

# Cytokinins contents and dry matter accumulation at different position and types of grains within a spike of wheat

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## ABSTRACT

Dry matter accumulation and cytokinin (zeatin and zeatin riboside) level of grains in various positions within the wheat (*Triticum aestivum* L. Var. PBW-343) spike and spikelet were investigated during the grain filling period. Main shoot ears were partitioning into proximal, middle and distal regions and further into bold (basal) and small (apical) grains. Ten labeled spikes were sampled every 4 days from 3 days after anthesis (DAA) to 23 DAA, and every 7 days from 23 DAA to maturity. Results indicate that the cytokinin level increased rapidly from about 7 until 15 days after anthesis and then decreased depending upon the position of grains in spike and spikelet. The differences in cytokinin levels, both among spikelets in different regions of the spike and also among grains within a spikelet, were positively correlated with the differences in dry matter accumulation. Higher zeatin and zeatin riboside contents in the grains at the early grain filling stage, may promote the division of endosperm cells, thus constitute a powerful sink, and enhance assimilate migration and its accumulation in the developing grains. The results suggest that cytokinins in the grains during the early phase of grain development play an important role in regulating grain filling pattern and consequently influence grain filling percentage.

## INTRODUCTION

The position of a grain within an ear to some extent determines its final size, which can range from about 20 to 60 mg. Generally the grains from spikelets in the central regions of the ear and from the proximal florets within each spikelet are more towards the upper level of this range. Whereas those from spikelets in the basal and apical regions of the ear or from distal florets within each spikelet are closer to the lower level<sup>1,2</sup>. Various explanations such as assimilate availability and/or transport capacity<sup>3,4</sup> or hormonal differences<sup>5</sup> are offered in the literature to explain these difference.

Cytokinins play an essential role in regulating plant growth and development<sup>6,7</sup>. In addition to regulating the rate of cell division and cell elongation, cytokinins influence the intensity and direction of assimilate flow<sup>8</sup>. In cereals, peas, and beans, high levels of cytokinins are generally founding the endosperm of developing seeds, which may be required for active cell division during the early phase of grain setting<sup>9,10</sup>. Morris et al.<sup>10</sup> reported that zeatin (Z) and zeatin riboside (ZR) in developing

rice and wheat grains showed large transient in criases following pollination, which coincided with the period of seed setting and maximum endosperm cell division. For these reasons the concentration of cytokinins as well as dry matter accumulation of grains, during the filling period was determined at various positions within the ear.

## MATERIALS AND METHODS

Single plants of the wheat (*Triticum aestivum* L. Var. PBW-343) were grown in soil-filled plastic containers with a diameter of 4.5cm and depth of 20cm, and placed into a screen-covered hall under otherwise natural conditions. For all experiments only the ear of the mean shoot was used, starting 3 days after anthesis and then at intervals of about 4 days, ears were harvested frozen in liquid N<sub>2</sub>, and freeze-dried. Thereafter, they were divided into proximal (spikelet No. 1-5), middle (spikelet No. 6-15) and apical (spikelet 16 upward), regions. Each spikelet was further separated into basal (grain No. 1and2) and distal (grain No.3 upward) grains, and stored at -20°C for cytokinins determinations. The methods for extraction and purification of Z and ZR were modified from those described by Bollmark et al.<sup>11</sup>.

## RESULTS AND DISCUSSION

Cytokinins concentrations in the proximal and distal grains started to increase about 8 days after anthesis from moderate levels of a bout 11 ng grain<sup>-1</sup> to surprisingly high values (18 ng grain<sup>-1</sup>) after reaching a peak about 20 to 30 days after anthesis, dependent on spikelet position within the spike, the concentration of cytokinins dropped almost to its value before the increase. The rapid increase in cytokinin concentration closely paralleled the dry weight accumulation, and a correlation is obvious, at least until the peak of cytokinin is reached. The correlation hold true both for comparisons between spikelets in different regions of the spike and also between florets within spikelets.

The proximal grain of spikelets in the middle region of the spike accumulated more dry matter during the later stages of grain filling than comparable grains in the basal region of the spike. This deviation from the general pattern was preceded by a corresponding change in cytokinin concentration. The proximal grain of spikelets in the apical region of the spike showed higher cytokinin concentrations in spite of their lower weight,

when compared to proximal grains from spikelets in the basal region. The close spatial and developmental correlation between cytokinin concentration and dry matter accumulation described above indicates a causal relationship. Several physiological processes implicated in grain growth could be affected by the concentration of cytokinins. Some of those already considered in the literature include effects on assimilate transport to the grain<sup>12</sup>.

Evidence that cytokinin concentration really affect these processes in wheat grains is, however, in most cases

substantial. The alternative possibility that the observed correlation between cytokinins concentration and dry matter accumulation only represents an accidental parallel variation is that the higher cytokinin level is the result of the higher growth rate of the heavier grains, cannot be excluded. Clearly, only more experiments designed to specifically alter cytokinins and/or dry matter accumulation will give us a closer insight as to whether cytokinins is a causal factor in or a by-product of grain growth.

**Table1: Changes in cytokinins (Z+ZR) contents (ng grain<sup>-1</sup>) of grains in different parts of spike and spikelet after flowering**

Position		Days After Anthesis (DAA)							
		3	7	11	15	19	23	30	37
Proximal	B	2.1±0.24	11.3±2.36	20.4±3.28	16.2±3.27	12.4±2.42	7.9±2.48	4.1±1.24	1.1±0.98
	S	1.8±0.25	9.2±1.36	18.3±2.47	13.5±2.98	10.2±1.42	5.6±1.37	3.1±0.92	0.8±0.61
Middle	B	3.1±0.28	13.2±2.27	23.2±3.88	19.9±4.07	15.6±2.42	10.3±2.38	6.4±1.24	2.7±0.86
	S	1.9±0.27	11.1±1.28	19.2±2.62	17.4±3.46	13.6±1.45	8.8±1.38	4.1±1.12	1.7±0.69
Distal	B	2.1±0.18	12.1±2.24	21.2±3.28	18.4±3.37	14.6±2.40	8.9±2.32	6.8±1.21	1.9±0.89
	S	2.2±0.16	10.3±1.21	17.5±2.22	16.2±3.26	12.4±2.21	6.7±1.12	5.8±0.94	1.1±0.78

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