the tunnels, the mortalities of all groups fluctuated at a low level, indicating that sulfoxaflor had no long-term lethal effect on bees.

Flight intensities

During the whole exposure period, the bee flight intensities of all groups at the same time of the same day were not significantly different. When compared among different time periods, the bee flight intensities at 7:00 and 11:00 were higher than that at 16:00 (figure 4).

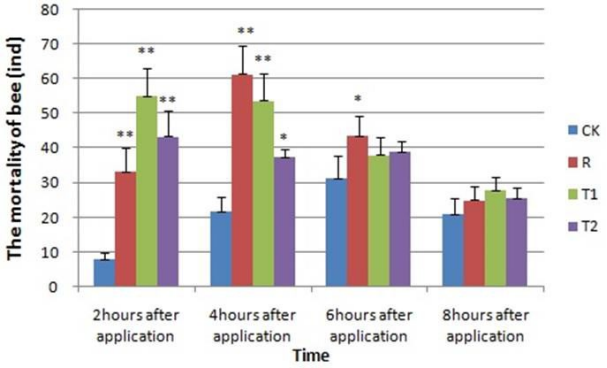

Figure 2. Bee mortality of each group on the day of second application (July 8). CK: water control; R: reference substance treatment, dimethoate 600 g a.i./ha applied on July 8; T1: sulfoxaflor treatment, 75 g a.i./ha applied on July 1 and July 8; T2: sulfoxaflor treatment, 100 g a.i./ha applied on July 1 and July 8. (*) = significantly different between blank control and positive control group or treatment groups, $P < 0.05$; (**) = highly significantly different between blank control and positive control group or treatment groups, $P < 0.01$.

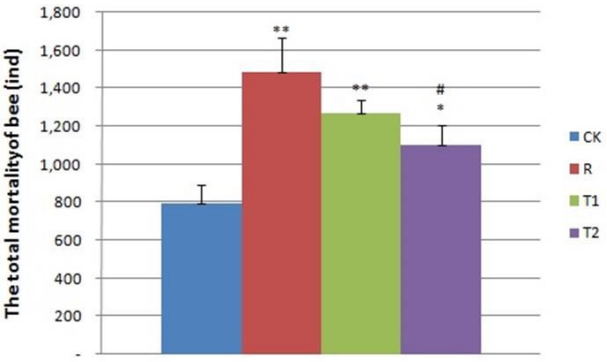

Figure 3. Total amount of dead bees in each group during the whole experiment period. CK: water control; R: reference substance treatment, dimethoate 600 g a.i./ha applied on July 8; T1: sulfoxaflor treatment, 75 g a.i./ha applied on July 1 and July 8; T2: sulfoxaflor treatment, 100 g a.i./ha applied on July 1 and July 8. (*) = significantly different between blank control and positive control group or treatment groups, $P < 0.05$; (**) = highly significantly different between blank control and positive control group or treatment groups, $P < 0.01$; (#) = significantly different between positive control group and treatment groups, $P < 0.05$.

