and according to another view, by carboxylation to succinate.[3]

We have compared the metabolic patterns of propionate and lactate in rat liver slices with the aid of C^{14}-labeled compounds and a radiochromatographic technique.[4] In general the metabolic patterns of lactate resembled those of acetate. As was also observed by Daus, *et al.*,[5] propionate carbon appeared in all compounds labeled by acetate and, in addition, in β-hydroxyvalerate. However, the distribution of propionate carbon in these compounds differed greatly from that of lactate or acetate. Thus the incorporation of propionate carbon into fatty acids and ketone bodies was much lower, and that into glucose and especially in succinate was much higher than was the case with lactate.

The effects of malonate inhibition on the metabolism of propionate and lactate differed strikingly. As shown in Table I, malonate inhibited greatly

TABLE I

EFFECT OF MALONATE ON THE METABOLISM OF PROPIONATE AND LACTATE

200 mg. of rat liver slices incubated at 37° for 2 hours in 2 ml. of Krebs phosphate buffer, *p*H 7.6, in Warburg flasks with KOH in center wells. Gas phase, air. The flasks contained 10 μm. of propionate-1-C^{14} or 10 μm. of *dl*-lactate-1-C^{14}. Malonate concentration, 0.02*M*. The incorporation of C^{14} in the various compounds was determined as described in ref. 4.

		Per cent of added C^{14} in:					
Substrate	Malonate	CO_2	Succinate[b]	Beta hydroxyvalerate	Glucose	Alanine	Others[a]
Propionate-1-C^{14}	−	9.2	0.1	1.5	2.8	0.5	2.3
	+	1.6	0.9	1.4	0.1	0.1	1.0
Lactate-1-C^{14}[c]	−	22	0	0	0	13	0
	+	26	0	0	0	6	0

[a] Contains also isosuccinate. [b] Lactic and glutamic acids, glutamine, and small amounts of other compounds. [c] It should be noted that under other conditions, such as those used in ref. 4, lactate-1-C^{14} is incorporated into glucose, glutamate and other compounds.

the $C^{14}O_2$ yield from propionate-1-C^{14} but enhanced that from 1-C^{14}-lactate.[6] Also, in the case of the former, labeled succinate accumulated, but none whatsoever could be detected in experiments with lactate-1-C^{14}. Our results indicate that, in rat liver, propionate is not metabolized via acrylate and lactate. These results are, however, consistent with the carboxylation of propionate to succinate.

When the labeled spots on the chromatograms corresponding to succinate were eluted and rechromatographed with unlabeled succinate carrier, it was noted that although the radioactive area and the acid area as determined by indicator spray overlapped quite closely, the superimposition was not perfect. Upon closer study it was established that the "succinate" spot was composed of two acids; the major component was succinate and the smaller was another acid which moved with succinate in many common chromatographic systems. Under optimal conditions the mixture was resolved, and the minor component identified as isosuccinic (methylmalonic) acid. The best solvents for resolution of these two acids are described by Kalbe.[7]

C^{14}-labeled isosuccinate was isolated from a large scale experiment and its identity established by (a) co-chromatography with carrier isosuccinate in three different solvents; (b) crystallization to constant specific activity and (c) preparation of derivatives. In a series of three experiments, it was found that in the presence of malonate about twice as much succinate as isosuccinate is formed. Thus, it appears that rat liver contains enzyme systems that can add carbon dioxide to the β- and α-carbons of propionate, yielding succinate and isosuccinate, respectively.

Wolfe has shown (*Fed. Proc.*, **14**-1, 306 (1955)) that in rabbit liver homogenate propionate is metabolized via succinate. The biosynthesis of isosuccinate from propionate and carbon dioxide by pig heart homogenates also has been reported by Flavin (*ibid.*, **14**-1, 211 (1955)).

(3) H. A. Lardy and R. Peanasky, *Physiol. Res.*, **33**, 560 (1953).

(4) J. Katz and I. L. Chaikoff, *J. Biol. Chem.*, **206**, 887 (1954).

(5) L. Daus, M. Meinke and M. Calvin, *J. Biol. Chem.*, **196**, 77 (1952).

(6) J. Felts and M. J. Osborn in this laboratory have found that the addition of malonate increased greatly the incorporation of C^{14} of pyruvate-2-C^{14} into fatty acids by rat liver slices.

(7) H. Kalbe, *Z. physiol. Chem.*, **297**, 19 (1954).

