Locally produced honey is one of the foods least contaminated with residues of environmental pollutants and pesticides. Although the 1.5 kg per capita consumption of honey in Germany is negligible in comparison with the amounts of meat, vegetables and fruits that find their way into the shopping basket, honey is nevertheless subject to analytical testing as if it belonged to one of the main foods for human consumption.

Since 1988, pesticide residue analysis has been carried out at the Federal Institute for Apiculture at the University of Hohenheim in Germany on several thousand honey samples every year. Quality control is an important issue but the main focus is the identification of factors which may jeopardize the image of locally produced honeys. The results are primarily important for informing beekeepers. The criteria in honey analysis are very close to those of drinking water analysis, with determination limits in the low ppb range.

**Why these procedures?**

Where no maximum levels have been assigned, a reliable maximum value, usually 10 to 50 µg/kg, is used for plant food sources. As honey was considered a plant food source (today it is recognized as an animal food source) analytical methods were developed and established that comply with these legal maximum levels. In this way, honey – which as a natural product is regarded as being especially pure – is subject to very stringent quality control using highly sensitive analytical methods.

But even when using very sensitive methods, it is still quite difficult to detect pesticide residues in honey. The problem is that these pesticides are frequently used on cultivated plants in full bloom, while they serve at the same time as an important source of nectar and pollen for honeybees and other insects. Although pesticide contamination of nectar in orchard flowers or rapeseed fields can be detected easily, the search for pesticide residues in harvested honey often produces negative results. Why is this so?

**Honey in GC analysis**

To investigate this contradiction, laboratory, field and semifield studies as well as tent tests were carried out in recent years where the pesticides were traced from the flower up to the ready-for-harvest honey in the beehive.

The availability of reliable miniaturized extraction methods and highly sensitive measuring instruments was indispensable in these studies. The objective was to...
detect the active pesticide compounds originally applied to the plants, in the collected nectar of honeybees as transported in their honey sacs.

Analyses were carried out using Shimadzu’s GC-17A and GC-2010 gas chromatographs, each equipped with ECD detectors as well as a GCMS-QP5050A.

Depending on the type of plant and weather, honeybees usually visit between 50 and several hundred flowers before they return to the beehive with a full honey sac. During their nectar gathering flights they select one particular type of flower, meaning that they visit only one type of plant and also safeguard the pollination process (Figures 1 and 2). Under suitable experimental conditions, it is possible to guarantee that all collecting bees actually fly into the experimental plot – a rapeseed field that was sprayed with pesticide. Each bee transports on average approximately 40 µL of nectar. This nectar contains the active compound of the pesticide spray subsequently determined in the laboratory.

After returning from the field, the forager bees must be intercepted at the hive entrance. This is done using a converted automobile vacuum cleaner thatinstantaneously covers the bees with CO2 snow. In the laboratory the honey sacs, up to 2000 per experiment, are dissected individually (Fig. 3) and the target active compounds then isolated using liquid-liquid extraction methods and subsequently determined using gas chromatography.

The results show that the bees encounter mid ppm range pesticide concentrations in the flowers, which then find their way into the nectar. An interesting observation is that very high fluctuation margins are measured between individual bees in one series, although the bees have all been released into the flowering fields simultaneously. In the honey sacs of some of the nectar collecting bees extremely low pesticide levels – with 0,1 pg/µL being below the quantitation limit – were found. Based on the high number of visited flowers, a more evenly distributed pesticide level was expected in the honey sacs.

Bees under laboratory conditions

In the laboratory, caged groups of bees were fed with sugar solutions containing active pesticides. After a predetermined timeframe, the honey sacs of these bee samples were examined in order to determine whether active pesticide levels were being reduced within the honey sacs. Indeed, for some lipophilic compounds a radical decrease in the active pesticide levels could be determined. The active pesticide molecules apparently diffused into the tissue of the honey sac, which in turn suggests that the bees already reduced the pesticide content in the collected nectar during their collecting flights.

As the times that bees spend collecting nectar in the flower fields vary, this may be the cause for the strongly fluctuating measuring values. Bees clearly reduce the pesticide content acquired from the sprayed flowers already during their flight. The nectar delivered when the bees return to their hives already shows clearly reduced pesticide contaminations (Figure 4).

In the beehive, the nectar is processed into honey. The collected nectar is passed on from bee to bee. The worker bees enrich the nectar with endogenous substances and extract water from the honey during honey production. In this way, a self-preserving food substance is produced and stored in the cells of the honeycomb (Figure 5).

Further measuring sequences have shown that additional pesticide reduction processes take place during storage of honey, such as diffusion processes from the collected nectar into the beeswax of the honeycomb cells walls. These processes apply especially to the lipophilic active compounds and take place at the start of honey production when the water content is still relatively high.

The question as to why pesticides rarely present a problem with respect to honey quality, even though beekeepers have always been very critical regarding the use of pesticides in orchards or rapeseed fields, is now close to being answered.