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Suppression of pyr-3 mutants by arg-12 mutants

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Abstract

Suppression of *pyr-3* mutants by *arg-12* mutants

McDougall, K. J. and V. W. Woodword. Suppression of pyr-3 mutants by arg-12 mutants.

Until recently the arg-12 locus, the locus which structures ornithine transcarbamylase (OTCase), was represented by a single mutant, arg-12^{s} (37301). arg-12^{s} possesses about 3% of wild-type OTCore activity and is capable of suppressing the pyrimidine requirement of pyr-3 mutants characterized by in vitro aspartic transcarbamylase (ATCase) activity. (The pyr-3 mutants used here are denoted by the KS-prefix. KS16 and KS20 ore ATCase^{+}; KS23 and KS43 are ATCase^{-}. The arg-12 mutants ore designated as 6-1, 6-2, 6-3, 6-8 and 7.0.) The mechanism of suppression is thought to be due to metabolic *cross-feeding* of *carbamyl* phosphate (CAP), a substrate common to both pathways. Independent efforts by us and by Davis and Thwaites (1963 Genetics 48: 1551) resulted in the isolation of OTCore-less mutants phenotypically distinguishable from arg-12^{s} by reduced growth rates and by 99% or more reduction of OTCore activity. There mutants were found to be located near arg-5 on linkage group II (Woodword and Schwarz 1964 Genetics 49: B45). It was not possible to demonstrate suppression of pyr-3 ATCase^{+} mutants by the new arg-12 mutants, since the required arginine supplement offsets suppression, possibly by *repression* or inhibition of carbomyl *phosphokinase*, shutting off the *remaining* source of CAP. The effect of exogenous arginine on pyr-3 ATCase^{+}; arg-12 double mutants can be overcome, however, by adding lysine to the culture medium (Houlahan and Mitchell 1947 Proc. Natl. Acad. Sci. U. S. 33: 223). This procedure was employed to demonstrate that the new arg-12 isolates are capable of suppressing pyr-3 ATCase^{+} mutants.

Table 1. Dry weights, in mg, from 125 ml stationary flask cultures at 30°C containing 40 ml of medium; supplements were used at a concentration of 0.3 mg/ml.

Strain	Time (days)	Medium: Minimal	Aroinine	Uridine	Arginine + Uridine	Lysine + Aroinine		
KS20	4	0	0	84.4	79.5	0	0	0
	8	0	0	110.3	110.4	0	0	0
b-9	4	0	85.4	0	86.7	97.7	90.1	97.2
	8	0	75.9	0	78.9	99. 1	102.0	103.8
KS43	4	0	0	90. 1	87.3	0	0	0
	8	0	0	119.4	112.1	0	0	0
KS43;6-9	4	0	0	0	91.3	0	0	0
	8	0	0	0	79.9	0	0	0
KS20;6-9	4	0	0	0	90.2	0	tr	4.1
	8	0	4.9	0	77.6	12.4	24.5	30.0

The data in Table 1 illustrate suppression of a pyr-3 ATCase^{+} mutant (KS20) by an arg-12 isolate (6-9). It is seen that the pyr-3 ATCase^{-} strain (KS43) is not suppressed, which is in agreement with earlier findings (Davis and Woodward 1962 *Genetics* 47: 1075). Suppression is also observed on arginine medium providing the double mutant is cultured for a prolonged period, in much the same way as the RU-suppressors *of* pyr-3 mutants (McDougall and Woodward 1965 Genetics 50: 397). Although data are presented for only two double mutants, suppression was observed in the following combinations involving an ATCase^{+} pyr-3 mutant and an arg-12 mutant: KS16;7-0, KS20;6-2, KS20;6-8, KS20;7-0. In addition, all of there double mutants eventually grew on arginine medium. No suppression was evident in the following ATCase^{-} pyr-3;arg-12 double mutants: KS23;6-1, KS23;6-3, KS43;6-1, KS43;6-3.

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