Indian J. Plant Physiol., Vol. 10, No. 2, (N.S.) pp. 168-170 (April-June, 2005)

SHORT COMMUNICATION

GLUCOSIDE CONTENT AND ITS ACCUMULATION IN BACH (*ACORUS CALAMUS* LINN.) AS INFLUENCED BY NITROGEN AND PHOSPHORUS APPLICATION

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Received on 26 Feb. 2004, Revised on 13 July, 2005

An experiment was conducted during 1999-2000 and 2000-2001 to assess the effects of different levels of nitrogen and phosphorus on total glucoside accumulation in rhizomes and roots of bach (*Acorus calamus* Linn.). Results revealed that application of nitrogen and phosphorus @ 150 kg/ha each led to maximum glucoside accumulation in rhizomes (3.29%) and roots (5.19%), which was higher by 225% and 192%, respectively compared to control. Glucoside accumulation was more in roots as compared to rhizomes with increase in nitrogen and phosphorus doses.

Key words: Acorus calamus, **glucoside, nitrogen, phosphorus**

Herbal Materia Medica emphasizes that active principle content of medicinal plant has relevance due to their action, rather than the crude drug. Chief active principles are alkaoids, glucosides, saponins, neutral principles, resins, volatile oils, and gums. Gluosides are generally non-nitrogenous substances found in plants. These contain carbon, hydrogen and oxygen. A few have nitrogen or sulphur also in addition to C, H and O. Glucosides have many important medicinal properties.

Acorus calamus L., a herbaceous medicinal plant (family –Araceae), is commonly known as sweet flag in english and bach, ghorabach, safed bach in hindi. It is, known as calamus in international trade. This plant is found almost throughout India upto an altitude of about 2000 m, growing mainly in marshy or moist situation of northern and eastern Himalayas. It is now commercially cultivated in few states of Indian sub-continent, *viz*. M.P., U.P., Bihar, Kashmir, Manipur, Nagaland, W. Bengal and Karnataka states (Singh *et al*. 1990, Jain 1994). The dried rhizomes of the plant constitute the drug 'Calamus'. It has wide therapeutic uses, *viz.* carminative, tonic and stimulant to central nervous system (Singh *et al*. 1990). Due to its essential oil (calamus oil) it acts as an expectorant, by promoting flow of bronchial secretions thus it is useful herbal drug in asthma. It is also used as remedy for flatulent colic. It also contains tannins and is therefore used in controlling diarrhoea and dysentery. In addition it is emetic and heavier doses can cause violent vomiting (Jain 1994). The oil from the rhizomes is a good nerve stimulant and the essential oil-free alcoholic extract shows marked sedative and analgesic (*i.e.* pain relieving) properties which causes moderate depression in blood pressure thus justifies its uses in treatment of mental diseases also (Jain 1994, Singh *et al*. 1990).

Root and rhizomes bark contains 1.5-3.5 per cent essential oil. Major part of this essential oil is asaryl aldehyde which has two active elements called A-asarone and B-asarone. Besides, it contains acorin (a pungent glycoside), eugenol, caffein and small amount of alkaloids (Sharma 2001). Glycoside reported from plant is flavone diglycoside called luteolin 6, 8 –C- diglycoside (Husain *et al*. 1992). This plant is widely used by pharmaceutical industries. Thus, there is need to cultivate this plant commercially. Development of efficient agro-technique is first step towards commercial cultivation of any medicinal plant. Optimization of nutrient requirement is

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one of the important factors in agro-techniques development. There is need to investigate impact of major nutrients (N and P) uptake on synthesis and accumulation of glycoside. Present investigation is the beginning to understand the same.

A field experiment was carried out during the rainy seasons of 1999-2000 and 2000-01 at the experiment field of JNKVV, Jabalpur. The soil type of the experimental area was clayey loam having pH value 6.5. with 20, 30 and 195 kg/ha available N, P_2O_5 and K_2O respectively, prior to planting. Rhizome slips with two live buds had been planted at 50 x 50 cm spacing in 5 x 5m plots in first week of July. Half the dose of nitrogen and full dose of phosphorus were applied as basal. Remaining half quantity of nitrogen was applied one month post transplanting. Twelve treatments as given in Table 1 were applied in three replications in RBD.

Randomly selected 10 plants per plot were taken for chemical analysis after 12 months of transplanting. Oven dried samples were partitioned into roots and rhizomes and finally grounded in mechanical grinder separately. Chemical analysis was carried out as per procedure suggested by Trim (1955) for extraction and crystallization of sparingly soluble flavones and flavonols glucosides.

