

Country Code: _____

Student Code: _____

The 21st INTERNATIONAL BIOLOGY OLYMPIAD

11th – 18th July, 2010

Changwon, KOREA



PRACTICAL TEST 1

PLANT AND ANIMAL SYSTEMATICS

Total Points: 50

Duration: 90 minutes

Dear Participants,

- ☺ In this test, you have been given the following 3 tasks:

Task I: Reconstruct the phylogenetic tree of six plant species using parsimony method

(25 points)

Task II: Reconstruct the phylogenetic tree of six insect species using the UPGMA method

(18 points)

Task III: Co-evolution between plants and insects (7 points)

- ☺ Write down your results and answers in the **Answer Sheet. Answers written in the Question Paper will not be evaluated.**
- ☺ Please make sure that you have received all the materials listed for each task. If any of the listed items is missing, please raise your hand.
- ☺ Stop answering and put down your pencil **immediately** after the end bell rings. The supervisor will collect the Question Paper and the Answer Sheet.

Good Luck!!

PLANT AND ANIMAL SYSTEMATICS

This practical test is composed of 3 tasks.

**TASK I. (25 points) Reconstruct the phylogenetic tree of six plant species
using parsimony method**

This task is composed of 4 parts.

Plant taxa (species)

A, B, C, D, E, and F.

Plant materials

Each set consists of the following materials:

- 1) The flowers of six species preserved in 70% ethanol (Flowers A-F).
- 2) The fruits of six species preserved in 70% ethanol (Fruits A-F).
- 3) Six dried flowering specimens (Flowering specimens A-F).
- 4) Six dried fruiting specimens (Fruiting specimens A-F).
- 5) Three prepared pollen slides (Each slide contains the pollen grains of two species, labeled A-B, C-D and E-F.).

Instruments

Stereomicroscope (20X), microscope (400X), razor blade, dissecting forceps, dissecting needles (2), petri-dishes (2), 20-cm ruler.

Part I-1. (9 points) Using the suggested materials and methods in Table 1, observe the following 10 characters. Enter each character state in the Data Matrix 1. Each character state should be recorded as the appropriate number (0, 1, or 2) based on the following descriptions. Figure 1 is provided as a reference of the descriptive terminology.

Table 1. Character descriptions for plants (See Figure 1 for illustration of the character states for the character 1, 2, 4, 7, 8, 9 and 10.)

| No | Character | Character state | Materials | Methods |
|-----------|----------------------------------|--|-------------------------------|--|
| 1. | Flower petal | 0: Sympetalous 1: Polypetalous | Flowers | Naked eye |
| 2. | Inflorescence | 0: Corymb or umbel 1: Raceme 2: Axillary or terminal | Flowering specimens | Naked eye |
| 3. | Fruit stalk | 0: Longer than 1 cm 1: Shorter than 1 cm | Fruiting specimens | Naked eye |
| 4. | Ovary position | 0: Superior 1: Half-inferior 2: Inferior | Flowers | Dissect with razor blade, Stereomicroscope (20X) |
| 5. | Habit | 0: Shrub 1: Tree | Given answer | Given answer |
| 6. | Trichomes on the fruit surface | 0: Hairless 1: Densely haired | Fruiting specimens | Naked eye |
| 7. | Fruit shape and size at maturity | 0: Cylindrical or circular with emarginated tip (Dia.<1 cm) 1: Circular with pointed tip (Dia.< 1 cm) 2: Circular-elliptic with pointed tip (Dia.≥ 1 cm) | Fruits and Fruiting specimens | Naked eye |
| 8. | Fruit types | 0: Pome or capsule 1: Drupe | Fruits | *Dissect with razor blade, Naked eye |
| **9 | Pollen grains | 0: Tetrad 1: Monad | Pollen slides | Microscope (400X) |
| 10. | Leaf margin at maturity | 0: Entire or undulate 1: Serrate or dentate | Fruiting specimens | Naked eye |

*If the endocarp is hard, carefully remove exocarp and mesocarp to identify the fruit type.

