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AUSTRALIA–JAPAN RESEARCH AND COLLABORATION FORUM

Case studies on research partnerships

< Self-introduction of Kazumi Kita, Dr. Agr. >

Vice President/Executive Director for General Affairs
(Faculty of Agriculture, Department of Animal Science)

1984 BS (Nagoya University)

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1989 PhD (Nagoya University)

1998 Assist. Prof. (Nagoya University)

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Department of Biochemistry, Adelaide Children's Hospital
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1998 Assoc. Prof. (Nagoya University)

2006 Prof. (Iwate University)



Journal of Endocrinology

Influence of nutrition on hepatic IGF-I mRNA levels and plasma concentrations of IGF-I and IGF-II in meat-type chickens

K Kita, F M Tomas, P C Owens, S E Knowles, B E Forbes,
Z Upton, R Hughes¹ and F J Ballard

Cooperative Research Centre for Tissue Growth and Repair, PO Box 10065 Gouger Street, Adelaide, South Australia 5000, Australia and ¹Parafield Poultry Research Centre, South Australian Research and Development Institute, 336 Salisbury Highway, Parafield Gardens, South Australia 5107, Australia (Requests for offprints should be addressed to K Kita who is now at Laboratory of Animal Nutrition, School of Agricultural Sciences, Nagoya University, Chikusa-ku, Nagoya 464-011, Japan)

Abstract

We have examined the influence of nutrition on plasma IGF-I, IGF-II and IGF-binding protein (IGFBP) levels and on hepatic IGF-I gene expression in young meat-type chickens. Plasma IGF concentrations were measured by using RIA with recombinant chicken IGFs as standards.

In chickens fed the control diet containing 200 g/kg dietary protein *ad libitum* for 7 days, plasma IGF-I concentrations increased significantly from those found in the initial control group. Food restriction for either 4 or 7 days decreased plasma IGF-I by 30% from the initial control. When chickens were refed *ad libitum* for 3 days after 4 days of restricted feeding, plasma IGF-I levels recovered to those of the control birds fed *ad libitum*. In chickens eating a low protein diet (100 g/kg protein), the plasma IGF-I tended to be lowered but the decrease was not significant. Although the intensity of IGF-I and β -actin mRNA bands protected in the RNase protection assay was changed by nutrition, no statistical effect of nutrition on the ratio of IGF-I to β -actin was observed. The nutritional treatments had no effect on plasma IGF-II concentrations.

Western ligand blot and chromatographic analyses were used to investigate the influence of nutrition on IGFBP profiles. Both IGF-I and IGF-II ligands in the Western ligand blot revealed the most intense binding at 30 kDa for plasma obtained from chickens with restricted food intake. The 30 kDa band also appeared at a lower intensity in the group fed a low protein diet but not in any other groups. These observations were confirmed by neutral gel chromatography. The chicken IGF-II ligand revealed an intensely labelled band corresponding to 75 kDa and this was not affected by nutrition.

IGF-I and IGFBP concentrations in the plasma of young broiler chickens were influenced by nutritional state but IGF-II concentrations were not. The lack of a response in circulating IGF-II levels may have been due to the presence of high concentrations of a 75 kDa specific binding protein which did not respond to nutrition in this experiment.

Journal of Endocrinology (1996) **149**, 181–190

Introduction

The insulin-like growth factors (IGF-I and -II) found in chickens have been characterised and shown to be 70 and 66 amino acid polypeptides respectively (Dawe *et al.* 1988, Ballard *et al.* 1990, Kallincos *et al.* 1990). Recently some findings pointing to an important role for IGF-I in the control of growth and metabolism in chickens, as in mammals, have been reported. When exogenous IGF-I was added to the chicken embryo, growth and differentiation were stimulated (Girbau *et al.* 1987). After hatching, plasma concentration and hepatic gene expression of IGF-I increase rapidly with ageing, reach a peak before sexual maturity, and then decline (Huybrechts *et al.* 1985, Johnson *et al.* 1990, McGuinness & Cogburn 1990, Kikuchi *et al.* 1991). Plasma IGF-I levels are also respon-

sive to nutrition and are reduced in chickens fed low protein diets (Rosebrough *et al.* 1992a,b, Rosebrough & McMurtry 1993).

