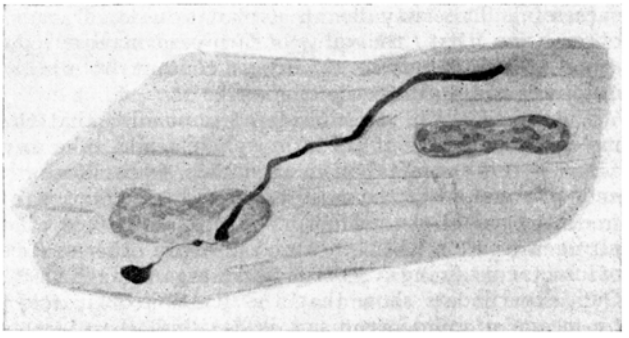

Fig. 2. – Gonflement d'un granule à l'extrémité d'un filament métaterminal, qui va disparaître. Même grossissement.

appareil métaterminal dans le tissu fibreux de la dure-mère, que les granules gonflés ou les rondelles. Comme ces débris persistent un certain temps, ils forment en quelques points de petits amas fortement teintés par le dépôt d'argent.

Les filaments métaterminaux sont tout à fait comparables aux collatérales transitoires que G. Levi et H. Meyer[1], J. Fabre et H. Mégevand[2], puis C. C. Speidel[3] ont observées dans les cultures de tissu nerveux ou bien lors du développement de nerfs moteurs ou sensitifs. L'émission de ces minces prolongements correspond sans doute à des variations brusques et réversibles de la tension superficielle des fibres nerveuses

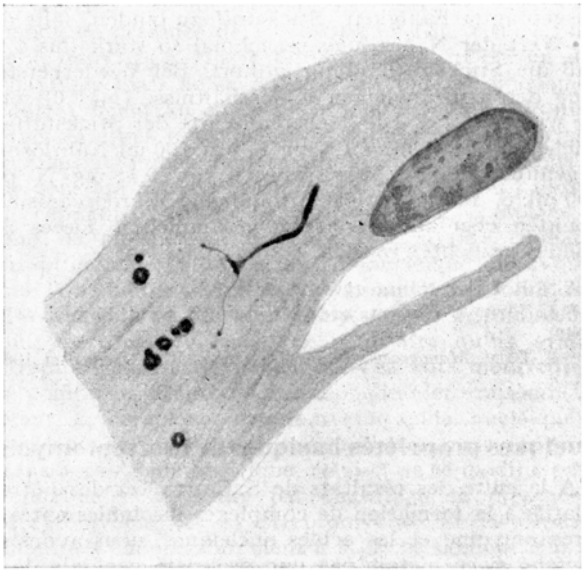

Fig. 3. – Rondelles et granules laissés en place par des filaments atrophiés. Reconstitution de l'appareil métaterminal après sa disparition. Même grossissement.

durant leur croissance. En ce qui concerne les terminaisons sensitives de la dure-mère chez le Rat, les modifications qui se produisent au niveau du bouton terminal, semblent soumises à une succession de phases. Après leur disparition, les filaments métaterminaux se reconstituent et un nouveau cycle recommence. On voit alors ces fins prolongements s'allonger en direction des rondelles ou des granules laissés en place par les filaments précédents. Il est possible que ces variations cycliques de la structure des terminaisons sensitives dans la dure-mère soient en rapport avec des différences périodiques dans la nature des sensations qu'elles transmettent.

A. Weber

Laboratoire de neuro-histologie, Institut d'anatomie de Genève, le 29 juin 1948.

Summary

All sensory endings of the trigeminal nerve in the dura mater of the rat are formed by a small swelling, from which continues the *metaterminal apparatus*, fine filaments at the limit of visibility. Periodically the fine granules at the extremity of the filaments swell, whereas the fibrils themselves disappear, leaving a small, round, argentophil mass. Later the filaments reappear, then redeposit their debris, which persists for a time.

Thus the metaterminal apparatus manifests cyclic variations, which recall the transitory existence of collaterals observed during *in vivo* or *in vitro* development of nerve fibres.

Nitrogen Fixing Microorganisms in the Alimentary Canal of Herbivorous Farm Animals

It is generally known that ruminants are able to live for a long time on a diet excessive in carbohydrate but deficient in nitrogen compounds. The data found in the literature indicate that the daily minimal physiological nitrogen requirement is about 10 g per 100 kg of living weight. But, in many cases, the amount of nitrogen lost daily in fæces and urine is often greater than that contained in the fodder. This indicates that nitrogen of the air probably enters into the metabolism of ruminants in the same way as in insects living on a diet poor in proteins; that is, through symbiotic bacteria.