** Search pollen grains within a red circle of each specimen.

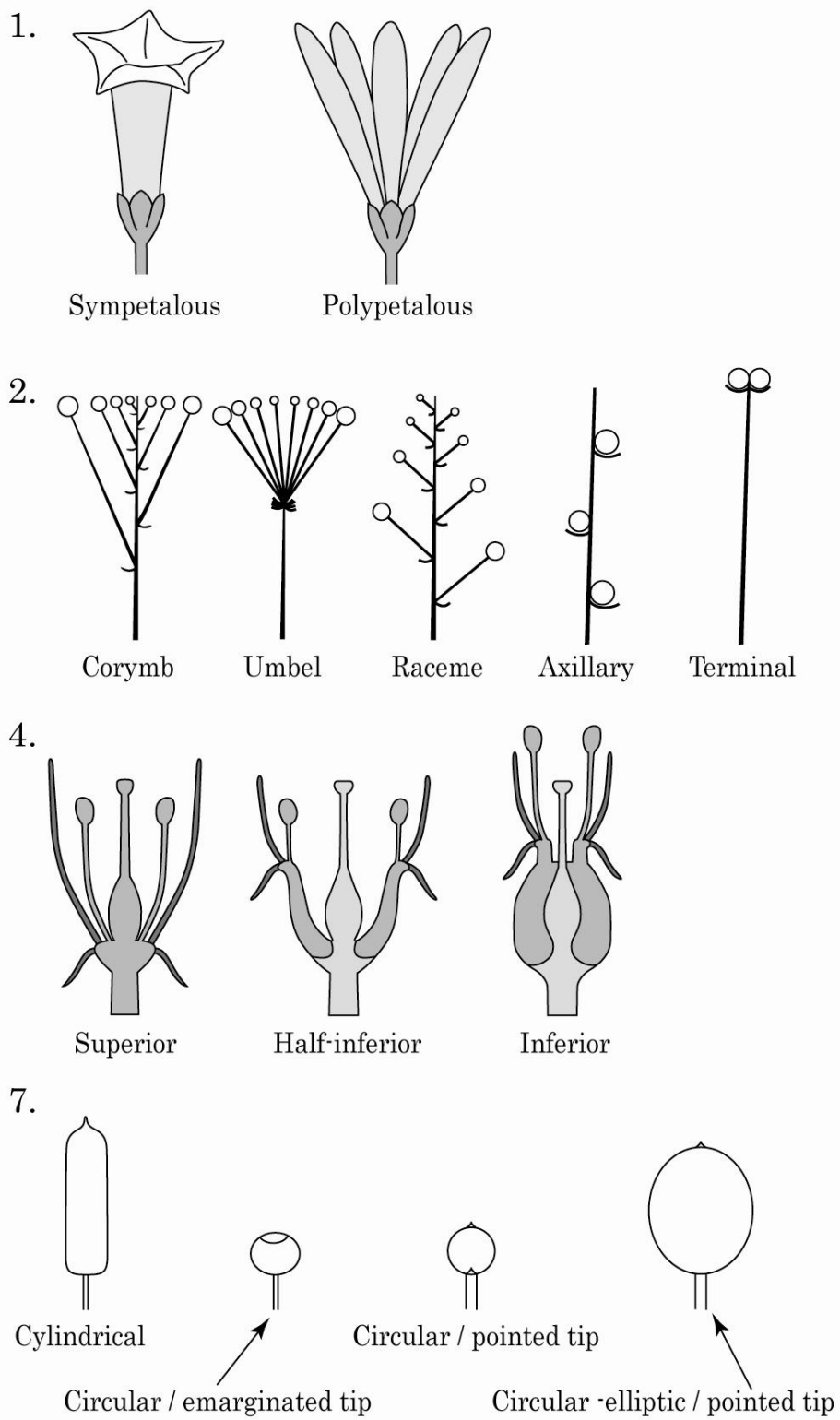


Figure 1. Illustration of character states for the character 1, 2, 4, 7, 8, 9 and 10.

