

vertebrates, are: (1) The abundant occurrence in nature of a form of demonstrated hybrid origin, having nearly all of the characteristics of a natural species; (2) the occurrence of a form as females only, over a wide portion of its range; (3) the consistent and abundant production of wholly female and purely matroclinous young; (4) apparent parthenogenesis in nature.

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THE VITAMIN C ACTIVITY OF HEXURONIC ACID FROM SUPRARENAL GLANDS¹

THE isolation and identification of vitamin C (from lemons), first reported by the writers,^{2, 3} has been followed by independent and in part concurrent evidence from other laboratories,^{4, 5, 6, 7, 8} which adds support to our conclusion that vitamin C is identical with the hexuronic acid previously studied as a reducing agent by Szent-Gyorgyi⁹ and Kendall.¹⁰ The earlier papers from other laboratories did not include quantitative assays of the acid prepared from suprarenal glands. Only one feeding level (1 mg per day) was recorded, so that the minimum protective dosage was not evident, and from this point of view there was need of further evidence to answer Zilva's criticism.¹¹ This point was further emphasized by an indication that the original crystals from animal glands could be further purified,¹² even though they were of reasonable purity when tested. Reasoning from analogy between the vitamin C content of lemon juice and its hexuronic acid content is conditioned by a variation of over 100 per cent. in the antiscorbutic activity of the juice and by very little data regarding its hexuronic acid content. The paper by Harris and others¹³ included an assay using a different technique than ours, but their product was evidently about the same as our own in activity. It

is clearly evident from his later publications¹⁴ that Szent-Gyorgyi had no knowledge of the vitamin function of the acid previous to the experimental work published in 1932.

To provide further evidence that the crystalline vitamin was not a case of inactive crystals accompanied by active material, we have made an assay of the hexuronic acid prepared by Dr. E. C. Kendall¹⁵ from suprarenal glands. This product was prepared according to a distinctively different procedure from that used in our laboratory and from a different type of starting material. Hence a quantitative study of its activity provided strong evidence regarding the identity of the vitamin. This product corresponded with our own in crystalline form, melting point, reducing value, titration equivalent and rotatory power. The essay showed strikingly that the two preparations were identical in activity within the limits of measurement. A dosage of 0.5 mg daily provided a minimum protective level with slightly suppressed growth (133 g. gain in 55 days) and with two of five animals showing slight scurvy symptoms (av. score 2). The 0.75 mg level was above the amount required for protection, and the 0.25 mg level was distinctly too low.

The procedure for isolating the hexuronic acid from suprarenal glands involved an ether extraction of the solution when neutralized by sodium bicarbonate. This would have removed such a substance as the o-diphenol derivatives of narcotine, which has been considered by Rygh¹⁶ to be identical with vitamin C.

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