# SPECIFIC DYNAMIC ACTION OF ACETIC ACID AND HEAT INCREMENT OF FEEDING IN RUMINANTS

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*Summary*

Attention is drawn to the lack of adequate explanation of the phenomena of the high specific dynamic action of acetic acid and the high heat increment of feeding in ruminants.

The theory is advanced that the high specific dynamic action of acetic acid is related to the following facts: it is non-glyconeogenic; it is not utilized in protein synthesis; it is oxidized by most, if not all, tissues; and it is utilized significantly for lipogenesis by only a few tissues, excluding in particular muscle and kidney. Furthermore, the metabolism of the acid appears to be relatively "uncontrolled," the uptake by tissues being directly dependent on the arterial level and unaffected by insulin, at least in ruminants, in contrast to glucose.

Finally, there is very little storage of absorbed acetic acid in the body fluids. Consequently, it is metabolized almost as fast as it is absorbed: in some tissues, notably intestinal wall, liver, and adipose tissue and lung, it is partitioned between oxidation and lipogenesis; in others, particularly muscle and kidney, it is of necessity largely utilized oxidatively. The high specific dynamic action of acetic acid indicates that the net partition is in favour of oxidation.

The high heat increment of feeding in ruminants is considered as due to the quantitative importance of acetic acid and butyric acid, which also has a high specific dynamic action, as products of ruminal digestion, and of acetic acid and β-hydroxybutyric acid, derived from acetic and butyric acids, as peripheral metabolites.

## I. Introduction

Two long-recognized phenomena, the high specific dynamic action of acetic acid and the high heat increment of feeding in ruminants have as yet failed to receive any adequate interpretation or explanation.†

The high S.D.A. of acetic acid was first demonstrated by Lusk in 1921 when it was shown that, per unit weight of material, the increase in the metabolic rate of a dog following feeding of acetic acid was very considerably greater than that following feeding of glucose; 3 g. of acetic acid increased the resting metabolic rate by 3.1 cal./hr., and 50 g. of glucose by 4.7 cal./hr. The principle of this observation has been several times confirmed. In nephrectomized dogs, the average excess oxygen consumption following intravenous

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† "Specific dynamic action" and "heat increment" are usually regarded as synonymous but in this paper "specific dynamic action" or "S.D.A." will be used in referring to the effect of specific nutrients or metabolites in increasing metabolism and "heat increment of feeding" to the post-feeding increase in metabolism that follows ingestion of food in general.

injection of sodium acetate was found to be 49 per cent. of that required for complete oxidation of the acetate (Dye and Masters 1944). In the sheep Marston has been reported (Barcroft 1947) as recording a specific dynamic action of 70 per cent. for acetic acid, as against 30 per cent. for the glycogenic propionic acid. In man, continuous intravenous infusion of sodium acetate resulted in the same rate of increase of serum carbon dioxide as did administration of an equivalent amount of sodium bicarbonate (Mudge, Manning, and Gilman 1949), suggesting a very rapid oxidation of the acetate.

The high heat increment of feeding in ruminants, amounting to 30-60 per cent. of the metabolizable energy of the food (gross energy minus energy lost in faeces, urine, and methane) was first described by Armsby and Fries in 1903, and has been repeatedly confirmed (Ritzman and Benedict 1938; Forbes, Braman, and Kriss 1928, 1930; Marston 1949). The findings that acetic acid is a major product of ruminal digestion (Barcroft, McAnally, and Phillipson 1944; Elsden *et al.* 1946; Marston 1948; Kiddle, Marshall, and Phillipson 1951; McClymont 1951*a*), that on a roughage diet virtually no enzymically digestible carbohydrate escapes ruminal fermentation to the volatile fatty acids (Heald 1951; McClymont 1949), and that acetic acid is a major tissue metabolite of ruminants (Reid 1950; McClymont 1951*b*), strongly suggest that the high heat increment of feeding of ruminants may be largely attributable to the high S.D.A. of the acetic acid produced in ruminal fermentation. It has been shown by Marston (1948), by experiments *in vitro*, that the actual heat of fermentation in the rumen could probably account for not more that 15 per cent. of the heat increment. However, despite the importance of acetic acid and heat increment of feeding in ruminant physiology and nutrition, no theory providing any fundamental interpretation or explanation of the phenomena has, as yet, been put forward, although the probable relation of the heat increment to metabolism of the volatile fatty acids produced in the rumen has been suggested by Ritzman and Benedict (1938) and Marston (1948). Also, Reid (1950) has made the significant suggestion that "when fermentation in the rumen is intense, acetate might be absorbed at such a rate that much of it would be oxidized without contributing useful energy to the animal." The picture now emerging of the pathways by which acetic acid and glucose are metabolized and of the metabolism of the volatile fatty acids by ruminants, is considered to provide sufficient evidence on which such a theory may be based.

