MEETING HELD AT THE CRITERION RESTAURANT, PICCADILLY, W., ON MONDAY, DECEMBER 9TH, 1912.

Mr. JULIAN L. BAKER in the Chair.

The following paper was read and discussed :—

Further Observations on Cider Sickness.

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THE present paper gives a *résumé* of earlier work on the subject and includes the results of additional work in so far as they bear on the subject as a disorder of cider. It is proposed to defer the publication of details concerned with the biological character of the organism responsible for the malady and of the chemical action on various organic compounds until a later occasion.

Cider sickness is an extremely common disorder of sweet ciders, which generally makes its first appearance in the late spring or early summer of the year following the making; the liquor at that period is normally in a quiescent condition and in a fit state for consumption. When the disorder sets in, a sudden violent fermentation arises, the flavour and aroma of the beverage are radically altered, and in the course of a short time the liquor becomes very turbid. It is rendered quite unsaleable.

The malady is one with which cider makers have been familiar for a great number of years, although in the earlier writers on the subject of cider there are no sufficiently detailed references to the disorder to allow of its positive identification with the subject of this paper; nevertheless, their references to sudden violent fermentations of the richer kinds of cider in warm weather indicate that in all probability it was cider sickness to which they referred. It has also always been recognised in the cider industry that cider is a bad "traveller," and since we know that the onset of cider sickness is often brought about by the disturbance of the liquor, this affords further presumptive evidence of its existence in earlier times. It may here be remarked that there is an exactly analogous disorder in perry, and that all

remarks which are here made in respect of cider apply equally to perry.

The disorder is apparently a common one in the French cider districts, being known there as "Maladie de la Pousse." It is improbable that the disorder is so frequent in the German or American cider districts, since the fruit dealt with there is appreciably more acid than that in this country and France. As will be seen, a high acidity tends to prevent the appearance of the disorder.

I referred briefly to the subject in a previous paper to this Society (this Journal, 1911, **17**, 425), and I have more recently given a detailed account of my work on the disorder, in conjunction with Mr. V. F. Hillier, in the *Journal of Agricultural Science*, **5**, pt. 1, Oct. 1912.

In this country the distribution of the disorder is more or less general through the cider-producing areas, but certain districts suffer more severely than others, this being due partly to the fact that in some localities varieties of apples of low acidity predominate. Consequently cider made there is more susceptible. It is also partly due to the character of the soil. The rate of fermentation of the juices from certain soils is much slower than that from soils of other character, and it is the slow-fermenting juices which in general yield ciders which are most easily attacked. The character of the season also has a great deal to do with the frequency of the trouble. After warm, sunny summers the acidity of the fruit and the rate of fermentation of the juice tend to be less than the normal; consequently, ciders made from such fruit are more liable to attack.

Taking a typical case of the disorder, it is characterised by the following features. First, the cider, which is apparently in normal condition, will begin to show a more or less frothing head. This frothing is very persistent and is a most reliable preliminary indication of the onset of the trouble. Very shortly afterwards there is a sudden and abundant production of gas, which is accompanied by a loss of sweetness in the liquor. If the cider is in bottle at this stage, a considerable amount of loss ensues, owing to the wholesale bursting of bottles. Almost simultaneously with this outbreak of fermentation, a radical change in the aroma and flavour of the cider takes place; the pleasant fruity character is lost or overwhelmed, and a disagreeable and pungent aroma and flavour are developed. In the course of a

short time the cider begins to grow turbid, and this may continue to develop until the liquid is in a condition of opaque milkiness.

Such are the typical features of the disorder. There are, however, several variations which must be recorded. In the first place, the violence and extent of the fermentation, as measured by the evolution of gas, is very variable. Sometimes practically no gas is evolved, and the only signs of the trouble are the alteration in aroma and flavour and the development of the turbidity. On other occasions there may be the typical violent fermentation at the outset, but this may suddenly cease long before the whole of the sugar has been destroyed, and the cider then remains in a comparatively sweet condition. Occasionally, again, the typical violent fermentation may continue until all the sugar has been broken up, but the characteristic sickness flavour and aroma may be entirely absent. I have been in some doubt until recently as to whether such cases were to be regarded as true cases of sickness, but, as will be seen later, the evidence is now in favour of their relation to the disorder. In certain instances the turbidity stage is entirely, or almost entirely, lacking.

When the previous paper was published, I was not in a position to explain these variations satisfactorily. Recent work has now afforded a complete explanation.

Ciders attacked by the disorder are obviously rendered undrinkable; after long keeping, however, they show a more or less complete recovery. The main distinction, then, between them in their original form and in the later stage, is that in the first instance they were more or less sweet, and in the latter instance they had lost to a greater or less extent that sweetness. The aroma and flavour characteristic of sickness disappear, and the turbidity gradually vanishes, the substance to which it is due being deposited as a thick crust on the sides and bottom of the vessels containing the liquor.

I have from time to time noted the occurrence of the typical sickness flavour and aroma in the apples themselves, the fruit to all appearance being in a perfectly sound and normal condition and absolutely free from disease. Such cases could only be understood on the assumption that the same type of chemical change was occurring in the fruit, as the result of abnormal physiological processes, as that which occurs in the cider under the action of the organism which causes the sickness fermentation. These curious cases of the occur-

rence of the sickness character in the fruit seem now explainable by the results which I have recently obtained in connection with the study of the disorder.

