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Homarine promotes the Growth of Duckweed and Radish Seedlings

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Abstract

The effect of homarine (*N*-methylpicolinic acid) on the growth of duckeweed *Lemna paucicostata* 151 and radish seedlings *Raphanus sativus* L. were investigated. It was found that homarine promoted the growth of duckweed most strongly at 10^{-6} M and its flowering induction was not observed. The growth rate of treated plants was 2.1 times higher than that of the control. Homarine also promoted the growth of radish seedlings. The hypocotyl elongation at 10^{-4} M was 1.9 times longer than that of the control. It is postulated that the methyl group at the *N*-position of pyridine ring may be involved in the plant growth promotion by homarine.

Key Words: homarine • plant-growth promotion • duckweed • radish seedlings • picolinic acid

Introduction

Homarine (*N*-methylpicolinic acid) was named after American lobster *Homarus americanus* from which the compound was first isolated by Hoppe-Seyler in 1933[1]. The structure of homarine is shown in Fig. 1. This compound has been found in marine invertebrates[2-5], but not in insects, fishes, amphibians, and mammals[2]. Its physiological function was thought to regulate osmotic pressure in marine invertebrates[4]. Recently, homarine was found to inhibit metamorphosis of marine hydroid[6-8]. Netherton III and Gurin reported that homarine was biologically synthesized from succinyl CoA and glycine *via* 7 steps in shrimp muscles[9,10]. Furthermore, quite recently we found a homarine-synthesizing enzyme, i.e., picolinate methyltransferase, which converts picolinic acid into homarine in the presence of *S*-adenosyl-L-methionine in a cell-free extract of turban shell *Batillus cornutus*[11].

During the study of the effects of nicotinic acid-related compounds on the growth of plants, we found that trigonelline (*N*-methylnicotinic acid) and cinchomeronic acid (4-carboxynicotinic acid) strongly promoted the growth of duckweed[12] and radish seedlings[13,14]. Homarine is biosynthesized from picolinic acid after methylation with *S*-adenosyl-L-methionine. Picolinic acid is derived from α-amino-

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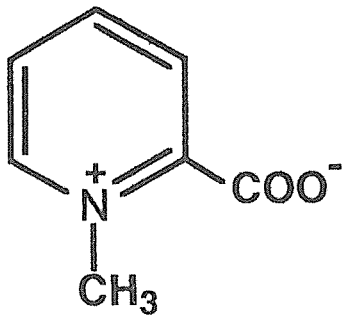

Fig. 1 Structure of homarine.

β-carboxymuconate ε-semialdehyde which is one of intermediates in the *de novo* biosynthetic pathway of NAD. Homarine is a structural isomer of trigonelline, which is the final metabolite of NAD in plants. Growth promoting effects of these compounds may closely relate to the biosynthesis and metabolism of NAD[15, 16].

In this study, we examined the effect of homarine on the growth of duckweed and radish seedlings. And finally we found the plant-growth promoting activity of homarine for the first time.

Materials and Methods

Materials Germ-free duckweed *Lemna paucicostata* 151 was a gift from Prof. Dr. Atsushi Takimoto, Faculty of Agriculture, Kyoto University. Radish seeds *Raphanus sativus* L. cv. Wase shijunichi were purchased from Atariya Farm, Chiba. Homarine was extracted from cuttlefish *Todarodes pacificus* and purified according to the methods described in the reference [9]. The purified product was identical with an authentic sample in all respects.

Plant growth bioassay The growth of duckweed[12] and radish seedlings[13] were assayed by the methods described previously.

Results and Discussion

The effect of homarine on the growth of duckweed at various concentrations is shown in Fig. 2. The effect was the strongest at 10^{-6} M and the growth rate was 2.1 times higher than that of the control. However, flowering induction of the plant was not observed at any concentrations studied.

The effect of homarine on the growth of radish seedlings at various concentrations is shown in Fig. 3. The hypocotyl was the longest at 10^{-4} M and hypocotyl elongation was 1.9 times larger than that of the control. However, leaf area, hypocotyl diameter, and root length were not affected significantly within the range of homarine concentrations tested.

