(Research Department, British Vinegars Ltd., Frome, Somerset, England, and Research Station, Long Ashton, Bristol, England).

THE GENUS *ACETOMONAS*

by

J. L. SHIMWELL and **J. G. CARR**

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INTRODUCTION.

It is now known, through the work of LEIFSON (1954), confirmed by SHIMWELL (1958), that the acetic acid bacteria do not comprise, as previously believed, a homogeneous group classifiable in a single genus *Acetobacter* in the family *Pseudomonadaceae* on account of the alleged possession, if motile, of polar flagella. LEIFSON has shown that there are two biochemically and morphologically distinct (and therefore probably phylogenetically unrelated) types of acetic acid bacteria. He has proposed that the genus *Acetobacter* should be retained and reserved for those bacteria which oxidize alcohol through acetic acid to CO_2 and H_2O and which, if motile, have peritrichous flagella (not polar ones as previously stated in the literature). For those which do not oxidize alcohol further than acetic acid, and which, if motile, have polar flagella, he has proposed the appropriate generic name *Acetomonas*. *Acetobacter*, by virtue of its peritrichous flagellation, should be removed from *Pseudomonadaceae*, whilst *Acetomonas* must obviously remain therein.

The genus *Acetobacter*, as redefined by LEIFSON, has recently been studied and re-assessed by one of us (SHIMWELL, 1959) and need not be further reviewed here. The present paper is concerned with the non-acetate-oxidizing genus *Acetomonas*.

HISTORICAL.

The first acetic acid bacterium incapable of oxidizing alcohol further than acetic acid was described by HENNEBERG (1897) under the name *Bacterium oxydans*. It was particularly active towards sugars, the

oxidation of ethyl alcohol being relatively less pronounced than with most (but not all) other types of acetic acid bacteria, rarely exceeding about 2.7% acetic acid, whereas selected strains of acetic acid bacteria found in vinegar and vinegar generators could sometimes produce up to 8% and occasionally more.

In addition to emphasizing the non-oxidation of acetic acid, HENNEBERG also particularly commented on the type of motility observed. This he described as apparently due to polar flagella on account of the direction of cell-motion. He did not, however, demonstrate the type of flagellation by staining. As HENNEBERG went out of his way to mention this probably polar flagellation, and as he did not stress the probable type of flagellation of other species, he seemed to think it notable and perhaps novel. Further comment on his full description of *B. oxydans* will be made later.

In the following year HENNEBERG (1898) described another similar non-over-oxidizing species which attacked precisely the same sugars and alcohols as *B. oxydans*, but differed from that species in rarely giving rise to involution forms, and in producing intense viscidity in liquid media containing dextrin. He regarded this as a separate species under the name *Bacterium industrium*.

BEIJERINCK (1911) isolated from beer another non-acetate-oxidizing acetic acid bacterium which produced, in addition to acid, a brown to black pigment in peptone and similar media containing either glucose or maltose.The colour-intensity of the pigment (which, it is important to note, was water-soluble) varied greatly from strain to strain. He also stressed the rapid oxidation of mannitol and sorbitol respectively to fructose and sorbose. BEIJERINCK named this species *Acetobacter melanogenum*. The further detailed description given by BEIJERINCK will also be discussed later.

BAKER, DAY and HULTON (1912) made a technological study of an acetic acid bacterium producing ropiness in beer. This organism they named *B. aceti viscosum*. It did not over-oxidize acetic acid and the nature of the constituent of beer from which the organism made the slime could not be discovered. Nor did they associate their organism with *Bacterium industrium* Henneberg.

KLUYVER and DE LEEUW (1924) described a species also not oxidizing acetic acid or acetate, and producing much gluconic acid from glucose, and dihydroxyacetone from glycerol. They named this organism *Acetobacter suboxydans*. In later years this name has become very popular, completely obscuring the priority of HENNEBERG (1897)

who was the first to isolate and describe non-acetate-oxidizing acetic acid bacteria. Cultures stated to be "*A. suboxydans*" have become perhaps the most extensively used of all acetic acid bacteria in biochemical studies.

