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# CARYOPSIS TRANSFER SYSTEM IN AN AVENA MAGNA MURPHY ET TERRELL × A. LONGIGLUMIS DUR. AMPHIPLOID

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**Abstract.** Evaluation of the structure of the caryopsis transfer system is presented for an *Avena magna*  $\times$  *A. longiglumis* amphiploid. Each component of the system such as vascular bundle, pigment strand, nucellar projection and ventral aleurone layer varies between the amphiploid and its parental species. The number of xylem vessels present in the caryopsis bundle expressed heterosis-like inheritance. The position of the caryopsis xylem bundle in *A. longiglumis* shows a lower efficiency in assimilate transport. Some anomalies in the development of the ventral aleurone layer and parenchyma, adjacent to the transfer system, are presented.

Key words: Avena amphiploid, caryopsis, transfer tissues, xylem

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#### Introduction

The caryopsis transfer complex is composed of various tissues like vascular bundle, pigment strand, nucellar projection, and ventral aleurone layer. These tissues are in the crease area. In the Hordeum mutants, this complex decides on successful assimilate storage in the endosperm (FELKER et al. 1985). In many grasses, transfer cell walls have been detected in these tissues, especially in nucellar projection. A distinct correlation has been discovered in wheat tetraploids between the number of xylem vessels presented in the vascular bundle of the caryopsis and the morphogenesis of starchy-protein endosperm tissue (KOSINA 1988). Bundles composed of many vessels were positively correlated with the development of a thick high-protein subaleurone layer. Demethylation of genomes in a Triticum timopheevii Zhuk. × Aegilops umbellulata Zhuk. amphiploid led to the development of a poor nucellar projection, acellular pigment strand and sclerification of the

tissue adjacent to pigment strand (KOSINA *et al.* 2013; KOŹLIK 2013). Two wheats, *Triticum kiharae* Dorof. & Migush. and *T. fungicidum* Zhuk. differ from each other in size of the transfer ability in the xylem bundle, pigment strand and nucellar projection (KOSINA & BURES 2011). Structural differences detected in the nucellar projection of *Brachypodium distachyon* (L.) P. Beauv. appeared to be almost qualitative, where three distinct types have been distinguished (KOSINA & KAMIŃSKA 2013a).

## Material and methods

Accessions of the parental species, Avena magna Murphy et Terrell and A. longiglumis Dur., and an A. magna  $\times$  A. longiglumis amphiploid were cultivated on small plots under the same soil and climatic conditions in R. Kosina's grass collection. The studied material was treated as originating from a completely randomised one-way classification design. Samples of size n = 30 were elaborated.

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Caryopses were fixed in FAA, in the following proportions: formaldehyde (40%) : ethanol (50%) : glacial acetic acid (10%). After fixation, caryopses were rinsed three times in tap water and cut in the central part, in a plane perpendicular to the caryopsis axis. For cutting, a freezing microtome K TS-II (USSR) was used. Cross-sections of caryopses about 40 µm in thickness were mounted in glycerin as semipermanent slides. The slides were documented in a polarizing Amplival microscope (Carl Zeiss, Jena, Germany). A natural fluorescence of tissues has been observed in an Olympus BX60 epifluorescence microscope with triple filter (DAPI, TRITC, FITC). Images were taken with an Olympus E-520 camera (Olympus Imaging Europa GMBH, Hamburg, Germany).

