if occurring *in vivo*, could produce modified body proteins which might interfere with enzymic activities or cell permeability, or might possess antigenic properties.

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A natural guanine-containing analogue of vitamin B_{12}

A preparation containing two vitamin B_{12} analogues has been obtained from the fermentation broths of a *Nocardia* strain isolated in our laboratory[1].

The extraction from the microorganisms was performed by autoclaving a cell suspension supplemented with cyanide. The substances under investigation have been separated from vitamin B_{12} and factor B, likewise present in fermentation broths, and purified by a procedure involving: adsorption on charcoal, elution in *iso*propanol–water, removal of vitamin B_{12} as dicyanidocomplex in benzyl alcohol, adsorption on ion-exchangers (Amberlite IRC-50, Amberlite IRA-401, Dowex 1) and electrophoresis in a column of Whatman cellulose powder[2]. The eluate from the cellulose column was concentrated *in vacuo* and the red substances were precipitated by acetone addition.

The cobalt content of the product, dried over $CaCl_2$ *in vacuo* at room temperature, was found to be 3.2%; two atoms of phosphorus and one molecule of ribose are present per atom of cobalt.

No benziminazole derivatives were found after hydrolysis by 6*N* HCl. Hydrolysis by 2*N* HCl at 100° for 2–4 h sets free a purine whose ultra-violet spectrum and chromatographic behavior are similar to those of guanine. Differential spectrophotometry of the hydrolysate in acid and alkaline solutions and FOLIN-CIOCALTEU's reaction show that the preparation contains one molecule of guanine per molecule of ribose.

The absorption spectrum of aqueous solutions of the product has maxima at 273, 320, 356, 500, 530 mμ over a wide range of pH values (from 1 to 10). By addition of cyanide to neutral and alkaline solutions the maxima are shifted to 276, 307.5, 367.5, 540 and 580 mμ.

Paper chromatography in butanol–acetic acid–water and paper electrophoresis in 0.5*N* acetic acid followed by bioautography[3] revealed the presence of two growth factors for *E. coli* 113/3. Most of the activity was due to an electronegative component showing an R_F value relative to vitamin B_{12} of about 0.12. The other factor, which is present in smaller amount, has an R_F of about 0.27 and behaves like vitamin B_{12} in electrophoresis. Deamination by HNO_2 apparently does not affect the latter component while the former is changed into a more electronegative growth factor.

A purine, not distinguishable from xanthine in its chromatographic behavior and ultra-violet absorption spectrum, has been found after hydrolysis of the deaminated mixture by 2*N* HCl.

Therefore we may conclude that the preparation consists mainly of a guanine-containing analogue of vitamin B_{12}. The chromatographic and electrophoretic properties of this substance are closely similar to those described for factor C[3].

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