Note

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Bisphenol A Stimulates Growth and Shoot Differentiation in Plants

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Abstract

Bisphenol A (4, 4′-isopropylinediphenol), an intermediate in the production of polymers and polycarbonates and an ingredient used in plastic dental fillings, is known to be an endocrine disruptor. Bisphenol A has been shown to exert estrogenic effects in animals, but its effects in plants were not known. We thus examined whether it has phytohormone effects. A cytokinin bioassay system that assessed the growth of soybean (*Glycine max* cv. 'Acme') calli showed that bisphenol A stimulated growth, and had maximum activity at a concentration of 10^{-1} μg ml^{-1}. Bisphenol A also induced shoot differentiation in carrot (*Daucus carota* L. var. *sativa* DC) calli. Thus, bisphenol A showed cytokinin-like activity.

Key words: bisphenol A (4, 4′-isopropylinediphenol), cytokinin, *Daucus carota, Glycine max* cv. 'Acme', growth, shoot differentiation.

Bisphenol A is used in the production of polycarbonate plastics and epoxy resins and can be solubilized from them (Brontons *et al.,* 1995; Olea *et al.,* 1996). It was shown to be estrogenic in ovariectomized rats (Steinmetz *et al.,* 1998) and by means of MCF-7 (human breast cancer) cell culture assay (Krishnan *et al.,* 1993). That is, it promotes growth.

On the other hand, its effect in plants has not been examined. In plants, cytokinins induce growth and shoot differentiation (Mok and Mok, 1994; McGaw and Buech, 1995). Typical cytokinins such as kinetin and zeatin, as purine derivatives, are structurally different from bisphenol A. Bisphenol A is not a derivative. We examined whether bisphenol A similarly promotes growth in plants.

We used a bioassay system that detects cytokinin activity (Miller, 1968; Haberer and Kieber, 2002). Soybean calli were used in the assay. They were placed on medium containing either bisphenol A or zeatin, a positive control.

On both bisphenol A and zeatin, the fresh weight of the calli increased (**Fig. 1**). The fresh weight increased linearly in relation to the concentration of zeatin (**Fig. 1**). In contrast, a peak in fresh weight occurred at 10^{-1} μg ml^{-1} bisphenol A. At that concentration, the activity was higher than that at the same concentration of zeatin (**Fig. 1**). These results suggest that bisphenol A induced growth. However, the growth pattern in relation to concentration differed between bisphenol A and zeatin. Bisphenol A may thus induce growth at a specific concentration only (**Fig. 1**).

Bisphenol A also induced the development of shoots in carrot (**Fig. 2**). Calli induced from carrot roots were transferred onto Murashige and Skoog (MS) medium (Murashige and Skoog, 1962) containing naphthaleneacetic acid (NAA) and bisphenol A, and were cultured at 26℃ under a 16-h light (30 μmol m^{-2} s^{-1}) / 8-h dark cycle. Shoots were again induced at a low concentration (10^{-1} μg ml^{-1}) of bisphenol A after 40 d (**Fig. 2A**). On the other hand, shoots were not induced at a high concentration (10 μg ml^{-1}) of bisphenol A (**Fig. 2B**). On zeatin, shoots were induced at both 10^{-1} and 10 μg ml^{-1} (**Figs. 2C, D**). They were not induced at very low concentrations of bisphenol A or zeatin (10^{-6} or 10^{-4} μg ml^{-1}) or on NAA alone (control) (data not shown). These results were consistent with the change in the fresh weight of calli in the bioassay (**Fig. 1**).

These results confirm that bisphenol A promoted growth and shoot differentiation. The induction of

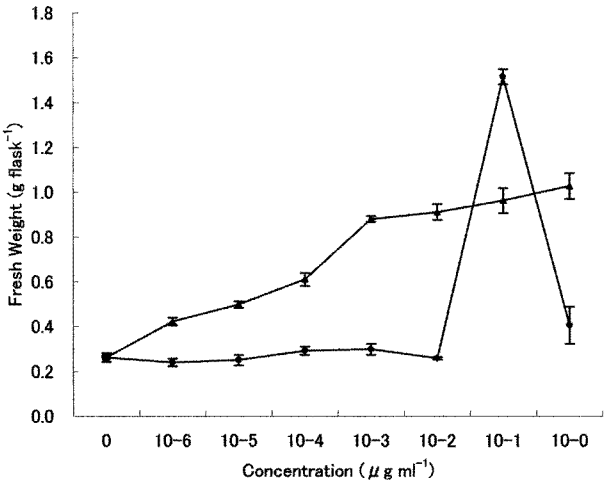

Fig. 1 Changes in the fresh weights of soybean calli exposed to bisphenol A (●) and zeatin (▲). Error bars indicate S. E.

cell growth and the development of shoots is a characteristic of cytokinin activity (Mok and Mok, 1994; McGaw and Buech, 1995). Thus, bisphenol A shows cytokinin-like activity.

