STUDIES ON THE HOST RANGE OF *PIRICULARIA ORYZAE* CAV. CAUSING BLAST DISEASE OF RICE

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A fungus resembling *Piricularia oryzae* Cav., the causal organism of blast disease of rice, has been recorded on several wild grasses occurring in rice fields and *bunds* as well as on species of Cyperaceae, Commelinacea, Musaceae, Zingiberaceae, and Cannaceae (Thirumalachar *et al.*, 1956; Meredith, 1962; Asuyama, 1963). Whether these hosts serve as collateral hosts for the pathogen on rice, enabling it to survive through the off-season when rice is not cultivated, is an important question. A considerable amount of attention has, therefore, been paid by workers to study their comparative morphology and their infectivity on rice. The literature on the subject has been critically reviewed by Padwick (1950) and more recently by Asuyama (1963); however, contradictory results have been obtained in cross-inoculation studies carried out by several workers. The present study was therefore undertaken to find out whether the grasses studied served as collateral hosts for the rice blast pathogen or not.

Isolates obtained from the grasses were compared with one another and with *Piricularia oryzae* occurring on rice, in respect of morphology, growth-characters, and pathogenicity on the different hosts. The results of the study are presented below.

Materials and Methods

Five isolates of *Piricularia* spp. were obtained from the following grasses:

1. *Panicum repens* Linn.
2. *Brachiaria mutica* Stapf.
3. *Leersia hexandra* Swartz.
4. *Digitaria sanguinalis* (Lim.) Scop.
5. *Digitaria setigera* Roth apud Roem *et* Schultt.

These isolates were compared with *Piricularia oryzae* Cav. from *Oryza sativa* L. (variety *Co. 13*).

The spore-morphology of the above-mentioned six fungi was studied with freshly-harvested spores from host-lesions. Two hundred conidia were measured in each instance.

The growth-characters and sporulation of the isolates were studied on oat meal agar with trace of thiamine and biotin, the cultures being maintained at 25° C. The amount of sporulation was described as 'Nil', if absent, sparse (+), fair (++), or profuse (+++), judged from the density of spores per uniform microscopic field of a spore-suspension prepared from identical amount of fungal matrix.

In addition to the study of the morphology, growth-characters of the isolates were observed on special media prepared with the extracts of leaves of the respective hosts. A quantity of twenty-five grams of fresh leaves of the individual grasses or rice was taken in 250 ml. of distilled water, and host extract was prepared and utilised in making a host-media with 2% agar.

For cross-inoculation studies, the different grasses were raised in pots along with the rice variety, *Co. 13*, in an enclosure and seven days before inoculation the entire aerial portions of the different hosts were cut off and the plants were placed under cover. The fresh foliage which emerged during the ensuing seven days were inoculated with spore-suspension, which was prepared by harvesting spores from 15 days old culture of the respective pathogens cultivated on oat meal agar with traces of thiamine and biotin, kept at 25° C. Appropriate uninoculated controls were kept in each case. Inoculations were carried out in the evening when the temperature was favourable, *i.e.*, below 26° C. The plants were kept under moist cloth curtain overnight. Observations on the infection which developed, if any, were recorded a week after. Only typical blast-lesions were taken to represent successful infection. Flecks and pinpoints of penetration were noticed in most cases but were not considered successful infection. Reisolation of the fungus was resorted to for confirming successful infection.

RESULTS

Morphological Studies

The isolates studied were of the same shape with typically obpyriform conidia, with 2 septa each, a characteristic hilum marking the place of detachment from the conidiophore; the hilum was occasionally eccentric in its disposition; the conidiophores were septate, pale olivaceous to brownish.

