ΑΝΑΚΟΙΝΩΣΕΙΣ ΜΗ ΜΕΛΩΝ

ΒΟΤΑΝΙΚΗ.— The assimilation of nitrates by higher plants*, *by* ***C. P. Sideris.*** Ἀνεκοινώθη ὑπὸ κ. Ι. Πολίτου.

The problem of nitrate assimilation by higher plants still seems to occupy the attention of students of nitrogen metabolism. It appears that the views of the different investigators on this matter are confusing as certain of them have claimed that nitrates may be assimilated by all the different tissues of the plant either in the presence or absence of light as long as there are available carbohydrates while others are still questioning such assertions.

Schimper in 1888 claimed that nitrates become depleted in the green leaves of plants exposed to daylight but that they are retained in those kept in the dark. Warburg and Negelein in 1920, and Moore in 1924 have supported Schimper's claim. The work, however, of other investigators such as Acqua in 1899, Thomas in 1927, Eckerson in 1924 and the more recent one of Davidson and Shive in 1934 complicate the issue of nitrate assimilation considerably because their views do not coincide with those of the previous investigators. The literature on nitrogen metabolism is a very extensive one and I do not wish to extend my comments beyond these few and brief remarks particularly in a paper of this kind.

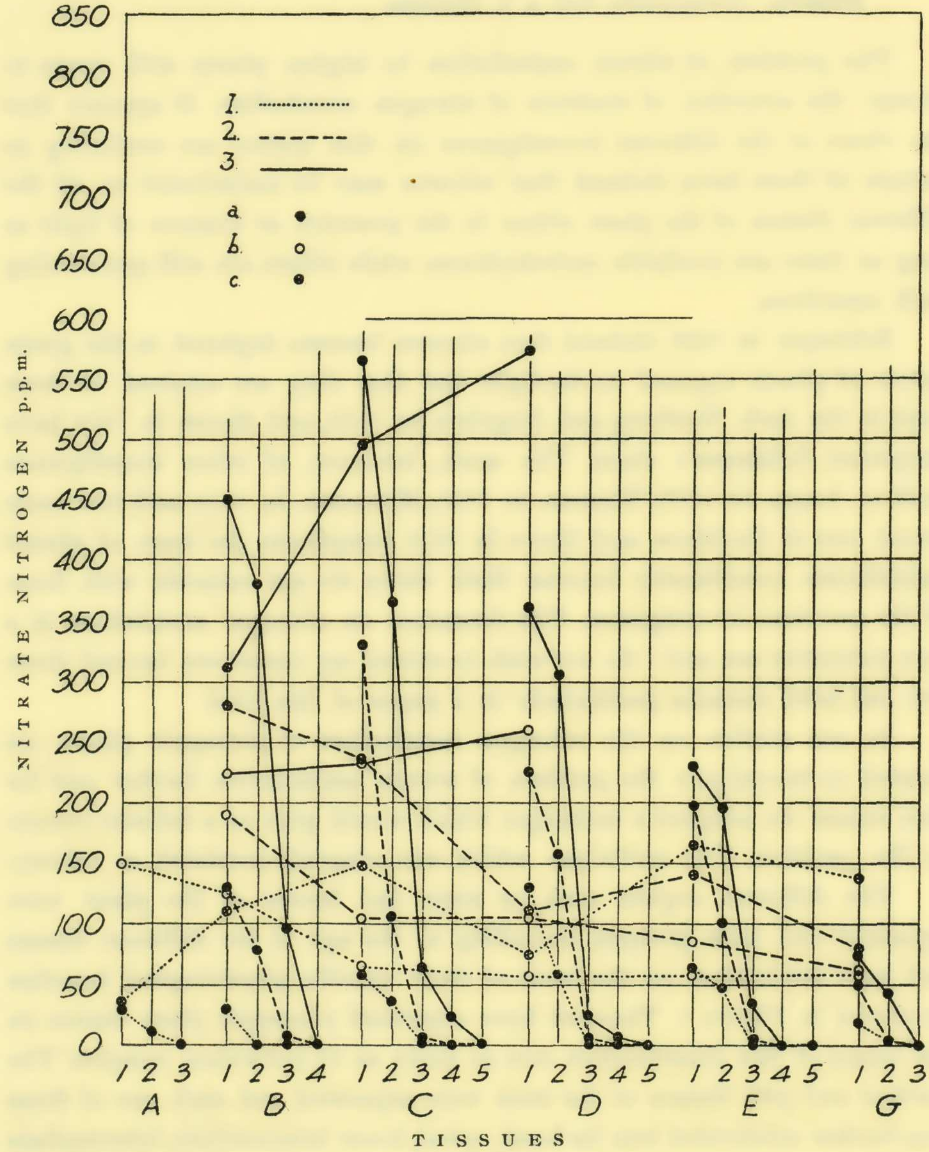
In our studies on the nitrogen metabolism of pineapple plants we wanted to investigate the problem of nitrate assimilation further and for that reason we adopted a technique which would give us a definite answer to the problem. The technique which was adopted consisted as follows:

