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To cite this article: Giulio Loglio, Giovanni Formato, Marco Pietropaoli, Riccardo Jannoni-Sebastianini, Norman Carreck & Jozef van der Steen (2019) An Innovative Home-Made Beebread Collector as a Tool for Sampling and Harvesting, *Bee World*, 96:1, 16-18, DOI: [10.1080/0005772X.2018.1556905](https://doi.org/10.1080/0005772X.2018.1556905)

To link to this article: <https://doi.org/10.1080/0005772X.2018.1556905>



Published online: 14 Feb 2019.



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An Innovative Home-Made Beebread Collector as a Tool for Sampling and Harvesting

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Introduction

A honey bee colony represents a practical tool for bio-indication, as it collects material from a large area while foraging for food (Grodzinski & Yorks, 1981; Porrini, Ghini, & Girotti, 2002; Raes, Cornelis, & Rzeznik, 1992; van der Steen, 2016; van der Steen, Cornelissen, Blacquiere, Pijnenburg, & Severijnen, 2016). There are several matrices available for sampling from the hive: nectar, honey, pollen, beebread, bees, and wax. Beebread as a food differs from corbiculate pollen loads in being fermented bee pollen (De Grandi-Hoffman, Chen, & Simonds, 2013; Fuenmayor et al., 2014) and it includes pollen, honey, and secretions of bees' salivary glands (Barajas, Cortes-Rodriguez, & Rodriguez-Sandoval, 2012; Vásquez & Olofsson, 2009) (Figure 1). In general, it has a higher nutritional value than pollen, better digestibility, a richer chemical composition (Anderson et al., 2014; Carroll et al., 2017; Habryka, Kruczek, & Drygaś, 2016) and is better absorbed thanks to the partial fermentation of components (Barene, Daberte, & Sikсна, 2015).

Beebread contains 14–37% proteins, 24–34% carbohydrates, 3.2% lactic acid, 6–13% lipids, and 2.7% cellulose (Anđelković et al., 2012; Barene et al., 2015; Campos et al., 2008; De Grandi-Hoffman et al., 2013; Estevinho, Afonso, & Feás, 2011; Fuenmayor et al., 2014; Karmakar, 2015; Nagai, Nagashima, Myoda, & Inoue, 2004; Zuluaga, Serrato, & Quicazan, 2015). It further contains mineral components, vitamin K, and free amino acids (Gilliam, 1979; Nagai et al., 2004).

Mainly, technical reasons limit the use and commercialization of beebread. When beebread is intended to be used for research, medicinal, cosmetic or food purposes, it is necessary to collect it by hand, since this is the only way to guarantee the highest microbiological or chemical purity (Kieliszek et al., 2017). In this case, beebread is extracted manually, breaking the wax comb at room temperature or after freezing; using a large-mesh sieve, it is possible to collect the beebread in the form of small cylinders, separating them from the wax. It is possible to speed up the harvesting process using a strong flow of air that separates wax from the beebread. Otherwise, operators may use small metal or plastic tools to extract the beebread from the combs and send it to a laboratory. This procedure crumbles beebread and may not provide a clean sample, as there is the risk of including other foreign material (e.g., wax or exuviae) that may contaminate the purity of the sample and make the laboratory analyses and results less affordable.

Collecting beebread samples could be useful to:

- Detect residues of pollutants in the environment such as heavy metals, agrochemicals and varroacides (Conti & Botrè, 2001; Fleche, Clement, Zeggane, & Faucon, 1997; Gooley, Gooley, & Fell, 2018; Kubik et al., 1999; Leita, Muhlbachova, Cesco, Barbattini, & Mondini, 1996; Mullin et al., 2010; Orantes-Bermejo, Pajuelo, Megías, & Fernández-Piñar, 2010; Sattler et al., 2016).
- Evaluate the qualitative and quantitative composition of pollen collected by the bees.
- Verify the presence of toxic or allergenic pollens.

- Detect the presence of bee pathogens (e.g., spores of *Paenibacillus larvae*, *Melissococcus plutonius*, *Ascosphaera apis*, *Nosema* spp.) (Chen, Evans, & Feldlaufer, 2006; Gilliam, Prest, & Lorenz, 1989; Gilliam, Taber, Lorenz, & Prest, 1988; Hale & Menapace, 1980; Higes, Martín-Hernández, Garrido-Bailon, García-Palencia, & Meana, 2008; Mehr, Menapace, Wilson, & Sackett, 1976; Moffet, Wilson, Stoner, & Wardecker, 1978).