Though closely resembling one another in shape, distinct differences in spore size could be seen amongst the isolates. The fungus from *Leersia*

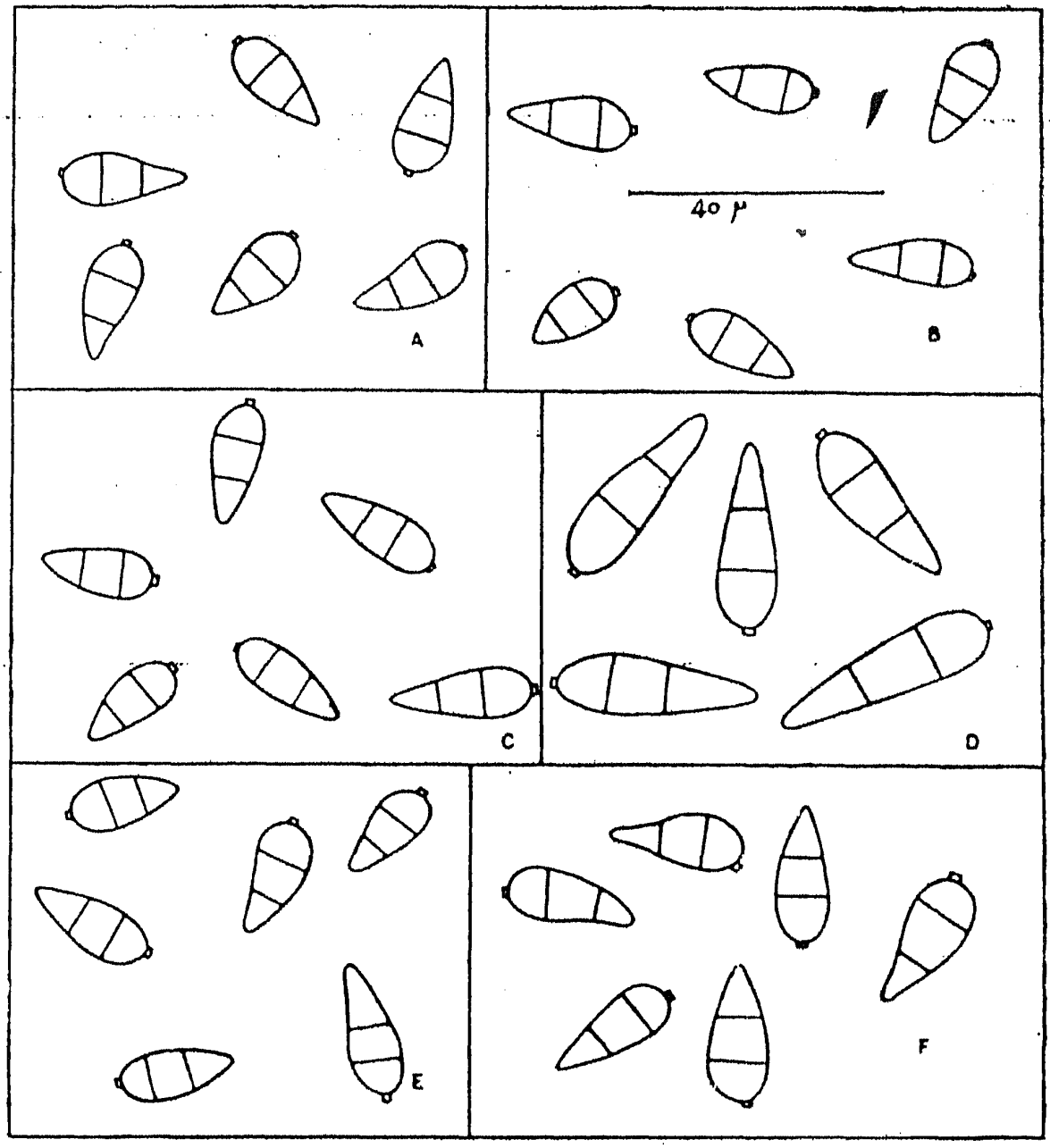

FIG. 1. Showing the morphology of conidia of the different isolates of *Piricularia* studied from host-lesions. A. *Piricularia oryzae*. B. *Piricularia grisea* f. sp. *brachiariae* comb. nov. C. *Piricularia grisea*. D. *Piricularia leersiae*. E. *Piricularia grisea* f. sp. *digitariae setigera*. F. *Piricularia grisea* f. sp. *panicae*.

hexandra had the longest spores being 25–35 μ in length, the spores from the isolates from *Brachiaria mutica* and *Oryza sativa* resembled each other closely in size, being 17–25 μ in length; while the spores of isolates from *Panicum repens*, *Digitaria sanguinalis* and *Digitaria setigera* fell into a third categor being 19–26 μ in length. The measurement of the conidia from host-lesion: are presented in Tables I A and I B.

