stored at −20 °C until assayed. The plasma LH and FSH concentrations were determined by the double antibody RIA method, using the NIAMDD kits[11] and following the directions supplied with the kits with minor modifications. Results were expressed in terms of the NIH-LH-S1 reference preparation for LH and the NIAMD-Rat FSH-RP-1 reference preparation for FSH. Statistical evaluation of the differences between means was performed using Student's t-test.

*Results and discussion.* As can be seen from the figure, which summarizes the results obtained in this work, mean plasma concentrations of LH and FSH 2 days following EB injection (i.e., at zero time) were similar among all 4 groups of chronically ovariectomized rats that were subjected to stereotaxic manipulation, ranging from 2.3±0.16 to 3.3±0.30 ng/ml for LH and from 883±29.5 to 948±49.9 ng/ml for FSH. The values did not differ significantly from those found in ovariectomized controls (4.9±0.35 ng/ml for LH and 930±45.4 ng/ml for FSH). As should be expected from earlier work on the stimulatory action of gonadal steroids on gonadotropin release[12], the systemic injection of P at zero time induced 6 h later in the spayed EB primed rats a profound elevation of plasma LH (22.2±3.85 ng/ml) and FSH (1393±76.2 ng/ml). The values were again low by the next morning, and remained low (LH) or increased slightly (FSH) in the afternoon. Similar triggering effects of P on LH and FSH release were observed in sham-operated animals, in which respective values for LH and FSH, 6 h later, were 24.2±6.04 ng/ml and 1583±132.1 ng/ml. On the other hand, LH surge was completely blocked (3.0±0.42 ng/ml) by crystals of P implanted into the MRF, and partially but significantly inhibited by inserting the tip of the cannula alone into this brain structure (9.1±1.96 ng/ml and 11.1±2.19 ng/ml for groups c and d, respectively). The latter effect is attributable to non-specific damages to the brain tissue, produced by the penetration of the cannula. Contrary to the observed effects on LH release, however, neither P crystals nor cannula alone, when inserted into the MRF, were significantly effective in altering FSH release, as compared to that in ovariectomized EB-P-treated controls. Thus, 6 h following P treatment, the FSH levels in the circulation of animals from groups b, c, d and e were, respectively, 1227±65.0, 1273±69.8, 1494±92.3 and 1583±132.1 ng/ml, as compared to 1393±76.2 ng/ml for group a. These findings suggest that the investigated area of the MRF is a site of the inhibitory feedback action of P in the control of LH release, but not of FSH release. The dissociation of LH from FSH release, observed in this work as well as in other laboratories and under different experimental conditions[6, 13–14], which cannot be explained by direct action of gonadal steroids on the anterior pituitary to change its responsiveness, keeps the question of the existance of 1 or 2 different releasing hormones still open.


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## A dramatic role of terpenoids in increasing rice production

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*Summary.* Earlier work has established the role of terpenoids and their derivatives as a new group of plant growth regulators. Some of these terpenoids have now been tested in field trials and have been found to increase rice production significantly.

A major breakthrough in the rice improvement program has been achieved by the introduction of semi-dwarf, tropical indica plant type varieties. These semi-dwarf varieties had a much higher yield than the conventional tall varieties because of their improved plant type, and increased response to heavy doses of nitrogen. The improved production technology worked out for these varieties further made it possible to realize the potential yield of these varieties (10 t/ha) particularly in Punjab (India).

The rice breeders are continuing their global efforts towards further genetic improvement of the yield potential of rice varieties by incorporating disease and insect-pest resistance. Considerable success has been achieved in this, but no variety has so far been released which could potentially outyield the high yielding varieties like IR 8 and PR 106. The agronomists and physiologists, on the other hand, are trying hard to raise the yield level of the present high yielding varieties by innovating new production technology. To raise the yield levels beyond the present varietal potential of 10 t/ha is a real challenge for the rice scientists in future. The productivity can be increased either by breeding more high yielding varieties, which is a time-consuming method or by the use of new growth regulators.

