A STUDY OF WATERMELON RUST

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A new disease, the rust of watermelon, first observed in 1949, seems to be endemic to the region and as yet confined therein. Although the fungus *P. citrulli* is specifically identical with the one occurring elsewhere on coloycnthis (*C. colocynthis*), it is considered to be a different variety and named *P. citrulli* variety *valgari*. The pathogen has been seen to enter both through the stomata and through the cuticle. The teleutospore have been found to need a long resting period and to germinate only if subjected to freezing and thawing for a period of 45-60 days. So far the method of perennation remains unsolved. The possibilitiy of an alternate host being responsible is being investigated.

Introduction

The rust disease of wetermelons was first observed at Karachi in the course of a routine survey of plant diseases in the outlying region of Malir.[1] The disease was severe enough in that year as well as in 1950, to materially decrease the yield as well as the quality of watermelons in the affected areas. In fact, some of the fields were so heavily affected in 1950 that the owners were advised to burn up the affected crop. At the same time, a vigorous compaign of dusting and spraying was undertaken and the disease brought under control. However, there have been occurring since then outbreaks of this rust in these same areas of Malir and Landhi, off and on, in various years.

Curiously enough, even though the crop of watermelon is sown all over the Sind and Punjab to the north, and in the Rajisthan in India to the east, there have been no report of the occurrence of watermelon rust in any of these areas. Indeed as far as is known the disease is endemic to Karachi, even though the allied plant *Citrullus colocynthis* was reported some fifty years ago as being parasitised by a rust named *Puccinia citrulli* by Butler[2] as far down as Madras in the Indian peninsula.

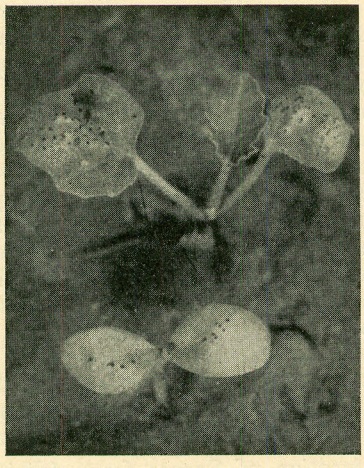

Plate 1.— Formation of uredial sori on the leaves of colocynthis (*Citrullus colocynthis*).

In view of the importance of the crop, an intensive investigation of the disease was undertaken in 1957-58 to determine the variety of the parasite, its mode of infection, perennation and multiplication, collateral host, if any, and to isolate any variety of watermelon that may be resistant.

Uredospores of the fungus were taken from the natural local host, *Citrullus vulgaris* Linn. and inoculated on *C. colocynthis Schrad.* using the technique detailed elsewhere.

The inoculated plants of *C. colocynthis* after the usual period of incubation produced sori (Plates 1 and 2) which were similar to those on *C. vulgaris*.

These experiments were repeated four times successfully. The results, summarised in Table 1 below, definitely indicate that the rust observed to occur at Karachi on *C. vulgaris* is specifically the same as the one reported by Butler from Madras occurring on *C. colocynthis*, namely *Puccinia citrulli*.

However in spite of careful search, no colocynthis plants have been found infected in nature with this rust, even on plants growing in the fields of watermelon heavily rusted. It seems reasonable to conclude that while in artificial inoculations, rust on watermelon can be successfully transferred on to the colocynthis plant, the same is not the case in nature.

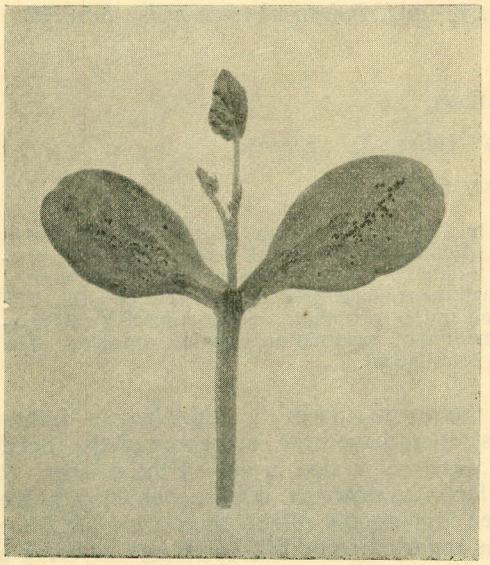

Plate 2.—Formation of uredial sori on the leaves of watermelon (*Citrullus vulgaris*).

