



**AGRI-FOOD  
& BIOSCIENCES  
INSTITUTE**

*Vespa velutina* nest  
dissection report

Belfast, October 2025

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## About the Agri-food and Biosciences Institute

AFBI is Northern Ireland's largest provider of agri-food and environmental sciences. As a non-departmental public body (NDPB) sponsored by the Department of Agriculture, Environment and Rural Affairs (DAERA) AFBI science plays a crucial role in providing statutory diagnostics and surveillance, research and development work, emergency response capability. On top of this it provides an expert scientific advice required to support the various work programmes of our sponsoring department and the wider requirements of the agri-food industry. AFBI has a broad remit of work including providing vital support to an industry which is a key economic driver within Northern Ireland, while delivering wider societal benefit in areas such as the environment and food safety. AFBI performs this against a backdrop of global and local challenges, including the threats posed by climate change, new and emerging animal and plant diseases, and the increasing requirement to balance environmental and economic sustainability. AFBI aims to meet these challenges through our work which centres on our three key themes of:

- Leading improvements in the agri-food industry to enhance its sustainability.
- Protecting animal, plant, and human health.
- Enhancing the natural and marine environments.

The work carried out by AFBI enables the agri-food industry to trade nationally and internationally and brings forward innovations to improve production efficiency and environmental sustainability.

## Plant Health and Integrated Pest Management Branch

Plant Health and Integrated Pest Management Branch (PHIPM) is a scientific branch within SAFSD. It provides diagnostics, surveillance, research, knowledge transfer and expert advice on plant and bee health, their pests and pathogens, as well as integrated pest management and pesticides. The Branch is organised to support the delivery of statutory diagnostic and surveillance programmes as required by the Official Control Regulations (EU) 2017/625, and supplementary regulation (EU) 2022/C 467/02.

AFBI has been assigned by DAERA as the Official Laboratory for plant health testing in Northern Ireland. It helps DAERA comply with 'plant health regulations and official controls' by providing statutory testing and surveillance of plants and their pests and pathogens agreed upon through an Assigned Work Programme (AWP). AFBI staff also provide guidance to DAERA on planning for, monitoring and reacting to plant and bee health threats, as well as conducting plant pest and pathogen horizon scanning and integrated pest management. AFBI offers a range of bee health tests including American and European foulbroods, small hive beetle, nosema and acarine disease.

## SUMMARY

The first *Vespa velutina* nest in Northern Ireland was found in October 2025. It was taken down by the Northern Ireland Environment Agency (NIEA) from the side of an unoccupied building in Dundonald, Belfast and was analysed by the Agri-Food and Biosciences Institute (AFBI). The nest contained 6-7 combs with active brood and was estimated to have produced ca. 3,300 individuals. It is therefore a moderate sized nest, of average size for the time of year and stage of development. A total of 84 adults were found in the nest, including 51 males and 33 females. The queen was absent from the nest, likely having fled when the nest was destroyed. There was a marked dominance of males in the brood as well as the adult stage. Caste analysis of the females indicated that there were at least 25 gynes in the brood, while most of the adult females were workers, with 3-6 individuals being potential newly emerged gynes. The nest appears to be in the early reproductive phase, beginning production of reproductive females. While it is possible that the nest was taken down in the short interval where reproductive males are produced before gynes emerge, it is not possible to say with certainty that no fertile gynes have left the nest, though it is unlikely that many would have been able to do so. It is therefore ***recommended that precautionary trapping for new queens takes place in spring 2026*** to prevent the establishment of any subsequent nests and to monitor the area closely for sightings of individuals.

## INTRODUCTION

In 2004, a single queen of the Asian or Yellow-legged hornet (*Vespa velutina*) was accidentally introduced into France and established a colony. Since then it has rapidly spread across France, neighbouring countries and then across most of Europe. There have been increasingly numerous incursions into Great Britain since 2016, with large-scale removal of nests annually.

Before 2025, very few Asian or Yellow-legged hornets have been sighted on the island of Ireland. However, in August 2025 a large nest was found in Cork City and then a smaller nest was found nearby in Cobh in September.

An individual hornet was reported by a member of the public in the Dundonald area of Belfast on 10<sup>th</sup> October 2025 and was identified as *Vespa velutina*. Bait traps erected by the Northern Ireland Environment Agency (NIEA) captured more individuals, indicating the presence of a nearby nest. By 17<sup>th</sup> October the nest had been located under the soffits of an abandoned building. It was sprayed with Vulkan (i.e. permethrin) at dusk that evening and delivered to AFBI on 18<sup>th</sup> October 2025.

The nest had dropped to the ground during the removal process and thus was damaged when received at the laboratory, with much of the outer casing destroyed, exposing the combs inside. Some of the combs were damaged and at least one had separated from the rest.

## METHODS

### Comb analysis

Following removal of the nest by NIEA, AFBI received the nest in a box sealed in biohazard bags. It was stored in -20 freezer for 48 hours before dissection and analysis. Analysis took place within a fume hood, with full PPE worn. The external structure and casing were examined for entrances and measurements

taken of the perimeter and height. As the nest was damaged, much of the outer casing was in pieces and these were carefully removed from the combs.

Each comb was extracted, numbered in order of removal and photographed with a scale bar. Brood pattern was observed in or each comb (regular or non-regular).

Measurements were taken of comb area, largest comb diameter, number of cells per comb, number of capped cells and their approximate size, the number of cells with eggs and the number of cells with larvae.

The cell-building rate per day was calculated using the formula given by Dieguez-Anton et al. 2022:

$$Cbr = Ce/6$$

where Cbr is the cell-building rate, Ce is the empty and cell eggs without meconium, and 13 is taken as the mean length of the development of the egg state in a nest from which the first worker or male has emerged.

Each comb was examined for all life stages: eggs, larvae, pupae, teneral adults, and adults. Larvae and pupae were sampled by uncapping cells, removing larvae and pupae, and placing them into labelled vials. An attempt was made to find and remove all teneral adults from each comb, as well as a large proportion of late-stage pupae (those who were coloured rather than white). Each stage was identified by the following features and sexed where possible:

**Eggs** were extracted whole from uncapped cells and frozen directly at -20 °C. The number of eggs per cell was also recorded.

**Larvae:** White and without legs, of various sizes and with lateral ridges along the body.

**Prepupae:** White and without legs, in capped cells, large in size and without lateral ridges along the body.

**Early-stage pupae:** White, with legs, and with abdomen, thorax and head.

**Late-stage pupae:** Legs and head formed and darker, in capped cells, wings present but not fully developed. All late-stage pupae were sexed.

**Teneral adults:** Fully formed adults in sealed cells prior to exoskeleton hardening and final colouration: wings present and fully developed; legs and head present. All teneral adults were sexed.

**Adults** found on the combs and within the nest or packaging were collected, counted and sexed.

**Meconium** is the excrement left behind by the larvae when it turns into a pupa. Contrary to bees, hornets do not clean out the cells after the adults emerge and thus the number of meconiums per cell can be used to estimate the number of individuals produced in the cell. Samples were taken from 5 cells per comb sampled from a cross-section of the comb. They were weighed using a fine-scale balance and the individual meconium weight calculated with the formula given by Dieguez-Anton et al. (2022):

$$Tcs = mMn / 0.06$$

where Tcs is the number of hornets emerged from each cell in secondary nests, and mMn is the mean weight of the meconium. Dieguez-Anton et al. (2022) calculated the mean weight of meconium for a secondary nest to be 0.06 g from analysing 44 nests in Galicia, Spain.

Samples were taken from all development stages and sent to FERA for genetic analysis. Eggs and larvae were sent intact while a single leg was removed from pupae, teneral adults, and adults. All samples were either kept frozen or stored in 100% ethanol until DNA extraction and genetic analysis (carried out by FERA).

