POLLEN CONTENT AND BEE PREFERENCES: FACTS AND HYPOTESIS*

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Abstract

The types of substances found in and on pollen are discussed, especially those most readily absorbed by honey bees by virtue of their being superficial and/or soluble. Pollen contains proteins, lipids and carbohydrates. Lipids and proteins may be inside or outside the pollen grain, whereas carbohydrates are exclusively internal, and can be distinguised as monosaccharides (glucose and fructose), disaccharides (sucrose) and polysaccharides (starch and fructans). Starch and long-chain fructans are insoluble, whereas short-chain fructans are soluble. Honey bees seem to prefer pollen with external lipids, polysaccharides of the fructan type and a high sucrose content.

Key words: *pollen reserves, bee preferences, carbohydrate, starch, fructans.*

Introduction

Pollinator bees find rewards in flowers. Pollen is a non specialized reward because it has two purposes, namely, to transport the gametes and to reward pollinators. Nectar is a specialized reward because its only purpose is to attract pollinators. Pollen which is not collected and transported or consumed by pollinators is an investment that cannot be recovered. In certain species, nectar which is not consumed is reabsorbed by the plant (NEPI et al., 1996). Indirectly, plants also offer animals honey-dew. Pollen, nectar and honey-dew differ in composition and in their contribution to bee nutrition.

It was originally thought that pollen contributed to pollinator diet principally by virtue of its protein content. However it was recently found that pollen is a complete food, as it also contains lipids and carbohydrates (FRANCHI et al., 1996; PACINI, 1997). The viewpoint of this minireview is mainly botanical, its aims are to review recent research on pollen composition and to examine bee preferences with regard to pollen.

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Chemical composition of pollen and possible rewards for bees

Water content

Pollen may contain 2-50% water (PACINI, 1990). Pollen with a water content below 30% is known as partially dehydrated, it is the smallest pollen (30-100 µm) and is preferred by bees. Pollen with a water content above 30% is known as slightly dehydrated (SPERANZA et al., 1997); it is larger in size (100-200 µm) and rarely consumed by bees. It is taken up involuntarily by bees seeking nectar, as in *Cucurbita pepo*, *Lavatera arborea* and species of the genus *Heliconia* (NEPI and PACINI, 1993, 1999; PEDERSEN and KRESS, 1999).

Proteins

Pollen proteins may be located externally or internally. External proteins are known as sporophytic and may be located in pores (PACINI et al., 1981) or between the pores (PACINI et al., 1994). Because of their external location, sporophytic proteins are among the first to be digested. Whether internal proteins are accessed and digested depends on whether bee digestive juices can penetrate the pollen. The pores (which are where the pollen tube is emitted) are the weakest part of the pollen surface. In most pollen, the pores open when they find themselves in an acid environment; this process is known as "instant pollen tubes" (STANLEY and LINSKENS, 1974). Bee gastric juices create an acid environment and therefore gain access to the pollen cytoplasm.

Lipids

Like proteins, pollen lipids may be located internally and externally. External lipids include pollenkitt and tryphine (PACINI, 1997). These two materials probably have many of, if not all, their functions in common, however they differ in how they develop, and as far as we know, tryphine is limited to the family of the *Cruciferae*. Pollenkitt and tryphine, which consist mainly of lipids, surround the pollen grains of plants whose pollen is collected by bees.

The few exceptions to this rule are the genera *Oenothera* and *Acacia*, the *Poaceae* and some *Solanaceae*. Pollenkitt and tryphine have many functions (PACINI, 1997); those that regard bees are:

1. To hold pollen grains together in the air and stick them to the insect during flight.

2. To determine pollen colour. Pollen without pollenkitt is invariably yellow because it is pollenkitt that confers the colours black, red and green. The flower of *Lagestroemia indica* has two types of anthers which produce different coloured pollen: the yellow pollen is gathered actively and consumed by bees and does not fertilize; the green pollen, which has fertilizing capacity, is collected passively by

bees while they gather food pollen. The two types have similar morphology (PACINI and BELLANI, 1986) but differ with regard to polysaccharide reserves (our unpublished data). In *Lagestroemia*, bees are not attracted by the true (green) pollen but by the food pollen (yellow). Pollenkitt therefore hides pollen from bees so that they gather the other type.

3. To attract pollinators with odours or substances (DOBSON, 1991).

4. To allow compaction of pollen in corbicules and clumps (PACINI, 1997). The pollen of the tomato, *Acacia*, *Oenothera* and the *Poaceae* does not have pollenkitt and pollen compaction on the third pair of legs is achieved with other viscous substances or by other mechanisms.

Lipids may also be found inside pollen. Here they occur as spherosomes, namely spheres with a single membrane unit (PACINI, 1994). The types of lipids found in pollen may be neutral such as carotenoids, glycerides and unsaturated fats, or polar, such as phospholipids (PIFFANELLI et al., 1998). Carotenoids are presents only in pollenkitt.

