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PERSPECTIVE

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Making way for the implementation of automated bee counters in regulatory risk assessment

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Abstract

Measuring adverse effects on honey bees and their colonies requires a suitable methodology. For example, due to the large number of bees in a hive and the foraging activity, measuring the mortality of individuals is a difficult task that has not yet been adequately addressed. Knowing the natural daily mortality rate of a bee colony would be of great benefit in assessing whether and to what extent external influences and stress factors affect mortality. More precise mortality data could in turn help refining specific protection goals for regulatory purpose. The European Food Safety Authority recently published a document that estimated such mortality rates based on a systematic literature review, but none of these rates were assessed from continuous monitoring of colonies. Currently, bee mortality is routinely evaluated with various types of dead bee traps that prevent deceased bees from being removed from the colony. Both the literature review and the dead bee traps are relevant to regulatory risk assessment, but in our opinion are not describing the total mortality. Bee counters capable of precisely determining daily loss rates meet the above points and combine them with generating automated and continuous monitoring data. Lately, the field has gained a lot of importance in research and technological advances offer new possibilities in regulatory risk assessment. We will highlight these possibilities and discuss their future application in practice.

KEYWORDS
automated bee counter, background mortality, dead bee traps, precision beekeeping, regulatory risk assessment

1 | BACKGROUND

The regulatory risk assessment of pesticides on bees follows a tiered approach that integrates both exposure estimation and effect assessment. This method progresses from conservative to more realistic evaluations, beginning with a simple screening based on standard data. As needed, complexity is added to refine the risk assessment, particularly when a high risk cannot be ruled out at the lower tier. In such instances, additional data from field or semi-field studies may be incorporated to enhance the precision of the assessment (EFSA, 2023). Hence, EFSA experts aimed to estimate the daily mortality rate for an entire honey bee colony or population in order to refine the specific protection goal of 7% acceptable loss on colony strength after pesticide exposure (it was recently set to 10%) (EFSA, 2021). In practice, the assessment of this so called "background mortality", i.e. the natural loss of adult bees from a honey bee colony during the season, proves to be difficult and challenging (EFSA et al., 2020). Continuous and automated measurements are required throughout a bee season, enabling a comprehensive understanding of the dynamic and fluctuating nature of bee populations over time (Odemer, 2022). Noting that background mortality is also highly dynamic, underscores the complex challenges of assessing the effects of pesticides on bee colonies under free-flying conditions.

Risk assessors and authorities agree that meaningful and realistic data can only be obtained by using whole bee colonies under such conditions (EFSA, 2023; EPPO, 2010). However, mortality rates can change depending on the state of the colony (nucleus, full-sized), seasonally high or low brood intensity, intensive foraging activity, and/or the presence of parasites or diseases. Hence, the background mortality of a bee colony is a rather dynamic parameter that requires continuous data assessment to capture these issues. Given the complex nature of bee behaviour, which is influenced by various factors, assessing non-lethal or so called sublethal effects of pesticides proves difficult (Chmiel et al., 2020). These effects include physiological or behavioural changes in individuals that survive pesticide exposure (Desneux et al., 2007). Yet isolating and attributing specific effects to pesticide exposure is not easy. For example, the successful return of foragers to the hive, which is critical for colony survival, may reveal potentially negative effects on bee cognition and memory. These effects may impact entire colonies and even affect the health of the population (Chmiel et al., 2020). Consequently, pollination success and food safety could be at risk.

In the past, several methods were established for the higher tiers to assess and monitor bee mortality which are used in risk assessment to obtain specific endpoints (Medrzycki et al., 2013). These standard risk assessment methods, though, have mutual shortcomings. None of them jointly covers the difficulties described above; (i) to continuously measure the daily mortality rate of a bee colony (i.e., background mortality) and to account for (ii) lethal, and (iii) sublethal effects caused by external stressors. Most importantly, however, there are very few methods that are fully automated. The vast majority are based on manual assessments and are therefore laborious and susceptible to bias, which requires constant quality assurance. Moreover, these methods are not designed to collect data over longer periods of time, such as a whole bee season.