The different organs such as stem and leaves of the plant were separated and were grouped according to the age of the different tissues and were subdivided on the basis of their specific physiological function as shown in Figure 1. Thus, we have separated pineapple plant tissues on the bassis of this classification into as many as 40 individual samples. The cortical and pith tissues of the stem were separated and each one of these was further subdivided into its basal, apical, lower intermediate, intermediate and upper intermediate tissues all of which are designated by the letters A, B, C, D, E etc. as in Figure 1. The leaves have been classified on the basis of the stem classification. Thus, those which were attached to the

* **ΧΡ. ΣΙΔΕΡΗ.— Μελέται ἐπὶ τῆς ἀφομοιώσεως τοῦ νιτρικοῦ ἀζώτου ὑπὸ τοῦ Ananas Sativus.**

stem tissue belonging to any one of these sections mentioned were grouped together and assigned the same symbol as the one possessed by the stem.

Graf 1.

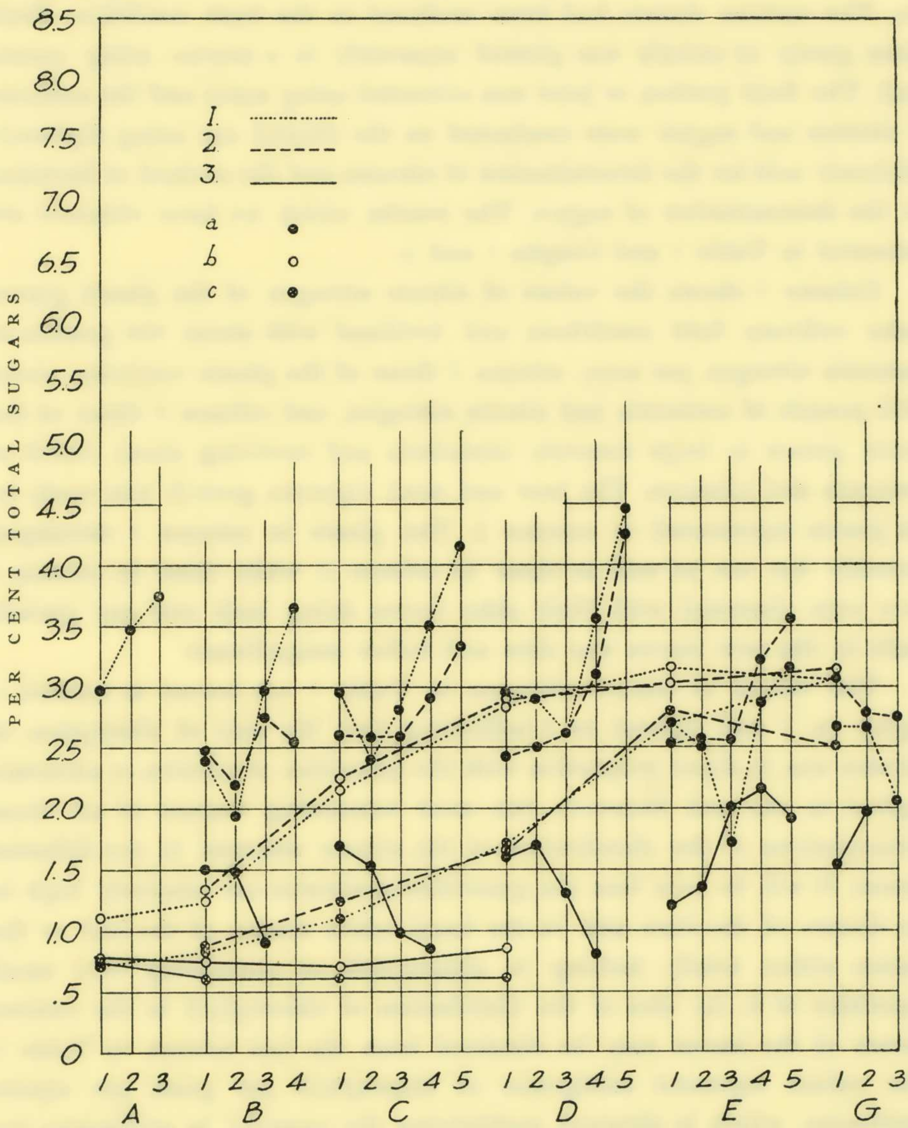


Leaf and stem groups and tissue sections.

Legend: 1.— 400 lbs N. per acre a.—Leaf tissues
2.— 2000 » » » » b.—Stem cortex
3.—20000 » » » » c.—Stem pith

The different tissues of each leaf were further separated into basal, lower intermediate, intermediate, upper intermediate and terminal tissue sections which are designated by arabic numerals as 1, 2, 3, 4 and 5.

Graf 2.



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Legend: 1.— 400 lbs N. per acre a.—Leaf tissues
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3.—20000 » » » » c.—Stem pith

The plants which were employed for these studies varied in age, some of them being very young and others mature plants. The quantities of nitrate nitrogen which had been applied also varied. Certain of the plants received very small while others very great quantities of nitrates or ammonia. The various tissues had been analyzed in the fresh condition. Each tissue group or sample was ground separately in a mortar using quartz sand. The fluid portion or juice was extracted using water and the analyses of nitrates and sugars were conducted on the cleared sap using diphenol-sulphonic acid for the determination of nitrates and the method of Bertrand for the determination of sugars. The results which we have obtained are presented in Table 1 and Graphs 1 and 2.