TABLE 1.—SIZE OF UREDOSPORE IN MICRONS, OF *P. Citrulli* ON THE HOST.

On *C. vulgaris* collected from Malir	On *C. vulgaris* inoculated in Labs.	On *colocynthis* inoculated in. Labs	On *C. colocynthis* as reported by Butler
Length :			
23.10 - 29.70	23.10 - 29.70	22.38 - 29.52	20 - 30
Breadth :			
19.80-24.75	19.80 - 24.75	19.98 - 24.08	18 - 24

This conclusion is strengthened by the fact that areas where the rust has been reported on colocynthis[2] have not reported it on watermelons.

In spite of specific identity therefore it is reasonable to regard the rust on watermelons as belonging to a variety distinct from that occurring on colocynthis. For this former, which is the subject of present investigation, the name *Puccinia citrulli* var. *vulgari* (var. novo) is proposed to distinguish it from *Puccinia citrulli var. colocynthi*.

Perennation of the Fungus

The following could be the possible sources of perennation through the uredial stages:

1. Volunteer plants of watermelon.
2. Collateral hosts.
3. Old plant debris.
4. Migration of spores from the adjoining areas.
5. Some alternate host producing aecia and aecial spores.

1. *Volunteer Plants.*—The possible perennation of the fungus in the field through volunteer plants has been studied during the years 1956-57, 1957-58 and 1958-59. Off-season plants of watermelon were kept under weekly observation in the field previously infected by the fungus but no case of infection could be detected on these plants till the main crop had come into its own from April to June. Only rarely the infection was carried up to the first week of July, even though there is an abundance of watermelon plants both before and after this period. Volunteer plants therefore do not seem to play any significant part in the perennation as far as our present observation goes.

2. *Collateral Host.—Citrullus colocynthis* which is seen growing through out the year has also been kept under constant observation month by month but as described elsewhere it has never been found attacked by this fungus in the field, even when growing in close proximity of watermelon plants and in those same fields where there is year after year fungus infection in the month of April, May, and June. It is only under controlled conditions that *C. colocynthis* has been found to act as a collateral host as described above. Colocynthis therefore does not seems to act as collateral host for this rust in nature.

3. *Old Plants Debris.*—In late June and July after the majority of the fruits have matured and before the crop dries out completely, some of it is fed to the cattle, and the rest usually just left over in field. During this season the temperature often shoots up from 112°F. to 118°F. in the shade and the plants soon dry up. The dried up debris is then ploughed in the field for manure. Thus there is not much plant material left over, which could act as source of infection. However some of the rusted plant debris was kept under room temperature in the Laboratories, and the spores were taken therefrom week after week and studied for their viability. It was noted that material which was collected on 2nd June had the viability of 75% of spores when tested the same day, but after two days only 10% spores were viable. After ten days when the material was tested on 12th June, 1959 there were no viable spores. It thus seems that plant debris either could not serve as source of uredo spore infection from one season to the other.

4. *Migration of Spores from Adjoining Areas.*—In areas adjoining Karachi, i.e. Baluchistan and Sind, watermelon crop has two seasons of cultivation: (a) March to June and (b) August to

October. In none of these areas however the disease has ever been reported either on the watermelon or on colocynthis. In fact from all evidence so far available the fungus seems to be endemic to Karachi on the watermelon only; thus there appears no outstanding source for the spores or spore material to be brought to Karachi to start the infection. Further, the prevailing direction of wind during this season is south westerly, i.e. from the sea, thus the wind blows from Karachi to Sind rather than in the opposite direction.