SHIMWELL (1936) isolated from beer the organism responsible for the ropiness thereof. It somewhat resembled the earlier *B. aceti viscosum* of BAKER, DAY, and HULTON, and the starting material from which the slime was produced was later identified (SHIMWELL, 1947) as the residual dextrin in beer. SHIMWELL did not, at that time, associate his "*A. capsulatum*" with HENNEBERG's *B. industrium*.

FRATEUR (1950) recognised that those acetic acid bacteria failing to over-oxidize acetic acid were different from others, but considered that taxonomically this property was more one of degree rather than absolute. He classed such bacteria in his "suboxydans group" as distinct from his (over-oxidizing) "mesoxydans group" (defining them both, however, as ketogenic). He recognised two non-overoxidizing species, namely *A. suboxydans* and *A. melanogenum*, with several varieties of each.

LEIFSON (1954) showed, for the first time, that the non-overoxidizing strains not only differed from other acetic acid bacteria in the type of their oxidative attack on alcohol, but were also morphologically entirely different types of bacteria. This has already been referred to under "Introduction". LEIFSON's flagella photomicrographs proved unacceptable to the editors of BERGEY's Manual (1957), who awaited electron micrographs before accepting LEIFSON's findings.

SHIMWELL (1958) confirmed LEIFSON's work, and published photomicrographs of the peritrichous flagellation of LEIFSON's amended *Acetobacter* genus, and of the polar flagellation of his new *Acetomonas* genus. He also published an electron micrograph of his *A. rancens* strain, confirming the peritrichous flagellation of *Acetobacter* proper (by generous permission of Dr. JAMES M. SHEWAN, of Torry Research Station, Aberdeen, where the micrograph was taken).

ASAI and SHODA (December, 1958) rejected LEIFSON's *Acetomonas* on the alleged grounds that such bacteria "are scarcely able to oxidize ethanol to acetic acid". They suggested the genus "*Gluconobacter*" instead. They also (wrongly) implied priority over SHIMWELL in showing that some strains were monotrichous and that not all were multitrichous, as stated by LEIFSON. SHIMWELL's publication was dated March 1958.

Definitions.

"Over-oxidize". This technical vinegar-maker's term is here occasionally used for the sake of brevity, to mean that alcohol is not only oxidized to acetic acid, but that the latter is then further oxidized to CO_2 and H_2O.

"Ketogenic". This is used, for simplicity, to denote the production of dihydroxyacetone from glycerol, as this is virtually the sense in which it is used by Frateur (1950). "Ketogenic" acetic acid bacteria may also produce 5-ketogluconic acid from glucose, fructose from mannitol, and so on. The production of 2-ketogluconic acid is a property probably common to most acetic acid bacteria except Frateur's "peroxydans group" (Frateur, Simonart and Coulon, 1953). It was not used, however, as a criterion in his classification, and is not so used in the course of this paper.

"Suboxydans Group". This was the term given by Frateur to the 4th of his 4 groups (the highly ketogenic, non-overoxidizing ones). In this paper "suboxydans group" only denotes the lack of over-oxidation of alcohol to CO_2 and H_2O. In our opinion this property alone probably suffices to separate such acetic acid bacteria from all others, as will be seen.

Specific Names. Although we finally propose, after Leifson (1954), that "*Acetomonas*" should be adopted henceforth as the name of the genus of non-overoxidizing acetic acid bacteria, the original names of existing species have been retained throughout this paper until the diagnosis of the genus *Acetomonas* comes to be given at the end. This is to enable such species to be identified and linked with their original creators.

Methods.

In the main these have been those used by Frateur (1950), but have been combined with others. In addition, feeble or doubtful oxydogram results have been augmented by growing the strains on slopes of the same media for a longer period. The all-important criterion of non-overoxidation of acetic acid or acetate has been given particular attention, and has been checked by a variety of methods by

both of us separately; *e.g.*, in addition to yeast-extract (YE)-alcohol-$CaCO_3$-agar oxydograms and slopes, slopes of YE-alcohol-agar, without $CaCO_3$ but with brom-cresol-green as internal indicator, have been used, as have also cultures in liquid media.

