der starken Verwitterung der Gesteine schwierig zu deuten. Vielleicht werden chemische Analysen des Alkaligehaltes der weniger verwitterten Glieder dieser feinkörnigen Gesteine noch nähere Aufklärung über die komplizierten Verhältnisse in dieser Serie geben.

In einem dritten Komplex (Keknenoserie) wurden örtlich viele grosse Eruptivblöcke (bis 3 m in Diameter) gefunden, die vielleicht dem Perm dieser Serie entstammen. Diese Gesteine gehören auch zu den Albititen. Teilweise sind diese Eruptivgesteine sicher als Ganggesteine zu betrachten, da etwas feinkörnigere Gänge in den grossen Eruptivblöcken auftreten. Die Gesteine sind in etwa ähnlicher Weise entwickelt wie die aus dem erstbeschriebenen Komplex. Der Feldspat ist ausnahmslos Albit. Als dunkle Gemengteile spielen Augit und brauner Amphibol eine wichtige Rolle. Die Pyroxenkristalle zeigen öfters unregelmässige Ränder von Aegirin, während die Amphibole mitunter eine sehr schmale Randzone von crossitischem Amphibol erkennen lassen.

Die Albitite mit Alkaliamphibolen und Alkalipyroxenen, welche dieser Serie entstammen, sind sicher nicht von metamorpher Herkunft. Die Gesteine zeigen eine weniger komplizierte Zusammenstellung als die Albitite des erstbeschriebenen Komplexes, z.B. ohne beträchtliche Mengen von Mineralien der Epidotgruppe. Es ist wohl wünschenswert, diese Gesteine, welche auch vom Mother Lode District in Kalifornien [17]) beschrieben worden sind, in Gegensatz zu den Albititen ohne Alkaliamphibole oder Alkalipyroxene als einen Gesteinstypus für sich abzusondern.

Die Gesteinstypen der drei genannten Komplexe sind also alle durch einen Reichtum an Natronfeldspat ausgezeichnet, während nur das Perm der Sonnebaitserie auch viel Kalifeldspat enthält.

Eine eingehendere Beschreibung der verschiedenen Gesteinstypen wird in einer späteren Publikation gegeben werden.

[17]) Siehe z.B.: A. KNOPF, The Mother Lode System of California. U.S.G.S. Prof. Paper 157 (1929).

Microbiology. — *Manometric investigations on bacterial denitrification.* By E. VAN OLDEN. (Communicated by Prof. A. J. KLUYVER.)

(Communicated at the meeting of April 27, 1940.)

Introduction.

In the course of a study on denitrification with cellulose as a substrate, it was found desirable to carry out some preliminary experiments regarding the mechanism of denitrification with simpler hydrogen donators.

Until now denitrification — which term is used here in the restricted sense of nitrate reduction leading to nitrogen evolution — has nearly always been studied in growing cultures. In order to simplify the problem, it seemed of importance to attempt to study in experiments of short duration the behaviour of a given population of denitrifying bacteria towards nitrate, both in the absence and in the presence of suitable organic substrates. This led to the testing of the applicability of the manometric method for this purpose [1]).

It will be clear that denitrification, *i.e.*, the dissimilation process by which hydrogen is transferred from an organic substrate to nitrate as a hydrogen acceptor, can only be studied with the aid of the manometric method, if nitrogen production is a true measure for the oxidation of the substrate. As soon as accumulation of intermediate non-gaseous products of the nitrate reduction would occur, the manometric readings will not give any more a correct indication of the oxidation of the substrate.

The question whether under the conditions of the experiment denitrification is restricted indeed to the conversion:

$$5\,AH_2 + 2\,HNO_3 \rightarrow 6\,H_2O + N_2 + 5\,A$$

can be decided by studying the course of the gas evolution as a function of time. If nitrogen production starts immediately after adding the nitrate, and if a rectilinear relation between nitrogen production and time exists, it is highly improbable that an accumulation of intermediate stages of the nitrate reduction takes place.

However, if nitrogen production is delayed and the rate of gas evolution increases slowly with time, this indicates that nitrate reduction does not lead directly to free nitrogen, and that accumulation of some intermediate stage — probably nitrite or hyponitrite — occurs.