## Sexing and caste differentiation

Adults, teneral adults and late-stage pupae were separated into male and female by examining the ventral side of their abdomens. Males had blunt-ended abdomens with two parallel ridges near the apex while females had pointed abdomens with a sting (Fig. 1).



**Figure 1.** Image showing male (left) and female (right) features on the *Vespa velutina* abdomen.

Wet weights and dry weights were taken of all adult and teneral females and some late-stage pupae. Dry weight measurements were completed by placing the female adults and tenerals in an oven set at 60 °C and weighing from 12hrs until the weight no longer changed giving a final dry weight.

Mesoscutum measurements: This measurement (mm) was taken of the mesoscutum, located on the dorsal side of the thorax between the wings, from tegula to tegula (fig. 3), using vernier callipers in all adult and teneral females and some late-stage pupae according to Pérez-de-Heredia *et al* (2017.).



**Figure 2.** Image showing mesoscutum width measurement from tegula to tegula.

Distribution plots were generated of the wet weights, dry weights and mesoscutum widths, using various lifestages to obtain enough data. Gaussian finite mixture models were run in R (4.5.1; packages stat and mclust), fitted by EM algorithm, on the three types of data to identify clusters in the data and the cutoff points between the castes.

## RESULTS

### Nest dimensions

The hive had a diameter of approximately 33 cm and was approximately 30 cm high, with a circumference of 95 cm. This indicates a volume of approximately 17.1 l. It was not possible to establish the number of entrances but from images it appeared there was one main entrance at the front of the nest. The casing thickness was 1-3.5cm. The number of combs was 6 or possibly 7, with the first and possibly second being empty of brood or eggs. Table 1 summarizes the nest dimensions.



**Figure 3.** Image showing intact nest. *Credit: Richard Gray (NIEA)*

**Table 1.** Approximate nest dimensions.

<b>Hive number</b>	1
<b>Location</b>	Dundonald
<b>Date</b>	Oct-25
<b>Type</b>	Secondary
<b>Circumference (cm)</b>	95
<b>Diameter (cm)</b>	33
<b>Height (cm)</b>	ca. 30
<b>No. entrances</b>	Unknown
<b>Entrance position</b>	Front centre
<b>Min. casing thickness (cm)</b>	1
<b>Max. casing thickness (cm)</b>	3.5
<b>No. combs</b>	6-7 (likely 6)

## Comb analysis

The individual combs are pictured in figs 4-11. while data related to the combs is presented in Table 2. Combs are layered horizontally on top of each other in the nest, connected with multiple sturdy pillars which had to be broken carefully in order to separate the individual combs.



**Figure 4.** All combs and comb fragments extracted from the 2025 Dundonald nest, shown to scale.

**Table 2.** Data relating to all combs in the *Vespa velutina* hive found in Dundonald, Belfast October 2025. Numbers are approximate for data marked with an asterisk.

Comb no.	1	2	3	4	5	6	Fragment 1	Fragment 2	total
Comb area (cm <sup>2</sup> )	44.6	287.9	337.7	340.2	271.3	133.5	79.5	153.2	1647.9
Comb diameter (cm)	6.8	23.3	21.5	21.2	19.5	16.3	13.0	17.6	n/a
Number of cells*	72	512	516	548	401	197	105	205	2556.0
Avg. cell area (cm <sup>2</sup> )	0.606	0.571	0.556	0.549	0.518	0.684	0.757	0.747	0.624
Cells per day*	0	11.5	0	0	0	0	17.2	33.8	10.4
Brood pattern	Regular	Regular	Regular	Regular	Regular	Regular	Regular	Regular	Regular
Number of capped cells	0	353	334	292	123	113	0	0	1215
Number of large cells*	0	26	34	12	5	12	0	0	89
Number of medium cells*	0	300	264	239	96	100	0	0	999
Number of small cells*	0	27	36	41	22	13	0	0	139
Number of uncapped cells	72	159	182	256	278	84	0	0	1031
Number of larvae*	0	52	50	36	23	44	2	2	209
No of cells with eggs*	72	103	123	22	8	34	103	203	668
Number of empty cells*	0	4	9	198	247	6	0	0	464
Number eggs per cell	1	1	1	1	1-2	1	1	1	n/a
Number of prepupae*	0	261	226	211	93	113	0	0	904
No. of late stage (coloured) pupae*	0	60	52	43	16	0	0	0	171
Number of male coloured pupae*	0	50	46	28	16	0	0	0	140
Number of female coloured pupae*	0	10	6	15	0	0	0	0	31
Number of teneral adults	0	16	28	19	7	0	0	0	70
Number of teneral males	0	15	24	15	4	0	0	0	58
Number of teneral females	0	1	4	4	3	0	0	0	12

### Comb 1

Comb 1 (uppermost comb) was the smallest of the six combs and almost perfectly round in shape (fig. 5). No brood was present but single eggs had been laid in almost all the cells in a regular pattern. All the other combs contained brood, empty cells and eggs in a regular pattern.



Figure 5. Image showing Comb 1.

### Comb 2

Comb 2 had some large extensions to the side that contained eggs but no meconium. It contained dense and mostly capped brood (fig. 6) which on investigation contained 16 (or 4.5%) teneral adults (all but one were morphological males), 60 (or 17.0%) late-stage pupae of which 50 (83.3%) were males, ca. 261 (or 73.9% of capped cells) early-stage pupae and approximately 52 larvae (or 32.5% of uncapped cells). Caste analysis indicated that the single teneral female was a worker while all the late-stage pupae were gynes.

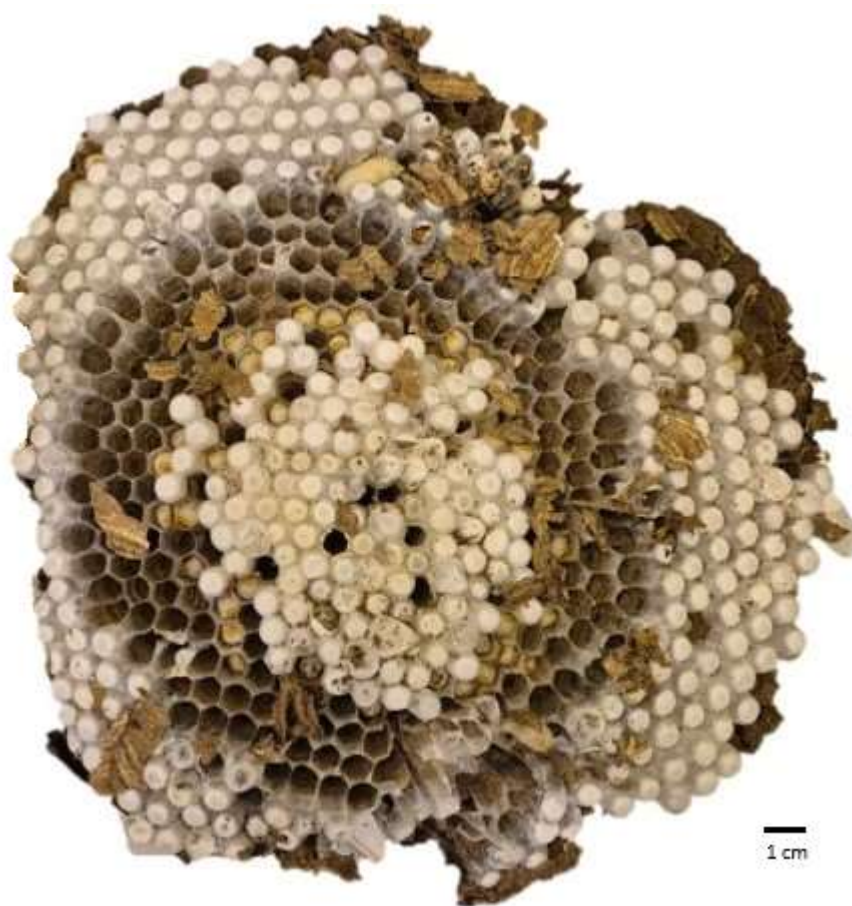


Figure 6. Image showing Comb 2.