Turning to the actual changes brought about in the cider by sickness, they may conveniently be grouped under three heads, the first relating to the nature of the fermentation, the second to the cause of the aroma and flavour, and the third to the nature and cause of the turbidity. Analysis shows that in sickness fermentation there is a type of alcoholic fermentation which in many respects approaches very closely the normal alcoholic fermentation produced by yeast. Its marked feature is the destruction of dextrose, accompanied by the evolution of carbon dioxide and the formation of ethyl alcohol. In addition to the carbon dioxide evolved, there is also a small quantity of hydrogen; but this has never yet been observed to exceed 3 per cent. of the total quantity of gas produced. Accompanying the breaking up of the sugar there are slight changes in the acidity of the liquid. In ciders of low acidity there is eventually a slight increase, which does not approximate to more than the equivalent of 0·2 per cent. malic acid; in ciders of rather higher acidity the final result may be a slight lowering of the acid contents. It seems likely that the malic acid naturally present in the cider is slightly attacked during the fermentation, thus producing a small diminution of acidity in respect of that substance. This, however, in cases of ciders of low acidity is rather more than counterbalanced by the formation of small quantities of volatile and fixed organic acids, resulting from the fermentation. The formation of the following acids has been proved: formic, acetic, propionic, butyric or valeric, and oxalic. No succinic acid has yet been found, and the formation of lactic acid is doubtful. Slight changes in the so-called tannin contents of the cider also occur, there being a slight reduction in the permanganate-reducing figure of the liquid.

The question of the production of aldehydes and alcohols may be considered in connection with that of the aroma and flavour. There is no doubt that the latter is due mainly to the presence of acetaldehyde. Comparatively large quantities of this substance are generally formed. Formaldehyde is also produced, and it is highly probable that minute quantities of higher aldehydes also are present. The quantity of these latter, however, is so small that it has not been possible to recognise any individual substance definitely.

With regard to alcohols, by far the largest constituent is ethyl alcohol. Appreciable quantities of methyl alcohol, however, also occur. Tests for higher alcohols have so far given negative results.

It is to the presence of the substances named, together with the volatile fatty acids previously referred to, that we must look for the cause of the aroma and flavour. Doubtless compounds such as esters result from the interaction of these substances, and these too add to the composite character of the sickness aroma and flavour.

The third section in the chemistry of sickness is that relating to the cause and nature of turbidity. Microscopical examination of a sick cider shows that whilst there are living and dead cells of various micro-organisms present in suspension in the liquor, nevertheless they do not occur in sufficient quantity to account for the density of the turbidity. There are present, in addition to them, large numbers of minute oily or resinous looking droplets, some single and others aggregated together so as to resemble very closely small colonies of cocci. The single droplets are easily mistaken for individual bacterial cells. This substance was believed in the earlier paper already mentioned to be due to the action of aldehyde on the tannins or related constituents present in the cider, and further work has now shown that there are good grounds for this assumption. The exact character of the substance is at present under examination by Dr. Nierenstein in the Bio-Chemical Department of the University of Bristol, and I do not propose to deal further with that here.

Turning now to the conditions which favour the development of the disorder, the first to which attention may be paid is that of the susceptibility of the individual ciders themselves. It has been found that the organism which causes sickness is one upon the growth of which the commoner organic acids have an unfavourable effect. Consequently those ciders with relatively high acidity are more or less immune to attack, whilst those of low acidity are very susceptible. The quantity of tannin in the cider does not appear to have any appreciable influence, since, provided that the acidity is low, ciders of high and low tannin content appear almost equally susceptible. The original rate of fermentation of the juice from which the cider is made is a factor of primary importance. Juices fermenting rapidly generally give ciders more or less resistant to sickness, whilst those fermenting slowly yield very susceptible ciders. The rate of fermenta-

tion seems to act in two ways. In the first place ciders which have fermented rapidly generally have low sugar contents, while those which have fermented slowly have high sugar contents; and, as already seen, it is sugar which is the substance mainly attacked. Apart, however, from the question of its bearing upon the sugar contents, the rate of fermentation itself seems to have a definite influence, since if two ciders with an approximately equal amount of sugar are taken, one of which has fermented slowly, and the other rapidly, it is found that the former is generally more easily attacked than the latter. It is evident from what has just been said that the question of the varieties of apples used in the making of cider is important, since not only does the acidity of different varieties vary, but also the rates of fermentation of their juices. In the same way, any factors influencing the quality of the fruit, such as the soil on which it was grown and the nature of the season, are important in connection with the question of susceptibility. Temperatures about 18° to 20° C. are particularly favourable to its development, whilst below 12° C. there is little, if any, action. Another factor of some influence in connection with the outbreak of the disorder is that of the mechanical disturbance of the liquor. It is found that so long as cider is left in the cellar undisturbed, there may be no signs of an outbreak of sickness, whereas if it is moved, either in connection with racking or with distribution from the factory, sickness may quickly set in.

It has been generally accepted that the immediate cause of sickness is an organism capable of acting upon the sugars in the cider. French writers have described a bacillus which is found in sick cider, to which they attribute the disorder, but they do not appear to have isolated it. In the earlier stages of this work definite proof that it was due to the action of an organism was obtained by sterilising various types of cider and infecting them with a few drops of a cider in an active state of sickness. The infected liquid quickly showed all signs of sickness, and examination of the liquor showed abundant development of small bacteria. The isolation of the organism proved a matter of considerable difficulty. Many series of plate cultures were made without success, the plates becoming covered with growths of various yeasts and bacteria, none of which were found capable of producing sickness. Eventually it was found that the sickness organism developed so slowly that a modification of the direct plate culture method was necessary in

order to isolate it from the other forms. The organism is a small motile bacterium, which is facultatively anaërobic, and which has an optimum temperature for growth of from 25° to 30° C. Its maximum temperature for growth is about 40° C., and its minimum temperature from 10° to 12° C. The temperature at which the cells are killed on heating for five minutes is about 55° C. No spore formation has been observed. There is little doubt that the organism finds its way into the cider at the time of making, and probably it is to be found on the surface of the fruit, in conjunction with wild yeasts and acetic and other bacteria.

A large number of infection experiments with pure cultures on sterilised ciders of various types show that the composition of the cider has a very great influence upon the susceptibility of attack by the organism. They bear out exactly the results which have already been recorded in connection with the susceptibility of cider under natural conditions.

Variations in the Nature of the Disorder.

