A GENE AFFECTING THE RATE OF PYRIMIDINE DEGRADATION IN MICE[1]

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IN mammals, the principal pathway of uracil catabolism is by way of reduction to dihydrouracil, followed by ring opening to form β-ureidoproprionic acid, and then decarboxylation to produce β-alanine, NH_3, and CO_2 (CANELLAKIS 1956; FINK 1956; FINK, MCGAUGHEY, CLINE and FINK 1956; FRITZSON 1957; FRITZSON and PIHL 1957; RUTMAN, CANTAROW and PASCHKIS 1954). This pathway is shown in Figure 1. Uracil degration takes places primarily in the liver, and the carbon in the number 2 position of uracil (indicated by an asterisk in Figure 1) is released as respiratory CO_2.

The initial purpose of the investigations reported in this paper was to determine whether inbred mouse strains differed in rates of uracil degradation as measured by the production of $C^{14}O_2$ from uracil-2-C^{14}. An interstrain difference was found. The strains could be classified as having either a rapid or a slow rate of uracil catabolism, without overlap of extreme values for either class.

FRITZSON (1957) reported that the rates of conversion of uracil to dihydrouracil, dihydrouracil to β-ureidopropionic acid and β-ureidopropionic acid to β-alanine were in the ratio of 0.15:13:10 in rat liver slices. Therefore, the observed interstrain differences in mice could have been due to a change in rate of the first reaction only. To determine whether the second and third enzymes were also involved, the rates of $C^{14}O_2$ formation from dihydrouracil-C^{14} and β-ureidopropionic acid-C^{14} were measured. The combined results from tests made in vivo and in vitro were interpreted as showing that a single gene affected all three reaction rates, so that they were consistently and uniformly more rapid in some

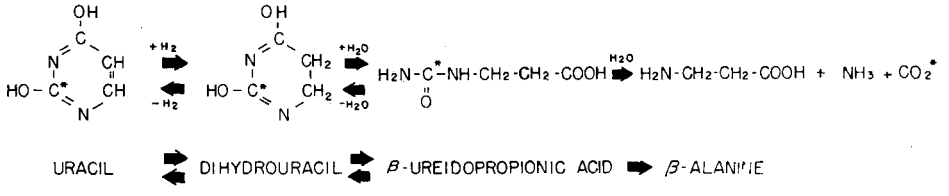

FIGURE 1.—The pathway of uracil degradation. The number 2 carbon of uracil, which is released in CO_2, is indicated by an asterisk.

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mouse strains than in others. The name Pyrimidine degrading, with symbols Pd^a for the slow rate and Pd^b for the fast rate, is proposed for this gene.

Thymine, like uracil, is degraded via a dihydropyrimidine, a β-ureido acid, and finally a β-amino acid (CANELLAKIS 1956; FINK, CLINE, HENDERSON and FINK 1956; FRITZSON 1957). To determine whether *Pd* affected thymine catabolism also, the release of respiratory $C^{14}O_2$ from thymine-2-C^{14} was measured in a few, selected inbred mice and hybrids. In every case the individual mice exhibited either rapid rates for uracil and thymine or slow rates for both substrates.

Livers from young rats, from birth to 11 days old, do not degrade uracil or thymine. The capacity to degrade pyrimidines appears at about 12 days after birth and increases to near adult levels at 30 days (STEVENS and STOCKEN 1960). For comparison, the age at which adult levels of the uracil degrading enzymes are attained in mice were determined in a Pd^a/Pd^a and a Pd^b/Pd^b strain.

MATERIALS AND METHODS

Inbred mice were obtained from the Production Department of The Jackson Laboratory or from research colonies maintained by investigators at this laboratory. Hybrid mice were produced under the authors' supervision in their animal colonies.

Uracil-2-C^{14} (U-C^{14}) and thymine-2-C^{14} were purchased from a commercial source. β-Ureidopropionic acid-C^{14} (UPA-C^{14}) labeled in the ureido group, was prepared by condensing β-alanine with potassium cyanate-C^{14}. The UPA-C^{14} was heated with HCl to yield dihydrouracil-2-C^{14} (DHU-C^{14}).