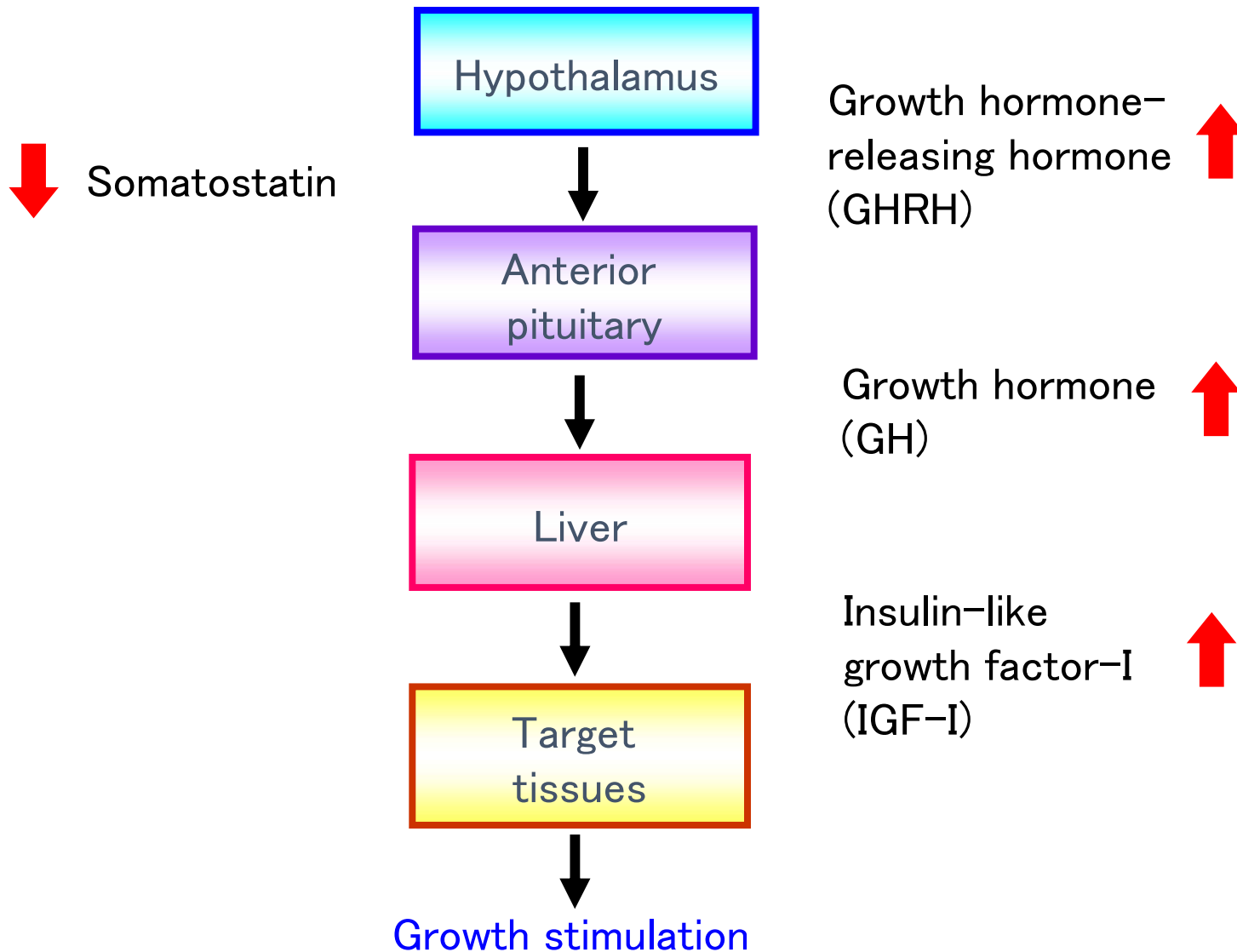
In contrast to the studies related to IGF-I, there is virtually no information on circulating IGF-II concentrations in avian species. Scanes *et al.* (1989) measured plasma concentrations of IGF-II in young chickens and found no correlation between IGF-II concentration and body weight. No data which allow assessment of the functional role of IGF-II in growth and metabolism in chickens have been reported. In this study, we have examined the influence of nutrition on hepatic IGF-I mRNA levels using a ribonuclease protection assay and on plasma IGF-I and -II concentrations in young meat-type chickens using an RIA with recombinant chicken IGFs as standards.