A New Tool for Sampling Beebread: the “Beebread Collector”

The purpose of this article is to describe a very simple and low cost tool designed to allow the easy extraction of beebread from comb cells (Figure 2A, B), invented by Giulio Loglio. It is intended to be home-made by interested beekeepers, such as those participating in citizen science projects, but also for those harvesting

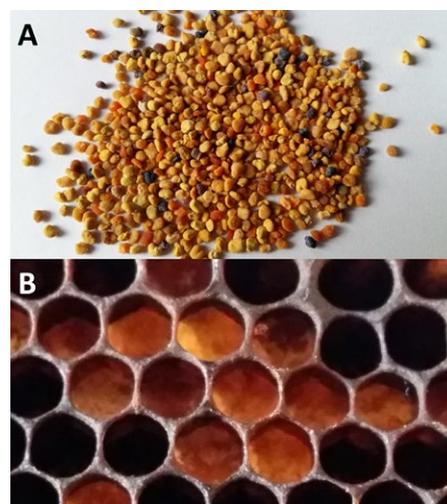


Figure 1. (A) Corbiculate pollen loads collected using a pollen trap. (B) Beebread cells stored in comb.



Figure 2. (A) The ready assembled beebread collector. (B) Components of the beebread collector: a small rigid tube; a plunger and a spring. (C) How to collect beebread: it is important to insert the beebread collector vertically into the cell. (D) A small pellet of beebread has been expelled from the beebread collector.

beebread for commercial use. The beebread collector can be made of various materials that can be in contact with food avoiding contamination of the beebread (glass, plastic, and steel). The main components of the tool are:

- A rigid tube, several centimeters long, with a diameter between 4.8 and 5.15 mm, the same as the honey bee cells. Commonly available drinking straws often have an external diameter of 4.8 mm. The tube, which has very thin walls, has a flattened top end.
- A plunger, such as a large steel nail with the point cut off, with a diameter of about 4.5 mm, which can slide into the rigid tube. The plunger has a flattened top end and must have a length so as to protrude from the rigid tube for 1–2 mm when fully depressed.
- A small spring 2–3 cm long inserted into the plunger able to bring the plunger in back position when it is not pressed.

To use the tool, the beebread collector is grasped like a pen and inserted vertically into a cell containing the beebread, exerting at the same time both a light pressure and rotary movement to allow the rigid tube to reach the bottom of the cell (Figure 2C). The outer part of the tube, sliding in contact with the inner walls of the cell, detaches the beebread from the cell wall and conveys it into the central part of the tube. To extract the beebread from the collector, simply press the plunger. The beebread is then pushed out of the tube in the form of a small pellet (Figure 2D).

Usually each cell of a bee comb contains about 75 mg of beebread and on average with the beebread collector it is possible to sample about 50 mg of it. To collect 10 g of beebread (the amount that is often required for laboratory analysis), it is thus necessary to insert the beebread collector into about 200–250 cells. The beebread collector can be cleaned with hot water and a small brush (e.g., dental brush), but if necessary to avoid cross contamination, due to its low cost it can be disposed and replaced by a new one.

It is possible to make the beebread collector more efficient and rapid by connecting the rigid tube directly to a vacuum pump so that the beebread, after being extracted from the cell, is deposited in a hermetic container. Two different solutions could be adopted:

1. An electric motor rotates the rigid tube which, inserted in the cell and acting like a small drill or a small cutter, crumbles the beebread.
2. An electric motor rotates in the rigid tube a small metal rod which, ending

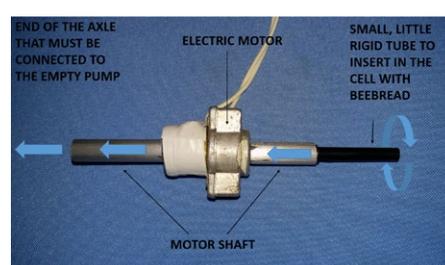


Figure 3. Beebread collector equipped with a small electric motor.



Figure 4. Differences between the beebread extracted with the manual beebread collector (pellets, on the left) and the motorized beebread collector (crumbles, on the right).

with a small corkscrew, crumbles the beebread (Figure 3).

In both solutions the beebread, vacuumed with a vacuum pump, is deposited in a hermetic container in a crumbled form and not in the form of pellets (Figure 4).

Conclusions

Beebread is an underused hive product that could have greater value both as a novel food and as a matrix for laboratory analysis to detect residues of agrochemicals, veterinary medicines used by beekeepers, presence of heavy metals in the environment or the presence of several honey bee pathogens. Our beebread collector is a tool that allows sampling in a fast, clean and uniform way, without altering the bee comb structure and guaranteeing the quality of samples for laboratory analysis. This simple and cheap tool will be used in the new INSIGNIA project, which is an innovative monitoring program using beekeepers as citizen scientists to study pesticide exposure of honey bee colonies and investigation of pollen sources.

Acknowledgements

This publication is part of the project “Citizen science investigation for pesticides in apicultural products” (INSIGNIA) – Multi Call PP-1-1-2018 – which has received funding from the European Union. Grant Agreement number: SANTE/E4/SI2.788418 – INSIGNIA – PP-1-1-2018. The content of this publication represents the views of the Authors only and is their responsibility; it cannot be considered to reflect the views of the European Commission or any other body of the European Union. The European Commission does not accept any responsibility for use that may be made of the information it contains.

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