Firstly a theory is presented concerning the specific dynamic action of acetic acid and then this theory is incorporated in a consideration of the heat increment of feeding of ruminants.

## II. Theory Relating to the Specific Dynamic Action of Acetic Acid

The evidence and arguments involved in this theory are presented hereunder, and summarized in diagrammatic form in Figure 1.

(*a*) *Acetic acid is non-glyconeogenic in animal tissues.* This has been evidenced by a diversity of experiments, including use of phloridzinized dogs (Ringer and Lusk 1910; Deuel and Milhorat 1928) and sheep (Jarrett and Potter 1950), liver glycogen formation experiments (Deuel *et al.* 1937), and failure of acetic acid, but not propionic acid, to affect the insulin coma or hypoglycaemia of sheep (Reid 1951). The appearance of labelled carbon from acetate in all carbon atoms of glucose can be readily explained by the tricarboxylic acid cycle theory without assuming glyconeogenesis (Potter and Heidelberger 1950). Nor is acetic acid utilized as a building block in formation of body proteins (Potter and Heidelberger 1950; Greenberg and Winnick 1949).

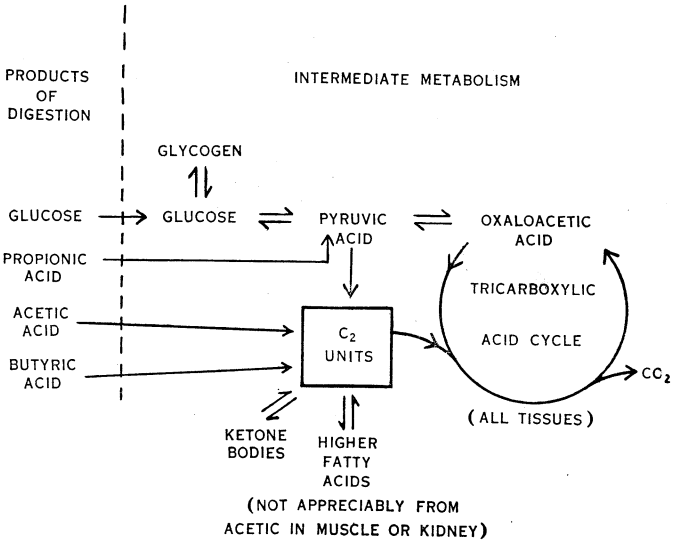


Fig. 1.—The major alternative metabolic pathways available to fatty acids and to glucose. In addition glucose may be utilized in synthesis of the non-essential amino acids.

The irreversibility of the pyruvic acid ⟶ $C_2$ unit step is considered to be the key factor determining the wide difference between the metabolism of glucose and of acetic acid, and the restriction of the $C_2$ unit ⟶ higher fatty acid step to certain tissues a major factor in the high S.D.A. of acetic acid.

(*b*) *Acetic acid is oxidized by many,* possibly all, individual tissues, including heart muscle (Lorber *et al.* 1946; Pearson, Hastings, and Bunting 1949; Pearson *et al.* 1949), diaphragm muscle (Pearson *et al.* 1949), gastrocnemus muscle (Lifson, Omachi, and Cavert 1951), lung, liver, and kidney (Pardie, Heidelberger, and Potter 1950), and mammary gland (Folley and French 1950; Popják *et al.* 1951). Brain has been reported not to utilize acetic acid *in vitro* (Long and Peters 1939) but it is possible that the finding may be due to initial exhaustion of the tricarboxylic acid cycle by which acetic acid is oxidized.