TABLE I A

Showing conidial measurements of Piricularia *spp. from host-lesions, as described by original authors*

Sl. No.	Name of fungus	Name of host from which obtained	Dimensions in μ	Name and author	Year
1	*Piricularia grisea*	*Digitaria sanguinalis*	18× 9	Saccardo	1880
2	*Piricularia oryzae*	*Oryza sativa*	20–22×10–12	Cavara	1891
3	*Piricularia leersiae*	*Leersia oryzoides*	20–35× 7–10	Sawada	1917

TABLE I B

Showing conidial measurements of Piricularia *spp. from host-lesions obtained by present authors*

Sl. No.	Name of fungus	Name of host from which obtained	Dimensions in μ	Mean length	C.D. for comparing mean length except for No. 6
1	*Piricularia grisea*	*Digitaria sanguinalis*	19·2–25·6×8·0– 9·6	6·50	..
2	*Piricularia grisea* f.sp. *brachiariae*	*Brachiaria mutica*	17·6–25·6×8·0– 9·6	6·04	..
3	*Piricularia grisea* f. sp. *digitaria setigera*	*Digitaria setigera*	19·2–25·6×8·0– 9·6	7·13	0·14
4	*Piricularia grisea* f. sp. *panicae*	*Panicum repens* ..	19·2–25·6×9·6–11·2	6·82	..
5	*Piricularia oryzae*	*Oryza sativa* ..	17·6–24·0×8·0– 9·6	6·04	..
6	*Piricularia leersiae*	*Leersia hexandra* ..	25–35·2 ×8·0– 9·6	9·27	..

Cultural Studies

(*a*) *On oat meal agar with thiamine and biotin.*—The colour of the aerial mycelium, the amount of mycelium, the nature of growth (submerged or aerial) and sporulation varied from one isolate to the other (Table II). The variation observed amongst the five grass isolates was within the range of variability of these characters recorded for *Piricularia oryzae* from rice (Henry *et al.*, 1948).

TABLE II

Showing cultural characters of different isolates of Piricularia *spp. on oat meal agar with thiamine and biotin at* 25° *C.*

Sl. No.	Host isolate from	Colour of mycelium	Amount of aerial mycelium	Type growth	Sporulation (after 15 days of growth)
1	*Panicum repens*	Gray with white patches	++	Aerial and flat with fair amount of mycelium	Present
2	*Barchiaria mutica*	Blackish-white	+++	Fluffy and aerial	Present
3	*Leersia hexandra*	Whitish-brown	+	Little surface growth and aerial	Present
4	*Digitaria setigera*	Dark black	Nil	Submerged	Present
5	*Digitaria sanguinalis*	Brownish-black	Nil	Submerged	Present
6	*Oryza sativa*	Ash	+	Aerial	Present

+ Fair; ++ very fair; +++ abundant.

(*b*) *On host-media.*—The observations made on the radial growth, amount and colour of aerial mycelium of the different isolates on the host-media are presented in Table III.

The media prepared from *Oryza sativa*, followed by the one prepared from *Leersia hexandra*, were more favourable than the other host-media for radial growth of all the isolates studied except for the fungi from *Leersia*

hexandra and *Digitaria setigera*; in the case of these two isoates all the host-media were equally favourable for their radial growth. The medium prepared from the grass, *Digitaria setigera*, was found to be relatively better suited for the production of mycelium than other media. The colour of aerial mycelium was similar in the different host-media, but the isolate from *Leersia hexandra* became slightly darker when grown on media prepared with *Brachiaria mutica* or *Oryza sativa*.

TABLE III

Showing results of cross-inoculation on hosts with different isolates of Piricularia *spp.*

Name of host from which isolate was obtained	Name of hosts inoculated					
	Brachiaria mutica	*Leersia hexandra*	*Digitaria setigera*	*Digitaria sanguinalis*	*Panicum repens*	*Oryza sativa*
Brachiaria mutica	+	−	−	−	−	−
Leersia hexandra	−	+	−	−	−	−
Digitaria setigera	−	−	+	−	−	−
Digitaria sanguinalis	−	−	−	+	−	−
Panicum repens	−	−	−	−	+	−
Oryza sativa L. (Co. 13)	−	−	−	−	−	+

+, Typical lesions with spores. −, No typical lesions with spores.