Flagella staining has been by SHIMWELL's (1958) modification of FISHER and CONN's method. Electron micrographs have also been made of some doubtful strains. The presence or absence of visible motility has in all cases been determined in hanging drop, for, as HENNEBERG (1897) and FRATEUR (1950) both noted, motility under a cover-slip may sometimes cease in a matter of seconds, owing to the absence of air.

Pigment production, the main (perhaps only) difference between *A. suboxydans* and *A. melanogenum* in FRATEUR's system, has been mainly studied on YE-agar media containing 10% of the particular sugar and 3% $CaCO_3$. Lower and various concentrations of sugar and $CaCO_3$ have also been used additionally in certain cases.

Colony form has been examined under a magnification of × 40, using a × 4 objective and a × 10 eyepiece, by transmitted light or oblique transmitted light (SHIMWELL, 1957, 1958, 1959).

Cultures have been maintained on agar slopes of either unhopped beer agar (in the case of beer strains) or apple juice agar (in the case of cider strains), both incubated at 26°C. until growth appeared, thereafter at room temperature or at 10°C. in the refrigerator. Most strains do not remain viable for long on these media if continuously incubated at 26°C. Freeze-drying has proved very satisfactory.

EXPERIMENTAL.

In all, 50 strains have been studied. Most of these were isolated from beer, brewery yeast, fermenting brewery wort, apple juice, fermenting apple juice, and cider. In addition, strains and varieties labelled *A. suboxydans* and *A. melanogenum* have been obtained from culture collections. Particular care has been taken to obtain the authentic strain of *Acetobacter suboxydans* Kluyver and de Leeuw. Most of the strongly pigmented strains have been isolated from apple juice and cider, in which these types proved plentiful. It is believed that the strains of FRATEUR's varieties of *A. suboxydans* and *A. melanogenum* are his authentic ones. All cultures from collections were carefully typed before the label was accepted, as were also our own isolates. A surprisingly large number of strains labelled "*A. suboxy-*

dans" from collections were found to be overoxidizing *Acetobacter* species and were rejected. It has become clear, indeed, that in some widely cited investigations the "*A. suboxydans*" used was not of this type at all.

Criteria for Differentiation of Species and Varieties.

For this purpose FRATEUR used the production of acid and/or pigment and/or mucilage on glucose, sucrose and maltose. All strains producing a brown to black pigment on YE-glucose-$CaCO_3$-agar were identified as *A. melanogenum* or as a variety of that species. Those not producing pigment on that medium he identified as *A. suboxydans* or as a variety of that species. It is important to note, however, that his *A. suboxydans*, and all its varieties, did produce a brown pigment if fructose was substituted for glucose. Thus the difference between the two species was really a matter of on which of these sugars the pigment was produced.

We have adopted the same criteria, but with the addition of fructose, in attempting to differentiate our strains. In addition to the species and varieties observed by FRATEUR a number of other combinations of acid, pigment, and mucilage production on the four sugars have been found. Altogether the strains in our collection have fallen, so far, into one or another of 13 groups with different combinations of FRATEUR's criteria, *i.e.* 6 more than the 7 of FRATEUR. In Table 1 these 13 different combinations are listed together with their specific or varietal names (if any) whilst it is also indicated in the first column whether the combination of properties found correspond with the description of the creator of the particular species or variety concerned.

We think it likely that if an even larger collection were to be studied further combinations of the criteria might be encountered. Thus the picture presented begins to resemble that found by SHIMWELL (1957, 1958) in the case of *Acetobacter* proper.

A word is necessary about the significance of "P" in the table, where it indicates water-soluble pigment production. The colour and intensity of the pigmentation varied greatly between all our own strains, and amongst named species and varieties also, varying from pale brown to darker brown to black, if one allowed incubation to proceed until the maximum colour intensity had developed; this often required 3 or 4 weeks or more at 26°C. In addition, the partly

TABLE I.

Acid, Pigment, and Mucilage Production.