Column 1 shows the values of nitrate nitrogen of the plants grown under ordinary field conditions and fertilized with about 400 pounds of ammonia nitrogen per acre; column 2 those of the plants receiving about 2000 pounds of ammonia and nitrate nitrogen, and column 3 those of the plants grown in large concrete containers and receiving about 20,000 of ammonia and nitrogen. The best and most vigorous growth was made by the plants represented in colnmn 2. The plants in column 1 developed normally but not as well as those in column 2, while those in column 3 were very abnormal with their older leaves dying back and any growth made by the new leaves was slow and rather insignificant.

The values of nitrate nitrogen in Table 1 are lowest in column 1, higher in 2 and highest in 3 indicating that the rate of absorption of nitrates was in direct proportion with the quantities of nitrates or ammonia applied to the soil. However, the most interesting feature of all these investigations is the distribution of the nitrate nitrogen in the different tissues. It will be seen that the quantities of nitrates are relatively high in the tissues of the stem and in the basal white tissues of the leaf or the tissues either totally lacking in chlorophyll or containing very small quantities of it. An idea of the distribution of chlorophyll in the various tissues of the leaves may be obtained from the last column in Table 1. The values represent milligrams of chlorophyll per gram per square centimeter, which is obtained multiplying the quantity in milligrams per gram of tissues by that per square centimeter of leaf surface. The tissues with relatively great quantities of chlorophyll do not contain any nitrate nitrogen except those of the abnormal plants appearing in the third column where the chlorophyll has undergone considerable breaking down.