Assays in vivo: A brief preliminary description of this procedure has been published (DAGG 1963). The rates of production of respiratory $C^{14}O_2$ from the four radioactive substrates were measured in vivo with commercial instruments (Dynacon 6000, Nuclear-Chicago) consisting essentially of a continuous flow ionization chamber connected to an electrometer and a graphic recorder. Mice were assayed singly by injecting one of the substrates intraperitoneally and placing the mouse immediately into a cylindrical glass chamber. Compressed air, from commercial cylinders of water-pumped gas, was directed through a column of Drierite, to remove moisture, and into the mouse chamber. Gas flow was regulated to one liter per minute. The effluent gas was redried and passed through the ionization chamber.

The apparatus was calibrated with commercial standardized samples of $BaC^{14}O_3$. Weighed samples of $BaC^{14}O_3$ and a small amount of water were placed in a flask connected to the inlet and outlet gas tubes, and dilute HCl was dripped onto the suspension. Areas under the curves drawn by the recorder were plotted against the amounts of C^{14} in the samples and a straight line relation was obtained.

Results obtained from the animals were converted to μmoles of $C^{14}O_2$ by using the equation describing the standardization curve. A maximum rate of $C^{14}O_2$ production for each mouse was computed from measurements of areas under its recorded curve at successive intervals of time. The maximum rates were reached in 10 to 15 min after injection, and for some strains, these rates persisted for 10 to 30 minutes. For others, the maximum rates were transitory, and for these the fastest rate for a 4 to 5 min interval was calculated. The differing characteristics of the curves were dependent upon dosage, substrate, and strain of mice.

Mice tested with more than one substrate were allowed one to two weeks between injections to eliminate stored radioactive compounds which might interfere with the subsequent assays and to permit pyrimidine metabolism to return to normal in case it had been altered.

Assays in vitro: The enzymes, dihydrouracil dehydrogenase (DHUase), dihydropyrimidinase (DHPase), and 3-ureidopropionase (UPAase) were assayed by the method described by CANELLAKIS (1956) using U-C^{14}, DHU-C^{14}, and UPA-C^{14} as substrates. High speed supernatant fractions (105,000 $\times$ *g*) prepared from mouse livers homogenized in 0.5 parts 0.1M phosphate buffer,

pH 7.5, were used as the source of enzymes. From one to three livers were used for the assays of adult animals. For the tests on neonatal mice, 12 to 20 pooled livers were used. Incubation was carried out in air at 37°C in a reaction mixture containing 0.5 ml of enzyme preparation, 1.0 ml of 0.05 M Tris(hydroxymethyl)aminomethane (Tris) buffer at pH 7.5, 5 μmoles of reduced triphosphopyridine nucleotide (NADPH), 0.625 μmoles of U-C^{14} (0.25 μcurie) and distilled water to make a total volume of 3 ml. When DHU-C^{14} and UPA-C^{14} were used as substrates the NADPH was omitted. In some cases, the substrate concentrations were increased to obtain reaction rates which were independent of substrate concentration. Aliquots (0.5 ml) were removed after 0, 7.5, 15, and 30 minutes of incubation. Trichloroacetic acid was added to stop the reactions, and an aliquot of the mixture was plated onto stainless steel planchets, dried, and counted. The rate of substrate utilization was linear for the first 10 minutes in the liver supernatants from mice of strains with rapid catabolic rates and for about 30 minutes for strains with the slow rates.

RESULTS AND DISCUSSION

Assays in vivo: Initially, in the tests for interstrain differences, U-C^{14} was injected intraperitoneally at 5 mg/kg in 0.4–0.6 ml containing 1.14 μc/ml. The mice were adult males and females, 13 to 30 weeks of age, weighing 22 to 30 grams. The results are presented in Table 1. The strains were separable into two distinct groups. Respiratory $C^{14}O_2$ was liberated at average maximum rates of .055–.065 μmoles/min by strains SJL/J, RF/J, and BUB/Bn. In contrast, an average maximum rate of .009–.020 μmoles/min was obtained for strains BDP/J, SWR/J, C3HeB/FeJ, A/J, C57BR/cdJ, C58/J, LP/J, and PL/J. To the latter group can be added strains 129/Rr, BALB/cGn, BALB/cJ, C57BL/6J, C57BL/10J, and C57BL/Ks, which were reported previously (DAGG 1963).