(*c*) *Acetic acid is utilized for fat synthesis.* This is so in the intact body and liver of the rat (Bloch 1947), in the mammary gland of ruminants (Folley and French 1950; Popják *et al.* 1951), in the liver, intestinal wall, and lung of the rabbit and lung of the ruminant (Popják and Beeckmans 1950), and in adipose tissue of the rat (Hangaard and Marsh 1952); but it is not detectably utilized for this purpose by at least heart and diaphragm muscle (Pearson, Hastings, and Bunting 1949; Pearson *et al.* 1949) or kidney of the rat (Pardie, Heidelberger, and Potter 1950; Elliott and Kalnitsky 1950), the kidney experiments showing that practically all acetate metabolized was oxidized. It is also utilized in synthesis of other minor tissue constituents, notably cholesterol and porphyrins (Bloch 1947). It has also been shown that with glucose as substrate the ratio of the conversion *in vitro* of glucose to fatty acids and to carbon dioxide was 1 : 6 in liver and 1 : 8 in muscle (Chernick, Masoro, and Chaikoff 1950). This coupled with evidence of a higher rate of turnover of fatty acids in liver than in muscle (Chernick, Masoro, and Chaikoff 1950), is evidence of a far greater tendency for oxidation than lipogenesis in muscle. The ability of the peripheral tissues, largely muscle, to metabolize acetic acid is well demonstrated by the rapid metabolism of the acid in eviscerated nephrectomized dogs (Dye and Masters 1944).

(*d*) *On absorption acetic acid does not accumulate in the tissues;* and of course, could not be accumulated to any extent without precipitating an acidosis, the maximum content of the blood during the height of ruminal fermentation usually being only about 15 mg. per cent. (Reid 1950; McClymont 1951*b*). That is, acetic acid is metabolized practically as fast as it is absorbed. The extremely high arterio-venous differences of acetic acid in ruminants, of the order of 20 per cent. of the arterial level in the head of the sheep (Reid 1950) and 40-80 per cent. in the lactating bovine mammary gland (McClymont 1951*b*), are reflections of this rapid metabolism.

To summarize, the evidence shows that acetic acid is oxidized by many, possibly all, tissues, but is utilized to a significant extent for lipogenesis by only a small proportion of them, since the muscles, which constitute the largest mass of the body, and the kidney, which is another very metabolically active tissue, apparently are not included in this category; and it is not available in any tissue for glyconeogenesis or protein synthesis. The evidence also indicates that acetic acid absorbed from the gastro-intestinal tract will be successively partitioned between oxidation and lipogenesis by the intestinal wall (at least in the rabbit; there is as yet no evidence of lipogenesis in the intestinal wall of the ruminant), by the liver, and by the lung; then on reaching the general circulation it will be distributed in equal concentration to all tissues, some of which, namely intestinal wall, liver, lung, mammary gland, and adipose tissues, are capable of using it for both lipogenesis and oxidation. But a large proportion of the tissues, at least skeletal and cardiac muscle and kidney, will be capable of metabolizing it to any significant extent only by oxidation. Even in a tissue where acetic acid is lipogenic, as the lactating ruminant mammary gland (Folley and French 1950), it appears that only a small proportion of

the acetic acid utilized is actually used for lipogenesis, the remainder being necessarily oxidized (McClymont 1951*b*).

*It is considered that this rapid metabolism of acetic acid, with partition between oxidation and lipogenesis in lipogenic tissues, and necessarily oxidative metabolism by non-lipogenic tissues, causes a high proportion of all acetic acid to be rapidly oxidized, and that this accounts largely for the high specific dynamic action of acetic acid.*

### *Discussion of S.D.A. Theory*

It is also probable that the specific dynamic action values recorded for acetic acid are underestimates of the amount actually oxidized, since there is an inverse relationship between glucose and acetic acid uptakes, at least by the head tissues of the sheep (Reid 1950): a rise of 2 mg. per cent. in arterio-venous difference of acetic acid depressed glucose uptake by approximately 1 mg. per cent. Competitive depression of metabolism of two-carbon-atom units from glucose by acetic acid might be involved in this example, but a more complex and profound relationship between acetate and glucose metabolism is indicated by the specific effect of acetate in depressing glucose oxidation and stimulating glycogenesis in rat tissues *in vitro* and *in vivo* (Parnas and Wertheimer 1950). However, in the dog the specific dynamic actions of acetic acid and glucose were reported by Lusk (1921) to be independent.

The fundamental divergence in the metabolism of glucose and acetic acid, leading to the marked difference in the efficiency of utilization of energy from the two metabolites, is considered to be that ingested glucose may be stored as glycogen in practically all tissues pending its disposition to oxidation, lipogenesis, synthesis of non-essential amino acids, or formation of oxaloacetic acid for the tricarboxylic acid cycle. Such storage is also at very little energy cost (Baldwin 1947) and pending such storage glucose may be built up to considerable levels in the body fluids without ill effect. Even adipose tissue, under the influence of a sudden large glucose intake or of hyperinsulinism, may store appreciable quantities of glycogen that is later utilized for lipogenesis (Wertheimer and Shapiro 1948). On the other hand, since acetic acid is non-glycogenic and thus cannot be temporarily stored as glycogen, and since it is not stored in the blood, it must, unlike glucose, always be rapidly partitioned between oxidation and lipogenesis.