TABLE I

Tissues	Nitrates p. p. m. 1*	2	3	Total Sugars % 1	2	3	Chlorophyll mg. per gm.×cm.²
LEAVES							
A: 1	30.1	—	—	2.89	—	—	0
2	7.8	—	—	3.40	—	—	0.0800
3	0	—	—	3.76	—	—	0.0845
B: 1	30.1	130.2	450.4	2.41	2.51	1.50	0
2	0	77.6	382.0	1.90	2.19	1.49	0.0177
3	0	8.7	97.0	2.98	2.74	0.90	0.1115
4	0	0	dead	3.65	2.53	dead	0.1050
C: 1	58.4	333.0	564.0	2.96	2.61	1.67	0
2	38.2	105.0	364.0	2.40	2.60	1.53	0.0400
3	0	5.0	65.5	2.81	2.58	0.99	0.1670
4	0	0	24.3	3.51	2.92	0.84	0.1490
5	0	0	dead	4.14	3.32	dead	0.1180
D: 1	129.5	224.0	360.0	2.93	2.42	1.59	0
2	60.0	158.0	303.0	2.89	2.49	1.71	0.0237
3	0	4.6	26.1	2.60	2.60	1.30	0.0558
4	0	0	8 8	3.55	3.09	0.80	0.1015
5	0	0	0	4.47	4.26	dead	0.0864
E: 1	64.4	197.0	231.0	2.71	2.51	1.19	0
2	47.0	100.0	196.5	2.52	2.56	1.36	0.0137
3	0	4.8	35.8	1.71	2.56	2.00	0.0355
4	0	0	0	2.86	3.21	2.14	0.0702
5	0	0	0	3.15	3.56	1.90	—
PEDUNCLE							
G: 1	17.0	50.0	76.6	2.67	3.04	1.54	0
2	5.0	5.0	42.8	2.67	2.78	1.94	0.0166
3	0.	0	0	2.06	2.75	—	0.0412
STEM							
A: Cortex	150.0	—	—	1.09	—	—	0
Pith	36.8	—	—	0.73	—	—	0
B: Cortex	124.0	190.7	226.0	1.25	1.33	0.77	0
Pith	111.8	278.0	312.0	0.84	0.86	0.76	0
C: Cortex	63.2	102.8	236.5	2.15	2.24	0.70	0
Pith	149.6	237.0	493.5	1.10	1.22	0.62	0
D: Cortex	59.2	106.4	262.5	2.86	2.89	0.87	0
Pith	76.3	109.2	564.0	1.69	1.66	0.62	0
E: Cortex	58.4	83.8	—	3.14	3.02	—	0
Pith	167.5	138.5	—	2.68	2.81	—	0
PEDUNCLE							
G: Cortex	56.2	58.2	—	3.02	3.12	—	0
Pith	137.5	78.2	—	3.05	2.47	—	0

* 1=400 pounds nitrogen per acre. 2=2000 p. n. p. a. 3=20000 p. n. p. a.

Therefore, the presence of nitrates in chlorophyllous tissues may indicate either a pathological condition or one where the quantities of chlorophyll are not sufficiently high to assimilate quantities of nitrates proportionately greater than their capacity. That this latter condition is possible sometimes, one may look at Table 2 as an illustration as it contains certain results which were obtained on maize plants grown in nutrient solutions of different initial concentrations of iron, the different iron concentrations having influenced the production of different quantities of chlorophyll.

TABLE 2

Total plant weight, chlorophyll, total sugars and nitrate nitrogen values obtained on Zea mays plants grown in nutrient solutions at different initial concentrations of iron

Iron initial concentration p. p. m.	Growth* per plant weight gms.	Chlorophyll mg. per $cm.^2$	Sugars per cent	Nitrate Nitrogen p. p. m.
1.75	41.0	0.003	0.2	206
3.50	61.0	—	0.4	100
7.00	92.0	0.037	1.0	29
14.00	140.0	0.044	1.3	4
28.00	101.0	0.051	1.7	1

It will be seen in Table 2 that the concentration of nitrates in the leaf tissues of the maize plants is in inverse proportion with the quantities of chlorophyll and the initial iron concentrations, indicating that the assimilation of nitrates was more or less in proportion with the quantities of chlorophyll present.

CONCLUSIONS

The results of these studies indicate beyond any doubt that nitrates are assimilated in the chlorophyllous tissues of the pineapple plant. Nitrate assimilation by maize plants has been found to increase with greater quantities of chlorophyll in the tissues and vice versa.