Nitrogen fixing bacteria may be found in the rumen of ruminants and in the cæcum of Equidæ. To test this possibility, slant agar tubes containing

0·5 %	glucose
0·2 %	succinic acid
0·5 %	NaCl
0·1 %	$CaCO_3$
0·1 %	$MgSO_4 \cdot 7H_2O$
0·07 %	K_2HPO_4
0·3 %	KH_2PO_4
0·05 %	$CaCl_2$
0·01 %	$MnSO_4$
0·0001 %	$NaM_0O_4 2H_2O$
2·0 %	agar-agar
	Na_2CO_3 (to p_H 8·0)
	sterile tapwater,

were inoculated with material taken from healthy animals (cows, sheep, horses) under sterile conditions. Tests revealed that the cultures contained many strains of nitrogen fixing bacteria. 24 hours after inoculation, vigorous strains appear as numerous small (1 mm diameter) colonies. Development proceeds rapidly; within 5–6 days (30°C) the whole surface of the agar slant is covered with a gelatinous mass of bacteria.

The capacity of the bacteria to fix nitrogen on a nitrogen-free culture medium does not diminish after a series of inoculations. In testing the nitrogen fixation of these bacteria the culture medium mentioned above was used, only the amount of agar was 0·05 % and the

[1] G. Levi et H. Meyer, Arch. de Biol. *52*, 133 (1941).

[2] J. Fabre et H. Mégevand, C. R. Soc. de Phys. et d'Hist. nat. de Genève *58*, 79 (1941).

[3] C. C. Speidel, J. compar. Neurol. *76*, 57 (1942).

glucose concentration 0·2%. Some of this culture liquid was added to the agar slant tubes. The bacterial coat was first lifted off and mixed with culture liquid, and then the solution was poured into an Erlenmeyer flask. After vigorous shaking a uniformally distributed suspension is obtained. This suspension is used to inoculate the liquid culture media of 125 ml Erlenmeyer flasks (in each 50 ml culture liquid + 2 ml suspension). Every day three of these flasks are analysed for total nitrogen by the PARNAS-WAGNER micro-Kjeldahl method. The analyses furnish the following data in respect to the nitrogen fixing capacities of the different strains:

Table I

Amount of nitrogen in 100 ml suspension of different strains of nitrogen fixing bacteria of cows, sheep, and horses

Day of experiment	mg N in 100 ml liquid culture medium inoculated with bacteria of the:					
	Cow			Sheep		Horse
	I. d.	V. f.	VI. f.	I. c.	I. e.	I. l.
Start	0·20	0·19	0·18	0·16	0·16	0·17
1st day	0·29	0·27	0·25	0·16	0·19	0·22
2nd day	0·38	0·40	0·28	0·19	0·23	0·35
3rd day	0·40	0·42	0·33	0·27	0·30	0·54
4th day	0·39	0·45	0·35	0·28	0·32	0·55
5th day	0·38	0·55	0·35	0·38	0·38	0·59
7th day	0·39	0·65	0·30	0·56	0·67	0·77
10th day	0·39	0·95	0·55	0·63	0·71	0·79
14th day	0·37	1·18	0·76	1·14	1·13	0·94
20th day	0·40	1·10	0·84	1·11	1·22	0·75

The given data are average values of 3 samples. The differences between these 3 values are statistically insignificant. The increase in nitrogen is most remarkable; this again shows that through these bacteria nitrogen of the air can enter into the metabolism of our farm animals. But the data also show that there are great differences in regard to nitrogen fixing ability between the different strains of bacteria even within the same animal (for instance cow I. d. and cow V. f.).

With regard to the nutrient solution, the question is raised whether succinic acid or glucose or both of them are indispensable for nitrogen fixation. Experiments in this line show that each of these substances alone or both together can effect nitrogen fixation. Glucose alone is more efficient than succinic acid alone. But none of them enables such vigorous nitrogen fixation as do both together.

During the period of fixation, the p_H value often drops, sometimes down to p_H 4·5. At this degree of acidity no more nitrogen fixation can take place, but if the optimum value (p_H 8·0) is experimentally reestablished, nitrogen fixation begins again. This is the case with strain Horse 1.l., where the p_H value dropped from 8·0 to 5·5 within four days. Re-establishment of the p_H value to p_H 8·0 through addition of $NaCO_3$ causes fixation to become vigorous again and results in the maximum amount of nitrogen or 1·8 mg per 100 ml of culture liquid, while the uncharged controls remain at 1·0 mg per 100 ml.

