

High sporulating media for *Piricularia oryzae* Cavara.

Hoo Sup CHUNG and Yong Joon LA

稻熱病菌 分生孢子形成을 促進하는 培地

鄭 厚 燮^①· 羅 瑒 俊^②

SUMMARY

In an attempt to find a satisfactory medium which would facilitate abundant conidial production of *Piricularia oryzae*, agar and liquid media containing various nutrients were tested as to the efficiency of conidial production.

Among the media tested most number of conidia occurred on V-8 juice broth and tomato juice agar and the least on rice straw medium. Thus in conidial productivity, V-8 juice broth and tomato juice agar far exceeded the rice polish and yeast starch agar which were known to be best by previous workers.

INTRODUCTION

During the course of research on the pathogenicity of the blast disease of rice caused by *Piricularia oryzae*, it was necessary to obtain conidia in quantity for use as inoculum.

Henry et al (2) and Hirata (3) reported several media for the production of *P. oryzae* among which the best was rice-polish agar. Yeast starch agar produced most conidia among the media tested by Satomi et al (5) and Ahn and Chung (1). Although these various media reported by previous investigators produced conidia of *P. oryzae* they are nevertheless not satisfactory for the production of abundant conidia which can be used for the large scale artificial inoculation.

La (4) reported that *Cercospora beticola*, which was known to be poor sporulating fungus on various media, sporulated abundantly on both of the agar and liquid medium of V-8 juice and

tomato juice.

Thus in an attempt to find a satisfactory medium which would facilitate abundant spore production of *P. oryzae*, agar and liquid media of V-8 juice and tomato juice along with those previously reported media were tested as to the efficiency of conidial production of *P. oryzae*.

MATERIALS AND METHODS

Conidia of *P. oryzae* were isolated from the diseased leaves collected at the College Farm, Suwon and grown on potato dextrose agar in petri plates. Agar and liquid media were prepared according to the formula given below which are based on liter quantities. Difco-Bacto agar was added to each of the solid media in the amount of 17g per liter and no agar was added to liquid media.

1. Rice polish medium
Rice polish 50g.
Sucrose 20g.
2. Rice straw medium
Rice straw 50g.
Sucrose 20g.
3. Potato sucrose medium
Potato 200g.
Sucrose 20g.
4. Yeast starch medium
Yeast extract 2g.
Soluble starch 10g.
5. Tomato juice medium
Tomato juice 300ml.
6. V-8 juice medium
V-8 juice 300ml.

The media were sterilized in an autoclave at 15 lbs pressure for 20 minutes and the pH of each medium was adjusted to 5.5-6.0 by addition of 50% lactic acid or calcium carbonate. Agar

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College of Agriculture,
Seoul National University

medium was placed in each petri plate in the amount of 20 ml and liquid medium in the amount of 50 ml in 125-ml Erlenmeyer flasks.

On agar media 2 ml of conidial suspension prepared from 14 days old culture of *P. oryzae* was spread evenly throughout the entire surface of the plates and then incubated for 7 days at room temperature. A piece of cultural mat (5 mm in diam) cut by biscuit cutter was placed in a concave watch glass and diluted with 2 ml of water. Spore counts were made by flooding the haemocytometer with the conidial suspension.

On liquid media, a piece of cultural mat (5 mm in diam) from 7 days old culture on potato dextrose agar was placed and incubated at room temperature. After 20 days of growth, 50 ml of sterile distilled water was added to each flask and the fungal mat grinded for one minute in Waring Blendor. Spore counts were made by taking the spore suspension and flooding the haemocytometer. In addition, air dry weight of the fungal mat was measured after spore counts.

RESULTS AND CONCLUSION

1. Conidial production on agar media.

On agar media, most abundant conidia occurred on tomato juice agar and the least on rice straw agar. Yeast starch agar which was known to be best for conidial production of *P. oryzae* by previous workers were not as efficient as tomato juice, and rice polish agar produced far less number of conidia than tomato juice agar. Moderate sporulation resulted on V-8 juice agar.

The results are summarized in Table 1.

Table 1. Number of conidia of *P. oryzae* produced per unit surface area on 6 different agar media.

Media	Number of conidia
Rice straw agar	8 ^a
Rice polish agar	16
Potato sucrose agar	27
Yeast starch agar	65
Tomato juice agar	96
V-8 juice agar	42

^aEach datum represents the average of six 9-mm² haemocytometer counts made on the spore suspension obtained from each of three plates.

2. Conidial production on liquid media.

V-8 juice broth produced most abundant conidia among the media used whereas the rice straw broth yielded the least as was in the agar medium. Moderate sporulation occurred on rest of the media except potato sucrose broth.

The luxuriant development of mycelium was not always accompanied by the abundant production of conidia as it can be seen from the potato sucrose agar which produced considerable amount of mycelium but yielded small amount of conidia. Table 2 summarized the results of this experiment.