It would seem that the rapid rate of oxidation of acetic acid by the tricarboxylic acid cycle, resulting as it does in extremely inefficient utilization of the energy liberated, is evidence of a relatively "automatic" or "uncontrolled" oxidation of the acid by the cycle in non-lipogenic tissues. This is in marked contrast to glucose, which is subjected to a multiplicity of enzymic and, until the last step, reversible, degradations to a two-carbon-atom unit, which is then oxidized by the tricarboxylic acid cycle or utilized for fatty acid synthesis.

As acetic acid appears to be the stabilized form of the two-carbon-atom units from pyruvic acid (Coxon 1950) and fatty acids (Bloch 1947) which enter the tricarboxylic acid cycle, it would seem that the metabolic disposal of $C_2$ units in non-lipogenic tissues, being a final step, is not very delicately

controlled. Such a relatively "uncontrolled" metabolism of the $C_2$ unit acetic acid, in comparison with glucose, is evidenced by the straight-line relationships between arterial levels and arterio-venous differences of acetic acid in the head of the sheep (Reid 1950) and bovine mammary gland (McClymont 1951*b*), suggesting a simple mass-action effect, and by the high percentage arterio-venous differences in ruminants, which are not affected, *in vivo,* by hyperinsulinism (Reid 1949; McClymont 1951*b*). Furthermore *in vitro,* insulin does not stimulate lipogenesis from acetate in ruminants although there is some evidence that it may do so in non-ruminants (Balmain and Folley 1951). In contrast, at least in man, arterio-venous differences of glucose are far from directly dependent on the arterial level and are greatly modified by hyperinsulinism and the resultant "opposing factors" (Somogyi 1948, 1949).

Another consideration is that the condensation of glucose or acetic acid to higher fatty acids, being an endergonic reaction, must necessitate an exothermic oxidation of substrate to provide the necessary energy: the energy transfer cannot be completely efficient, as has been discussed by Borsook and Winegarden (1930). There is evidence that when carbohydrate is metabolized under conditions where it must be largely used for lipogenesis, i.e., following massive ingestion or infusion of carbohydrate, the increase in heat production is equivalent to the oxidation of 20 per cent. or more of the glucose metabolized (Rapport, Weiss, and Csonka 1924; Wierzuchowski and Ling 1925; Wierzuchowski 1937). If this figure is applicable to lipogenesis from acetic acid, which has nearly the same combustible energy per gram and the same empirical formula as glucose, and if there is not sufficient "uncontrolled" oxidation of acetic acid in lipogenic tissues to provide the energy required for the condensation of other acetic acid molecules, then this factor also could explain an appreciable proportion of the S.D.A. of acetic acid.

An experimental finding that requires reconciling with the present theory is that where tri-acetin replaced 15 per cent. of glucose in a rat diet on an equicaloric basis, paired feeding experiments over 7 months gave equal results as regards growth and metabolic rates (McManus, Bender, and Garrett 1943), an unexpected finding in view of the high S.D.A. of acetic acid reported by others. However, this result can be reconciled if it is assumed that the food was eaten and the acetic acid hydrolysed from the tri-acetin at such a rate that the acetic acid could join the pool of endogenous "acetic acid" in the liver, which undergoes a rapid turnover (Bloch 1947), without raising the peripheral blood level of acetic acid to any appreciable extent. Without this raised blood level the necessarily oxidative utilization of acetic acid by muscle and kidney tissue would not occur and the net energy value of calories from glucose and acetic acid could be approximately equal.

## III. Theory Relating to the Heat Increment of Feeding in Ruminants

Turning now the problem of the high heat increment of feeding in ruminants, it is considered in the light of evidence cited above that this heat increment is attributable in a very large degree to the high S.D.A. of the acetic

acid produced during ruminal digestion and carried to all tissues in the arterial blood.

However, butyric acid, another major product of ruminal digestion, and a fatty acid with an even number of carbon atoms, will also be metabolized as $C_2$ units. These will follow, perhaps in somewhat different proportions because of the two types of $C_2$ units, acetyl and carboxy-methyl, from butyric acid (Potter and Heidelberger 1950; Kennedy and Lehringer 1950), the same metabolic pathways as acetic acid and will thus also contribute to the heat increment of feeding. The high S.D.A. (Dye and Masters 1944), the non-glycogenicity (Deuel *et al.* 1937), and the lipogenicity (Brady and Gurin 1950) of butyric acid have all been demonstrated. That much of the butyric acid and some acetic acid is probably converted in the rumen wall (Pennington 1951) and the liver to ketone bodies, largely $\beta$-hydroxybutyric acid, which are then metabolized by the peripheral tissues (Shaw and Knodt 1941; McClymont 1949), would not affect the fate of rapid partition between lipogenesis and oxidation. Ketone bodies are metabolized, in at least the fed bovine, at a rate dependent on the arterial level (McClymont 1949), as is acetic acid, and are non-glycogenic (Stadie 1945), which is only to be expected, since they are products of the non-glycogenic acetic acid.

