



Identification of bee and wasp taxa relevant in systemic allergic reactions to Hymenoptera stings in Central Europe

Volker Mauss · Franziska Ruëff

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Abstract

Background Correct identification of the Hymenoptera that was responsible for stinging a patient is crucial when establishing the indication for specific immunotherapy (SIT) using Hymenoptera venom immunotherapy (VIT), advising the patient on allergen avoidance, and in terms evaluating accidental stings or sting challenges during VIT. There are around 15 species of social bees and wasps that most frequently cause stings in Central Europe. Even lay people are normally able to correctly classify causal insects into their respective families, i. e., Apidae or Vespidae. On the other hand, a more precise taxonomic classification and distinction within the subfamily and the genus is often imprecise or incorrect; for example, within the family Apidae (true bees), *Apis* (honeybees) is often mistaken for *Bombus* (bumblebees), while within the family Vespidae (folded-winged wasps), there is confusion between *Polistes* (paper wasps), *Vespa* (hornets), *Dolichovespula* (yellow jackets, long-cheeked wasps), and *Vespula* (yellow jackets, short-cheeked wasps).

Results The morphological characters used for the typing of relevant genera are described and illustrated. A redetermination of wasps used at university hospitals for sting challenge tests revealed that 7% did not belong to the desired genus *Vespula*, but mainly to *Dolichovespula* instead. However, sting challenge tests using *Dolichovespula* species elicit ap-

proximately the same percentage of systemic allergic reactions as do *Vespula* stings.

Conclusion With training, even nonentomologists are able to distinguish between taxa using simple techniques. The fact that a similar number of systemic allergic reactions are seen with sting challenge tests using *Dolichovespula* as with challenge tests using *Vespula* may be due to the high allergen affinity between the venoms. Another possible explanation for this could be that allergic reactions are more commonly triggered by *Dolichovespula* venom compared with *Vespula* venom than originally assumed.

Keywords Allergen immunotherapy · Desensitization, immunologic · Insect venom · Allergy · Determination

Introduction

Information on the occurrence and stinging risks of the various Hymenoptera species is important to allergists in their evaluation of which insect was causal to a systemic sting reaction. This applies not only to Germany, but also to other countries, since it is not uncommon that the stinging incident responsible for presentation occurred in a country other than Germany.

It is often necessary and technically possible to determine which species the stinging insect belongs to. Particularly when interpreting sting challenge tests or field stings in patients during specific Hymenoptera venom immunotherapy (VIT), it is important to ensure that the stinging insect belongs to the species with whose venom the treatment is being performed. Only then—alongside taking other aspects into consideration such as strength of the local reaction (did the insect actually discharge a noteworthy amount of venom with the sting?) and the use of an emergency kit—is it possible to draw any conclusions about the

Dipl. Biol. V. Mauss (✉)
 Abteilung Entomologie, Staatliches Museum für Naturkunde
 Stuttgart, Rosenstein 1, 70191 Stuttgart, Germany
 volker.mauss@gmx.de

F. Ruëff
 AllergieZENTRUM, Ludwig Maximilian University, Munich,
 Germany

Table 1 A guide to distinguish between stings from honeybees (*Apis mellifera*) and social wasps (*Polistes*, *Vespa*, *Dolichovespula*, *Vespula*) in Central Europe

Honeybee	Social wasps
The sting apparatus and abdominal tip is detached from the rest of the body following the sting and remain anchored by the stinger in the sting canal at the site of the sting	The stinger does not remain at the sting site (exceptions are possible if the patient compresses the wasp as it is stinging)
Lancets have long barbs	Lancets have short barbs
Brown in color	Striking black and yellow in color
Sting in the immediate vicinity of a bee colony or beehive (wild nests are virtually never seen in Central Europe)	Sting sustained close to a free-hanging nest with a paper envelope or an open paper comb without covering, or close to an entrance hole in the ground or on a facade
Sting sustained in area abundant with blooms, clover lawns are typical. Triggered by mechanical interference/irritation of the insect while visiting a flower, often when walking barefoot	Sting sustained around anthropogenic food sources (sugary drinks, beer, cake, barbecue food). Typically at the table, often after the insect has been struck. Also at bakeries or around sweet berries, grapes, fruit, often fallen fruit, e. g., from ornamental plum trees
Sting sustained around a garden pond or swimming pool where insects are floating stranded. On hot days, large numbers of workers may collect water, meaning that they get into water more frequently than wasps	Also collect water on hot days, but get into water less frequently, probably also due to the lower numbers
May collect sugar-containing solutions at refuse bins	Often at refuse bins, where they take up food
Stings are possible during the entire vegetation period, i. e., roughly between late February and early November. Significantly increased activity in the open in spring and summer	Colonies are founded from May onwards, increased activity from late June to early November, peaking in August and September. Hibernating queens may be brought into homes with firewood during the winter

onset of a clinically protective effect. Precise information on the insect species responsible for eliciting an allergy is also helpful in counselling sessions on allergen avoidance.