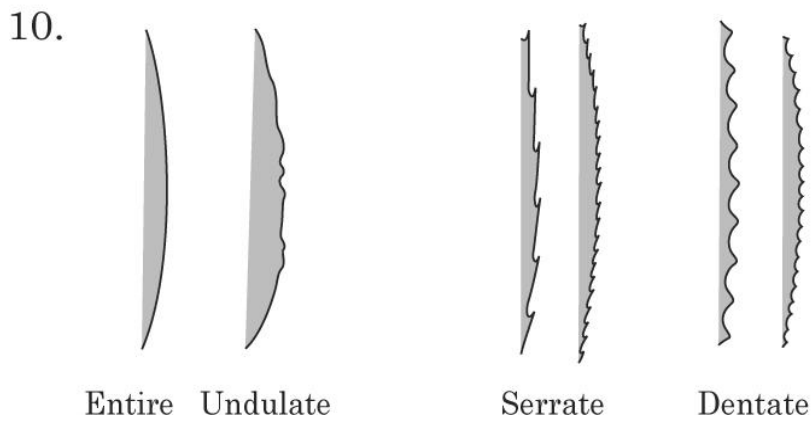
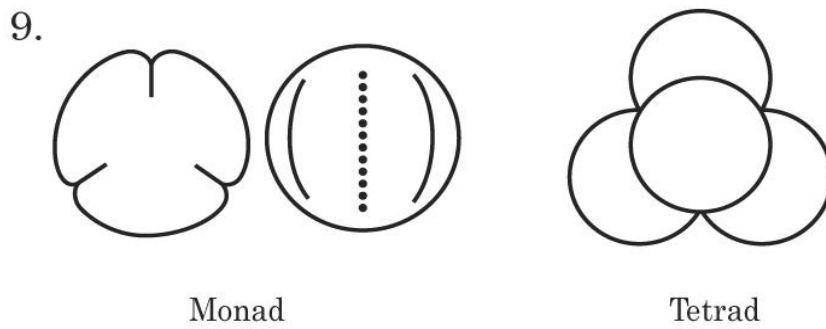
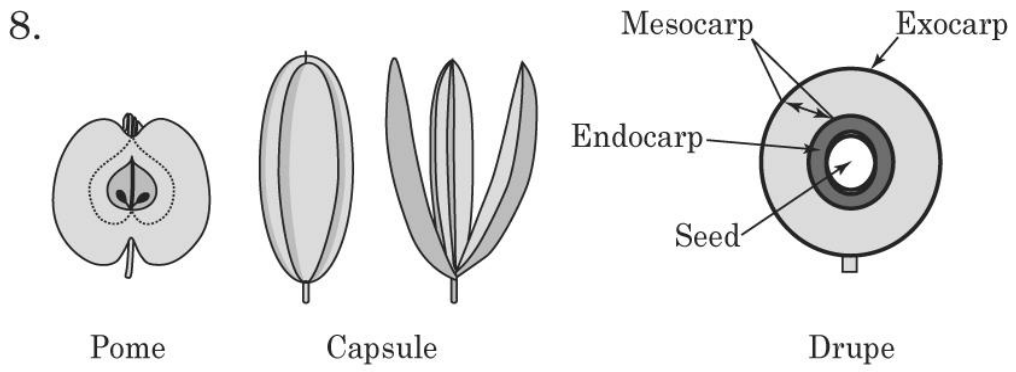


Figure 1. continued

Q1. (9 points) Fill the empty cells of Data Matrix 1 on your Answer Sheet. The sheet consists of 6 x 10 cells. Taxon F and character 5 are already filled for your reference.

| Character Taxa | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|-------------------|---|---|---|---|---|---|---|---|---|----|
| A | | | | | 1 | | | | | |
| B | | | | | 1 | | | | | |
| C | | | | | 0 | | | | | |
| D | | | | | 0 | | | | | |
| E | | | | | 1 | | | | | |
| F | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Part I-2. (4 points) Refer to your completed Data Matrix 1 to answer the following questions

Q2.1. (1 point) Indicate with checkmarks (✓) which of the characters are phylogenetically informative.

Q2.2. (1 point) Indicate with checkmarks (✓) which of the characters are polymorphic (more than two states).

Q2.3. (2 points = 1 × 2) Use the following numerical formulae to define the number of possible trees for a given number of taxa (n).

The number of possible unrooted trees = $(2n-5)!/2^{n-3}(n-3)!$

The number of possible rooted trees = $(2n-3)!/2^{n-2}(n-2)!$

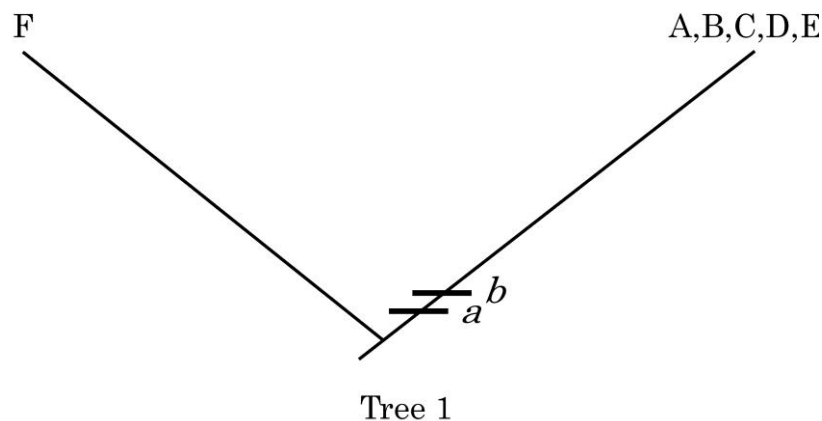
The symbol ‘!’ in the formulae indicate the factorial.