Considering now the results which have been obtained since the previous paper on the subject was written, those bearing upon the causes of the variations in character of the disorder may first be dealt with. It has been stated that the extent and intensity of the fermentation show a considerable variation in individual cases. Quantitative experiments with pure cultures in sterilised ciders and sugar solutions show that there are great differences in the amounts of alcohol and carbon dioxide which are produced from a given quantity of sugar. In some instances the amount of carbon dioxide produced is almost equal to that of alcohol, and the combined weights of alcohol and carbon dioxide are approximately equal to the total weight of sugar destroyed. This type of fermentation is thus very closely allied to normal yeast alcoholic fermentation. In other cases the amount of carbon dioxide may be very much less than that of alcohol, and in extreme instances no carbon dioxide is evolved.

During the earlier part of the work some confusion in the results was introduced owing to this behaviour. The absence of evolution of gas somewhat naturally led to the belief that the infection had not been successful, and that no destruction of the sugar had taken place. When, however, the quantitative work was started, it was found that

there had been destruction of the sugar. The amounts of alcohol produced in such cases vary, but the total does not exceed about 50 per cent. of the weight of sugar destroyed. Consequently there must be some other product, or products, formed in considerable quantity, and the nature of these is at present doubtful. Aldehydes, organic acids, and methyl alcohol partly account for the deficiency, but the total quantity does not at present appear to be sufficient to account for the whole of the deficiency. It has been found that the heaviness of the infection has considerable influence upon the amount of carbon dioxide produced, but whether or not this is the sole factor determining the result remains to be proved. Generally speaking, above a certain limit, the heavier the infection the less carbon dioxide is found; whilst at the other extreme a similar result follows if the infection falls below a certain minimum limit.

Reference has already been made to cases of sudden fermentation which occur in certain ciders during the summer season, which, however, lack the characteristic flavour and aroma of sickness. It now seems probable that these fermentations are caused by the sickness bacterium, and that the lack of the characteristic aroma and flavour is due to an almost entire absence of aldehyde formation. Experiments with pure cultures on various nutrient solutions have shown that the amounts of various aldehydes formed vary considerably. In certain cases the amount formed is not very much more than the extremely small quantity formed in ordinary alcoholic fermentation by yeast, whereas in other cases the amounts are relatively large. The factors determining the amount of aldehyde formed have not yet been ascertained.

Instances where sickness fermentation suddenly ceases, although relatively large amounts of sugar remain in the cider, have been frequently recorded. The explanation appears to be that certain substances formed by the organism during the course of fermentation accumulate and exert an inhibitory effect on its further development. Now that the products of fermentation have been more fully investigated, it seems probable that this inhibition may be attributed to the formaldehyde, and possibly some of the other products mentioned. As already just stated, the amounts of these products of fermentation vary considerably, and in cases where the amount of these substances is relatively high, it is clear that the further growth

of the bacterium must be influenced on account of their well-known antiseptic properties.

Sickness Aroma and Flavour.

Dealing now with the question of the aroma and flavour of sickness, experiments of adding varying quantities of acetaldehyde to sound cider show that to a very great extent this substance must be held responsible for those features. Whilst the exact flavour and aroma of sickness cannot be reproduced by the addition of aldehyde, a close approximation is obtained.

It has been recorded that in course of time the flavour and aroma tend to disappear, and that, if a cider be kept long enough, eventually all traces will be lost. Analysis has shown that in such cases the amount of aldehyde is very much less than during the early stages of sickness fermentation, and it may therefore be concluded that the loss of the sickness aroma and flavour may be attributed to the disappearance of the aldehyde.

Sickness Turbidity.

The fate of the latter leads on to the subject of the turbidity which develops during sickness. There is now very strong reason to suppose that the turbidity is due to the action of aldehydes on those constituents of the cider which have been classed under the head of tannins. The evidence in favour of this is as follows.

If a sterilised cider is infected with a pure culture of the bacterium and the gaseous products are passed through a wash bottle containing sterilised cider, the latter will gradually become turbid and at the same time acquire the aroma and flavour of sickness. It has been proved that this liquid remains in a sterile condition, so that the development of the sickness character cannot be attributed to the presence of the bacterium. It follows, therefore, that the products of fermentation which cause both aroma, flavour, and turbidity, are volatile and are carried over by the carbon dioxide which is evolved. Examination of the sterile cider shows the presence of aldehydes just as in the case of infected cider.

If formaldehyde or acetaldehyde themselves are added to sterile cider, the liquor gradually becomes turbid as in sickness, and to some extent the flavour and aroma of sickness are also developed. The action of formaldehyde in producing the turbidity is very much more rapid than that of acetaldehyde. In the former case considerable

turbidity is produced within 24 hours; in the latter more than a week may elapse before any signs of haziness appear. Estimation of the tannin contents by the usual direct permanganate titration shows a diminution. The amount of this diminution is determined by the amount of aldehyde added, but a limit is reached beyond which the addition of a greater quantity of the aldehyde does not result in any further diminution. The extent of the diminution is apparently much greater in the case of formaldehyde than in the case of acetaldehyde. The liquor after treatment with sufficient aldehyde no longer gives the ordinary qualitative tests for tannin.

The nature of the substance which is the cause of the turbidity is, as already stated, under investigation by Dr. Nierenstein.

The variations which have been recorded as to the extent of the turbidity developed during sickness may be accounted for by the varying amounts of aldehydes which are formed, just as the variations in aroma and flavour may also be explained.

Sickness Flavour in Apples.

The striking part which the production of the aldehydes plays in sickness fermentation in respect of aroma and flavour at once suggests that those curious cases of the occurrence of those features in perfectly sound apples referred to earlier may also be accounted for by the production of aldehyde. It is known that during the course of the ripening of certain fruits there is a destruction of sugar, generally resulting in the evolution of a corresponding amount of carbon dioxide. Under certain conditions, however, ethyl alcohol may be formed, and it is only necessary to assume that one further stage of oxidation takes place, converting the alcohol into aldehyde, in order to account for this abnormal flavour in the fruit.

Frothing during Sickness.

