

# The Genomics Of Migration: Learning From The Painted Lady

A differential gene expression study using *Vanessa cardui* as a model species for migratory behaviour

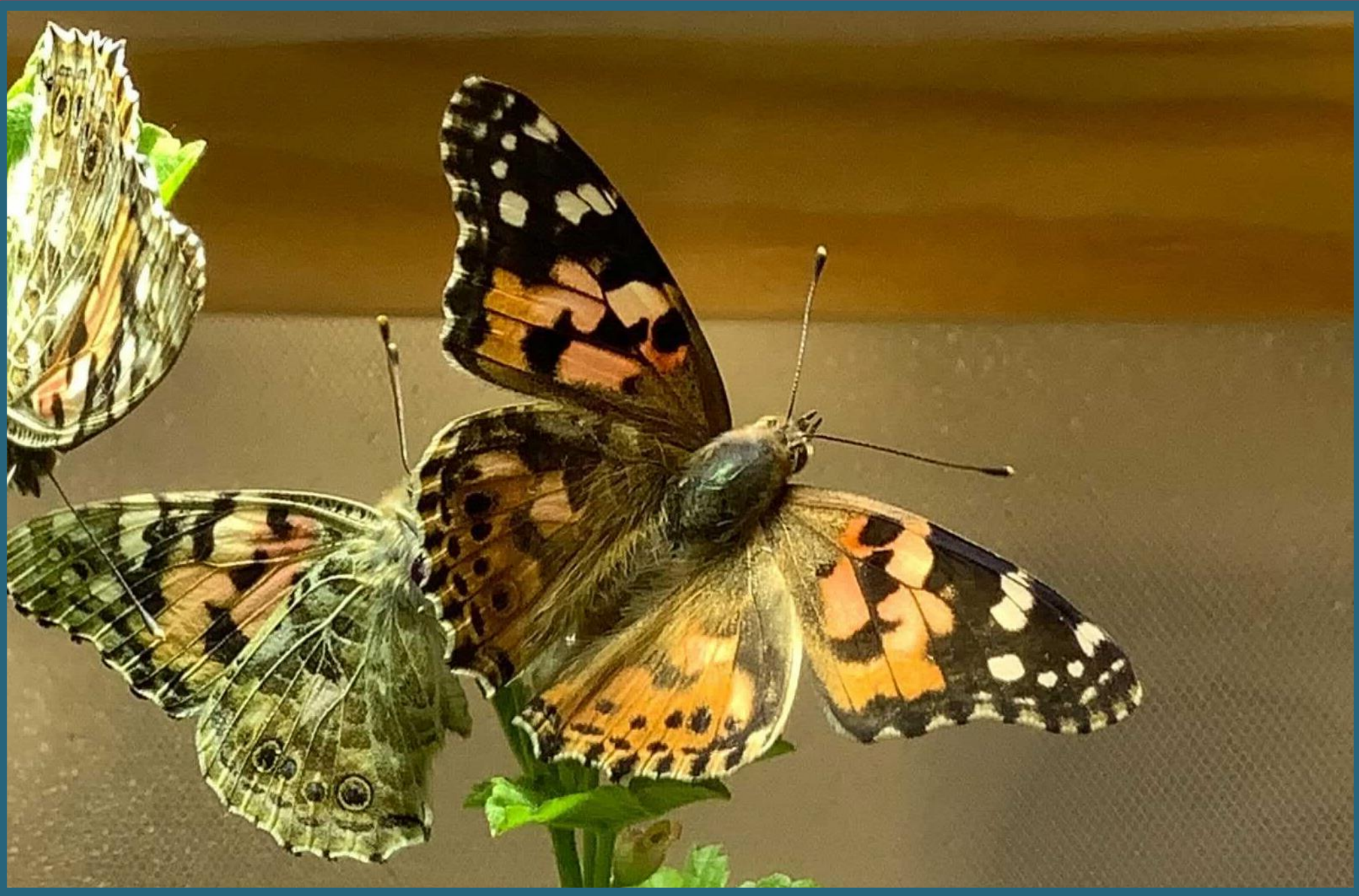
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## Migration