What is the numbers of rooted and unrooted trees in this case?

Part I-3. (8 points) Cladistic analysis can be used to construct a phylogenetic tree of this species group. The primitive character state (plesiomorphy) is hypothesized to be the same as the state found in the outgroup F. Therefore, the character state 0 represents the primitive state for all given characters. Any change in state from that primitive trait is considered to be a derived character state, representing an evolutionary event (apomorphy). The character states of 1 and 2 represent derived condition. In this analysis, all characters are given equal weight. Tree construction is done in a step-by-step process. Place the appropriate character numbers and group members on the tree at each step.

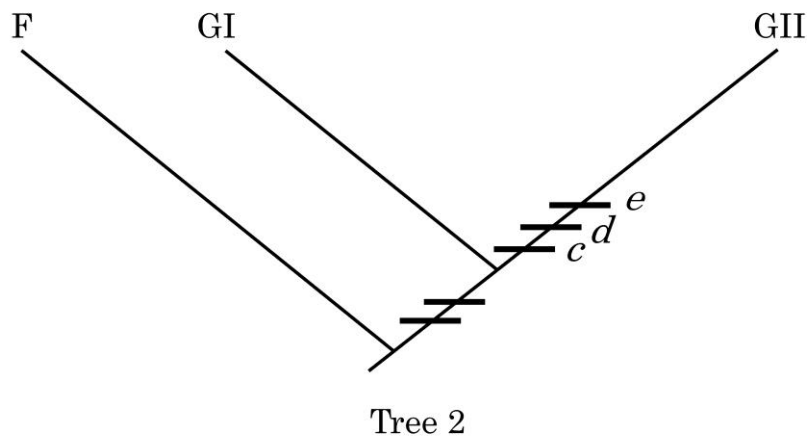
Q3.1. (1 point) The initial Tree 1 can be created if we separate the ingroup (A, B, C, D, E) from the outgroup (F) using the two characters that distinguish all members of the ingroup (shared derived traits) from the outgroup F.

Identify these two characters (*a* and *b*) shown in Tree 1 and write them in the Answer Sheet. Character state should be given in parenthesis if the character is a polymorphic one.



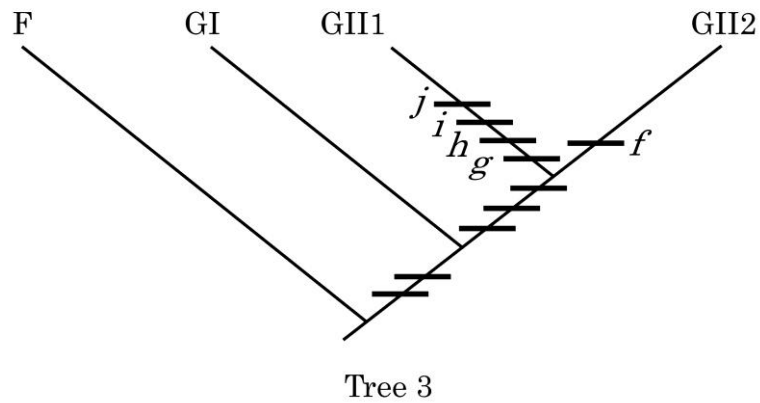
Q3.2. (2 points) The step-by-step method to create the final tree from this initial tree can be illustrated by the concept of membership. Analysis proceeds to progressively less inclusive groupings supported by other derived characters. A less inclusive group(s) can be separated from the more inclusive group by the supported character change(s) on the tree. In the second step, the five ingroup taxa can be further divided into two subgroups (GI and GII) based on three synapomorphic characters.

Identify the subdivided species group members of GI and GII and the three character numbers (*c*, *d* and *e*) shown on Tree 2 and write them in the Answer Sheet. Character state should be given in parenthesis if the character is a polymorphic one.



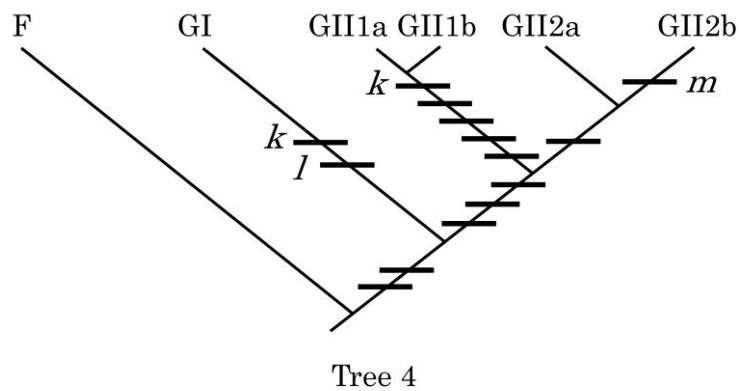
Q3.3. (3 points) The group II (GII) can be further divided into two less inclusive subgroups (GII1 and GII2) by four and one synapomorphic character(s), respectively.