### Comb 3

Combs 3 & 4 had the largest number of cells and the highest comb area. In the centre of comb 3 was an area of capped cells containing (pre-)pupae followed by an area of uncapped cells containing both larvae and eggs, followed by an outer ring of capped cells with brood including teneral (fig. 7). Only 4 of the 28 teneral were female with the remainder (85.7%) being male. Forty-six of 52 late-stage pupae were male (88.5%) with while approximately 226 (or 67.7 % of the capped brood) were early-stage pupae or pre-pupae. Approximately 50 larvae were counted in uncapped cells.

Caste analysis indicated that all four of the teneral females were gynes. Of the six late-stage female pupae found in comb 3, caste analysis further indicated that 4 were gynes while 2 were workers.



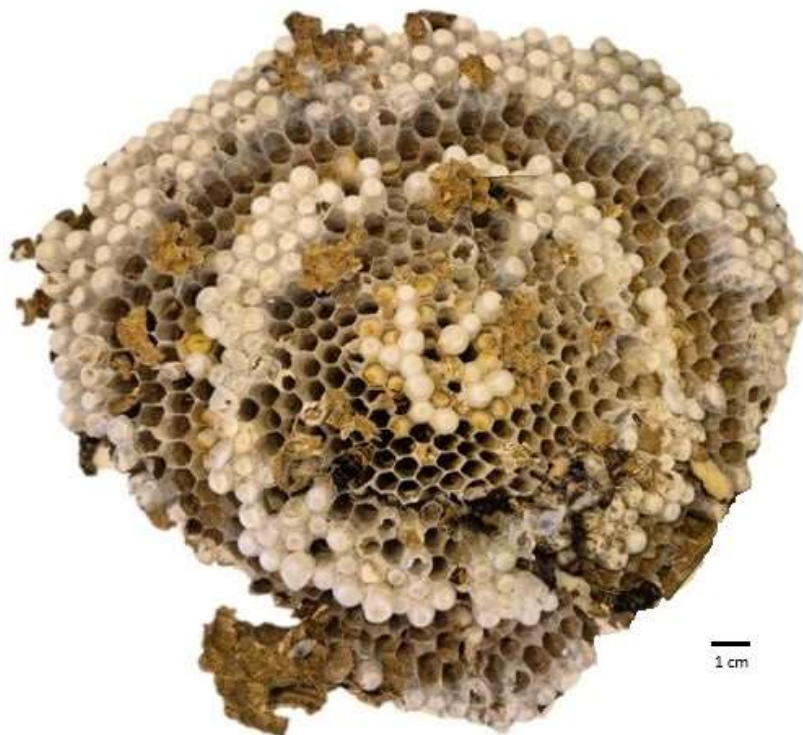
**Figure 7.** Image showing Comb 3.

### Comb 4

Comb 4 had the largest area and showed a regular pattern of capped and uncapped brood (fig. 8). Most of the uncapped cells that did not contain larvae were empty, with only few eggs present.

The brood contained 19 (or 6.5%) teneral adults, 43 (or 14.7%) late-stage pupae of which 28 (65.1%) were males, ca. 211 (or 72.3%) early-stage pupae or prepupae and ca. 36 (12.3%) were larvae.

The 15 late-stage female pupae were separated into 7 gynes and 8 workers while all four of the teneral females were workers.

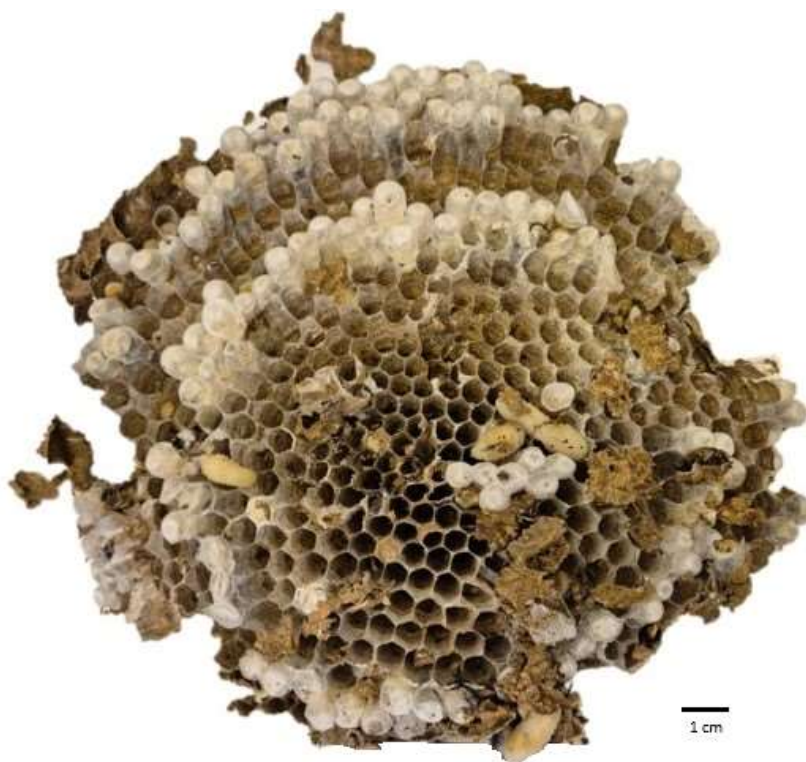


**Figure 8.** Image showing Comb 4.

### Comb 5

Comb 5 also showed a mostly regular pattern of capped and uncapped brood (fig. 9). However, where larvae would have been expected in the centre of the comb, only empty cells remained without eggs and with rather damaged side walls. Only a small number of cells contained eggs and some appeared to contain two eggs.

Seven teneral adults were found, of which 4 were males and 3 were females. The three the teneral females were classed as workers. Of 16 late-stage pupae all were males. Approximately 93 (or 75.6% of capped brood) early stage or prepupae and 23 larvae were counted.



**Figure 9.** Image showing Comb 5.

### Comb 6

Comb 6 had a large average cell area but fewer cells and a smaller comb area. It was included separately from the rest of the nest and its positioning is not entirely certain. The cells included dense brood consisting mainly of capped cells (fig. 10). These all appeared to be prepupae or early-stage pupae as no teneral adults or late-stage pupae were found. The sex of the hornets could not be distinguished from these early stages of development.



Figure 10. Image showing Comb 6.

### Comb fragments 1 and 2

Some loose broken comb fragments without brood or meconium but with eggs was present in the nest (fig. 11), which could possibly have been a separate comb in position 2 from the top or 7 at the bottom. However, it could also have represented broken edges from other combs (perhaps comb 6) and they were not analysed as a separate comb.



Figure 11. Image showing fragments 1 (smaller) and 2 (larger).

## Meconium

Apart from about 78 newly built cells in comb 2 and the two loose fragments of comb found in the nest, almost all the cells contained at least one meconium. The possible number of layers was estimated for each comb and 5 pellets were samples from across combs 2-6 and weighed (Table 3). When the formula given in Dieguez-Anton et al. (2022) was applied, the figures indicated that there was only one layer of meconium per cell on average in the hive.

**Table 3.** Meconium wet weight per cell.

comb	wet weight of 5 cells (g)	avg wet weight per cell (g)	Min. meconiums	Max. meconiums	No cells with at least 1 meconium
1	Not measured	Not measured	2	4	72
2	0.308	0.062	0	3	434
3	0.224	0.045	1	3	516
4	0.274	0.055	1	3	548
5	0.225	0.045	1	3	401
6	0.376	0.075	1	3	197
total	1.407	0.563	n/a	n/a	2168

Using estimates for the meconium and the numbers of larvae, eggs and prepupae obtained through examination of the combs as described above, the total production of the colony was estimated as shown in Table 4.