2 | STANDARD METHODS

Normally, dead bee traps are used to monitor mortality inside the hive. So-called undertaker bees carry deceased bees and brood out of the hive to protect the colony from disease (Wen et al., 2023). The trap prevents the deceased bee from being permanently removed from the hive and catches it in front of the hive entrance (Human et al., 2013; Illies et al., 2002). In this way, the daily deadfall can be recorded. Dead bee traps are easy to apply and account for lethal effects. They are widely used in risk assessment, covered by several international test guidelines (EPPO, 2010; OECD, 2014; Oomen et al., 1992).

An issue with using such traps is that long-term monitoring over several weeks or months is challenging. Even if dead bees are removed daily, which alone is labor-intensive, the bees see the trap as part of their hive and begin to clear it out the longer they are used to the trap (Illies et al., 2002). Furthermore, the recovery rates of different trap models vary between 71% and 96% (Human et al., 2013, table 16), which may yield biased and less comparable results in the end. The recovery rate is the efficiency of a trap model to capture all dead bees within a given time. Although alternative trap models exist, they all face another problem. Predators such as wasps, hornets, ants or earwigs decimate the dead bees more or less quickly (Human et al., 2013). As a result, recovery rates further decrease and long-term monitoring of mortality with this method suffers from severe shortcomings.

Moreover, dead bee traps only capture bees that have died inside the hive, were sick or of old age. Bees that die outside or bees that cannot navigate back to the hive are not recorded by this method, although this is relevant for risk assessment. In semi-field studies, where hives are placed in tunnel tents, outside bee mortality can be partly assessed by counting dead bees on the ground in surfaces covered with sheets near the foraging area (EPPO, 2010; Pistorius et al., 2012). Of course, this is only possible to a very limited extent in field studies. There, only a small area in the field is covered with sheets on which dead bees are counted. In addition, sublethal effects of pesticides may increase the number of bees unable to return to the hive because their orientation was affected (Chmiel et al., 2020). Dead bee traps cannot capture the impact of such effects on honey bee navigation. The nuanced behavioural changes induced by pesticide exposure that are not measured by conventional traps reveal a crucial gap in our understanding.

Addressing this gap, the most recent approach to monitoring sublethal effects on honey bee navigation involves the adoption of OECD Guidance Document No. 332 "Honey bee (*Apis mellifera* L.) homing flight test, using single oral exposure to sublethal doses of test chemical" (OECD, 2021). This innovative method uses RFID (radio-frequency identification) technology to tag individual bees that have been exposed to an acute dose of a pesticide. Bees released at a known location and distance are recorded on their return to the hive. The effect is measured by the proportion of bees that do not return within a certain time in relation to negative and positive controls.

While RFID technology is valuable for tracking the lifespan of individuals, it may have certain limitations that need to be considered, especially in this test system. For example, the additional weight of the tag or chemicals in the glue may interfere with the bees' natural behaviour and alter flight dynamics (de Souza et al., 2018; Susanto et al., 2018; Toppa et al., 2021). In addition, the technology may be too costly for large-scale experiments. Given these limitations, it is important to carefully evaluate the potential impact of this method on the physiology and natural behaviour of bees, especially in the context of pesticide exposure studies in risk assessment.

Recording bees that do not return to their hive for natural or environmental reasons would in fact benefit from a more automated and holistic approach. By tracking individual foragers, specific age cohorts or sexes (drones, queens, or workers) can be investigated. At the same time, evaluating the total traffic of a colony provides insights into daily losses and general activity patterns (Odemer, 2022). A combination of both would therefore be highly preferable.

3 | THE PATH FORWARD

The first automated bee counter was introduced 100 years ago, but development in the sector has been slow to date. Recently, there have been more counters on the market than ever before, and scientific approaches are published in a large variety (Odemer, 2022). Over the past decade, more than 50% of the scientific literature on bee counters spanning a 40-year period has been published, with $n=270$ publications (Figure 1, Web of Science Core Collection, Method S1). The steep increase in publications is an unmistakable sign that this field is highly relevant and advancing. We expect that within the next 5–10 years, bee counters and automated hive monitoring (i.e., precision beekeeping) will be available in a quality suitable for scientific and regulatory needs. This means that it must be possible to validate any automated device that records, for example, flight activity, individual bee activity, hive weight, colony sound, temperature, and other factors. Without device validation, i.e. without the certainty that the counter is counting bees accurately, no transparent and reproducible data can be generated (Bermig et al., 2020).