The strains that produced $C^{14}O_2$ rapidly also released a much larger fraction of the total injected C^{14}. By 2 hours after injection, $C^{14}O_2$ was barely detectable

TABLE 1

Maximum rates of production of respiratory $C^{14}O_2$ from uracil-2-C^{14} by 15 strains of mice

Strain	Number	$C^{14}O_2$ production, μmoles × 10^{-3}/min*	
		Average	Range
BUB/Bn	5	76	67–85
RF/J	4	53	45–63
SJL/J	18	51	41–71
BDP/J	2	20	18–21
C57BL/6J	5	19	16–25
AKR/J	2	18	17–18
C3HeB/FeJ	3	18	15–20
PL/J	3	16	13–17
BALB/cJ	3	14	13–18
LP/J	4	14	11–16
P/J	1	14	.....
SWR/J	3	13	12–16
C58/J	1	12	.....
A/J	2	10	8–11
C57BR/cdJ	1	8	....

* Uracil-2-C^{14} was injected intraperitoneally at 5 mg/kg body weight.

by this apparatus under these conditions. But during this 2 hr period, 70 to 80 percent of the injected C^{14} had been recovered from SJL, RF, and BUB mice, whereas only 35 to 50 percent could be accounted for in the strains with slower rates.

To study the inheritance of the interstrain differences, crosses were made between SJL and C57BL/6 and between RF and BDP (Table 2). The F_1 mice from reciprocal crosses of these paired strains produced $C^{14}O_2$ at rates which were indistinguishable from those for SJL or RF. These data were consistent with the interpretation that the factors which determined the capacity to degrade uracil at the faster rate were completely dominant to factors for the slower rate, in vivo.

A simple dominant-recessive relationship was questionable when the shapes of the recorded assay curves were examined. The curves for most animals with the slow rate showed clear 15 to 25 minute plateaus during the period of maximum rates, indicating that uracil was present in excess of amounts required for expres-

TABLE 2

Maximum rates of production of respiratory $C^{14}O_2$ from uracil-C^{14}, dihydrouracil-C^{14} and β-ureidopropionic acid-C^{14} by inbred mice and hybrids

		Respiratory $C^{14}O_2$, μmoles × 10^{-3}/min					
		Rapid rate			Slow rate		
Substance injected	Mouse strains and hybrids	Number	Average	(SD)	Number	Average	(SD)
Uracil*	RF/J	6	59	(5.4)			
	BDP/J				5	17	(4.2)
	(BDP × RF)F_1	6	60	(3.3)			
Dihydrouracil*	RF/J	6	41	(4.1)			
	BDP/J				5	14	(3.0)
	(BDP × RF)F_1	6	44	(3.0)			
β-Ureidopropionic* acid	RF/J	6	7.5	(1.1)			
	BDP/J				5	2.2	(1.8)
	(BDP × RF)F_1	6	6.3	(1.0)			
Uracil	SJL/J	15	57	(9.9)			
	C57BL/6J				15	21	(6.6)
	(C57BL/6 × SJL)F_1	12	63	(7.1)			
	(C57BL/6 × SJL)F_2	44	62	(8.9)	14	20	(6.7)
	(C57BL/6 × F_1)BC_1	56	64	(8.8)	66	21	(6.5)
	(C57BL/6 × BC_1)BC_2†	18	61	(9.1)	21	20	(7.2)
Dihydrouracil	SJL/J	8	40	(5.0)			
	C57BL/6J				8	10	(6.6)
	(C57BL/6 × SJL)F_1	10	41	(5.0)			
β-Ureidopropionic acid	SJL/J	10	6.6	(1.5)			
	C57BL/6J				6	2.4	(1.1)
	(C57BL/6 × SJL)F_1	10	4.7	(0.8)			

