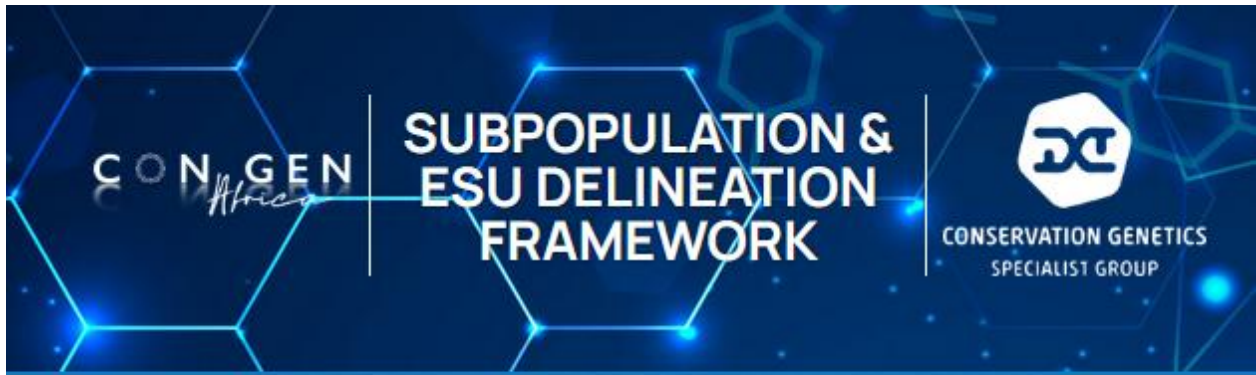


Step-by-step guide:

to support

**From Framework to Practice: Operational tools for Identifying
Subpopulations and Evolutionary Significant Units to Support Conservation
Management**

(DOI: <https://doi.org/10.32942/X2RK9Q>)



Subpopulation and ESU delineation framework by the
IUCN Conservation Genetics Specialist Group (CGSG)

How to cite:

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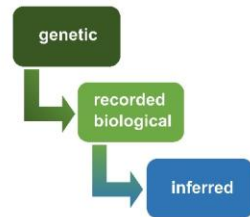
Introduction

The Purpose of this guide is to help users navigate a two-phase webtool to apply a framework for identifying subpopulations and Evolutionarily Significant Units (ESUs) in species based on different data types (genetic, recorded biological and inferred; Figure 1A). It first explains the general features of the webtool and then presents two case studies to demonstrate how the tool can be applied.

Overview of the testing process/framework (Figure 1B)

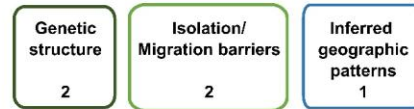
1. Species are assessed in so-called 'candidate scenarios' (see below for more details)
2. Within the framework three different lines of evidence ("genetic evidence", "recorded-biological evidence" and "inferred evidence") are used to accommodate different data availabilities for species (Figure 1A)
3. Four different scenarios can be assessed simultaneously
4. In Phase 1, genetically meaningful subpopulations are identified (Figure 1B)
5. Candidate scenarios scoring ≥ 2 points qualify for Phase 2
6. In Phase 2, the subpopulations identified in Phase 1 may be tested individually or grouped into candidate ESU scenarios (Figure 1B). As with Phase 1, different lines of evidence are used to assess their ESU status.
7. Please note: Genetic or genomic data are required in Phase 2 to achieve a score of ≥ 10 and delineate genetically meaningful ESUs.

A) Lines of evidence



B) Two - Phase Framework

Phase 1: Identify subpopulations



Score candidate **subpopulation scenarios**

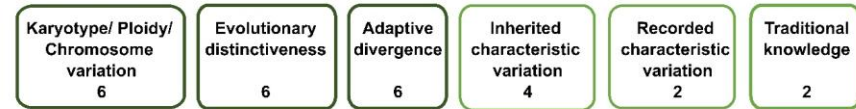
***Null - Hypothesis:** 1 subpopulation = no subdivision within species = 1 ESU (Phase 1B)

Tested Scenarios & Results:

Subpop - Scenario A) x subpopulations
Subpop - Scenario B) n subpopulations
...

Scoring: **≥ 2 points** is required to proceed to **Phase 2**
(group into 'meaningful' ESU - Scenarios)

Phase 2: Delineate Evolutionarily Significant Units (ESUs)



Score candidate **ESU scenarios** (note: these scenarios can differ from the subpopulation scenarios)

***Null - Hypothesis:** 1 ESU (Phase 2B)

Tested Scenarios & Results:

ESU - Scenario X) k ESUs
ESU - Scenario Y) j ESUs
...

Scoring: **≥ 10 points** is required to delineate **distinct ESU(s)**
< 10 points due to various situations:

- a) final score could not reach or exceed 10 points (even if all missing data were supportive of the tested scenario)
= **no support for the ESU - Scenario**
- b) final score does not currently reach or exceed 10 points*, but could do so if missing data subsequently prove to be supportive of the scenario
= **scenario indicates possible ESUs (pESUs)**

*if only non-genetic evidence is currently scored, the ESU-scenario should be rather considered as 'data-deficient'

* Scenarios falling under the **Null - hypothesis - 1 subpopulation or ESU -** can be formally tested using the link provided in each Phase (Phase 1B and Phase 2B).

Figure 1. Lines of evidence (A) and the Two-phase framework (B) to assess the strength of evidence for subpopulations and Evolutionarily Significant Units (ESUs). Different lines of evidence are highlighted in color (dark green, green and blue). Maximum scores are the numbers provided below each category in the boxes based on genetic or non-genetic evidence from the literature and other documentation. Below each phase the scoring system is explained by exemplary imaginary 'scenarios'.

Getting Started

The webtool can be accessed from www.congenafrika.com/demo-survey/.

On the landing page you will get an overview of the framework, including its purpose, different sections of the framework, and its scoring system.

Purpose of the framework

This two-phase framework is intended to help identify appropriate units for biodiversity threat assessments as employed by the IUCN (i.e., IUCN Red List and IUCN Green Status of Species) and other entities. The IUCN currently conducts assessments at three levels: species, subspecies, and subpopulations. The vast majority of assessments are carried out at the species level with subpopulations identified in only 5% of assessments. Expanding these assessments below the species-level through a standardised definition of subpopulations, together with the inclusion of Evolutionarily Significant Units (ESUs), would better capture patterns of genetic diversity and ensure sustainable conservation efforts. For more information, please look at the scientific paper accompanying this framework (<https://ecoevovxiv.org/repository/view/9129/>).



Subpopulation and ESU delineation framework by the IUCN Conservation Genetics Specialist Group (CGSG)

Purpose:

This two phase framework is intended to help IUCN based species assessments (i.e. IUCN Red List and IUCN Green Status of Species). Species assessments are currently done at three levels: the species as a whole, different sub species, and subpopulations. Most assessments are done however on the species level (~95% of species assessed have tested subpopulations). Below species level

assessment in form of a broader implementation and standardisation of subpopulation settings, as well as the inclusion of Evolutionarily Significant Units (ESUs) would reflect genetic diversity patterns more accurately and aid its protection. For more information please look at the scientific paper accompanying this framework.

