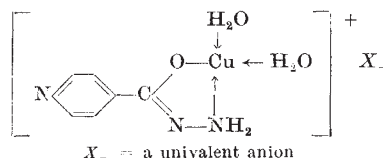


Mode of Action of Isonicotinic Hydrazide

In our earlier report we directed attention to the importance of chelation as an essential step in the anti-tuberculous activity of isonicotinic hydrazide (isoniazid)¹. It was impossible to decide whether the chelating action with a metal or metals took place before (in the medium) or after adsorption on the bacterial cell.

Since then we have had the opportunity of examining the activity *in vitro* and *in vivo* of the relatively insoluble cupric complex of isoniazid:



The results now reported (see Table 1) indicate that this complex is highly active. Thus, it might now be postulated that isoniazid is probably inactive in the absence of a chelating metal, and that its anti-tuberculous activity is, in fact, due to its metal complex, in this instance the cupric derivative. These findings are strongly reminiscent of those obtained in earlier studies of the mode of action of 8-hydroxyquinoline (oxine)².

Table 1. ACTIVITY OF ISO NICOTINIC HYDRAZIDE AND ITS CUPRIC CHELATE COMPLEX

Compound	Anti-tuberculous activity		
	<i>In vitro</i> (ref. 5) Tuberculostatic endpoint. Medium: synthetic serum-synthetic		<i>In vivo</i> (ref. 3) Healing ulcer response
Isonicotinic hydrazide (isoniazid)	2,048,000 (0.07 μgm./ml.)	2,048,000 (0.07 μgm./ml.)	20 mgm./kgm. weekly I.M. HR 100 in 28 days.
Cupric complex of isoniazid	4,096,000 (0.08 μgm./ml.)	512,000 (0.6 μgm./ml.)	20 mgm./kgm. weekly I.M. HR 99 in 28 days.

It will be noted that the activity *in vivo*, examined by the healing-ulcer technique previously reported³, is extremely high, two injections effecting 85 per cent cure (HR value 85) in three weeks and 99 per cent cure in 28 days after three injections.

This copper derivative appears to be as active as isoniazid, verazide⁴ or any drug we have tested so far. However, it exhibits a profound local and general toxicity in mice and guinea pigs. During treatment of guinea pigs by intramuscular injection, a severe gangrenous necrosis and ulceration developed at the site of injection, and in mice the acute toxicity (LD₅₀) was as low as 36 mgm./kgm. body-weight compared with 200 mgm./kgm. for isoniazid itself.

The interest in these findings centres on the demonstration that the copper-isoniazid chelate complex is active *in vitro* and *in vivo* under conditions in which it is unlikely to break down to isoniazid. This observation, taken in conjunction with our earlier findings¹, has led us to conclude that the anti-tuberculous activity of isoniazid is expressed after it has formed a chelate metal complex in the medium or the animal body in which it is acting. The identity of the complexing metal *in vitro* or *in vivo* is still unknown, and the activity of other metal complexes is being investigated.

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S. D. RUBBO
JANICE EDGAR

J. CYMERMAN-CRAIG
G. N. VAUGHAN
D. WILLIS

School of Bacteriology,
University of Melbourne.

Department of
Organic Chemistry,
University of Sydney.

Nov. 4.

¹ Cymerman-Craig, Rubbo, Willis and Edgar, *Nature*, **176**, 34 (1955).

² Rubbo, Albert and Gibson, *Brit. J. Exp. Path.*, **31**, 425 (1950).

³ Rubbo and Pierson, *Amer. Rev. Tuberc.*, **68**, 48 (1953).

⁴ Rubbo and Cymerman-Craig, *Nature*, **176**, 887 (1955).

⁵ Cymerman-Craig, Rubbo and Pierson, *Brit. J. Exp. Path.*, **35**, 478 (1954).

Reduction to Normal Levels of the High Erythrocyte Sedimentation-Rates in Apparently Healthy South African Bantu Men

In the tropics and semitropics, unusually high values for erythrocyte sedimentation-rates are common among apparently healthy individuals. Thus, among seventy-five medically fit young Indian medical students, the mean rate (Wintrobe's method)¹ was 11.2 ± 9.4 , range 0.5–30 mm.; high values are assumed to be pathological, although the cause was unknown².

Neither malnutrition nor under-nutrition (unless very severe) is believed directly to influence erythrocyte sedimentation-rate³. Nevertheless, we find that although very high rates are common among different regional groups of apparently healthy Bantu mine-workers, the mean high values become reduced to within normal limits after subjects, without any medical treatment, have consumed for four-fifteen months the excellent diet provided in the mine compounds.

The Bantu mine-workers were eighteen to forty years old, in good physical condition, not anemic, and free from radiological tuberculosis. No significant enlargement of liver, spleen or lymph nodes was apparent. Subjects were variously parasitized, particularly the tropical group. The Kolmer-Wassermann test for syphilis was positive in 4 per cent of cases. The majority showed liver dysfunction as revealed by the common biochemical tests on serum.

Workers came from areas where the diet is usually adequate in calories and gross protein, but is low in animal protein, fat and certain mineral salts and vitamins. The mine compound diet, which may be eaten to satiety, is nutritionally adequate.

The erythrocyte sedimentation-rate was determined on oxalated blood using Wintrobe's method¹, two technicians working in parallel, and determinations being completed within 3 hr.

In Table 1, only in the mixed group do data refer to the same individuals examined at the beginning and end of periods of service.

Our results revealed an equal degree of abnormality of erythrocyte sedimentation-rate among recruits with and without previous periods of service in the mines: this finding suggests that the normal values found on leaving the mines rise again on returning for some months to conditions of tribal life.

The dramatic and unexpected changes in erythrocyte sedimentation-rate found in these outwardly healthy subjects are not due to any medical treat-