Identify the members of GII1 and GII2 and write the corresponding character numbers (shown in locations *f-j* in Tree 3) in the Answer Sheet. Character state should be given in parenthesis if the character is a polymorphic one.



Q3.4. (2 points) At the final stage of tree construction, all autapomorphic (singly derived) characters should be located on the tree, and any conflicting characters should be adjusted using the parsimony principle. There are two autapomorphic characters (*l* and *m*) and only a single conflicting character (*k*) in this case.

List the taxon name for each of the five ingroup species on the fully resolved Tree 4, and give the character numbers that correspond to *k*, *l* and *m*, respectively, in the Answer Sheet. Character state should be given in parenthesis if the character is a polymorphic one.



Part I-4. (4 points) Use the complete phylogenetic tree to answer the following questions.

Q4.1. (1 point) What is the number of character changes (steps) on the maximum parsimonious tree?

Q4.2. (1 point) The consistency index (CI) is defined as the minimum number of character state changes required in an absolutely consistent tree (all character states changed only once) divided by the observed number of character state changes in the final tree.

What is the CI of the final Tree 4?

Q4.3. (1 point) What is the maximum number of genera that can be recognized from the final tree if taxa C and D are congeneric species?

Q4.4. (1 point) How many monophyletic groups can be recognized from the final tree?

TASK II. (18 points) Reconstruct the phylogenetic tree of six insect species using the Unweighted Pair Group Method with Arithmetic mean (UPGMA) method

This task is composed of 3 parts.

Insect taxa: Six beetles (Coleoptera)

T1, T2, T3, T4, T5 and T6

Insect materials

Pinned and dried specimens of six beetles, labeled T1~T6.

Experiment tools

Insect stage, ruler, stereomicroscope (20X)

Note: Please be careful. Do not break the legs or antennae of the beetle specimens. There will be three point deduction penalty if you break the parts of any specimen.

Most insect specimens and their parts can be observed directly from the plastic case after removing the cover.

Part II-1. (8 points) The character states are defined in Table 2. Carefully observe the morphological characters of the beetle specimens using the naked eye and the stereomicroscope. Then, complete the Data Matrix 2. Figure 2 is provided as a reference for the Coleoptera body parts.

Table 2. Character descriptions for Coleoptera

| Character | Character state | Methods |
|---|--|--|
| 1. Longitudinal discontinuous ridges on elytra | 0: Present 1: Absent | Stereomicroscope |
| 2. Horns on head and pronotum | 0: Absent 1: Present | Naked eye |
| 3. Compound eye | 0: Does not surround antennal socket 1: Surrounds about half of antennal socket | Stereomicroscope |
| 4. Mandible length | 0: Shorter than prothorax length 1: Longer than prothorax length | Naked eye, stereomicroscope if the part is small |
| 5. Antennae length | 0: Shorter than body length 1: Longer than body length | Naked eye |
| 6. Antennae shape | 0: Filiform or serrate 1: Distal segments clubbed or lamellated | Naked eye, stereomicroscope if the part is small |
| 7. Antennae | 0: Not elbowed 1: Elbowed | Naked eye |
| 8. Hind tarsi | 0: 5 segments 1: 4 segments or less | Stereomicroscope |
| *9. Notopleural sutures of the prothorax | 0: Fused 1: Not fused | Given answer |
| *10. 1st sternum and hind coxa | 0: Separated 1: Not separated | Given answer |
| *11. Food preference | 0: Zoophagy or saprophagy 1: Phytophagy | Given answer |

* Character states are provided in the Answer Sheet.