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THE Si–F BOND MOMENT

Sir:

In a recent paper by Altshuller and Rosenblum[1] estimates of the dipole moments of the bonds H–Si, R–Si, and C–Si were based on a value of 2.3 *D* for the moment of the Si–F bond obtained from infrared intensity measurements on SiF_4.[2] While this value seems a not unreasonable one to choose for the static Si–F bond moment, the grounds for basing it on infrared intensity data are weak. Evidence is presented here that the infrared bond moment in SiF_4 is 3.3 *D* rather than 2.3 *D*; it is also emphasized that the infrared bond moment may be somewhat different from the static moment.

On the first point, it is to be recalled that the infrared intensity data yield two alternative values of μ_{SiF}, *viz.*, 2.3 and 3.3 *D*, respectively. A precisely similar situation exists in CF_4, SF_6 and in the in-plane vibrations of BF_3, the intensities of which have recently been measured.[3]

Table I shows the alternative values of μ and $\partial\mu/\partial r$ for these molecules. They have been collected into two sets, the first set containing the solutions having high values of μ and low and positive values of $\partial\mu/\partial r$. The strong resemblance between the individual solutions of set (1), and the irregularity of the solutions in set (2) favor the selection of set (1). In addition it is not possible to conceive of a simple mechanism whereby $\partial\mu/\partial r$ should be negative (with respect to μ, assumed to be positive, $^{+}X–F^{-}$) and of such magnitude as is found in set (2).

Further evidence exists in the case of BF_3 to support the higher bond moment. The intensity of the out-of-plane bending vibration yields a

(1) A. B. Altshuller and L. Rosenblum, THIS JOURNAL, **77**, 272 (1955).

(2) P. N. Schatz and D. F. Hornig, *J. Chem. Phys.*, **21**, 1516 (1953).

(3) D. C. McKean, unpublished work.

B–F moment of 1.7 *D.*: considerations of orbital following suggest that this is equal to or less than the static bond moment. The structural resemblances between these molecules make it fairly certain that μ_{SiF} should be taken to be 3.3 *D.* It may be noted in passing that there is an approximate correlation between the value of μ in set (1) and the quantity Δr_0 where Δ is the electronegativity difference between the atoms in the bond and r_0 is the bond length.

On the second point, it may be predicted that the static Si–F bond moment is actually less than the vibrational one. The model used is one in which the orbitals of the central atom rehybridize during the vibration so as to continue to point in the direction of the fluorine atoms. In the motion in which three Si–F bonds rotate away from the fourth, orbital following may be achieved by an increase in the p character of the three bonds and a decrease in that of the fourth.

The negative charge on the three atoms will then tend to increase on the three moving fluorine atoms and to decrease on the fourth, resulting in a vibrational bond moment greater than the static one.

TABLE I

		BF_3 (E' class)	CF_4	SiF_4	SF_6
Set (1)	μ	2.6	2.4	3.3	2.7
	$\partial\mu/\partial r$	4.0	3.4	3.7	3.9
Set (2)	μ	0.9	1.1	2.3	0.6
	$\partial\mu/\partial r$	−6.1	4.9	−7.5	−6.6
		(A_2'' class)			
	μ	1.7			
	Δr_0[a]	2.6	2.0	3.4	2.4

[a] Δ = electronegativity difference (Pauling), r_0 = bond length.

It must be emphasized that the bending of bonds may produce considerable electron rearrangement throughout a molecule, particularly where multiple bonds and unshared valency electrons are present,[4] and that in no case yet has the identification of vibrational and static bond moments been justified.

(4) D. F. Hornig and D. C. McKean, Symposium on Bond Moments, 125th National Meeting Am. Chem. Soc., Kansas City, Missouri, 1954.

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ENZYMATIC AMINATION OF URIDINE TRIPHOSPHATE TO CYTIDINE TRIPHOSPHATE[1]

Sir:

No pathway of synthesis of cytidine nucleotides has been previously elucidated. With an enzyme purified about 45-fold from extracts of *Escherichia coli* B evidence has now been obtained for the amination of UTP[2] to yield CTP in a reaction involving NH_3 and ATP, as illustrated in equation (1).

$$UTP + NH_3 + ATP \longrightarrow CTP + ADP + Pi \quad (1)$$

With this enzyme preparation, uracil, uridine and U5P were totally inactive, and with UDP the reaction rate was approximately half of that with UTP. Whether UDP serves as an amino acceptor without prior phosphorylation to UTP is not yet known. No reaction occurred in the absence of ATP except when ADP replaced it, in which case the rate was 12% of that with ATP. The following nucleotide pairs resulted in no cytosine nucleotide synthesis: A5P plus UTP; ADP plus UDP; and ITP plus UTP.