The perennation of the fungus in Karachi area is thus still something of a puzzle. Further studies including those on the role of possible alternate hosts might solve the riddle.

Seasonal Variation in Infection in Field

The watermelon crop at Karachi is normally in the field from March to July. A survey of the crop was therefore undertaken during the season and the extent of infection recorded month by month. A summary of these observations is given in Table 2.

TABLE 2.—SEASONAL VARIATIONS IN WATERMELON RUST INFECTION IN THE FIELD.

Aug. to Mar.	April 1	2	3	4	May 1	2	3	4	June 1	2	3	4	July 1	2	3
	←						Weeks								→
No crop	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:
Traces or nil	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Very slight	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Slight	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Mild	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Mildly epiphytotic, all fields	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:
Moderately epiphytotic	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Severe	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:
Severe	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Very severe	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:
Very severe	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Mild; infection dying out	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:
Slight	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Very rare	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:
Nil	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.
Nil	:	:	:	:	:	:	:	:	:	:	:	:	:	:	:

The fall of infection at the end of June was due to the uprooting of plants as the fruits matured by the 3rd week; in 4th week the rains started. After the rains the infection comes to an end.

Varietal Susceptibility

Several varieties of watermelons are cultivated commercially. These are chiefly distinguished by the colour of the ripe fruit and of seed. Some of these varieties have been tested for possible variations in susceptibility to the rust. All these commercial varieties were found as highly susceptible as one cultivated locally. Results of these experiments are given in Table 3.

Signs and Symptoms

The uredia and the telia are produced mainly on leaves, but occasionally also on the petiole and stem. The uredia while occurring on both sides of the leaves are commonest on the upper surface. As the mycelium extends through the thickness of the leaf, it is common to find a pustule on the upper surface and vice versa. When uredia are few and not crowded it is customary to find a ring of smaller uredia around a larger central one (Plate 5).

Uredia are without spot, scattered, round, 0.5 to 2.5 mm. in diameter. With the increasing intensity of infection, the pustules formed are generally smaller in size than at the beginning of the infection. At first they are sub-epidermal but soon the epidermis ruptures exposing the powdery masses of spores. When uredia first appear, there is usually little or no discolouration of the surrounding leaf tissue, but as the infection proceeds and especially when sori are numerous the leaf becomes partially or completely yellowed and dies. In some cases it has been noticed that the sori are surrounded by partially discoloured areas of leaf, in some cases typical necrosis takes place (Plate 3). Whether these reactions could be indicative of the presence of more than one physiological race in the population has not yet been ascertained.

TABLE 3.—SUMMARY OF THE RESULT OF INOCULATION ON DIFFERENT VARIETIES OF WATERMELON.

	Karachi black	Karachi green	*Citrullus colocynthis*	Sind black	Sind green with red seed	Sind green	Punjab black	Punjab green
No. of plants inoculated	450	100	100	50	30	30	350	100
No. of plants got infection	450	100	100	50	30	30	350	100

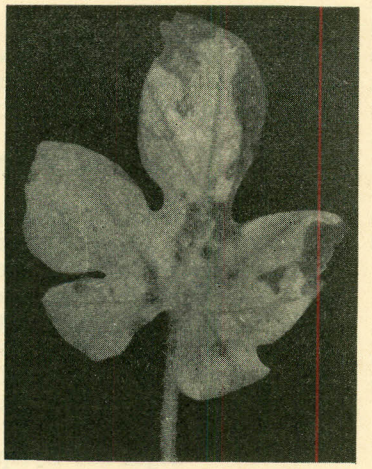

Plate. 3.—Necrotic zones on the leaf of watermelon (*Citrullus vulgaris*).