Characters correspond to:	Glucose	Fructose	Sucrose	Maltose
1. *A. suboxydans* (authentic strain)	AP	AP (pale)	O	AP
2. *A. suboxydans* (FRATEUR's description)	A	AP	O	A
3. *A. suboxydans* (N.C.I.B. 3734)	A	A	O	A
4. *A. suboxydans* var. *muciparum*	A	AP	AM	A
5. *A. suboxydans* var. *biourgianum*	A	AP	AM	O
6. *A. suboxydans* var. *hoyerianum*	A	AP	O	O
7. *A. melanogenum* (BEIJERINCK's description)	AP	O	O	AP
8. *A. melanogenum* (FRATEUR's description)	AP	AP	O	O
9. *A. melanogenum* var. *maltovorans*	AP	AP	O	AP
10. *A. melanogenum* var. *malto-saccharovorans*	AP	AP	AP	AP
11. *A. capsulatum* (N.C.I.B. 4943)	AP	AP	O	A
12. *A. melanogenum* (our strain C. 1)	AP	AP	AMP	O
13. *A. suboxydans* (our strain C1A)	A	AP	AM	O

A = acid; P = pigment; M= mucilage; O = no action.

developed pigmentation was sometimes "plain" brown, sometimes orange-brown, and sometimes orange-red; the black final colour produced by some strains appeared to be largely a very deep red.To have indicated this in the table would have complicated it unduly; "P" merely means, therefore, that some definite degree of pigmentation developed, without indicating its intensity of colour. It may be mentioned, however, that the colour produced by the authentic (supposedly colourless) strain of *A. suboxydans* was by no means the palest, being red-brown on glucose and fructose, and orange-brown on maltose, after 4 weeks at 26°C.

Ketogenic Power.

All our strains, except one, produced dihydroxyacetone from glycerol. The one exception was, surprisingly, a strain stated to have been deposited by KLUYVER himself in the National Collection of Type Cultures (N.C.T.C.) in 1932. This strain is now No. 3734 in the National Collection of Industrial Bacteria (N.C.I.B.).

No dihydroxyacetone-production was detectable with this strain, nor did it produce fructose from mannitol. Mr. K. R. BUTLIN, however, (private communication) says that when he studied this strain in 1934 it produced much dihydroxyacetone from glycerol (see also SHIMWELL, 1958).

This strain is also the only one in our collection which has not produced any signs of pigment on any sugar. The authentic strain of *A. suboxydans* from Delft, however, is both ketogenic and pigment-producing, which suggests that if N.C.I.B. 3734 and the Delft culture were originally of the same strain, the former has now become replaced by colourless non-ketogenic mutants, a supposition supported by the finding of SHIMWELL (1957) and by the fact that both strains still produce acid from the same sugars.

DISCUSSION.

The data in Table 1 together with our other observations, require some discussion to bring out their taxonomic significance. A surprising result is that the authentic *A. suboxydans* strain produces brown pigment from glucose, and is therefore virtually classifiable as *A. melanogenum* or a variety thereof. It is also to be noted that its production of acid is on the same sugars as with *A. capsulatum*, (which is similarly strongly ketogenic).

We have found, however, that this *A. suboxydans* strain differs from *A. capsulatum* in two other characters studied. It freely produces hypertrophied cells and does not make dextrin media viscid. *A. capsulatum* rarely produces hypertrophied cells and does produce viscidity in beer-dextrin media. These two differences are precisely the same as those emphasized by HENNEBERG in the case of his *B. oxydans* and *B. industrium* respectively; he particularly stated that the sugar oxidation of his two species were identical. He did not study pigment production on sugar-chalk media.

Ketogenic power, extensively used by FRATEUR in his system, was not specifically recorded by HENNEBERG; he merely stated that gly-

cerol (and mannitol) were "oxidized". KLUYVER and DE LEEUW, however, noted it in the case of their *A. suboxydans*. On the other hand, KLUYVER and DE LEEUW did not use some of HENNEBERG's criteria. Where criteria are common to both investigations, however, they closely correspond. We feel, in view of the above, that *A. suboxydans* was probably a later isolate of *B. oxydans* Henneberg, and *A. capsulatum* of *B. industrium* Henneberg (see Table 2).