Carbohydrates

Pollen carbohydrates may be mono-, di- or polysaccharides. Polysaccharides belong to the pollen walls or cytoplasm. Wall polysaccharides are mainly cellulose and pectins, the composition of which varies particularly in the pore regions. It was thought that the only polysaccharide reserve in pollen was starch (BAKER and BAKER, 1979), until FRANCHI et al., (1996) demonstrated the existence of cytoplasmic polysaccharides. These were recently shown to be fructans (our unpublished data). Fructans are a family of polysaccharides originating from sucrose molecules, to which a variable number of fructose molecules become attached (POLLOCK and CHATTERTON, 1988). They differ in degree of polymerization and solubility. Soluble fructans may diffuse when pollen is moistened. Fructans polymerize and depolymerize more quickly than starch because are not located inside plastids (KANDLER and HOPF, 1990). Fructans form by total or partial hydrolysis of stored starch in the last stages of pollen maturation (PACINI, 1996).

Sucrose concentrations in pollen vary from species to species (SPERANZA et al., 1997). This sugar is practically absent in *Cucurbita pepo*. In the apple and other species whose pollen is gathered by bees, sucrose content is high and may even exceed 10% (SPERANZA et al., 1997, and our unpublished data). The presence of sucrose in pollen is related to pollen life-span (HOEKSTRA et al., 1989); pollen containing little sucrose, such as that of the *Poaceae* (grasses with pollen rarely gathered by bees), only lives for a few hours.

Pollen that has a long life has a high sucrose content, such as pollen of *Chamaerops humilis*, which is gathered by bees (PACINI et al., 1997; SPERANZA et al., 1997, and our unpublished data).

The pollen cytoplasm also contains variable quantities of glucose and fructose which diffuse readily (SPERANZA, et al. 1997). No interpretation has yet been offered as regards the presence of these simple sugars, however in pollen gathered by bees they occur in quite high concentrations.

Characteristics of pollen gathered by bees

Three types of pollen can be distinguished on the basis of presence and types of polysaccharides: 1. pollen containing starch only; 2. pollen containing starch and fructans; 3. pollen containing fructans only. Table I shows the main characteristics of pollen in relation to whether or not it is gathered by bees.

Table I

Composition of pollen gathered by bees

Cytoplasmic features	Pollen not collected by bees Examples: *Poaceae* with few exceptions Pollen rarely collected by bees Example: *Zea mays*	Pollen not collected by bees Examples: *Malvaceae, Violaceae* Pollen collected by bees Examples: some *Rosaceae*	Pollen collected by bees Examples: *Boraginaceae, Compositae, Cruciferae, Labiatae, Leguminosae, Myrtaceae,* some *Rosaceae, Schrophu-lariaceae, Umbelliferae, Liliaceae*
starch	+	+	-
fructans	+ or -	+	+
pectines	+ or -	+	+
sucrose	low % or absent	Medium or high %	high %
glucose	low amount	Medium or high %	high %
fructose	low amount	Medium or high %	high %
pollenkitt	-	+ or -	Always present with few exceptions: *Oenothera, Acacia*, tomato

Bees prefer pollen in which fructans are the only polysaccharide reserves; pollen that contains starch also contains fructans. Bees also prefer pollen with a good sucrose content and surrounded by pollenkitt.

Families such as the *Cucurbitaceae*, *Euphorbiaceae* and *Ranunculaceae* are visited principally for nectar and their pollen contains starch and few fructans. Families visited for pollen and nectar, such as the *Boraginaceae*, *Compositae*, *Cruciferae*, *Labiatae*, *Leguminosae*, *Myrtaceae*, *Scrophulariaceae*, *Umbelliferae* and *Liliaceae* have pollen with little starch and many fructans. The *Rosaceae* are a special case, because species or varieties such as *Malus domestica* cv. Top red, *Mespilus germanica* and *Spiraea media* have starchless pollen whereas others, such as *Malus domestica* cv. Golden delicious and *Prunus persica* cv. Maycrest, have pollen containing starch (FRANCHI et al., 1996). Both types, however, have fructans as cytoplasmic polysaccharide reserves and are visited by bees for pollen and nectar (RICCIARDELLI D'ALBORE and PERSANO ODDO, 1978).

The *Poaceae* are another special case. The pollen of this family always contains starch, but the species visited more than any other, at least in Italy, is *Zea mays* (FRANCHI et al., 1996, RICCIARDELLI D'ALBORE and PERSANO ODDO,

1978) which has pollen with modest quantities of fructans (our unpublished data). Bees may gather maize pollen for lack of other available pollen within their range of flight (RICCIARDELLI D'ALBORE and PERSANO ODDO, 1978).

To conclude, we can say that honeybees prefer pollen without starch and in which the only polysaccharides are fructans. If there is starch, however, there are always fructans as well, i.e. cytoplasmic polysaccharides, some of which diffuse readily and are easily digested. Another reason for this preference is a high percentage of sucrose, except in *Zea mays* (SPERANZA et al., 1997). Other apoidea, such as *Osmia cornuta*, actively gather and are able to digest pollen starch (NEPI et al., 1997). Various species of the genus *Bombus* gather pollen of both *Helleborus foetidus* and *H. bocconei*, the former containing starch and the latter not (VESPRINI, unpublished data).

The fact that bees prefer pollen with fructans rather than starch could be due to the solubility of certain fructans and their easier break-down by bee gastric juices. Fructans are dispersed in the cytoplasm and are therefore in tiny compartments, whereas starch is in larger compartments with a smaller contact area. There may be other reasons for bee preferences: a. pollen size: bees gather pollen in the 35-100 µm range; b. the presence, abundance and composition of pollenkitt; c. the ease with which ingested pollen releases its contents, i.e. cytological structure of the pore region.

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