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Concluding Remarks

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Concluding remarks

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These past two days have been a fitting recognition of the achievements of a century of researchers in nitrogen fixation. There has been a fascinating combination of history, exciting current developments and speculation about directions for future research and applications. This was the intention of the organizers, and the contributors of papers and those engaging in discussion have each made their marks. The contributions have largely concentrated on science, the 'Natural Knowledge' that is fitting both for the occasion and for this location.

There has been a wide variety of presentations and discussion from several representative parts of the subject of this meeting. I have been impressed by the growing cohesion between genetical, biochemical, chemical and physiological work. This was amply illustrated by the presentations and discussion about the alternative nitrogenases of *Azotobacter* species.

In these days in which there is an emphasis on 'scientific accountability', but also because a matter of principle is involved, it is necessary to make a connection between two important aspects of our subject. Firstly, the study of nitrogen fixation, as an important component of the terrestrial nitrogen cycle, has many features that encourage the involvement of a number of scientific disciplines and produces good science with intrinsic value in each of them. Secondly, nitrogen fixation is, and must continue to be, used for the benefit of people and their environment, largely through agriculture and the conservation of natural resources. Dr Nutman's paper clearly showed the origins of the subject to have been in agriculture. Consequently, some contributions have mentioned features of applications of research to these matters.

In spite of the recognition that connections between fundamental aspects and agriculture need to be developed, it is often difficult to reconcile differences in the terminology of measurements which are made in the field and in the laboratory. In an attempt to relate laboratory measurements and nitrogen fixation in agriculture, I have prepared the following examples.

For several years I have been involved with improvement of methods for using ^{15}N to estimate nitrogen fixation more reliably in various agricultural systems which utilize nodulated legumes in Australia, Thailand and Malaysia. Table 1 shows some of the rates of nitrogen fixation which have been calculated from our experiments in Australia; in table 2, the rates obtained for irrigated soybeans at one experimental site have been transposed from agricultural terms to terms more related to those measured in laboratory experiments. The rates in the field are of the same order as those in the best laboratory experiments with bacteroid suspensions, in which I am rather pleased to have achieved activities of 2–5 nmol NH_3 min^{-1} mg^{-1}.

We have heard about research whose objective is the improvement of nitrogen-fixing systems and about some directed towards the construction of new ones. An important additional consideration arises from application of our better understanding of the properties of existing systems; this may offer more immediate prospects for application. I illustrate this also from some recent Australian studies.

It has long been known that nitrate in the soil, if above certain limits, restricts nodule development in many legumes and may also diminish nitrogen fixation by those nodules that are formed. The limits are often exceeded when legumes are planted in soils previously fertilized with nitrogen or after a period of nitrification during fallowing. Consequently, scientific effort is being directed to the production of legumes and nodule bacteria that are more tolerant of nitrate and so allow nitrogen fixation to proceed promptly, rather than being delayed until the level of soil nitrate is sufficiently depleted by the growing crop. However, sometimes changes to the management of crops may enable better utilization of existing material, with advantage to the farmer. This is illustrated in table 3. By growing winter oats, which removed, with profit, 130 kg N ha^{-1} in the harvested crop, and by using high rates of inoculation with standard commercial inoculants, excellent levels of nitrogen fixation were achieved by the following irrigated summer crop of soybeans. Grain yields were very satisfactory, with improved protein content (data not shown). If used by a farmer, this practice would improve the productivity

TABLE 1. DAILY RATES OF NITROGEN FIXATION: SOME AUSTRALIAN EXAMPLES

(Rates calculated from data in the papers cited, with ^{15}N methods.)

system (location)	measurement period/d	N_2 fixation kg N ha^{-1} d^{-1}	source
sub. clover–ryegrass (A.C.T.)	0–40	1.5	Bergersen & Turner (1983)
	50–56 (after cutting)	6.1	
	0–25 (after cutting)	0.8	Ledgard *et al.* (1985)
lucerne–ryegrass (A.C.T.)	0–25 (after cutting)	0.4	Ledgard *et al.* (1985)
soybeans (Leeton, N.S.W.)	50–114	3.8	Bergersen *et al.* (1985)
	98–114	6.0	
lupins Wagga Wagga (N.S.W.)	129–193	2.2	Evans *et al.* (1987)

TABLE 2. CALCULATED SPECIFIC NITROGEN FIXATION ACTIVITIES OF BACTEROIDS IN FIELD-GROWN SOYBEAN NODULES

(Rates calculated from the values for soybeans in table 1 and data of the author and collaborators from various sources.)

parameter	measurement
measured field rate	3.8 kg N ha^{-1} d^{-1}
	6.0 kg N ha^{-1} d^{-1}
number of plants	270220 ha^{-1}
N_2 fixation per plant	14.1 mg N $plant^{-1}$ d^{-1}
	22.2 mg N $plant^{-1}$ d^{-1}
dry mass of nodules (per plant)	0.4 g
	0.66 g
proportion of bacteroids	25% nodule dry mass
dry mass of bacteroids (per plant)	100 mg
	165 mg
specific N_2 fixation activity of bacteroids *in vivo* (24 h d^{-1} basis)	7.0 nmol NH_3 min^{-1}(mg bacteroid dry mass)$^{-1}$

TABLE 3. SOIL NITRATE, NODULATION, NITROGEN FIXATION AND YIELD OF IRRIGATED SOYBEANS

(Unpublished data of F. J. Bergersen, G. L. Turner, R. R. Gault, L. Morthorpe, M. B. Peoples and J. Brockwell, obtained at Trangie, N.S.W., for Forrest soybeans planted on 11 December 1985.)

soil pretreatment	soil nitrate at planting (p.p.m. by mass)	days from planting	parameter	inoculation rate[a] 0	$N/100$	N	$100N$
fallow	37.7	105	nodulation[b]	0	0	0.75	1.75
		105	P[c]	0	4.4	4.6	7.2
		147	grain (t ha^{-1})	3.0	3.1	3.2	3.3
cropped[d]	18.5	105	nodulation[b]	0	0.92	2.50	3.92
		105	P[c]	0	11.9	29.7	56.8
		147	grain (t ha^{-1})	1.8	2.5	3.1	3.5

[a] N is the normal commercial rate of inoculation.
[b] Nodulation assessed on a scale of 0–5.
[c] P is the percentage of plant-N derived from N_2 fixation, determined from measurements of ^{15}N natural abundance.
[d] 130 kg of cereal-N ha^{-1} was harvested in October 1985.

of his irrigated area and, if the crop residues were properly conserved, would preserve the nitrogen status of the soil, without recourse to nitrogenous fertilizers.

I present these illustrations to emphasize that both the laboratory and field sciences need to develop the connections between them if the benefits of past and future nitrogen fixation research are to be realized in accordance with their potential. Further, the connections are important to the framing of appropriate objectives for improvement by application of the vastly improved techniques of molecular genetics, biochemistry and chemical technology which are now available. Without proper development of these connections, there is some danger that our science will wither on the vine (or, perhaps more appropriately, many potentially fruitful pods will fall unfilled).

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