One other subject in connection with a feature of the fermentation may be referred to, that being the characteristic frothing which occurs at the beginning of fermentation. The froth which is formed at the outset, and which becomes more noticeably marked when any of the cider is heated for distillation purposes, is far more pronounced and persistent than any produced by normal yeast fermentation. Recent work has shown that probably the presence of aldehyde has a great

deal to do with this feature. It was observed by chance in the laboratory that the addition of lime water to sterilised cider, to which a quantity of aldehyde had been added some weeks before, gave rise to the evolution of a considerable amount of carbon dioxide, and a characteristic head entirely resembling that produced during the sickness fermentation was produced. This action only occurs after the aldehyde has been added to the cider long enough beforehand for the turbidity to be developed. If the mixture is immediately tested with lime water after the addition of aldehyde, there is no evolution of gas, nor is any frothing head formed.

I should like to take this opportunity of acknowledging the valuable assistance rendered by Mr. Wm. Camps, my laboratory assistant, in the chemical part of the work.

Remedial Measures.

In the paper previously quoted, brief reference was made to certain lines of treatment for the prevention of the disorder to which attention had been given. Since its publication the results of practical experiments at the National Fruit and Cider Institute during the cider-making season of 1911–12 have been obtained, and this aspect of the subject may be now more fully considered.

In dealing with a disorder of this kind, where the responsible organism is in all probability present in the liquor from the start, the question of control can be given attention in at least four different directions.

1. The prevention of the occurrence of the organism in the freshly pressed juice.
2. The elimination or destruction of the bacterium when it is actually in the liquor.
3. The production of a type of liquor unsuited to the growth of the bacterium.
4. The treatment of the liquor after it has been attacked by the disorder, in order to render it again marketable.

1. In order to prevent the access of the living organism to the juice it is necessary—assuming it to occur naturally on the surface of the fruit—either to kill it *in situ* on the latter or to remove it before milling by some means such as washing. From experiments carried

out in France a few years ago by Warcollier it is evidently possible to obtain a sterile juice, if the fruit is washed prior to milling with a dilute solution of formaldehyde. That worker found that after such treatment the juice was not only sterile, but also practically free from traces of formaldehyde. Possibly other antiseptic solutions could be utilised with similar success. As a practical measure, however, this method of dealing with the disorder presents considerable difficulties. The average cider maker is not a chemist; whilst in the case of the larger factories where chemists are employed, the quantity of fruit to be dealt with in a very limited space of time makes it desirable that any treatment of the fruit before milling should be of the simplest character. This objection may also be urged, although with less force, against the method of washing the fruit with water. The latter procedure has been adopted in certain cases, and it is claimed by those using it that the ciders are afterwards less liable to sickness owing to the fact that the washing removes many, if not all, of the bacteria. My own experiments with cold-water washing point in the same direction, but they show at the same time that the method cannot be relied upon as a sure safeguard against the disorder.

In view of the comparatively low temperature (55° C.) at which the cells of the bacterium are killed, the cold-water washing experiments were supplemented by others in which hot water at a temperature ranging between 60° C. and 65° C. was used. The results in these experiments have been much more promising. In no case did sickness develop after the hot-water treatment, although in some instances in the control tests with fruit of the same kind unwashed or washed with cold water the disorder occurred. The actual number of these trials at present completed is, however, too small to arrive at a final conclusion, and the work in this direction needs considerable extension under varied conditions before anything more positive may be claimed. It is important to note that the hot-water treatment has practically no effect on the flavour of the juice or cider. The objection to dealing with the disorder by the direct pasteurisation of the juice on account of the so-called cooked flavour imparted to it by heat does not occur when the fruit itself is subjected to the hot treatment. The flavour of the mature cider in the latter case is not quite so good as that of the control sample made from fruit washed in the cold, being slightly coarser and, perhaps, rather more bitter; but

the effect is comparatively insignificant, and would not be noticed unless the two ciders were tasted side by side.

2. Several attempts to prevent sickness by the elimination or destruction of the bacterium from the juice or cider after it has gained access have been made. Filtration of the liquor through the ordinary type of pulp filter has proved useless for the purpose. As indicated in the previous paragraph, pasteurisation is equally a failure on account of the effect on the flavour.

Antiseptics such as salicylic acid are effective in suitable doses, but their use is to be deprecated on other grounds. Sulphurous acid in one or other of its various forms is not so much open to objection, but it does not give altogether satisfactory results. For a time this substance does undoubtedly check sickness, but its effects—except in doses so large as to ruin the flavour of the cider permanently—are transient, and sickness is only delayed and not permanently warded off. Its use, however, may prove of service in cases where the cider is likely to be consumed comparatively early in the summer. In such instances it is probably better to sulphur the liquor at the end of the primary fermentation rather than directly after pressing, since, as will be seen from the succeeding section, a vigorous primary fermentation is of material service in rendering the cider resistant to sickness.

Sterilisation by exposure to ultra-violet rays of light has been suggested as a possible method. Arrangements are now being made to test their effect, but even in the event of success this method seems impracticable for general adoption under present conditions in the cider-making industry.

3. Undoubtedly the surest method of combating the disorder is the production of a type of cider in which the organism is unable to flourish. The early investigations on the subject proved that it was possible to render a cider practically immune by raising the acidity to at least 1 per cent. of malic acid. With that degree of acidity, however, cider is too sharp for ordinary palates. Unless there is an exceptionally large amount of sugar present, it may be accepted that the acidity cannot be raised much above 0·6 to 0·7 per cent. of malic acid with advantage from the consumer's point of view. By the use of a suitable admixture of sharp and sweet or bittersweet types of apples it is comparatively simple to attain that standard of acidity without occasion for the addition of foreign acids, such as citric or tartaric.