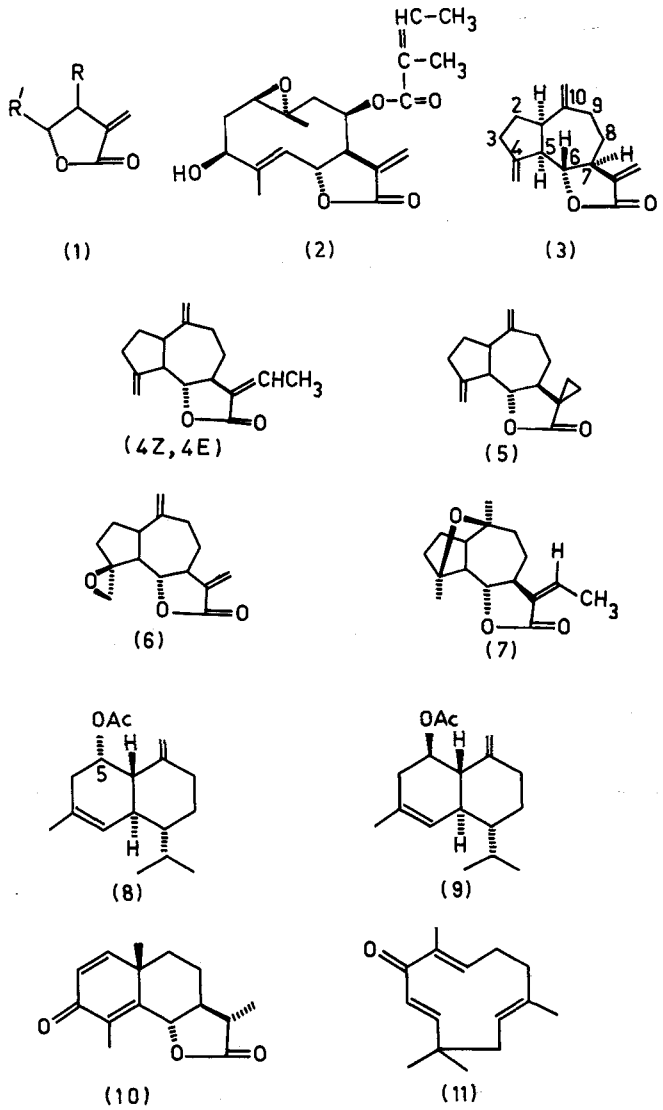
The biological potentialities of terpenoids[3] in general and

Performance of PR-106 in seed and seedling treated trials

| Compound | Seed treatment trial Average yield (kg/ha) | Percent increase over control | Seedling treatment trial Average yield (kg/ha) | Percent increase over control |
|---|---|---|---|---|
| Santonin (**10**) | 10271* | 10.04 | 9927 | 14.17 |
| Zerumbone (**11**) | 10500* | 12.50 | 9927 | 14.17 |
| $C_{16}$-Guaianolide (**4Z**) | 10000 | 7.14 | 9347 | 7.50 |
| Control | 9333 | - | 8695 | - |
| C.D. (0.05) | 903 | | 1352 | |

*Statistically significant at 5% level of significance.

of terpenoid lactones in particular as plant growth regulators have been explored intensively. Sesquiterpene lactones have recently become interesting to both chemists and biologists, because of their structural diversity, chemical transformations, physiological activities and importance as genetic markers in various plant taxa. It has been established that the physiological activity of the terpenoid lactones is due to the conjugated exomethylene moiety (**1**) which is most probably essential[4]. The germacranolide heliangine (**2**), a sesquiterpene lactone isolated from the leaves of *Helianthus tuberosus,* and the guaianolide dehydrocostus lactone (**3**) isolated from costus roots *Saussurea lappa,* which contain this moiety, are biologically active. Previous work[5] from our laboratory showed that some $C_{16}$terpenoid $\gamma$-lactones in which conjugation is of a different type are physiologically more active than the parent $\alpha$-methylene-$\gamma$-lactones. Thus the $C_{16}$ guaianolides (**4Z**) and (**5**) are biologically more potent than their parent dehydrocostus lactone. It is observed that the double bond isomers have different biological activities[6]. These differences of stereochemistry about the C=C reflect their significance in the case of conjugated $\gamma$-lactones which act as plant growth regulators. The root-forming potential of the isomerized product (**4E**) underwent a significant decrease when compared with its parent compound (**4Z**). The placement[7,8] of either an epoxy group at C−4 or an ether linkage at C−4, C−10 affects favorably the plant growth activity of the parent conjugated $\gamma$-lactone moieties in the case of guaianolides[3,9,10]. Among the huge number of conjugated lactones and their oxygenated derivatives tested in our laboratory, compounds (**6** and **7**) are biologically the most active. A dramatic effect of stereochemistry on the plant growth activity of the cadinane group of terpenoids has also been recorded[10]. Significantly, it was found that epi-khusinol acetate (**8**) was almost 4 times more active at 20 mg/l when compared with khusinol acetate (**9**). Cross conjugated ketones[4] like Santonin (**10**) and Zerumbone (**11**) have been reported as a new group of plant growth regulators. Santonin is the sesquiterpene lactone based on the eudesmane skeleton belonging to the keto type. It occurs extensively in various species of the genus *Artemisia* and has commanded the attention of organic chemists for well over a century. This is chiefly due to the vast array of novel and interesting chemical and photochemical transformations associated with this sesquiterpenoid. Zerumbone is a crystalline monocyclic sesquiterpene ketone which is isolated from the rhizomes of *Zingiber zerumbet,* which grow abundantly in the west coast of India. The activity of zerumbone is greater[9] than that of with indole acetic acid (IAA).