- Migration patterns are characterized by seasonal changes <sup>1</sup> and time/length of migration is affected by temperature and humidity meaning the behaviour is variable.
- Climate change causes shift in the phenology and movement of animals and can affect their migration <sup>2</sup>, for example Pacific pink salmon faced an increased risk of upstream migration failure and experienced higher mortality when held in water warmer than their natural habitat <sup>3</sup>.
- Harsh winters and warm summers drive phenology and extent of migration in aphids, a common agricultural pest <sup>4</sup>, which could pose a threat to food security as their migratory behaviour becomes unpredictable.
- The displacement of organisms toward the Poles is a repeated pattern and is estimated a rate of 17 km per decade in terrestrial species <sup>5</sup>, 72 km per decade in marine ones <sup>6</sup> and is likely to keep increasing to affect a greater number of species in the coming years<sup>7</sup>.
- Assisted migration for non-migratory species has been applied as a method to maintain biodiversity in systems whose habitats are severely impacted by climate change<sup>8</sup>; understanding the underlying mechanisms for migration is key to this approach.
- Study into the genetics of migratory response in existing migratory species is essential for making more accurate predictions of future changes to migration patterns.

## Genetics of migration

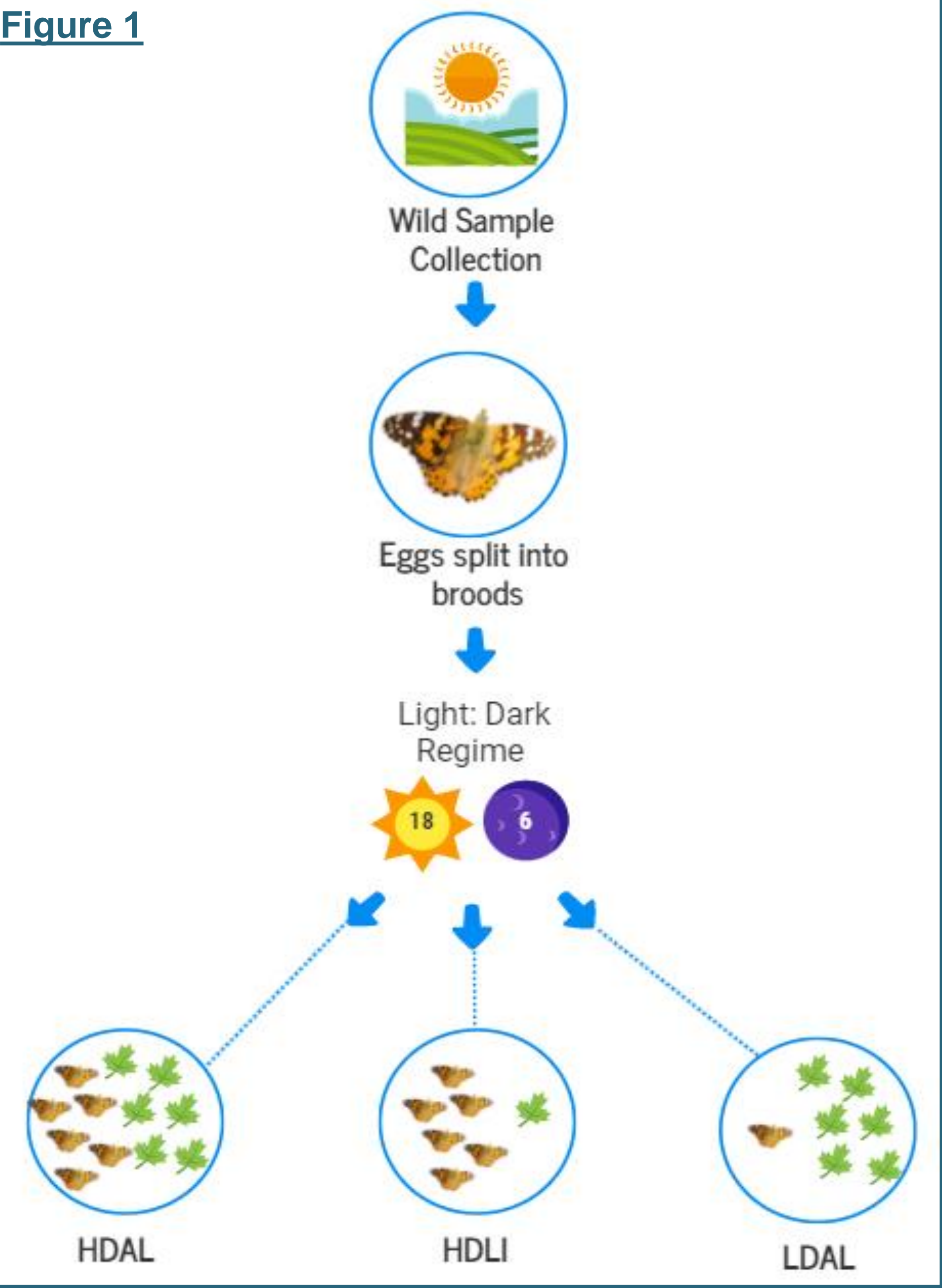
- Food availability and population density are factors that impact whether a population will migrate to or from a specific location <sup>9</sup> but the genetic basis is relatively unknown.
- The North American Monarch butterfly (*Danaus plexippus*) responds to changes in photoperiod and temperature to trigger their seasonal migration <sup>1</sup> but a single migration cycle occurs over multiple generations and re-migrants travel in the opposite orientation to their predecessors <sup>1</sup> suggesting epigenetic control of migration.
- Two populations of Monarch which display very different migratory behaviour were found to have no genomic differences <sup>10</sup> implying migration is not encoded in the genome but an effect of gene expression changes triggered by environmental cues.
- Difference in selection pressure does not reveal the mechanisms triggering the migratory response, so comparative genomics studies must be complemented by in-depth investigations of gene expression differences when comparing individuals predisposed to migrate vs reproduce.