The framework:

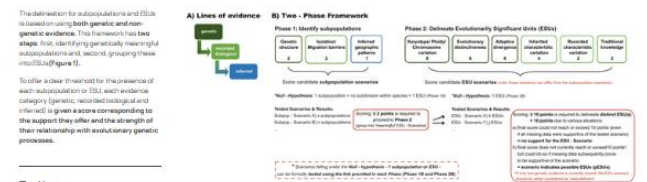


Figure 1. Lines of evidence (A) and the Two-Phase Framework (B) to assess the strength of evidence for subpopulations and Evolutionarily Significant Units (ESUs). Different lines of evidence are highlighted in color (dark green: genetic; light green: recorded biological; blue: inferred). Maximum scores are the numbers provided below each category in the boxes based on genetic or non-genetic evidence from the literature and/or documentation. Below each phase the scoring system is explained separately according to category, scenario.

Scoring:

In Phase 1, scenarios with 2 points or higher indicate the presence of distinct subpopulations. Each scenario with 2-2 points can then be further investigated in Phase 2.

In Phase 2, scenarios comprising potential ESUs can be assessed (number of subpopulations and ESUs might differ). To delineate distinct ESUs, we suggest the need of some genetic/genomic evidence. Only scenarios scoring 10 points or higher are considered to contain distinct ESUs.

Scoring <10 points can have several reasons, such as competing evidence indicating no distinctiveness, or data deficiency in some evidence categories. With scores below 10 we have two options:

a) The candidate scenario could not reach or exceed 10 points (even if all missing data were supportive of the tested scenario)
→ no support for the ESU - Scenario.

b) The candidate scenario does not currently reach or exceed 10 points, but could do so, if missing data subsequently proved to be supportive of the tested scenario → scenario indicates possible ESUs (pESUs).

*However, if a candidate scenario currently only scores in recorded biological evidence types (i.e. non-genetic evidence), the scenarios should be regarded as data deficient rather than as a pESU scenario.

Start Survey >>

Overview of the framework

The delineation of subpopulations and ESUs in this framework draws on both genetic and non-genetic evidence. The framework follows a two-tiered approach: first, identifying genetically meaningful subpopulations and, second, testing subpopulations or groups of subpopulations as ESUs (Figure 1).

To provide clear thresholds for recognising each subpopulation or ESU, evidence is categorised as “genetic”, “recorded biological” or “inferred”. Each category is assigned a score that reflects both the strength of the evidence and its relevance to evolutionary processes.

Scoring system

In **Phase 1** only scenarios with 2 points or higher indicate the presence of distinct subpopulations. Scenarios meeting this threshold can move to Phase 2 to investigate whether the subpopulations or groups of subpopulations qualify as ESUs.

In **Phase 2** scenarios scoring 10 points or higher indicate the presence of ESUs. This can only be achieved with genetic/genomic evidence.

Scores below 10 points may result from either:

- **No support for ESUs.** In this situation, even if missing data were assumed supportive for the tested scenario, a score of 10 could not be reached.
- **Possible ESUs (pESUs).** In this situation, the scenario does not **currently** score 10 points, but could if new evidence later proves to be supportive.

*Note: If the score of a candidate scenario is based **only** on recorded biological (non-genetic) evidence, the scenario should be regarded as having insufficient data, and hence not a pESU.

Species selection

It may not be necessary to assess all species, although there is no restriction either. This framework is intended to help clarify population structure and guide conservation decision-making. Priority should be given to species with likely or suspected genetic structure, to test the relevance of subspecies designations, to understand appropriate management units for translocations, and to taxa that may be experiencing different threats across their range.

Scenario selection

Species should be divided into the most likely groups of subpopulations and ESUs, hereafter referred to as *candidate scenarios*. These scenarios may be derived from the scientific literature or developed using other relevant criteria based on existing knowledge. This framework is intended for users who are familiar with the species' biology, life history traits and other pertinent information. We therefore advise against the exhaustive testing of all possible scenarios (e.g., 1-7+ subpopulations or ESUs), as many are likely to lack sufficient supporting data. Different scenarios can be tested in Phase 1 and Phase 2, as multiple subpopulations may be grouped into a single ESU. An option is also available to test the likelihood of a single metapopulation or ESU. If users choose this route, they will be redirected to a separate survey, as the questions for assessing a single grouping differ from those used for multiple groupings.

To start the assessment, go to www.congenafrika.com/demo-survey/, click the **“Start Survey”** button.

Conducting the survey

Navigating the survey pages

Left-hand information panel

On every testing page, a panel appears on the **left** side of the screen. This panel provides definitions for technical terms, explanations of principles used in the framework, and links to relevant reference materials. It is intended to help clarify concepts and ensure consistent interpretation of the criteria during scoring. Users are encouraged to refer to this panel regularly, especially when encountering unfamiliar terminology or when making decisions that require an understanding of the key principles.

Survey questions and layout

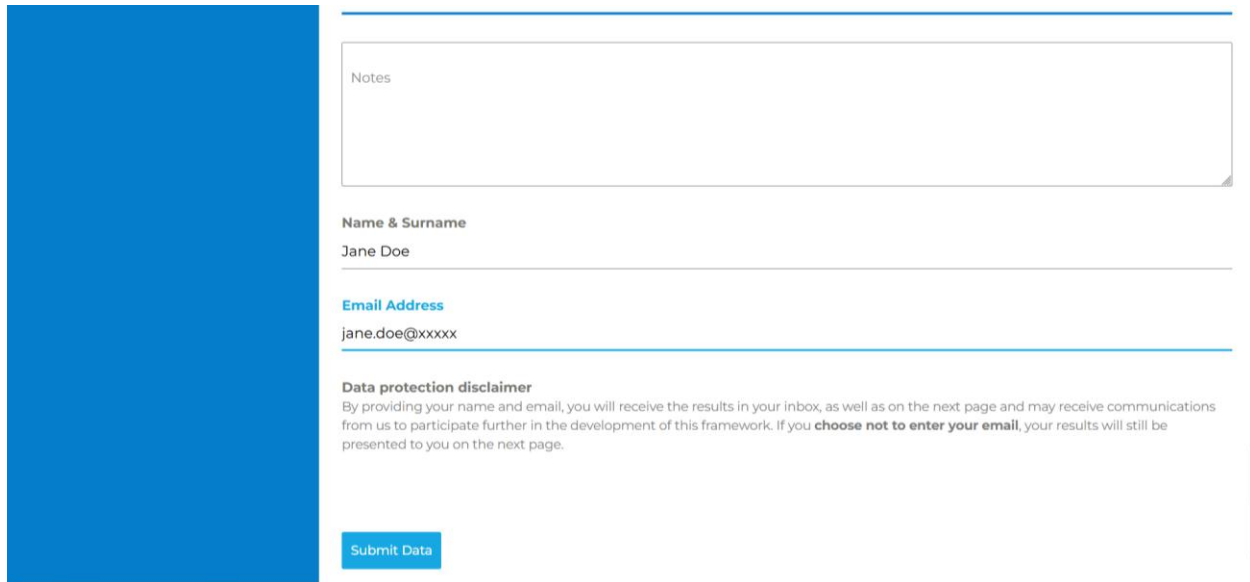
At the start of the survey, users are asked to enter the name of the species and the scenarios being investigated to allow for easier recall and context should an email of the results be requested.

Questions are grouped according to evidence categories (“genetic”, “recorded biological”, and “inferred”). To help users understand the type of information each question requires, information bubbles have been included. Hovering over the “i” icon will display additional guidance.