Generally a cider with this degree of acidity withstands sickness very well and in most cases escapes the disorder, even when kept under conditions favourable for the development of the disorder. Experiments have repeatedly shown that a cider of low acidity, to which is added a sufficiency of a sharper cider to raise the acidity to the figure mentioned, remains sound and in excellent condition, whilst a control sample of the same cider unblended succumbs to the disorder as soon as the summer approaches. Nevertheless, there are occasional instances where this treatment fails owing to the fact that 0·7 per cent. of malic acid is not high enough to check the development of the bacterium, when other conditions are exceptionally favourable for its growth.

It has also been remarked that the ciders prepared from rapidly fermenting juices, even though they may contain relatively large amounts of unfermented sugar, resist the disorder better than those of similar type produced from more slowly fermenting juices; and it has been found that if the latter kind of juices are blended with the former and their rate of fermentation thus raised, the chances of resisting the disorder are correspondingly increased.

A vigorous primary fermentation appears to exert a lasting effect on the cider in aiding it to resist sickness. This has been well shown in some filtration experiments. In these, certain juices were taken as they came from the press and divided into two equal volumes. In the one case they were filtered immediately, and then left until fermentation eventually set in. Fermentation was allowed to continue until a definite point of gravity, generally about 1025, had been reached, and then the liquor was again filtered, after which no further appreciable fall in gravity was recorded. By this treatment the start of the normal primary active fermentation was generally delayed two or three weeks, and the subsequent rate tended also to be slower. In the other case the juices were left unfiltered until the gravity had dropped to the point already referred to. They were then filtered, and no further appreciable fall took place. In certain cases the latter remained sound, whilst the corresponding cider treated in the former way turned sick. In other instances where both succumbed to the disorder, the cider filtered direct from the press developed the trouble much more quickly than the other, and suffered more severely. In cases where the acidity was high neither kind was affected. These experiments also illustrated very clearly the determining influences

of combinations of factors and showed how in critical cases a comparatively slight difference may suffice to turn the scale one way or the other. For instance, in dealing with a cider the acidity of which stood at between 0·45 and 0·7 per cent., the region which from the sickness standpoint is doubtful, the portion which was filtered direct from the press turned sick, whilst that in which the primary fermentation was permitted to develop naturally remained sound.

Other experiments in which the yeast fermentation apparently played a large part may also be mentioned here. In one instance a cider of low acidity standing at a gravity of 1027 continued to fret after being filtered. It was divided into two equal portions, one of which was left untouched until the gravity had fallen to 1020, when it was again filtered and fermentation permanently checked. To the other sufficient brewers' yeast was added to produce an active fermentation. After this had continued until the gravity had dropped to approximately 1020, the cider was filtered and fermentation arrested. In the former instance the cider became sick during the following summer, but in the latter no signs of sickness appeared.

In the other instances the effect of early bottling is concerned. Several seasons ago it was shown that occasionally cider in cask became sick, although the same kind of cider in bottle escaped. Later it was noticed that a cider which had been bottled in March escaped sickness, whilst a stock of the same sort bottled in May developed the disorder. A series of tests during the past three years was therefore arranged, ciders of a susceptible character being selected. Each was bottled from cask at three or four different times extending from January to May, at intervals of about four or five weeks. In some cases the earlier lots bottled remained sound, whilst the later ones became sick: and in other cases where all developed sickness, the disorder appeared in order according to the time of bottling, setting in first in the latest lots bottled, and last in the earliest bottled lots. This result corresponds with the amount of secondary yeast fermentation that takes place in bottle. The earlier a cider is bottled the greater is the amount of fermentation which occurs in bottle, and the more highly charged with carbonic acid does the liquor become. There is here, therefore, a case which corresponds very closely with those already mentioned in which the susceptibility to sickness varies inversely with the vigour of the primary fermentation.

Since in all the cases here referred to there was enough unfermented sugar left in the cider to permit the full manifestations of sickness, if the bacterium had been in a condition to grow actively, it follows that the results recorded must be in some way dependent upon the antagonism of the yeasts of the liquor and the sickness organism. Whether active yeast fermentation keeps the bacterium in check by the production of a substance or substances injurious to its growth, or whether it is simply a question of dominant fermentation on the part of the yeasts, the bacteria being crowded out, is a question not yet settled; but it seems now definitely established that the development of the bacterium is inversely proportional to that of the yeasts. It follows, therefore, that the encouragement of an active primary fermentation should reduce the liability to sickness. There is still a wide field for further research in this direction, and at the same time considerable promise that the disease organisms may be successfully held in check by those necessary for the production of a sound beverage.

4. When a cider is attacked by sickness, it is for a time not fit for consumption. After remaining untouched for some months, however, the sickness aroma and flavour may pass off and the turbidity settle to form a crust. The liquor then, to some extent, regains its original character, except for the loss of sweetness which it has sustained. At present no quicker method of dealing with it so as to render it in fit condition for sale appears to have been suggested. Now that it is known, however, that the aroma and flavour are due to very volatile substances, it seems possible that the affected cider might be dealt with immediately the active sickness fermentation ceases and before the greater part of the turbidity appears. The passage of a steady stream of carbon dioxide through the liquor until practically the whole of the aldehydes are removed, followed by filtration of the cider, might be effective. It is proposed during the coming summer to take an opportunity of testing this suggestion.

Discussion.

The Chairman said that they had listened to a most interesting and suggestive paper. Those who had followed Mr. Barker's work on cider would congratulate him on the important results arising out

of investigations carried out under such able leadership at the Cider Institute. In cider manufacture fermentation was started in a non-sterile liquid, and, in addition to the yeast or yeasts which carried on fermentation, there were naturally present large numbers of acid-forming bacteria. In brewing, on the other hand, the position was quite different. The brewer started with a clean slate. After the wort was boiled in a copper it was sterile. It really seemed that a great deal was known about the diseases and troubles which attacked cider. He only wished they could say as much concerning similar problems in their own industry. The organism of which Mr. Barker had given them an account was an extremely interesting one. Though it caused such trouble, yet it was not what they might call, bacteriologically speaking, a very strong organism. It grew slowly, its death temperature was fairly low, and, unlike many other organisms with which they were acquainted, it did not grow easily in an acid medium. In some respects there was an analogy between the organism responsible for cider sickness and one that most brewers knew of, namely, *Saccharobacillus Pastorianus*. Mr. Barker had told them that sterilisation caused a bad flavour in the bottled cider; but as the organism died at 60° C., one wondered whether a temperature of 55° C., or perhaps even a little lower than that, would alter the flavour. He would like to ask Mr. Barker if he had compared the organism producing the sickness with *Bacterium albumenosum* (Lindner). In some respects there appeared to be a resemblance. In conclusion, he would ask a question which would be obvious to brewers. Had any experiments been tried in the direction of maturing the cider in hogsheads or other suitable vessels, then chilling, carbonating, and filtering?

