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A preliminary evaluation of the effects of pollinator enhancement and gibberellins on the fruit set and fruit shape of 'Conference' pears – Short Communication

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Abstract: 'Conference' (*Pyrus communis* L.) is a self-incompatible cultivar, although it can also set fruit parthenocarpically. Stimulating parthenocarpy through gibberellin (GA) applications increases the fruit set, but it may also negatively affect the fruit size and shape. The aim of this study was to investigate the effects of a bumblebee (BB) amendment in combination with a GA treatment on the fruit set and fruit shape of 'Conference' pears. In the first experiment, we applied three treatments (GA, GA + BB & control) in a 'Conference' monoculture. In the second experiment, we applied two treatments (GA & GA + BB) in a 'Conference' orchard inter-planted with 'Concorde' as pollinizer trees. Both experiments showed that the GA application and bumblebee supplementation did not affect the fruit set. However, the BB treatment resulted in significantly higher amounts of normally shaped pears. Trees closer to the bumblebee hive had more normal shaped pears than trees further away.

Keywords: Bombus spp.; bumblebees; phytohormones; polliniser trees; *Pyrus communis* L.

'Conference' is the most widely grown European pear cultivar (*Pyrus communis* L.) in NW-Europe. It is a self-incompatible cultivar although it can also set fruit parthenocarpically. Successful pollination in pome fruits depends on many factors, such as compatible polliniser trees and the transfer of the pollen by pollinators (Free 1993). Due to insufficient pollination and fertilisation, both the fruit set and fruit quality remains inadequate in some apple and pear cultivars (Geslin et al. 2017). This is mainly due to a lack of suitable polliniser trees and/or pollinator insects that disperse the pollen, as previous studies have shown that the fruit set and fruit quality increases when pear cultivars are cross-pollinated (Moriya et al. 2005; Quinet, Jacquemart 2015). The current declines in both wild pollinators and managed honeybees (Potts et al. 2010) urges experimentation with other insect pollinators to guarantee an optimal fruit set. For example, bumblebees (*Bombus* spp.) are well adapted to wind, rain and cold weather, they can forage over substantially longer periods of the day and can visit more flowers per minute than honeybees (Lundberg 1980). Yet, it is not common to introduce bumblebees in orchards. One study revealed that the number of seeds per fruit, fruit set, fruit size and fruit quality of 'Spadona' and 'Coscia' pear trees were increased when bumblebee hives (10/ha) were established (Zisovich et al. 2012). Furthermore,

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most pear cultivars are self-incompatible and need to be inter-planted with polliniser trees for an optimal fruit set and fruit quality (Jacquemart et al. 2006; Kemp et al. 2008). Warnier (2000) showed that the fruit size and shape of 'Conference' pears were improved when the flowers were cross pollinated with 'Comice', compared to self-pollination. Quinet and Jacquemart (2015) reported an increase in the fruit set between 10 and 40% when 'Conference' flowers were cross pollinated, whereas parthenocarpic fruits remained 12% smaller compared to the fruits with seeds. To increase the fruit set, growers can also stimulate the parthenocarpic fruit set by spraying phytohormones such as gibberellins (GA). Although a high fruit set is often obtained, the fruit quality has been reported to be poor, often with long bottle shaped pears having a lower market value (Vercammen, Gomand 2008). Also, in cultivars such as the Portuguese 'Rocha', the cross-pollinated pears were found to be superior in terms of their fruit shape compared to pears treated with GA (Silva et al. 2008).

The aim of this study was to investigate the effects of a bumblebee amendment and a GA treatment on the fruit set and fruit shape of 'Conference' pears, in both a monoculture orchard and in an orchard inter-planted with polliniser trees. Furthermore, we aimed at evaluating the mediating role of bumblebees by comparing the fruit set and fruit shape among the different distances from the established bumblebee hives.

