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EFFECT OF PHYTOHORMONES ON UREASE ACTIVITY OF MARROW COTYLEDONS

El-Shora, H. M. and Lamiaa F. M. El Said
Botany Department, faculty of Science, Mansoura University, Mansoura, Egypt

ABSTRACT

Urease (EC 3.5.1.5, urea amidohydrolase) was isolated from marrow cotyledons of 5-day old seedlings. The effect of exogenous phytohormones namely indolebutyric acid, naphthaleneacetic acid, indoleacetic acid, indolepropionic acid, indoleacetic acid and zeatin at various concentrations (100-600 µmol) on urease activity in the cotyledons of marrow was investigated. The optimum concentrations of the five compounds that gave the highest activities of urease were 500 mol for indolebutyric acid indoleacetic acid, and 400 mol for naphthaleneacetic acid and indolepropionic acid However, the optimum concentration for zeatin was 100 µmol .

INTRODUCTION

Urease (EC 3.5.1.5, urea amidohydrolase), a nickel-dependent metalloenzyme, catalyzes hydrolysis of one molecule of urea results in the release of two molecules of ammonia and one molecule of carbon dioxide (Lam *et al*., 1996).

Urea + H_2O→ Ammonia + Carbon dioxide

Urease is an abundant seed protein in many members of the *Leguminosae, Cucurbitaceae, Asteraceae, and Pinaceae* **(**Bailey & Boulter, 1971), urease is also found at lower levels in the vegetative tissues of most or all other plants (Hogan *et al*., 1982).

This enzyme is present in two isoforms (Hollander *et al.,* 1971**);** the embryo-specific urease, which is synthesized only in the developing embryos (Turley& Ching*,* 1986) and the ubiquitous urease, which is found in all vegetative tissues (Pollaco *et al*., 1993).

Moreover, urease is the only nickel-containing metalloenzyme yet identified in plants (Polacco & Holland, 1993; 1994).

The present paper aimed to investigate the effect of some phytohormones on urease activity of marrow (*Cucurbita pepo* L.) cotyledons. The tested phytohormones were indolebutyric acid, naphthaleneacetic acid, indoleacetic acid, indolepropionic acid, Indoleacetic acid (IAA) and zeatin.

MATERIALS AND METHODS

The germination of marrow *(Cucurbita pepo* L.) seeds This method was carried out according to El-Shora, (2001). The preparation of urease was carried out according to El-Shora, (2001**).**This assay of enzyme is made according to El-Shora **(**2001).

Total protein in the extract was determined by the method of Meyer-Bothling & Polacco, J.C. (1987).

Phytohormones were tested at 100-600 µmol for 3 days. Samples of treated and non treated plants were taken and analyzed.All values are means of three measurements± S.E.

RESULTS

Indolebutyric acid on urease activity in the cotyledons was studied. This compound was tested at various concentrations (100 – 600 µmol). The results were shown in Fig 1. These results indicate that by increasing indolebutyric acid concentration, urease activity increased until concentration 500 µmol after which the activity decreased. Naphthaleneacetic acid induced the enzyme activity. The induction of urease was dependent on the concentration. It was observed that concentrations range from 100 to 400 µmol (Fig. 2) induced the enzyme activity whereas the higher concentrations 500 and 600 µmol were inhibitors. Indoleacetic acid (IAA) at the same different concentrations (100- 600 µmol).The results in Fig. 3 indicate that urease activity was induced by IAA up to 600 µmol; however at 600 µmol the enzyme activity was reduced. It is noted that the enzyme activity at 600 µmol IAA was still higher than the control samples to which no IAA was added. Indolepropionic acid was stimulator at the range of 100 to 400 µmol (Fig. 4) whereas 500 and 600 µmol reduced the activity of the enzyme**.**When the cotyledons were incubated with zeatin (10 – 100 µmol). The results shown in Fig 5 illustrate that the enzyme activity increased with increasing zeatin concentration. It is apparent that the enzyme activity reached 52.8 U mg^{-1}protein after treatment with 100 µmol zeatin.

Discussion

Urease activity of zeatin-trated cotyledons increasede to a level higher than in water –treated control cotyledons. In support, urease was induced by *Aspergillus niger* in the presence of zeatin in groeth medium (El-Shora *et al*., 2005**).**

It is possible that zeatin may act at the transcriptional or posttranscriptional regulation. However, this needs further investigation. Another possibility for the increase of the enzyme activity by zeatin is that it may be involved in urease phosphorylation. To test this hypothesis saturosporine a well known inhibitor of protein kinase was added with zeatin to see whether the induced-activity of the enzyme by zeatin could be inhibited.