No significant differences were observed with respect to sporulation of the different isolates studied on the various host-media.

(*c*) *Cross-inoculation studies on hosts.*—The results obtained from cross-inoculations made with the isolates on the different hosts are presented in Table IV.

The cross-inoculations made during two crop seasons clearly indicated that the pathogen, *Piricularia oryzae*, from rice did not cross over to any of the grasses studied; nor did the isolates of *Piricularia* from the grasses studied

TABLE IV

Showing rate of radial growth cultural and physiological characters of different isolates of Piricularia *spp. on host media at 25° C.*

Sl. No.	Name of host from which medium was obtained	Name of host from which isolate was obtained	Rate of radial growth in cm./day	Type of growth	Amount of aerial mycelium	Colour of mycelium	Sporulation after 14 days growth
1	*Panicum repens*	*Panicum repens*	0·46	Flat	+	Gray-white	+
		B. mutica	0·46	Little aerial mycelium little	++	White	+
		L. hexandra	0·59	Aerial mycelium	+	White	+
		D. setigera	0·45	Submerged	Nil	Dark	—
		O. sativa	—	—	—	—	—
2	*Brachiaria mutica*	*Panicum repens*	0·44	Flat aerial	+	Grey with white patches	Nil
		B. mutica	0·35	Flocculent and aerial	++	White	Nil
		L. hexandra	0·36	Little surface growth and aerial	+	Whitish-brown	Nil
		D. setigera	0·52	Submerged	Nil	Dark	Nil
		O. sativa	0·40	Fluffy and aerial	++	Grey	Nil
3	*Leersia hexandra*	*P. repens*	—	—	—	—	—
		B. mutica	0·56	—	—	Grey-white	+++
		L. hexandra	—	—	—	—	—
		D. setigera	0·69	—	—	Dark	—
		O. sativa	0·55	—	—	Ashy grey	+

TABLE IV (*Contd.*)

Sl. No.	Name of host from which medium was obtained	Name of host from which isolate was obtained	Rate of radial growth in cm./day	Type of growth	Amount of aerial mycelium	Colour of mycelium	Sporulation after 14 days growth
4	*Digitaria setigera*	*P. repens*	—	Little fluffy at central and aerial	++	White	+
		B. mutica	0·47	Fluffy and flocculent and aerial	+++	White	+++
		L. hexandra	0·48	Flat and aerial	+	White	++
		D. setigera	0·50	Flat and aerial	+	Whitish-grey	Nil
		O. sativa	—	—	—	—	—
5	*Oryza satica*	*P. repens*	0·56	Flat and aerial	+	Grey-white	+
		B. mutica	0·54	Fluffy and aerial	++	White	—
		L. hexandra	0·68	Flat with little aerial mycelium	+	Grey-white	—
		D. setigera	0·63	Submerged	Nil	Dark	—
		O. sativa	0·56	Flat and aerial	+	Grey	—

Amount of mycelium

— Information not available.
\+ Sparse.
++ Fair.
+++ Profuse.

Sporulation

— Information not available.
\+ Sparse.
++ Fair.
+++ Profuse.

cross over to rice. The pathogen isolated from any particular host was pathogenic on its own host only.

Discussions

Successful infection of other species of Gramineae with the isolate from rice has been reported by Thomas (1940), Kawamura (1941), De Guitterrez (1954), Latterell *et al.* (1960) and several others; infection of rice with isolates from grasses has been reported by Kawakami (1902), Ramakrishnan (1948), Kuribayashi (1953), ICAR Report (1954), Srinivasan and Vijayalakshmi (1958), Krishnaswamy *et al.* (1959), Piagiatgong (1961); while a few workers like Sawada (1917), Mc rae (1922), Mehta *et al.* (1953), Thirumalachar *et al.* (1956), Govinda Rao and Koteswar Rao (1958) and Wilson (1962) have reported that they could not establish cross-over in either direction with the isolates studied by them.

