HORMONAL BALANCE AND PHYSIOLOGICAL BACKGROUND FOR DECISION SUPPORT ON FRUIT SET IMPROVEMENT ON PEAR TREES

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ABSTRACT

Since the growth regulator chlormequat chloride (CCC) was removed in Belgium in 1998, pear growing in intensive training systems has become more difficult and fruit growers are looking for alternative ways to control the vegetative growth of the pear trees. Prohexadione-Ca, a new growth regulator with good results on apple trees, is much less effective on pear trees and can have a negative effect on return bloom. The best way to control the vegetative vigour of a fruit tree is a regularity in the productivity, which can be achieved by a treatment with gibberellins, but the results of these treatments are not consistent. In this study, we investigate the effect of treatments of 'Conference' pear trees with Prohexadione-Ca and different gibberellins on proteome and gene expression, in order to obtain a better understanding of the physiological background of the process of flower initiation and fruit set on 'Conference' pear.

INTRODUCTION

Growth regulation in modern intensive pear growing systems is considered to be an important cultural measure for crop regularity and for fruit quality. Since the growth regulator chlormequat chloride (CCC) was removed in Belgium in 1998, pear growing in intensive training systems has become more difficult and fruit growers are looking for alternative ways to control the vegetative growth of the pear trees. Prohexadione-Ca, a new growth regulator with good results on apple trees, is much less effective on pear trees and can have a negative effect on return bloom (Deckers and Schoofs, 2004). For lack of efficient chemical growth regulators, the best way to control the vegetative vigour of a fruit tree is a regularity in the productivity, which can be achieved by a treatment with gibberellins, but the results of these treatments are not consistent. In this study, we investigate the effect of treatments on pear trees with Prohexadione-Ca and different gibberellins on proteome and gene expression in order to obtain a better understanding of the physiological background of the process of flower initiation and fruit set on 'Conference' pear.

MATERIALS AND METHODS

The trials were conducted on adult pear trees of the cultivar 'Conference' on Adams rootstock. There are five different groups in the experiment: control trees without treatment, trees treated with the gibberellin GA_3, trees treated with the gibberellin $GA_{4/7}$, trees treated with a combination of GA_3 and $GA_{4/7}$ and trees treated with Prohexadione-Ca. Because growth and flowering capacity of shoots can be different depending on the place on the tree, samples were taken of three different types of shoots and of flowers and fruits on different key moments in the

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growth season (Fig.1). Flower bud quality, fruit set, yield, fruit quality and return bloom were successively examined in detail on marked branches.

Fig.1: Different key moments in flower development

A total protein fraction is obtained using the phenol extraction procedure and is analyzed by two-dimensional gel electrophoresis. Isoelectric focusing is performed on the Ettan IPGphor system and the second dimension on the Ettan DALT six system. For each experimental group, four biologically independent gels are constructed. These gels are analysed and compared using ImageMaster 2D Platinum. Corresponding gel spots that are different in intensity between the different treatments are excised, trypsinised and identified by mass spectrometry (MALDI-TOF/TOF).

Gene expression of the pear homologues of LFY, AP1 and TFL, three key genes in the flower initiation process, is analysed by real-time quantitative RT-PCR. The RNA expression levels of the most important proteins that are shown to be up- or downregulated by the 2-D electrophoresis, will also be analysed by real-time PCR.

RESULTS AND DISCUSSION

We observed that fruit set on terminal buds tended to be higher in trees treated with gibberellines, but it was never statistically signifant. There was no observed effect of Prohexadione-Ca. The yields of all trees were high in 2005 (ca. 30T/ha) and not noticeably different between the treatments. In this context it would be interesting to analyse the endogenous levels of plant growth regulators to check if a high gibberellin level was already naturally present in the trees. There was, on the other hand, a significant reduction of fruit size in trees treated with GA_3, which was not observed in trees treated with $GA_{4/7}$. There were no statistical differences in return bloom, although a tendency to a reduced return bloom was observed in trees treated with Prohexadione-Ca.

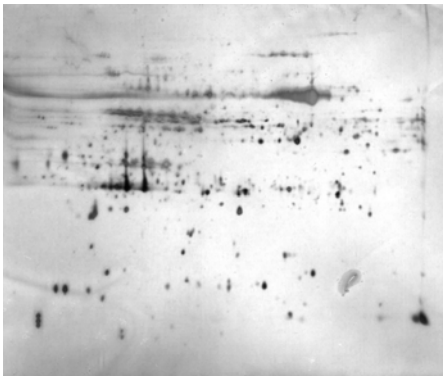

Fig.2: 2D-electrophoresis analysis of flower buds

In order to obtain a clean and analysable protein extract and 2D-electrophoresis gel of pear tissue, several optimisation steps were necessary: a new extraction buffer (based on the phenol extraction method for recalcitrant plant tissues from Carpentier et al., 2005), some extra rinsing steps and extra centrifugation of the sample for 90 minutes at 75000 x g to remove DNA and other contaminants; an other pH-gradient (pH 4-7) and a new running protocol for first (IEF) and second (SDS-PAGE) dimension (Fig. 2).

The real-time quantitative RT-PCR analysis showed that gene expression levels of the 3 genes of interest are very variable within treatments; untill now no significant differences could be found between treatments.

ACKNOWLEDGEMENTS

This project was sponsored by The Institute for the Promotion of Innovation by Science and Technology in Flanders (project nr.030794).

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