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GHRH – GH – IGF-I axis





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Influence of nutrition on hepatic IGF-I mRNA levels and plasma concentrations of IGF-I and IGF-II in meat-type chickens

Animals

Female broilers

Nutritional conditions

- 1) Ad libitum for 7 days (CP 20% diet) (A7)
- 2) Low protein (CP 10%) for 7 days (L7)
- 3) Restricted for 4 days (CP 20% diet) (A4)
- 4) Restricted for 7 days (CP 20% diet) (R7)
- 5) Restricted for 4 days and ad libitum
for 3 days (CP 20% diet) (R4A3)

Plasma IGF-I concentration

Radioimmunoassay

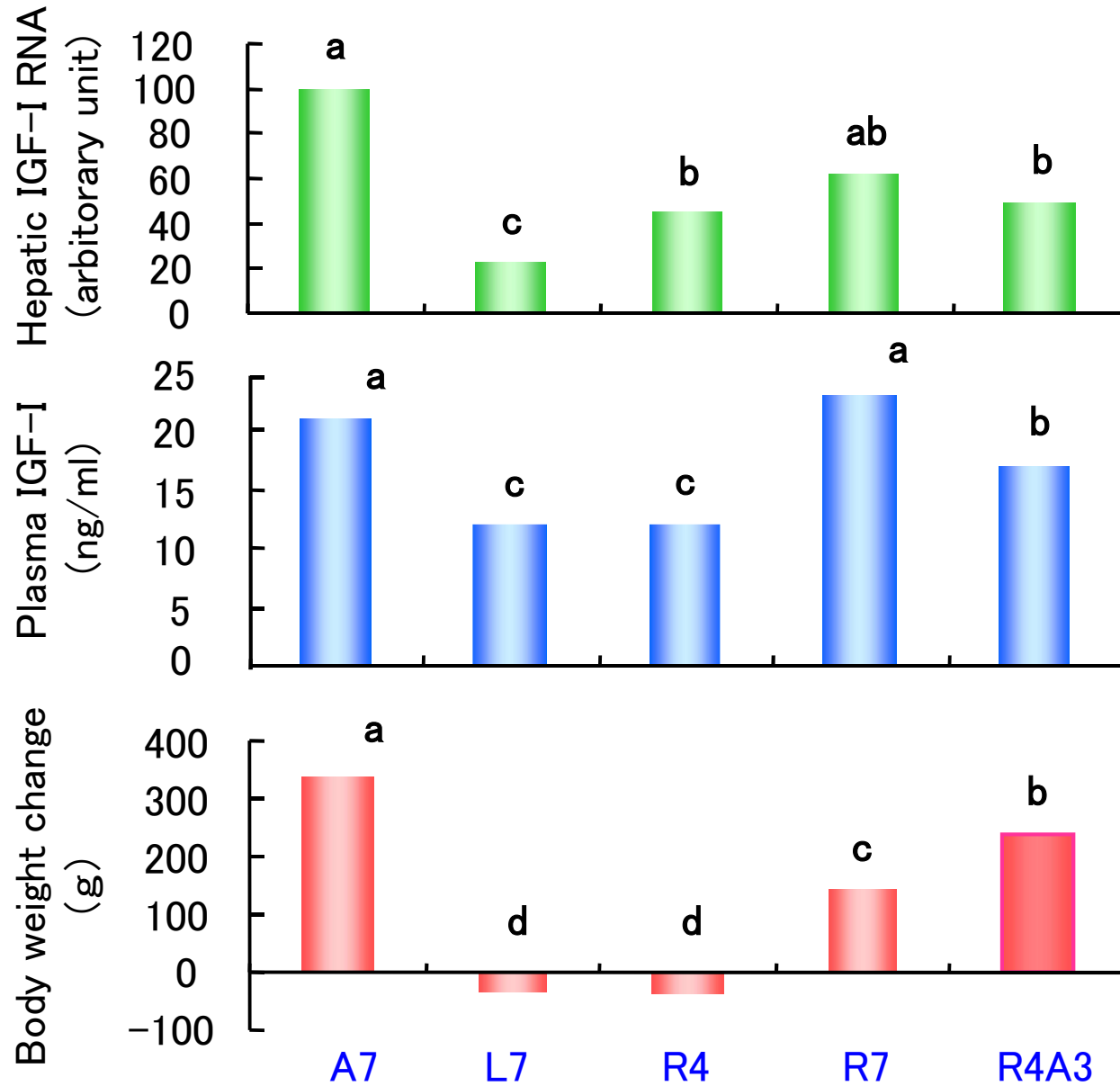
Hepatic IGF-I mRNA

Ribonuclease protection assay



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Nutritional Conditions



Hepatic IGF-I Gene Expression



Plasma IGF-I Concentration



Protein Metabolism



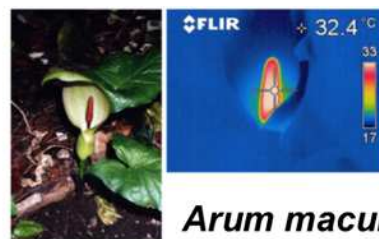
Growth Regulation



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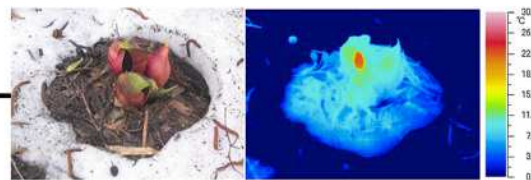