The risk of stings by Central European Hymenoptera species

Patients are often unable to give precise information on which species of insect they have been stung by, and the causal insect is only very rarely captured and brought to a medical visit, at least upon first sting reaction. In such cases, certain details on the circumstances of the sting can yield important information (Table 1).

Social bee and wasp species are the chief elicitors of Hymenoptera allergies in Germany; they are regularly found in residential areas, sometimes in dense numbers [1]. Stinging events involving bees (Apidae) can be attributed to the honeybee (*Apis mellifera*) in the vast majority of cases. In addition, eight other social bumblebee species (*Bombus* spec.) are regularly seen in residential areas, meaning that here again bumblebee stings are possible [2]. Bumblebees, as well as the honeybee races kept in Germany, are essentially considered placid. Stings primarily occur when the insect feels attacked and are more likely in close proximity to, rather than at a distance from, the beehive. Particularly worthy of note in this context is the readiness of the tree bumblebee (*Bombus hypnorum*) to defend its nest [2].

Within the folded-winged wasps (family Vespidae), stings can be predominantly attributed to two species in the *Vespula* genus, i. e., the common wasp (*Vespula vulgaris*) and the German wasp (*Vespula germanica*). These two species regularly nest in residential areas and develop significantly larger nest populations and

resultantly higher densities of individuals outdoors compared with other social wasps [3]. Moreover, since they are the only species to seek anthropogenic food sources [4, 5], they are most likely to be responsible for wasp stings in individuals eating or drinking outdoors, e. g., at coffee tables, bakeries, beer gardens, or while barbecuing.

At least four other social wasp species also live in the immediate environment of humans in Central Europe [2]. Stings by these species occur primarily at the nests and near ripe fruit, e. g., when plucking berries or collecting fallen fruit. A sting that occurs at a wasps nest made of paper and consisting only of a single exposed comb, i. e., with no outer envelope, is a sting from a paper wasp (*Polistes*). Stings incurred in the vicinity of pear-shaped nests with an outer envelope and hanging above ground level in bushes or hedges can be attributed to *Dolichovespula* species. These stings typically occur while cutting hedges.

More detailed information on the bionomics and defensive behavior of Central European bee and wasp species relevant in Hymenoptera venom allergies has already been discussed in earlier publications [1, 2, 6], as has information on the risk of stings at various holiday destinations [7].

Identifying insects for sting challenge tests and following field stings

Whereas it is virtually impossible to reliably identify the causal insect in the case of field sting reactions prompting patients to present to an allergist, such identification is technically possible and also necessary on other occasions. When performing sting challenge tests [8, 9] to identify patients not protected by VIT, reliably determined animals should be used or determinations verified in the context of quality con-

Fig. 1 Species membership of the social wasps used in sting challenge tests at two allergy centers in Germany between 2006 and 2016 based on the redetermination of the specimens as part of quality assurance

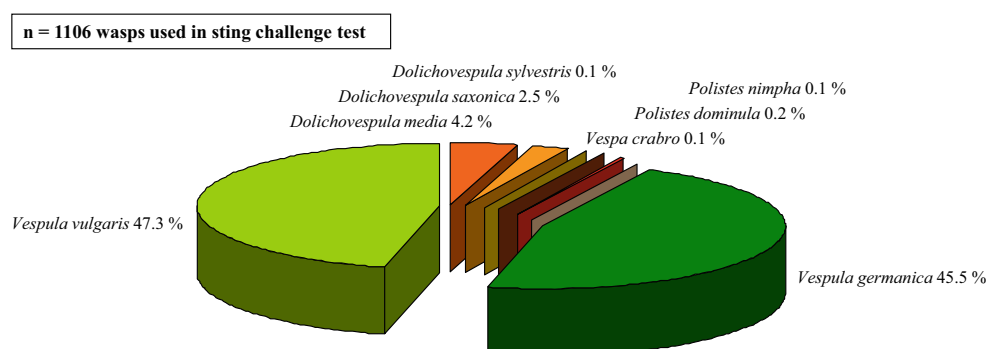


Table 2 Reactions in 623 patients to sting challenge tests using different social wasp species from the genera *Dolichovespula* and *Vespa* conducted at the Munich University Hospital, Germany