Up to four scenarios can be tested simultaneously, displayed as adjacent columns. Answers are required for each scenario to enable direct comparison and ensure consistent scoring. If more than 4 scenarios need to be assessed, additional surveys must be completed, and the scores for each scenario should be recorded and retained for reference. This process is supported by the email response function which sends results directly to the user if an email address is provided.

After completing all questions, users have the option to enter additional notes and their email address before submitting the survey. These notes and the email addresses **are not stored or**

retained by the survey developers. They are used solely for the user’s convenience. The email address is used to send a copy of the results to the user. Clicking the **“Submit Data” button** will display a results screen showing the total scores for each scenario.



Notes

Name & Surname
Jane Doe

Email Address
jane.doe@xxxx

Data protection disclaimer
By providing your name and email, you will receive the results in your inbox, as well as on the next page and may receive communications from us to participate further in the development of this framework. If you **choose not to enter your email**, your results will still be presented to you on the next page.

Submit Data

Phase 1: Identify subpopulations

Phase 1 has two survey options. The main survey option takes users through scenarios where the existence of multiple subpopulations is being queried; while Phase 1 B allows users to assess the possibility the species exists as a single metapopulation.

To identify meaningful subpopulations or the presence of a single metapopulation three different evidence categories are used. Based on existing data and knowledge it is assessed: (1) if **genetic structure** can be determined/detected, (2) if there is **natural isolation of fragmentation** of the subpopulations/metapopulation tested and (3) in case there is no direct (observed) data available of the focal species, if there is **inferred evidence of a likely fragmentation** of the subpopulations/metapopulation.

Testing a single metapopulation scenario

To access the single metapopulation survey, click on the blue button “**Test a metapopulation**” on the main Phase 1 page.

Phase 1: Identify Subpopulations

This survey is best views on Desktop, or rotate your phone for a better view.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Possible Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Null-hypothesis: If no candidate scenarios can reach the minimum threshold of 2 points, the species may comprise a single subpopulation and therefore a single ESU.

Scoring:

- 2 points or higher = distinct subpopulations

Which species are you testing? *

If you want to specifically assess a scenario with only a **single metapopulation** press here:

TEST A METAPOPOPULATION >>

To assess scenarios with several subpopulations please continue.

How many Scenarios do you want to assess?

1 Scenario

1 Scenario

2 Scenarios

3 Scenarios

4 Scenarios

This will take you to a separate survey, with a set of questions that are designed to test for a single metapopulation.

Phase 1B: Identify a single metapopulation

This survey is best views on Desktop, or rotate your phone for a better view.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Possible Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 2 points or higher = one continuous population


Which species are you testing?

Scenario A

Scenario 1 name

Genetic evidence

Is there no genetic structure?

Evidence through (multiple can apply): 

- No clear population cluster
- Low pairwise Fst Values
- No private Alleles

Caution: If there is knowledge of human-mediated/controlled genetic structure or extreme bottleneck events, genetic clustering detected in a species does not automatically indicate the presences of separate subpopulations.

Testing scenarios with multiple subpopulations (up to 4)

To test scenarios of multiple subpopulations, users can simply stay on the main Phase 1 page or go back to it (if Phase 1 B was used first).

Remember, only four scenarios can be assessed simultaneously.

Phase 1: Identify Subpopulations

This survey is best views on Desktop, or rotate your phone for a better view.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Possible Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Null-hypothesis: If no candidate scenarios can reach the minimum threshold of 2 points, the species may comprise a single subpopulation and therefore a single ESU.

Scoring:

- 2 points or higher = distinct subpopulations

Which species are you testing? *

If you want to specifically assess a scenario with only a **single metapopulation** press here:

TEST A METAPOPOPULATION >>

To assess scenarios with several subpopulations please continue.

How many Scenarios do you want to assess?

4 Scenarios

Scenario A	Scenario B	Scenario C	Scenario D
Scenario 1 name	Scenario 2 name	Scenario 3 name	Scenario 4 name

Disclaimer

The following questions are assessed between **ALL** subpopulations **WITHIN** a scenario. So if a candidate scenario comprises 3 subpopulations, and there are only differences between subpopulation 1 and subpopulation 2, but subpopulation 3 is similar to 2, then you need to test for a new scenario merging subpopulation 2 and 3.

Phase 2: Delineate Evolutionarily Significant Units (ESUs)

Similar to Phase 1, two survey options are available. One to investigate whether the species exists as a single ESU (Phase 2B) or whether multiple ESUs exist (Phase 2).

To delineate subpopulation (groupings) into single or multiple ESUs, six evidence categories are used. Based on existing data and knowledge it is assessed: (1) if **inherited variation can be detected at the chromosome level** (number, structure or ploidy level), (2) if there is genetic evidence of **long-term reproductive isolation**, (3) if **local adaption** can be detected, if there are **characteristic differences** recorded due to (4) **inheritance** or (5) **transmission** via e.g., learning behaviour, so not robustly shown to be inherited and (6) if there is any indication of distinctives of tested ESUs based on **traditional and local knowledge** (not scientifically tested).

Testing a single ESU scenario

To access the single ESU survey, click on the blue button “**Test 1 ESU**” on the main Phase 2 page.

Phase 2: Delineate Evolutionarily Significant Units (ESUs)

Please use the information provided by the email sent, to complete the following ESU survey.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Probable Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 10 points or higher = distinct ESU
- Below 10 points:
 - a) 10 points cannot be reached or exceeded (even if all missing data were supportive of the tested scenario) = **no support for the ESU - Scenario.**

Which species are you testing?

If you want to specifically assess a scenario with only **1 ESU**, press here:

TEST 1 ESU >>

To assess scenarios with several ESUs please continue

How many scenarios are you testing?

1 Scenario

1 Scenario

2 Scenarios

3 Scenarios

4 Scenarios

This will take you to a separate survey as a different set of questions are needed to test for a single grouping.

Phase 2B: Identify 1 ESU

Please use the information provided by the email sent, to complete the following ESU survey.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Probable Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- Score a minimum of 10 points = one Evolutionarily Significant Unit (ESU)

Which species are you testing?

Scenario A

Scenario 1 name

Genetic evidence

Is there no/limited variation in chromosome numbers, ploidy level or chromosome structure?

Evidence through (multiple can apply):

- No or limited karyotype variation
- No or limited difference in ploidy levels between units
- No or limited structural variation of chromosomes between units

Testing scenarios with multiple ESUs (up to 4)

To assess scenarios with multiple ESUs, simply stay on (or go back to) the main Phase 2 page. Remember, only four scenarios can be assessed simultaneously

Phase 2: Delineate Evolutionarily Significant Units (ESUs)

Please use the information provided by the email sent, to complete the following ESU survey.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Probable Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 10 points or higher = distinct ESU
- Below 10 points:
 - a) 10 points cannot be reached or exceeded (even if all missing data were supportive of the tested scenario) –no support for the ESU – Scenario.

Which species are you testing?

If you want to specifically assess a scenario with only **1 ESU**, press here:

TEST 1 ESU >>

To assess scenarios with several ESUs please continue

How many scenarios are you testing?

4 Scenarios

Scenario A	Scenario B	Scenario C	Scenario D
Scenario 1 name	Scenario 2 name	Scenario 3 name	Scenario 4 name

Disclaimer

The following questions are assessed between **ALL** ESUs **WITHIN** a scenario. So if a candidate scenario comprises 3 ESUs, and there are only differences between ESU 1 and ESU 2, but ESU 3 is similar to 2, then you need to test for a new scenario merging ESU 2 and 3.