As far as we know, the plant growth-promoting activity of homarine was found for the first time in this study. Trigonelline which is a structural isomer of homarine, also has the plant growth-promoting

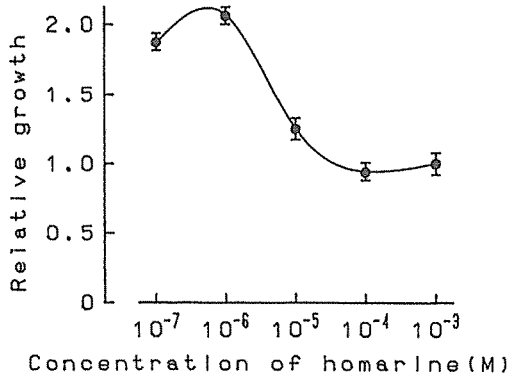

Fig. 2 Effect of homarine concentration on the growth of duckweed.
The plant was cultured with homarine at various concentrations for 7 days at 25℃ under continuous illumination at 4,000 lux according to the previous paper[12]. The relative growth means the growth rate relative to that of the control experiment without homarine. Values shown here are mean ± SE from 10 samples.

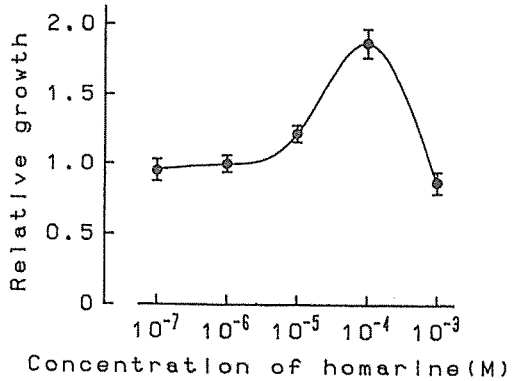

Fig. 3 Effect of homarine on hypocotyl elongation of radish seedlings.
Single radish seed was sown on a medium (2 ml with 0.8% agar, pH 5.5) containing homarine at different concentrations in a test tube (18×180 mm) with an aluminum cap and incubated for 10 days at 25℃ in the dark to the previous paper[13].
The relative growth means the hypocotyl elongation relative to that of the control grown in distilled water. Values shown here are mean ± SE from 10 samples.

activity[12, 13]. Although it is probable that the effect of homarine is principally the same as that of trigonelline, the effectiveness of homarine was comparatively higher than that of trigonelline. Nicotinic acid, which is formed after demethylation of trigonelline, is esily converted into NAD[16], but the demethylation of homarine in plants can not be detected in our experiments.

On the other hand, picolinic acid which is a demethylated compound of homarine, has flower-inducing activity[12, 17]. Furthermore, nicotinic acid, which is a demethylated compound of trigonelline and is a sructural isomer of picolinic acid, also has plant growth-inhibiting activity and flower-inducing activity[12, 17]. These results suggest that a methyl group on the *N*-position of pyridine ring is necessary to promote plant growth. Existence, biosynthesis, and metabolism of homarine in plants are not clear at present. Further investigation is now in progeress to elucidate the mechanism of plant growth-promotion by homarine.

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アオウキクサならびにダイコン幼植物に対するホマリンの生長促進効果

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アオウキクサならびにダイコン幼植物の生長に及ぼすホマリン（*N*-メチルピコリン酸）の影響について検討した。アオウキクサにおいて，ホマリンは10^{-6} M の濃度で生長を最も促進し，その相対生長度はコントロールに対して2.1倍であった。開花誘導効果は，いずれの濃度においても認められなかった。ダイコン幼植物において，ホマリンは10^{-4} M の濃度で下胚軸の伸長を最も促進し，その相対伸長度はコントロールに対して1.9倍であった。ニコチン酸関連化合物の構造と活性の相関に関する研究結果から，ホマリンの *N* 位に位置するメチル基が，ホマリンの植物に対する生長促進作用に関与していると思われる。