Naturally occuring Terpenoids and their derivatives **1**, $\alpha$-Methylene-$\gamma$-lactone moiety; **2**, heliangine; **3**, dehydrocostus lactone; **4** and **5**, $C_{16}$-Guaianolides (**E&Z** isomers) derived from dehydrocostus lactone; **6**, C-4 epoxy derivative of dehydrocostus lactone; **7**, C-4, C-10 ether of **4Z** isomer; **8**, epi-khusinol acetate; **9**, khusinol acetate; **10**, santonin; **11**, zerumbone.

The major biological effect observed with these compounds was prolific rooting in stem cuttings of *Phaseolus aureus* and some other plant species. An increase in the root formation (both number and length) is likely to affect the physiology and morphology of the plant which in turn could affect the total yield. With these views in mind we set out to screen these compounds in field trials to detect increase in production on PR-106, the widely grown variety of rice.

Two separate field trials were laid out at Punjab Agricultural University, Ludhiana (India) one by dipping the seeds in 100 ppm solution and the other by dipping the roots of 35 days old nursery plants in 25 ppm solution of the compounds for 24 h in each case. The solutions were prepared by dissolving the weighed quantity of the com-

pounds in small quantity of alcohol and making the desired volume with warm water. The 35 days old nursery plants from treated seeds and the root-treated nursery plants were planted in 3 and 4 replications respectively in the field. The plant-to-plant and row-to-row distances were kept at 15 and 20 cm respectively. The plot size in the seed-treated trial was 8 $m^2$, whereas it was 3.45 $m^2$ for the seedling-treated trial. The yield was determined on a dry weight basis and the results obtained with different compounds are presented in the table.

Interestingly, we report that Santonin (**10**), Zerumbone (**11**) and $C_{16}$-Guaianolide (**4Z**) enhanced the rice yield. The highest increase in yield over control, 12.50 and 14.17%, was shown by Zerumbone, followed closely by Santonin which showed 10.04 and 14.17% increase in yield over control in seed and seedling treated trials respectively. To the best of our knowledge this is the first report of its kind. The authors are hopeful, therefore, that these terpenoids may be in the future prove to be valuable agrochemicals for further improving rice production.

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## Elimination kinetics of iopamidol, a new water soluble nonionic radiographic contrast medium, analyzed by radioactivation

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*Summary.* We have studied the elimination kinetics of iopamidol employing radioactivation and radiochemical separation. This method offers the advantage of guaranteeing absolutely no interference by radiation with tissue distribution or elimination kinetics of the analyzed compound. Our results show that iopamidol does not cross the blood-brain barrier, has no effect on thyroid iodine uptake and does not accumulate in the liver.

Modern radiographic contrast media (RCM) are derived from benzoic acid, into which 3 iodine atoms are introduced in the 2-4 and 6 positions of the aromatic ring. The carboxyl group has been salified to obtain soluble products which unfortunately possess some toxicity. Studies achieved to correct this toxicity have shown that solubility, viscosity, tolerability, elimination and radioopacity are linked to substitutions in the 6 positions of the ring[3]. Certain salt derivatives may cause a selective toxic action on some parenchymas, as in the case of the toxicity at the myocardial level, where the need for maintaining a physiological concentration of $Na^+$ ions is especially important. In any case these RCM dissociate in solution to form anions and cations affecting the osmolarity of the solution. The resulting high osmotic pressure has been cited by several authors as a probable cause of diverse side effects seen in their clinical use, including hemodynamic changes[4,5], agglutination of red blood cells and endothelial membrane permeability changes[6], and neurotoxic effects[7].

These distressing side effects prompted the search for water soluble 'nonionic' RCM[8]. Recently, the synthesis of iopamidol, a new water soluble 'nonionic' radiographic contrast medium, has caused great interest, owing to its satisfactory viscosity and solubility characteristics. The analysis of iopamidol solutions has revealed a further unexpected advantage. Lower osmolarity values than expected have been measured, possibly due to the spontaneous formation of polymolecular aggregates. Therefore the use of iopamidol is a decisive step forward in neuroradiology, where osmolarity changes can cause serious consequences[9]. Looking at this, we decided to study the elimination kinetics of iopamidol from the most significant tissues by radioactivation. This method is the most sensitive presently available and in contrast to methods employing previously activated molecules, it offers the advantage of guaranteeing that there will be absolutely no interference by radiation with either the distribution into tissues, or the elimination kinetics of compounds studied. The substance to be analyzed is activated by a capture (n, $\gamma$) reaction in a nuclear reactor only as the final step of the experiment, i.e. after animals have been treated and sacrificed. After radioactivation it is possible qualitatively and quantitatively to discriminate between various chemical elements by means of a $\gamma$-spectrometry Ge-Li detector.

In our experiments iodine was measured as an index of the iopamidol presence in tissues. The analytical determination of iodine as an index for measurement of modern RCM levels in biological materials is universally accepted, since iodine is covalently bound to the benzene ring. Studies done in vivo on the metabolism of these RCM in different mammals, including man, have not demonstrated the presence of any enzyme capable of breaking the bond between