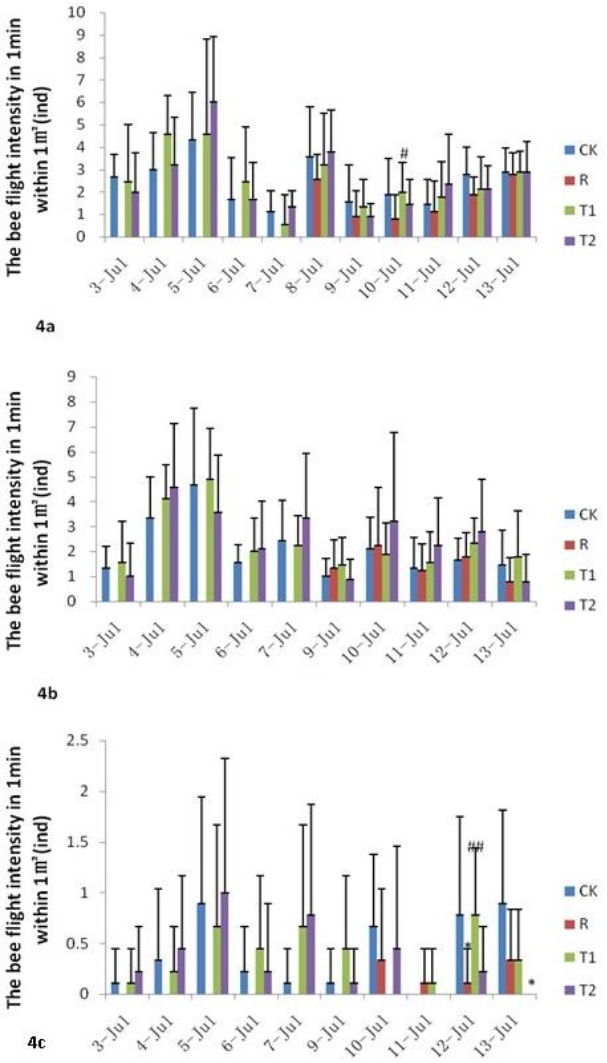

Figure 4. Change of bee flight intensity of each group from the beginning to the end of exposure. CK: water control; R: reference substance treatment, dimethoate 600 g a.i./ha applied on July 8; T1: sulfoxaflor treatment, 75 g a.i./ha applied on July 1 and July 8; T2: sulfoxaflor treatment, 100 g a.i./ha applied on July 1 and July 8. **4a:** bee flight intensity at 7:00; **4b:** bee flight intensity at 11:00; **4c:** bee flight intensity at 16:00. (#) = significantly different between positive control group and treatment groups, $P < 0.05$; (##) = highly significantly different between positive control group and treatment groups, $P < 0.01$.

Colonies conditions and sizes

Before exposure, there were no significant differences in the total amount of bees between groups ($P > 0.05$). On the second day of the end of exposure, the total amount of bees of positive control group became the highest, and the total amount of bees of T1 group was lower than that of the positive control group but higher than that of the blank control group. The total amount of bees of T2 group was the lowest, significant lower than the positive control group ($P < 0.05$). On 14 days after the end of exposure, the differences in the total amount of bees between groups were not significant ($P > 0.05$) (figure 5).

The total amounts of bees in each group on the second day of the end of exposure and on 14 days after the end of exposure were not significantly different with that of the first assessment ($P > 0.05$) (figure 6).

Before exposure, there was pollen stored in each hive, and the colonies were healthy with different portions of different brood stages, including pupae, larval, young bees. At the end of exposure, the pollen storage in each hive decreased to zero while the proportion of bees at each developmental stage did not change obviously. On 14 days after the end of exposure, there appeared pollen in each hive, also with different portions of different brood stages (figure 7).

Colony condition assessment showed that: the pollen storage during the exposure period decreased to zero, while other endpoints such as the colony strength, the proportion of bees at each developmental stage did not change obviously. Under this experiment condition, sulfoxaflor had no obvious adverse effect on the strength and the condition of the test bees.

Sulfoxaflor residues on cucumber flowers

Limit of Detection and Limit of Quantitation

The limit of detection (LOD) and limit of quantitation (LOQ) of sulfoxaflor in cucumber flower were calculated according to 0.01 mg/kg added recovery experiment. LOD was three-time standard deviation (0.0018 mg/kg) while LOQ was ten-times standard deviations (0.006 mg/kg).

The limit of detection (LOD) and limit of quantitation (LOQ) of dimethoate in cucumber flower were calculated according to 0.02 mg/kg added recovery experiment. LOD was three-time standard deviation (0.0009 mg/kg) while LOQ was ten-times standard deviations (0.003 mg/kg).

