WORKER BEES (*APIS MELLIFERA* L.) ARE ABLE TO UTILIZE STARCH AS FUEL FOR FLIGHT WHILE DRONES ARE NOT

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Abstract

When old worker bees, sampled from a feeding station, were fed a 2 molar glucose solution which additionally contained an equivalent of soluble starch of 1 molar glucose, they increased the period of flight in a roundabout by 24 percent. Flight-drones which were fed with the same amounts of sugar and starch did not increase, but instead reduced flight time significantly by 23.6 percent, compared to when they were fed a pure 2 molar glucose solution. The two feeding regimes did not alter the mean velocity in workers and drones as well. Our result indicates that workers can utilise starch during flight while male honey bees cannot. This provides evidence that the enzyme amylase found in the hypopharyngeal glands of older workers increases their efficiency of utilising complex carbohydrates and underlines their important role as food processors in the highly evolved social system of the honeybee society.

Keywords: *drone / flight metabolism / starch / enzyme / digestion*

Introduction

In honeybees the highly evolved division of labour includes the performance of many various tasks with age and even involves digestion. Workers of nursing age consume most of the pollen brought into the colony by foragers and produce proteinaceous jelly (brood food) in their brood food glands, which is distributed to the brood, and also to other workers, drones and the queen (CRAILSHEIM 1992; CRAILSHEIM et al, 1992; HRASSNIGG and CRAILSHEIM, 1998a, b). Workers are highly effective in digesting the ingested food, nectar and pollen. This is reflected by the production of enzymes like saccharase (SIMPSON et al., 1968) or amylase in the hypopharyngeal glands, which digest carbohydrates, as well as by the presence of proteolytic enzymes in their midgut, mainly used to degrade pollen proteins (MORITZ and CRAILSHEIM, 1987). The production of these enzymes strongly depends on the workers' occupation. While in younger workers (nurses) the hypopharyngeal glands produce mostly proteinaceous jelly, they change their synthesising activity in older workers (foragers) producing mainly carbohydrate digesting enzymes (HALBERSTADT, 1980; KUBO et al., 1996). Amylase and saccharase degrade complex carbohydrates into their monomers, while glucose oxidase helps to preserve the honey by the production of small amounts of hydrogen peroxide. These carbohydrates are also found in honey stored in the combs.

The male gender lacks hypopharyngeal glands (SNODGRASS, 1956). Drones are not involved in the process of collecting or storing food reserves for the colony. Their primary and, as far as known, their only task is to produce sperm and to mate with a queen. In order to meet this challenge they are strong and forceful flyers (GMEINBAUER and CRAILSHEIM, 1993). They derive the energy for these flights from honey, which they ingest from the stores in the colony.

It is known that workers are able to digest starch, but whether this process is quick enough to sustain flight metabolism is not clear so far. It is neither known whether drones can digest starch nor if they can utilise it for flight. Therefore we address the question: Are honey bee workers and drones able to utilise starch in their flight metabolism?

Material and Methods

To investigate flight metabolism, workers and drones were prepared to fly in a roundabout already described by HRASSNIGG and CRAILSHEIM (1999). If starch is utilised in flight metabolism of bees, an additional supply of starch in a glucose solution should increase flight time and/or the number of rounds flown by the bees in a roundabout. If starch is not utilised no such increase in flight time should be found.

The workers were sampled from a feeding station, established near the colony, providing a 1 molar sucrose solution. Drones were taken from side frames of a colony, where older drones are usually found. The bees were prepared to be attached to the arm of the roundabout. First, the animals were stimulated to fly without additional feeding to deplete their energy (i.e. sugar) reserves. Then they were fed defined amounts of glucose or glucose-starch solutions and were stimulated to fly again. Every turn flown by the bees was recorded by computers and the overall flight time was additionally clocked by hand, so that only the active flight period was taken into account for further calculations.

After the first flight the workers and drones were fed with 10µl of a pure glucose solution (2M), and stimulated for flight after a resting period of five minutes. When they were exhausted, they were taken from the arm of the flight mill and fed with a glucose-starch solution (concentration of glucose 2M and of starch calculated as anhydroglucose 1M). For the experiments we used pure glucose, and soluble starch (according to ZULKOWSKY) from Merck. The amount of starch was calculated for the monomers of it, as anhydroglucose ($C_6H_{10}O_5$) with a molecular weight of 162 g. We increased the amount of starch by 10%, to compensate for the water bound to it physically.

Results

The flight period of workers in the roundabout was 19% longer than in drones, when same amounts of glucose (10µl, 2M) were fed (fig. 1).

When drones were fed a pure glucose solution (10µl, 2M glucose), they flew significantly longer than when they were fed glucose plus starch (10µl, 2M glucose and 1M anhydroglucose). So, the flight period of drones, which were additionally fed with starch, was reduced by 23.6% (fig. 1).