**Table 4.** Colony production calculation.

eggs	668
larvae	209
meconiums (high estimate assumes 1.3 per cell average)	2818
prepupae	904
<b>max total production:</b>	<b>4,600</b>
eggs	668
larvae	209
meconiums (low estimate assumes 1 per cell average)	2168
<b>min total production:</b>	<b>3045</b>

According to Rome *et al.* (2015), the overall production of a nest is strongly correlated to the size of its broadest comb and follows the quadratic function:

$$Y = 7.12 * x^2 - 37.72 * x - 9.68$$

where 'y' is the number of individuals produced by the colony, and 'x' is the diameter of the largest comb. Thus, the overall estimated production of a nest could be inferred from this result, enabling one to evaluate how many individuals have been produced by a given nest only by measuring the diameter of its widest comb.

Using this formula and taking comb 2 to have the widest diameter (23.3cm), we obtain a figure of

$$Y = 7.12 * 542.89 - 37.72 * 23.3 - 9.68$$

$Y = 3865.4 - 513.75 = 3,351$

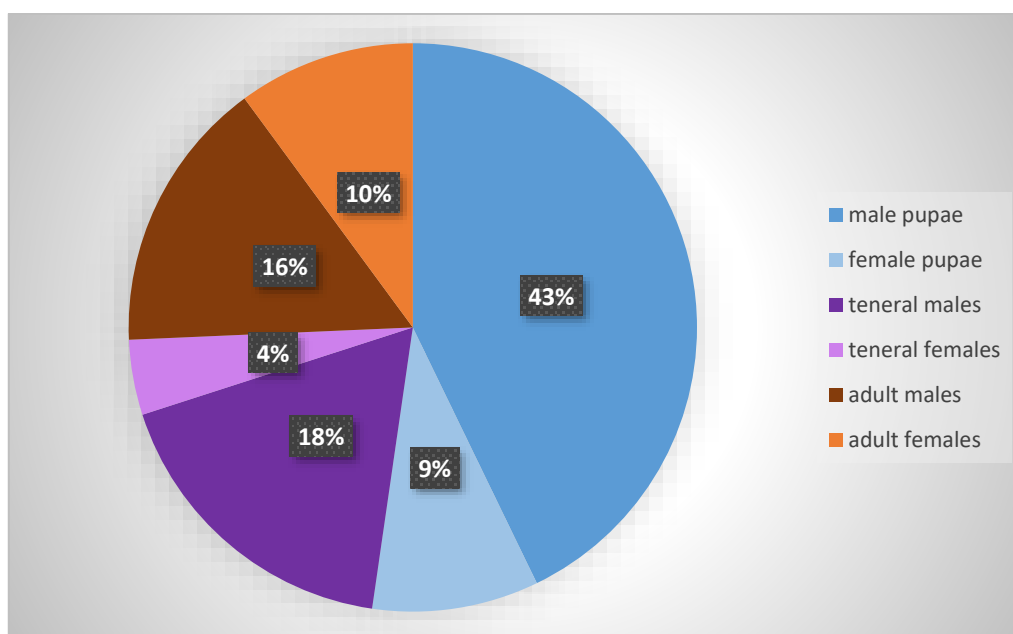
### Adult hornets and sex ratios

A total of 84 adult hornets were found in the nest, of which 33 were female and 51 were male. While there were several female hornets of the right size, none of them had clear signs that marked it as the mother queen, such as smooth and distended abdomen and frayed wings. It was therefore presumed that the mother queen was not in the nest at the time it was taken down.

All teneral adults and late-stage pupae were also sexed. Numbers of each are given in table 5 and summarized in figure 12 below.

**Table 5.** Numbers of males and females of adult, teneral adult and late stage pupae found in the 2025 Dundonald *Vespa velutina* nest.

Lifestage	male	female
Adult	51	33
Teneral adult	58	14
Late stage pupa	140	31

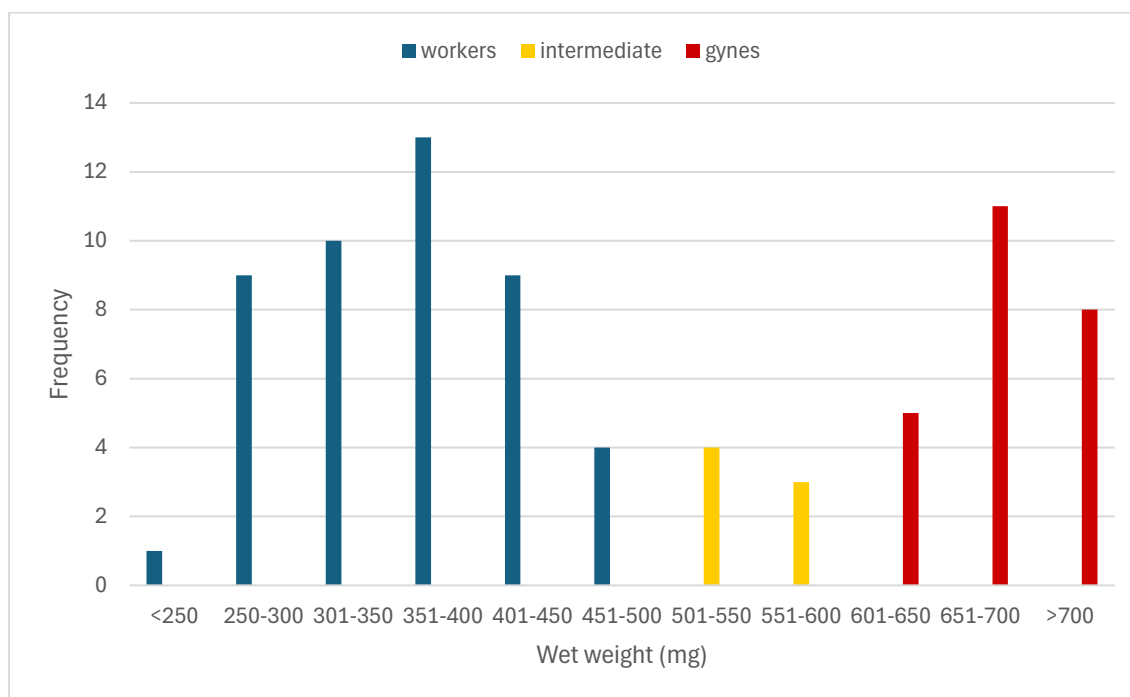


**Figure 12.** Percentages of males and females of adult, teneral adult and late stage pupae found in the 2025 Dundonald *Vespa velutina* nest.