Case Studies

To assist in navigating the framework, two species examples are provided. For both case studies, existing literature and previous knowledge was used to determine candidate scenarios which were tested in Phase 1 and 2. We did not intend to test all possible scenarios, since that would result in many “data-deficient” scenarios. We rather built on existing theories and based our scenarios on them.

Case Study 1 – The Hawaiian koa tree (*Acacia koa*)

The Hawaiian koa is an endemic tree species of high ecological and economical importance, growing in a range of different habitats (various elevation levels with wet to drier conditions¹). It is distributed on the four main Hawaiian Islands (Hawai‘i, Maui, O‘ahu and Kaua‘i) and genetic as well as morphological studies have been conducted, therefore the Hawaiian koa represents a good case study.

1. Fill in species and scenario information.

Based on two scientific studies utilising genetic data and the distributional pattern of the Hawaiian koa, three different candidate scenarios are tested here: (1) two subpopulations based on microsatellite markers² (2) seven subpopulations based on genomic (SNP) data³ and (3) four subpopulations based on their occurrence of the four main Hawaiian oceanic islands (geographic distribution)¹.

Please use the dropdown menu below for additional information.

Subpopulations

Evolutionarily Significant Units (ESU)

Possible Evolutionarily Significant Unit (pESU)

Gene Flow

Potential IUCN Species assessment

References

Null-hypothesis: If no candidate scenarios can reach the minimum threshold of 2 points, the species may comprise a single subpopulation and therefore a single ESU.

Scoring:
+ 2 points or higher = distinct subpopulations

Phase 1: Identify Subpopulations

This survey is best views on Desktop, or rotate your phone for a better view.

Which species are you testing? *

Acacia koa

If you want to specifically assess a scenario with only a **single metapopulation** press here:

TEST A METAPOPOPULATION >>

To assess scenarios with several subpopulations please continue.

How many Scenarios do you want to assess?

3 Scenarios

2 subpopulations

7 subpopulations

4 subpopulations

Scenario 1 name

Scenario 2 name

Scenario 3 name

Disclaimer

The following questions are assessed between **ALL** subpopulations **WITHIN** a scenario. So if a candidate scenario comprises 3 subpopulations, and there are only differences between subpopulation 1 and subpopulation 2, but subpopulation 3 is similar to 2, then you need to test for a new scenario merging subpopulation 2 and 3.

¹ Baker, P. J., Scowcroft, P. G., & Ewel, J. J. (2009). Koa (*Acacia koa*) ecology and silviculture. Gen. Tech. Rep. PSW-GTR-211. Albany, CA: U.S. Department of Agriculture, Forest Service, Pacific Southwest Research Station. 129 p, 211.

² Fredua-Agyeman, R., Adamski, D., Liao, R. J., Morden, C., & Borthakur, D. (2008). Development and characterization of microsatellite markers for analysis of population differentiation in the tree legume *Acacia koa* (Fabaceae: Mimosoideae) in the Hawaiian Islands. *Genome*, 51(12), 1001–1015.

³ Gugger, P. F., Liang, C. T., Sork, V. L., Hodgskiss, P., & Wright, J. W. (2018). Applying landscape genomic tools to forest management and restoration of Hawaiian koa (*Acacia koa*) in a changing environment. *Evolutionary Applications*, 11(2), 231–242.

2. Answer the questions for each scenario.

a. Genetic evidence

For two of the three candidate scenarios: (1) two subpopulations and (2) seven subpopulations, **genetic structure was detected based on clustering analyses and F_{ST} values**. In scenario (1) two distinct genetic clusters were identified: one on the island of Kaua'i and the second cluster was distributed across the other three islands: Hawai'i, Maui and O'ahu ². In scenario (2) seven distinct subpopulations were distributed across the various islands. For the islands of Maui, O'ahu and Kaua'i one subpopulation dominated on each, whereas on the island of Hawai'i five of the seven detected subpopulations occurred, with detectable admixture among those. However limited gene flow was detected between the four oceanic islands of Hawai'i³.

Genetic evidence

Is there evidence of genetic structure?

Evidence through (multiple can apply) ⓘ

- Population Clustering Analyses
- Pairwise F_{ST} Values
- Private Alleles

Caution: If there is knowledge of human-mediated/controlled genetic structure or extreme bottleneck events, genetic clustering detected in a species does not automatically indicate the presences of separate subpopulations.

2 subpopulations	7 subpopulations	4 subpopulations
<input checked="" type="radio"/> Yes	<input checked="" type="radio"/> Yes	<input type="radio"/> Yes
<input type="radio"/> No	<input type="radio"/> No	<input checked="" type="radio"/> No
<input type="radio"/> No data	<input type="radio"/> No data	<input type="radio"/> No data

b. Recorded biological evidence

For Scenario (1) and (2) no clear indication of natural isolation through migration barriers can be detected. In scenario (1) one of the two subpopulations is distributed across three independent islands, highlighting that migration is possible between islands. In scenario (2) three of the seven subpopulations occur on three separate islands, however the remaining four subpopulations are only recorded from the island of Hawai'i where admixture has been detected³. In scenario (3), four subpopulations are tested according to the distribution of the Hawaiian koa on the four oceanic islands¹. Therefore, each of the four subpopulations constitutes an island which is indicative of four biogeographical zones.

Recorded biological evidence

Is there evidence of natural isolation or subpopulation fragmentation (through migration barriers)?

Evidence through (multiple can apply): ⓘ

- Occupation of different biogeographical zones
- Occupation of discrete remnants of historical habitats with little/no natural migration

Caution: In this framework, the natural fragmentation of subpopulations should be distinguished from human-mediated/controlled fragmentation of subpopulations, since the latter does not automatically indicate the presences of separate subpopulations.

2 subpopulations

- ☐ Yes
☒ No
☐ No data

7 subpopulations

- ☐ Yes
☒ No
☐ No data

4 subpopulations

- ☒ Yes
☐ No
☐ No data

c. Inferred evidence

For all three scenarios, no information could be found for indirect evidence of subpopulation fragmentation and therefore all scenarios were scored as having “No data”.

Inferred evidence

Is there inferred evidence of likely subpopulation fragmentation?

Note: Only relevant if genetic or recorded evidence from the focal species is absent. Evidence of inferred patterns can be used. If nothing is known about the focal species, co-occurring or closely related species could give indication of subpopulation structure.

Evidence through (multiple can apply): ⓘ

- **inferred** subpopulation disjunction (e.g., species distribution modelling)
- Phylogeographic and/or biogeographic evidence **from co-occurring species**
- Assumption of biogeographical data/refugia **without direct evidence**
- **inferred** dispersal distance and buffer according to the CBD indicators

