help to decide which may have greater importance in the tissue. The much lower K_m for the NADPH_2 -linked reductase would suggest that it may be favoured, but at optimum substrate concentrations the maximal rate of glyoxylate reductase is about twice as great with NADH_2 as with NADPH_2 . Thus more experimental work will be needed before the physiological role of the two reductases in leaves will be better understood.

SUMMARY

1. A new glyoxylate reductase, which catalyses the reduction of glyoxylate to glycollate in the presence of NADPH_2 , has been demonstrated and separated to a large extent from the NADH_2 -linked glyoxylate reductase in both tobacco and spinach leaves. Unlike the NADH_2 -linked glyoxylate reductase of leaves, the new enzyme has a low Michaelis constant for glyoxylate, 0.13 mM, and reacts only slowly with hydroxypyruvate. Pyruvate and oxaloacetate are not substrates.

2. The kinetic properties and favourable equilibrium should make the NADPH_2 -linked glyoxylate reductase a useful reagent for the analytical estimation of glyoxylate. Together with NADH_2 -linked glyoxylate reductase, both enzymes may be used to provide a convenient method for determining the concentration of NADPH_2 and NADH_2 in mixtures of the two dinucleotides.

3. Although there is increasing evidence of the importance of glycollate and glyoxylate in the carbon metabolism and respiration of leaves, especially in sunlight, the physiological role of the glyoxylate reductases remains uncertain.

This work was begun while I. Z. was a Fellow of the John Simon Guggenheim Foundation in the Departments of Botany and Biochemistry, University of Oxford. A. M. G. was a Predoctoral Fellow of the National Science Foundation, Washington, D.C., U.S.A. We wish to thank Professor Sir Hans Krebs and Dr V. S. Butt for their hospitality and encouragement. Partial support by a grant from the National Science Foundation is acknowledged.

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Biochem. J. (1962) **84**, 546

Further Studies on the Entry Rates of Acetate and Glucose in Sheep, with Special Reference to Endogenous Production of Acetate

By E. F. ANNISON AND R. R. WHITE

School of Rural Science, University of New England, Armidale, New South Wales, Australia

(Received 15 January 1962)

The measurement of acetate and glucose turnover in sheep in separate experiments was described by Annison & Lindsay (1961) and Annison & White (1961). Isotope-dilution techniques were used to measure the rates of entry of substrates into body pools and a considerable range of turnover rates was observed in animals maintained on similar diets. These differences between animals, which

were presumably due to differences in endocrine balance, emphasized that the interrelations of acetate and glucose turnover could be more conveniently studied by methods allowing simultaneous measurement of the rates of entry of both acetate and glucose. In the present studies, this has been achieved by the infusion of both labelled acetate and labelled glucose. Measurements of the

rates of entry of glucose and acetate were made on fed and starved sheep.

Isotope-dilution methods have also been used to measure the contribution of the endogenous production of acetate to total acetate turnover. The magnitude of the entry rate of endogenous acetate was found to be related to the concentrations of blood acetate and blood glucose. Raised concentrations of plasma glucose or blood acetate reduced the entry of endogenous acetate, but glucose entry was unaffected by raised concentrations of blood acetate. Annison & White (1961) obtained evidence of inhibition of glucose entry under glucose load.

MATERIALS AND METHODS

Merino wethers were maintained on diets of lucerne (500 g./day—diet 1) or lucerne chaff and maize (400 g./day and 500 g./day respectively—diet 2), as described by Annison & White (1961). The animals were prepared with permanent rumen fistulae.