Also, treatment of cotyledons with zeatin in the presence of Furd-cordycepin and α-aminitin fails to cause an increase in urease activity. This result strongly supports the suggestion that zeatin-induced protein synthesis is necessary for the increased urease activity by zeatin

Urease activity showed remarkable enhancement in triazoles treatments. Triazole compounds increased other plant enzymes such as amylase and invertase (Kishorekumar *et al.,* 2007). This induction of acid phosphatase activity varies with concentration of triazoles. Application of triazole compounds may be proved to be a good tool in enhancing urease in cotyledons of marrow plants.

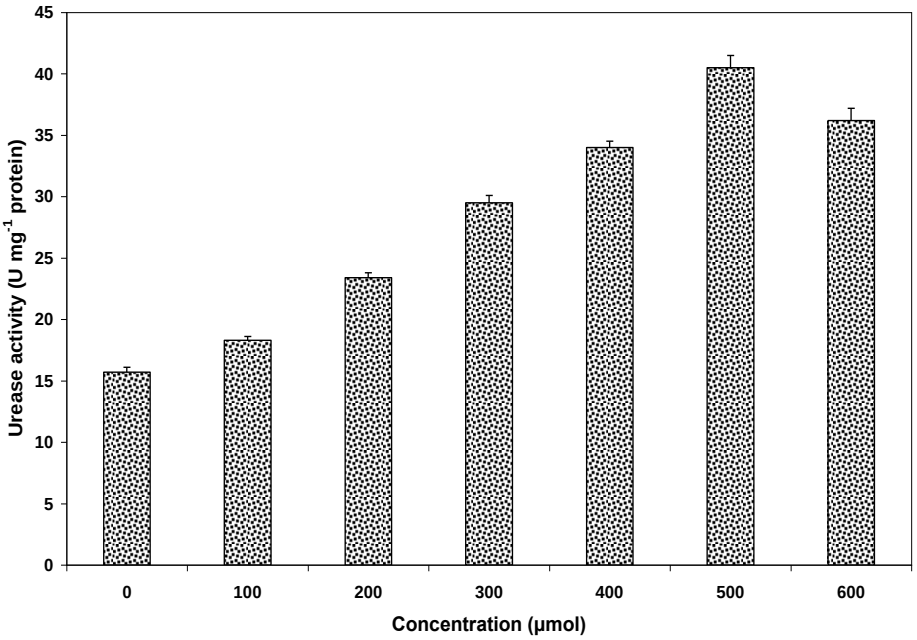

Fig 1: Effect of indolebutyric acid on urease activity.

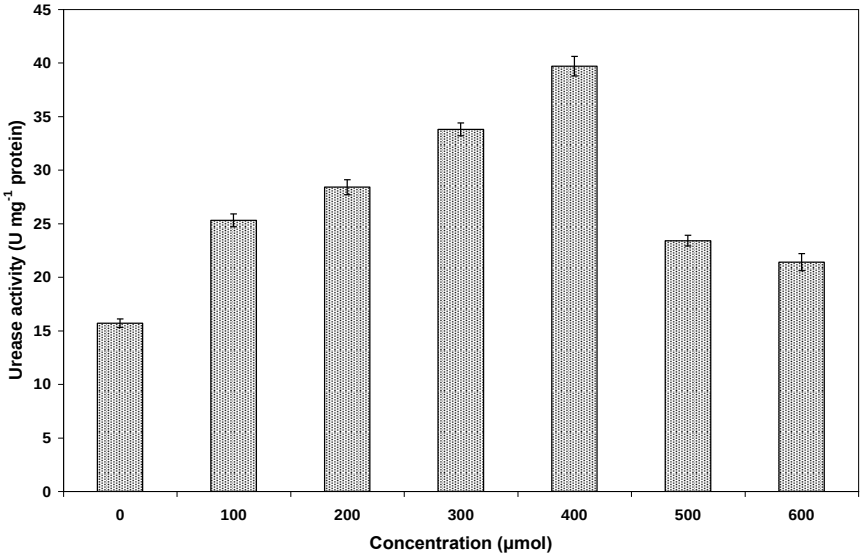

Fig 2: Effect of naphthaleneacetic acid on urease activity.