Other minor products of ruminal fermentation such as valeric and hexanoic acids (McClymont 1951*a*; Gray *et al.* 1951) will contribute three or nil carbon atoms for glyconeogenesis, depending on whether they contain an odd or even number of carbon atoms, and the remaining atoms will follow the pathways of acetic and butyric acids.

There is some dispute as to the actual proportions of the volatile fatty acids produced in ruminal digestion (Gray and Pilgrim 1950; Kiddle, Marshall, and Phillipson 1951).

### *Discussion of Heat Increment Theory*

It is intriguing that acetic acid, such a major product of digestion in ruminants, is so inefficiently utilized. It would seem that although plants, e.g. developing oil seeds, can convert fats to carbohydrates and microorganisms can utilize acetic acid as a source of carbohydrate, mammalian (and perhaps all animal) tissues evolved without the enzyme systems capable of converting two-carbon-atom compounds to glucose precursors. The ability of bacteria to carry out carbohydrate synthesis from acetic acid depends apparently on enzyme systems catalysing the condensation of two molecules of acetic to the glycogenic succinic acid (Barron, Ardao, and Hearon 1950). Condensation of formate and acetate to pyruvate has not yet been proven (Strecker 1951). It appears that the only enzyme systems involved in quantitatively significant metabolism of two-carbon-atom units in animal tissues are those involved in the metabolism of such compounds from fatty acids and from pyruvic acid, in oxidation, in acetylation, in synthesis of fatty acids and of some other minor body constituents such as cholesterol, and in metabolism of glycine.

The evolution of ruminants has led to many changes that can be interpreted as adaptations to the radical change in the nature of digestion products occasioned by the circumstance that all feed is first exposed to bacterial digestion. These include low blood glucose, reduced sensitivity to insulin, and small glucose arterio-venous differences (Reid 1951). However, it has apparently failed to lead to an enzymic adaptation permitting glyconeogenesis from acetic acid. If this had occurred there would have been, if the present theory is correct, a considerable increase in the ruminants' low efficiency of utilization of metabolizable energy. It is interesting to speculate upon the effects this might have had on the evolution and characteristics of the ruminant.

Glyconeogenesis from propionic acid, the other major product of ruminal digestion, appears to be due to an enzyme, reported so far in rabbit liver but not in kidney (Huennekens, Mahler, and Nordmann 1951), which may be the result of enzymic adaptation to bacterial digestion. It will be of interest to learn how widely this enzyme is distributed among species with varying degrees of dependence on bacterial digestion. Possibly the enzyme is an adaptation, to the special case of the three-carbon-atom fatty acid, of the enzyme responsible for the normal oxidation of fatty acids, and glyconeogenesis may be only incidental to $\beta$ oxidation of propionic acid, leading via acrylic acid to the glycogenic lactic acid (Huennekens, Mahler, and Nordmann 1951).

It is recognized that some key assumptions have been made in the above arguments. For example, it has been assumed that the finding of lack of appreciable fat synthesis by rat muscle *in vitro* is applicable to ruminant muscle *in vivo*, and that the finding of the non-glycogenicity of butyric acid in rats is true for ruminants. Experimental confirmation of these points and further data on the lipogenicity of various tissues would assist considerably in affirming or denying the validity of the present theory.

The theory suggests some intriguing problems in ruminant physiology. Forbes, Braman, and Kriss (1928, 1930) have, for example, concluded that the net energy of feeds decreases with increasing food intake. Does the rate of ruminal fermentation of food, which may be affected by amount fed, rate of feeding, and physical condition of the feed, influence, through an effect on blood levels of acetic acid, the proportions of the acid wastefully oxidized and used for fat synthesis, or alternatively does it influence the proportions of the glycogenic propionic acid, and the non-glycogenic acetic and butyric acids, and thus influence the net energy value of feed? Or does Marston's (1949) finding of a constant proportion of metabolizable energy lost as heat increment indicate that a constant proportion of acetic acid is always used for lipogenesis?

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