The material was dried and powdered. 5 g sample was extracted by boiling in 40 ml water. The solution was filtered hot and then cooled until tepid and then rapidly extracted with twice of its volume of ether so as to remove the chlorophyll and other lipid soluble material, which interfere in the separation of glycoside. Unshaken glucosides were crystallized on cooling. Higher the glycoside content in solution faster is the crystallization and *vice-versa*. The crude crystalline product was filtered. It was re-crystallized in boiling methanol amounting five times to the previous volume. Left over glucoside in the filtrate was precipitated by adding distilled water. Process was repeated till precipitation discontinued. Total precipitated glucoside was finally crystallized by boiling the solvent in quantity of distilled water two hundred times to that of solvent volume. Crystallized glucoside was finally weighed after cooling. The crude crystalline product was again refluxed with methanol and recrystallized as above to remove the impurities. Final crystals were weighed. The percentage of total glucosides was calculated by formula given below:

$$\text{Total glucosides in sample (\%)} = \frac{\text{Weight of final crystallized glucoside}}{\text{Weight of dried sample}} \times 100$$

The results revealed significant increase in per cent glucoside accumulation in rhizome (3.290%) due to application of nitrogen and phosphorus @ 150 kg/ha each (T_{12}) when compared to control (1.460%). Thus supplementing nitrogen and phosphorus nutrients through inorganic sources resulted in increased glucosides accumulation in rhizome. Study revealed that inorganic nitrogen and phosphorus individually and in combination enhanced glucoside accumulation in rhizome through carbohydrate and protein metabolism (Table 1). As glucosides are made up of glucose (carbohydrate) attached to non-sugar part (David *et al*. 1972), phosphorus increased glucosides accumulation through their role in energy transfer *via* ATP and ADP. Thus

Table 1. Effect of nitrogen and phosphorus application (kg/ha) on total glucoside content (% on dry weight basis) in rhizome and roots of Bach.

S.No.	Treatments	Glucoside in rhizome (%)	Glucoside in roots (%)
1	T_1 (N_0P_{50})	1.46	3.20
2	T_2 (N_0P_{100})	1.84	3.24
3	T_3 (N_0P_{150})	1.18	3.63
4	T_4 ($N_{50}P_{50}$)	1.86	3.36
5	T_5 ($N_{50}P_{100}$)	1.93	3.34
6	T_6 ($N_{50}P_{150}$)	2.07	3.45
7	T_7 ($N_{100}P_{50}$)	2.13	3.52
8	T_8 ($N_{100}P_{100}$)	2.50	3.72
9	T_9 ($N_{100}P_{150}$)	3.07	3.85
10	T_{10} ($N_{150}P_{50}$)	2.35	4.60
11	T_{11} ($N_{150}P_{100}$)	3.15	4.78
12	T_{12} ($N_{150}P_{150}$)	3.29	5.19
	SEm ±	0.14	0.04
	CD (P=0.05)	0.29	0.12

phosphorus had indirect but effective role in glucoside synthesis and accumulation.

Accumulation of total glucosides in roots followed similar trend to that of rhizome (Table 1). There was significantly higher glucoside accumulation (5.190%) in roots when N and P were applied @ 150 kg/ha each (T_{12}) when compared to control. It is apparent (Table 1) that application of nitrogen increased comparatively more glucoside percent both in roots and rhizome than phosphorus. Hence significance of phosphorus was next to nitrogen. From the investigation, it is concluded that nitrogen and phosphorus application @ 150 kg/ha each is the best combination to achieve maximum glucoside yield in bach (*Acorus calamus* L.).

Data revealed that roots were the major sink for glucoside accumulation amounting to 50–90% higher active principle than rhizome. Normally roots are thrown as useless plant part and the rhizomes are only marketed. Present study reveals that roots have glucosides content higher than rhizome. Hence, root should also be processed for value addition and marketing. This would lead to higher economic returns per hectare.

ACKNOWLEDGEMENT

Authors are thankful to All India co-ordinated Research Project on Medicinal and Aromatic Plants, College of Agriculture, Indore for all kind of support given for chemical analysis work. We are also thankful to Dr. J.P. Tiwari, Ex-Prof. & Head, Deptt. of Plant Physiology, JNKVV, Jabalpur for his guidance in conducting field trial of the experiment.

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