Hence, to serve scientific and regulatory purposes, automated counters must meet important requirements. The following specifications of Struye et al. (1991) still apply to modern counters: (a) monitoring of all colony sizes, (b) no interference with normal bee behaviour, (c) ventilation and orientation behaviour should not be affected, (d) fully autonomous operation under field conditions, (e) user-friendly and low-maintenance design, (f) affordable enough to monitor more than one colony, and (g) low energy consumption to allow continuous operation. In addition, counters must record (h) reproducible data that can be easily extracted, and, (i) allow a robust validation of the counter error which is required to achieve reproducible results (Borlinghaus et al., 2022; Odemer, 2022). Without these last two points, use in the context of good laboratory practice (GLP), which is a prerequisite for pesticide risk assessment, would not be possible (Tausch et al., 2022).

In the past, sensor-based approaches have been used to establish reliable and field-robust counters. However, until now, the model by Struye et al. (1994) called "BeeScan" was the only one that was scientifically validated and commercially available. They used optical sensors, which were sufficiently reliable and inexpensive. More recently, capacitive sensor technology has shown the most promise in terms of reliability, maintenance, and precision. Bermig et al. (2020) introduced the "BeeCheck," which appears to be superior to Struyes' device in terms of ease of use. Capacitive sensors can be maintained with little effort, as frequent cleaning of the components is not necessary. Counters can be operated independently for several months

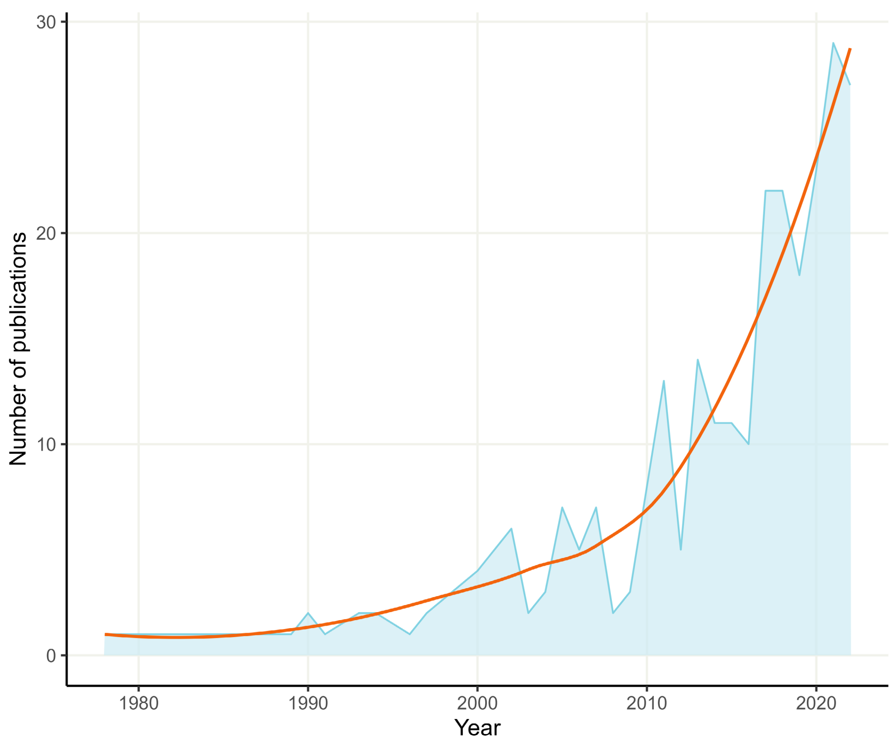

FIGURE 1 Publication trend related to bee counters over the last ~40 years (for more information see Method S1).

or even an entire bee season and have a replaceable data memory from which counting data can be easily retrieved (Odemer, 2022).

In addition to the more simplistic sensor technology, bee counters with video-based bee detection are commonly used to count traffic at the hive entrance. We can state that scientific publications describing video-based bee counters are growing beyond proportion compared to other types (Odemer, 2022). A major advantage of this technology is the ability to implement counting algorithms that use artificial intelligence (AI) and deep learning approaches to detect bees of any species (Bilik et al., 2021; Marstaller et al., 2019) if individually tagged. Moreover, the problems described by Odemer (2022) for sensor-based counters that lead to erroneous recordings, such as bees sitting in front of or in the entrance constantly triggering the sensor, oncoming or stuck bees, and bees moving back and forth in the sensor without actually leaving or entering the hive, can be adressed more easily with modifications to the software. We therefore expect this type of counter to be predominant in the long term even though there are some current limitations that need to be overcome.

