How many samples are sufficient for testing the reliability of mating stations in honey bees (*A. mellifera)?*

Andonov, Dahle, Kovačić, Marinč, Moškrič, Prešern, Pavlov, Puškadija, Uzunov
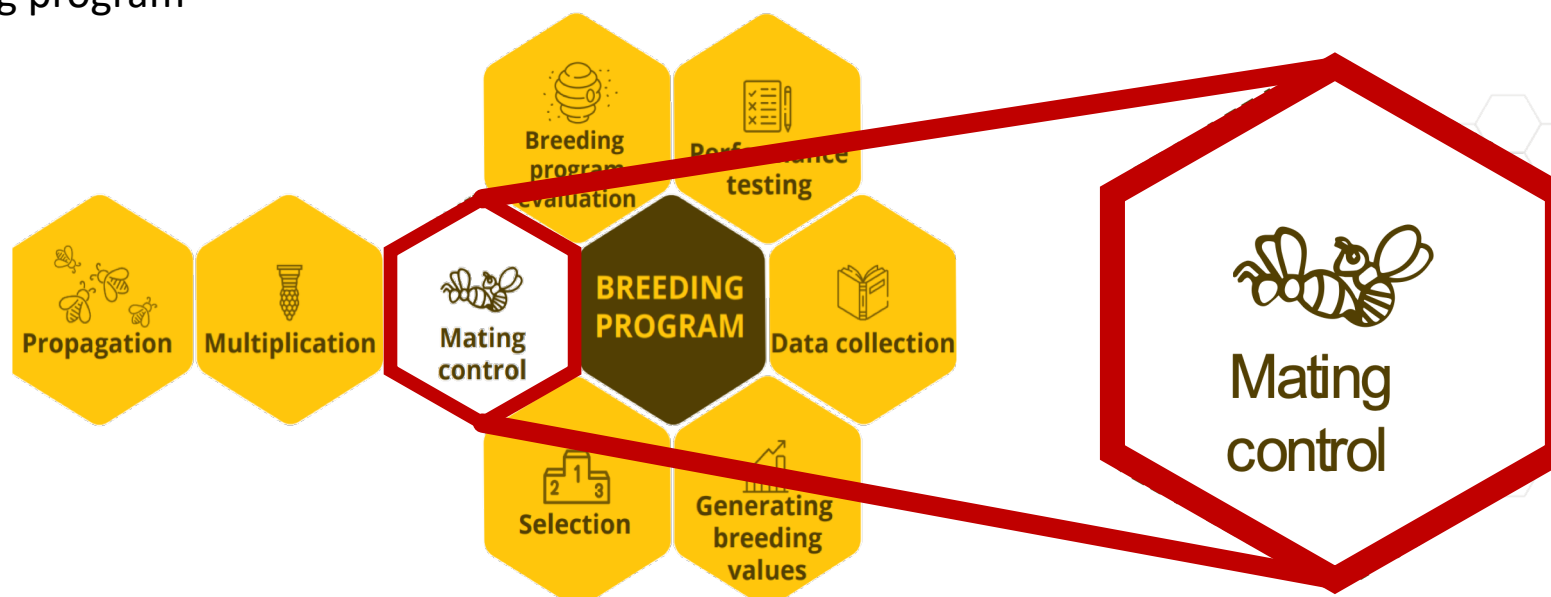
Introduction

- Genetic structure

  - Queens - diploid

  - Drones - haploid

- Reproduction peculiarity

  - Young –virgin queens - mated naturally in mating flights with 10-20 drones once in live time

  - The depot of sperm (Spermotheca), sperms are alive for 4-5 years.