Experimental procedure

Simultaneous measurement of the rates of entry of glucose and acetate. The general procedure described by Annison & White (1961) for the measurement of the rate of glucose utilization was used, except that both [^{14}C]glucose and sodium [^{14}C]acetate were infused. A priming dose of labelled glucose was used, as described by Annison & White (1961). Equilibrium conditions are established in 40–60 min. during the infusion of labelled acetate (Annison & Lindsay, 1961) and, to minimize the incorporation of acetate carbon into glucose, the infusion of [^{14}C]acetate was not commenced until 80 min. after the start of the glucose infusion, infusions of both substrates being continued for a further 80 min. Labelled acetate was infused at a rate of $0.34\ \mu\text{C}/\text{min.}$, labelled glucose at a rate of $0.17\ \mu\text{C}/\text{min.}$

Preparation of animals with emptied and washed-out rumen. Rumen contents were removed through the rumen fistula, and the rumen was washed out with warm (39°) 0.9% sodium chloride. A solution (3 l.) of salts approximating in composition to that of saliva (McDougall, 1948) was placed in the rumen, and the animal allowed to stand quietly for 1–2 hr. before utilization rates were measured.

Analysis of rumen contents during this period showed that the total volatile fatty acid concentration never exceeded 2 mM, or about 1–2% of the normal post-feed concentration.

Entry of endogenous and exogenous acetate. The rate of utilization of acetate, which is a measure of the total entry of acetate into body pools, may be conveniently partitioned into two fractions: (a) entry of exogenous acetate, i.e. that absorbed from the alimentary tract; (b) entry of endogenous acetate, i.e. that produced by metabolic processes (Davis, Brown, Staubus & Nelson, 1960; Annison & Lindsay, 1961). The entry of exogenous acetate may be roughly equated with acetate produced in the rumen, since acetate production in the remainder of the alimentary tract is relatively low (Annison & Lewis, 1959).

Radioactive materials. Uniformly labelled D- ^{14}C]glucose ([U- ^{14}C]glucose) and sodium [^{14}C]acetate were obtained from The Radiochemical Centre, Amersham, Bucks.

Chemical methods

The methods for measuring the concentration of plasma glucose, the specific activity of glucose by using the glucosazone derivative, and the collection and assay of radioactive barium carbonate were described by Annison & White (1961).

Specific activity of acetate in blood and rumen contents. Special precautions were necessary in the present investigation to avoid contamination of acetate with formate (Annison, 1954a), or with traces of partially steam-volatile keto acids and hydroxy acids derived from labelled glucose. Blood (10 ml.) was deproteinized with metaphosphoric acid, and the filtrate concentrated *in vacuo* to 3–5 ml. (Annison, 1954a). Mercuric oxide (0.2 g.) and 10N- H_2SO_4 (1 ml.) were added to the filtrate, and the mixture was refluxed for 1 hr. The filtrate was then steam-distilled and 100 ml. of distillate collected. After being neutralized (phenolphthalein) with 0.1N-KOH, the filtrate was concentrated by boiling to 3–5 ml., adjusted to pH 3.8–4.0 with M- NaH_2PO_4 (1 ml.) and 2N- H_2SO_4 , and redistilled. Two 100 ml. portions of distillate were collected and titrated, the second portion serving as a blank. Carrier sodium acetate (20 μ -moles) was added to the first distillate, which was evaporated to dryness and oxidized by the method of Van Slyke & Folch (1940), $^{14}\text{CO}_2$ being collected and assayed as described by Annison & White (1961).

Volatile fatty acids in the rumen were estimated by steam-distillation (Annison, 1954b), and assayed for radioactivity by wet oxidation as described for blood acetate.

RESULTS

Cross-labelling of glucose and acetate during the infusion of [^{14}C]acetate and [U- ^{14}C]glucose

Substantial cross-labelling of substrates would vitiate attempts to measure simultaneously the utilization rates of glucose and acetate, which depend on the specific activities of substrates in the blood. Control experiments established, however, that, under the experimental conditions used, incorporation of glucose carbon into acetate carbon, and vice versa, was only 2–5% (Table 1). A much greater degree of incorporation of acetate into glucose was observed when starved (24 hr.) sheep with emptied and washed-out rumens were examined (Table 1).