MATERIALS AND METHODS

Orchard and experimental set-up. The field trials were conducted in 2015 and were located in two, 6 year old 'Conference' orchards in Bierbeek, Belgium. The flowering period took place from April 18th until April 29th 2015. One 'Conference' orchard (6 600 m²) was inter-planted with 'Concorde' polliniser trees and the other one was a 'Conference' monoculture only (10 800 m²). The distance between the rows was 3 m and the distances between the trees within the row was 1 m. Both orchards had a similar crop load in the year before the experiment. In the monoculture orchard, three plots (each ± 2 000 m²) were randomly established. Each plot was assigned a different treatment and within each plot, 24 trees were selected. In the first plot, the trees were treated with gibberellins (GA), in the second plot, the trees were treated with GA and a bumblebee (BB) hive was established at the centre of the plot, and the third plot was a control treatment (without GA and bumblebees). In the second orchard, 'Concorde' trees were inter-planted within each row as polliniser trees at a density of 10%. In this orchard, all the trees were treated with GA, and two plots (± 2 000 m²) were randomly established. In the first plot, one bumblebee hive was established, and the second plot acted as a control. Due to technical reasons, it was not possible to establish a plot with trees that were not treated with GA, as in the first experiment. In the two experiments, 'turbo hives' (Biobest nv, Belgium) were used as the bumblebee hives, which consisted of 200 workers and were placed in the orchards when 20% of the flowers were open (20th of April). The GA treatment (Promalin® BASF) was conducted just before full bloom (22nd of April) when the temperatures were optimal. The trees were sprayed in the morning following the manufacture's guidelines.

Fruit set and fruit shape analysis. On each selected tree, we labelled one branch on which the total number of clusters and flowers were counted at the beginning of the bloom period. After the initial fruit set, all the fruits on the branch were counted and this was repeated after the June drop and just before harvest at the end of August. From each labelled branch, 8 pears were randomly harvested at the beginning of September at their optimal picking time.

We analysed the fruit shape and size (length and width), and we counted the number of seeds. The pears were divided into 5 groups according to their shape (Figure 1): normal pears (A); slightly misshapen pears (B); strongly misshapen pears (C); bottle shaped pears (D); different shapes (A–D), but too small (length < 110 mm).

RESULTS

Monoculture orchard. A Kruskal-Wallis test showed that there were no differences (χ^2 = 1.2, d.f. 2, P = 0.5) in the fruit set among the treatments (Table 1). Only 23 out of the 544 harvested pears had one or more fully developed seed(s) and no significant differences in the number of seeds were found among the treatments. There were significant differences in the pear length between the treatments (χ^2 = 9.3, d.f. 2, P = 0.01) (Table 2). The GA treatment and the combination treatment (GA + BB) yielded significantly longer pears compared to the pears from the control treatment (P = 0.044 and P = 0.003, respectively). No differences were found in the length of the pears between the GA and

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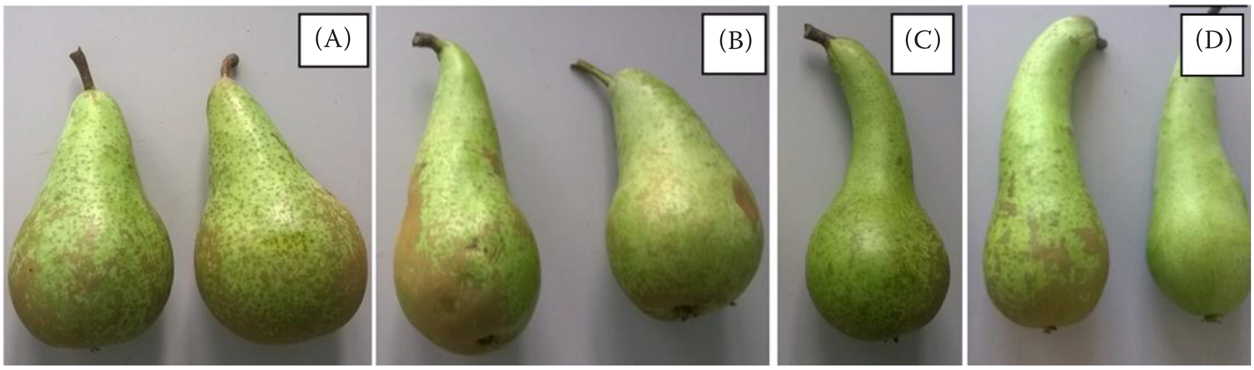