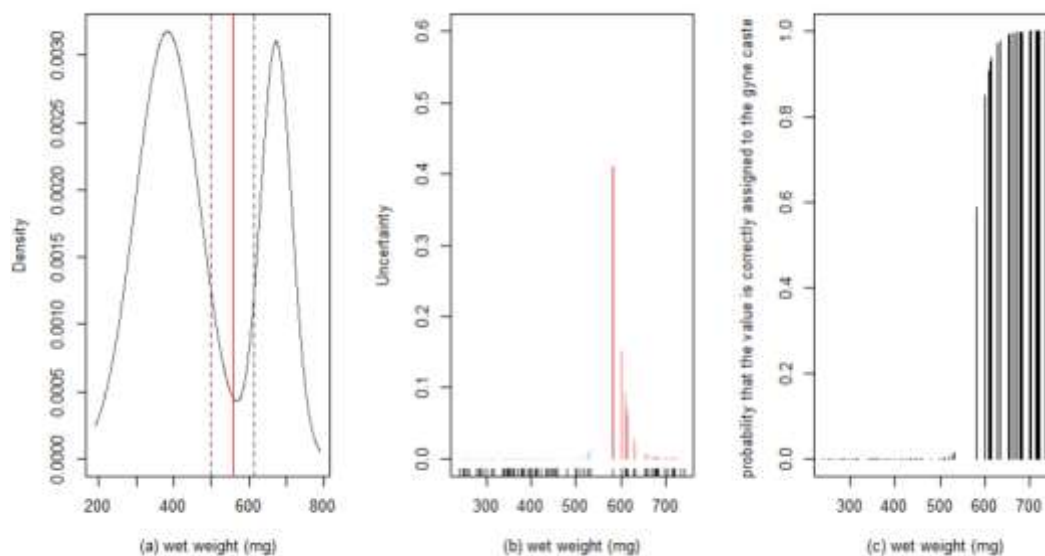
### Caste differentiation

Plotting a frequency distribution of the wet weights of female *Vespa velutina* in the adult, teneral adult and late pupal stages resulted in a bimodal distribution with an intermediate range of 545-615 mg. Weights below this were clearly indicative of workers while weights above this were indicative of gynes

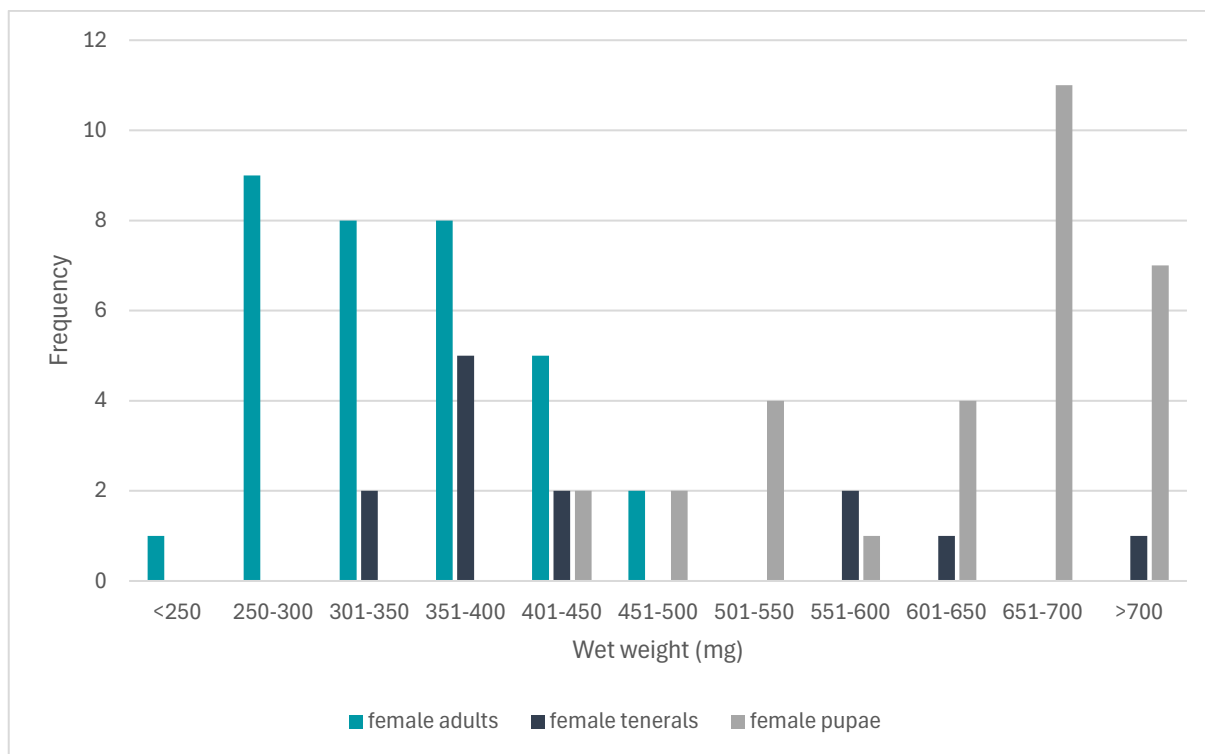
(figs. 13 & 14). Separating out the various lifestages on the plot indicates that none of the adult hornets of the Dundonald hive are gynes, but several teneral adults and many pupae are (fig. 15).



**Figure 13.** Wet weight of female *Vespa velutina* (adults, tenerals and pupae) from the 2025 Dundonald nest assigned to castes.

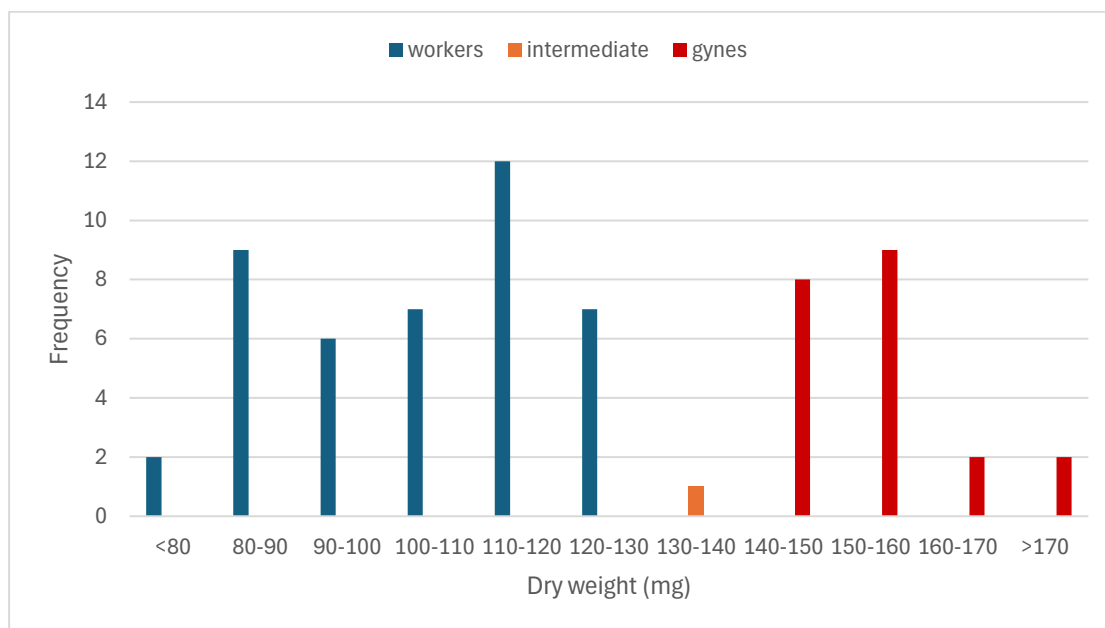


**Figure 14.** Cluster analysis results for wet weights of *V. velutina* female adult, teneral adult and pupae of the 2025 Dundonald nest. (a) univariate model distribution with 2 component of unequal variance. (b) Uncertainty of individual allocations in model. (c) Probability that individuals are correctly assigned to the gyne caste.

