

The favourable effect of vitamin B₁₂ could not be observed in attempts at *in vitro* activation of the creatine-forming system by additions of vitamin B₁₂ solutions.

An incidental observation that emerged in the course of the foregoing studies was a high mortality-rate in the vitamin B₁₂-deficient groups; the correction of thyrotoxic symptoms by vitamin B₁₂ has since been recorded¹².

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Isolation of Sarcosine from an Acid Hydrolysate of Groundnut Protein

A TWO-DIMENSIONAL paper chromatogram of an acid hydrolysate of groundnut protein has shown, in addition to the spots due to the amino-acids previously reported present in groundnut protein¹, a glycine spot ($R_F = 0.32$, 80 per cent aqueous phenol; $R_F = 0.22$, butanol-acetic acid) and an unidentified spot ($R_F = 0.78$, 80 per cent aqueous phenol; $R_F = 0.24$ butanol-acetic acid mixture). The R_F values of the unidentified spot did not correspond with any of the values reported for amino-acids² or amino-sugars³.

Dalgleish, Johnson, Todd and Vining⁴ have recently isolated sarcosine from the antibiotic actinomycin, and it was noted that the R_F values they recorded for sarcosine were similar to those found for the unidentified spot on the groundnut protein hydrolysate chromatogram. A synthetic sample of sarcosine was prepared⁵, and its R_F values using different solvents were measured. In the accompanying table the R_F values of the unidentified spot are compared with the values found for sarcosine.

From the R_F values it seemed very likely that the unidentified spot was due to sarcosine. The unidentified spot was still present on the two-dimensional

	R_F value		
	80 per cent phenol	Butanol-acetic acid (ref. 3)	80 per cent phenol/1 per cent ammonia
Unidentified spot	0.78	0.24	0.75
Sarcosine	0.78	0.24	0.76
Sarcosine values found by Dalgleish <i>et al.</i> (<i>loc. cit.</i>)	0.78	0.25	—

paper chromatogram of the deaminated hydrolysate, obtained by treatment with nitrous acid under conditions which re-form the secondary amine from its nitroso derivative⁴, showing that the spot is derived from a secondary amino-compound.

Final proof that the unidentified spot was due to sarcosine was obtained by isolating sarcosine from the groundnut protein hydrolysate. The groundnut protein hydrolysate was shaken up with charcoal to remove tyrosine and phenylalanine and then fractionated on a column of 'Zeocarb. 215'⁶, when a fraction containing alanine, proline, valine and sarcosine was obtained. This fraction was further separated on a cellulose column⁴, when a fraction containing sarcosine and alanine was obtained. Purification by fractional crystallization from alcohol and a methanol-acetone mixture gave sarcosine (m.p. 209–210° C. decomp.), 2–3 mgm. from 7.5 gm. protein. Cocker and Lapworth⁵ give a melting point of 212° C. (decomp.) for sarcosine.

So far as is known, sarcosine has not been previously reported present in a protein hydrolysate, although it occurs in the free state in biological extracts.

The groundnut protein used in the above experiments was supplied by Imperial Chemical Industries, Ltd., Nobel Division, and was isolated from groundnuts by the procedure outlined by Haworth, MacGillivray and Peacock⁷. It is a mixture of arachin and conarachin. The sarcosine spot was observed in two different samples of groundnut protein isolated by the above method.

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Evidence of a Complex Compound of Cobalt with a Purine Base (Adenine)

It has been known for a long time that most amino-acids have the property of forming complexes with heavy metals, and it has now been shown¹ that the cobaltous ion, Co⁺⁺, combines reversibly with histidine, giving a tetra-co-ordinated compound, cobalto-dihistidine, which takes up oxygen reversibly to form a compound of octahedral configuration, oxy-*bis*-cobalto-dihistidine.