Figure 1. The different fruit shape categories of the 'Conference' pears: (A) normal, (B) slightly misshapen, (C) strongly misshapen, and (D) bottle shaped

combination (GA + BB) treatment. Also, the width of the pears was significantly different between the treatments (χ^2 = 12.6, d.f. 2, P = 0.002) (Table 2). The pears from the combination treatment and the control treatment were significantly wider than those treated with GA only (P = 0.001 and P = 0.003, respectively). No significant differences were found in the width of the pears between the control and combination treatment. The differences between the treatments became even clearer for the length/width ratio (χ^2 = 31.0, d.f. 2, P < 0.0001) (Table 2). The pears from the GA treatment had a significantly higher length/width ratio compared to the combination treatment (P = 0.03) and compared to the control treatment (P < 0.0001). The pears from the combination treatment had a significantly higher length/width ratio compared to the ones from the control treatment (P < 0.0001). The fruit shape categories differed significantly among the three treatments (χ^2 = 26.6, d.f. 8, P = 0.0008) (Table 2). The pears that were only treated with GA had less normal shaped pears in comparison with the combination treatment and the control ones (based on the adjusted residuals, P = 0.0002). The pears from the combination treatment also had less fruits that were strongly deformed compared to the GA treatment and the control ones (P = 0.001). No significant differences between the treatments were found for the other fruit shape categories.

Orchard with polliniser trees. A Kruskal-Wallis test showed that the fruit set did not differ significantly between the treatment with (GA + BB) and without bumblebees (GA) at harvest (χ^2 = 2.0, d.f. 1, P = 0.2) (Table 1). Twenty two out of the 384 investigated pears had one or more developed seeds. No significant differences were found in the seed number between the treatments (χ^2 = 2.3, d.f. 1, P = 0.1), but a significant difference was found among the distances from the bumblebee hive (χ^2 = 9.8, d.f. 3, P = 0.02) (Table 3). The pears at 10-20 m from the bumblebee hive had more seeds compared to the ones at 30–40 m (P = 0.023) and 50-60 m (P = 0.023), no signifi-

Table 1. Fruit set (n = 24) and seed set of the 'Conference' pears in the experiment conducted in the monoculture orchard (gibberellins + bumblebees (GA + BB); gibberellins (GA)) and the orchard with polliniser trees (gibberellins + bumblebees (GA + BB); gibberellins (GA))

	Fruit set*			Seed set*
	initial	after June drop	at harvest	seeds per fruit (# pears)
Orchard monoculture				
Control	95.1%	40.8%	33.0%	0.057 (192)
GA + BB	96.4%	41.1%	32.5%	0.016 (192)
GA	93.2%	39.3%	30.3%	0.056 (160)
Orchard with pollinizer trees				
GA + BB	95%	42%	31%	0.063 (192)
GA	95%	40%	28%	0.089 (192)

*No significant differences were found between the treatments

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Table 2. The length, width, length/width ratio and percentage fruit shape of the normal and strongly deformed 'Conference' pears per treatment in the monoculture orchard (gibberellins + bumblebees (GA + BB); gibberellins (GA) and the orchard with the polliniser trees (gibberellins + bumblebees (GA + BB); gibberellins (GA))

	Length (mm)	Width (mm)**	Length/width**	Fruit shape (%)	
				normal*	strongly deformed*
Orchard monoculture					
Control (*n* = 192)	118.44^{b}	59.5^{a}	2.00^{c}	41^{a}	11^{a}
GA + BB (*n* = 192)	122.92^{a}	59.8^{a}	2.06^{b}	47^{a}	4^{b}
GA (*n* = 160)	121.74^{a}	57.94^{b}	2.11^{a}	29^{b}	12^{a}
Orchard with polliniser trees					
GA + BB (*n* = 192)	117.64	57.51	2.06	46	14
GA (*n* = 192)	118.35	57.93	2.05	46	15

*Different letters indicate a significant difference between the treatments at $P \leq 0.002$ (adjusted Bonferroni *P*-value); **different letters indicate a significant difference between the treatments at $P \leq 0.05$

cant differences were found for the other distances. The width ($\chi^2 = 0.2$, d.f. 1, $P = 0.7$), length ($\chi^2 = 0.08$, d.f. 1, $P = 0.8$) and length/width ratio ($\chi^2 = 0.01$, d.f. 1, $P = 0.9$) did not significantly differ between both treatments (Table 2). The width of the pears differed significantly among the distances to the hive ($\chi^2 = 12.1$, d.f. 3, $P = 0.007$) (Table 3). The pears at 50–60 m were significantly smaller compared to the pears at 0–10 m ($P = 0.001$) and the ones at 30–40 m ($P = 0.011$). For the other distance comparisons, no significant differences were found in the width of the pears. The fruit shape categories did not differ significantly between the treatments ($\chi^2 = 0.8$, d.f. 4, $P = 0.9$) (Table 2), but did differ among the distances from the bumblebee hive ($\chi^2 = 21.9$, d.f. 12, $P = 0.04$) (Table 3). The pears at 50–60 m from the hive had significantly ($P = 0.0008$) less normal shaped pears in comparison to the other distances. On the other hand, the pears at a distance of 50–60 m were significantly more bottle shaped compared to the other distances ($P = 0.002$).