TABLE 2.

	B. oxydans (HENNEBERG'S description)	*A. suboxydans* (authentic strain)	*B. industrium* (HENNEBERG'S description)	*A. capsulatum* (authentic strain)
Oxidation of acetic acid	—	—	—	—
Gluconic acid from glucose	+	+	+	+
Gluconic acid from maltose	+	+	+	+
Action on glycerol	"oxidized"	dihydroxyacetone	"oxidized"	dihydroxyacetone
Pigment	N	N	N	N
Cell-chains	common	common	rare	rare
Hypertrophied cells	common	common	rare	rare
Ropiness in beer dextrin	—	—	+	+

N = not originally studied.

Production of Water-Soluble Pigment from Glucose.

This criterion was used by BEIJERINCK (1911) to establish his *Acetobacter melanogenum*, and maintained by FRATEUR (1950) for its differentiation from *A. suboxydans*. However, quite apart from the fact that there seems no reason why pigment production on glucose should be regarded as so much more important than similar pigment production on fructose, maltose, and sucrose, it seems to have been overlooked that BEIJERINCK himself (1911) wrote (page 174), "Strains of varying pigment producing potency were isolated from various beers . . .". And, on page 175, "Long-maintained cultures show two main types of variability. They can completely lose the power of producing pigment . . . also their usually slight power of slime formation can considerably increase". He added that, when having lost pigment production, such strains became indistinguishable from colour-

less forms. Thus it seems that the specific insignificance of pigment production on any particular sugar, brought out in Table 1, was implicit in BEIJERINCK's original work of 1911.

The probable mechanism of such loss of pigmentation has recently been indicated by SHIMWELL (1957) who found that a strain of *A. melanogenum* (the only one thus examined) gave rise to two colony forms, one pigment-producing and the other not. Moreover, the colourless mutant could regain pigment production, simultaneously regaining its original colony form (Fig. 1).

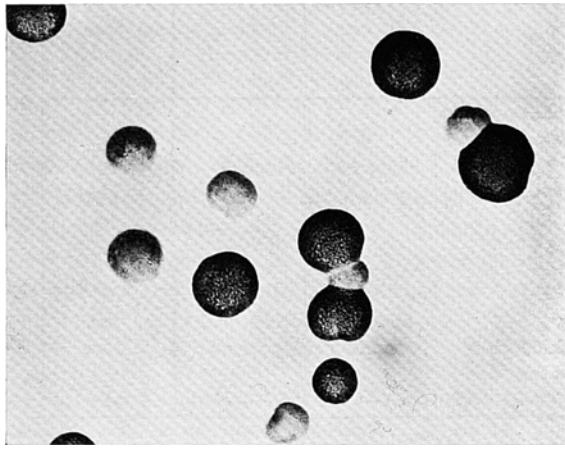

Fig. 1. *A. melanogenum* N.C.I.B. 8086.
Pigmented and non-pigmented colonies. × 40.

Finally the remaining results reported in Table 1 need perhaps a further word. Those familiar with the organisms concerned, and with the appropriate literature, will notice that the results recorded in Table 1 sometimes conflict somewhat with the statements of others. We have recorded the results of our work, however, and where such confliction was encountered the tests were repeated several times.

CONCLUSIONS.

Apart from the fact that our study of the literature, and our experimental results, together with those of SHIMWELL (1958), show that LEIFSON's differentiation of *Acetomonas* from *Acetobacter* is funda-

mentally essential and should be accepted, we consider that the specific distinction between "*A. suboxydans*" and "*A. melanogenum*" is based on criteria (e.g. pigment and/or acid production on a particular sugar) too slight, too variable from strain to strain, (and probably mutable within a strain) to be worthy even of varietal status. There appears, therefore, to be no alternative to limiting the number of species to one. To do otherwise would necessitate innumerable species based on variable and/or mutable criteria.

Taxonomy.

Acetomonas Leifson amended Shimwell.