A second possible complication seemed to be the following.

In manometric experiments on dissimilation the conditions are usually

[1]) In a recent paper of YAMAGATA (*7b*) the author mentions in a footnote, that he also has successfully applied the Warburg technique to the study of the process in question.

chosen in such a way that they are as unfavourable as possible for growth and proliferation of the cells. For this purpose use is made of bacterial suspensions of great density, whilst as a rule the medium is devoid of any nitrogen source. However, it results from the investigations made by BARKER (2), GIESBERGER (5) and CLIFTON (3), that even under such conditions assimilation is not altogether prevented.

Under the conditions of my experiments, where nitrate had to be added in order to act as a hydrogen acceptor, there seemed to be great danger that a non-negligible part of the nitrate would be involved in assimilatory processes, and in doing so, would seriously interfere with the study of the dissimilation process.

As will appear from the following pages, it proved possible to find conditions under which the discussed complications could be avoided.

Methodical remarks.

All experiments were carried out with *Micrococcus denitrificans* BEIJERINCK. Initially a strain from the collection of the "Laboratorium voor Microbiologie" at Delft was used, which was the same strain as has extensively been studied by ELEMA in connection with his investigations on the redox potentials occurring in media containing denitrifying bacteria. ELEMA (4) has given experimental proof that under certain conditions this strain is able to reduce nitrate quantitatively to nitrogen with simultaneous oxidation of various simple organic hydrogen donators to carbon dioxide and water.

However, this strain proved to have some drawbacks which will be discussed in the following chapter.

For this reason a number of cultures of denitrifying bacteria were tested on their respiratory activity, both in the absence and in the presence of substrates, and on their denitrifying ability in media containing different substrates.

As a result of this investigation another strain of *M. denitrificans*, isolated from an enrichment culture in a cellulose nitrate medium, was chosen for the further experiments.

For details concerning the manometric method of Warburg I refer to: DIXON, Manometric methods, Cambridge 1937.

The bacteria were collected by washing the crop of several agarplate-cultures either with distilled water, or with 0.9 % NaCl solution, and by centrifuging these suspensions in an ordinary centrifuge at the speed of 4000 r.p.m. In some cases they were obtained from a culture in a liquid medium by centrifuging in a Sharples Super-centrifuge at the speed of 35000 r.p.m. The centrifuged bacterial mass was washed twice with water, and either finally suspended in a Na_2HPO_4—KH_2PO_4 buffer solution of pH 7.1 and molarity 1/15, or suspended in distilled water in which case enough concentrated buffer solution was given in the Warburg vessel to yield also a solution of molarity 1/15.

The volume of the liquid in the Warburg vessel was always 3 cc. All manometric experiments were made in duplicate, at 30° C.

In the denitrification experiments the vessels were filled with nitrogen gas which was first led over a glowing copper spiral to free it from traces of oxygen, always present in the commercial product. The nitrate was added from one of the sidebulbs of the vessel at the same time as the substrate, or the substrate was already present in the main vessel in contact with the bacterial suspension, and the nitrate only added, when the vessels had attained temperature equilibrium in the waterbath. The substrate was always given in excess to the nitrate in order to enable the bacteria to reduce all nitrate present to nitrogen.

In the interpretation of the pressure changes observed it was assumed that under anaerobic conditions with nitrate as hydrogen acceptor the gases evolved were only carbon dioxide and nitrogen. As will be seen, the results obtained justify this assumption. The separate estimation of the gases in question was accomplished in the usual way by determining both the gas evolution in the presence of 0.2 cc 20 % potassium hydroxyde in one manometer vessel, and in another vessel in the absence of KOH.

Qualitative reactions on nitrite were performed with the aid of GRIESS–ROMIJN's nitrite reagent. The presence of nitrate was tested after TILLMANS, the nitrite, if present, being destroyed by boiling the solution with urea and sulfuric acid.

Regarding the presentation of the results in Tables 1 to 5 it needs scarcely be remarked that the term Q_{O_2} is the amount of oxygen consumed in one hour by a quantity of bacteria corresponding to one mg. dry weight.