Utilization of glucose and acetate in fed and starved sheep

The production of volatile fatty acids in the rumen reaches a peak some hours after feeding, and concentrations of blood acetate in this period follow a similar pattern (Annison & Lewis, 1959). Since entry rates can only be measured by constant infusion under equilibrium conditions, when constant rates of entry of substrate are reflected by a largely unchanged concentration in the blood, preliminary experiments were essential to establish the most suitable time after feeding at which to

measure acetate entry. The period of maximum concentration of blood acetate chosen was that when, for 30–60 min., concentrations of blood acetate were fairly constant. During this period, which coincided with the maximum concentration of acetic acid in the rumen, entry of acetate from the rumen was presumably maximal. In the present experiments, infusion of glucose was commenced 120 min. before the period of maximum concentrations of acetate, when specific activity measurements were made. Concentrations of plasma glucose show only slight changes after feeding (Annison & White, 1961).

The results of measurements on fed animals are shown in Table 2. The rates of entry of acetate were considerably higher than those obtained for starved sheep (Table 2). The results obtained with starved (24 hr.) sheep by the simultaneous infusion of glucose and acetate (Table 2) were similar to those obtained by Annison & Lindsay (1961) and Annison & White (1961). In further experiments

the entry rates of glucose and acetate were measured after feeding, and after starving for 24 and for 48 hr. (Table 2).

Measurement of entry of endogenous acetate. The entry rates of acetate and glucose were measured by the double-infusion technique in sheep after feeding (Table 3). The sheep were starved for 48 hr., and the rumen contents of the same animals were then removed and replaced with artificial saliva. The animals were allowed to settle down for 90 min. before the entry rates of acetate and glucose were again measured (Table 3). The rates of entry of endogenous acetate under these conditions were 43–46% of the values obtained for total entry after feeding. The rates of entry of glucose were similar to those reported for fed and starved sheep by Annison & White (1961).

The virtual absence of any entry of exogenous acetate in these experiments possibly affected the endogenous production of acetate, and in a second approach to the problem the entry of endogenous

Table 1. *Cross-labelling of plasma glucose and blood acetate after infusion of labelled acetate and labelled glucose*

Labelled acetate and labelled glucose were infused in independent experiments with sheep starved for 24 hr. Carrier substrates were negligible in all experiments. Glucose was infused for 160 min. after injection of a priming dose of 15 μ C (Annison & White, 1961), and acetate from 80 to 160 min., blood samples being removed for analysis immediately before the end of the infusion. Specific activities are expressed in terms of substrate carbon (C).

Substrate infused	Infusion rate (μ C/min.)	Concn. of blood acetate (mm)	Concn. of plasma glucose (mg./100 ml.)	Sp. activity of acetate (μ C/g. of C)	Sp. activity of glucose (μ C/g. of C)
Glucose	0.17	0.42	49	0.11	8.0
Glucose	0.17	0.47	52	0.09	7.5
Glucose	0.17	0.37	46	0.14	7.2
Acetate	0.34	0.45	61	12.3	0.16
Acetate	0.34	0.41	59	11.7	0.21
Acetate*	0.34	1.37	87	6.0	0.33
Acetate†	0.34	0.34	65	11.2	0.53

* After feeding.

† Starved sheep (48 hr.), with rumen contents removed.

Table 2. *Entry rates of glucose and acetate measured simultaneously in fed and starved sheep*

Results were obtained after feeding (A), and on starving for 24 hr. (B) and for 48 hr. (C).