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Research on Plant Thermogenesis & Collaboration with Australian Researcher

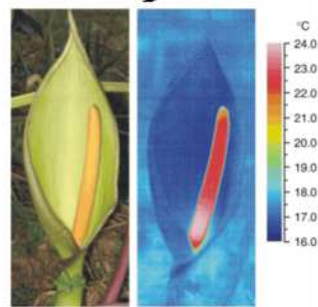


Arum maculatum

(Wagner et al., 2008, Ito et al., 2012)

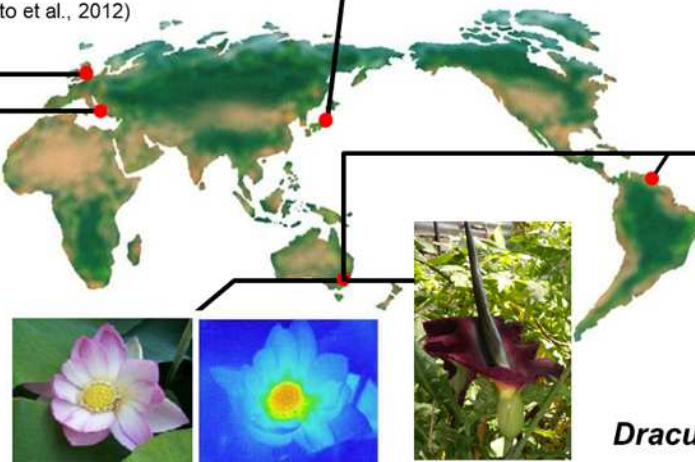


Symplocarpus renifolius (Kamata et al., 2009)



Arum concinatum

(Kakizaki et al., 2010, Onda et al., 2015)

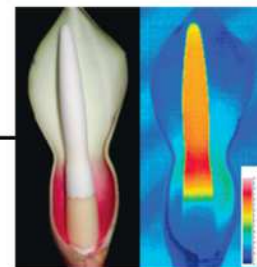


Nelumbo nucifera (Onda & Ito, 2007)



Dracunculus vulgaris

(Ito et al., 2013)



Philodendron

(Seymour & Gibernau, 2008)



Prof. Kikukatsu Ito
(Iwate University)



Prof. Roger Seymour
(University of Adelaide)



PRIME MINISTER'S EDUCATION ASSISTANCE PROGRAM FOR JAPAN 2011



Japanese students &
Prof. Roger Seymour



Prof. Kikukatsu Ito



Prof. Roger Seymour

- ✓ On April 22, 2011, Australian Prime Minister Julia Gillard announced an education program for Japan following the Great East Japan Earthquake.
- ✓ Prof. Kikukatsu Ito's team at Iwate University, Japan, studied mitochondria from thermogenic plants with Prof. Roger Seymour at the University of Adelaide.