Video-based bee monitoring faces significant challenges, primarily rooted in its dependence on lighting conditions, leading to sensitivity variations. When wide-angle lenses are used to capture the entire hive entrance, lens distortion contributes to false-negative registrations and false-positive results, further complicating accurate monitoring. Tracking algorithms show reduced efficiency with an increasing number of targets, and the variable flight traffic of honey bee colonies poses difficulties in maintaining accuracy. Additionally, the technique generates large storage requirements for video data, presenting challenges in data transfer and accessibility, a concern that may persist until the widespread availability of high-speed networks (Odemer, 2022).

4 | MOST RECENT ADVANCES

Despite all existing challenges, the integration of automated bee counters is gaining importance in research and extends to risk assessment by authorities (More et al., 2021). Current bee counters are capable of detecting both incoming and outgoing bees. With standardized methods to validate error rates, future counter development must focus on reducing errors and providing accurate counts (Odemer, 2022). This advance will allow background mortality in bee colonies to be determined more accurately, taking into account the condition of the colony, environmental influences, and seasonal changes. This is particularly relevant given that EFSA used constant mortality rates of forager bees derived from their flight span to establish an assumption for their background mortality rate (EFSA et al., 2020). Automated bee counters could validate these assumptions in line with the dynamics of a bee colony. As a result, a more nuanced assessment of factors affecting bee colonies, such as the specific effects of pesticides, can be achieved with less bias, significantly improving the rigour of risk assessment processes. In addition, video-based counters, as an example, can already identify different bee casts as well as pollen foragers returning to the hive (Yang et al., 2018), Varroa-infested foragers (Bilik et al., 2021; Bjerge et al., 2019; Voudiotis et al., 2022) and predators (pers. Observation, please see video footage in our data). As mentioned earlier, these counters allow detection of other bee species such as non-*Apis* bees with a simple adaptation of the algorithm (Borlinghaus et al., 2023; Knauer et al., 2022), and of course tracking of individual honey bees to follow their entire lifespan (Chen et al., 2012). Similar if not superior to what is currently described in OECD GD No. 332 (OECD, 2021) where the homing flight ability of only a limited number of forager bees is investigated.

This would obviate the need for RFID technology in this test system, and the insight into both the effects on individuals and the entire colony could be greatly enhanced by video-based counters (Figure 2). Sublethal effects occurring at the individual level rather than the colony level could be more easily detected and new meaningful endpoints generated. For example, total flight performance, forager recruitment or precise daily colony, and individual bee loss rates of certain cohorts would provide detailed information resulting from any environmental event that might have a negative impact on bee health. Furthermore, additional endpoints are proposed in the revised EFSA Bee Guidance Document (EFSA, 2023). It is suggested that the amount of pollen collected per flight and the number of bees returning with pollen should be monitored by an automated device to draw conclusions on adverse effects on foraging behaviour. EFSA is generally open to the inclusion of new technologies in risk assessment, even with regard to the assessment of bee colony strength (EFSA, 2023, annex C). They predict these technologies to substitute human observers in the near future.

Recent EU initiatives significantly contribute to advancing the precision beekeeping sector through substantial support, as highlighted in projects like B-GOOD (More et al., 2021). These developments are crucial due to the current lack of an accurate counter model to record daily bee losses, whether from natural causes like background mortality (Odemer, 2022) or other factors. Low error rates are essential for generating meaningful data from automated bee counters. The separation of hardware and software in video-based counters, as opposed to traditional sensor-based systems, offers a promising path for rapid progress. This is underscored by recent publications that show a recognizable trend towards video or image recognition in counter design (see Odemer, 2022, Figure 1) and highlight the industry's commitment to advancing high-resolution cameras independently of software development.