2 subpopulations

- ☐ Yes
☐ No
☒ No data

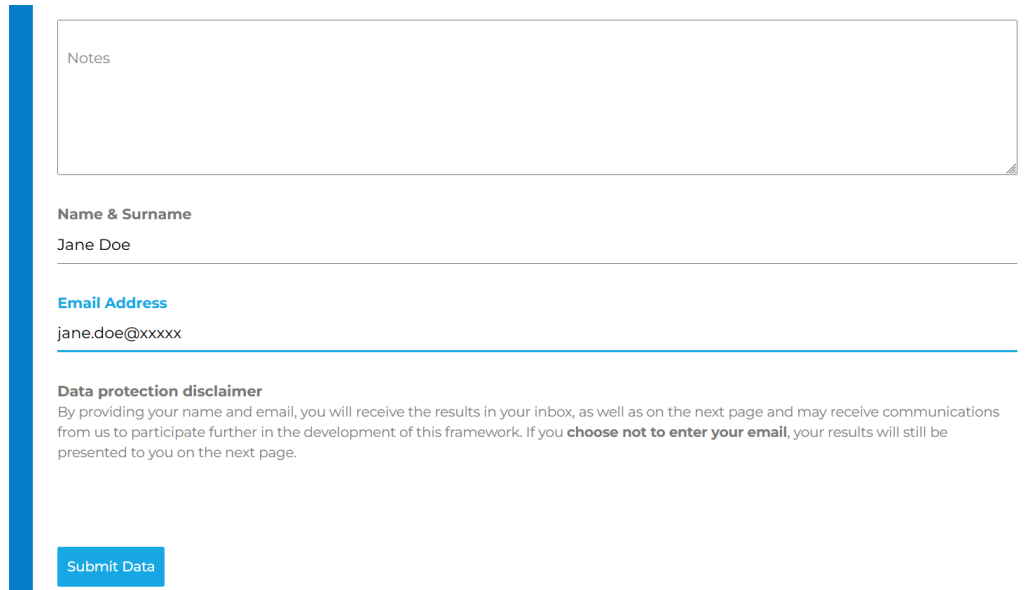
7 subpopulations

- ☐ Yes
☐ No
☒ No data

4 subpopulations

- ☐ Yes
☐ No
☒ No data

3. Provide your name and e-mail address (optional). This will allow the results to be sent to your email address.



Notes

Name & Surname
Jane Doe

Email Address
jane.doe@xxxxx

Data protection disclaimer
By providing your name and email, you will receive the results in your inbox, as well as on the next page and may receive communications from us to participate further in the development of this framework. If you **choose not to enter your email**, your results will still be presented to you on the next page.

Submit Data

4. Click the **“Submit Data”** button to receive the results (mandatory to get results).

5. Interpreting the results.

This is the results page. It lists each question and the responses entered by the user as verification. Below the questions is a summary of the scores for each scenario.

For the Hawaiian koa tree, all three candidate scenarios meet the minimum threshold of two points, and therefore qualify as subpopulation scenarios. Where scores are tied, we recommend selecting the one based on the most robust data to maximize the conservation of genetic diversity. In this case, Scenario (2), based on high-resolution genomic markers (11,000 SNPs from >300 samples via genotyping-by-sequencing³), identified seven distinct subpopulations, while using nine microsatellites from 170 samples², identified only two. Although microsatellites remain valuable in population genetics, the two-subpopulation scenario (2) is unlikely to represent the true subpopulation structure. Instead, it might better reflect higher-level structuring at the ESU level, along with Scenario (3) (4 islands subpopulations). Overall, the seven subpopulation scenario better represents subpopulation structure.

All three scenarios scoring equally higher (two points) and therefore can be tested in Phase 2.

Phase 1: Identify Subpopulations

This survey is best views on Desktop, or rotate your phone for a better view.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Possible Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Null-hypothesis: If no candidate scenarios can reach the minimum threshold of 2 points, the species may comprise a single subpopulation and therefore a single ESU.

Scoring:

- 2 points or higher = distinct subpopulations
- Less than 2 points = stop assessment

Lines of evidence

Thank you , for participating in the CGSG Subpopulation and ESU survey.

The species you tested is the **Acacia koa**

Your Survey Results:

1. Is there evidence of population structure? (2 points)

2 subpopulations Yes	7 subpopulations Yes	4 subpopulations No	
-------------------------	-------------------------	------------------------	--

2. Is there evidence of natural isolation or subpopulation fragmentation (through migration barriers)? (2 points)

2 subpopulations No	7 subpopulations No	4 subpopulations Yes	
------------------------	------------------------	-------------------------	--

3. Is there inferred evidence of likely subpopulation fragmentation? (1 points)

2 subpopulations No	7 subpopulations No	4 subpopulations No	
------------------------	------------------------	------------------------	--

Total score for the scenario: **2 subpopulations** is 2
Total score for the scenario: **7 subpopulations** is 2
Total score for the scenario: **4 subpopulations** is 2
Total score for the scenario: is 0

For scenarios that scored 2 points or higher, please use the button below to continue to Phase 2.

GO TO PHASE 2 >>

You'll be receiving the results in your inbox soon!

5. Click on the “Phase 2” button to proceed.

6. As in Phase 1, enter the species and scenario information. This does not get carried over from Phase 1.

Based on the results of Phase 1, we tested three candidate scenarios for delineating Evolutionarily Significant Units (ESUs): (1) two ESUs and (2) seven ESUs and (3) four ESUs.

Phase 2: Delineate Evolutionarily Significant Units (ESUs)

Please use the information provided by the email sent, to complete the following ESU survey.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Probable Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 10 points or higher = distinct ESU
- Below 10 points:
 - a) 10 points cannot be reached or exceeded (even if all missing data were supportive of the tested scenario) = no support for the ESU - Scenario.
 - b) 10 points are currently not reached or exceeded, but could if missing data subsequently prove to be supportive of the tested scenario = scenario indicates possible ESUs (pESUs)*

Which species are you testing?

Acacia koa

If you want to specifically assess a scenario with only **1 ESU**, press here:

TEST 1 ESU >>

To assess scenarios with several ESUs please continue

How many scenarios are you testing?

3 Scenarios

2 ESUs	7 ESUs	4 ESUs
Scenario 1 name	Scenario 2 name	Scenario 3 name

Disclaimer

The following questions are assessed between **ALL** ESUs **WITHIN** a scenario. So if a candidate scenario comprises 3 ESUs, and there are only differences between ESU 1 and ESU 2, but ESU 3 is similar to 2, then you need to test for a new scenario merging ESU 2 and 3.

7. Answer the questions for each line of evidence for each scenario being investigated.

a. Genetic evidence

For the three ESU scenarios tested, no karyotype variation was detected among different samples collected from all four islands (all are tetraploid ($2n=52$), independent of the island⁴ .

Genetic evidence

Is there inherited variation in chromosome numbers, ploidy level or chromosome structure?

Evidence through (multiple can apply): ⓘ

- Karyotype variation
- Difference in ploidy levels between units
- Structural variation of chromosomes between units

2 ESUs	7 ESUs	4 ESUs
<input type="radio"/> Yes	<input type="radio"/> Yes	<input type="radio"/> Yes
<input checked="" type="radio"/> No	<input checked="" type="radio"/> No	<input checked="" type="radio"/> No
<input type="radio"/> No data	<input type="radio"/> No data	<input type="radio"/> No data

Taxonomic and phylogeographic knowledge of the Hawaiian koa is still very scarce and only disentangled to a limited extent¹. Therefore, scenarios (1) and (3) were scored as having "No data". For Scenario (2) – testing for seven ESUs – "No" was entered, because on the island of Hawai'i (admixture was detected³. In the framework, ESUs are defined lineages with highly restricted gene flow from others within the species⁵, such that each follows its own evolutionary trajectory⁶ (i.e., no gene flow).

Is there evidence of long-term reproductive isolation?