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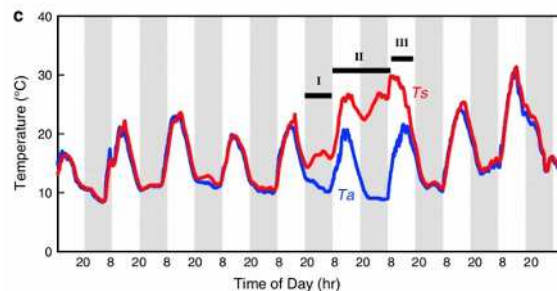
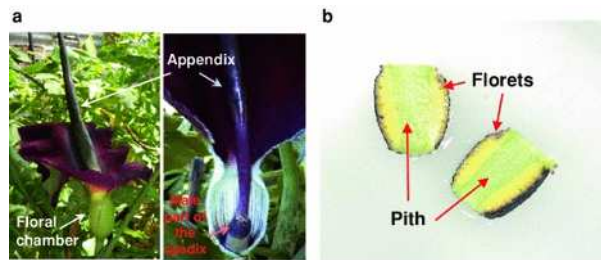
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Research Outcomes Published in Metabolomics

Metabolomics (2013) 9:919-930

Metabolite profiling reveals tissue- and temperature-specific metabolomic responses in thermoregulatory male florets of *Dracunculus vulgaris* (Araceae)

Kikukatsu Ito · Hideyuki Takahashi · Yui Umekawa · Tomohiro Imamura ·
Shuji Kawasaki · Takafumi Ogata · Yusuke Kakizaki · Roger S. Seymour



Our study uncovered significant metabolic shifts related to plant heat-generation.



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<https://doi.org/10.1093/plphys/kiad059>

PLANT PHYSIOLOGY 2024, 195: 1561–1585

Plant Physiology®

2024

Gene expression and metabolite levels converge in the thermogenic spadix of skunk cabbage

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The author responsible for distribution of materials integral to the findings presented in this article in accordance with the policy described in the Instructions for Authors (<https://academic.oup.com/plphys/pages/General-Instructions>) is Kikukatsu Ito (kikuito@iwate-u.ac.jp).

Abstract

The inflorescence (spadix) of skunk cabbage (*Symplocarpus renifolius*) is strongly thermogenic and can regulate its temperature at around 23 °C even when the ambient temperature drops below freezing. To elucidate the mechanisms underlying developmentally controlled thermogenesis and thermoregulation in skunk cabbage, we conducted a comprehensive transcriptome and metabolome analysis across 3 developmental stages of spadix development. Our RNA-seq analysis revealed distinct groups of expressed genes, with *selenium-binding protein 1/methanethiol oxidase* (SBP1/MTO) exhibiting the highest levels in thermogenic florets. Notably, the expression of *alternative oxidase* (AOX) was consistently high from the prethermogenic stage through the thermogenic stage in the florets. Metabolome analysis showed that alterations in nucleotide levels correspond with the developmentally controlled and tissue-specific thermogenesis of skunk cabbage, evident by a substantial increase in AMP levels in thermogenic florets. Our study also reveals that hydrogen sulfide, a product of SBP1/MTO, inhibits cytochrome *c* oxidase (COX)-mediated mitochondrial respiration, while AOX-mediated respiration remains relatively unaffected. Specifically, at lower temperatures, the inhibitory effect of hydrogen sulfide on COX-mediated respiration increases, promoting a shift toward the dominance of AOX-mediated respiration. Finally, despite the differential regulation of genes and metabolites throughout spadix development, we observed a convergence of gene expression and metabolite accumulation patterns during thermogenesis. This synchrony may play a key role in developmentally regulated thermogenesis. Moreover, such convergence during the thermogenic stage in the spadix may provide a solid molecular basis for thermoregulation in skunk cabbage.