* All three substrates were injected intraperitoneally at 1.14 μmoles per mouse.
† The BC_2 mice were obtained from crosses between C57BL/6 and (C57BL/6 × F_1)BC_1 which had shown the rapid rate of uracil degradation.

sion of this maximum rate. In contrast, the strains with fast rates and the F_1 mice generated curves with relatively sharp peaks, without plateaus, suggesting that they would be capable of more rapid rates with larger amounts of uracil. To determine whether the SJL and the (SJL × C57BL/6)F_1 could be distinguished from each other at higher doses, additional mice were injected with either 10, 20, or 40 mg/kg. The average values for strain SJL at these doses were .097, .123 and .150 μmoles/min, respectively. The average values for the F_1 mice at the same doses were .111, .103, and .149 μmoles/min. The curves recorded for 20 and 40 mg/kg showed that uracil was present in excess. It was concluded that, under these conditions, the factors controlling the fast rate were dominant, in vivo. The assays with liver supernatants, however, which are presented later, showed that the F_1 mice had intermediate values, and therefore the alleles exhibited incomplete dominance.

The average rate for C57BL/6 mice injected with 10 or 20 mg/kg was not significantly different for the rate at 5 mg/kg.

Inspection of the data for strain SJL and the (SJL × C57BL/6)F_1 treated with 5 mg/kg revealed that the heavier mice, which received proportionately larger absolute amounts of uracil, tended to have proportionately faster rates. Therefore, in order to avoid possible misleading results, and for convenience, all subsequent tests were made with a fixed dose: 0.125 mg of U-2-C^{14} per mouse (approximately 5 mg/kg) at 4.56 μc/mg of uracil (Table 2).

Among the 58 F_2 mice derived from (SJL × C57BL/6)F_1, 44 showed the rapid rate and 14 the slow rate after injection with U-C^{14}. The expected values for single-factor inheritance in this cross are 43.5 rapid and 14.5 slow. The BC_1 was produced by mating C57BL/6 to (C57BL/6 × SJL)F_1. In this group, 56 showed the rapid rate and 66 showed the slow rate; which can be compared to an expected ratio of 61 to 61 ($P = .5–.3$). The BC_2 was produced by mating C57BL/6 to BC_1 animals which degraded uracil at the rapid rate. The observed ratio of rapid to slow was 18 to 21, and the expected ratio was 19.5 to 19.5 ($P = .8–.7$).

From these observations, it was concluded that the difference between strains SJL and C57BL/6 in the rates of uracil degradation was controlled by a single gene. The name Pyrimidine degrading is proposed, with symbols Pd^a for the slow rate, as typified by C57BL/6, and Pd^b for the rapid rate, characteristic of SJL.

If the slow rates of degradation in the various strains were due to repressors, and if the genes controlling the repression were not closely linked, then matings between slow strains might be expected to produce an F_1 with a rapid catabolic rate. Matings were made between C57BL/6 and BDP, and all ten of the F_1 which were assayed had the slow rate. Such data support the conclusions that strain BDP, like C57BL/6, is Pd^a/Pd^a.

Since the enzymes involved in pyrimidine catabolism have been shown to be nonspecific toward a variety of pyrimidine substrates (Reichard and Sköld 1963), it was expected that thymine would be degraded rapidly in vivo by Pd^b/Pd^b and Pd^a/Pd^b animals and degraded slowly by Pd^a/Pd^a. Thymine-2-C^{14} was injected into SJL, C57BL/6, (SJL × C57BL/6)F_1 and (C57BL/6 × F_1)BC_1. The rates were rapid in SJL and F_1 and were slow in C57BL/6 (Figure 2).