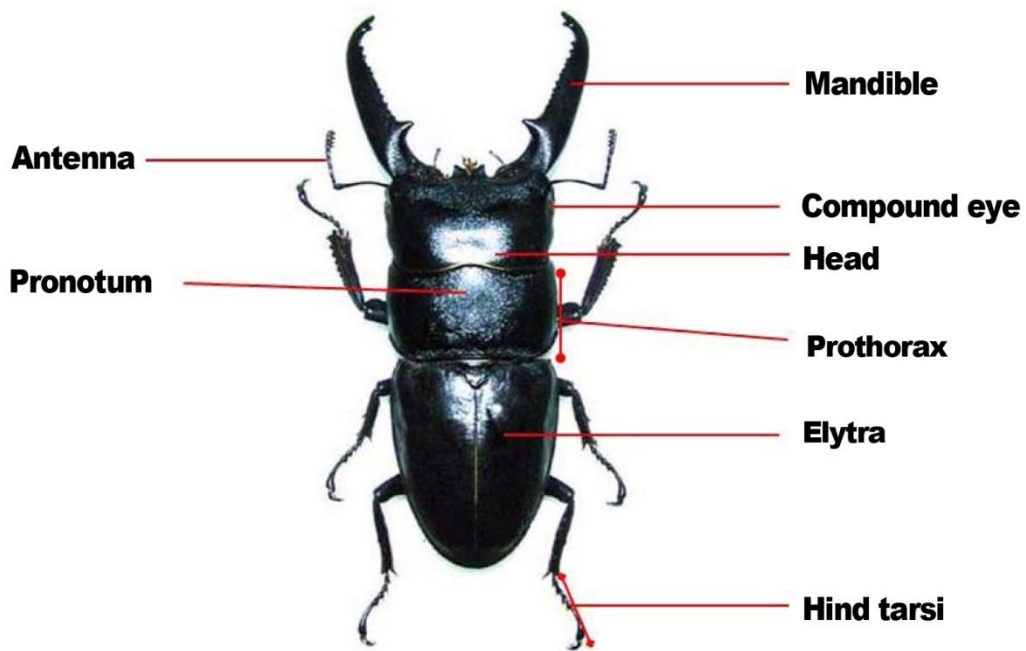


Figure 2. The body parts of Coleoptera to be observed.

Q5. (8 points) Complete the Data Matrix 2 in your answer sheet. The sheet consists of 6 x 11 cells. Characters 9, 10, and 11 are already filled in.

Data matrix 2.

| Character \ Taxa | C1 | C2 | C3 | C4 | C5 | C6 | C7 | C8 | C9 | C10 | C11 |
|------------------|----|----|----|----|----|----|----|----|----|-----|-----|
| T1 | | | | | | | | | 0 | 0 | 0 |
| T2 | | | | | | | | | 1 | 1 | 0 |
| T3 | | | | | | | | | 1 | 1 | 1 |
| T4 | | | | | | | | | 1 | 1 | 1 |
| T5 | | | | | | | | | 1 | 1 | 1 |
| T6 | | | | | | | | | 1 | 1 | 1 |

Part II-2. (3 points) Create a character difference matrix between all possible pair-wise taxa from the completed Data Matrix 2. The difference value (D_{ij}) between taxon i and j is the sum of the character (C) numbers for which $C_i \neq C_j$. Calculate the difference values, and fill the table below (Difference Matrix 1). The values of three pair-wise comparisons (T1/T2, T3/T4, and T5/T6) are already provided as examples.

Q6. (3 points) Complete the following Difference Matrix 1 on your answer sheet.

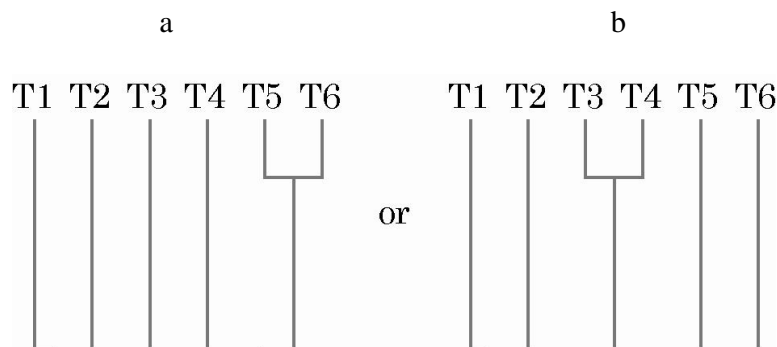
Difference Matrix 1. The pair-wise difference matrix calculation.

| D_{ij} | T1 | T2 | T3 | T4 | T5 | T6 |
|----------|----|----|----|----|----|----|
| T1 | - | - | - | - | - | - |
| T2 | 4 | - | - | - | - | - |
| T3 | | | - | - | - | - |
| T4 | | | 2 | - | - | - |
| T5 | | | | | - | - |
| T6 | | | | | 2 | - |