An analogous term can be introduced to describe the rate of denitrification: Q_{N_2}. Q_{N_2} is the amount of nitrogen liberated by the reduction of nitrate under anaerobic conditions in one hour by a quantity of bacteria corresponding to one mg. dry weight.

For the sake of comparison between aerobic oxidation and oxidation by means of nitrate oxygen, it must be observed that the amount of gaseous nitrogen produced, multiplied by 2.5, is equal to the nitrate oxygen consumed in the oxidation of the substrate, assuming that the equation: $2HNO_3 = H_2O + N_2 + 5O$ holds.

Experimental.

As already mentioned the first experiments were made with the strain of *Micrococcus denitrificans* from the collection of the "Laboratorium voor Microbiologie" at Delft.

The bacteria used in the first experiment were cultivated aerobically on tapwater-glycerol-agar containing 0.1 % nitrate. In the manometric experiments gas evolution only started two hours after the nitrate had been added, both in the absence and in the presence of the glycerol.

In the vessel in which both glycerol and nitrate were present, the rate of gas production slowly increased till after $5\frac{1}{2}$ hours the rate rapidly

decreased. It appeared that at the end of the experiment the theoretical amount of nitrogen had been evolved; in agreement herewith neither nitrate nor nitrite were found.

When only nitrate — but no substrate — had been added to the bacterial suspension the rate of nitrogen evolution was almost constant, very slowly decreasing at the end of the experiment. The nitrogen produced was only part of the theoretical amount which could be expected from the nitrate added. At the end both nitrate and nitrite were present.

The second experiment was made with bacteria cultivated in a liquid medium containing 2 % glycerol and 0.5 % potassium nitrate, in which medium active denitrification took place during the growth of the bacteria.

This time nitrogen production started immediately after the addition of the nitrate. Moreover the rate of the gas evolution was constant, both in the absence and in the presence of glycerol. When glycerol was present, the rate of the gas production was somewhat more rapid than in the presence of nitrate only. In both cases the rate of the nitrogen production only decreased at the moment that almost the theoretical amount of gas was evolved. In the absence of nitrate no gaseous metabolism was observed under anaerobic conditions.

In a way these results may be deemed quite satisfactory, in so far as they prove that it is possible to find conditions under which nitrogen evolution — manometrically established — is a correct measure of the rate of denitrification. Nevertheless, the fact that there was only a small difference in the rate of nitrogen evolution in the experiments with and without substrate was quite unexpected, and undesirable from the standpoint of getting a clear insight into the nature of the dissimilation process. The only possible explanation of this phenomenon seemed to be that the bacteria in question were able to oxidize their own reserve materials with the aid of the nitrate added, and that we are dealing here with a process which may be termed: "endogenous denitrification". Until now this process has escaped the attention of the investigators who have confined themselves almost completely to a study of denitrification in growing cultures.

The correctness of the interpretation given is strongly supported by the observation that the bacteria were also characterized by a high endogenous respiration.

In order to avoid the difficulty of the high endogenous denitrification steps were taken to find, if possible, a strain which did not show this complication, and as a result hereof the further experiments were made with a strain of *Micrococcus denitrificans*, already referred to in the preceding chapter.

The result of the first experiment as recorded in Table 1 was not encouraging. No gas production whatever was observed when a suspension of bacteria grown on plain peptone agar was brought together with nitrate under anaerobic conditions. Even no nitrite, the occurrence of which is the first indication of nitrate reduction, could be detected. Aerobically the

TABLE 1.

Micrococcus denitrificans.

Denitrification and respiration.

Substrate: Na-acetate 0.2 cc of a 5 % solution. KNO_3 added in the anaerobic experiments: 0.4 cc of a 0.5 % solution (corresponding with a nitrogen production of 221 mm³, 0° C. and 760 mm Hg).

Bacteria cultivated aerobically on peptone agar without nitrate; 48 hours at 30° C.