Evidence through (multiple can apply): ⓘ

- Reciprocal Monophyly
- Molecular Estimate of Divergence Time

2 ESUs	7 ESUs	4 ESUs
<input type="radio"/> Yes	<input type="radio"/> Yes	<input type="radio"/> Yes
<input type="radio"/> No	<input checked="" type="radio"/> No	<input type="radio"/> No
<input checked="" type="radio"/> No data	<input type="radio"/> No data	<input checked="" type="radio"/> No data

⁴ Shi, X. (2003). Genetic improvement of *Leucaena* spp and *Acacia koa* Gray as high-value hardwoods [University of Hawaii at Manoa]. <http://hdl.handle.net/10125/6908>

⁵ Fraser, D. J., & Bernatchez, L. (2001). Adaptive evolutionary conservation: Towards a unified concept for defining conservation units. *Molecular Ecology*, 10(12), 2741–2752.

⁶ Funk, W. C., McKay, J. K., Hohenlohe, P. A., & Allendorf, F. W. (2012). Harnessing genomics for delineating conservation units. *Trends in Ecology & Evolution*, 27(9), 489–496.

Only scenario (3) (four ESUs) showed signals of adaptive divergence through genome-wide association analyses with environmental variables. Genetic divergence among the four Hawaiian Islands correlated most strongly with precipitation, highlighting its important role in water stress adaptation³. Additionally, each island is considered an ecoregion and therefore so called “seed zones” (confirmed by genomic data) are established based on the four islands.⁷

Is there evidence of local adaptation or adaptive divergence?

Evidence through (multiple can apply): ⓘ

- Genomic signals of local adaptation
- Evidence from hybrid zones

2 ESUs

☐ Yes

☒ No

☐ No data

7 ESUs

☐ Yes

☒ No

☐ No data

4 ESUs

☒ Yes

☐ No

☐ No data

b. Recorded biological evidence

Provenance tests across all four islands revealed inherited phenotypic differences (growth form and seed shape) reflecting general adaptations to each islands’ environment¹. We therefore scored a “Yes” for scenario (3) (4 ESUs). This phenotypic data qualifies as an ‘inherited line of evidence’ because it was statistically tested in a controlled provenance trial.

Due to the differences among the four islands, scenarios (1) and (2) were scored as a “No”. Scenario (1) considers three of the four islands as one ESU (based on the microsatellite study conducted²) and scenario (2) is characterised by several ESUs on one island (Hawai’i, based on the genomic study conducted³).

Recorded biological evidence

Is there evidence of inherited characteristic differences?

Evidence through (multiple can apply): ⓘ

- Heritable differences in focal traits observed in experiments or confirmed by analytical tests

2 ESUs

☐ Yes

☒ No

☐ No data

7 ESUs

☐ Yes

☒ No

☐ No data

4 ESUs

☒ Yes

☐ No

☐ No data

⁷ Dudley, N., Jones, T., James, R., Snieszko, R., Wright, J., Liang, C., Gugger, P. F., & Cannon, P. (2017). Applied Genetic Conservation of Hawaiian Acacia koa: An Eco-Regional Approach. *Banking on the Future*.

Evidence of recorded characteristic variation is not available for the Hawaiian koa. Therefore, all scenarios were scored with a “No data”.

Is there evidence supporting transmitted characteristics differences?

Evidence through (multiple can apply): ⓘ

- Cultural or learnt behavioral differences unique or specific between units
- Acquired or transmitted traits (e.g., migration pattern, methylation differences, body size differences)

2 ESUs	7 ESUs	4 ESUs
<input type="radio"/> Yes	<input type="radio"/> Yes	<input type="radio"/> Yes
<input type="radio"/> No	<input type="radio"/> No	<input type="radio"/> No
<input checked="" type="radio"/> No data	<input checked="" type="radio"/> No data	<input checked="" type="radio"/> No data

We could not find evidence of traditional knowledge about the different Hawaiian koa subpopulations. Therefore, all scenarios were scored with a “No data”.

Is there traditional knowledge suggesting distinctiveness between units?

Evidence through (multiple can apply): ⓘ

- Recorded differences based on traditional and local knowledge

2 ESUs	7 ESUs	4 ESUs
<input type="radio"/> Yes	<input type="radio"/> Yes	<input type="radio"/> Yes
<input type="radio"/> No	<input type="radio"/> No	<input type="radio"/> No
<input checked="" type="radio"/> No data	<input checked="" type="radio"/> No data	<input checked="" type="radio"/> No data

8. Enter your name and e-mail address (optional).

Notes

Name & Surname

Jane Doe

Email Address

jane.doe@xxxxx

Data protection disclaimer

By providing your name and email, you will receive the results in your inbox, as well as on the next page and may receive communications from us to participate further in the development of this framework. If you **choose not to enter your email**, your results will still be presented to you on the next page.

Submit Data

9. Click the “**Submit Data**” button to receive the results.

10. Interpreting the results.

As with the subpopulation survey results, each question and the answers provided are presented on the results page. A summary of the total scores for each scenario is presented below that.

Only scenario (3) (four ESUs) met the minimum thresholds of 10 points, providing strong support for four distinct ESUs. This score combines one genetic line of evidence (“Evolutionary distinctiveness”) with one recorded line of evidence (“Inherited characteristic variation”). Other lines of evidence were either not applicable or lacked data. Scenario (1) (two ESUs) and scenario (2) (seven ESUs) received zero points due to lack of supporting evidence.

Phase 2: Delineate Evolutionarily Significant Units (ESUs)

Please use the information provided by the email sent, to complete the following ESU survey.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Probable Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 10 points or higher = distinct ESU
- Below 10 points:
 - a) 10 points cannot be reached or exceeded (even if all missing data were supportive of the tested scenario).
=no support for the ESU – Scenario.
 - b) 10 points are currently not reached or exceeded, but could if missing data subsequently prove to be supportive of the tested scenario = scenario indicates possible ESUs (pESUs)*

*However, if a candidate scenario currently only scores in recorded biological evidence-types (so non-genetic), the scenario should be regarded as “data-deficient” rather than as a pESU scenario.

Lines of evidence

(No Title)

Thank you , for participating in the CGSG Subpopulation and ESU survey.

The species you tested is the **Acacia koa**

Your Survey Results for Phase 2:

1. Is there inherited variation in chromosome numbers, ploidy level or chromosome structure? (6 points)

2 ESUs No	7 ESUs No	4 ESUs No	
--------------	--------------	--------------	--

2. Is there evidence of long-term reproductive isolation? (6 points)

2 ESUs No data	7 ESUs No	4 ESUs No data	
-------------------	--------------	-------------------	--

3. Is there evidence of local adaptation or adaptive divergence? (6 points)

2 ESUs No	7 ESUs No	4 ESUs Yes	
--------------	--------------	---------------	--

4. Is there evidence of inherited characteristic differences? (4 points)

2 ESUs No	7 ESUs No	4 ESUs Yes	
--------------	--------------	---------------	--

5. Is there evidence supporting transmitted characteristic differences? (2 points)

2 ESUs No data	7 ESUs No data	4 ESUs No data	
-------------------	-------------------	-------------------	--

6. Is there traditional knowledge suggesting distinctiveness between units? (2 points)

2 ESUs No data	7 ESUs No data	4 ESUs No data	
-------------------	-------------------	-------------------	--

Total score for the scenario: **2 ESUs** is 0
Total score for the scenario: **7 ESUs** is 0
Total score for the scenario: **4 ESUs** is 10
Total score for the scenario: is 0

Scenario scoring:

- Scenarios scoring 10 points or higher = distinct ESUs
- Scenarios scoring below 10 points:
 - without potential to reach/exceed 10 points = no support for tested scenario
 - with potential to reach/exceed 10 points = possible ESU (pESU) status

You'll be receiving the results in your inbox soon!