Diagnosis: Bacteria oxidizing ethyl alcohol to free acetic acid, but not further to carbon dioxide and water. Motile or non-motile. If motile the cells possess one or more polar flagella.

Type (and sole) species:
Acetomonas oxydans (Henneberg) Shimwell and Carr.
Diagnosis: The same as that of the genus.

We consider that the above generic and specific diagnoses are adequate. The inclusion of other and variable or mutable properties which "may or may not" be possessed, such as pigment on glucose and oxidation of this or that substrate, are mere varietal or strain characteristics, not only unnecessary in the present context, but likely to lead to excessive future "splitting" into innumerable unstable "varieties".

Some such minor properties (for instance the production of viscidity in beer containing dextrin) may be of considerable technological, but scarcely of taxological, significance. The latter is the question we are here considering.

Nor do we think it necessary to specify that the cells should even be rod-shaped, gram-negative, or catalase-positive, although the strains we have studied possessed these properties. Some strains of "*A. capsulatum*" however, when growing in beer or malt extract, are almost coccoid; in fact they have at times been microscopically mistaken for cocci by brewery technologists. Similarly catalase-positivity was formerly claimed for all acetic acid bacteria, but it has since been found that many strains of *Acetobacter* proper are comple-

tely devoid of catalase, although strictly aerobic. The diagnoses of both *Acetomonas* and *A. oxydans*, although brief, are considered sufficiently exclusive to admit only bacteria of the type under discussion.

Strain Variations.

The following characteristics, amongst others, have been found to vary from one strain to another (and often even within the same strain): Cell-shape and size; motility; number of flagella per cell; colony form and other cultural characteristics; pigment production on glucose, fructose, maltose, or sucrose; intensity of ketogenic power; slime production; and relatively small variations in apparent optimum temperature (as the medium used, and its acidity and/or pH, may affect this, as may also the definition of "optimum").

Type Culture.

As far as we can discover, no authentic cultures of HENNEBERG's original isolate of *B. oxydans* exist today, although some strains with this label are to be found in culture collections. Furthermore, we have not encountered any strains in which every characteristic, morphological, cultural, biochemical, and technological is identical with those described in detail by HENNEBERG (1897). As such absolute identity, however, rarely (if ever) occurs in nature, we consider that this does not prevent the establishment of a neotype. We are therefore depositing with the N.C.I.B. two cultures which closely correspond to HENNEBERG's description of *B. oxydans* except in respect of one sugar fermentation in each case. The first culture (from beer) differs from *B. oxydans* (and/or *B. industrium*) in that it does not acidify sucrose; the second (from cider) does acidify sucrose but does not acidify maltose, phenomena probably associated with their respective isolation from a sucrose-free and a maltose-free habitat. An electron micrograph of the cider strain is shown in Fig. 2, where it will be seen that one cell has 5 polar flagella and another a single flagellum, in accordance with the diagnosis of LEIFSON (1954) as amended by SHIMWELL (1958).

"*Acetimonas*" Orla-Jensen.

In 1909, ORLA-JENSEN, in the course of propounding a new bacteriological classification, coined many new generic names ending in

Fig. 2. *Acetomonas oxydans* (cider strain).
Electron micrograph.

"monas". For the acetic acid bacteria as a whole he proposed "*Acetimonas*" to indicate the alleged polar flagellation of this group.

We submit that this name must be rejected because (1) He did not describe the genus so named. (2) He did not designate a type species. Indeed the only species mentioned was *A. schützenbachi*; this, however, was a non-motile species, and, even if it had been motile, should have had peritrichous flagella, as it overoxidized alcohol to CO_2 and H_2O (HENNEBERG, 1926; LEIFSON, 1954).

As ORLA-JENSEN's "*Acetimonas*", therefore, was not only superfluous, but also terminologically wrong and inadequately described, there can be no grounds for considering that mere similarity in spelling to the properly described and published *Acetomonas* Leifson

justifies any priority, particularly as this would only perpetuate the former belief that all motile acetic acid bacteria have polar flagella.

Gluconobacter Asai and Shoda.