Among BC_1 mice which had been selected on the basis of their rates for uracil, the rates for thymine were rapid in Pd^a/Pd^b and slow in Pd^a/Pd^a. These data support the conclusions that the Pyrimidine degrading genes also affected the rate of thymine degradation, and add further support to the evidence previously cited that these pyrimidines are degraded by the same series of enzymes.

DHU-C^{14} and UPA-C^{14} were injected intraperitoneally to determine whether the other enzymes involved in this sequence: DHPase and UPAase, were affected by the *Pd* locus. The DHU and UPA were given at 1.14 μmoles per mouse, as was uracil. The results are presented in Table 2 and Figure 3. The rates of $C^{14}O_2$ production from DHU-C^{14} and UPA-C^{14} were rapid in Pd^b/Pd^b and Pd^a/Pd^b mice, and slow in Pd^a/Pd^a mice.

It is important to note, however, that DHU and UPA were degraded more slowly than uracil. FINK (1956) and FRITZSON (1957) have made similar observations and have suggested that the more polar compounds, DHU and UPA, are more slowly transported across cell boundaries. It should be pointed out, also, that endogenous DHU and UPA (formed in situ from injected uracil) were more

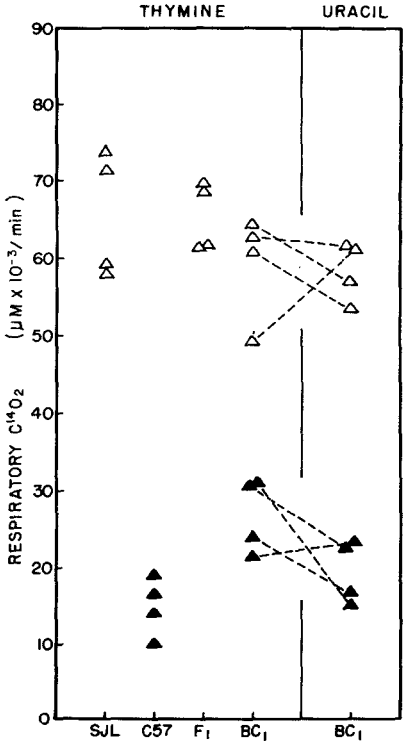

FIGURE 2.—Maximum rates of production of respiratory $C^{14}O_2$ by mice injected with 1.14 μmoles of thymine-2-C^{14}. The BC_1 mice were derived from crosses between C57BL/6 and F_1 animals. The BC_1 mice were selected on the basis of their rates of $C^{14}O_2$ production from uracil-2-C^{14}. The values obtained with uracil and thymine in each animal are indicated by connecting dotted lines.

rapidly utilized than exogenous DHU and UPA. Thus, the relative rates of degradation of the injected substrates, i.e. U > DHU > UPA, can be partially accounted for by the unequal rates of transport of these substances to the site of degradation.

The data obtained in vivo could have been interpreted in two ways. First, that in mice, unlike rats, the last enzyme in the sequence, UPAase, was rate-limiting and that *Pd* affected only UPAase. Second, that the first enzyme, DHUase, was rate-limiting and that *Pd* affected all three enzymes. The second interpretation is supported by the assays in vitro which showed that the relative activities of the enzymes were DHUase > DHPase > UPAase, and that the activities of all three enzymes were affected by *Pd*.

Assays in vitro: Table 3 shows the reaction rates in vitro of the three enzymes involved in pyrimidine degradation. The enzyme preparations from the SJL mice had much greater activities for all three enzymes when compared with similar preparations from the C57BL/6 strain. Hybrid mice (C57BL/6J × SJL/J)F_1 had intermediate activities for all three enzymes. In both of the inbred strains and in the hybrid the relative amounts of the three enzymes increased in the following sequence: DHUase < DHPase < UPAase.