	In air				In nitrogen, with nitrate			
	with substrate		without substrate		with substrate		without substrate	
	oxygen consumed mm³	Q_{O_2}	oxygen consumed mm³	Q_{O_2}	nitrogen produced mm³	Q_{N_2}	nitrogen produced mm³	Q_{N_2}
0— 60 min.	373	47	43	5	6		8	
60—120 „	496	63	30		2		—1	
120—180 „	525	67	30		—2	0.4	0	0.3
180—240 „	173	22	22	2.8	4		3	
240—300 „	(118)	(15)	(16)		4		2	
Total	1685		141		14 [1])		12 [1])	

[1]) Nitrate present, no nitrite formed.

bacteria used behaved normally and oxidized the substrate: sodium acetate rapidly.

Hereupon, the same experiment was repeated with bacteria, grown aerobically on peptone agar containing 0.5 % potassium nitrate. (Table 2). However, only a slow nitrogen production was observed, when nitrate was added to the suspension already in contact with the substrate. Moreover, the rate of this gas evolution showed a distinct rise with time, whilst the final volume of the nitrogen obtained corresponded only to a part of the nitrate added.

Although these results meant real progress, when compared with the outcome of the first experiment, they still were unsatisfactory, because of the inconstant rate of gas evolution and because of the incomplete conversion of the nitrate added. That this behaviour was not due to a general lack of activity of the bacteria was clearly shown by the fact that the aerobic oxidation of sodium acetate proceeded at a quite normal rate, the Q_{O_2} being no less than 60.

The fact that the bacteria which had been cultivated in the absence of nitrate (Experiment 1) were quite unable to denitrify in the "resting state", whilst the bacteria which had been cultivated in the presence of nitrate (Experiment 2) showed an unmistakable activity in this respect,

TABLE 2.

Micrococcus denitrificans.

Denitrification and respiration.

Substrate: Na-acetate 0.2 cc of a 5 % solution. KNO_3 added in the anaerobic experiments: 0.4 cc of a 0.5 % solution (corresponding with a nitrogen production of 221 mm³, 0° C. and 760 mm Hg).

Bacteria cultivated aerobically on peptone agar, containing 0.5 % KNO_3; 48 hours at 30° C.

	In air				In nitrogen, with nitrate		
	with substrate		without substrate		with substrate		without substrate
	oxygen consumed mm³	Q_{O_2}	oxygen consumed mm³	Q_{O_2}	nitrogen produced mm³	Q_{N_2}	nitrogen produced mm³
0— 60 min.	340	44	37		12	1.5	4
60—120 „	457	59	34		9	1.2	—1
120—180 „	467	60	20		14	1.8	0
180—240 „	281	36	19		24	3.0	5
240—300 „	(130)	(17)	(30)		22	2.8	0
Total	1675		140		81 [1]		8 [1]

[1] Nitrate present, nitrite formed.

TABLE 3.

Micrococcus denitrificans.

Denitrification.

Substrate: Na-acetate 0.2 cc of a 5 % solution. KNO_3 added: 0.4 cc of a 0.84 % solution (corresponding with a nitrogen production of 372 mm³, 0° C. and 760 mm Hg).

Bacteria cultivated on peptone agar containing 2 % KNO_3; 48 hours in an anaerobic jar at 30° C.

	In nitrogen, with nitrate			
	with substrate		without substrate	
	nitrogen produced mm³	Q_{N_2}	nitrogen produced mm³	Q_{N_2}
0— 30 min.	112	33.4	45	13.4
30— 60 „	78	23.3	20	5.97
60— 90 „	75	22.4	20	5.97
90—120 „	62	18.5	20	5.97
120—180 „	17	2.5	37	5.5
180—240 „	0	0	(26)	
Total	344 [1]		168 [2]	

[1] Nitrate and nitrite absent.

[2] Nitrate present, nitrite trace.

made it seem probable that a still better activity could be expected for bacteria which had been completely depending on denitrification during the period of their growth.

For this reason a third experiment was made with bacteria, cultivated anaerobically on peptone agar containing 2 % nitrate. Table 3 shows that these bacteria were indeed capable of a very vigorous denitrification. After a short initial period of abnormal high activity the rate of nitrogen production arrived at a fairly constant level which was maintained till almost the theoretical amount of gas was produced. Then the rate rapidly declined to zero. In this case a Q_{N_2} ! was observed which corresponded with a Q_{O_2} of the same order of magnitude as that actually observed in the previous aerobic experiments.