It is known that new-born animals do not possess any intestinal micro-organisms. They acquire them with the first lot of grass or hay, soiled by excrement. If this is so, the same nitrogen fixing bacteria may be found in the fæces too. Indeed, with the method described above, the cultivation of nitrogen fixing bacteria from cow excrements collected at the moment of defecation was successful. This may be an explanation for DARH'S[1] observation, that "the value of farmyard manure (cow dung) is due not only to its nitrogen content, but also to its power to fix nitrogen of the air".

Until recently it was considered impossible that the molecular nitrogen of the atmosphere could take any active part in the metabolism of animals. TÓTH, WOLSKY, and BÁTORI[2] were the first to show that fixation of free nitrogen of the air occurs in aphides. Since then nitrogen fixation has been found in many other species of insects containing symbiotic micro-organisms (TÓTH[3]). Our experiments show that the fixation of nitrogen by means of microorganisms is not limited to insects but exists also in farm animals.

I am indebted to the Swedish Wenner-Gren Foundation for a grant which has fianced this work.

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Microbiological Institute, Uppsala, Sweden, May 14, 1948.

Zusammenfassung

1. Aus steril entnommenem Inhalt des Pansens vom Rind, dem Schafe und des Colcums des Pferdes konnten auf einem stickstofffreien Agarnährboden verschiedene Stämme stickstoffbindender Mikroorganismen gezüchtet werden. 2. In stickstofffreier Nährlösung vermögen diese Rohkulturen Luftstickstoff in verschieden starkem Maße zu binden. 3. Als C-Quelle erwiesen sich Glukose und Bernsteinsäure als geeignet. Glukose allein ergibt bessere Resultate in der Stickstoffanreicherung als Bernsteinsäure allein. Noch besser aber wirken beide Substanzen zusammen. 4. Bei Stämmen mit ausgeprägter Fähigkeit, Stickstoff zu binden, fällt der p_H-Wert der Nährlösung manchmal so stark (bis 4,5), daß die Stickstoffbindung aufhört. Bei Wiederherstellung der ursprünglichen p_H-Verhältnisse (p_H 8,0) setzt sie wieder ein. 5. Die höchsten Werte der Stickstoffanreicherung erreichten 1,8 mg N pro 100 ml Nährlösung gegenüber einem Ausgangswert von 0,18 mg N pro 100 ml. 6. Ähnliche stickstoffbindende Mikroorganismen konnten ebenfalls aus frisch gesammelten Fæces des Rindes gezüchtet werden.

[1] N. R. DARH, Nature *159*, 65 (1947).

[2] L. TÓTH, A. WOLSKY, and M. BÁTORI, Z. vergl. Physiol. *30*, 67 (1942).

[3] L. TÓTH, *Monographs Natural Sciences*, V, 1 (Budapest 1946).

Quelques propriétés basiques de la streptomycine

A la suite des résultats de S. COHEN[1] et des nôtres[2] relatifs à la formation de complexes insolubles entre la streptomycine et les acides nucléiques, nous avons recherché si ce n'était pas une propriété générale de la basicité de la streptomycine de donner des complexes insolubles avec des substances organiques à point isoélectrique acide. Nous avons ainsi obtenu de ces complexes avec l'acide tannique, la céphaline, des acides gras à longues chaînes (oléique, palmitique)[3].

En ce qui concerne plus spécialement les phosphatides, signalons tout d'abord que nous n'avons pu obtenir de

[1] S. COHEN, J. Biol. Chem. *168*, 511 (1947).

[2] F. GROS, B. RYBAK, M. MACHEBŒUF et U. RAMBECH, C. R. Acad. Sci. *226*, 1550 (1948). – F. GROS et B. RYBAK, Helv. chim. acta (sous presse).

[3] En ajoutant une solution de streptomycine à des émulsions en eau distillée ou même salée à 9‰ d'acide oléique ou palmitique amenées à p_H 7,5–8, on constate la formation d'un précipité très abondant qui floculе facilement et adhère aux parois du tube. Avec l'oléate de sodium, la glucosamine-HCl, la triméthylamine-HCl ou encore $ClNH_4$ donnent une très importante précipitation.