## **Results and discussion**

The organization of the caryopsis transfer system in oats is similar to that of other grasses. In the maternal species, A. magna, the xylem bundle is composed of nearly 19 vessels. Some of them are clearly visible in Fig. 1 A. In the paternal species, A. longiglumis, the xylem bundle is distinctly smaller, and consists from about 9 vessels (Fig. 1 C). In A. longiglumis, the distance between the xylem and the pigment strand is larger compared to that in A. magna (Fig. 1 C vs 1 A), and this distance can be several times larger (Fig. 1 D, see a dotted line between pigment strand and vascular bundle). Thus, the efficient transport of assimilate provided by vascular bundle into endosperm is distinctly lower in A. longiglumis. On the other hand, in the amphiploid, the vascular bundle and adjacent parenchyma are isolated from other parts of pericarp by a thickwalled structure (Fig. 1 E, see white arrows). Also, a pericarp epidermis with thick internal and external tangential walls additionally isolates this area. Under polarizing light, these walls show a strong reaction, because of the presence of high cellulose content. Sclerification of the caryopsis transfer system has also been noted in other oat species such as A. brevis Roth and A. strigosa Schreb. (GRABIŃSKA 2008). The xylem bundle in

the amphiploid is large and composed of up to 30 vessels (Fig. 1 B). Such a significant transgression of parental characteristics in the amphiploid can be a result of heterosis. After colchicine treatment of F1 plants, genomes of each parent occur as homologous sets in an amphiploid; however, interactions between the genomes of both parents and multiple translocations observed in the oat hybrid progeny create a heterozygous complex with expression of heterosis. This phenomenon of inheritance observed in successive generations in plant hybrids can be considered as a multiple heterosis (PALILOV 1976). In the amphiploid, the morphology of the xylem bundle, but also multivariate characteristics of spikelet and abaxial epidermis of lemma show that the uniparental dominance is expressed, showing a distinct shift towards the maternal species, A. magna (Swietlikowska 2008). In another cereal amphiploid, Triticum timopheevii × Aegilops umbellulata, the number of placental xylem vessels is highly positively correlated with the width and thickness of caryopsis and height of nucellar projection (KOSINA 2014).

As a rule, the aleurone layer develops between the nucellar projection and the starchy endosperm. In this area, the aleurone cells have transfer walls that are not present in others parts of the caryopsis. Some anomalies in the development of transfer system are detected in this area in A. longiglumis (Fig. 1 D, green arrow) and to a greater extent in the amphiploid. The aleurone layer is not developed there and the cells of the starchy endosperm adjoin nucellar projection. Such a pattern of development has been observed in other grasses such as Brachypodium sylvaticum (Huds.) P. Beauv. (Kłyk 2005), B. distachyon (Kosina et al. 2012; Kosina & Kamińska 2013b), Avena wiestii Steudel (GRABIŃSKA 2008) and Bromus secalinus L. (KOCHMAŃSKI 2008), but it seems to be more frequent in plants of hybrid origin (Bureś 2008; Tomaszewska 2009; Zając 2009; Koźlik 2013).

The development of the amphiploid caryopses appeared to be less stable than that in parental species (Świetlikowska 2008). In amphiploid plants, the most frequent



**Fig. 1.** Caryopsis xylem bundles in parental species *Avena magna* (**A**) and *A. longiglumis* (**C**), and in the amphiploid (**B**). Caryopsis transfer system in *A. longiglumis* (**D**) and in the amphiploid (**E**). **np** – nucellar projection; **ps** – pigment strand. Xylem bundles are encircled by **green dotted lines**. Thick cell walls isolating the vascular bundle are indicated by **white arrows** in **E**. Absence of aleurone layer at the border between the nucellar projection and starchy endosperm is indicated by a **green arrow** in **D. A-C** – as observed under polarising light; **D**, **E** – epifluorescence images. Microscope magnification for **A-C** – ×200; for **D**, **E** – ×80.

chromosome number is 41. Cytogenetic behaviour, with bridges, laggards and telocentrics, can lead to this hipohexaploid level.

## **Concluding remarks**

The 4x/2x Avena amphiploid expressed some cytogenetic and developmental instability

(ŚWIETLIKOWSKA 2008). Also, such a status was detected for the ventral aleurone layer development. In the amphiploid plants, heterosis and maternal dominance were noted. A positive transgression in the number of caryopsis xylem vessels creates some assimilate storage advantage of the amphiploid over its parental species. Perhaps, this phenomenon can be more enhanced in cytogenetically stable hexaploid plants.

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