The results of the experiments given in Tables 1, 2 and 3 are represented graphically in Figure 1. In this figure the difference in bacterial quantities added pro vessel is accounted for.

The figure clearly demonstrates the marked difference in denitrifying ability of the bacteria in its dependence on cultural conditions.

It seemed desirable to corroborate these results in some further experi-

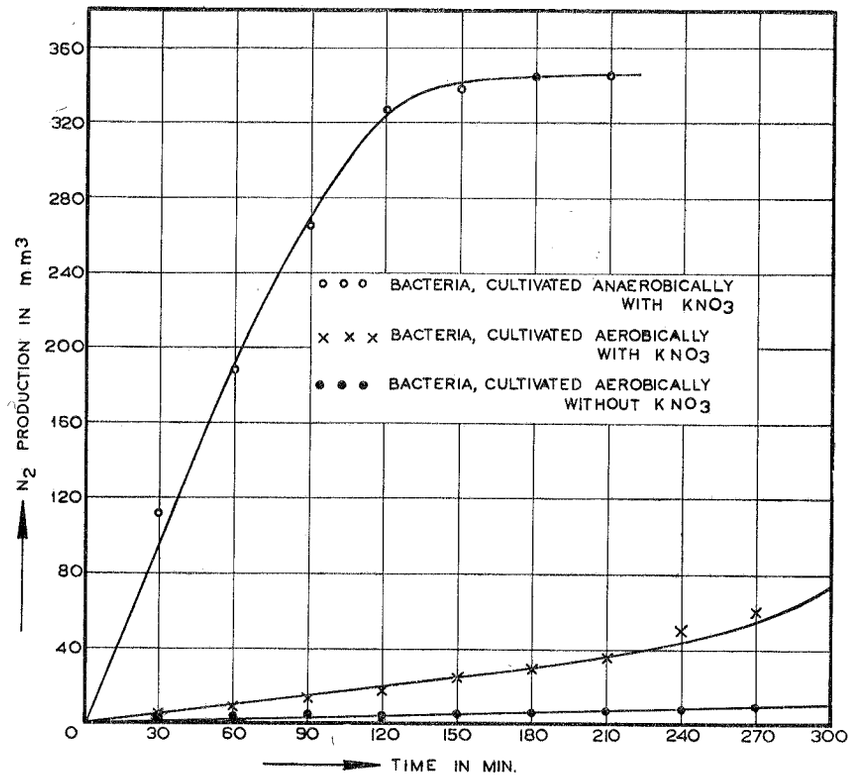

Fig. 1.

Micrococcus denitrificans; denitrification with Na-acetate as substrate.

ments. For this purpose comparative experiments were made with bacteria partly grown aerobically on peptone agar with 2 % nitrate, partly on the same medium in an atmosphere of pure hydrogen. As it appears from Tables 4 and 5 it was again found that only the latter bacteria were able to bring about denitrification at a rate of the same order of magnitude as that of the aerobic dissimilation ($Q_{O_2} = 2.5\ Q_{N_2}$!).

The bacteria cultivated aerobically only slowly produced nitrogen from the nitrate added, the rate of the gas evolution, moreover, increasing with time. At the end of the experiment nitrate was left, and nitrite was shown to be present in the medium.

In contrast herewith, the bacteria cultivated anaerobically immediately started to reduce the nitrate to free nitrogen in the experiment with substrate. During the first two hours the rate of nitrogen evolution was practically constant, and then quickly dropped to zero. The nitrogen produced corresponded with the theoretical amount for complete conversion of the nitrate. Neither nitrate, nor nitrite, were present in the medium at the end of the experiment.

TABLE 4.

Micrococcus denitrificans.

Denitrification and respiration.

Substrate: Na-acetate 0.2 cc of a 5 % solution. KNO_3 added in the anaerobic experiments: 0.4 cc of a 0.84 % solution (corresponding with a nitrogen production of 373 mm³, 0° C. and 760 mm Hg).
Bacteria cultivated aerobically on peptone agar, containing 2 % KNO_3; 48 hours at 30° C.