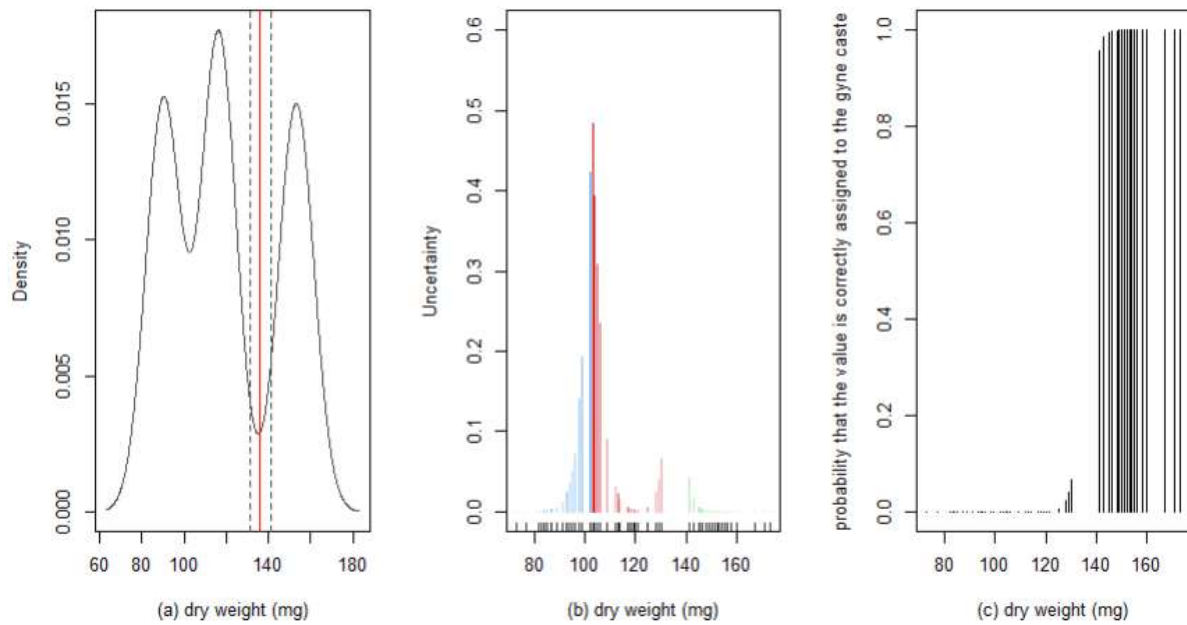


**Figure 15.** Wet weight of female *Vespa velutina* (adults, teneral and pupae) from the 2025 Dundonald hive.

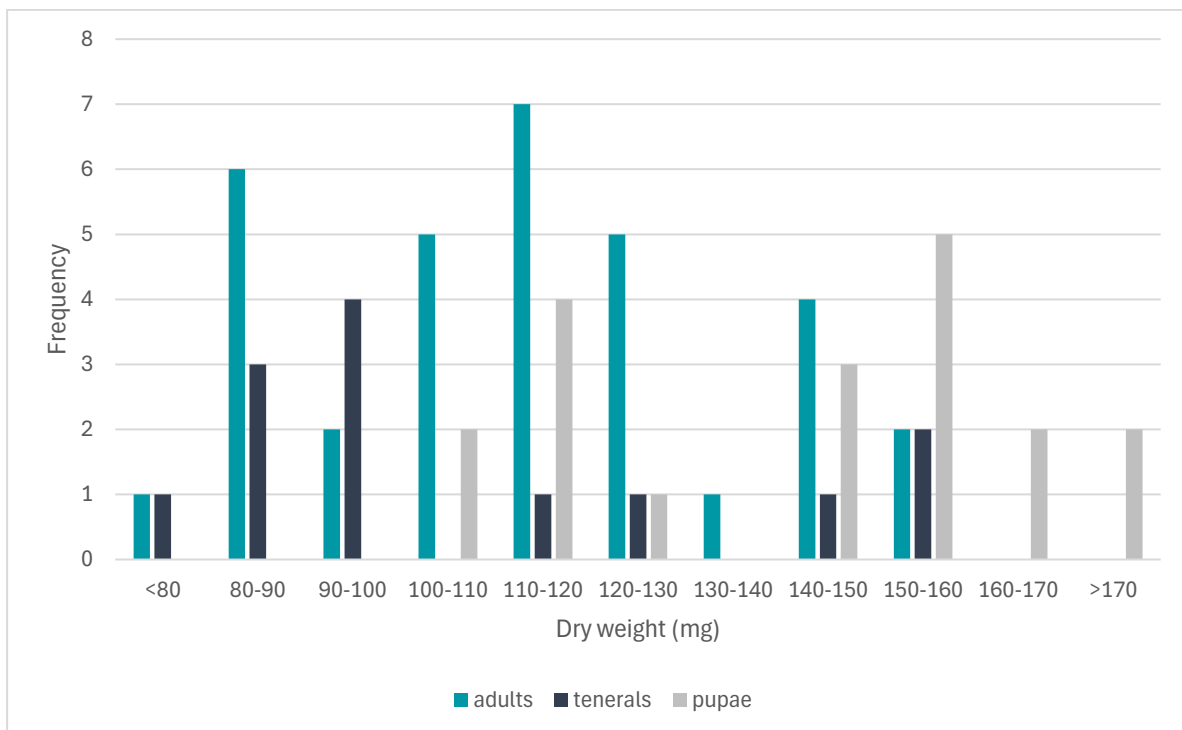
A frequency distribution of the dry weights of female *Vespa velutina* in the adult, teneral adult and late pupal stages resulted in a bimodal distribution with an intermediate range of 130-140 mg. Weights below this were clearly indicative of workers while weights above this were indicative of gynes (figs. 16 & 17). Separating out the various lifestages on the plot indicates that potentially 6 of the adult hornets of the Dundonald hive are gynes, as well as 3 teneral adults and many pupae (fig. 18).



**Figure 16.** Dry weight of female *Vespa velutina* (adults, teneral and pupae) from the 2025 Dundonald nest assigned to castes.

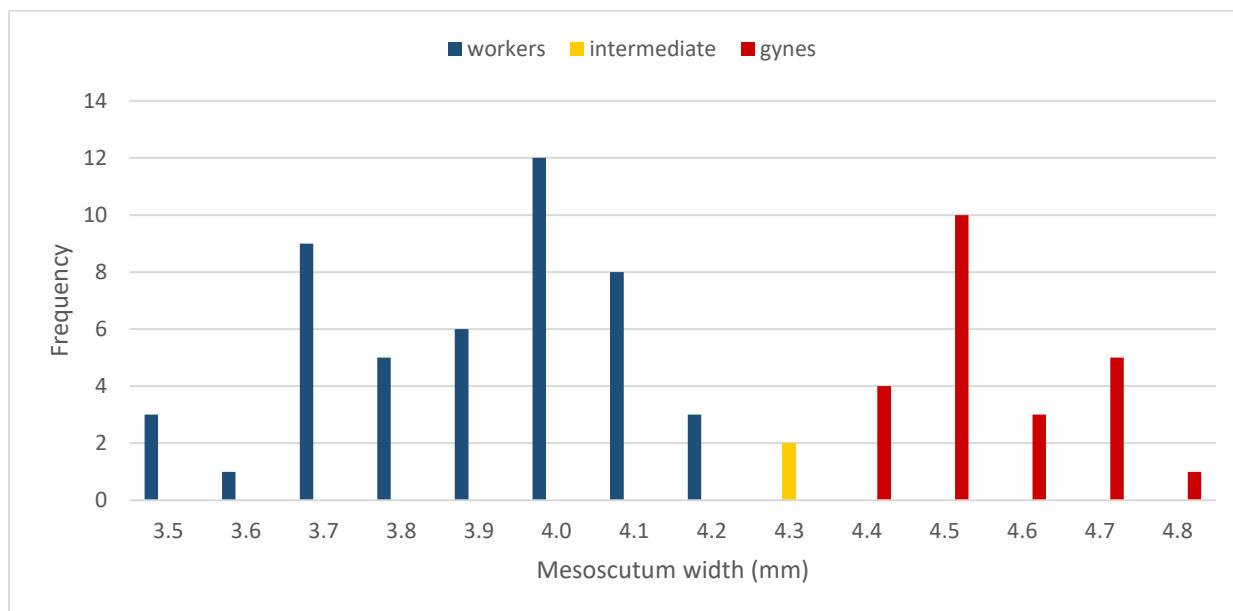


**Figure 17.** Cluster analysis results for dry weights of *V. velutina* female adult, teneral adult and pupae of the 2025 Dundonald nest. (a) univariate model distribution with 3 components of equal variance. (b) Uncertainty of individual allocations in model. (c) Probability that individuals are correctly assigned to the gyne caste.