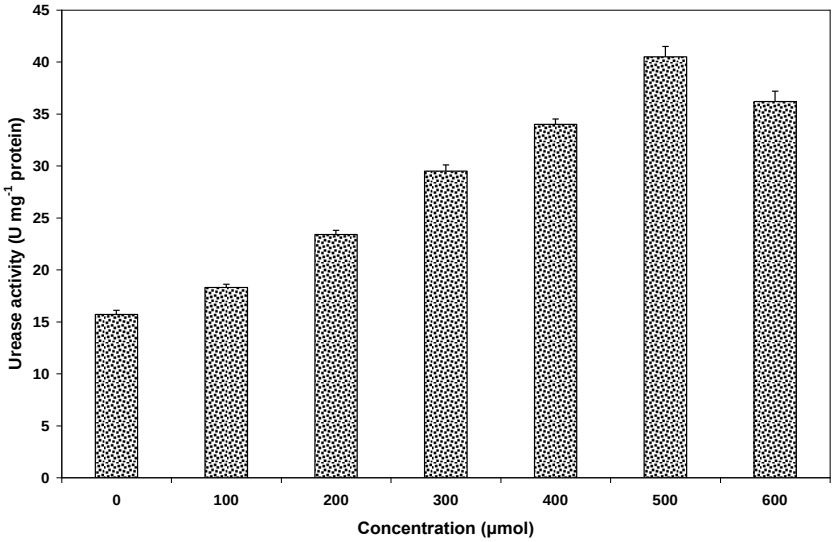

Fig 3: Effect of indoleacetic acid on urease activity

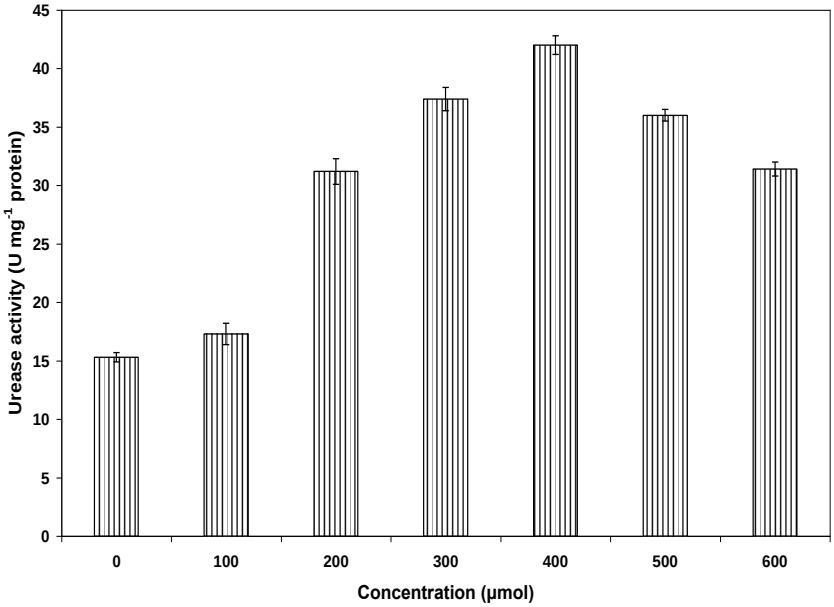

Fig 4: Effect of indolepropioic acid on urease activity

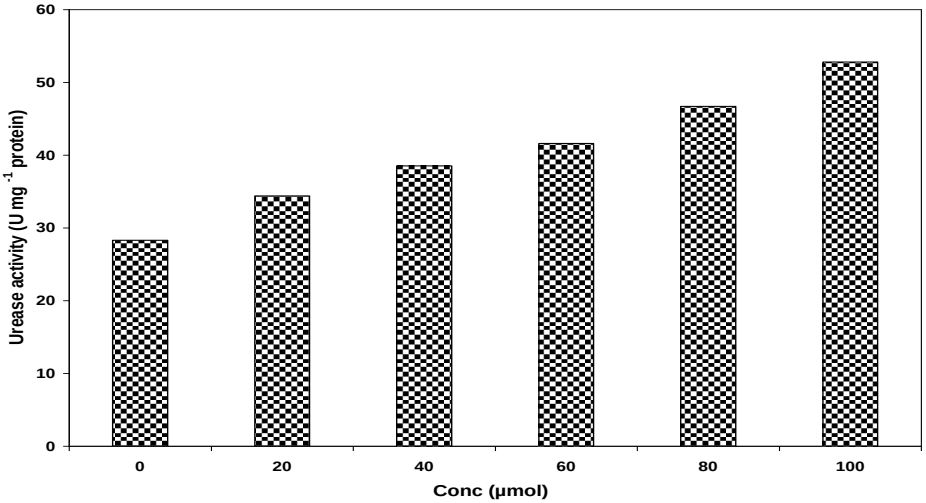

Fig 5: Effect of different concentration of zeatin on urease activity

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تأثير هرمونات النمو النباتية على نشاط انزيم اليوريز فى فلقات نبات القرع.

حامد محمد الشورى و لمياء فؤاد محمد السعيد.

قسم النبات-كلية العلوم-جامعة المنصورة.

هدف هذا البحث ال دراسة تأثير بعض هرمونات النمو مثل اندول حمض البيوتريك و اندول حمض الخليك و نفثالين حمض الخليك و اندول حمض البروبيونيك و الزياتين على نشاط انزيم اليوريزفى فلقلت نبات القرع المعزولة من بادرات نمت لمدة خمسة ايامز و اتضح من الدراسة ات معاملة الفلقات بكل من هذه المركبات الخمس قد ادت الى زيادة النشاط الانزيمى حيث كان التركيز الامثل لهذه المركبات كالاتى ١٠٠،٤٠٠،٤٠٠،٥٠٠،٥٠٠ميكرومول.