#### November 23, 2023

### Summary 2023 Splits Data

#### Methods

Three types of splits were made from single box 8-frame Comfort hives between May 29, 2023 and July 29, 2023:

- 2-Day Cells (48 hr.) on May 29, 2023
- 10-Day Cells on July 1, 2023
- Walk-away on July 29, 2023

The queen cells were obtained from grafted eggs from over-wintered queen stock so that the exact ages were known. All splits were started with approximately the same population density of workers.

Approximately 3 weeks after the splits were made, they were inspected for split success and hive population density. Split success was determined by visual evidence of a queen and evidence of young brood. Absence of a live queen, failure of queen cells to emerge, the presence of a damaged queen (e.g., deformed wings), a dead hive or evidence of laying workers, were considered failures. However, the presence of a queen without eggs or young brood was considered evidence of a virgin queen and marked as a success.

Hive population density was assessed at inspection for split success and was categorized as low (2 or fewer frames), medium (3-4 frames) or high (5 or more frames).

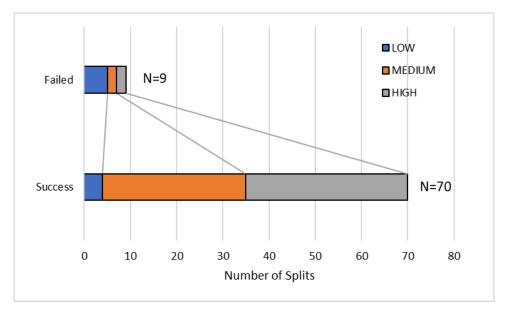
Two generalized logit models (GENMOD) were run on split success and population density with type of split as the independent factor to determine the influence of type of split on success and population density, which can affect ability to perform subsequent splits. For modeling purposes, population density was coded to 1=low, 2=medium and 3=high. A 2-sided t-test was performed to compare each type of split to the others, if the model indicated that types of splits were an influential factor.

## Results

A total of 87 splits were made between over the testing period and were evenly distributed among the types:

Type of split	N (%)
Walk-away	29 (33.3%)
10-day old queen cells	29 (33.3%)
2-day old queen cells	29 (33.3%)
TOTAL	87 (100%)

Of the 87 splits, 70 (80.5%) were successful, 9 (10.3%) were failures and the remaining 8 (9.2%) had no data reported. With regards to population density, 13 (14.9%) of splits were categorized as having low density, 33 (37.9%) were categorized as medium density and the remaining 41 (47.1%) were categorized as high density. These outcomes were highly correlated (Spearman's rho=0.27, p=0.01) with the failed splits tending to be of low population density while the successful splits of higher population density. Below illustrates the number of hives by population density for failed and successful splits, plus a cross tabulation of splits by population density and split success.



Population Density	Outcome Unknown n (%)*	Failed Split n (%)*	Successful Split n (%)*	Total N (%)
LOW	4 (30.8%)	5 (38.5%)	4 (30.8%)	13 (14.9%)
MEDIUM	0	2 (6.1%)	31 (93.9%)	33 (37.9%)
HIGH	4 (9.8%)	2 (4.9%)	35 (85.4%)	41 (47.1%)

Total	8 (9.2%)	9 (10.3%)	70 (80.5%)	87 (100%)
*percentages are based on number of splits within each population density				

The type of split did not influence the success rate. All types of splits had at least 89.7% success.

	Type of Split		
	10-Day Queen Cells	2-Day Queen Cells	Walk-away
% Success	89.7%	96.6%	82.8%
vs 10-Day		NS	NS
vs 2-Day	NS		NS
NS = not statistically significant (p>0.05)			

With regards to population density, the 2-Day cell splits had the highest populations with more that 86% in the high-density category. This was significantly different from 10-Day cell splits (p=0.025) and walk-away splits (p<0.0001). There was no difference between 10-Day cell splits and walk-away splits.

	Type of Split			
Density	10-Day Queen Cells	2-Day Queen Cells	Walk-away	
Low (1)	6.9%	13.8%	24.1%	
Medium (2)	58.6%	0	55.2%	
High (3)	34.5%	86.2%	20.7%	
vs 10-Day		0.025	NS	
vs 2-Day	0.025		<0.0001	
NS = not statistically significant (p>0.05)				

<u>Caveat:</u> Each type of split was performed at different parts of the summer, with walk-away splits occurring in late July, 10-Day queen cell splits occurring in early July and 2-Day queen cell splits occurring in late May. Since weather, food availability and other environmental factors can vary throughout the season, this introduces a potential bias in outcomes based on types of splits. Future studies should include all 3 types of splits equally throughout the season in effort to minimize temporal bias. For example, in May approximately of third of the splits should be walk-away, a third be 10-Day queen cells and the remainder 2-Day queen cells.