Part II-3. (7 points) Construct a phylogenetic tree based on the UPGMA algorithm using the pair-wise difference matrix that you created in Part 2. During the procedure, you will create nested clusters of taxa (smaller clusters into larger clusters) using successive difference matrices and phenograms until you construct a single cluster. The order for clustering is: 1) Pick the smallest entry D_{ij} . 2) Join those two species into a cluster, 3) Compute new distances from that cluster to the other taxon k , using the Unweighted Pair Group Method with Arithmetic mean (UPGMA). A new distance between a new species k and a cluster (i and j) is defined as $D(k(ij)) = (1/2)((D(ki)+D(kj)))$. Repeat the clustering process 1) ~ 3) to create the next cluster. This process should be continued to construct a single cluster for the entire group.

Q7.1. (1 point) Two alternate initial trees (a and b) can be constructed from the Difference Matrix 1 as shown below. Combine the two alternative trees and draw as a single tree (Tree 1). Draw Tree 1 on your Answer Sheet.

Initial Trees a and b: (T1, T2, T3, T4, (T5, T6)) or (T1, T2, (T3, T4), T5, T6)



Q7.2. (2 points) Complete the Difference Matrix 2. Calculate new difference values

between cluster and taxon (or between cluster and cluster) using UPGMA algorithm and fill the cells in the answer sheet. Find the taxon pair(s) that shows the lowest difference values and make a newly clustered tree (Tree 2). Draw the tree in the answer sheet.

Difference Matrix 2:

| <i>D_{ij}</i> or <i>D_{k(ij)}</i> | T1 | T2 | T(3,4) | T(5,6) |
|---|----|----|--------|--------|
| T1 | - | - | - | - |
| T2 | 4 | - | - | - |
| T(3,4) | | | - | - |
| T(5,6) | | | | - |

Q7.3. (2 points) Complete the Difference Matrix 3. Again calculate the new difference

values between cluster and taxon (or between cluster and cluster) using UPGMA algorithm and fill the cells in the answer sheet. Find the taxon pair that shows the lowest difference values and make a newly clustered tree (Tree 3). Draw the tree in the answer sheet.

Difference Matrix 3:

| <i>D_{ij}</i> or <i>D_{k(ij)}</i> | | | |
|---|---|---|---|
| | - | - | - |
| | | - | - |
| | | | - |

Q7.4. (2 points) Complete the Difference Matrix 4. Calculate the new difference values between cluster and cluster using UPGMA algorithm and fill the cells in the answer sheet. Make a complete clustered tree (Tree 4) and draw it in the answer sheet.

Difference Matrix 4:

| | | |
|---|---|---|
| <i>D_{ij}</i> or <i>D_{k(ij)}</i> | | |
| | - | - |
| | | - |

TASK III. (7 points) Co-evolution between plants and insects

Plant-herbivore relationships are one of the core explanations for the rapid diversification of insects and flowering plants. For this task, use the final plant phylogeny (Task 1) and beetle phylogeny (Task 2). Under the assumption that the larvae of each insect species feed on a single plant species, compare the insect and plant phylogenies and answer the following questions.

- Q8.** (3 points) If the insect species T2, T3, and T5 feed on the plant species E, D, and A, respectively, what kinds of plant species are fed upon by insect species T1, T4, and T6, respectively?
- Q9.** (2 points) Which plant and insect species pair shows different phylogenetic positions in the trees?
- Q10.** (2 points) Which are the two best possible reasons to explain the differences between the insect and plant phylogenies? (Select the **two** best answers).
- A. Host shift of insect species
 - B. Adaptive radiation of plant species
 - C. Genetic bottleneck during the insect species evolution
 - D. Different tree reconstruction methods
 - E. Genetic drift of plant species

PRACTICAL TEST 1

Answer Key

PLANT AND ANIMAL SYSTEMATICS

Total Points: 50

Duration: 90 minutes

TASK I. (25 points)

Part I-1. (9 points)

Q1. (9 points)

Data Matrix 1. (One point per character)

| Character Taxa | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|-------------------|---|---|---|---|---|---|---|---|---|----|
| A | 1 | 2 | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 |
| B | 1 | 2 | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 |
| C | 1 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 |
| D | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 |
| E | 1 | 0 | 0 | 2 | 1 | 0 | 0 | 0 | 1 | 0 |
| F | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Part I-2. (4 points) Place "√" symbol on the right characters.