The sugar content of tissues which do not take part in the assimilation of nitrates such as the stem tissues, plays no important part in the assimilation of nitrates within such tissues. The sugar content of the leaves, however, is influenced by the nitrate content of such leaves being higher with a low nitrate content and vice versa.

Further studies on the immediate by-products of nitrate assimilation are under way.

The writer wishes to express his very sincere thanks to his assistants

Miss B. H. Krauss and Mr. H. Y. Young for their very kind cooperation in these studies.

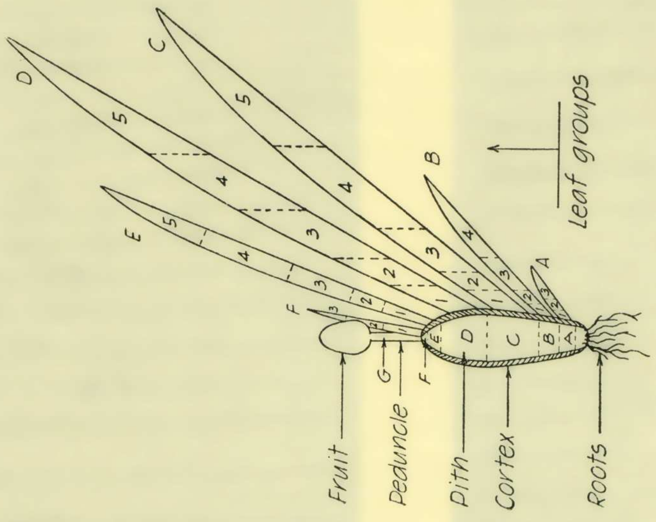

Fig. 1.—*Diagrammatic representation of the different plant tissues as reported in Table I.*