You've reached the end of the survey. We trust that the information will provide insight into the management of your species.

Case Study 2 – The black wildebeest (*Connochaetes gnou*)

The black wildebeest is endemic to southern Africa and has an historical range spanning South Africa, Eswatini and Lesotho⁸. While overhunting caused population declines, recent re-introduction efforts in Eswatini and Lesotho have halted further losses and, in some cases, documented growth.^{9,10} The species inhabits diverse habitats such as grasslands, open plains and karoo shrublands¹¹.

1. Fill in species and scenario information.

In this case, we tested whether the species exists as a single metapopulation across its range, so we selected the Phase 1B survey. Mitochondrial DNA¹² and microsatellite markers¹³ exists to test this scenario. We expect no evidence for genetic structure or subpopulation fragmentation.

Phase 1B: Identify a single metapopulation

This survey is best views on Desktop, or rotate your phone for a better view.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Possible Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 2 points or higher = one continuous population

Which species are you testing?


Connochaetes gnou

1 metapopulation

Scenario 1 name

Genetic evidence

Is there no genetic structure?

Evidence through (multiple can apply): 

- No clear population cluster
- Low pairwise Fst Values
- No private Alleles

Caution: If there is knowledge of human-mediated/controlled genetic structure or extreme bottleneck events, genetic clustering detected in a species does not automatically indicate the presences of separate subpopulations.

⁸ Vrahimis, S., Grobler, J. P., Brink, J. S., Viljoen, P., & Schulze, E. (2016). *Black wildebeest. Red List of South African Species. South African Biodiversity Institute.*

⁹ Lundrigan, B., & Bidlingmeyer, J. (2000). *Connochaetes gnou* (black wildebeest). Animal Diversity Web.

¹⁰ Vrahimis, S., Grobler, J. P., Brink, J. S., Viljoen, P., & Schulze, E. (2017). *Connochaetes gnou*. The IUCN Red List of Threatened Species.

¹¹ Skinner, J. D., Chimimba, C. T. (2005). *The mammals of the southern African subregion*. Cambridge University Press, pp. 642-645.

¹² Corbet, S. W., & Robinson, T. J. (1991). Genetic divergence in South African Wildebeest: Comparative cytogenetics and analysis of mitochondrial DNA. *The Journal of Heredity*, 82(6), 447–452.

¹³ Grobler, P., van Wyk, A. M., Dalton, D. L., van Vuuren, B. J., & Kotzé, A. (2018). Assessing introgressive hybridization between blue wildebeest (*Connochaetes taurinus*) and black wildebeest (*Connochaetes gnou*) from South Africa. *Conservation Genetics*, 19(4), 981–993.

2. Answer the questions for each line of evidence.

a. Genetic evidence

The mitochondrial DNA haplotypes¹² and microsatellite markers¹³ showed no evidence of genetic clustering.

Genetic evidence

Is there no genetic structure?

Evidence through (multiple can apply): 

- No clear population cluster
- Low pairwise Fst Values
- No private Alleles

Caution: If there is knowledge of human-mediated/controlled genetic structure or extreme bottleneck events, genetic clustering detected in a species does not automatically indicate the presences of separate subpopulations.

1 metapopulation


- ☒ Yes
☐ No
☐ No data

b. Recorded biological evidence

Individuals are continuously distributed across central South Africa, extending into Eswatini and Lesotho, with historical migration among these regions⁸. Consequently, no direct subpopulation fragmentation was detected.

Recorded biological evidence

Are there records of continuous occupation of the range with frequent migration between all regions?

Evidence through (multiple can apply): 

- Occupation of the **same** biogeographical zones
- Occupation of remnants of historical habitats **with frequent migration**


- ☒ Yes
☐ No
☐ No data

c. Inferred evidence

No evidence was found indicating subpopulation fragmentation; therefore, the tested scenario is scored as “No data”.

Inferred evidence

Is there inferred evidence of no subpopulation fragmentation?

Evidence through (multiple can apply): 

- **No** inferred Population disjunction
- Phylogeographic and/or biogeographic evidence from co-occurring species hint towards **no** subpopulation fragmentation
- Dispersal distance and buffer according to the CBD indicators indicate **no** differentiation

☐ Yes

☐ No

☒ No data

3. Enter your name and e-mail address (optional).

Notes

Name & Surname

Jane Doe

Email Address

jane.doe@xxxxx

Data protection disclaimer

By providing your name and email, you will receive the results in your inbox, as well as on the next page and may receive communications from us to participate further in the development of this framework. If you **choose not to enter your email**, your results will still be presented to you on the next page.

Submit Data

4. Click the “Submit Data” button to receive the results.

5. Interpreting the results.

The scenario testing whether the black wildebeest comprises a single metapopulation scored 4 points, exceeding the 2-point threshold. Thus, the black wildebeest can be considered a single metapopulation and one Evolutionarily Significant Unit (ESU).

Phase 1B: Identify a single metapopulation

This survey is best views on Desktop, or rotate your phone for a better view.

Please use the dropdown menu below for additional information.

- Subpopulations
- Evolutionarily Significant Units (ESU)
- Possible Evolutionarily Significant Unit (pESU)
- Gene Flow
- Potential IUCN Species assessment
- References

Scoring:

- 2 points or higher = one continuous population

Lines of evidence

Thank you for participating in the CGSG Subpopulation and ESU survey.

The species you tested is the *Connochaetes gnou*

Your Survey Results for Phase 1B:

1. Is there no genetic structure? (2 points)

1 metapopulation
Yes

2. Are there records of continuous occupation of the range with frequent migration between all regions? (2 points)

1 metapopulation
Yes

3. Is there inferred evidence of no subpopulation fragmentation? (1 point)

1 metapopulation
No data

Total score for the scenario: 1 metapopulation is 4

If the metapopulation scenario scored 2 points or higher, the Null-hypothesis is accepted. Therefore the *Connochaetes gnou* can be considered as one single metapopulation.

You'll be receiving the results in your inbox soon!

FAQs

Understanding the framework

What's the difference between a subpopulation and an ESU?

Subpopulations as used here most closely relate to the IUCN's classification, whereby population refers to the species as a whole, and subpopulations represent demographic groupings (often with limited gene flow), which others may refer to as populations.

ESUs (or Evolutionarily Significant Units) on the other hand are evolutionary lineages that typically reflect deeper genetic and ecological divergence. Should we state here which definition we used? There are multiple definitions so would be good to be clear here.

Why is there the option to test a single metapopulation or ESU?

While users can infer the rejection of all multi-group scenarios to mean the lack of structure and hence a single metapopulation or ESU for the species as a whole, others may wish to systematically assess this. This can be particularly useful when testing the validity of an existing designation (e.g., a recognised subspecies or management unit).

Since the logic of the questions differs when testing for a single grouping (often reversing the framing of comparison questions), a separate survey path is provided to ensure clarity and accuracy in scoring.

How does this framework align with the current IUCN process and international agreements such as the Kunming-Montreal Global Biodiversity Framework?

This framework complements and strengthens existing IUCN processes by providing a more standardised and biologically grounded method for identifying intraspecific units—such as subpopulations and Evolutionarily Significant Units (ESUs)—that may warrant separate consideration in Red List or Green Status assessments. While the IUCN permits assessments below the species level (e.g., subspecies and subpopulations), the criteria for defining these units are often inconsistently applied. This framework addresses that gap by offering clear, evidence-based guidance to support more consistent and defensible decisions.