Species used for sting challenge	SAR upon sting challenge						Σ
	Negative		Positive		Unclear ^a		
<i>Vespula vulgaris</i>	241	(92%)	8	(3%)	13	(5%)	262
<i>Vespula germanica</i>	308	(95%)	11	(3%)	7	(2%)	326
<i>Dolichovespula saxonica</i>	18	(95%)	1	(5%)	0	(0%)	19
<i>Dolichovespula media</i>	11	(69%)	3	(19%)	2	(12%)	16
Σ	578	(93%)	23	(4%)	22	(3%)	623

SAR Systemic allergic reaction, Σ, total

^aNonverifiable reaction not requiring treatment, cannot be conclusively defined

trol. In the case of honeybees, the procurement of bees presents no difficulties prior to sting challenge tests, since workers can be obtained directly from beekeepers, thereby ensuring the identity of the species. Where possible, bees that leave the beehive should be used for sting challenge testing, since one can assume that their sting apparatus is fully developed and that the venom reservoir is filled with venom.

Social wasps, on the other hand, can only be collected from the outdoors or outdoor nests and therefore it has to be verified which species they belong to. If insects can be taken repeatedly from particular nests, conclusive identification of an individual is worthwhile. Since the determination of live insects is not straightforward for the nonentomologist, nor is it without hazard, insects should be killed following sting challenges and redetermined as part of quality management. When redetermining wasps used for sting challenge tests, it became evident—and that in hospitals with great expertise in sting challenge testing—that, although insects belonging to the desired species in the genus *Vespa* had been used for sting challenges in the vast majority of cases, other species, in particular members of the genus *Dolichovespula*, had also been used inadvertently on occasion (Fig. 1).

A subanalysis of 623 sting challenges in patients hyposensitized to wasp venom conducted at the Munich University Hospital, Germany, [as yet unpublished data] revealed that, taking the highly varying case numbers into consideration, approximately the same percentage of patients exhibited systemic reactions to stings from *Dolichovespula* as they did to stings from *Vespa vulgaris* or *V. germanica* (Table 2).

It is essentially unlikely that treatment with *Vespa* spp. venom would also provide sufficient protection against *Dolichovespula* stings; having said that, the same percentage of systemic sting reactions seen with *Dolichovespula* shows that one does not necessarily, at least, need to fear a “false negative” result in sting challenges using *Dolichovespula*. Conversely, one should even consider the possibility that patients who react to sting challenges using *Dolichovespula* despite VIT with *Vespa* venom more often have a primary allergy to *Dolichovespula* rather than to *Vespa*.

If it can be reliably shown that a patient has been stung by the “wrong” insect, and if this insect is *Polistes* spp., sting challenge should be repeated, since the *Polistes* sting sustained is of no relevance to the question of whether VIT has produced a protective effect. *Dolichovespula* stings are likely to be more reliable. In the case of systemic sting reactions to a *Dolichovespula* sting, one should investigate whether the original stinging event was not possibly caused by *Dolichovespula* spp. Where this is the case, attempts should be made to additionally perform the treatment with *Dolichovespula* venom. Although not commonly used in Germany, venom from *Dolichovespula arenaria* (aerial yellow jacket), *Dolichovespula maculata* (bald-faced hornet), and *Polistes* spp. are produced by ALK Denmark, for instance, and are approved in the US. With a certain degree of organizational effort, they can be obtained through international pharmacies. Since the therapeutic allergens are not approved in Germany, one needs to bear in mind all the arrangements required

for off-label use, including the clarification of cost-coverage.

As a basic principle, the predictive value of a sting sustained in the field is lower compared with a sting sustained during sting challenge [10], likely due primarily to the unreliable identification of the insect. In the case of field stings, patients on VIT should attempt to retrieve the causal insect as evidence—only assuming, naturally, that this can be done without hazard to themselves or others. If emergency medication was not required and also not used prophylactically and a significant local reaction occurred, the field sting sustained from the disease-causing insect also yields information on the presence of a protective effect.