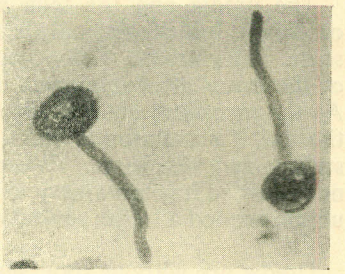

Plate. 4.—Germinating uredo spores showing hyaline germ tube.

Uredia at first are yellowish brown, later they change to brown or slightly dark brown. Telia are dark brown in colour, occur both on dorsal and ventral surfaces of the leaves. They are minute, rounded, usually mixed with uredia and very occasionally separate. The formation of telia does not seem to be general. It seems to depend upon a combination of various climatic conditions, e.g., higher temperature accompanied by low humidity.

Nature of Pustules

The pustules are identical on both the surfaces. Small uredia may be surrounded by necrotic zones. At first the leaves show only a little necrosis but later it becomes more clear, and the leaves become more chlortic and die prematurely. In some cases two or three sori which are broadly scattered show a whitish or discoloured area surrounding them together. After a few days a ring of numerous small pustules is produced around it (Plate 5). Occasionally the tissues where the sori are formed is raised due to the excessive growth in the leaf tissue resulting in the small cavity below the raised portion. The arrangement of sori is irregular, i.e. scattered on the whole surface.

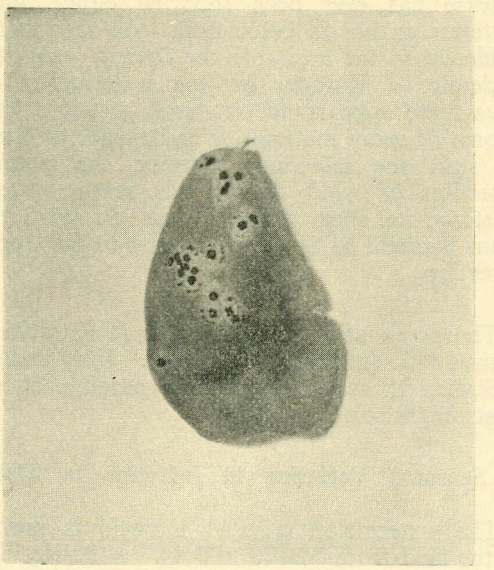

Plate 5.—Ring of small uredia around the larger central one.

The uredo spores are 23.10-29.70X19.80-24.75 M (cf. Table 4). They are yellowish brown to light brown in colour, spherical or ovate in shape. There is a single thick echinulated wall with 4 equatorial germ pores. The spores germinate readily in water. At 14°C. it takes only two hours for germination of fresh spores giving a glassy hyaline germ tube (Plate 4). The germ tube is 6.6 M in diameter.

TABLE 4.—SIZE OF UREDOSPORES IN MICRONS.

Leaf collected from Malir			Leaf inoculated in Lab		
Length/ no. of spores	Breadth/ no. of spores	Total no. of spores	Length/ no. of spores	Breadth/ no. of spores	Total no. of spores
29.7/19	23.97/14	100	29.70/13	24.75/3	100
28.00/13	23.10/36		28.00/7	23.93/5	
27.20/6	22.28/7		26.40/40	23.10/40	
26.40/24	21.45/26		25.57/3	22.28/22	
24.75/19	20.60/10		24.75/16	21.28/22	
23.10/19	19.80/7		23.10/211	9.80/8	

Size, Colour and Form of Teleuto-spore

The teleuto-spores are 26.40-33.00X31.35-42.2 M (cf. Table 5). They are two-celled, dark brown to chestnut brown in colour, ellipsoid, slightly constricted in the middle, with a smooth thick wall. The germ pore of the upper cell is just at the centre ofthe apex, while germ pore of the lower cell is just

below the septum. Stalk is not very persistent and is 19.30 - 36.30X4.95 - 6.6 M (Table 6).

TABLE 5.—SIZE OF TELEUTO-SPORE (IN MICRONS).