Mr. STENHOUSE said that he was certainly struck with the fact that fermentation might go on without any outward or visible signs, and he would like to know what, if any, increase in temperature took place during the process.

Mr. R. L. SIAU said that Mr. Barker seemed to have shown pretty definitely that it was only the sweet ciders of fairly high gravity that were particularly liable to the disease. He was under the impression that he had read in some of Mr. Barker's earlier works that the speed of fermentation of a cider depended more or less closely upon its nitrogen-content. That being the case, he would like to ask if

Mr. Barker had tried hurrying up those dangerous ciders through their fermentation by adding to them nitrogenous food so that their primary fermentation, so to speak, would get them through the dangerous zone by getting them down to practically the same position as a rapidly fermenting cider. As regards, from his point of view, the interesting matter that Mr. Barker had touched upon, the fact that cider was very liable to enter into a stage of sickness as a result of transit, he was sorry to say that he had seen that on several occasions; but he was thankful to have the information that if a potentially sick cider was bottled earlier the fermentation might protect it from risk of future sickness.

Mr. H. C. BURGESS said that the very large amount of work which Mr. Barker had brought before the meeting made it somewhat difficult to criticise it fully. He thought that much praise was due to Mr. Barker in having identified the organism which was apparently responsible for cider sickness, and no doubt much further research would be done in cultivating the organism in various media to bring about the same peculiar odour which was characteristic of sick cider. The whole burden of Mr. Barker's paper seemed to point to the presence of a slight excess of malic acid to avoid sickness, but was it really due to malic acid or rather to some acid, such as lactic acid, which was well known to retard the development of certain organisms? Further, the author did not appear to have studied the bearing of albuminoid matter in relation to malic acid; it seemed likely that the albuminoid matter would be less in a sour apple than in the sweeter apple. A careful study of the relationship of nitrogen-content to acid might illuminate the cause of this sickness further and might also help in the study of aldehyde formation. Had Mr. Barker discovered any simple practical way of determining if a cider was likely to develop sickness at a later period of its existence, or was the organism present in all ciders and only developed in some when an internal change of the cider made it become an available medium for the growth of the organism?

Dr. L. T. THORNE said that the new observation which Mr. Barker had made of alcoholic fermentation going on without apparently any production of carbonic acid was a most striking one, and it was especially striking when, as Mr. Barker said, in hardly any case was there a larger production of alcohol than that corresponding with 50 per

cent. of the sugar acted upon, and yet the other 50 per cent. seemed to have more or less completely disappeared. The author had suggested that aldehyde was a general constituent that was formed; but he took it, from what Mr. Barker had said, that even in the cases where he had found most aldehyde produced, it did not seem to account for anything like the additional quantity of sugar fermented. From the observation that Mr. Barker had made, that the sickness might be, at all events, partly cured by the passage of carbonic acid through the sick cider, by which some of the aldehydes were carried off, it might be possible that in some cases of sickness in barrels some of the aldehyde produced might evaporate; but of course that could not be the case with the sickness in bottles, and they would certainly look forward to Mr. Barker's further experiments with great interest, for the elucidation of that very remarkable reaction. Another very striking point was the influence of rapid and slow fermentation, although in the brewing industry they had a certain amount of analogy in the stopping of infection from an infected yeast where fermentation was very active. Undoubtedly the active fermentation by the yeast had a tendency to inhibit the growth of—and, in some cases, probably to destroy—the infecting organisms, and he took it that might very likely be the case in the cider industry; this infecting bacterium seemed to be rather a feeble individual, and might be actually killed by the more active yeast ferments when fermentation took place rapidly, whereas when there was slow fermentation there might be a chance for that infection to go on developing and get a more firm hold on the liquid. The blending of the cider was a very interesting point, and certainly from Mr. Barker's observations seemed to go far towards stopping the infection. He thought the observation made by one of the speakers as to the effect of trying varying acids might be a very useful one, and possibly lactic acid might have a greater inhibitory effect than malic acid which was present normally.

Mr. W. M. BREWIS said that he was connected with the cider industry, and he must say he had come across the sickness, and it was very peculiar. They got it in various ways. One year it was entirely different to another. And in some ciders that they had bottled out of the same cask, part would be absolutely sound, brilliant and perfect, whereas others would be sick. Of course, it

was very difficult for large merchants to wash all their fruit. His firm crushed 100 tons a day, and he thought it would be very difficult to wash, either with cold or hot water or with formalin, as had been suggested.