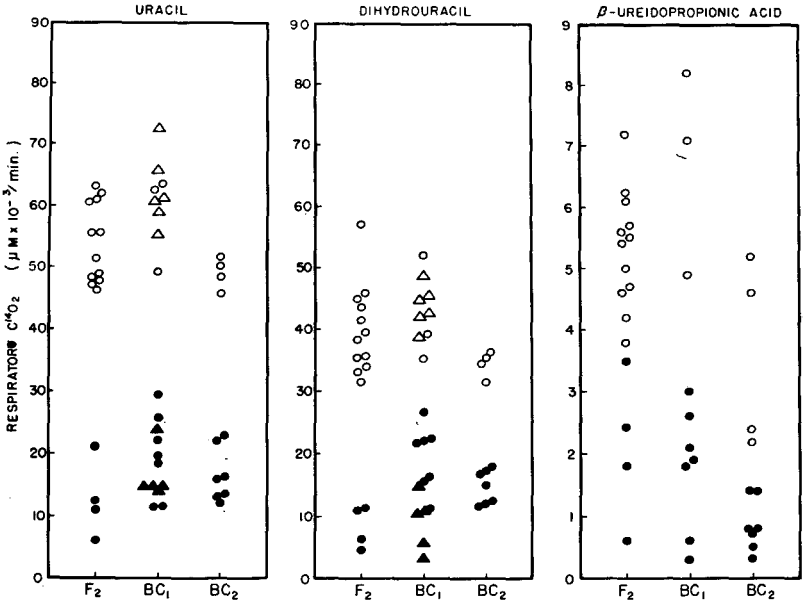

FIGURE 3.—Maximum rates of respiratory $C^{14}O_2$ formation from radioactive uracil, dihydrouracil, and β-ureidopropionic acid by F_2, BC_1, and BC_2 mice derived from crosses between inbred strains SJL/J and C57BL/6J. The BC_1 were offspring from matings between C57BL/6 and (SJL × C57BL/6)F_1. The BC_2 were offspring from matings between C57BL/6 and BC_1 mice which showed the rapid rate of $C^{14}O_2$ formation from uracil. All three substrates were injected intraperitoneally at 1.14 μmoles/mouse. The circles represent animals tested with all three substrates while the triangles stand for animals tested with uracil and dihydrouracil only. The empty circles and triangles represent mice which formed $C^{14}O_2$ rapidly from uracil and the dark circles and triangles represent those which formed $C^{14}O_2$ slowly.

TABLE 3

Rate of conversion of labeled substrates to $C^{14}O_2$ by various genotypes of mice

Strain	Substrate		
	Uracil	DHU	3-UPA
SJL/J	.035 (.027–.042)*	.074 (.060–.086)	.120 (.10–.15)
C57BL/6J	.010 (.009–.011)	.013 (.012–.015)	.016 (.013–.020)
(SJL/J × C57BL/6J)F_1	.021 (.018–.025)	.056 (.053–.060)	.097 (.077–.113)

* Figures represent μmoles of substrate converted to CO_2 per minute per one gram of liver. Figures in parentheses represent range of values.

The genetic evidence suggested that the rate of pyrimidine catabolism in intact mice was controlled by a single genetic factor, but it was impossible to establish whether the gene was controlling only the rate-limiting step or all three enzymes in the sequence. The data obtained from assays in vitro show that the rate-limiting step is the first reaction in the sequence, an observation in agreement with that found in rats (CANELLAKIS 1956). Further, since the activities of all three enzymes segregated together and in a predictable fashion it appeared that the three enzymes were controlled by the same genetic locus.

Although the genetic analysis was carried out for the most part in vivo, certain classes of animals were assayed in vitro for one or all of the three enzymes after classification had been made in vivo. In all cases the results of the classifications made in vivo were confirmed. Furthermore, it became evident that those mice of backcross generation (C57BL/6J × F_1) which were classified as high in vivo (Figure 3) were intermediate in activity with respect to all three enzymes in vitro, and gave activities corresponding to those obtained from F_1 hybrid mice. These mice would be of the genotype Pd^a/Pd^b and such intermediate values would be expected if Pd^a and Pd^b exhibited incomplete dominance. The assays with liver supernatants allowed a study of the reciprocal backcross generation (SJL/J × F_1) since the homozygous Pd^b/Pd^b could be distinguished from the heterozygous Pd^a/Pd^b. Twelve of these BC_1 animals, all having rapid uracil degradation in vivo, were studied with respect to DHUase and UPAase activity; five of these proved to be high with respect to both enzymes studied and therefore homozygous, and seven were intermediate and heterozygous for the *Pd* gene.