A guide to determining the relevant taxa

In order to minimize errors in determination, the following summarizes the characteristic and readily identifiable characters that permit conclusive identi-

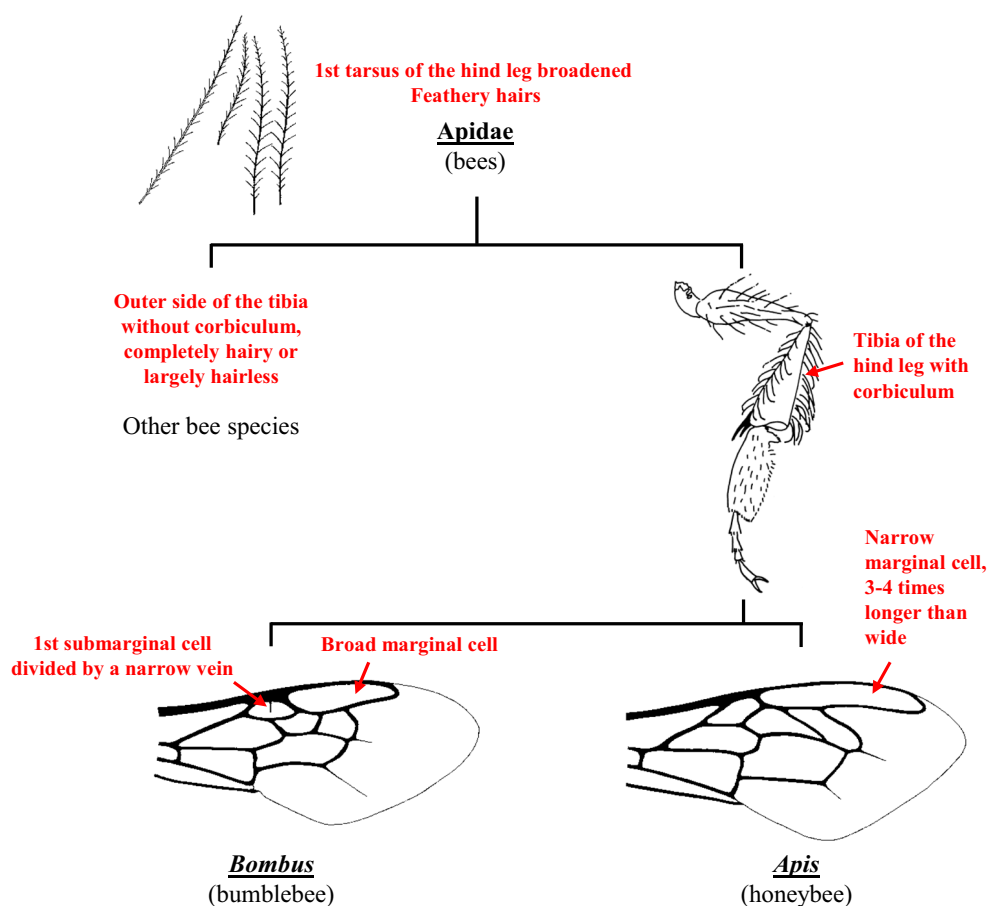
fication of the groups. It is generally not necessary in routine practice to distinguish on a species level between all the ca. 23 social bee and wasp species belonging to the subfamilies Apinae, Polistinae, and Vespinae [2] regularly or occasionally found in residential areas. Instead, assigning the insects to the correct genus is sufficient in order to provide patients with, e.g., information on the need to move nests or take other precautionary measures, as well as to exclude unsuitable individuals from sting challenge testing. In this way, determination problems are reduced to distinguishing between only six taxa, i.e., the genera *Apis* (honeybees), *Bombus* (bumblebees), *Polistes* (paper wasps), *Vespa* (hornets), *Dolichovespula* (yellow jackets, long-cheeked wasps), and *Vespula* (yellow jackets, short-cheeked wasps).

The characters used to this end can all be identified on the live insect. Determination is made easier by using a magnifying glass. Alternatively, simple supermarket reading glasses (reading aid) can be used

Fig. 2 Members of the Aculeata relevant in Hymenoptera venom allergy and the position of important characteristics



Fig. 3 Quick-check key for the reliable identification of honeybees (*Apis mellifera*) and bumblebees (*Bombus spec.*) (Apidae: Apinae)



and also combined with existing glasses where necessary. Difficulties arising due to the fact that the insects are highly agitated can be resolved by putting them in a small, transparent container (e. g., a snap cap vial, 65 × 30 mm) and placing this in a cool, dark place, e. g., refrigerator or cooler bag, for a certain period of time. Insects need to be brought back to room temperature for sting challenge tests.