Lastly, it may be pointed out that reports of successful cross-infection of some isolates from some specific grasses to rice or *vice versa* have been contradicted by later workers.

The data presented in this contribution indicate that the isolates of *Piricularia* from the different graminaceous hosts studied are independent entities pathogenically. There were also some differences in respect of spore-size especially in the isolate from *Leersia hexandra.* The range of variability in other cultural characters like type of growth and amount of aerial mycelium was within the range for such characters of *Piricularia oryzae* from rice.

A possible explanation of successful cross-over reported hitherto may lie in the fact that pinpoints of penetration seen as flecks might have been considered as a positive infection, whereas in the present study the development of typical lesions was the criterion adopted to assess successful infection. If the possibility of natural infection could be ruled out in such studies, the presence of a virulent biotype of the pathogen which under certain conditions might successfully cross over to other hosts could also be considered. A sexual cycle in grass-hosts may give rise to such biotypes. Based on the present evidence it is very necessary to clarify further the position regarding the identity and nomenclature of the grass isolates studied.

Piricularia grisea on *Digitaria sanguinalis* is the type species of the genus (Saccardo, 1880). *Piricularia oryzae* was subsequently described on rice by Cavara (1891). The two fungi resemble closely each other morphologically and they have been considered to be synonymous by workers who had succeeded in cross-inoculation studies with the two isolates. Recently,

Latterell *et al.* (1960) had suggested that the pathogen on rice be named as *Piricularia grisea* f. sp. *oryzae* in view of the fact that the rice pathogen resembled closely *Piricularia grisea* as originally described but differred from it in its pathogenicity.

Asuyama (1963), however, has suggested the use of the name *Piricularia oryzae* for the pathogen from rice, due to its long usage and familiarity, although strictly speaking it should be deemed only as a *forma specialis* of the species, *Piricularia grisea.* The present authors agree with the view expressed by Asuyama (1963).

The valid criteria for the erection of a species in a genus like *Piricularia* would be the morphological differences and differences in host-range. A varietal name may accommodate fungi having morphological differences alone, having common host-range and the erection of *formae specialis* is justifiable for those biological entities which are morphologically alike but differ in their pathogenicity.

Based upon the above criteria, the isolates studied may be designated as follows:

1. *Piricularia oryzae* Cav. on *Oryza sativa* L.
2. *Piricularia leersiae* (Sawada) Ito on *Leersia hexandra* Swartz.
3. *Piricularia grisea* (Cke) Sacc. *forma specialis brachiariae* f. sp. comb. nov. on *Brachiaria mutica* Stapf.
4. *Piricularia grisea* (Cke) Sacc. *forma specialis digitaria setigera* f. sp. nov. *Digitaria setigera* Roth apud Roem *et* Schultt.
5. *Piricularia grisea* (Cke) Sacc. *forma specialis panicae* f. sp. nov. *Panicum repens* Linn.

SUMMARY

The grasses *Panicum repens, Brachiaria mutica, Leersia hexandra, Digitaria sanguinalis* or *Digitaria setigera* do not seem to serve as collateral hosts for *Piricularia oryzae* Cav. pathogen of blast disease of rice. The pathogens studied exhibited distinct host specificity.

The isolates of *Piricularia* on *Panicum repens, Brachiaria mutica* and *Digitaria setigera* have been identified as *formae specialis* of *Piricularia grisea* (Cke) Saccardo. The accommodation of the isolates of *Piricularia* on *Digitaria sanguinalis, Oryza sativa* and *Leersia hexandra* as *Piricularia grisea,* and *Piricularia oryzae* and as *Piricularia leersiae* respectively, is accepted.

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REFERENCES

1. Anonymous .. *Report on Agricultural Research, I.C.A.R., for* 1952–53, New Delhi, 1954, pp. 2–45.

2. Asuyama, H. .. "Morphology, taxonomy, host range and life-cycle of *Piricularia oryzae*," *Symp. on Rice Blast Disease*, Paper B, I.R.R.I., The Philippines, 1963.