Mr. M. J. CANNON said that he had investigated cases of cider sickness, and if he had not been successful in dealing with it, he must attribute his failure partly to the manufacturers, who were reluctant to provide the means by which a difficult investigation could be carried on, and partly to the elusive nature of the organism concerned. At quite an early stage of his work he had been impressed by the amount of turbidity in sick cider and the comparative freedom from yeast and bacteria. Although he felt sure that the source of the trouble was due to bacteria and not to yeast, he had failed to isolate the organism. As Mr. Barker had informed them, ordinary plate cultivations became crowded with colonies long before any appreciable development of the organism could be detected. The odour and flavour of a cider suffering from sickness were the most characteristic symptoms, but he was of opinion that the odour was not entirely due to aldehyde; there was a peculiarity which could only be described as a cross between aldehyde and garlic. Whilst he recognised that a bacterium was concerned in the development of this malady he did not believe that the presence of the organism was the determining factor. Cider making was dependent upon spontaneous fermentation and the erratic appearance of this disease in a factory suggested that the organism was always present in the apple juice. Whether it developed and caused sickness or whether it remained quiescent appeared to depend upon other factors, particularly the rate of fermentation. Mr. Barker had also proved that the amount of acidity exerted an important influence. There was little doubt that if the "primary" fermentation had been vigorous in cask the cider would be more resistant to sickness. He had obtained some success by stimulating fermentation by the addition of nitrogenous substances to the original apple juice. The cider industry was to be congratulated that it possessed an Institute well equipped for the solution of its difficulties. Ten years ago the methods of cider making were more crude than those existing in the brewery 50 years since, but during the last seven or eight years great improvements had been made in cider making, and its difficulties better understood, mainly as the result of

the admirable work carried on at the Cider Institute under the able direction of Mr. Barker.

The CHAIRMAN read a letter received from Mr. Walter A. Riley:—

"I should like to ask Mr. Barker if he can explain the reason why samples of ciders from a series of 50 or more pipes, if taken in sterilised flasks holding about 300 c.c., closed with cotton wool plugs, placed in an incubator, and kept at a temperature of 26° C. for five days, some will have developed the peculiar aroma of sickness, whilst others will be quite free of any taint; however, later on in the season, those pipes which were found to be sick by the incubator test do become sick, and, in some cases, *one or more pipes that were supposed to be free have by that time developed sickness;* was it because those samples from these pipes ought to have been placed at a higher temperature, or was the period of incubation too short?

"I might add that this happens very frequently, and with samples taken from the same cellars, so that the question of cellar temperature cannot arise.

"I should like to ask at what period samples ought to be taken after pressing. I have in some cases taken them after the first racking. I should also like to know what temperature is most favourable for development of the disorder, and how long the samples should remain in the incubator.

"I have found that 26° C. is the most favourable, and an incubation period of five days.

"Would Mr. Barker recommend that the contents of those pipes that are found by the forcing tray test to be infected should be immediately pasteurised, and then blended off with other ciders, or by blending with clean cider having a higher percentage of malic acid, or would he prefer to allow the cider to undergo further fermentation, and then prevent the course of the disease spreading by the addition of a vigorous yeast?

"I should like to learn whether Mr. Barker would have any objection to the addition of brewer's yeast which has had very careful washing, to cider must?

"I am afraid that until such time as the cider maker can protect his apples by washing with antiseptics in the first instance, he must apply the incubator test to all his pipes, and those that he can detect, cure at once by some of the methods already suggested.

"Personally I am convinced that if only the cider maker would apply some of the methods that have been adopted by the brewer, we should hear less of cider sickness."

Mr. BARKER, in reply, said that the Chairman had raised the question as to the extent of the alteration of the flavour of the cider by submitting it to a temperature of from 50° to 60° C. Unfortunately it was found that the effect on the flavour varied greatly with the different ciders, and the composition of individual ciders varied so much that a constant result at any definite temperature was not obtained. For example, if freshly pressed juice was submitted to a temperature of, say, 55° C. and fermented afterwards, and, again, if some of the same juice was fermented first and then submitted to a temperature of 55° C. under similar conditions, they would not get in the end ciders of the same type and flavour. The presence of sugar was a complicating factor. Also the degree of acidity of the juice had a great deal of influence upon the effect of the heat in modifying the flavour; so that the question of dealing with the juice by heating was by no means as simple a one as might appear at first sight. With regard to the possible identity of the bacterium with *Bacterium albumenosum*, the description given by Lindner of the latter differed in many respects from the sickness organism. So far as he had had an opportunity of consulting bacteriological literature, he had been unable to discover any organism hitherto described which corresponded with the one in question. In some respects the type of fermentation brought about by it resembled that of *B. coli communis* on glucose, as described by Harden; but there were several points of difference in the chemistry of the actions in the two cases. Their biological characters also differed considerably. With regard to the quantities of carbon dioxide and alcohol which were produced from a given weight of sugar, it had been found that in most cases there was a certain amount of sugar broken up which could not be accounted for by any of the products he had so far examined quantitatively. Since the paper was written, however, acetylmethyl carbinol had been identified among the products of fermentation, and it might possibly be found that this substance, together with 2·3-butylene glycol, which generally accompanied it, represented the sugar not accounted for. Harden found, under similar circumstances, that this was actually the case in his work on

the action of *B. coli communis* on glucose. With regard to the question of chilling, filtration, and carbonation, for quick consumption, that was a method which he thought was likely to play an important part in the cider industry. There were certain difficulties, but as far as the general character of the product went it did not compare unfavourably with the liquor dealt with under the ordinary system. He knew, however, of cases of sickness which had followed such treatment, and it could not therefore be regarded as a preventive of the disorder. He had been asked whether there was a rise in temperature during the sickness fermentation. He had no information on that point. All the quantitative work had been conducted at a temperature of 25—30° C., and he had not dealt with the question of temperature changes in the liquor itself. As to the effect of the addition of nitrogenous substances to ciders which were susceptible to sickness, in order to produce a more vigorous primary fermentation, he had carried out some experiments in that direction with successful results, but he had not yet experimented on a practical scale. He might say that, in so far as the addition of those nitrogenous foods for the yeasts aided the primary fermentation, they certainly appeared to be of assistance in rendering the liquor less susceptible. It would be observed that in the paper he had emphasised the importance of an active yeast fermentation. Whilst he had referred to early bottling as a promising method of dealing with some of the more susceptible cider, he ought certainly also to give a word of warning against certain dangers attached to such early bottling. It was true that in most cases susceptible ciders, being naturally slow-fermenting kinds, could be bottled at an early stage without giving rise to serious after-fermentation in bottle and too much deposit. Nevertheless it had always to be borne in mind that they might get too much fermentation by bottling too early, and they might also get too great a deposit unless such cider could be consumed within a comparative short space of time. As to the effect of the acidity on the liability to sickness, and the possibility that other acids of importance in this connection might be present in the cider in addition to the malic acid, he had not attempted to make any examination of the acids present in fresh apple juice. He had taken into consideration the work that had been done mainly in Germany on the composition of apple juice, and the results seemed to show that malic acid was certainly the only acid