  - Each queen carries 2 alleles in her own cells plus the sperm from 10-20 mates

  - <u>Single queens store most of the variation found in the bees of an area</u>

  - The mating season is once in year (May-July)
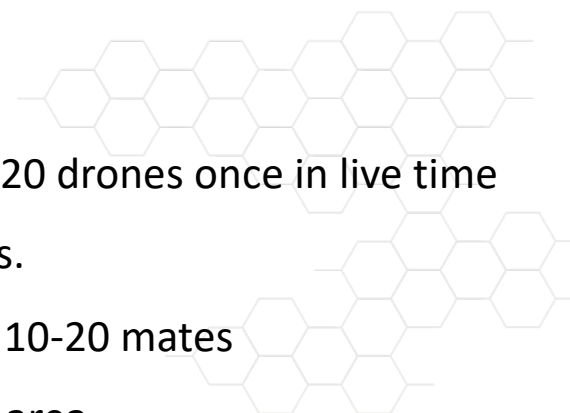
Introduction - Challenges

- Breeding program

Introduction - Challenges

- Mating options

  - Geographical isolation - islands, isolated sites (deep forest, wales ….)

  - Alternative time isolation - labyrinths, cooling systems …

  - Biological saturation on site selected drones outnumbering alien drones

  - Instrumental Inseminations - full pedigree known, but limited by cost labor and access, sometimes used in single drone insemination

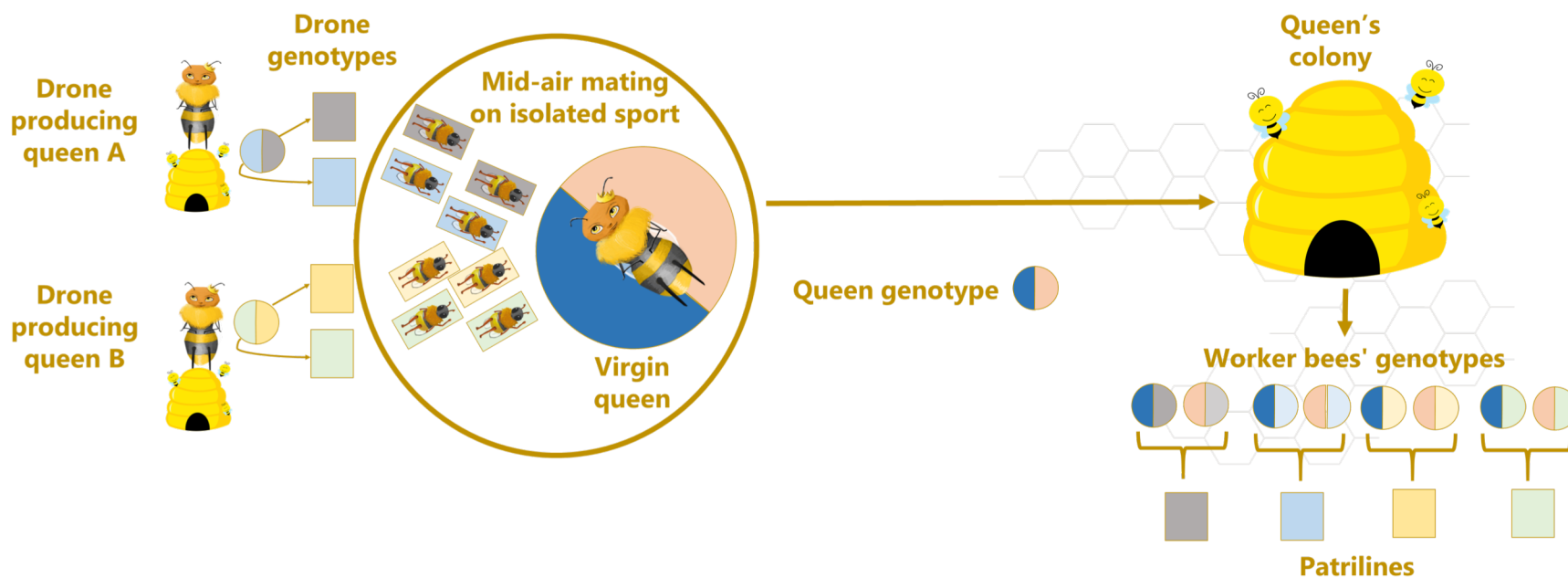 - Mating stations assuming to be sufficiently free of drones from unknown origins
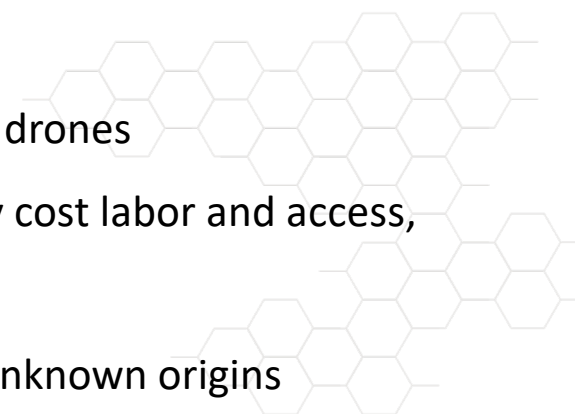
Mating station
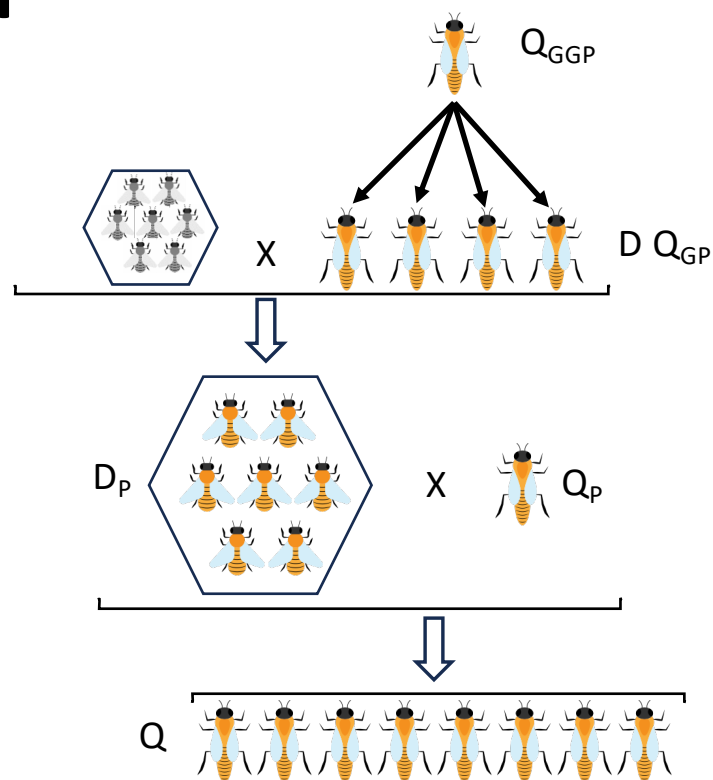
Introduction - Solution