# Conclusion

In this study there was no significant difference in the success of the split based on type of split performed. This may, in part, be due to the higher overall success rate of all splits (>80%) and the smaller sample size (number of hives) which reduces the statistical power to detect smaller differences.

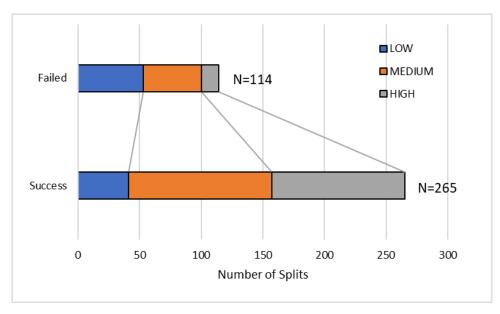
Population density was highest among the 2-Day cell splits than either 10-Day cell or walk-away. There was no difference between 10-Day cell splits and walk-away splits. Since each type of split occurred during different parts of the summer, where nectar flow, drone availability, mite load and weather patterns can affect successful mating and brood rearing, time of season may have had a greater influence on success and population density than type of split used.

In comparison to the 2022 splits experiment, the success rate of this 2023 experiment was higher (80.5% vs 64%). In addition, the population densities of the 2023 experiment were higher in general compared to 2022 (47.1% vs 27.7% for high density).

In the 2022 study, the order of splits type over time was walk-away (mid June), followed by 10-Day cells (late June, early July), then 2-Day cells (early August). By contrast, the 2023 experiment reversed order of walk-away (late July) and 2-Day cell splits (late May). In the 2022 experiment, the walk-away split was more successful than the others splits and with had higher density populations. Whereas, in 2023 the 2-Day cell splits were more successful than the other splits (although, not statistically significant) and had higher population densities. This observation suggests that time of season is more influential than type of splits in the success the splits and population density. A study that performed all three split types in late spring, then again in early summer and finally shortly after solstice would allow for any bias associated with time of season to be separated from type of splits.

# Bonus Analysis (pooled data from 2022 and 2023)

Data from 2022 and 2023 were pooled to see if a larger overall sample size would enhance any split type differences. The correlation between split success and population density increased to Spearman's rho=0.36 (p<0.0001) and was highly significant.



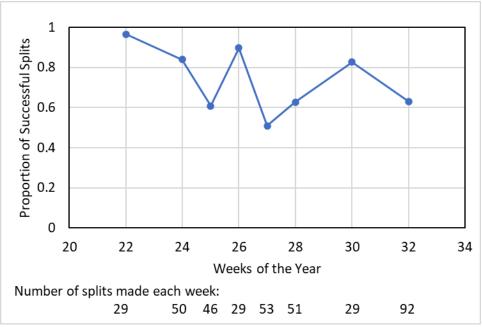
However, the differences between split types in success rate and population density was no longer significant. This was likely due to the transposition of timing for walk-away and 10-Day cells between years.

Over the 2 years, splits were performed during a window of time between late May and early August. To test whether time of season was influential in the success and population density of splits, the following analysis ignored split type and replaced it with weeks of the year. For example, Week 22 is the 22<sup>nd</sup> week of the year, usually late May, while Week 30 is the 30<sup>th</sup> week of the year, usually late July.

The split success and population density were analyzed using generalized logit models (GENMOD) with WEEKS as the independent factor to determine the influence of timing on success and population density. The following graphs illustrate the estimated proportion of splits that were successful over time and the estimated average population density over time from the models. Since the number of splits varied at each week, the number of splits made for each week are summarized below each graph.

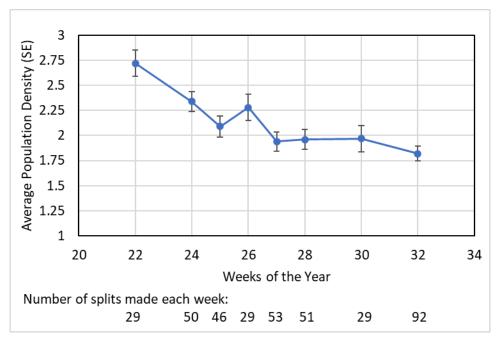
The highest success rate occurred late Spring, usually late May to early June. Split success generally trended downward over the remaining weeks. Summer solstice occurs between weeks 24 and 25, for reference.

**Proportion of Successful Splits over Time** 



The population density over time showed a stronger trend. On average, splits had the highest population density early in the split season and continued a steady decline through week 27 after which the density remained fairly stable.

Average Population Density of Splits over Time



# **Conclusions from Pooled Analysis**

The pooled results show a general decline in split success over time and a stronger decline in population density over time, independent of the type of split performed. In order to determine the impact of split type on success and population density, a study will need to be conducted that include all 3 split types during the same window of time in order to rule out influence of seasonal variability.