## Vanessa cardui as a study system

- *Vanessa* species display variation in migratory strategies even in closely related populations highlighting how important it is to study gene expression with regards to environment
- *Vanessa cardui* has a majority of migrating populations and minority sedentary populations.
- *V. cardui*, do not enter dormancy (diapause) during unfavourable conditions but continually migrate to suitable environments to reproduce there. This also means their migration can happen anytime <sup>11</sup>
- Some individuals fly > 4000km in their lifetime <sup>12</sup> as part of a 10 generation circular migratory route in the West Palearctic region and have been used as a model for migratory behaviour due to its abundance in varied climates across multiple continents <sup>12</sup>
- Host plant abundance was modelled to impact mating frequency/time and mated females can detect suitable breeding areas <sup>13</sup>

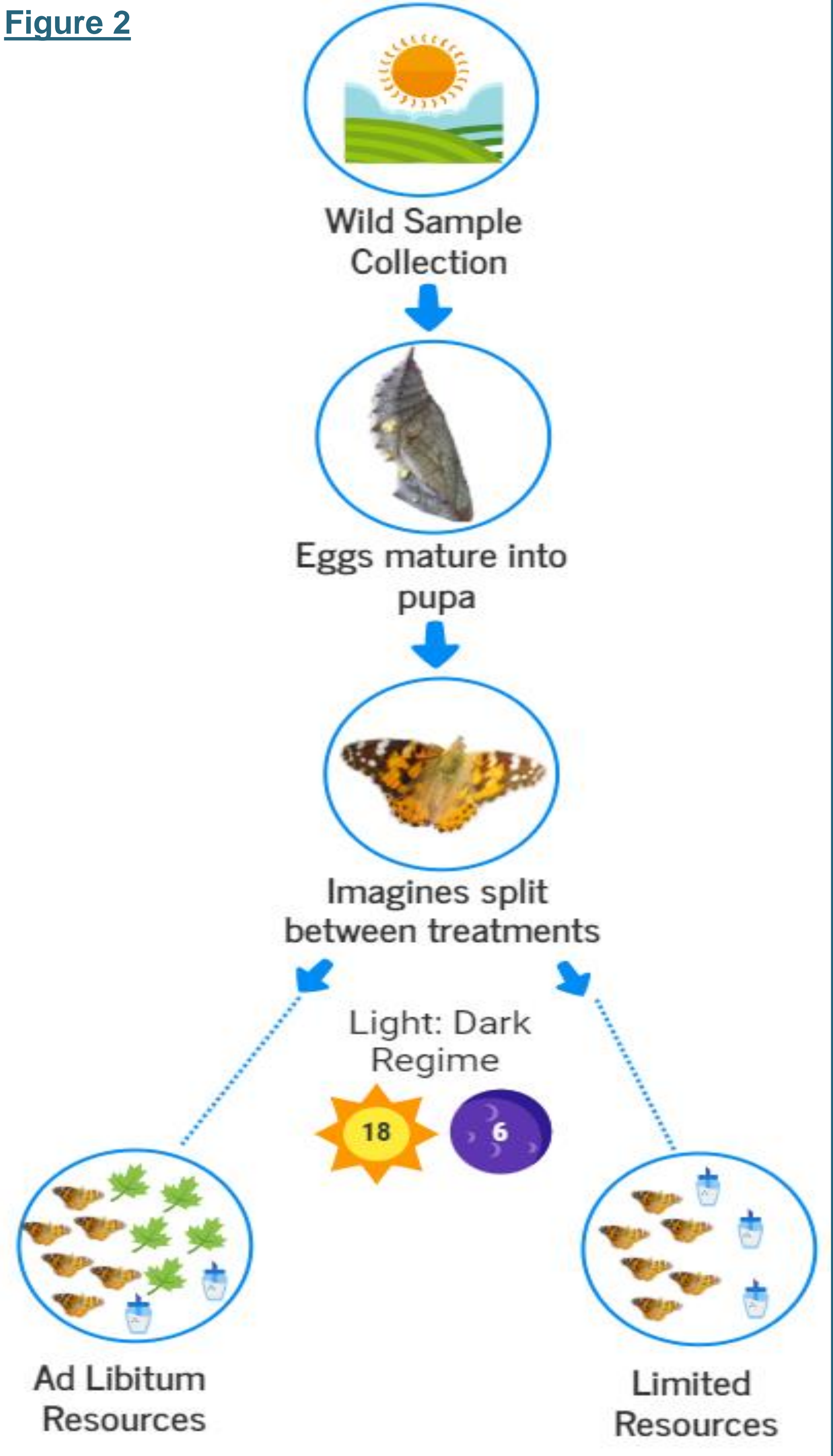
Figure 1



## Aims

1. Investigate difference in transcriptome profiles between populations of *V. cardui* in different treatments aimed to induce or inhibit migratory behaviour
2. Identify the genetic pathways associated with migratory behaviour and further understand the genetic underpinnings of migration
3. Use this informational as a tool for mitigating the consequences of climate change and promote biodiversity<sup>14</sup>

