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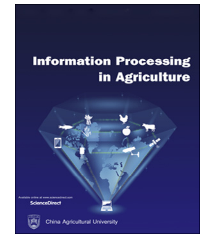

Effect of gravistimulation on amino acid profile of pea, rice, corn, wheat during early growth stages

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The amino acids are one of the major cellular components of plants, which are involved in different metabolic pathways. In present study, effect of artificial gravistimulation on amino acid profiles of pea, rice, corn, wheat during early growth stages was investigated. One-axis clinostat was used for gravistimulation application, which was applied at embryonic stage. Amino acid profile was measured in 10-days old seedlings of pea, rice, corn and wheat cultivars. The effect of clinostat rotation was also evaluated under salt stress and MS medium supplement. Germinated pea, rice, corn and wheat seedlings were grown under the gravity condition for specific time interval. Corn and wheat seeds showed slow germination as compared to pea and rice cultivars. The rate of amino acid formation under gravity condition was significantly higher than control (un-treated seedling). The variation in amino acid profile of pea, rice, corn and wheat cultivars vary deferentially. Results revealed that gravistimulation applied through clinostat has positive effect on amino acid profile in plant tissue and future studies should be focused on growth, biochemical, physiological at lateral stages of growth.

1. Introduction

Scientists demonstrated that the clinostat has ability to simulate the growth of plants. A clinostat continually rotates plant through 360° to eliminate a set direction for gravity, which also prevents the hormone, auxin, from accumulating on one side of the stem or roots. As a result, the clinostat may induce unusual growth in seedlings. Roots may grow toward stems, and stems may grow horizontally, rather than upwards under gravity effect [1].

The microgravity conditions can be produced by free fall or parabolic flight on earth, however, the effect of microgravity generation is small and resultantly, changes in plant growth and morphogenesis may change negligibly. A clinostat with

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a horizontal axis, produce unilateral gravity, which influence the plant growth and morphogenesis significantly. Microgravity influence the cellular functions and cellular components under constant microgravity [2]. This technique is highly interesting and attracted the attention of researchers, which opens new horizon in understanding the effect of microgravity plants morphogenesis, growth and metabolism regulation [3]. However, it needs to carry out research on plant genetic, which will be ultimately upgraded to cereals and vegetables for the production of high quality food and other valuable bioactive compounds [4–10].

To date, few researchers have been studied the effects of gravity on plant developmental processes i.e., the effect of microgravity on cell proliferation within the root meristem [11,12] and using parabolic flights, the studies of Hausmann et al. [13] and Aubry-Hivet et al. [14] showed that changes in gene and protein expression in *Arabidopsis thaliana* cells were significant and fundamental changes in metabolic pathways were also revealed by these researchers. It is reported by Hoson et al. [2,15] that microgravity supports that microtubules played important role in mediating gravity resistance in *Arabidopsis*. Furthermore, Nakashima et al. [16] evidenced that actin might also be an important modulator of root growth in space, whereas Scherer and Pietrzyk [17] reported the root coiling in *Arabidopsis* under the effect of microgravity. Galland [18] and Göttig and Galland [19] used sporangiophore of *Phycomyces blakesleeanus* to investigate graviperception and graviresponses. Analyses of spores revealed that gravity affected expression of a PM-type Ca^{2+} ATPase [20]. Scherer and Quader [21] revealed a transient increase of endocytosis in tobacco pollen tubes under the effect of microgravity. Kordyum [22] highlighted a short overview of real and simulated microgravity on cell components, including statolith positioning, mitochondria, tubulin and the endoplasmic reticulum. Although significant progress has been made in identifying stimulus-responsive elements, the nature of the sensors remains elusive and Iida et al. [23] summarized mechanosensitive channels in Arabidopsis, named MCA1 and MCA2, and their putative role in gravity sensing in Arabidopsis. Tatsumi et al. [24] described recent progress in mechanosensitive channels controlled by the actin cytoskeleton. The involvement of the actin cytoskeleton in gravity perception is further investigated in plant cells using imaging tools i.e., Grolig et al. [25] reported the role of actin in organelle movement in the sporangiophore of the *zygomycete P. blakesleeanus*. Auxin transport is an essential component of the signaling pathway of roots and shoots under gravitropism. The report of Ueda et al. [26] complemented this view by examining the relationship between polar auxin transport and graviresponse in the context of microgravity. A comprehensive analysis of gene expression of floral buds revealed that hypergravity substantially changes expression of genes involved in the biosynthesis of phytohormones such as abscisic acid and auxin [27]. Nasir et al. [28] demonstrated the changes in expression of stress related genes under microgravity in *Euglena gracilis*.

From above discussion, it was hypothesized that microgravity can alter the morphogenesis and growth during germination. Therefore, present study was aimed to appraise the effect of microgravity (generated by clinostat) on pea, rice, corn and wheat cultivars. The principle objective was to investigate the effect on microgravity on amino acid profile under under normal, salt stress and Murashige and Skoog medium supplementation.

2. Materials and methods

2.1. Microgravity stimulator

UNOOSA is launching the zero-gravity Instrument Distribution Project and in UN/Malaysia Expert Meeting on Human and Space Technology in November 2011, gravity instruments were distributed to selected institutions (Universities, and research laboratories worldwide). The main objective of the project was to raise awareness and launch this research in education and research institutes, particularly in developing countries. One-axis clinostat (UNOOSA, USA) was used for microgravity generation and experimental set up is shown in Fig. 1 (The clinostat has a horizontal rotational axis perpendicular to the gravity vector on the ground). Growing seeds were exposed around the axis by rotation. The clinostat provided a simulated microgravity condition in equalizing the gravity vector. The direction of the rotational axis can be varied from 0° (parallel to the ground) to 90° (perpendicular to the ground). The rotation speed can be freely selected from 0 to 90 rpm with a 0.5 rpm increment from 0 to 20 rpm and a 5 rpm increment from 20 to 90 rpm. A one-axis clinostat has one limitation if a sample placed on the end of the rotational axis, it remained away from the axis and sample can not be exposed equally. The two-dimensional clinostat rotation cannot effectively compensate for the gravity exerted on all parts of the sample body [29]. This limitation was avoided by selecting small samples size of seeds.

There are several physical factors which affect the performance of one-axis clinostat. The first factor is the angle between the rotational axis and the true horizontal plane [30]. If the angle is one degree, the axial residual acceleration is 0.02*g*. It is important to set the rotational axis within 0.5° from the true horizontal plane of 10–2*g*. The second factor is the centrifugal force if a sample is placed away from the rotational axis [31]. A simple calculation shows that if a sample is placed one centimeter away from the rotational axis

Fig. 1 – Experimental setup of one-axis clinostat.

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