This must be rejected for a number of reasons. The statement that "*Acetomonas*" is "unreasonable", because its strains "scarcely" acidify alcohol at all, obviously conflicts with the fact that until LEIFSON's work they were not differentiated from *Acetobacter* species, being recognised by all as typical acetic acid bacteria. ASAI and SHODA's definition of *Gluconobacter* describes acetic acid production from ethanol as "weak or none". Such bacteria giving "none" would be *Pseudomonas* strains, however, excluded from *Acetomonas* by definition. Apparently these authors' collection included both *Acetomonas* and *Pseudomonas* strains.

The bacteria dealt with in our present paper are, in contrast (like those of HENNEBERG, BEIJERINCK, KLUYVER and DE LEEUW, SHIMWELL, FRATEUR, and LEIFSON) acetic acid bacteria, thriving (usually preferentially) in acid media, and not bacteria with merely somewhat similar oxidative properties on substrates other than ethanol in media of near-neutral reaction.

ASAI and SHODA have also included in their genus *Gluconobacter* several strains which they state have polar flagella and yet (surprisingly) oxidize acetate to carbonate. Their electron micrographs appeared somewhat inconclusive, however, so by courtesy of Dr T. ASAI we have obtained and studied these strains of *G. melanogenus*. We find them to possess not polar, but peritrichous flagella. These are sparse (1 to 4) and become readily detached, so that some cells appearing monotrichous were perhaps really multitrichous before staining (Fig. 3). These acetate-oxidizing organisms are therefore *Acetobacter* strains, LEIFSON's generic criteria thus remaining undisturbed.

Acetobacter Beijerinck.

We draw attention to the fact that before LEIFSON's discovery of the dual composition of the previously defined genus *Acetobacter*, all published descriptions of its properties were incorrect, for they consisted of a single unified diagnosis of two morphologically, biochemically, (and therefore probably phylogenetically) different types of bacteria.

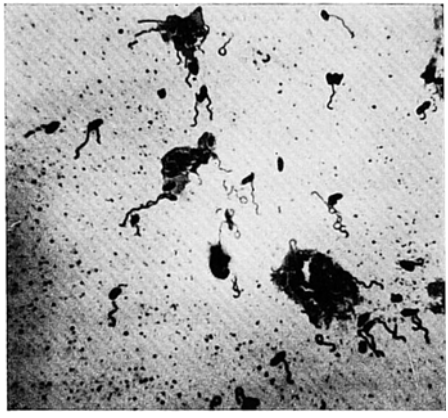

Fig. 3. Sparse peritrichous flagella of "*Gluconobacter melanogenus*". Also detached flagella. ×1000.

The description of a genus, to be valid, must not only be published, but surely must also be correct. This combination of requirements is fulfilled only by *Acetobacter* as described by LEIFSON (1954). It therefore seems that not only *Acetomonas*, but also *Acetobacter*, should probably be attributed to LEIFSON.

Summary.

A study of 50 strains of acetic acid bacteria incapable of further oxidation of acetic acid, together with a study of the literature, has led to the conclusion that the segregation of such bacteria in a new genus *Acetomonas* Leifson amended Shimwell is fundamentally and phylogenetically sound and should be accepted.

No objective grounds justifying the specific separation of pigment-producing strains from alleged colourless ones have been discovered, pigmentation on a particular sugar varying from strain to strain, and also (probably by mutation) within a single strain. Similar arguments apply to the other biochemical and morphological criteria studied. A single species "*Acetomonas oxydans* (Henneberg) Shimwell and Carr" is therefore proposed and diagnosed. It is considered that *A. suboxydans* Kluyver and de Leeuw is a synonym of, and a later isolate of, *Bacterium oxydans* Henneberg (1897).

It is suggested also that the previous necessarily inaccurate des-

cription of *Acetobacter* Beijerinck may invalidate the claims of anyone before LEIFSON to have originated, or properly described, this genus, and that "*Acetobacter* Leifson" may now be the correct designation.

"*Acetimonas*" Orla-Jensen, and "*Gluconobacter*" Asai and Shoda are rejected, and the reasons given.

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