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## Effect of indole-3-acetic acid and kinetin on non-cyclic electron transport in intact leaf discs and isolated chloroplasts of *Cephalandra indica* L.

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Indole-3-acetic acid (IAA) and kinetin have been found to accelerate photosynthetic non-cyclic electron transport in light reaction, whereby the artificial electron acceptor 2,6-dichlorophenolindophenol (DCPIP) is reduced at a faster rate by accepting electrons. The rate of reduction of DCPIP by IAA treatment is, however, higher than that affected by kinetin. Reducing power of chloroplasts is remarkably higher when these are isolated from leaf discs pretreated with hormones compared to the system when isolated chloroplasts are subjected to hormone treatments *in vitro*.

INDOLE-3-acetic acid (IAA) and kinetin are two major plant growth regulators. IAA has a remarkable role in apical dominance, cell enlargement, cell division, ethylene formation and it also acts on some enzymes. Besides these effects, IAA promotes photosynthesis in a wide range of species<sup>1–3</sup>, influences movement of nutrients within plants<sup>4</sup> and stimulates the transport of hydrogen ions across membranes<sup>5</sup>. The possible role of various cytokinins in the regulation of photosynthesis includes the reduction of the lag phase in the production of grana and promotion of synthesis of photosynthetic enzymes and plastidial ribosomal RNA<sup>6,7</sup>. However, the phytohormones have been shown to affect mainly the enzymatic dark reactions of photosynthesis, but virtually little information is available on the effects of IAA and kinetin on photosynthetic electron transport in light reaction. In the present work, IAA and kinetin action on photosynthetic non-cyclic electron transport in the leaves of *Cephalandra indica* L. was studied. To test whether there is any difference in

sensitivity between intact and isolated systems towards hormones, both intact leaf discs and isolated chloroplasts were chosen as the experimental materials.

Fresh and healthy leaves of *Cephalandra indica* L. were surface-sterilized with 0.1% mercuric chloride and washed thoroughly with distilled water. For experiment with intact system, discs measuring 8 mm in diameter were cut from such leaves. Leaf discs weighing about 1 g were floated in different concentrations of IAA and kinetin solutions in petri dishes together with a water control and kept in dark for 1 h, then washed thoroughly with distilled water. For the isolation of chloroplasts, leaf discs were homogenized with chilled sucrose-phosphate buffer (0.5 M, pH 6.0), centrifuged at 2000 *g* for 5 min<sup>8</sup>. Supernatant was taken and centrifuged again at 5000 *g* for 15 min. Residue-containing, partially intact chloroplasts were taken and suspended in the extraction buffer. The volume of the chloroplast suspension was made up to 5 ml by the addition of the same buffer from which 1 ml was taken in test tube and diluted to 4 ml. After mixing the suspension with 0.5 ml of 0.03% 2,6-dichlorophenolindophenol (DCPIP), the tubes were kept under continuous illumination for 10 min provided by two white fluorescent tubes (Philips TL 40 W/33) giving photon density of 150  $\mu\text{E m}^{-2}\text{s}^{-1}$  (400–700 nm) at the material level. Chlorophyll content of 4.5 ml reaction mixture containing either treated or untreated chloroplast suspension was about 0.18 mg. After light exposure, the change in optical density of DCPIP undergoing decolorization was measured in Spectronic 20 colorimeter. The control containing chloroplast suspension was kept in dark and used to denote the initial optical density of the dye.

For experiment with isolated system, chloroplast suspension was prepared from untreated leaves in a similar way as mentioned before. One ml aliquot was taken in each test tube, diluted suitably, mixed with different concentrations of hormones and kept in dark for 1 h. Another set of controls was made using mixtures of different hormones taken separately with DCPIP but without chloroplast suspension and kept both in light and dark. It was noted, however, that there was no reduction of the dye without chloroplast suspension. Following dark incubation, 0.5 ml of 0.03% DCPIP was added to each tube and exposed to light as before. Change in optical density was measured and result was expressed in  $\mu\text{mol DCPIP reduced per mg chlorophyll per h}$ . Disappearance of blue colour of DCPIP to different degrees indicated the rate of non-cyclic electron flow, i.e. hill activity. Chlorophyll content was measured from 1 ml aliquot of chloroplast suspension<sup>9</sup>.

The results show that both the hormones increased the non-cyclic electron transport in both the systems (Figure 1). The salient features of the changes brought about by IAA and kinetin were essentially similar in