	In air				In nitrogen, with nitrate			
	with substrate		without substrate		with substrate		without substrate	
	oxygen consumed mm³	Q_{O_2}	oxygen consumed mm³	Q_{O_2}	nitrogen produced mm³	Q_{N_2}	nitrogen produced mm³	Q_{N_2}
0— 30 min.	209	46.1	37	8.2	6	0.66	9	0.66
30— 60 „	253	55.8	34		0		−3	
60— 90 „	305	67.2	22		6	2.0	6	
90—120 „	388	85.5	18		12		9	
120—180 „	427	47.1	39		14	1.54	0	
180—240 „	173	19.1	37		19	2.09	5	
240—300 „	166	18.3	29	3.2	27	2.97	3	
Total	1921		216		84 [1]		29 [1]	

[1] Nitrate present, nitrite formed.

TABLE 5.

Micrococcus denitrificans.

Denitrification and respiration.

Substrate: Na-acetate 0.2 cc of a 5 % solution. KNO_3 added in the anaerobic experiments: 0.4 cc of a 0.84 % solution (corresponding with a nitrogen production of 373 mm³, 0° C. and 760 mm Hg).
Bacteria cultivated anaerobically on peptone agar containing 2 % KNO_3; 48 hours at 30° C.

	In air				In nitrogen, with nitrate			
	with substrate		without substrate		with substrate		without substrate	
	oxygen consumed mm³	Q_{O_2}	oxygen consumed mm³	Q_{O_2}	nitrogen produced mm³	Q_{N_2}	nitrogen produced mm³	Q_{N_2}
0— 30 min.	171	60.8	41	14.5	96	34.1	29	10.3
30— 60 „	204	70.7	17		79	28.1	12	4.3
60— 90 „	255	90.6	20		82	29.1	8	2.8
90—120 „	266	94.5	6		88	31.3	14	5.0
120—180 „	502	89.2	11	1.9	26	4.6	25	4.4
180—240 „	233	41.4	+2		0		22	3.9
240—300 „	120	21.3	+2		0		10	1.8
Total	1751		91		371 [1]		120 [2]	

[1] Nitrate and nitrite absent.
[2] Nitrate present, nitrite absent.

In the experiment in which only nitrate had been added to the suspension, but no substrate, the decreasing rate of the nitrogen production showed the rapid disappearing of the reserve materials present in the cells. It is worth-noticing that nitrite was absent at the end of this experiment.

Discussion of results.

As has been remarked in the Introduction the aim of the investigation made was to test the possibility of studying denitrification of "resting bacteria" with the aid of the manometric method.

The foregoing experiments show that, indeed, under certain conditions denitrification with "resting cells" of *Micrococcus denitrificans* can be obtained which answers the requirements made. Hereto it was found necessary to use cells which during their development had depended on denitrification as a sole source of energy, *i.e.*, which had been grown anaerobically in the presence of nitrate. With these cells in the manometric experiments an evolution of nitrogen could be observed of the same order of magnitude as that of the aerobic gaseous metabolism. Moreover, the

rate of nitrogen production proved to be constant over a long period, namely as long as a sufficient amount of nitrate still was present. This constant rate offers sufficient proof that under the condition of the experiment no intermediate stages of the nitrate reduction were formed in any appreciable amount. In agreement herewith the total amount of nitrogen evolved corresponded with the amount which should be obtained from the nitrate added on the basis of a complete conversion.

The constant rate of nitrogen evolution also shows that the anxiety expressed in the Introduction, *viz.*, that assimilatory processes might interfere with the study of denitrification as such, has been unjustified.

From all this we may conclude that in principle the manometric method is suitable for the study of denitrification. Experiments in which besides the nitrogen also the carbon dioxide produced from the substrate has been estimated are already in progress, and the results will be reported in a forthcoming communication.

Two more points should be brought to the front.

In the first place it has been found that with certain bacteria denitrification proceeds almost equally well in the presence and in the absence of an organic substrate. This can only mean that these bacteria are able to oxidize reserve materials present in the cells at the expense of the nitrate added. This process has been termed "endogenous denitrification" in analogy to the well-known process of "endogenous respiration". To a smaller extent this endogenous denitrification is encountered in all cases.

