INHIBITORS OF PEA ROOT INDOLEACETIC ACID OXIDASE

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SUMMARY

A list of 33 substances that inhibit the destruction of IAA by an extract from pea roots is presented. The effect on IAA-oxidase activity of 16 of these compounds was not tested before.

1. INTRODUCTION

As has been demonstrated by many authors, several compounds inhibit the destruction of indoleacetic acid (IAA) by plant extracts (see PILET & GASPAR 1968). IAA-oxidase inhibitors are usually polyphenols having at least one free hydroxyl group on an aromatic ring ortho or para to another hydroxyl group (HARE 1964) or reducing substances like ascorbic acid and cystein (BETZ 1963). The available results were obtained with IAA-oxidase preparations of various origin. During the course of our investigations of the IAA-oxidase from pea roots the effect of many compounds on this IAA destroying system was studied. The results obtained with cofactors have been published previously (JANSSEN 1969a). In this paper a list of compounds that inhibited the pea root IAA-oxidase will be presented.

2. MATERIAL AND METHODS

From root-tips of 65 hours old seedlings of *Pisum sativum* cv. "Vlijmsche Gele Krombek" an extract was prepared in phosphate-citrate buffer solution of pH 5 as described before (JANSSEN 1969a).

The IAA-oxidase activity was measured in a reaction mixture consisting of 0.2 ml IAA 10^{-3} g/ml, 0.5 ml p-coumaric acid 10^{-5} g/ml, 3.3 ml phosphate-citrate buffer solution of pH 5, 0.5 ml of the crude enzyme preparation (30 min. 27,000 g) and 0.5 ml of the substance to be tested. The (final) concentrations used were 10^{-5} g/ml to 10^{-8} g/ml. Ascorbic acid and cystein also at 10^{-4} g/ml. After an incubation period of 30 minutes the remaining IAA was determined with the Salkowski reagent according to TANG & BONNER (1947).

The experiments were performed in dim red light at 22°C. During the incubation time the reaction mixtures were carefully shaken.

3. RESULTS

The substances tested are listed in *table 1*. Except ascorbic acid and cystein they

Table 1.

Inhibitors of pea root indoleacetic acid oxidase.
The effect on IAA-oxidase activity of the compounds marked with a * has not been tested before.

a. para-diphenols
hydroquinone
gentisic acid
*2,5-dihydroxyhippuric acid

b. ortho-diphenols
catechol
protocatechuic acid
*hydrocaffeic acid methylester
*caffeic acid methylester
caffeic acid
chlorogenic acid
dihydroxyphenylalanine
*adrenalin
*3,5-dihydroxyphenyl-thiourea
*2,3-dihydroxybenzoic acid

c. triphenols
pyrogallol
gallic acid
*2,4,5-trihydroxybenzoic acid
*2,3,4-trihydroxybenzoic acid

d. methoxy-compounds
guaiacol
ferulic acid
syringaic acid
synapic acid
*p-anisidine

e. phenylenediamines
p-phenylenediamine
*m-phenylenediamine
*o-phenylenediamine
*N, N, N′, N′-tetraethyl-p-phenylenediamine

f. various compounds
*p-toluidine
*p-aminophenol
*4,4′-dihydroxybiphenyl
p-benzoquinone
*1,8-dihydroxy-3,6-dimethylnaphtalene
ascorbic acid
cystein

all caused a complete inhibition of the destruction of IAA during the incubation period of 30 minutes at the concentration of 10^{-5} g/ml. None of the concentrations of ascorbic acid and cystein used caused complete inhibition. Even the presence of 10^{-4} g/ml did not prevent that some of the IAA in the reaction mixture was converted.

If the phenolic substances of *table 1* are oxidized, quinones are formed. It seems that phenolic substances only inhibit IAA-oxidase activity if they can easily form quinones. This is supported by the fact that 4-4'-dihydroxybiphenyl, and 1,8-dihydroxy-3,6-dimethylnaphtalene which can form quinones, inhibit IAA-oxidase (*table 1*), while 4–4' dihydroxydiphenylmethane (JANSSEN 1969b) and 4-phenylphenol (GOLDACRE c.s. 1953), which do not form quinones, are cofactors of IAA-oxidase activity.

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