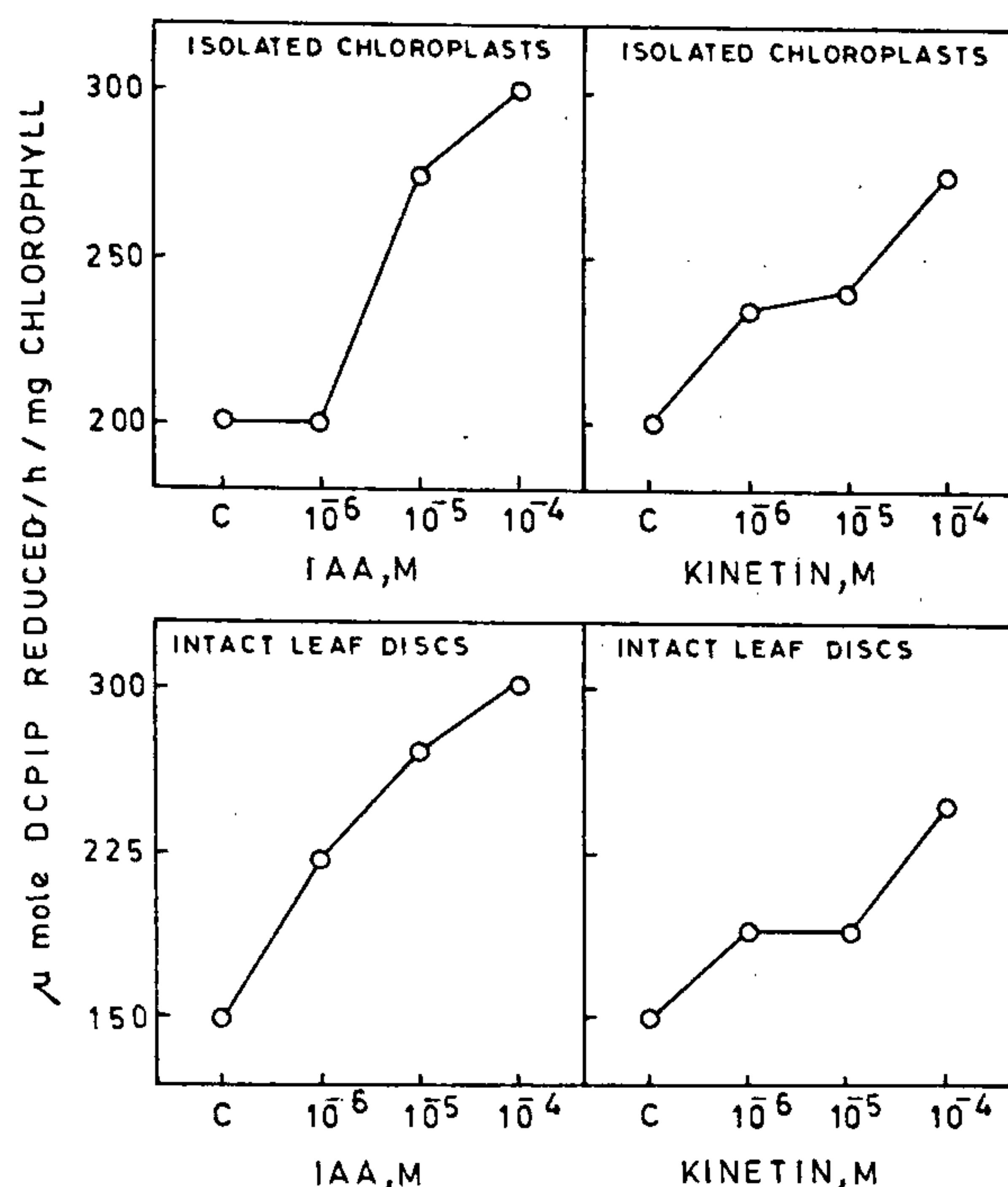


Figure 1. Effect of different concentrations of IAA and kinetin on the reducing power of chloroplasts. Data expressed as  $\mu\text{mol}$  DCPIP reduced per h per mg chlorophyll.

that the activity increased with increasing concentrations reaching the peak at  $10^{-4}$  M. However, the DCPIP photoreduction-capacity of chloroplasts of isolated system developed at a faster rate in presence of IAA (55% higher than control at  $10^{-4}$  M), whereas the effect produced by kinetin was relatively less (37% higher than control at  $10^{-4}$  M). In the intact system, on the other hand, the hormonal effect was a little more pronounced, showing 77% and 63% increments with IAA and kinetin respectively.

In the light reaction of photosynthesis, the rate of reduction of the electron acceptor dye DCPIP is directly proportional to the rate of electron flow from the excited chlorophyll molecules in a medium where photolysis of water takes place with generation of electrons. In our experiment, larger rate of reduction in presence of IAA and kinetin by chloroplasts from both sources indicates that they enhance the liberation of electrons and their flow towards the artificial electron acceptor DCPIP. Apparently, it is clear that IAA and kinetin accelerate the photolysis of water both in intact and isolated systems, giving rise to a greater rate of electron production. It is further noted that the hormone-induced development of reducing power of chloroplasts occurs in a more efficient manner in intact system compared to the isolated counterpart.

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## Introduction of rye genes into bread wheat by chromosome manipulations

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**Monosomic 5B ( $2n=41$ ) of wheat variety Chinese Spring was crossed with a rust-resistant strain of rye (*Secale cereale*,  $2n=14$ ). The 27 chromosome hybrid-lacking chromosome 5B of wheat and showing extensive chromosome pairing between wheat and rye chromosomes was backcrossed to rust susceptible wheat variety Sonalika. In the subsequent segregating generations only rust-resistant plants were selected and in  $BC_1-F_7$  several cytologically stable 21 bivalent-forming plants were identified. Many of these were resistant at the seedling stage to several races of leaf rust (*Puccinia recondita*) and to all the races of stripe rust (*P. striiformis*). These genetic stocks are expected to be a good source of rust resistance in wheat breeding.**

WHEAT is today an important cultivated plant to meet human needs and is the only crop with an annual world production of 520 million metric tons<sup>1</sup>. The present study reports our attempts towards transferring rust-resistant rye genes into bread wheat through chromosome 5B manipulations. This has been found necessary to prevent the loss of about 7–20% of grain yield every year amounting to 3.7 to 10.4 million tons of yield per year due to attack of rusts<sup>2</sup>. Chromosome 5B-deficient method was earlier employed for transferring desirable traits into bread wheat from its allied genera<sup>3,4</sup>. However this technique made it difficult to induce homoeologous recombinants between the chromosomes of wheat and rye<sup>5</sup>. Riley and Kimber<sup>5</sup> had colchicined the chromosome 5B-deficient hybrid of wheat and rye to yield a 54 chromosome amphidiploid.