- Uncertainty in pedigrees => Solution - genomic

Solution & Aim

- DNA samples - use of 5 microsatellite for:

| DPQ (or ~10 drones per DPQ) Mated queen | → reference genetic compositors of matting partners |

| Sampling ~20-30 WB brood from nucs | → verification and quantification of drones with known origin |

- **Aim**

  - Cut costs – sampling fewer samples per brood/colony.

  - Keep considerable reliability of confirmation

M & M

- Material:

  - Data - BeeConSel project

  - One year and one mating station data - 20 colonies mated

  - Samples genotyped (n=483),

  - 5 microsatellites (A0007, A0013, Ap043, Ap055, and B0124) reads, presence of 81 alleles.

  - From each nuc 30 worker bee brood was sampled => 16-30 DNA successful reads (mean 24.42)

  - The reliability of all samples set as a baseline (64.19%)

- Method:

  - Random sampling 1 to 5 DNA reads per colony, repeated 100x and summarized
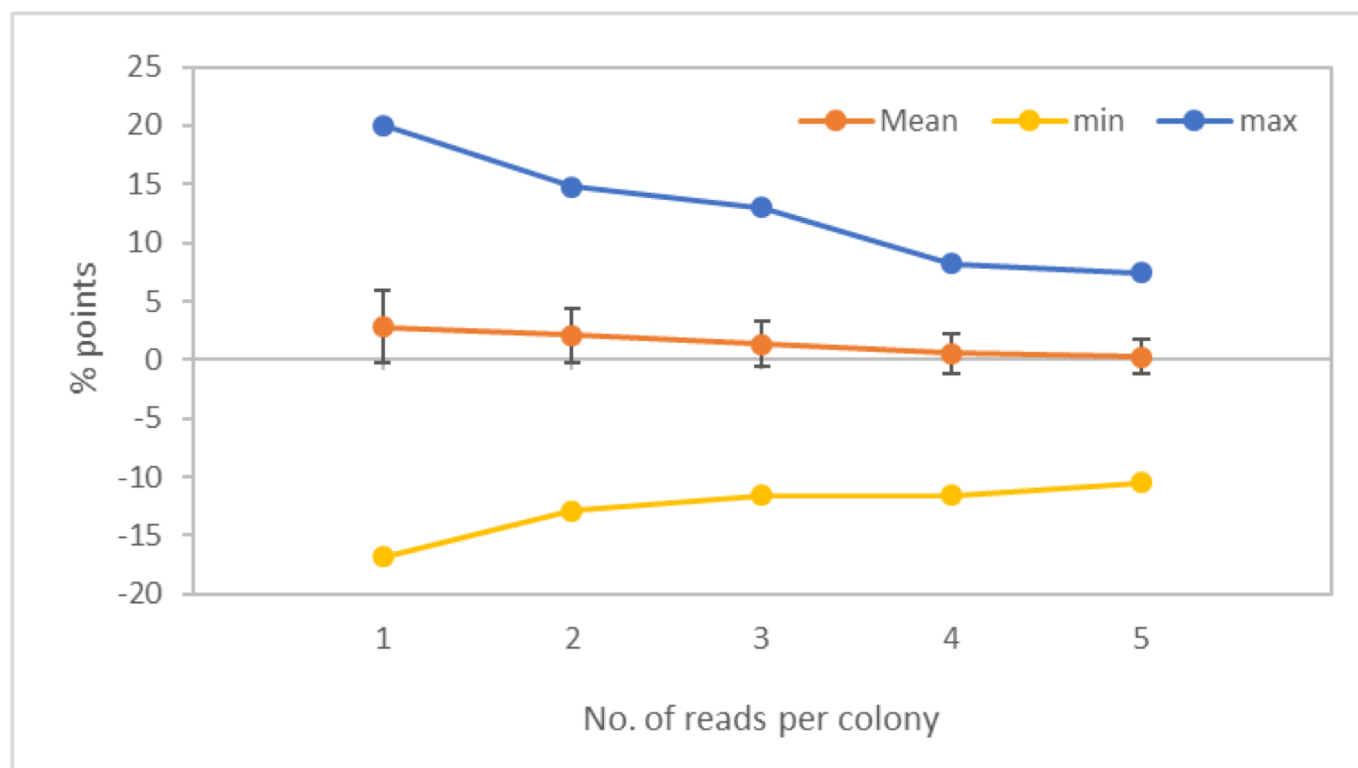
  - Linux bash script

Results

Conclusion

- Generally, high isolated mating station - higher reliability – less samples per nuc needed

- More samples per nuc – higher reliability

- At least 4 – 5 samples per nuc for testing the reliability of the mating stations

- Fewer samples (1-3) - most probably uncertain

- Sevier reducing cost would compromise reliability testing of mating stations

- Next stage different 11 mating station reliability tests (over 10000 reads)
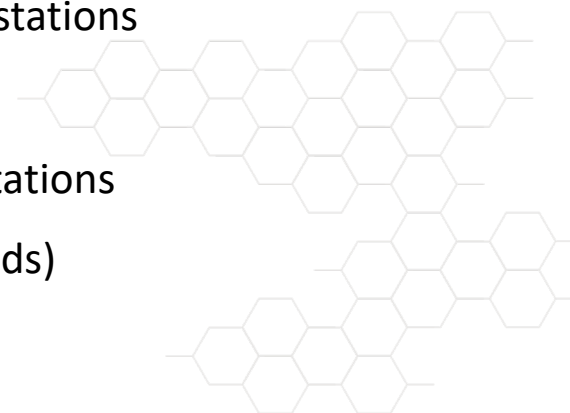
Thank you!

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  - Agricultural Institute in Slovenia

  - CAIS Croatia

  - CARPEA, Macedonia

  - Norwegian association of beekeepers

  - SLU