Introduction

Thermogenesis, an endogenous metabolic heat-production process in specific organs or tissues, is a physiological phenomenon observed across various angiosperm species. Plant thermogenesis is often associated with the release of an odor or scent from the thermogenic organ. Notably, this mechanism attracts pollinators during the reproductive stages and concurrently creates a warm habitat advantageous for energy conservation in visiting insects (Meeuse and Raskin 1988; Seymour et al. 2003; Edl et al. 2022). A number of plants, including the

titan arum (*Amorphophallus titanum*), dead-horse arum (*Helicodiceros muscivorus*), Crete arum (*Arum concinatum*), and lords-and-ladies (*Arum maculatum*) from the Araceae family and the sacred lotus (*Nelumbo nucifera*) and American lotus (*Nelumbo lutea*) from the lotus family, exhibit thermogenesis, although the extent of their temperature increase and the timing of this phenomenon vary among species (Wagner et al. 2008; Barthlott et al. 2009; Kakizaki et al. 2010; Dieringer et al. 2014; Zheng et al. 2022).

Skunk cabbage (*Symplocarpus renifolius*) is a thermogenic species that flowers in late winter or early spring, a period

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Research Article

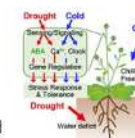
Most read

REVIEW ARTICLE

Regulatory networks in plant responses to drought and cold stress

June-Sik Kim and others

Drought and cold represent distinct types of abiotic stress, each initiating unique primary signaling pathways in response to dehydration and temperature changes, respectively. However, a convergence at the gene regulatory level is observed where a common set of stress-responsive genes is activated to mitigate the impacts ...



#1

RESEARCH ARTICLE

Gene expression and metabolite levels converge in the thermogenic spadix of skunk cabbage

Haruka Tanimoto and others

Comparative transcriptomic and metabolomic analyses unveil the crucial role of gene and metabolite expression convergence in the developmentally regulated thermogenesis of *Symplocarpus renifolius*.



#2

This paper achieved second place in the journal's Most Read articles worldwide.

Cooperative research between Iwate University (Morioka) (Ken-ichi Kimura) and La Trobe University (Melbourne) (Don. R. Phillips)



Prof. Ken-ichi Kimura
(Iwate University)

<Research fund>

- ◎JSPS Bilateral Programs (2004-2005, 2009-2010)
 - Biochemical studies on the mechanism of action of a new anticancer drug, barminomycin
 - Study on the anticancer mechanism based on analysis of barminomycin-DNA binding proteins
- ◎JSPS Invitational Fellowships for Research in Japan (Short period)(2009)

<Publications>

◎K. Kimura, D. M. S. Spencer, R. Bilardi, L. P. Swift, A. J. Box, R. T. C. Brownlee, S. M. Cutts and D. R. Phillips, Barminomycin, a model for the development of new anthracyclines. *Anti-cancer Agents in Med. Chem. (Review)*, 10, 70-77 (2010). Other 4 regular papers (2003, 2008, 2010, 2012).

Barminomycin is 1,000 times more active than the clinical drug adriamycin, and it has been discovered that barminomycin covalently bind to the 2 amino group of guanine in the 5'-GC-3' sequence of the DNA via a carbinolamine structure in cancer cells.

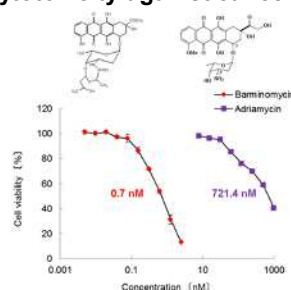
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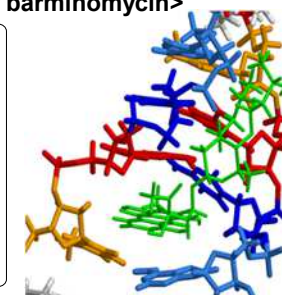
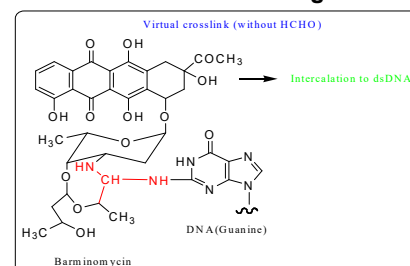
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<Cytotoxicity against cancer cells>