The reader is referred to earlier publications for the determination of bumblebee species [11, 12] and wasps [13, 14].

Characteristic features of the honeybee and bumblebees (Apinae)

Membership of aculeates to the bees (Apiformes) can be unequivocally identified by the fact that the first tarsus of the hind legs is considerably broadened, thereby differing markedly from the following “normal” tarsi (Fig. 2). In the other groups of aculeates, the first tarsus is the same width as the following tarsi. The bees also have feathery hairs¹, i. e., at least some of the hairs have fine branches (Fig. 3). This is

best visualized on hairs located on the thorax using a stereo microscope².

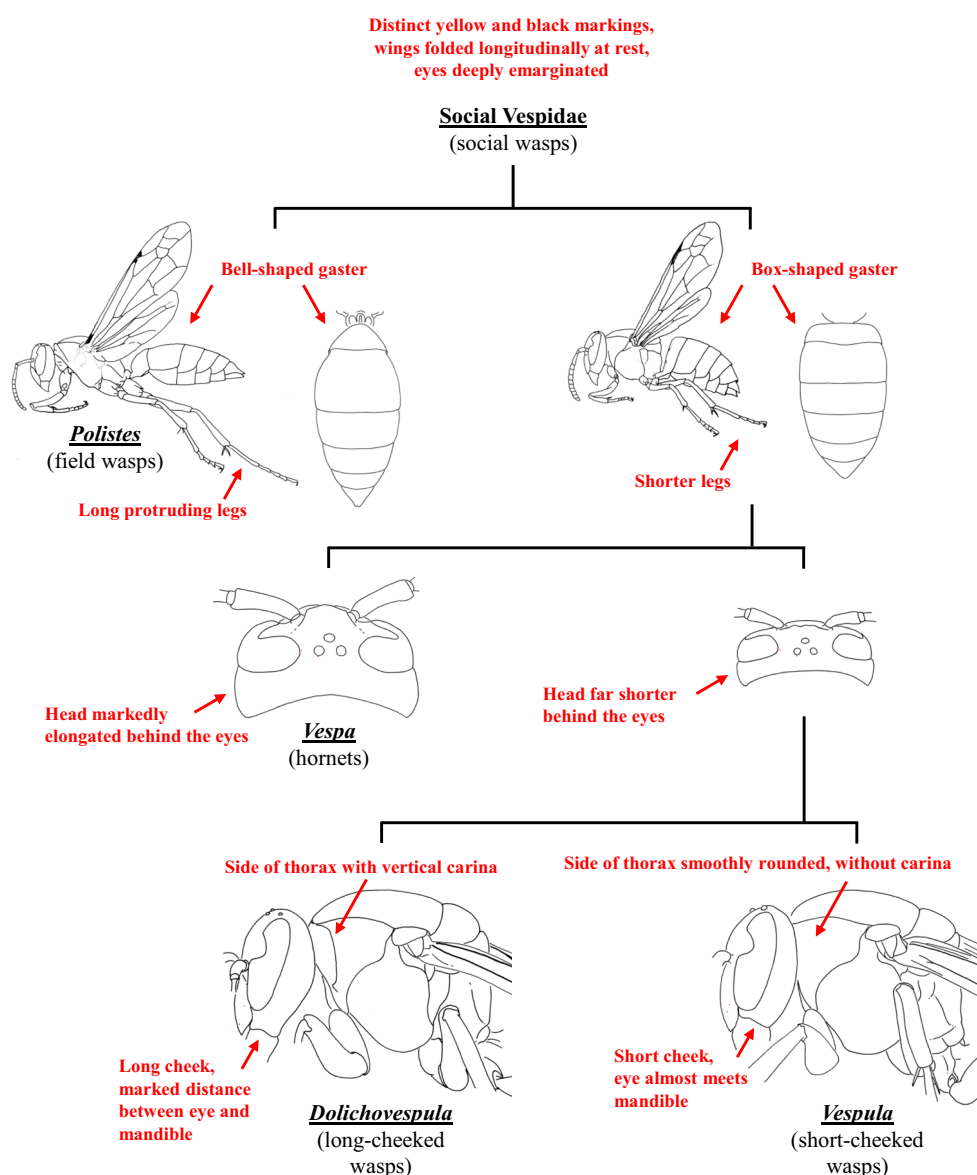
Honeybees and bumblebees are the only native bees to have corbicula (pollen-baskets) (Figs. 2 and 3). The corbiculum, located on the outer side of the hind legs, is designed to store pollen. It consists of long hairs along the anterior and posterior edge of the tibia of the hind legs, bordering the largely flat, highly shiny and hairless outer side of the tibia to the front and the back. This creates a cavity surrounded by long hairs, which is filled with pollen during foraging. When the corbicula are full of pollen, the bees look a little as if they are wearing trousers, which is why in German one also refers to the process of filling the corbicula with pollen as “Höseln”.