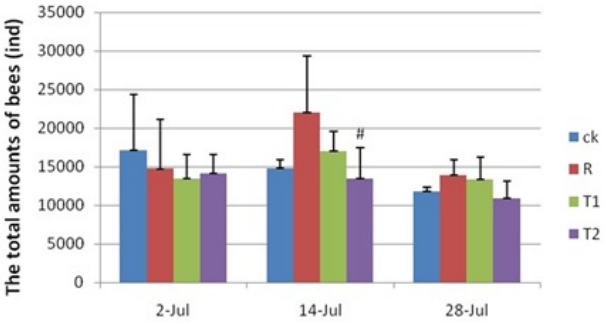

Figure 5. Comparison of the total amounts of bees at the same time. CK: water control; R: reference substance treatment, dimethoate 600 g a.i./ha applied on July 8; T1: sulfoxaflor treatment, 75 g a.i./ha applied on July 1 and July 8; T2: sulfoxaflor treatment, 100 g a.i./ha applied on July 1 and July 8.

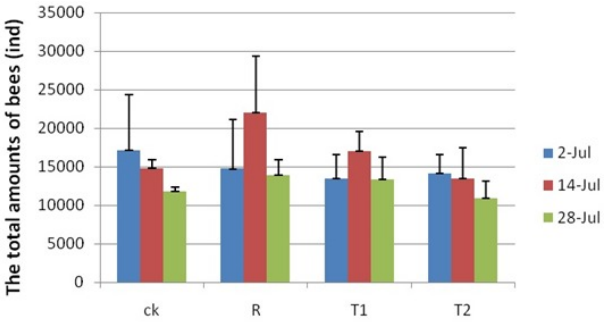

Figure 6. Comparison of the total amounts of bees in same group before exposure, on the second day of the end of exposure and on 14 days after the end of exposure. CK: water control; R: reference substance treatment, dimethoate 600 g a.i./ha applied on July 8; T1: sulfoxaflor treatment, 75 g a.i./ha applied on July 1 and July 8; T2: sulfoxaflor treatment, 100 g a.i./ha applied on July 1 and July 8.

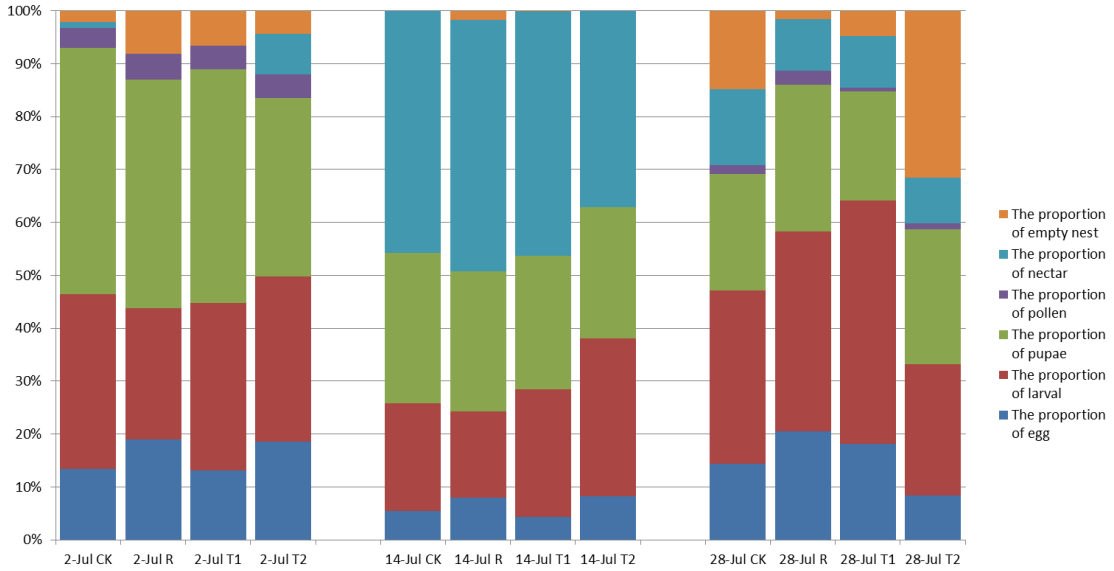

Figure 7. Change of colony conditions before exposure, on the second day of the end of exposure and on 14 days after the end of exposure. CK: water control; R: reference substance treatment, dimethoate 600 g a.i./ha on July 8; T1: sulfoxaflor treatment, 75 g a.i./ha on July 1 and July 8; T2: sulfoxaflor treatment, 100 g a.i./ha on July 1 and July 8.