Sheep	Expt.	Diet	Concn. of plasma acetate (mm)	Concn. of plasma glucose (mg./100 ml.)	Acetate entry rate (mg./min./kg.)	Glucose entry rate (mg./min./kg.)
1	A	1	0.80	85	2.5	2.6
2	A	2	1.31	70	3.9	3.2
3	A	2	1.84	58	4.2	1.9
4	B	1	0.40	54	2.2	1.8
5	B	1	0.45	60	1.7	1.4
6	B	2	0.61	63	1.9	1.7
7	B	2	0.65	70	1.5	1.4
8	A	1	1.05	99	1.6	2.2
8	B	1	0.80	57	1.0	1.8
8	C	1	0.65	45	0.8	1.3
9	A	2	1.67	61	3.9	2.1
9	B	2	0.94	56	2.6	1.6
9	C	2	0.46	54	0.7	1.2

acetate was assessed under conditions of approximately normal entry of exogenous acetate. A solution of labelled sodium acetate (3 l., 500 m-moles, 75 μ C, pH 5.8) was placed in the emptied rumen of a sheep starved for 24 hr., and after 70 min. samples of rumen contents and blood were taken at intervals of 10–20 min. and the specific activity of the acetate was assayed. Comparison of the specific activities of blood acetate and rumen acetate provided a direct estimate of the contribution of the entry of endogenous acetate to total entry. At the relatively high concentrations of rumen acetate employed (150 mM), concentrations of blood acetate rose to 1.5 mM, a value similar to that usually found in fed sheep (Table 2). The ratio of the specific activities of rumen and blood acetate indicated that entry of endogenous acetate initially accounted for only about 25% of total entry (Table 4). This value increased as the concentration of blood acetate declined, but quantitative evaluation was not possible, since the total entry rates at any stage were not known. The contribution of the entry of endogenous acetate to total entry (25%) obtained at concentrations of blood

acetate equivalent to those of the fed sheep (Table 2) was considerably lower than the values (43–46%) obtained from the difference between the entry rates measured in fed animals and in the same animals with emptied rumen (Table 3).

The effect of the entry of exogenous acetate on that of endogenous acetate was examined directly in sheep with emptied rumen by measuring the total acetate entry at increasing concentrations of blood acetate achieved by infusing carrier acetate with isotopic acetate. A marked fall of the entry rate of endogenous acetate was observed only during the first period of carrier-acetate infusion (Table 5). This effect was also seen in sheep starved for 48 hr., but not in fed animals or in animals with relatively high concentrations of blood acetate (Annison & Lindsay, 1961).

Effect of glucose on the entry of endogenous acetate. Sodium [1- 14 C]acetate was infused continuously into starved sheep with emptied and washed-out rumens by the normal procedure for the measurement of the entry rates of acetate. After 70 min., glucose was infused at 166 mg./min. with the labelled-acetate solution, the rate of infusion of

Table 3. *Measurement of the entry of endogenous glucose and acetate*

Entry rates were measured in fed animals (A), and in the same animals after starvation for 48 hr. and with rumen contents removed immediately before the experiment (B).

Sheep	Expt.	Concn. of blood acetate (mM)	Concn. of plasma glucose (mg./100 ml.)	Acetate entry rate (mg./min./kg.)	Glucose entry rate (mg./min./kg.)	Contribution of entry of endogenous acetate to total entry (%)
1	A	1.02	79	3.5	2.54	46
	B	0.31	73	1.6	1.44	—
2	A	1.37	—	4.6	—	43
	B	0.34	—	2.0	—	—
3	B	—	62	—	1.38	—
4	B	0.36	—	1.33	—	—

Table 4. *Assessment of the entry of endogenous acetate under conditions of changing entry of ruminal acetate*

The specific activity of rumen acetate was 6.22 μ C/g. of substrate carbon.