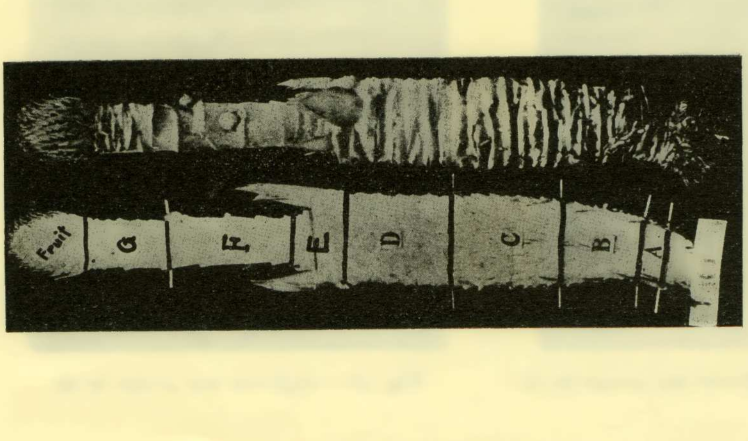

Fig. 2.—*Different Stem tissue groups.*

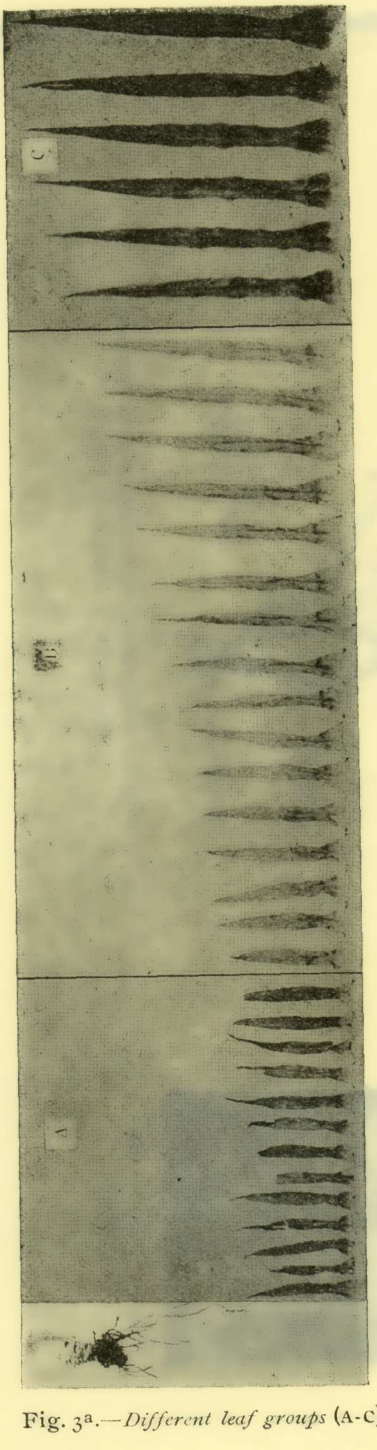

Fig. 3ª.—*Different leaf groups* (A-C).

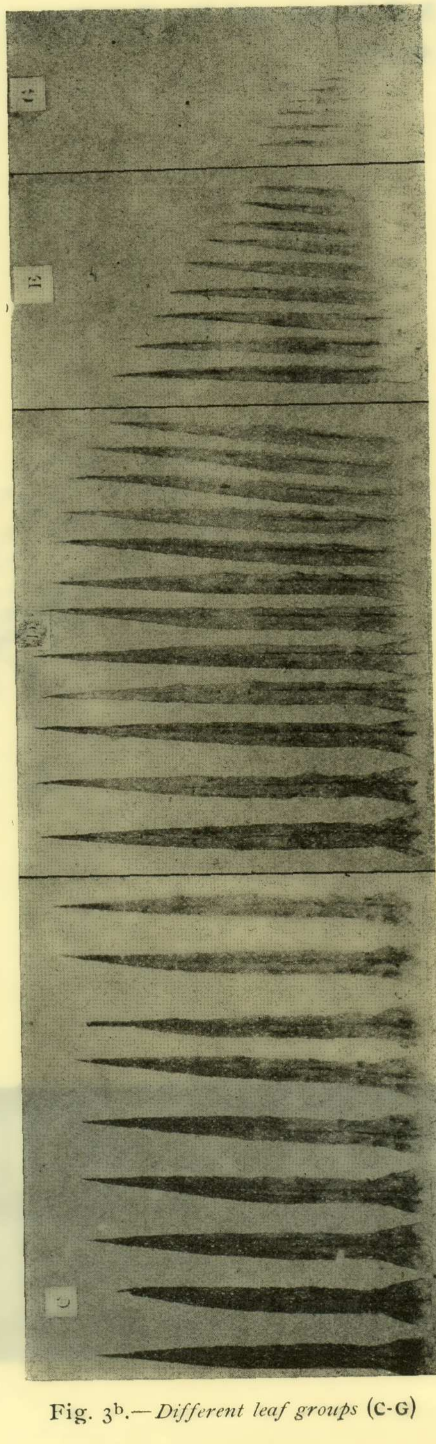

Fig. 3ᵇ.—*Different leaf groups* (C-G)

The entire leaf system of a pineapple plant.

Different leaf groups : size $^{47}/_{1000}$ natural. The different leaves show the extent to which their different tissues contain chlorophyll.