In the broader global context, the framework is also closely aligned with international policy targets, such as Target 4 of the Kunming-Montreal Global Biodiversity Framework (GBF), which emphasises the importance of preventing extinction, halting population declines, and maintaining genetic diversity within species. It supports this target by helping users recognise and conserve genetically meaningful units (subpopulations correlates to populations in GBF context), which is essential for long-term species resilience and adaptive potential.

By improving how we recognise and assess intraspecific diversity, the framework contributes directly to the implementation of the IUCN Red List Guidelines and the goals of the GBF, particularly around conservation planning, recovery prioritization, and tracking progress on the state of biodiversity at national and global levels.

Using the framework and webtool

How do I know which scenarios to test?

Start by identifying plausible ways that the species might be divided in subpopulations or ESUs, based on any existing information. Scenarios can be drawn from:

- Published scientific literature
- Expert knowledge or field observations
- Known geographic or ecological barriers
- Morphological or behavioural differences
- Suspected areas of genetic divergence
- Need to verify/validate subspecific designations

You do not need perfect evidence to define a scenario. This framework is designed to help *test* these possibilities. Try to focus on the most likely or relevant groupings, especially those that might face different threats, show signs of isolation, or are important for management decisions (e.g., translocation units).

You can test up to **four scenarios at a time**, and more can be assessed by completing additional surveys. Each scenario represents a hypothesis about how population structure may be organised.

What should I do with the results?

This framework was developed to help researchers, practitioners and government officials identify and assess genetically meaningful units to inform conservation management and planning. While its application may be broad, we feel it is especially relevant to IUCN assessments (i.e., Red List, Green Status). These assessments are primarily conducted at the species level; however, subspecies and subpopulation assessments are also permitted, but their use is limited and inconsistent.

Subspecies designations are often questioned due to modern genetic evidence revealing little or no support for traditional classifications. Similarly, only around 5% of IUCN assessments include information on subpopulation number or size, and there is considerable variation in how subpopulations are defined and delimited.

This framework was therefore created to help standardise the classification of intraspecific units in a way that is more biologically and evolutionarily meaningful. The results of the survey can guide the

appropriate level at which assessments should be conducted—particularly when different subpopulations or Evolutionarily Significant Units (ESUs) face different threats or extinction risks. More broadly, however, the results can strengthen species conservation planning by providing a clearer, evidence-based understanding of population structure.

For scenarios showing data deficiency (i.e., subpopulation scenarios scoring less than 2 or ESU scenario below 10), where population structure is suspected but not well supported, the results can serve as a basis for considering follow-up research (e.g., collecting genetic data).

In all cases, it is highly recommended that users retain all scenario scores and notes. This promotes transparency, facilitates the tracking of decision-making, and allows for reassessments as new data become available.

What if I'm unsure how to score a question?

If you're unsure how to answer a question, use the information bubbles (hover over the “i” icon) for guidance on what kind of evidence is relevant. If uncertainty remains, it's better to select “uncertain” or leave the score blank rather than guessing, as inaccurate scoring can affect the framework's overall result. You may also want to make a note explaining the uncertainty to support transparency and future reassessment.

What if I have evidence based on different genetic markers?

Molecular data has a multitude of forms, ranging from mitochondrial or candidate gene haplotypes, to microsatellite markers, and Single Nucleotide Polymorphisms (SNPs, which themselves can range from hundreds to millions in count). Each marker type provides information on the presence of a subpopulation or an ESU in a species but with different resolution. Higher resolution genomic tools are always likely to find more structure because there are more data available. This however does not always translate into biologically meaningful subpopulation divisions within species. Study design is also key, and can lead to various outcomes/conclusion, when e.g., comparing genomic markers with mitochondrial haplotypes or microsatellite markers, or comparing a range-wide study to a study only using very few sites.

If different genetic markers point to different evidence you may want to assess these evidences in different candidate scenarios.

Can I use this framework for species without genetic data?

Yes. The framework is designed to accommodate a wide range of evidence types, including ecological, morphological, behavioural, and geographic data. While genetic data generally carry the highest weight in determining population structure due to the ability to directly reveal patterns of gene flow,

divergence, and reproductive isolation, the tool can still be meaningfully applied where such data are lacking.

In such cases, users are encouraged to use other relevant indicators of structure, such as:

- Known barriers to dispersal (e.g., rivers, mountains)
- Habitat discontinuities or fragmentation
- Local adaptation or environmental differences
- Morphological variation or behavioural distinctions
- Expert opinion or unpublished field observations

The scoring framework reflects the relative strength of different data types, with genetic evidence typically contributing most strongly to higher scores. However, moderate or even low scores derived from non-genetic data can still be valuable for identifying candidate units and prioritising further investigation. The framework is especially useful for highlighting areas of uncertainty and guiding decisions on where additional data collection (including genetics) may be warranted.

Are there risks of oversplitting or undersplitting species in this framework?

Yes. As with any method used to assess population structure, there is a potential risk of either oversplitting (dividing a species into more units than is biologically justified) or undersplitting (failing to recognise meaningful structure). This framework aims to reduce those risks by offering a transparent, repeatable, and evidence-based approach.

The structured questions and weighted scoring system help users assess the strength of evidence for population or evolutionary distinctiveness. Genetic evidence carries the highest weight because it provides direct insights into gene flow and divergence, but other data types are also considered. By integrating multiple lines of evidence, the framework promotes balanced decision-making.

To further minimise misclassification:

- Users are encouraged to test only plausible scenarios grounded in available data.
- The scoring system accommodates uncertainty (e.g., “uncertain” responses don’t contribute to the total).
- The framework does not prescribe taxonomic action-it is a decision-support tool and does not replace formal taxonomic classification or phylogenetic studies.

In short, while some subjectivity is unavoidable, the framework helps make assumptions and limitations explicit, reducing arbitrary or inconsistent decisions and supporting more defensible classifications.

Survey functionality

Can I go back and edit a scenario after submitting the survey?

Unfortunately, no. Survey responses are not stored on a database and therefore they cannot be retrieved or edited once submitted. However, if you provide your email address before submission, a copy of the questions, answers and scoring results will be sent to you. This allows you to review the tested scenarios later, consider exploring additional scenarios, or update the assessment if new information becomes available.

Can I save my progress and return later?

Unfortunately, no. Surveys cannot be saved and must be completed in a single session.

What format will the results be in?

The results are sent as plain text in an email, summarising the scores for each tested scenario along with any notes you provided. Please note that results are not sent as PDF or Word (.doc) files.

Can I compare results across surveys?

Yes. Because each scenario receives a discrete score, results for the same species can be meaningfully compared across surveys. Each survey allows testing up to four scenarios, so completing additional surveys enables broader comparisons. While the tool does not automatically aggregate or analyse results, users can manually review and compare saved email summaries.

Can multiple users collaborate on a survey?

Yes, but only if working together in person. The survey can only be completed and submitted from a single computer, as progress cannot be saved or shared across devices. Collaborative input is encouraged, but responses must be entered and submitted for one session.

Are my results or email address stored?

No. All content entered in the survey is not stored or retained by the developers. The email address option provided at the end of each survey is simply available should the user want a saved copy of the survey questions, answers and results (scores) to refer to in future.

Contact Support

Should you have any questions regarding the functionality, use and interpretation of the survey, please do not hesitate to contact ConGen Africa at info@congenafrika.com.