Time after [14 C]acetate placed in rumen (min.)	Concn. of blood acetate (mM)	Concn. of rumen acetate (mM)	Sp. activity of blood acetate (μ C/g. of C)	Contribution of entry of endogenous acetate to total entry (%)
72	1.89	162	4.70	24
90	1.00	150	3.83	38
106	0.86	139	3.44	45
122	0.79	135	3.16	49
138	0.87	119	2.31	63
150	0.86	120	2.80	55
165	0.82	107	2.64	58
180	0.79	98	2.48	60
210	0.72	90	2.40	61
240	0.65	82	2.00	68
285	0.52	70	1.64	74

Table 5. *Effect of the entry of exogenous acetate on the entry of endogenous acetate*

Sodium [$1-^{14}\text{C}$]acetate was infused ($0.34\ \mu\text{C}/\text{min.}$) intravenously into starved (24 hr.) sheep with emptied rumen. The total acetate entry was measured by infusing: (a) labelled acetate without carrier for 0–103 min.; (b) labelled acetate with carrier acetate ($0.49\ \text{m-mole}/\text{min.}$) for 103–210 min.; (c) labelled acetate with carrier acetate ($0.98\ \text{m-mole}/\text{min.}$) for 210–313 min. The weight of the sheep was 33 kg.

Time after start of infusion (min.)	Concn. of blood acetate (mM)	Specific activity of acetate ($\mu\text{C}/\text{g. of C}$)	Mean total acetate entry (A) (m-mole/min.)	Rate of infusion of unlabelled acetate (B) (m-mole/min.)	Entry rate of endogenous acetate (A–B) (m-mole/min.)
90	0.33	11.11	1.33	0	1.33
94	0.40	9.03	—	—	—
97	0.36	9.63	—	—	—
102	0.37	11.68	—	—	—
185	0.46	9.65	1.18	0.49	0.69
189	0.37	12.30	—	—	—
195	0.50	11.37	—	—	—
200	0.50	11.18	—	—	—
210	0.46	13.66	—	—	—
297	0.74	8.63	1.57	0.98	0.59
302	0.75	8.73	—	—	—
307	0.71	9.27	—	—	—
313	0.76	8.43	—	—	—

Fig. 1. Effect of glucose load on the entry of endogenous acetate. Sodium [$1-^{14}\text{C}$]acetate was infused ($0.33\ \mu\text{C}/\text{min.}$) intravenously into a starved (24 hr.) sheep with emptied rumen for the period 0–140 min. Unlabelled glucose was infused during the period 75–150 min. ▲, Specific activity of blood acetate; ○, concentration of blood acetate; ●, concentration of plasma glucose.

[^{14}C]acetate remaining unchanged. Concentrations of blood acetate fell slowly, but the specific activity of acetate increased sharply in each experiment, indicating decreased entry of endogenous acetate (Fig. 1). Similar results were obtained in sheep starved for 48–72 hr.

Effect of acetate on the entry of glucose. Glucose entry was measured in starved (24 hr.) sheep with an emptied rumen, by the usual method, except that after 120 min. sodium acetate was infused with labelled glucose. Increased concentrations of blood

Table 6. *Effect of concentration of blood acetate on glucose entry*

Labelled glucose was infused during the period 0–120 min. at $0.17\ \mu\text{C}/\text{min.}$ (priming dose $15\ \mu\text{C}$). Sodium acetate was infused ($1.22\ \text{m-moles}/\text{min.}$) during the period 120–180 min., the rate of infusion of labelled glucose remaining unchanged.

Time after start of infusion (min.)	Concn. of plasma glucose (mg./100 ml.)	Concn. of blood acetate (mM)	Sp. activity of plasma glucose ($\mu\text{C}/\text{g. of C}$)
90	68	—	8.73
101	67	0.25	8.37
110	66	—	8.92
120	66	—	8.80
131	64	—	9.13
140	62	—	9.17
150	62	0.62	9.02
160	57	—	9.02
170	56	—	10.01
180	55	0.66	9.30

acetate had only a slight effect on glucose entry, the specific activity of blood glucose remaining largely unchanged. The results of a typical experiment are shown in Table 6.

