An Apparent Change from Cytoplasmic to Nuclear Control of a Form of Chlorophyll Deficiency in Gossypium arboreum L.

THE occasional occurrence of variegated plants in Gossypium herbaceum race Wightianum has been reported by Gopalratnam¹, and in Gossypium arboreum race sinense by Feng², who also reported typical maternal inheritance.

Variegated plants have frequently been observed at this Institute in G. arboreum race bengalense. On one of these plants a bud from a chlorophyll-deficient area was forced into growth by appropriate pruning, giving rise to two flowers with chlorophyll-deficient boll, calyx, bracts and pedicel. These two flowers were selfed and the pollen also used for pollinating several types of normal green.

Selfing resulted in two progenies with twelve and thirteen yellow seedlings which died shortly after germination.

The crosses of normal green \times deficient gave sixteen plants all of which were green. From the sixteen F_1 plants, sixteen F_1 families

were raised. The F_2 families exhibited two types of behaviour.

(1) Normal Mendelian segregation. Family B6 gave eighty-six plants of which sixty-seven were green and nineteen yellow deficient-a close approximation to the usual 3:1 ratio. The results of F_3 , F_4 and F_5 confirmed the mode of inheritance as monogenic. Chlorophyll deficiency, however, showed a progressive increase in viability, and in F_3 and subsequent generations it was possible to grow deficient plants to maturity. They flowered well in pots, the first year's growth producing yellow-green leaves, while the leaves in the second year were almost full green, with only the growing-point yellow-green. Crosses were then made reciprocally between the new type of viable deficient and normal green. In F_1 green was fully dominant with 3:1 segregation of green to deficient in F_1 , thus: green × deficient, 80:19; deficient × green, 108:29. A slight excess of green plants is found.

(2) Maternal inheritance. The F_2 families from the other fifteen crosses threw only greens—a total of 1,879 plants being recorded. This absence of segregation conforms to the usual type of cytoplasmically determined inheritance.

Therefore, out of sixteen crosses between female green and male chlorophyll-deficient from a deficient region of a variegated cotton plant, fifteen exhibited maternal (cytoplasmic) inheritance, while in the remaining family inheritance was nuclear and monogenic, with mutation from green to deficient in one locus as a possible explanation. This transition from cytoplasmic to nuclear control has apparently not been recorded in other plants. The improvement in viability of deficients from lethality to the condition of fertility is possibly due to progressive reverse plastid mutation.

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¹ Gopalratnam, Madras Agric. J., 20, 151 (1932). ⁸ Feng (in Yu, C. P.), J. Amer. Soc. Agron., 33, 756 (1941).

Synthesis and Properties of S-Alanyl Coenzyme A

S-ALANYL coenzyme A has been prepared by aqueous transfer of the alanyl residue from S-alanyl thiophenol hydrochloride to coenzyme A (CoA-SH), in a manner similar to that used for the synthesis of S-alanyl glutathione¹, and other S-acyl compounds²⁻⁴:

 $PhSCOCH(CH_3)NH_3^+Cl^- + CoA-SH \rightarrow$ PhSH + CoA-SCOCH(CH_a)NH+Cl-

Due to the small quantities of CoA-SH available and the lability of S-alanyl coenzyme A, the product could not be isolated by classical methods, that is, as a metal salt. However, S-alanyl coenzyme A was successfully isolated in the pure state by paper electrophoresis (50 V./cm.), as it migrates at a slower rate than CoA-SH.

50 mgm. (70 per cent pure) CoA-SH and 25 mgm. of S-alanyl thiophenol hydrochloride were dissolved in 0.2 ml. of water. The pH of the solution was adjusted to 5-6 by means of the dropwise addition of 2N sodium hydroxide solution. The reaction solution was then placed in a cold room at 4° C. for three days. The product was separated and purified by paper electrophoresis in a neutral buffer (pyridine : acetic acid : water, 10 : 1 : 89). (It was found necessary to use several 35-cm. wide paper sheets, owing to the low capacity of the paper.) The buffer solution was removed from the electropherogram by freezedrying, and the area which contained S-alanyl coenzyme A was cut out and eluted at low temperature with water. The water solution of S-alanyl coenzyme A was then freeze-dried. The yield was 20 mgm.

A fresh solution of S-alanyl coenzyme A, subjected to paper electrophoresis, exhibited one identical spot when developed with ninhydrin, in ultra-violet light, and with nitroprusside ammonia. The nitroprusside spot was slightly delayed, appearing within 1-2 min. The paper electropherogram of an aged solution also contained a positive ninhydrin spot for alanine.

When a sample of S-alanyl coenzyme A was incubated for 2-3 hr. with an excess of glutamic acid at pH 8 in a sodium bicarbonate buffer solution at 20° C., a new ninhydrin reactive compound arose. This compound we believe to be alanyl-glutamic acid (Fig. 1). In the presence of pigeon-liver homogenate,

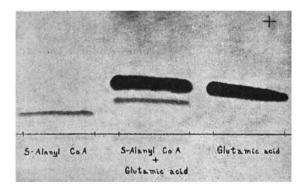


Fig. 1. Electropherogram of S-alanyl coenzyme A – glutamic acid incubation mixture. The formation of alanyl-glutamic acid is shown in the middle column where it has migrated at a slower rate than glutamic acid (right column), but faster than S-alanyl coenzyme A (left column). Composition of incubation mixture : 0.1 ml. of 0.5 M sodium bicarbonate, 0.05 ml. of 0.1 M glutamic acid, 0.05 ml. of 2 mgm./ml. S-alanyl coenzyme A