The classification of UPAase activity in vivo was ambiguous in some cases, probably due to poor absorption of UPA. In these cases, the results from six F_2 and six BC_2 animals were verified by analysis in vitro. The activities in vitro were 0.12, 0.097 and 0.016 μmoles/min/gm liver for genotypes Pd^b/Pd^b, Pd^a/Pd^b, and Pd^a/Pd^a respectively. In no case was there disagreement as to classification between the two assay systems.

A control mechanism that segregates as a single factor and which controls the activity of a series of reactions is unusual. For this reason experiments were undertaken to eliminate the possibility that the gene was controlling one step only, i.e., the rate-limiting reaction in the sequence. If the last enzyme, UPAase, was

the rate-limiting step then the addition of excess UPAase should increase the rate of uracil catabolism in C57BL/6 mice. Attempts to fractionate the three enzymes from mouse livers were unsuccessful and consequently pure UPAase was not available. However, supernatants from SJL liver, which contained three times the concentration of DHUase, had about seven times the concentration of UPAase as did preparation from C57BL/6 livers. Thus, the concentration of UPAase in C57BL/6 supernatants could be effectively doubled by the addition of SJL/J supernatant while increasing the concentration of the DHUase by less than 50 percent. Such additions produced only additive responses, equal to what would have been expected if DHUase alone had been added. This confirmed the conclusion that the conversion of uracil to dihydrouracil was the rate-limiting step in the sequence, and showed that the concentration of UPAase alone was not preventing maximum degradation of uracil in C57BL/6 liver supernatants. The 1:1 mixtures of liver supernatants from the two strains produced only an additive response of giving values very similar to those found in the F_1 hybrids. Addition of boiled SJL/J supernatant solution to the C57BL/6J supernatant had no stimulating effect which ruled out a lack of essential unknown cofactors as a cause of the decreased activities observed in C57BL/6 supernatants. Similarly no inhibitor was found associated with boiled C57BL/6 supernatant solution.

To see whether the *Pd* alleles acted by controlling the structures rather than the amounts of the enzymes, Michaelis constants were determined for the DHUase in supernatants from each strain of mice. The values obtained with uracil as substrate (1×10^{-4} M for C57BL/6 and 9.5×10^{-5} M for SJL) suggested that the DHUases from each source were enzymatically very similar. Thus the differences in rates represented a difference in actual quantity of the enzyme per unit weight of liver rather than a qualitative change.

The changes in DHUase activity with age are shown in Figure 4. Newborn mice of both strains had essentially no activity. The activity increased linearly after birth until 6 days, at which time the C57BL/6 strain had reached its adult value (0.1 μmoles/min/gm liver). After this time, the enzyme level remained the same in C57BL/6 mice but continued increasing in the SJL strain. At 14 days approximately 80 percent of the adult value was present in SJL mice.

A variety of mechanisms can be proposed to explain the apparent one-factor control of the activities of three enzymes. Although it seems unlikely, the possibility that the interstrain differences were controlled by three separate mutant genes, which were closely linked, cannot be completely ruled out. The assumption that there are not three distinct enzymes but, instead, a single macromolecule with three active sites, one for each of the three substrates, that is controlled by *Pd* also seems improbable because others (see REICHARD and SKÖLD 1963) have fractionated liver from various sources and established that three enzymes are involved in these reactions. Since the first reaction is rate-limiting, the lack of an adequate concentration of the reaction product (DHU) may have prevented the induction of the second and third enzyme of this sequence. However, attempts to induce a higher level of DHPase and UPAase in C57BL/6 mice with DHU were without effect.