3. De Guitterez, L. H. .. "Presence of *Piricularia oryzae* on sugarcane in Costa rica," *Pl. Dis. Rept.*, 1954, 38 (9), 695.

*4. Fulton, H. R. .. "Diseases affecting rice in Louisiana," *Bull. La. agric. exp. Stn.*, 1908, p. 105.

5. Govinda Rao, P. and Koteswar Rao, D. "A new record of *Piricularia* from India," *Indian Phytopath.*, 1958, 10 (2), 107.

6. Henry, B. W. and Anderson, A. L. "Sporulation of *Piricularia oryzae*," *Phytopathology*, 1948, 38 (4), 265–78.

*7. Kawakami, T. .. "On the blast disease of rice plant," *J. Sapporo agr. Soc.*, 1901, 2, 1–47; 1902, 3, 1–3.

*8. Kawamura, E. .. "Reaction of certain species of the Genus *Oryza* to the infection of *Piricularia oryzae*," *Bull. Sci. Fac. terk. Kyushu Univ.*, 1940.

9. Krishnaswamy, C. S., Govindaswamy, C. V. and Adyanthayya, N. R. "Blast and foot root diseases of rice in Madras State," *Rep. I.C.A.R.*, July 1959.

*10. Kuribayashi, K., Ichikawa, H. and Terazawa, M. "Study of physiological specialization in rice blast fungus," *Report for the years* 1951–62, *Nagano. Agr. Expt. Stn.*, 1953, pp. 31.

11. Latterell, F. M., Tullis, E. C. and Collier, J. W. "Physiologic races of *Piricularia oryzae*," *Pl. Dis. Rep.*, 1960, 44, 679–83.

*12. Mc rae, W. .. *Report of the Imperial Mycologist, Agric. Res. Inst.*, 1921–22 (*RAM*, 2, 258–60).

13. Mehta, P. R., Singh, B. and Mathur, S. C. "A new leaf-spot disease of bajra (*Pennisetum typhoides* Stapf and Hubb.) caused by a species of *Piricularia*," *Indian Phytopath.*, 1953, 5, 140–43.

14. Meredith, D. S. .. "*Piricularia musae* Hughes in Jamaica," *Trans. Brit. Myc. Soc.*, 1962, 45 (1), 137–42.

*15. Nishikado, Y. .. "Studies on the rice blast disease," *Jap. J. Bot.*, 1927, 3, 239–44.

*16. Ono, K. *et al.* .. "Studies on rice blast disease," *Rep. Res. Pl. for Dis. Hokuriku Agr. Exp. Stn. for* 1959–60, 1960–61.

17. Padwick, G. W. .. *Manual of Rice Diseases*, CAB, Kew, 1950.

18. Piagiatgong .. "Studies on blast disease of rice and grasses," *Madras agric. J.*, 1961, **48**, 25.

19. Ramakrishnan, K. V. .. "Studies on the morphology, physiology and parasitism of the genus *Piricularia* in Madras," *Proc. Ind. Acad. Sci.*, 1948, **27**, B (6) 174–93.

*20. Sawada, K. .. "Blast of rice plant and its relation to infective crops and weeds, with description of five species of *Dactylaria*," *Sp. Bull. Agr. exp. Stn. Govt. Farm*, 1917, **16**, 78.

21. Srinivasan, K. V. and Vijayalakshmi, U. "*Piricularia oryzae* Cav. on *Arumdo donax*," *Sci. and Cult.*, 1958, **23**, 490–91.

22. Thirumalachar, M. J., Kulkarni, N. B. and Patel, M. K. "The new records of *Piricularia* species from India," *Indian Phytopath.*, 1956, **9** (5), 48–51.

*23. Thomas, K. M. .. *Detailed Administration Report of the Government Mycologist, Madras, for the Years* 1939–40, 1940–41.

*24. ——— and Krishnaswamy, C. S. "The control of chief diseases of rice as a means of increasing rice production," *Madras agric. J.*, 1948, **34–35**, 1–3.

25. Wilson, K. I. .. "New record of *Piricularia* on *Rhyncheletrum repens* (Wild) Hubbard," *Indian Phytopath.*, 1962, **15**, 76–77.

*Originals not seen.