DISCUSSION

Both in the monoculture orchard and the orchard with polliniser trees, no differences were found in the fruit set between the different treatments. Although it could have been expected that trees treated with gibberellins would have a higher fruit set (Deckers, Schoofs 2002; Zhang et al. 2008). Also, no effect was observed with the distance from the bumblebee hive on the fruit set. When compared with other studies on 'Conference' pears, our fruit set at harvest was high overall, which may have blurred any potential differences between the treatments (Quinet et al. 2016).

Table 3. The length (mm), width (mm), length/width ratio, seed set and percentage fruit shape of the normal and bottle shaped 'Conference' pears for the treatment gibberellins + bumblebees (GA + BB) based on the distances from the bumblebee hive (*n* = 48) in the orchard with the polliniser trees

Distance from the bumblebee hive	Length** (mm)	Width** (mm)	Length/width**	Seed set**	Fruit shape (%)	
				seeds per fruit	normal*	bottle shaped*
0–10 m	120.11	59.12^{a}	2.04	0.083^{ab}	60^{a}	10^{b}
10–20 m	115.89	57.85^{ab}	2.01	0.167^{a}	52^{a}	15^{b}
30–40 m	118.47	58.28^{a}	2.04	0 b	46^{a}	6^{b}
50–60 m	116.08	54.80^{b}	2.13	0 b	25^{b}	29^{a}

*Different letters indicate a significant difference between the treatments at $P \leq 0.002$ (adjusted Bonferroni *P*-value); **different letters indicate a significant difference between the treatments at $P \leq 0.05$

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It was expected that adding bumblebees would result in more developed seeds compared to the gibberellin treatment (Zisovich et al. 2012). Yet, the seed set was not affected by the presence of the bumblebees, although the fruits closest to the hive (0–10 m and 10–20 m) had a significantly higher seed set compared to the fruits further away from the hive. This suggests that the bumblebees pollinated the flowers on trees close to the hive more than the trees further away (Zisovich et al. 2012). We only included the fully developed seeds and not the aborted seeds. Quinet et al. (2016) found a high percentage of aborted seeds in 'Conference' pears. The aborted seeds may have affected the development of the fruits during the first weeks after flowering, finally affecting the final fruit quality at harvest.

In the monoculture orchard, the fruits treated with gibberellins were significantly longer and had a reduced width, whereas this trend was not observed in the orchard with polliniser trees. The fruits from the treatment with both the bumblebees plus the gibberellins had a more normal shape. The positive effect on the fruit shape is likely caused by the bumblebee pollination which initially results in more seeds, improving the length/width ratio and creating a more pyriform fruit shape (Stern et al. 2004; Zisovich et al. 2012). The fruits from the gibberellin treatment are parthenocarpic and it is known that the absence of seeds negatively impacts the fruit shape (Silva et al. 2008). The fruits from the treatment with gibberellins not only deviated from the normal fruit shape, but had also significantly more strongly deformed fruits compared to the treatment with the bumblebees. We also observed a trend towards more slightly deformed pears, more undersized pears and bottle shaped pears in the gibberellin treatment, as was also reported earlier by Warnier (2000) and Vercammen and Gomand (2008). In the orchard with polliniser trees, the presence of the bumblebees did not have an overall effect on the length, width, length/width ratio and fruit shape categories. However, on the other hand, some effects were observed among the different distances from the bumblebee hive. For instance, the pears closer to the hive were wider compared to the pears further away. Moreover, the pears close to the hive had a higher percentage of normally shaped pears while an increase in bottle shaped pears was found further away from the hive. This suggests that although GA was applied, some effects of the bumblebees were observed, demonstrating the positive contribution of the bumblebees on the fruit shape of the 'Conference' pears.

Our results should be interpreted with care as they should be replicated through time and space. For instance, the weather conditions during the flowering and growing season may differ from year to year, influencing the fruit set and fruit growth. On the other hand, orchards also differ in terms of design, maintenance and age, which may all influence whether certain pollination measures succeed. As we could show a distance effect from the bumblebee hive, it may be necessary to establish sufficient number of bumblebee hives. Furthermore, unless there are specific reasons, such as frost or poor flower quality, we recommend to reduce the GA applications to a minimum in 'Conference' orchards, to improve the fruit quality.

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