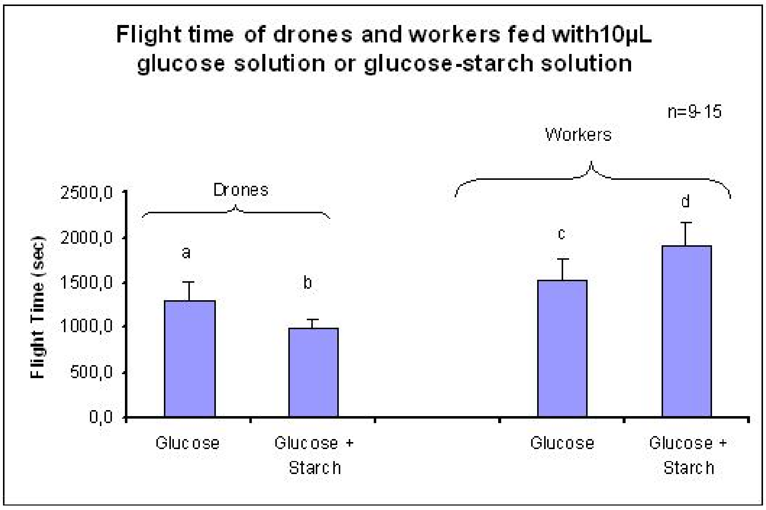

Figure 1 - Total flight time of drones and workers fed either 10µl of a 2 molar glucose solution or 10µl of a 2 molar glucose plus starch solution, which equals a 3 molar glucose solution. Identical letters above columns indicate no statistically significant difference (ANOVA; P<0.05)

Workers fed with the same amount of glucose and starch increased their flight time by 24.2%, compared to workers fed only glucose. No significant difference in the total flight period was found, when workers were fed with 10µl of a 3 molar glucose solution or 10µl of a solution containing 2M glucose and 1M anhydroglucose.

The distance flown in the roundabout by workers fed glucose (10µl, 2M) was significantly shorter than that flown by workers fed glucose plus starch (10µl, 2M glucose and 1M anhydroglucose), which was 1665.0±199.1 m (n=11) and 2249.4±331.1 m (n=15) respectively (U-Test, $P<0.001$). The same feeding regimes made drones fly with glucose a distance of 1727.0±357.3 m (n=9) and with glucose plus starch a significantly shorter distance of 1380.7±153.1 m (n=9) (U-Test, $P<0.05$). While workers and drones, which were fed pure glucose solution, flew nearly equal distances ($P=0.70$), drones flew significantly shorter when they were fed additional starch ($P<0.001$).

Our results demonstrate that the enzymatic equipment of foragers enables them to utilise not only sugars but also starch for their intense flight metabolism, while this is not the case in drones, which even show reduced flight performance when they ingest starch.

Discussion

Starch is not a main constituent of honey bee nutrition. It is present in pollen in different quantities, for *Zea mays* a relatively high content of 22.4% is documented (STANLEY and LINSKENS, 1985). It is estimated that a honey bee colony consumes on average 80 kg of honey and 20 kg of pollen per year (SEELEY, 1985). Assuming for the pollen a mean content of starch of 10% of dry weight will give an additional amount of 2.0 kg of anhydroglucose, which equals approximately 2.6 kg of honey. This probably is still an overestimation as it assumes that all the starch present in the pollen is digested and absorbed. Compared to the total amount of carbohydrates (i.e. 82.6 kg: honey plus starch) the energetic equivalent of starch makes only an estimated portion of 3.1 %. This seems to be relatively little, and it will probably be still less in the light of the fact that entomophilic plants produce less starch than aerophilic plants (cf STANLEY and LINSKENS, 1985). So that we may speculate that the amylase in honeybee workers is not only important to increase the energy supply, but may also play a role in the general process of pollen digestion.

Nevertheless, workers in contrast to drones are able to utilise starch in their flight metabolism, which enhances their energetic efficiency compared to that of drones. Our results also show that drones are specially adapted to the pre-processed food they take up from honey cells, so that there is no need for them to produce amylase to be able to digest starch. This is consistent with the findings that drones ingest very small amounts of pollen, which is also reflected in the reduced presence of proteolytic enzymes in their intestines (SZOLDERITS and CRAILSHEIM, 1993). Whether also queens, which belong to the reproductive female caste, are able to utilise starch, still remains to be investigated.

The reduced flight performance of drones in the experiment might be based on the adsorption of glucose to the molecules of the fed starch. Under natural conditions, before flights, drones provide themselves with honey from cells. In this way they also ingest enzymes produced by the workers. This was not the case in the present experiment where the drones were fed pure sugar or sugar-starch solution by the experimenter. So it might be possible that under natural conditions ingested starch could be utilised by the drones via the enzymes provided with honey or the workers' food. But this would just additionally underline the important function of the workers in processing food and in supporting the male gender.

Our results give an additional evidence for the highly evolved division of labor in honeybee colonies, where workers not only forage for food, but also pre-digest it for other members of the colony.

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