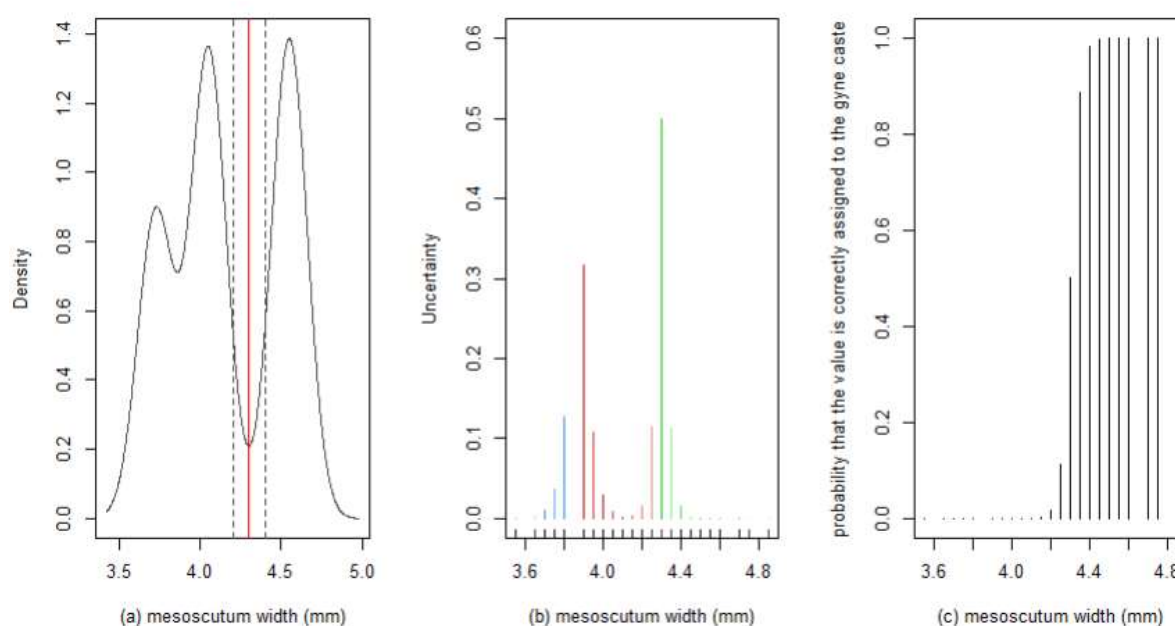


**Figure 18.** Dry weight of female *Vespa velutina* (adults, tenerals and pupae) from the 2025 Dundonald hive.

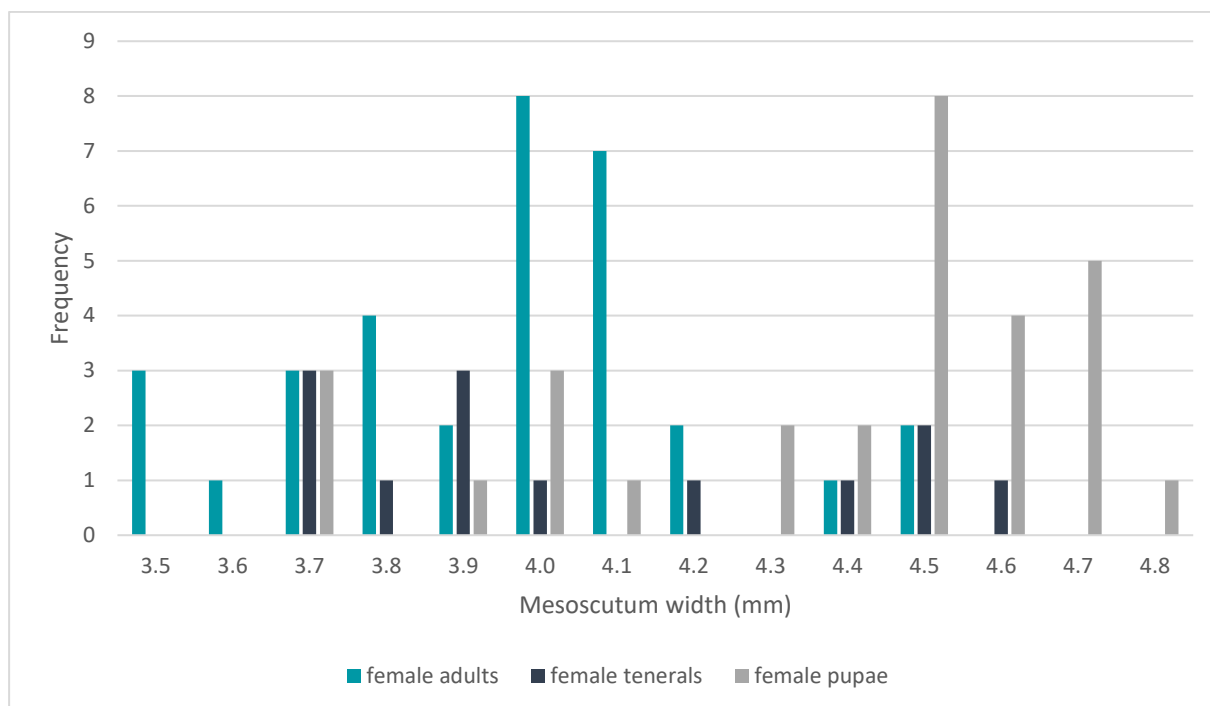
A frequency distribution of the mesoscutum widths of female *Vespa velutina* in the adult, teneral adult and late pupal stages resulted in a bimodal distribution with an intermediate range of 4.4 mm. Weights below this were clearly indicative of workers while weights above this were indicative of gynes (figs. 19 & 20). Separating out the various lifestages on the plot indicates that potentially 3 of the adult hornets of the Dundonald hive are gynes, as well as teneral adults and many pupae (fig. 21).



**Figure 19.** Mesoscutum widths of female *Vespa velutina* (adults, teneral adults and pupae) from the 2025 Dundonald nest assigned to castes.



**Figure 20.** Cluster analysis results for mesoscutum widths of *V. velutina* female adult, teneral adult and pupae of the 2025 Dundonald nest. (a) univariate model distribution with 3 components of equal variance. (b) Uncertainty of individual allocations in model. (c) Probability that individuals are correctly assigned to the gyne caste.