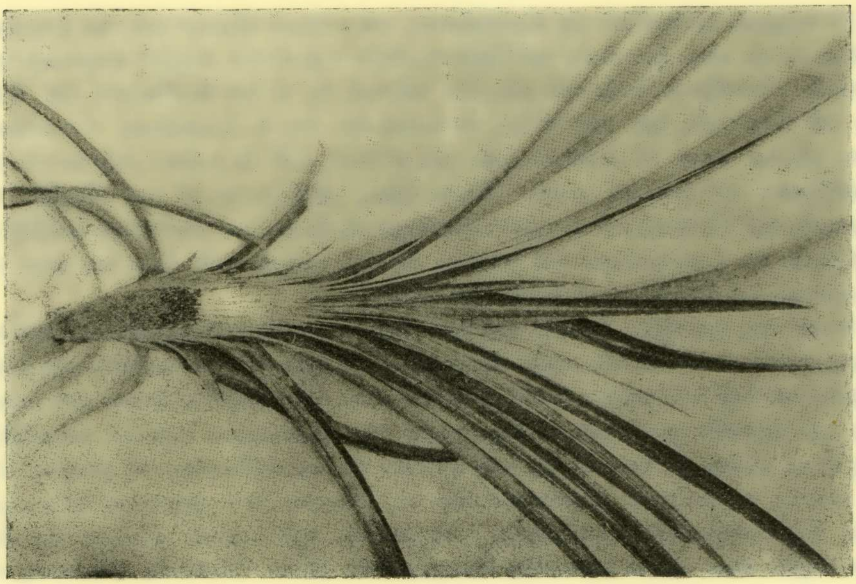

Fig. 4.—*A young plant of Ananas sativus.*

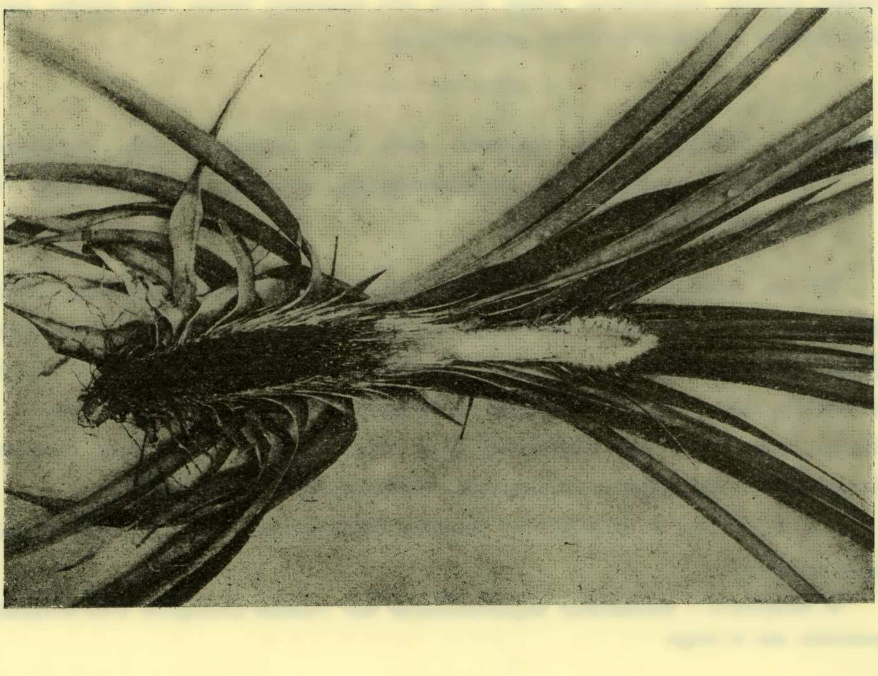

Fig. 5.—*A fruit bearing plant of Ananas sativus.*

ΠΕΡΙΛΗΨΙΣ

Αἱ διάφοροι μελέται ἐπὶ τῆς ἀφομοιώσεως τοῦ νιτρικοῦ ἀζώτου ὑπὸ τῶν ἀνωτέρων πρασίνων φυτῶν εἶναι ἡμιτελεῖς καὶ ἀβέβαιαι, διότι στεροῦνται ἀρκετὰ πειστικῶν πειραματικῶν ἀποδείξεων. Τινὲς τῶν μελετῶν τούτων, ὡς αἱ τοῦ Schimper, τοῦ Warburg καὶ Negelein καὶ τοῦ Moor ἀποφαίνονται ὅτι ἡ ἀφομοίωσις τοῦ νιτρικοῦ ἀζώτου γίνεται κατὰ τὸ πλεῖστον ὑπὸ τῶν φύλλων, καθ' ὃν χρόνον εὑρίσκονται ὑπὸ τὴν ἐπίδρασιν τοῦ ἡλιακοῦ φωτός. Ἄλλαι ὅμως μελέται, ὡς αἱ τοῦ Acqua, τῆς Eckerson, τοῦ Thomas καὶ αἱ τελευταίως δημοσιευθεῖσαι τοῦ Davidson καὶ Shive, ἰσχυρίζονται ὅτι ἡ ἀφομοίωσις τοῦ νιτρικοῦ ἀζώτου γίνεται καθ' ὅλας τὰς ὥρας τοῦ ἡμερονυκτίου καὶ ὑφ' ὅλων τῶν ἱστῶν τοῦ φυτοῦ.

