歟冬の細菌性斑點病

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Bacterial disease of Petasites.

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In July, 1921, the bacterial disease of Petasites (*Petasites Japonicus* Miq.) was first observed by the writer in the vicinity of Fukuoka. In March 1922, the same disease was observed at Dazaifu, and also in the farm of Fukuoka Agric. Exp. Station, near Fukuoka. In November 1923, the writer collected the diseased leaves of petasites in the farm of Kagosima Agric. Exp. Station, near Kagosima.

With these material numerous isolations were attempted, and the blackened areas of the lesions were found to contain the bacteria in large number and no fungi. Inoculations proved that the disease is caused by a bacterium hitherto undescribed. Therefore the brief descriptions are mentioned as follows:

Symptom: In the early stages the disease appears on the leaves as small circular brown spot, the spots gradually become enlarged and irregularly elongated. Sometimes the spots turn black and finally look like rag. On the under surface of the leaves, the spots at first take on dull green or dark purplish brown, and then usually turn dark brown.

Blackened area are cut into small pieces which are dipped in 80% of alcohol, for few seconds. The piece then are put in mercuric chloride solution 1:1000 for 1—2 minutes, rinsed in sterile water, and transferred in bouillon from which the dilution plate cultures are made. Successful isolation is also made from diseased pieces free from contamination with flamed knife instead of disinfection as above.

Inoculation experiments: Three methods were applied such as injection, smearing and spraying. The water suspension of the organism from one day old agar cultures was used as inoculum. In the case of injection and smearing the lesions appeared in 1 or 2 days but when spraying was applied they appeared in 5 days taking the characteristic symptoms. The upper side of the leaves is more susceptible to the disease than the under. By these inoculation experiments

the pathogenicity of the organism was confirmed, and the organism was found to infect the plant without wounds.

Morphological characters of the organism: The organism is a short rod with rounded ends, and motile by means of a polar flagellum. It occurs singly or occasionally in pairs. The limit of size is $1.1-1.7 \times 0.8-1.0 \mu$. No spore have been observed even in one year old cultures. Capsule and involution forms are not formed. It stains readily in basic anilin dyes. Gram's stain gives negative result.

Cultural and biochemical characters of the organism: (All cultural tests were made at 28°C ., with exception of gelatin culture.)

Agar plates ($+15^{\circ}$ in Fulles's scale): The surface colonies are circular or amoeboid flat with smooth and entire margine, and their color are pale whitish at first and finally dirty whitish taking granular structure.

Agar slant: In newly isolated cultures growth is filiform but when repeatedly transferred it becomes spreading. The surface is smooth of slightly verrucose and the consistency is butyrous.

Bouillon: Strong clouding occurs in 24 hrs., fine powdery pellicle and heavy viscid precipitation are formed in one or two months. The reaction of medium when titrated with phenolphthalein is $+0$ to -5 while that of control is $+15$.

Potato plate: Vigorous growth occurs in 24 hrs., the surface of the colonies is flat and verrucose; conspicious odor is emitted; starch destroyed feebly; color of the growth is marqurite yellow.

Litmus whey: At first litmus begins to turn blue, then the bleaching occurs at the bottom of the tube, and in 30 days the milk coagulates; in 3 weeks the bleaching is complete except 1 c.m. of the upper parts of the tubes.

Ushinsky's solution: Uniform clouding occurs, but rather feeble, in 10 days powdery white pellicle and the heavy viscid sediments are formed.

Fermentation tube tests: (1) In potato juice moderate clouding occurs in the open and closed ends of the tubes, and the abundant gas is produced. The fluid in the open arms turns, brownish black, and its reaction becomes alkaline, (2) in 2 % of Teruuchi's peptone water containing 2 per cent of dextrose, sucrose, lactose, mannit respectively, strong clouding occurs in both arm.

in 2 days and then gas and heavy precipitation are produced. The reaction of the fluid is alkaline, (3) In 2 % of Teruuchi's peptone water containing 3 per cent glycerine clouding occurs strongly in the open arm and slowly and feebly in the closed. The heavy precipitation is formed without gas and the reaction of the fluid become acid.

Aerobism: The organism grows in Buchner's tube, and also in the closed end of the tubes containing sucrose, dextrose or mannit.

Hydrogen sulfide: Hydrogen sulfide is not produced, the lead acetate paper suspended over the broth culture being not blackened.

Nitrite in nitrate bouillon: Nitrates is reduced in 3 days.

Temperature relation: The maximum temperature for growth on beef broth is about 47°C., at 5°C. the organism does not grow even in the course of 6 days, the optimum temperature for growth is 27—30° C., the thermal death point ranges between 55°C. and 56°C.

Moisture relation: The organism stands to dryness. The organisms from 3 days old broth cultures were smeared on the cover glasses and placed in sterile Petri-dishes which had been kept in desiccator. When each piece of these cover glasses was thrown into sterile bouillon at definite intervals there was a good growth in that of 9 days keeping, but none in that of 11 days keeping.

Tolerance of acids: There is a feeble growth in beef broth (originally +15) containing 0.1 % and 0.2 % of citric or tartaric acid, but none in that of 0.3 % of the acids.

Tolerance of sodium chloride: There was a feeble growth in beef broth containing 6 % of sodium chloride, but none in that of 8 or 9 %.

Effect of sunlight: The organism is not sensitive to bright sunlight. In 2 hours exposure one-third of the bacteria were destroyed, and in 3 hrs. exposure all were destroyed.

Vitality on culture media: It remained alive for 11 months on beef agar.

Technical description:

Bacterium Petasitis n. sp.

Short, rapidly growing rod with rounded ends, motile by means of a polar flagellum, occurring single or in pairs; average measurements 1.1—1.7 x 0.8—1.0

μ .; no spore, capsule, zoogloea nor involution form observed; Gram negative; stains readily in gentian violet and methylen blue; facultative aerobic. Colonies on beef agar white, circular or amoeboid, flat, with entire margins; smooth surface and butyrous consistency; gelatine not liquefied; milk curdled in 30 days; litmus whey reduced; indol not produced; gas formed; nitrates reduced; slight diastasic action on potato plate; good growth in Ushinsky's solution; Max. Temp. for growth 47°C .; opt. temp. $27-30^{\circ}\text{C}$.; thermal death point $55-56^{\circ}\text{C}$.; it not sensitive to dryness, destroyed in 3 hrs. exposure to bright sunlight; long vitality on culture media. Pathogenic on the leaf of *Petasites Japonicus* Miq. in Fukuoka and Kagosima prefectures.

摘 要

- (一) 一九二一年以後福岡縣地方の欽冬に一種の細菌性の病害發生し、葉を枯損し被害大なりき。
- (二) 本病は最初葉面に小なる褐色の圓形病斑を生じ漸次増大且不規則的に延長し遂に縞襌狀となりて枯損す。
- (三) 數回に亘り細菌の分離及接種を行ひたるに常に白色の細菌を分離し、該細菌は無傷の葉面に感染し得るものなることを知れり。
- (四) 分離せる細菌は從來記載せられたる植物の病原細菌より異なり、一の新種なるものと認めたるを以て之れに *Bacterium Petasitis* n. sp. の學名を附與せり。
- (五) 本研究は中田教授指導の下に九大、農學部植物病理學教室にて行ひたるものにして研究及發表に關し日野巖氏の助力を得たところ多し、共に感謝するところなり。

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