

SYSTEMIC ACTIVITY OF PROCAINE HYDROCHLORIDE ON POWDERY MILDEW¹

Met een samenvatting: Systemische werking van procaine-hydrochloride tegen echte meeldauw

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Investigating control of plant diseases with antibiotics at the University of California, DR. P. A. ARK (personal communication) observed systemic activity of a commercial penicillin preparation (procaine penicillin G) against powdery mildew on cucumbers after administering this antibiotic to the roots. Follow up of this observation at our laboratory indicated that not penicillin G but the procaine part of the molecule was responsible for the chemotherapeutic effect. The following is a preliminary report on the activity of procaine hydrochloride against several powdery mildews of plants.

Cucumber plants, variety "Lange gele tros", were grown on HOAGLAND nutrient solution. After development of two leaves, the plants were inoculated with a conidial suspension of *Erysiphe cichoracearum* DC, and procaine HCl was added to the nutrient solution. Transfer of the plants to a procaine free medium took place after 4 days and assessment for mildew after 14 days, using a scale ranging from 0, no mildew, to 5, leaf covered with mildew.

TABLE 1. Powdery mildew on cucumber plants, after addition of procaine HCl to the nutrient solution for 4 days. Average of 6 plants each with 2 leaves.

Ontwikkeling van meeldauw op komkommerplanten, die gedurende 4 dagen na de inoculatie op een voedingsoplossing met procaine-HCl hadden gestaan. Gemiddelde van 6 planten, elk met 2 bladeren.

Treatment <i>Behandeling</i>	Degree of powdery mildew development * <i>Mate van meeldauwontwikkeling</i>
control	5.0
75 p.p.m. procaine HCl	4.5
150 " " "	2.1
300 " " "	0.0

*0 = no mildew/*geen meeldauw*

5 = leaves covered with mildew/*bladeren geheel met meeldauw bedekt*

The highest concentration of procaine HCl caused a slight browning of the edge of the oldest leaf.

Procaine HCl was active against mildew development on the leaves, when added to the roots in HOAGLAND solution within 3 days after inoculation. Ad-

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ministering it at a later date did not eradicate the powdery mildew, but prevented its spread over the rest of the leaf.

Cucumber plants could also be kept free or almost free of mildew by spraying the leaves with a 1000 p.p.m. solution on the first and second day after inoculation, or by pouring 50 ml of a 0.5% solution on the soil of a 12 cm diameter flower pot with three plants, for four successive days after inoculation.

Uptake of procaine HCl by the roots and transport to the leaves was demonstrated by paper chromatography. Three weeks old cucumber plants were placed with their roots in a 300 p.p.m. solution for 48 hours. Eighty mm³ of the leaf juice was spotted on WHATMAN no. 3 chromatography paper and the paper developed with butanol- acetic acid - water 50:15:45 v/v as a descending solvent. Since procaine provides an orange colour after diazotation and coupling with α -naphthol (JAMINET, 1951), the paper chromatogram was sprayed with a mixture of an 1% aqueous NaNO₂ solution and glacial acetic acid, 10:1 v/v, and thereafter with a mixture of a 1% alcoholic α -naphthol solution and concentrated NH₄OH, 4:1 v/v. In the chromatogram of leaf juice from treated plants, an orange spot appeared with a R_f value equal to that of procaine HCl.

Spraying of procaine HCl was found to be effective against the following powdery mildews:

Erysiphe cichoracearum on cucumbers and gherkins,
Erysiphe graminis DC. on wheat,
Erysiphe polygoni DC. on beets, peas and lupins,
Sphaerotheca pannosa Lév. on roses,
Podosphaera leucotricha (Ellis & Ev.) Salm. on apples.

Phytotoxicity was observed, when frequently high concentrations of procaine HCl were sprayed with intervals of about three days: necrotic spots on leaves and fruits of apple trees (0.4 % procaine) and drop of the older leaves of roses (0.8 % procaine).

Since no systemic protection of procaine HCl was obtained against any other fungal or bacterial diseases tested, including rust on beans, early blight on tomatoes, cucumber scab, late blight on potatoes, downy mildew on lettuce, and halo blight on beans, it seems likely that it acts specifically on powdery mildews.

In vitro a concentration of 1000 p.p.m. was inactive against all ten fungi and bacteria tested. Even germination of conidia of *Erysiphe cichoracearum* appeared to be unaffected, either in vitro or on the leaves of plants in HOAGLAND solution, to which procaine HCl, 300 p.p.m., had been added. In the last case penetration of the fungus was observed into the epidermis, but no symptoms of the disease ever developed. So it seems that procaine HCl is only effective after it has penetrated into the plant. It is not yet known, whether it acts directly on the haustorium of the fungus, or via the metabolism of the plant.

Both constituents of procaine, namely p-aminobenzoic acid and diethyl-amino-ethanol were ineffective on pathogens for which procaine showed a remarkable activity.

More detailed results concerning the systemic action on powdery mildews of procaine HCl and a number of related substances will be published elsewhere.

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SAMENVATTING

In een voorlopige mededeling wordt de systemische werking van procaïne-hydrochloride tegen echte meeldauw op komkommers en enkele andere gewassen vermeld.

Komkommerplanten, die met hun wortels in een voedingsoplossing gezet werden welke 300 d.p.m. procaïne-HCl bevatte, bleven vrij of nagenoeg vrij van meeldauw (tabel 1). Dit was eveneens het geval wanneer een 0,5 % procaïne-oplossing gegoten werd bij de wortels van in grond opgroeiende planten, of wanneer een 1000 d.p.m. oplossing op het blad gespoten werd (fig. 1). Transport van de wortels naar het blad werd aangetoond met behulp van papierchromatografie en een kleurreactie.

Na bladbespuiting was procaïne-HCl ook actief tegen meeldauw op augurken, tarwe, bieten, erwten, lupine, rozen en appels. Herhaaldelijk spuiten met hoge concentraties (0,4–0,8 %) veroorzaakte evenwel op diverse gewassen fytoxische verschijnselen. Een aantal aan procaïne verwante stoffen werd bij de voortzetting der proeven betrokken.

Procaïne-HCl is vermoedelijk een specifiek op echte meeldauw werkend middel, daar het in vitro noch in vivo werkzaam was tegen alle andere getoetste planteparasieten. Tegen meeldauw werkt het pas nadat het door de plant opgenomen is; het heeft geen waarneembare invloed op de kieming der conidiën in vitro.

REFERENCE

JAMINET, FR., – 1951. Séparation de quelques anesthésiques locaux par chromatographie de partage sur papier. J. Pharm. Belg. 6:81.