amount of radioactivity be found in the adenine isolated from the paper (Table).

Radioactive contaminants were found on the paper between the spots of adenine and hypoxanthine which were responsible for the small amount of radioactivity found in adenine. This view was further supported by the fact that the radioactivity of the base was considerably reduced when the adenine isolated from the paper was treated with charcoal Darco G-60 (Table). Taking into consideration the specific activity of the hypoxanthine used in the reaction mixture, the specific activity of the



Mechanism proposed by BOWNE[2] for the conversion of hypoxanthine to adenine as it would take place in the presence of C^{14}-hypoxanthine.

Specific activity

Components	μmoles counted	Radioactivity cpm	cpm/μmole[a]
Adenine	2.3	133	0.0[b]
Hypoxanthine	0.4	$5.5 \cdot 10^4$	$1.37 \cdot 10^5$
Paper control No. 1		147	
Paper control No. 2		124	
Adenine (after charcoal)	3.0	50	6
Hypoxanthine (after charcoal)	0.5	$6.9 \cdot 10^4$	$1.38 \cdot 10^5$
Background		33	

[a] Background corrected. [b] After correction for the amount of radioactivity present in the paper controls No. 1 and No. 2.

[1] This work was supported by United States Public Health Service Grants No. AM-08897, No. AM-10126, and No. 2A-5124; a grant from The Arthritis Foundation (Northern California Chapter); and from the Committee on Research, University of California School of Medicine (San Francisco).

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isolated adenine after charcoal treatment is certainly negligible.

Our work, therefore, does not support the conclusion reached by BOWNE[2] that hypoxanthine and adenine interact with each other in concentrated ammonium phosphate solutions at pH 6 to form adenine.

Materials. Hypoxanthine (puriss) and adenine (puriss) were purchased from Fluka. Radioactive C^{14}-hypoxanthine was obtained from Schwarz Bioresearch Inc. The ammonium phosphates were purchased from Mallinkrodt Chemical Works.

Method. A 1.5 *M* ammonium phosphate pH 6 solution mixture (10 ml) containing adenine and C^{14}-hypoxanthine (10 μcurie), at the final concentration of $1.5 \cdot 10^{-3}$ *M* for hypoxanthine and $1 \cdot 10^{-3}$ *M* for adenine, was incubated under N_2 gas with gentle shaking for 100 h at room temperature. (Solubility of the ammonium phosphates in H_2O is limited to a concentration of about 1.8 *M*.) The mixture was then evaporated to dryness in vacuo at 40 °C and the residue extracted 3 times with 10 ml each of 0.5 *M* ammonium hydroxide in ethanol. This solution was prepared with concentrated aqueous ammonium hydroxide in absolute ethanol. The extraction volumes were combined and concentrated to dryness. The method of extraction was essentially quantitative. The residue containing no inorganic salts was dissolved in the minimal amount of 0.05 *M* aqueous ammonium hydroxide, applied to Whatman 3 MM paper, and submitted to descending chromatography with a mixture of *n*-butanol-H_2O (86:14) for 24 h. On the dry paper, the bases were visualized by an UV-lamp. The distribution of radioactivity was determined by placing the paper on non-screen X-ray film for 1 week. The specific activity of the components was determined in a scintillation counter after their elution from the paper chromatogram with 0.1 *M* HCl. Controls were strips of paper from both sides of the adenine spot which were eluted in the same way as the nucleic acid bases. Both the control paper strips and the paper strip containing the adenine component were of equal area.

Radioactive contaminants from the paper were removed by concentrating the acid eluates to dryness. The residue, dissolved in H_2O, was adsorbed in 100 mg of Darco G-60 and Cellite (1:1). After several washings with H_2O, the bases were desorbed by 25% ethanol in H_2O.

Résumé. Une conversion non-enzymatique d'hypoxanthine en adénine fut proposée sur la base d'observations spectrophotométriques obtenues de solutions contenant adénine et hypoxanthine dans 1–5 *M* de phosphate d'ammonium. Les résultats fournis par les expériences sur la radioactivité ne confirme pas la conclusion émise par BOWNE[2] que hypoxanthine et adénine agissent l'une sur l'autre en solution concentrée de phosphate d'ammonium pour former de l'adénine.

J. LUDOWIEG and J. D. BENMAMAN

Department of Orthopaedic Surgery and the Rheumatic Disease Group, Department of Medicine, University of California, San Francisco Medical Center, San Francisco (California, USA),
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Chlorophyll Stability Index, a Suitable Criterion for Rapid Screening of Tree Provenance in Arid Zones

Information on the provenance of a species is important in determining the best available sources of seeds to give well-adapted productive trees and in directing the breeding of interracial and interspecific hybrids towards adaptation to particular localities[1]. The development of provenance research to assist in the selection of a suitable provenance of a species in any afforestation and reforestation programme therefore ranks high. Among the different species of Eucalyptus tried, *Eucalyptus camaldulensis* has shown good promise for introduction into arid zones[2]. This species has a very wide range of distribution in its native home (Australia) and, as such, offers great scope for selection of suitable provenance which may be best adapted to the arid environment of western Rajasthan in India. In provenance trials, however, it becomes difficult at times to test all the required material in the field for either want of adequate space or of funds. It is, therefore, necessary that some criterion should be established for carrying out rapid screening of a large number of provenances, even in the nursery stage, for selecting the most promising ones for field trials. It is with this objective that the present study on 16 provenances of *E. camaldulensis* was taken up in the nursery of this Institute at Jodhpur to determine whether any relationship exists between the environmental characteristics, viz. mean annual rainfall and mean temperature of these provenances in Australia, with the % seedling survival and drought hardiness as expressed by the chlorophyll stability index[3] (CSI), recorded in the nursery at Jodhpur where seedlings from these provenances were grown under

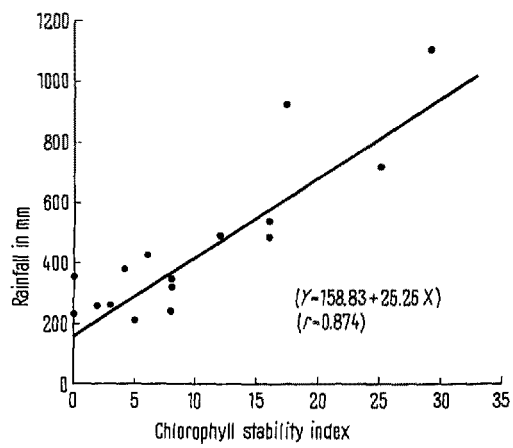

Fig. 1. Relation between chlorophyll stability index and rainfall of different donor localities of *E. camaldulensis*.

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