Collected from Malir			Produced in Lab.		
Length/ no. of spores	Breadth/ no. of spores	Total no. of spores	Length/ no. of spores	Breadth/ no. of spores	Total no. of spores
42.20/9	33.00/8	100	42.25/12	29.70/67	100
3.60/22	31.35/6		39.60/49	28.00/17	
37.90/15	29.70/43		37.90/12	26.49/13	
36.00/27	28.00/12		36.30/24	26.00/3	
34.60/10	26.40/31		34.60/3		
33.00/13					
31.35/4					

TABLE 6.—SIZE OF TELEUTO-SPORE STALK (IN MICRONS).

Length/no. of spores	Breadth/no. of spores	Total no. of spores
36.30/8	6.6/81	100
33.00/5	5.77/9	
31.55/3	4.95/10	
29.70/21		
28.00/4		
26.40/16		
23.10/21		
21.45/20		
19.30/3		

Teleutospores have a long resting period and could only germinate if subjected to freezing temperatures. The present experiment on germination (Table 7) shows that the spores needed alternate freezing and thawing for a period of 45-60 days before they germinate to produce the promycelia.

Method of Inoculation

The viable uredo spores have been used successfully for inoculating plants (before inoculation the spores were tested for their viability). Inoculations were made by two different techniques :

(*a*) By the simple spraying of a spore suspension on leaves, and
(*b*) By injecting spore suspension below the epidermis with a hypodermic syringe.

TABLE 7.—GERMINATION OF TELEUTO-SPORE (SPORES KEPT IN REFRIGERATOR ON 9-7-1956).

Date of examination	Observation	% of germination
9-7-1956	No germination	0
15-7-1956	,,	0
21-7-1956	,,	0
2-8-1956	,,	0
9-8-1956	,,	0
16-8-1956	,,	0
24-8-1956	,,	0
30-8-1956	,,	0
9-9-1956	2 spores germinated	4

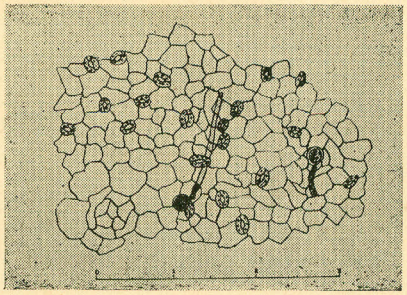

Plate. 6.—Path of infection through the cuticle.

After inoculation the plants were covered over by a bell jar, humidified and the plants together with the bell jar kept in a refrigerator for 16-24 hours at 14°C. Thereafter the bell jars were removed and the plants kept at room temperature and humidity. During winter with room temperature ranging from 24-16°C. or lower, it was not necessary to keep the inoculated plants in a refrigerator. Still the plants needed keeping under the bell jar for the sake of humidity.

The incubation period, i.e., the time from inoculation to the appearance of faint brown spots, was usually six days under the conditions of these experiments when temperature ranged between 26-32°C. and humidity was 100%. A further period of two days was required for the appearance of pustules. During winter when temperature falls down, the incubation period prolongs by one to two days depending on the fall of temperature.

Path of Infection

The uredo spore on germination on leaf surface gives rise to a germ tube which is unbranched

till it enters the host tissue. The germ tube usually enters the leaf through cuticle (Plate 6) but it also enters through stomata. In some cases, germ tubes were observed entering into the leaf through the basal cell of the leaf hair, and reaching through them into the pallisade tissue of the leaf.

Host-Parasite Relationship

After entering into the host tissue, the mycelium ramifies in all directions between and within the cells. At the time of spore formation, the hyphae collect below the epidermis and produce spores on small stalks which exert a pressure on the epidermis, resulting in its ultimate rupture and liberation of the spores.

References

1. S.Z. Hasanain and Azmatullah Khan, *Some new and interesting records of fungi from Karachi*, Proceeding of the Second Pakistan Science Conference (1950), Part. III, p. 32.
2. Syd and Butler, Ann. Mycol. Notitiam Sci. Mycol. Univ., **10**, 259 (1912).