LXXIV. THE WATER-SOLUBLE B-VITAMINS XIV. NOTE ON THE YEAST ELUATE FACTOR OF THE VITAMIN B₂ COMPLEX

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BIOLOGICAL and chemical investigation of the yeast eluate factor [Edgar & Macrae, 1937] had indicated that this factor was probably identical with vitamin B_6 [György, 1935] and factor 1 [Lepkovsky *et al.* 1936], and very potent concentrates of yeast eluate factor had already been prepared when it was reported that factor 1 [Lepkovsky, 1938, 1] and vitamin B_6 [Keresztesy & Stevens, 1938, 1; György, 1938; Kuhn & Wendt, 1938, 1] had been obtained in a pure state.

Dr Lepkovsky very kindly presented us with a generous sample of his crystalline factor 1 and we found that this material completely replaced our yeast eluate factor in the diet of the rat. Since the biological identity of yeast eluate factor and factor 1 was thus established, we did not pursue further our independent experiments on the isolation of yeast eluate factor. However, by submitting our concentrates to the methods of fractionation found successful for factor 1 [Lepkovsky, 1938, 2] we obtained with some difficulty a small amount of a crystalline material which appeared to be identical with the hydrochloride prepared from factor 1. M.P. of our crystals, $201-203^{\circ}$; mixed M.P. with the hydrochloride of factor 1, $202-203^{\circ}$. Our crystals had the same crystalline form as that of the hydrochloride of factor 1 and also gave similar red colorations with FeCl₃ [cf. Kuhn & Wendt, 1938, 2; Keresztesy & Stevens, 1938, 2]. The identity of vitamin B₈, factor 1 and yeast eluate factor is therefore established.

The activity of factor 1 in our rat growth tests for eluate factor

Rats were prepared as previously described [Edgar *et al.* 1938, 1] and received yeast filtrate factor and riboflavin for a preliminary period of 2 weeks. Some of the animals then received an additional daily supplement of $5\mu g$. factor 1, others $10\mu g$. and others $20\mu g$. In all cases increases in the growth-rate of the animals occurred (Table I). The extent by which the growth-rate of the animals was increased by administration of factor 1 was of the same order as that which has been repeatedly observed following administrations of our yeast eluate factor to rats receiving the same diet with the same supplements. Crystalline factor 1 also cured the florid type of rat dermatitis [Chick, unpublished experiments].

The growth-rate increase following administration of $20\mu g$. daily of factor 1 (free base) was somewhat greater than that observed with $10\mu g$. daily, which in turn was considerably greater than that observed with $5\mu g$. daily. The optimal daily requirement of the rat for factor 1 in growth experiments appears therefore to lie between $10\mu g$. and $20\mu g$. daily and is probably nearer $10\mu g$. Lepkovsky [1938, 1] observed optimal growth when he administered $10\mu g$. of factor 1

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Table I. Growth-promoting action of factor 1 for rats receiving a "vitamin B-free" basal diet with supplements of $10-15 \,\mu g$. aneurin, $50 \,\mu g$. riboflavin and yeast filtrate fraction equivalent to $1-2 \, g$. dry yeast

No. of rats	Av. weekly wt. increase of the group in each of 2 preliminary weeks	Av. weekly increase during 2 weeks g.	Factor 1 µg. daily	Av. weekly wt. increase of the group in each of 2 weeks subsequent to dosing with factor 1 g.	Av. weekly increase during 2 weeks g.
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4	17, 12.5	14.7	5	20, 19.8	19.9
5	19.6, 13.6	16.6	10	26.2, 22.8	24.5
4	16.5, 14.5	15.5	20	28, 24.8	26·4
6*	18, 12	15	Yeast eluate fraction = 2 g. dry yeas	24, 25 t	24.5

* Taken from a previous paper [Edgar et al. 1938, 2 (Table II)].

(free base) daily and Dimick & Schreffler [1939] found that $10\,\mu g$. daily produced nearly optimal growth; Kuhn & Wendt [1938, 3] stated that the rate of weight increase observed following administration of $2\cdot 5-10\,\mu g$. daily of vitamin B_6 hydrochloride increased with the dose of the vitamin given.

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