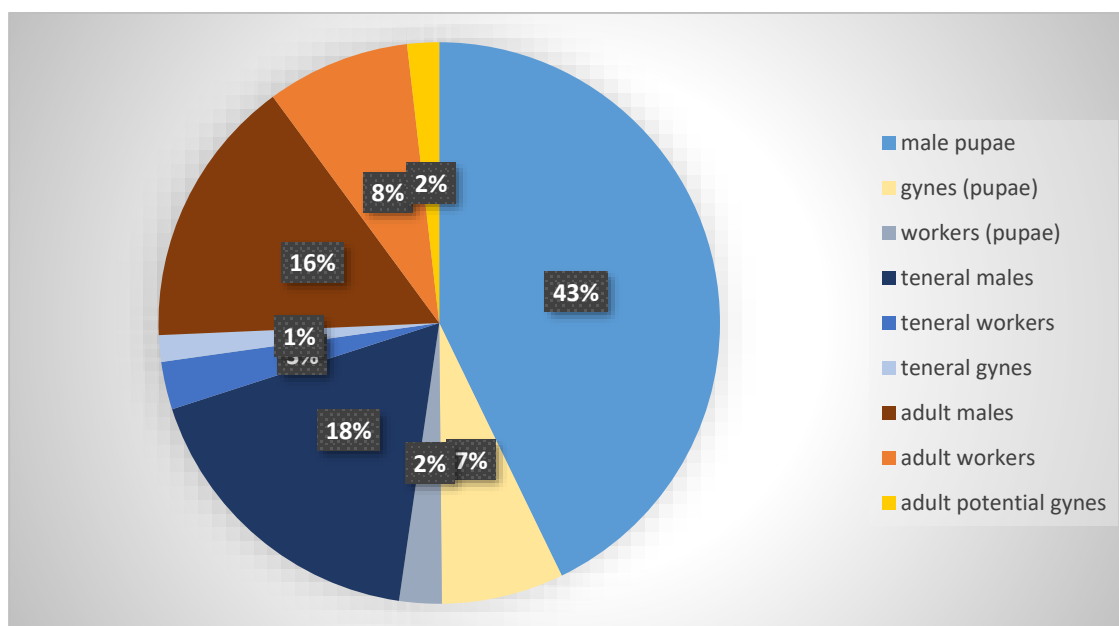


**Figure 21.** Mesoscutum widths of female *Vespa velutina* (adults, tenerals and pupae) from the 2025 Dundonald hive.

Using the cutoffs from the bimodal distributions in the data, the numbers of gynes and workers were estimates to be approximately 47 workers and 31 gynes across the lifestages, with most gynes in the pupal phase (table 6; fig. 22).

**Table 6.** Numbers of males and females of both castes of adult, teneral adult and late-stage pupae found in the 2025 Dundonald *Vespa velutina* nest.

Lifestage	male	female	workers	gynes
Adult	51	33	27-33	0-6
Teneral adult	58	14	9-11	3-5
Late stage pupa	140	31	8	23



**Figure 22.** Percentages of males and females of adult, teneral adult and lat- stage pupae found in the 2025 Dundonald *Vespa velutina* nest, including probable caste allocations.

## DISCUSSION

The size and production capacity of the Dundonald nest was moderate. The Dundonald nest was smaller than the nest found in Cork City, with the Dundonald nest containing around six combs with an estimated production capacity of 3,300 and the Cork City nest comprising seven combs with an estimated production of 9,000 hornets (O’Hanlon, 2026). However, the Dundonald nest was bigger than the Cobh nest, which had five combs and an estimated production of 2,000 hornets (O’Hanlon, 2026). Rome et al. (2015) analysed 77 nests from south-west France and reported a mean comb number of 6.3, mean number of cells of 5004 and mean colony production of 5320 individuals for mature nests in October, which is comparable to the Dundonald nest in terms of number of combs, but the number of cells present in the Dundonald nest was only about 2,500, or half the number found in France, with a lower colony production.

Assessing the developmental stage of the nest helps to determine whether the hornets were likely to have released the next generation of founder queens. Size and architecture of the nest, time of year, and the internal composition of the caste can help with determining the developmental stage. During summer the nest is primarily comprised of female workers, which drives nest growth and expansion, in autumn the colony starts to shift towards reproduction. Fertile males and females (gynes) develop in short succession and mate in the nest. The gynes leave the nest with liberal fat reserves to help survive the winter and start their own founder nests in spring. Diploid males early in the season are a common feature of severely inbred *V. velutina* nests such as those found in Europe (Darrouzet et al., 2015).

Hornet nests start small (Comb 1) with few cells. This indicates the initial nest building stage when resources and workers are short in supply. The rapid growth phase (Combs 3 & 4) shows the colony at its peak activity. Here the colony is maximising brood production and the storage of available resources. The later stage (Comb 6) shows fewest cells, but these have a greater average cell area. This can indicate shifting behavioural patterns, such as when the colony starts its reproductive phase.

The regular brood pattern and presence of eggs in the newly built comb indicated that this was an active secondary nest during the time it was taken down. The absence of the queen is relatively common, as she tends to escape the nest with her workers as soon as the nest is disturbed (Semence, pers comm). She may then try and rebuild a tertiary nest at the exact same location, which will certainly fail to be reproductively successful so far into the season.

Determining whether the nest was in the reproductive phase requires assessment of the presence of gynes. Differentiating between gynes and workers is challenging as there is no obvious morphological distinction between the castes. Various methods are proposed in the literature, the simplest being weight measurements and mesoscutum width measurements, which were deployed here. Chemical analysis of the cuticle (Perez-de-Heredia et al., 2017) and wing morphometrics (Perrard et al., 2012) have been suggested as alternative methods. Another option is dissection, although the internal differences between workers and gynes has not been clearly described. A successful dissection should however be able to indicate whether the females have been fertilised or not. Perez-de-Heredia et al. (2017) demonstrated that wet weights are less reliable than dry weights in distinguishing castes and that mesoscutum widths was more accurate than either. They indicated that the cutoff between workers and gynes is 4.5 mm in their study, although there were size differences between individual colonies. The bimodal distribution for mesoscutum width seen in the Dundonald nest indicated a cutoff value of 4.3 mm which is slightly smaller than found by Pérez-de-Heredia et al. (2017). No other studies appear to add in measurements from teneral adults and late-stage pupae and the consistency of measurements across the various life-stages should be treated with caution. The reason that the cluster analysis picked up three clusters rather than two in the case of dry weight and mesoscutum width measurements was primarily linked to differences between lifestages (figs. 18 & 21). The clustering could also reflect differences in developmental stage of the nest, as it has been demonstrated that hornet size varies over time within each nest (Rome et al. 2015; Pérez-de-Heredia et al. 2017). The sex of late-stage pupae in the Dundonald nest was however easily visible and measurements did appear to be able to predict the caste of the individual with high confidence in most cases. The wet weight cutoff of 600 mg was comparable with the thresholds reported in the literature. The dry weight values obtained were however considerably lower, with a cutoff of 225 mg reported by Perez-de- Heredia et al. (2017) and 250 mg by Rome et al (2017) whereas the Dundonald threshold appeared to be 140 mg. The range of dry weight values was 73-173 mg, substantially below that reported in other studies. The reason for this is unclear as the time of drying was comparable with that reported in other studies.

Division between the castes of adult hornets was not clear, varying between no gynes (wet weight metric) to three gynes (mesoscutum width metric) to six gynes (dry weight metric). Interestingly, the large mesoscutum widths did not always correlate with high dry weight in individuals. All the potential adult gynes were close to the threshold used to differentiate gyne from drone, leaving some uncertainty to their categorisation. Nonetheless, the overwhelming dominance of males present in the nest (70% of sexed individuals) suggests that the nest is mainly producing non-reproductive males (Darrouzet et al. 2015). The presence of some adult and/or teneral adult gynes in the nest suggests that at least some of the males are likely reproductive males. Data on the number of diploid vs haploid males would also help confirm the stage of development. The larger proportion of gynes in the pupal stage (7% of individuals) and a small presence of gynes in the teneral stage (1%) suggests that the nest is in the early stages of the reproductive stage.

There is a possibility that some adult gynes have already left the nest after mating and gaining sufficient fat reserves, leaving only a few gynes still in the nest when it was taken down.

***Overall, the caste composition indicates that the nest was in the early reproductive stage when it was taken down.***

## CONCLUSIONS

The Dundonald *Vespa velutina* nest found in October 2025 was a moderately sized nest in the early reproductive phase. It contained an overwhelming number of males and was beginning to produce gynes. From the analysis, it is not possible to say with certainty that no fertile gynes have left the nest, though it is unlikely that many would have been able to do so. It is therefore recommended that precautionary trapping for new queens takes place in spring 2026 to prevent the establishment of any subsequent nests and to monitor the area closely for sightings of individuals.

At approximately 54.5 degrees latitude, this is likely the most northerly nest found worldwide so far. The size and weights of individuals was smaller than reported from more southernly locations. However, the number of combs and cells with active brood as well as the production of what are very likely reproductive adults indicates the viability of *Vespa velutina* nests in Northern Ireland.

Further information on the nest, including hive lineage and the ploidy level of males, can be obtained via microsatellite analysis.

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