The stoichiometry with regard to the uracil and cytosine nucleotides was studied with the partially purified enzyme (Table I). The formation of UDP, CDP, and nucleoside monophosphates can be explained by the presence in the enzyme preparation of nucleoside diphosphokinase[3] and mixed myokinase.[4] CTP and CDP were identified by their absorption spectra (peaks at 280 mμ, $\lambda 280/\lambda 260$ = 1.99–2.02, $\lambda 290/\lambda 280$ = 0.74–0.75, at *p*H 2), by the molar ratios of cytosine, pentose,[5] acid-labile P, and total P of 1.00:1.01:1.96:3.06,[6] and 1.00:1.00:0.98:2.10, respectively, and by acid hydrolysis

TABLE I

STOICHIOMETRY OF URACIL AND CYTOSINE NUCLEOTIDES

The reaction mixture (64 ml.) contained 6.5 ml. of glycine (1 *M*, *p*H 8.5), 3.2 ml. of $MgCl_2$ (0.1 *M*), 3.8 ml. of NH_4NO_3 (0.2 *M*), 0.50 ml. of ATP (0.075 *M*), 3.2 ml. of UTP (0.012 *M*), and 9.6 ml. of enzyme fraction IV (containing 3.5 mg. of protein). An aliquot of the reaction mixture (29 ml.) was placed immediately in a boiling water-bath for 3 min., the remainder was incubated at 36° for 90 min. and then heated for 3 min. in a boiling water-bath.

	0 min.	90 min.		Δ
	μMoles			
UTP[a,b]	16.8	5.54		
UDP[c]	0.00	2.53	8.95	−7.85 (−7.46)[e]
U5P[c]	0.00	0.88		
ATP[b]	17.4	11.3		
ADP[b]	0.00	4.48	17.2	−0.20
A5P[c]	0.00	1.46		
CTP[d]	0.00	4.60		
CDP[d]	0.00	2.39	7.68 (7.74)	+7.68 (+7.74)
C5P[d]	0.00	0.69		

[a] Anion-exchange chromatography of an aliquot of the reaction mixture (29 ml.) yielded UTP, ATP, and ADP free from each other and the other nucleotides; half of the CDP and U5P fractions were free from each other and the other nucleotides, half were mixtures of the two nucleotides; CTP and UDP were eluted together except in the early fractions of CTP; C5P and A5P were eluted together. [b] Estimated spectrophotometrically at 260 mμ. [c] Estimated spectrophotometrically at 260 mμ correcting for the cytosine nucleotide present. [d] Estimated spectrophotometrically at 295 mμ. [e] Values in parentheses were determined by optical density measurements at 250 mμ (uracil nucleotides) and 295 mμ (cytosine nucleotides) before chromatography.

(1) This investigation was supported by a grant from the National Institutes of Health, Public Health Service.

(2) Abbreviations used: uridine-5′-phosphate, U5P; uridine diphosphate, UDP; uridine triphosphate, UTP; adenosine-5′-phosphate, A5P; adenosine diphosphate, ADP; adenosine triphosphate, ATP; cytidine-5′-phosphate, C5P; cytidine diphosphate, CDP; cytidine triphosphate, CTP; inosine triphosphate, ITP; inorganic orthophosphate, Pi.

(3) P. Berg and W. K. Joklik, *J. Biol. Chem.*, **210**, 657 (1954).

(4) I. Lieberman, A. Kornberg and E. S. Simms, THIS JOURNAL, **76**, 3608 (1954); J. L. Strominger, L. A. Heppel and E. S. Maxwell, *Arch. Biochem.*, **52**, 488 (1954); A. Munch-Peterson, *Proc. Swedish Biochem. Society*, June, 1954; *Acta Chem. Scand.*, in press; D. M. Gibson, P. Ayengar and D. R. Sanadi, Absts., 126th Meeting, Am. Chem. Soc., p. 41-C (Sept. 1954); E. Herbert, V. R. Potter and Y. Takagi, *J. Biol. Chem.*, in press.

(5) Unpublished method of C. E. Carter.

(6) Corrected for 7% contamination with UDP.