Secondly the investigation has clearly shown the decisive influence of the previous history of the cells on their denitrifying activity, only those cells being appreciably active which originate from cultures in which denitrification has been the main source of energy. In this connection it may be remarked that both AUBEL and GLASER (1) and YAMAGATA (7*a* and *b*) have recently given arguments in favour of the view that denitrification is depending on the presence of a special nitrate activating enzyme system in the cells. The experiments made show, therefore, that this "nitrate reductase" must be considered as an "adaptive enzyme" in the sense of KARSTRÖM (6).

REFERENCES.

1. E. AUBEL, C. R. Acad. Sciences **207**, 348 (1938).
   E. AUBEL et Mlle GLASER, C. R. Soc. Biol. **127**, 473 (1938); ibid. **127**, 976 (1938).
   E. AUBEL, C. R. Soc. Biol. **128**, 45 (1938).
2. H. A. BARKER, J. Cell. and Comp. Physiol. **8**, 231 (1936).
3. C. E. CLIFTON, Enzymologia **4**, 246 (1937).
4. B. ELEMA, De bepaling van de oxydatie-reductiepotentiaal in bacteriëncultures en hare beteekenis voor de stofwisseling. Delft (1932).
5. G. GIESBERGER, Beiträge zur Kenntnis der Gattung *Spirillum* Ehbg. Delft (1936).
6. H. KARSTRÖM, Ergebn. d. Enzymforschung **7**, 350 (1938).

7*a*. S. YAMAGATA, Acta phytochimica **10**, 284 (1938).

7*b*. ———, Acta phytochimica **11**, 145 (1939).

Hydrodynamics. — *On the application of viscosity data to the determination of the shape of protein molecules in solution* *). By J. M. BURGERS. (Mededeeling N°. 38 uit het Laboratorium voor Aero- en Hydrodynamica der Technische Hoogeschool te Delft.)

(Communicated at the meeting of April 27, 1940.)

15. *System of four spheres lying in the corners of a square.* — The system of 4 spheres can be treated along similar lines as the cubical system; account, however, must be taken of the circumstance that the components of the angular velocity which the system assumes under the influence of a shearing motion of the liquid will be different from those which were found for the cube.

The value of λ for the system of four spheres lying in the corners of a square becomes:

$$\lambda = \frac{1}{4}\left(1 + 2{,}70\,\frac{R}{a} - 0{,}04\,\frac{R^2}{a^2}\right) \quad . \quad . \quad . \quad . \quad . \quad (51)$$

In order to investigate the behaviour of the system and its influence upon the effective viscosity of a liquid in shearing motion, we take the plane of the square as the ξ_1, ξ_2-plane. The ξ_3-coordinates of the centres of all four spheres then will be zero. This makes it possible to define the rotation of the system and the quantities describing the remaining relative motion of the liquid in the following way, which at once refers to the case of a field of flow of general type, as given by eq. (45): — With the aid of eqs. (30) we obtain:

$$\dot{\xi}_i = \sum_{jkl} a_{ik}\, a_{jl}\, \varkappa_{kl}\, \xi_j \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad (52)$$

where the summation with respect to j now can be restricted to the terms $j=1$, $j=2$. We consequently can define the components ω_{31} ($=-$ angular velocity about the ξ_2-axis) and ω_{32} ($=+$ angular velocity about the ξ_1-axis) of the square in such a way that the value of $\dot{\xi}_3$ is wholly determined by them. When the components of the angular velocity about the three axes are denoted by p, q, r respectively, we then have:

$$p = \omega_{32} = \sum_{kl} a_{3k}\, a_{2l}\, \varkappa_{kl}; \qquad q = -\omega_{31} = -\sum_{kl} a_{3k}\, a_{1l}\, \varkappa_{kl};$$

and:

$$\dot{\xi}_3 = \omega_{31}\,\xi_1 + \omega_{32}\,\xi_2 = -q\,\xi_1 + p\,\xi_2.$$

*) Continued from these Proceedings **43**, 435 (1940).