DISCUSSION

The rates of entry of acetate and glucose have been measured simultaneously in fed and starved sheep. The results for glucose were similar to those reported by Annison & White (1961). Measurement of acetate entry in starved sheep is relatively simple (Annison & Lindsay, 1961), but the variable rates of the production of acetate in the rumen after

feeding limits the application of the technique to fed animals. Direct measurements were made, however, during periods of relatively constant concentrations of blood acetate which were determined in preliminary experiments. Correction of the total acetate entry for the entry of endogenous acetate provides a measure of the production of acetate in the rumen. The mean value (three sheep) for the total acetate entry for fed sheep (diet 2) was 4.0 mg./min./kg. (Table 2), or 3.0 mg./min./kg. when corrected for the entry of endogenous acetate (25%). Extrapolation of these values to daily production rates is not possible, however, since acetate entry was measured at the maximum concentration of blood acetate, which reflected maximum concentrations of ruminal acetate and presumably maximum production of acetate. The concentrations of acetate in the rumens of sheep fed once daily reach a maximum after 2-6 hr. and then decline until after the next feeding period. The total daily production of acetate can be roughly calculated from entry rates measured after feeding, and in the starved sheep (by applying appropriate corrections for the entry of endogenous acetate), but more reliable information would be obtained from sheep trained to eat food presented to them at intervals of 2-3 hr. (Moir & Somers, 1957). In these animals roughly constant concentrations of ruminal volatile fatty acids, and thus volatile fatty acid entry, would be achieved. The problem of measuring the ruminal production of acetate under normal feeding conditions remains largely unsolved.

The use of sheep with emptied rumens allowed measurement of the entry of endogenous acetate under various conditions of the entry of exogenous acetate. In the virtual absence of any entry of exogenous acetate, the entry of endogenous acetate amounted to almost half the entry rate observed in the fed animal, but under conditions of approximately normal acetate absorption from the rumen was only about a quarter of the total entry. A direct effect of the entry of exogenous acetate on the endogenous production of acetate was demonstrated, increased concentrations of blood acetate inhibiting the entry of endogenous acetate when examined at low concentrations of blood acetate, but no effect could be demonstrated in fed sheep. Similarly, infusion of glucose into sheep with very low concentrations of blood acetate, brought about by removing rumen content or by starvation, markedly inhibited the entry of endogenous acetate (Fig. 1). Annison & Lindsay (1961) indicated some reduction of acetate entry when the concentration of blood glucose was raised, but these experiments were carried out on animals with only moderately depressed concentrations of blood acetate. The observed effects of acetate and glucose on endo-

genous production of acetate are consistent with the hypothesis that free fatty acids are a major source of endogenous acetate. Concentrations of these acids in the plasma are high in starved sheep and in sheep with emptied rumen (Annison, 1960; Lindsay, 1961), i.e. under conditions where a marked effect of exogenous acetate or glucose on the entry of endogenous acetate can be demonstrated. Increased concentrations of free fatty acids in sheep plasma almost certainly reflect increased mobilization and oxidation of these acids, as in dogs (Armstrong *et al.* 1961), and administration of glucose or acetate is known to lower the concentrations of free fatty acids in sheep plasma (Lindsay, 1959; Annison, 1960), and probably diminishes their oxidation. The absence of a marked effect on the entry of exogenous acetate when the concentrations of blood acetate are reasonably high, and the concentrations of free fatty acids in plasma are low, is consistent with the hypothesis that the acids are a source of endogenous acetate. Some labelling of blood acetate has been observed during the infusion of sodium [$1-^{14}\text{C}$]palmitate in starved (24 hr.) sheep (C. E. West & E. F. Annison, unpublished work). The concentrations of free fatty acids in sheep plasma are raised when concentrations of blood sugar are depressed (Annison, 1960), and the decrease of acetate entry at low concentrations of blood sugar reported by Annison & Lindsay (1961) might appear to be inconsistent with the hypothesis that free fatty acid is a major precursor of endogenous acetate. In these experiments, however, low concentrations of blood sugar were achieved by the continuous infusion of insulin, which inhibits the mobilization of free fatty acids (Bierman, Schwartz & Dole, 1957).