Κατὰ τὰς ἡμετέρας ἐρεύνας ἐπὶ τοῦ *Ananas sativus* ἐκτελεσθείσας ὑπὸ τὰς κλιματολογικὰς συνθήκας τῶν νήσων Hawaii, τὸ νιτρικὸν ἄζωτον, κατόπιν τῆς ἀπορροφήσεώς του ὑπὸ τῶν ριζῶν, μεταφέρεται διὰ μέσου τῶν διαφόρων ἱστῶν τοῦ κορμοῦ μέχρι τῶν βασικῶν ἱστῶν τῶν φύλλων χωρὶς τὴν παραμικρὰν χημικὴν μεταβολήν. Μόλις ὅμως φθάσῃ εἰς τοὺς ἱστοὺς τῶν φύλλων οἵτινες ἀφθονοῦσιν εἰς χλωροφύλλην τότε ταχέως ἐμφανίζεται μεταβαλλόμενον εἰς διάφορόν τινα ἐνόργανον ἀζωτοῦχον οὐσίαν. Εἰς παθολογικὰς περιπτώσεις ἡ ἀφομοίωσις τοῦ νιτρικοῦ ἀζώτου δὲν εἶναι τόσον ταχεῖα ὅσον εἰς τὰ κανονικῶς λειτουργοῦντα φύλλα. Ὡσαύτως ἡ ποσότης τῆς χλωροφύλλης τῶν διαφόρων ἱστῶν τῶν φύλλων συντελεῖ κατὰ πολὺ εἰς τὴν βραδεῖαν ἢ ταχεῖαν ἀφομοίωσιν τοῦ νιτρικοῦ ἀζώτου.

Ὅθεν, ἐπὶ τῇ βάσει τῶν διαφόρων πειραματικῶν ἀποτελεσμάτων, τὰ ὁποῖα ἀναφέρονται ἀνωτέρω, δυνάμεθα νὰ εἴπωμεν μετὰ πεποιθήσεως ὅτι ἡ ἀφομοίωσις τοῦ νιτρικοῦ ἀζώτου συντελεῖται μόνον ἐντὸς τῶν πρασίνων ἱστῶν τῶν φυτῶν καὶ ὄχι ἐντὸς ἐκείνων οἵτινες στεροῦνται χλωροφύλλης.

ΙΑΤΡΙΚΗ. — Πειραματικαὶ ἔρευναι ἐπὶ τοῦ πιθήκου τῆς παθογόνου δράσεως ἀναεροβίου στρεπτότριχος, *ὑπὸ* ***Μ. Δ. Πετζετάκη***. Ἀνεκοινώθη ὑπὸ κ. Γερ. Φωκᾶ·

Ἐν τῇ συνεδρίᾳ τῆς 16 Δεκεμβρίου 1933 τῆς Ἰατρικῆς Ἑταιρείας Ἀθηνῶν, ἀνεκοινώσαμεν περίπτωσιν πνευμονικῆς καὶ πλευρικῆς μυκητιάσεως, καθ' ἣν παρετηρήσαμεν ἐν τοῖς πτυέλοις καὶ τῷ πλευριτικῷ ὑγρῷ μύκητα, οὗτινος ἡ ὀψιαιτέρα λεπτομερὴς ἔρευνα ἐν τῷ Ἑλληνικῷ Ἰνστιτούτῳ Pasteur, ἀπέδειξε τόσῳ ἐν τοῖς πτυέλοις ὅσῳ καὶ ἐν τῷ πλευριτικῷ ὑγρῷ τὴν ὕπαρξιν στρεπτότριχος ἀναπτυσσομένου μόνον εἰς ἀναεροβίους καλλιεργείας. (Cohnistreptothrix Pinoy).

Διὰ τοῦ οὕτως ἀπομονωθέντος μύκητος ἐξετελέσαμεν σειρὰν πειραματισμῶν ἐπὶ διαφόρων ζώων.

* **M. PETZETAKIS. — Recherches experimentales sur l'action pathogène d'un streptothrix anaérobie sur le singe.**