Our study is the first to show that bisphenol A has cytokinin-like activity. The growth at 10^{-1} μg ml^{-1} bisphenol A was different from that induced by zeatin (**Fig. 1**). We now plant to study the differences between bisphenol A and zeatin in the mechanisms that induce growth and shoot formation.

Acknowledgements

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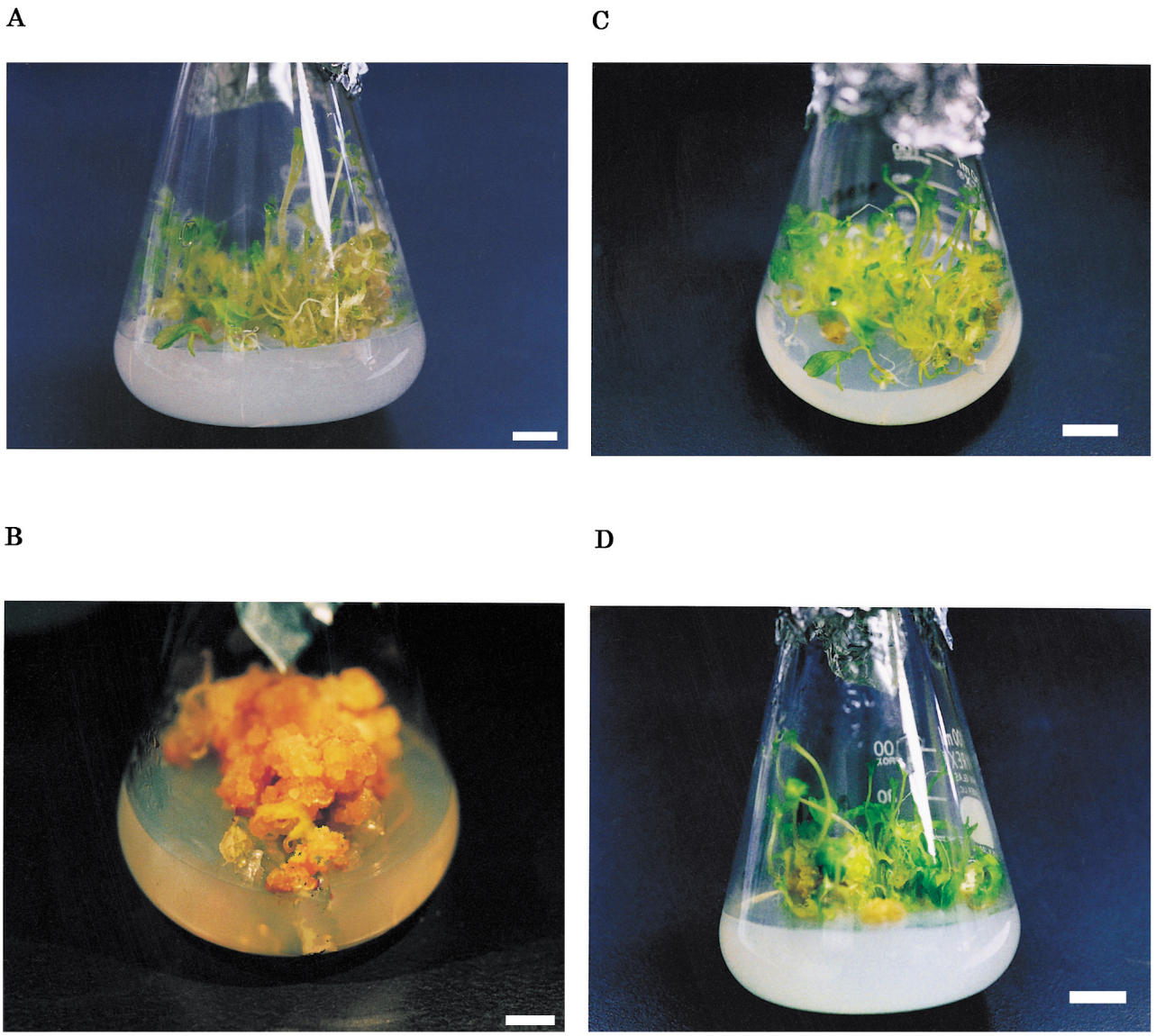

Fig. 2 Induction of shoots from calli in carrot. Explants of carrot root were cultured on MS medium containing NAA (10^{-1} μg ml^{-1}) at 26°C under a 16-h light (30 μmol m^{-2} s^{-1}) / 8-h dark cycle for 4 weeks. The induced calli were subcultured every 4 weeks. Calli that had been subcultured for 3 generations were used for shoot induction. The calli were transferred onto MS medium (NAA 10^{-1} μg ml^{-1} ; bisphenol A 10^{-1} or 10 μg ml^{-1}, or zeatin 10^{-1} or 10 μg ml^{-1}) and cultured as before. (A) Bisphenol A 10^{-1} μg ml^{-1}. (B) Bisphenol A 10 μg ml^{-1}. (C) Zeatin 10^{-1} μg ml^{-1}. (D) Zeatin 10 μg ml^{-1}. Bars = 1 cm.