Table 2. Number of conidia of *P. oryzae* and air dry weight of mycelium produced on six different liquid medium after 20 days of incubation at room temperature

Media	Number of conidia	Air dry weight(mg)
Rice straw broth	8 ^a	74 ^b
Rice polish broth	24	110
Potato sucrose broth	12	214
Yeast starch broth	18	63
Tomato juice broth	23	230
V-8 juice broth	100	185

a. Each datum represents the average of six 9-mm² haemocytometer counts made on the suspension obtained from each of three 125 ml Erlenmeyer flask cultures.

b. Average of three 125 ml Erlenmeyer flask cultures.

It was also noticed that the amount of sporulation on agar media was differed from the liquid medium in a few cases in the experiment. The results obtained in this experiment indicate that V-8 juice broth and tomato agar are highly efficient in producing conidia of *P. oryzae* and it is recommended to use V-8 juice or and tomato juice medium in the laboratory practice for obtaining abundant conidia of *P. oryzae*.

LITERATURE CITED

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要 約

稻熱病菌의 分生胞子를 多量으로 形成하는 培地를 찾고자 여러가지 榮養分을 가진 寒天 및 液體培地에 대하여 分生胞子形成度を 試驗하였다.

여러가지 供試 培地 中에서 V-8培養液과 토

마트汁寒天培地 上에서 가장 많은 分生胞子が 形成되었고, 벗질培地 上에서 가장 적은 數의 分生胞子が 形成되었다. 이와 같이 V-8汁培養液과 토마토寒天培地는 稻熱病菌 分生胞子形成度에 있어서 過去 研究者들이 最適이라고 報告한 쌀겨寒天培地와 酵母澱粉寒天培地를 훨씬 능가하였다.

—抄 錄—

STRONG, R. G. AND D.E. SBUR 1960. Influence of Grain Moisture and Storage Temperature on the Effectiveness of Malathion as a Grain Protectant. (穀物濕度和 貯藏溫度가 防蟲劑로서 使用되는 Malathion의 效果에 미치는 影響)

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穀物の 濕度を 10%, 12%, 14%, 16%, 18%, 20%로 하고 60°F에 貯藏한 小麥에 Malathion 10 p.p.m.을 使用하였을 때 濕度の 影響은 穀物の 濕도가 增加함에 따라 그라나리바구미 (*Sitophilus granarius*), 쌀바구미 (*Sitophilus oryzae*) 및 *Tribolium confusum*의 殺蟲率이 減少하였다. Malathion을 小麥에 가장 有效하게 持續시키는 面에서 生覺한다면 12%의 穀物濕도가 가장 安全한 有效水準이 있고 臨界水準은 14%에서 나타났다. 10 p.p.m.의 藥量을 10%의 穀物濕도와 60°, 70°, 80°, 90°, 100°, 120°F의 濕도에 貯藏한 小麥에 對한 Malathion의 效果는 溫度가 增加함에 따라 減少하였다. 그러나 Malathion이 持續的으로 有效하게 效果를 나타낼수 있는 安全한 有效溫度는 찾아 낼 수 없었다. 穀物濕度, 貯藏溫度 및 使用한 藥量間에 關한 實驗結果는 穀物の 實際 貯藏에서 볼수 있는 穀物濕도와 貯藏溫도의 逆效果를 맞추기 위해서는 malathion의 使用量을 調節함이 重要하다고 表示하였다.

(編 輯 委)

—文 獻 紹 介—

明日山 秀文·向 秀夫·鈴木直治 編集

1962, 植物病理實驗法, 日本植物防疫協會, 843頁.

植物病理學이 植物學者 특히 菌學者에 의해서 開拓되어 오늘날은 醫學과 함께 典型的인 綜合科學으로 불리워지고 있다. 따라서 그 論及範圍도 매우 넓으며 나날이 발전되는 새로운 지식과 기술이 요구됨에 비추어, 이에 따르는 植物病理學의 實驗指導書를 苦待하였던 것이다.

이 책은 明日山秀文教授를 비롯한 20人的 專門家들이 專攻別로 分擔하여 실험에 필요한 사항을 記述하려고 노력한 것 같다. 全編을 19章으로 나누었는데, 우선 病理學에 필요한 實驗器具와 施設의 해설, 현미경취급법, 植物病害의 診斷法, 病害標本만드는 법, 병원균의 分離 및 培養技術, 환경조건의 測定 및 그 調節法, 殺菌劑의 効力檢定方法, 被害査定法, 防除試驗, 實驗記錄과 그 整理法 등을 설명하고 있다. 그 외에 植物病의 生態, 病態解剖, 病態生理, 病原菌의 生理, 植物病原菌의 代謝生産毒素에 關한 實驗法, 血清反應, 바이러스實驗法, 電子顯微鏡技術 등도 提及하고 있다.

이와 같이 실험에 필요한 모든 事項을 취급한 결과, 實驗方法이라기 보다는 現象의 설명에 그친 點도 적지않으나, 이 책 한편으로 植物病理學에 關한 全般的인 것을 알 수 있으므로 매우 有用한 책이라고 생각된다.

(編 輯 委)