The amounts of acetate absorbed from the alimentary tracts of species (e.g. dog, cat and man) not dependent on the microbial fermentation of polysaccharides as the major energy source are probably very low; concentrations of blood acetate in these species (Annison, 1954*a*) are similar to those observed in sheep with emptied rumens. There is some evidence that acetate turnover in dogs (Ciaranfi & Fonnesu, 1954) and cats (Smyth, 1947) is high and it seems likely that free fatty acids are a major precursor of blood acetate in these species.

Glucose entering the body glucose pool of sheep is almost all derived from endogenous sources (Annison & White, 1961). The relatively high rates of the entry of glucose observed in sheep with emptied rumen (Table 3), or in animals starved for 48 hr. (Table 2), was further evidence that ruminal propionate accounts for not more than half of the glucose entering the body glucose pool (Annison & White, 1961).

Little is known of the factors that control the relative rates of entry and oxidation of acetate,

glucose and free fatty acids in the whole animal, but the use of isotopically labelled substrates has allowed some parameters to be defined.

SUMMARY

1. The rates of entry of glucose and acetate in fed and starved sheep were measured simultaneously by the constant infusion of labelled glucose and labelled acetate.

2. The entry of acetate after feeding was sufficiently constant to allow measurement of entry rates only during the period when a maximum concentration of rumen acetate was attained, which presumably coincided with maximum production and absorption of acetate.

3. The entry of endogenous acetate in sheep with emptied rumens was 40–50 % of the value for the total entry of acetate obtained in the same animals after feeding. Under conditions of normal acetate absorption, the entry of endogenous acetate accounted for about 25 % of the total entry.

4. Raised concentrations of blood glucose or acetate reduced the entry of endogenous acetate in sheep with emptied rumens, or in starved sheep. These results were consistent with the hypothesis that the oxidation of free fatty acids contributes substantially to the entry of endogenous acetate in sheep.

This investigation was made possible by the generous support of the Australian Wool Research Committee. Mr J. Roberts and Mrs J. Musgrave gave valuable technical assistance.

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Biochem. J. (1962) **84**, 552

Formate Metabolism in Sheep

By E. F. ANNISON AND R. R. WHITE

School of Rural Sciences, University of New England, Armidale, New South Wales, Australia

(Received 15 January 1962)

The extensive participation of formate in intermediary metabolism is well established. Hartmann & Buchanan (1959) have reviewed the role of formate in purine metabolism, where interconversions involving C₁-unit metabolism are mediated by tetrahydrofolic acid derivatives. The incorporation of formate into the glucogenic amino acid serine (Sakami, 1948; Kruhoffer, 1951), which also involves activation of the C₁ compound by reaction with tetrahydrofolic acid (see Coon & Robinson, 1958), provides a pathway for formate incorporation into carbohydrate (Wood, 1949), and the entry of formate into acetoacetate (Plaut & Lardy, 1950) and choline (du Vigneaud, Verly & Wilson, 1950) has also been demonstrated. A substantial proportion of formate administered to animal tissues, however, is directly oxidized to carbon

dioxide by a catalase-hydrogen peroxide complex (Nakada & Weinhouse, 1953; Oro & Rappoport, 1959).

The presence of formate in sheep blood was reported by Annison (1954), and McCarthy, Shaw, McCarthy, Lakshmanan & Holter (1958) demonstrated considerable formate production by a perfused goat-liver preparation. In the absence of information on the turnover of this substrate, however, no conclusions could be drawn concerning its metabolic significance. In the present studies the entry rates of formate have been measured by isotope dilution with the constant-infusion and single-injection methods.

Considerable incorporation of formate into plasma serine and plasma glucose occurred after the injection of labelled formate. The distribution