The honeybee is distinct from the bumblebees, as well as all other bees, by virtue of a narrow and distinctly elongated marginal cell on the forewing that is approximately three to four times longer than it is wide (Figs. 2 and 3). This marginal cell is significantly wider and shorter in bumblebees (Fig. 3). In addition, honeybees are considerably slimmer with a more elongated form compared with bumblebees, which have far stouter bodies (Fig. 2). Honeybees and

¹ The morphologically correct term is setae, since insect setae are not the same as mammal hairs. However, for the sake of easy comprehension, the term hair has been used throughout the text in place of seta.

² The morphologically correct term is mesosoma, since among Aculeata the thorax is fused with the first abdominal segment to form a new body section, the mesosoma. The remaining part of the abdomen forms the metasoma or gaster.

Fig. 4 Quick-check key for the reliable determination of the indigenous species of social wasps (Vespidae: Polistinae und Vespinae)



bumblebees also differ in terms of coloring: although honeybees have simple brown hair, the gaster appears striped due to lighter and darker rows of hairs (Fig. 2). Bumblebees, in contrast, are brightly colored, with yellow, orange, red, brown, white, and black elements in varying arrangements depending on the species.

Characteristic features of the social wasp (Polistinae, Vespinae)

Wasps differ from all other native insect groups by virtue of their wings, which are folded longitudinally at rest (Fig. 2) and their deeply emarginated, kidney-shaped eyes (Fig. 2). The surface of their body also shows the typically striking yellow and black warning colors (Fig. 2), which can also be seen in some other groups of insects that mimic wasp markings.

Paper wasps belonging to the genus *Polistes* (subfamily Polistinae) have a bell-shaped gaster and long

legs that protrude conspicuously beyond the end of the body particularly during flight (Figs. 2 and 4). In addition, the by far most common *Polistes* species in residential areas, the European paper wasp *Polistes dominula* (Christ), is distinct from all other social wasps by virtue of its striking antennal flagellum, which is entirely orange in color (Fig. 2). The gaster of vespine wasps (subfamily Vespinae), in contrast, is box-shaped and their legs are shorter (Figs. 2 and 4). Moreover, they have black or brown antennae.

Hornets (genus *Vespa*) differ from the other members of the Vespinae at first glance due to their considerable body size and far more robust stature (Fig. 2). Only in hornets is the head elongated behind the eyes, such that the posterior ocelli are significantly further from the posterior margin of the head than from the compound eye (Fig. 4). In the other genera, the posterior ocelli are roughly at the same distance from the

posterior margin of the head as they are from the compound eye (Fig. 4).

Long-cheeked wasps (*Dolichovespula*) and short-cheeked wasps (*Vespula*) have a similar appearance and look like typical “wasps” (Fig. 2) that one knows from outdoor eating places, waste bins, bakeries, etc. (whereby these are always members of the *Vespula* genus). *Dolichovespula* wasps have a markedly elongated head in a downwards direction and long “cheeks” (Fig. 2). As a result, there is a clear distance between the lower edge of the compound eye and the upper edge of the mandible (Fig. 4). *Vespula* wasps, in contrast, have very short cheeks and the distance between the lower edge of the compound eye and the mandible is minimal (Fig. 4). In addition, *Dolichovespula* wasps have a prominent angular carina anterior-laterally on the thorax, while such a carina is lacking in *Vespula* wasps so that this part of the thorax is smoothly rounded (Figs. 2 and 4).

Conclusion

Correct determination of the causal insect forms an important basis when establishing the indication for VIT, counselling patients on avoidance measures, and evaluating field or challenge test stings. With the help of simple techniques, even nonentomologists are able to identify the relevant bee and wasp genera.

Conflict of interest V. Mauss and F. Ruëff declare that they have no competing interests.

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