Figure 2



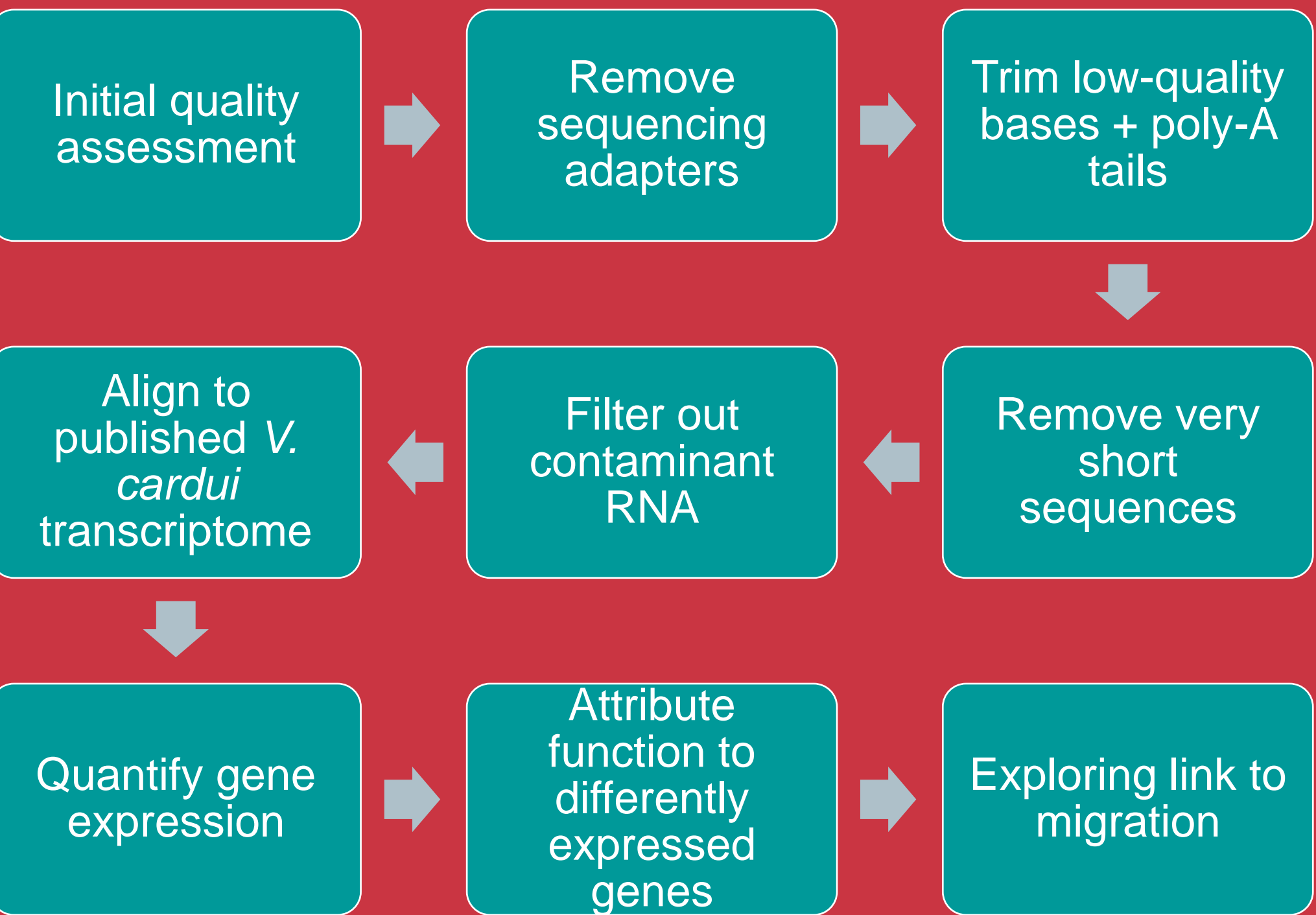
## Experimental Set Up

- Wild *V. cardui* females were collected and allowed to lay eggs on host plants (*Malva sylvestris*) in the lab.
- Eggs were split into treatment groups intended to induce migration (high density [HD], limited resources [LI]) or investment in reproduction (low density [LD], ad libitum [AL]) (Figure 1).
- Population density was investigated with a third group (low density [LD], ad libitum [AL]) (Figure 1).
- RNA samples were extracted from the head and abdomen of individuals from each treatment at four developmental stages (Instar III larva, Instar V larva, Pupa [male and female] and Adult [male and female])
- A simultaneous investigation with adult female *V. cardui* was set up, splitting imagines from pupa into two treatment groups with adult male HDAL (an abundance of the host plant and 10% sugar water) and HDLI (10% sugar water but no host plants) (Figure 2).
- Individuals were sampled after 5 days when the trade-off between mating and migrating should be decided <sup>13</sup>

## RNA Extraction + Sequence Analysis

- RNA extraction was performed on head and abdomen samples of individuals using the RNeasy Mini Kit (Qiagen) and RNA extractions suspended in nuclease free water
- Quality and quantity of the RNA was measured using Bioanalyzer, Qubit and Nanodrop
- RNA solutions were sent to NGI SciLife Lab for library preparation and sequenced using Illumina paired-end sequencing.
- RNA sequence analysis will be performed based on the pipeline outlined by Näsval et al, 2021 (Figure 3).

Figure 3



## Expected Results + Significance

It is difficult to predict precise gene families that will experience differential gene expression but we speculate they will be in these areas:

- genes associated with **metabolic pathways** are affected by resource allocation because timing of energy allocation is important in oogenesis : flight syndrome
- Monarchs experience reproductive diapause during migration <sup>15</sup> so genes associated with **reproduction** could be less expressed in the *V. cardui* treatment with limited resources, hence our focus on gonad RNA.
- **Flight muscle function** genes have been linked to migration in Monarchs <sup>15</sup> so our study could highlight phenotypic difference between migratory and sedentary populations

These results will be significant for:

- Understanding the genetic underpinnings of migratory behaviour
- Predicting the influence of climate change on migratory organisms
- The agricultural sector to anticipate potential changes to pollinator and pest species' migration
- Providing information to conservation groups mitigating the impacts of climate change and habitat loss on biodiversity

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