present in any appreciable quantity. Certainly acids like lactic acid were absent. Owing, however, to the mixture of bacteria present in the juice at the outset they might get other fermentations going on concurrently with the normal alcoholic fermentation, and certain acids, lactic acid for instance, might be produced by such subsidiary fermentations. In that way the inhibitory effect of such acids might at times occur; but, so far as the work referred to in his paper went, it could be accepted that the primary effect was produced by the malic acid, since repeated experiments with sterilised apple juices and pure cultures had been made. Similar results illustrating the effect of acidity had also been obtained with artificial nutrient culture solutions, to which citric and tartaric acids had been added in varying quantities. With regard to the aldehyde question, Dr. Thorne had indicated there might be evaporation of a certain quantity of aldehyde in cask, and that evaporation could not take place in the bottle. However, the tannins in the cider seemed to be the principal factors for reacting with and, therefore, reducing the amount of the aldehyde constituents. The point which Mr. Brewis had raised with regard to the variation in the results of the same bottling from the same lot of casks of cider, was one commonly met with, and he admitted that it was difficult to explain. Personally he was inclined to think that the difference of aëration which occurred during the course of bottling as the cask became empty might have something to do with it, but he had no sufficiently reliable details to put forward in support of that view. He merely suggested it as a possible explanation. With regard to the question of the odour characteristic of cider sickness which Mr. Cannon had raised, he quite agreed with him that there was something present in addition to the aldehyde odour. With such a mixture of volatile compounds various esters and other compounds were probably formed, and these might account for the additional feature in the aroma. In working with pure sugar solutions the aroma of the sugar solution after infection and fermentation with the sickness organism was entirely distinct from the cider sickness odour. It had the garlic character, to which Mr. Cannon had referred, very much more strongly pronounced. There were other variations in the character of the aroma. In the case of certain ciders where the acidity had been modified by partial or complete neutralisation by calcium carbonate, the aroma obtained had a decided resemblance to

that of chocolate. The enquiry as to the relation of albuminoid matter to the acidity of the juice and its bearing on the question of susceptibility to sickness was also concerned with the rate of fermentation of the juice. It was known as the result of investigations at Long Ashton that during the course of ripening of an apple the amount of soluble organic nitrogenous substances suitable for yeast nutrition in the juice diminished up to a certain point of ripeness. That might be regarded as the optimum point of ripeness for vintage purposes. Afterwards the fruit became overripe and the soluble nitrogenous constituents available for yeast nutrition gradually increased again. The acidity diminished throughout ripening. The rate of fermentation followed the same course as that of the nitrogenous bodies referred to. For any given apple, therefore, it followed from the results quoted in the paper, that its juice was most susceptible to sickness at the moment of optimum ripeness; so far the rate of fermentation factor alone was of influence. On the other hand, since the acidity was constantly diminishing, even after the stage of optimum ripeness was passed, it might very well happen that the juice from fruit in an overripe condition was even more susceptible. That, no doubt, was the case in many instances. Regarded solely from the sickness point of view, the safest method would be to mill the fruit in an underripe state. There had generally been little difficulty in detecting beforehand, at the National Fruit and Cider Institute, which of the ciders made there were likely to develop sickness. The general procedure adopted had been to take samples of each juice as it came from the press, placing them after analysis in bottles holding from 75 to 300 c.c. in an incubator at about 28° C. The smaller size had recently been found adequate for the purpose. The acidity determinations indicated in a preliminary way which samples should be regarded with suspicion. All those with acidities below 0·45 per cent. of malic acid might be considered as liable to the disorder. Those with acidities ranging between 0·45 and 0·7 per cent. were held to be doubtful, the chances being in favour of their immunity; and those showing acidities above 0·7 per cent. were in nearly every case safe. The records of the rate of fermentation were then taken, readings of the specific gravity being made every 48 hours. If the average daily fall was above 5 points, the cider might be considered as likely to resist sickness; if below that figure, sickness was probable if the degree of acidity was

low. Frequently the fermentation ceased when the specific gravity was still above 1015; and in those instances sickness almost invariably occurred, unless the original acidity of the juice exceeded 0·5 per cent. When these details had been obtained, it was a comparatively simple matter by applying the knowledge of the disorder now available to blend individual ciders in such fashion as to reduce in most instances the risk of sickness practically to vanishing point, provided that a suitable selection of ciders was at command. It would be gathered from these remarks that it was tacitly assumed that the sickness organism was invariably present in all juices. That was probably the case in many districts. There were, however, localities where sickness was comparatively rare, and possibly in those instances the bacterium might be frequently absent. The foregoing remarks covered many of the points raised in Mr. Riley's letter. The incubator test could be also made in the way he had described, but there were frequently complications in the results owing to the acetification which quickly occurred when fermented ciders were exposed to high temperatures. This caused a rise in acidity which might at times be responsible for negative results as regarded sickness. Possibly the cases he had mentioned, where certain pipes of cider turned sick contrary to expectation from the incubator tests, might be ascribed to this cause. They might also possibly be due to the fact that the bacterium was only present in the casks in very small numbers at the time the samples were taken, so that the relatively small volume taken for trial contained so few that they were unable to make headway within the five-day period over which the incubator tests extended. It had been frequently observed in the pure culture experiments with sterile media that the infection was not successful if the number of cells of the organism added was very small. It might also be asked if the pipes in question were safeguarded against infection from other sources at the time the samples were taken and subsequently. There appeared to be no serious objection to the addition of brewer's yeast in order to promote more vigorous fermentation, and in the paper itself reference was made to an experiment where its use gave very satisfactory results.

The proceedings terminated with a hearty vote of thanks to the Author.