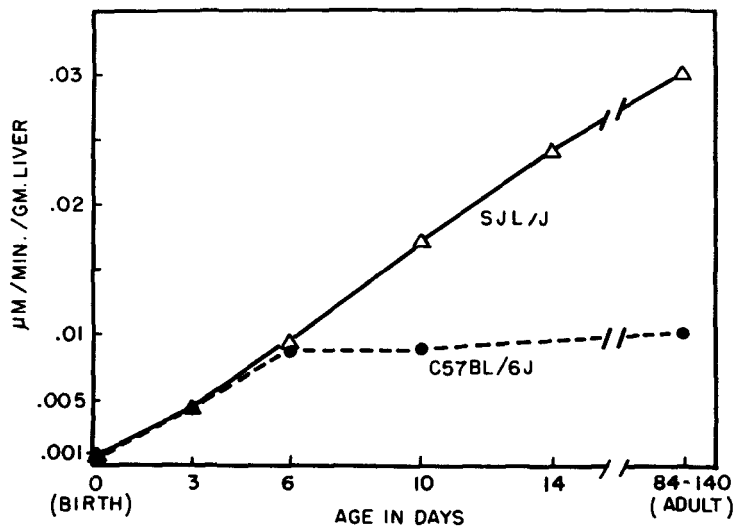

FIGURE 4.—Development of dihydrouracil dehydrogenase in C57BL/6 (Pd^a/Pd^a) and in SJL (Pd^b/Pd^b) mice. Activity units are μmoles of uracil-C^{14} degraded to CO_2 per minute per one gram wet weight of liver.

More likely possibilities for the mode of action of *Pd* involve control over the rates of synthesis or inactivation of the three enzymes simultaneously. In considering enzyme inactivation, it may be necessary to assume that the half-lives for the enzymes are considerably different, because the *Pd* alleles do not have proportional effects on the three enzymes. The rates of synthesis of the three enzymes may be controlled simultaneously if *Pd* is an operator gene, or if it is a regulator gene which determines the production of a repressor substance acting upon the operator (JACOB and MONOD 1961). Suggestive evidence for the production of such a repressor substance by *Pd* comes from the study on the development of the enzyme DHUase in baby mice where it is seen that something occurs in C57BL/6 mice (Pd^a/Pd^a) at six days of age which slows the rate of accumulation of the enzyme, resulting in an equilibrium concentration much lower than is seen in SJL mice (Pd^b/Pd^b). It is also possible that the primary action of the *Pd* gene is not directly involved with the synthesis of these enzymes at all. Instead, this gene may be exerting its control over three structural genes (possibly not even linked) through its control of another enzyme or pathway of which we are unaware.

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SUMMARY

The rates of catabolism of uracil-2-C^{14} to $C^{14}O_2$ were measured in vivo in several strains of inbred mice. A slow rate of degradation was found in strains C57BL/6J, BDP/J, SWR/J, C3HeB/FeJ, A/J, C57BR/cdJ, C58/J, LP/J, PL/J, 129/Rr,

BALB/cGn, BALB/cJ, C57BL/10J, and C57BL/Ks. A rapid rate of degradation was found in SJL/J, RF/J, and BUB/Bn. The F_1 mice from crosses between C57BL/6 and SJL and between BDP and RF showed a rapid rate of uracil degradation in intact animals and an intermediate rate in the supernatant fraction of liver homogenates. Data obtained from the F_2, BC_1, and BC_2 generations prepared from strains C57BL/6 and SJL were interpreted as showing that the interstrain differences in rates of uracil degradation were controlled by a single pair of alleles exhibiting incomplete dominance. The catabolism of two degradation metabolites of uracil: dihydrouracil and β-ureidopropionic acid, were assayed in vivo and in vitro. In every animal, the rates of degradation for all three substrates were either uniformly slow or uniformly rapid, and therefore it was concluded that a single pair of alleles affected the activities of three separate enzymes: dihydrouracil dehydrogenase, dihydropyrimidinase and 3-ureidopropionase. The name Pyrimidine degrading, with symbols Pd^a for the slow rate and Pd^b for the fast rate is proposed for this gene. The possible mechanisms by which *Pd* could affect these enzymes were discussed.

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