5 | CONCLUSION

Since EFSA has proposed the use of automated bee colony monitoring systems (EFSA, 2023), and the EU is actively promoting the development of such tools (More et al., 2021), we can expect their use in the future. Authorities outside the EU, such as the U.S. Environmental Protection Agency (US EPA) are also likely to be receptive to this progress, as we expect the existing test guidelines in

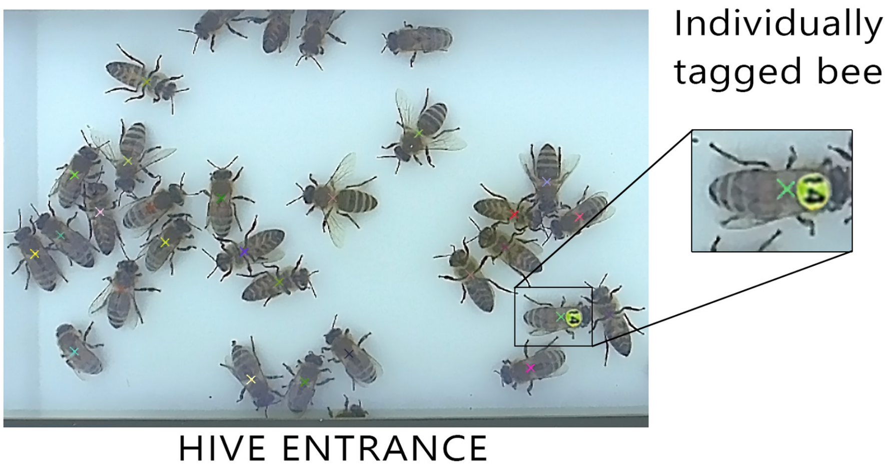

FIGURE 2 Recording both individually tagged worker bees (or drones) and the flight traffic of bee colonies with an automated video-based counter (from apic.ai).

risk assessment to be adapted accordingly. Once the current shortcomings have been overcome, a modern bee counter should have sufficient detection accuracy (i.e., low error rate) to meet the above tasks. With an accurate counter, daily background mortality can be measured and lethal impacts can be determined better and more conveniently than with current standard methods. Sublethal effects in terms of flight and foraging activity at both the individual and colony level, as well as homing success and forager recruitment, could be measured continuously with an automated bee counter. This will allow to fully assess the acute and chronic effects of natural and artificial stressors affecting honey bee colonies. Risk assessment would benefit greatly from these technological advances, and not just by designing more cost-effective studies. With a higher degree of automation, more replicates in honey bee studies are possible, and experiments gain reliability and validity. Given the possibility of adaptation to different bee species, a counter could also provide important data that account for differences in susceptibility to pesticides in other bees.

A look at the current possibilities shows that technological progress has already found its way into the approval process of pesticides, as demonstrated by the use of RFID to measure sublethal effects (OECD, 2021). Modern digital technology has also become an integral part of OECD protocols for recording the developmental stage of bee brood (OECD, 2014). Photographing brood combs and assessing individual brood cell development is now standard in the approval process, which raises great optimism for the use of bee counters in the time to come. On the other hand, beekeepers and bee research institutes in Germany, for example, have successfully used a network of automated beehive scales, the so-called "Trachtnet" (Johannesen et al., 2022; Otten & Berg, 2018), to monitor the nationwide honey flow. Upgrading such a network with bee counters capable of detecting negative environmental influences could serve as an early warning system across countries or even continents. The integration of bee counters as a universal tool in regulatory risk assessment involves not only monitoring bee health in terms of resource limitation or swarming, but also collecting data on climate change, landscape structure, pesticide use, and measures to reduce or offset them. In particular, bee poisoning incidents could be more easily reported, documented, and ultimately prevented in the future. This in turn would enable an automated feedback system providing information on whether farmers have complied with the regulations and whether the measures stipulated for a pesticide were sufficient to protect bees. We conclude that the integration of bee counters into regulatory risk assessment not only provides a comprehensive approach to bee health monitoring but also provides valuable data on broader environmental factors, potentially leading to more proactive measures to protect bee populations in agricultural ecosystems.

AUTHOR CONTRIBUTIONS

Richard Odemer: Conceptualization; data curation; formal analysis; investigation; project administration; resources; software; visualization; writing – original draft. **Oliver Jakoby:** Writing – review and editing; conceptualization; resources. **Markus Barth:** Writing – review and editing; conceptualization; resources. **Silvio Knäbe:** Writing – review and editing; conceptualization; resources. **Jens Pistorius:** Writing – review and editing; funding acquisition; resources. **Katharina Schmidt:** Writing – review and editing; conceptualization; resources; visualization.

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CONFLICT OF INTEREST STATEMENT

Katharina Schmidt is a founder of apic.ai GmbH, which manufactures and distributes electronic bee counters.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are openly available in the Open Science Framework (OSF): https://doi.org/10.17605/OSF.IO/YKN9B.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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