<DNA binding mode of barminomycin>

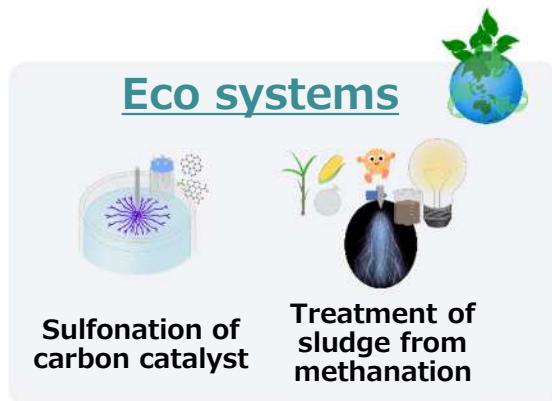


New applications of electrostatics and discharge plasma



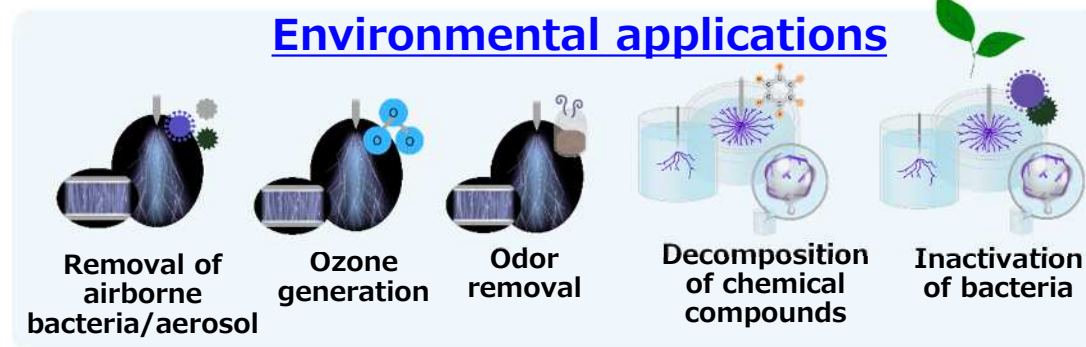
Assoc. Prof. Katsuyuki Takahashi
(Iwate University)

Eco systems



Sulfonation of carbon catalyst Treatment of sludge from methanation

Environmental applications



Removal of airborne bacteria/aerosol Ozone generation Odor removal Decomposition of chemical compounds Inactivation of bacteria

Agricultural applications



Ethylene removal for keeping freshness Water treatment for hydroponics Inactivation of bacteria Soil treatment Enzyme control Acceleration of dry process Inactivation of bacteria Destruction of inner cellular Improvement of mushroom production



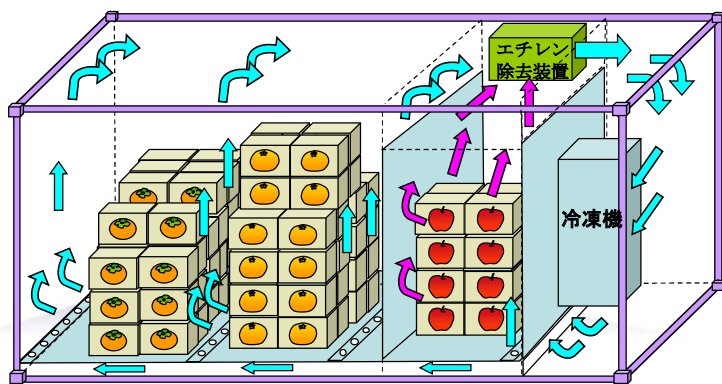
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Preservation

- Quality loss by ethylene, a plant hormone released by respiration
- Low temperature injury

Mixed loading in a transportation container



Inside of container

Ethylene release



Ripening:
Damage of fruit,
Quality loss

Plasma treatment



- High speed treatment by high density radicals
- High safety by removing by products using catalyst
- High maintainability
- Compact and light

Reactions in plasma :





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Thank you for your attention.