Q2.1. (1 point)

| Character Number | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---------------------------------|---|---|---|---|---|---|---|---|---|----|
| Phylogenetically informative | | √ | √ | √ | √ | √ | √ | √ | | √ |

Q2.2. (1 point)

| | | | | | | | | | | |
|------------------|---|---|---|---|---|---|---|---|---|----|
| Character Number | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
| Polymorphic | | √ | | √ | | | √ | | | |

Q2.3. (2 points = 1 x 2)

| |
|---|
| The number of possible unrooted trees: <u>105</u> |
| The number of possible rooted trees: <u>945</u> |

Part I-3. (8 points) Character state should be given in the parenthesis if the character is polymorphic one.

Q3.1. (1 point)

| | |
|---------------------------------|-------------------|
| | Character numbers |
| Character <i>a</i> and <i>b</i> | 1, 9 |

Q3.2. (2 points). Character number and taxon name will be assign one point, respectively.

| | | | |
|---|-------------------|-----|---------------|
| | Character numbers | | Taxon name(s) |
| Character <i>c</i> , <i>d</i> and <i>e</i> | 4(1), 8, 10 | GI | E |
| | | GII | A, B, C, D |

Q3.3. (3 points)

| | Character number(s) | | Taxon name(s) |
|---------------------------------------|---------------------|------|---------------|
| Character <i>f</i> | 7(1) | GII1 | A, B |
| Character <i>g, h, i</i> and <i>j</i> | 2(2), 3, 6, 7(2) | GII2 | C, D |

Q3.4. (2 points)

| | Character number | | Taxon name |
|--------------------|------------------|-------|------------|
| Character <i>k</i> | 5 | GI | E |
| Character <i>l</i> | 4(2) | GII1a | A |
| Character <i>m</i> | 2(1) | GII1b | B |
| | | GII2a | C |
| | | GII2b | D |

Part I-4. (4 points)

Q4.1. (1 point)

14 steps

Q4.2. (1 point)

CI = 13/14

Q4.3. (1 point)

4

Q4.4. (1 point)

5

Task II. (18 points)

Part II-1. (8 points)

Q5. (8 points)

Data Matrix 1

| Character Taxa \ | C1 | C2 | C3 | C4 | C5 | C6 | C7 | C8 | C9 | C10 | C11 |
|---------------------|----|----|----|----|----|----|----|----|----------|----------|----------|
| T1 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| T2 | 1 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 1 | 0 |
| T3 | 1 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 1 | 1 | 1 |
| T4 | 1 | 1 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 1 | 1 |
| T5 | 1 | 0 | 1 | 0 | 1 | 0 | 0 | 1 | 1 | 1 | 1 |
| T6 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 |

Part II-2. (3 points)

Q6. (3 points)

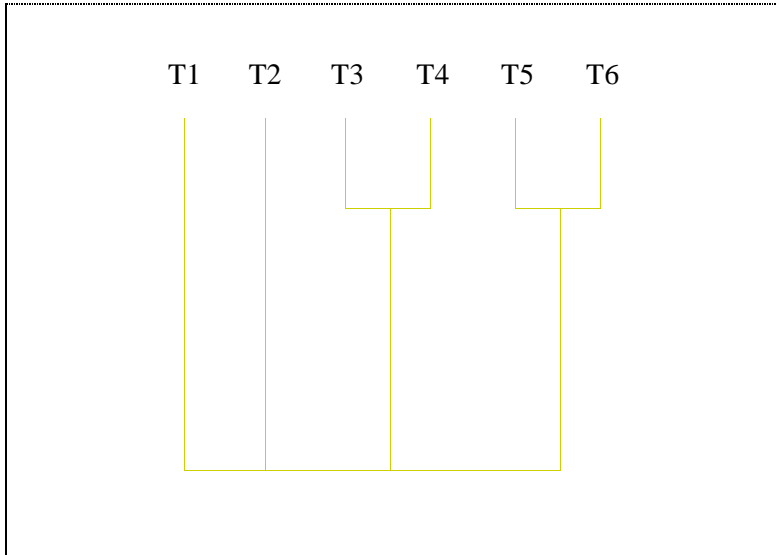
Difference Matrix 1

| Dij | T1 | T2 | T3 | T4 | T5 | T6 |
|-----|----------|----|----------|----|----------|----|
| T1 | - | - | - | - | - | - |
| T2 | 4 | - | - | - | - | - |
| T3 | 7 | 3 | - | - | - | - |
| T4 | 7 | 3 | 2 | - | - | - |
| T5 | 7 | 5 | 6 | 6 | - | - |
| T6 | 5 | 3 | 4 | 4 | 2 | - |

Part II-3 (7 points)

Q7.1. (1 point)

Tree 1: Combine and draw the two alternative trees as a single tree

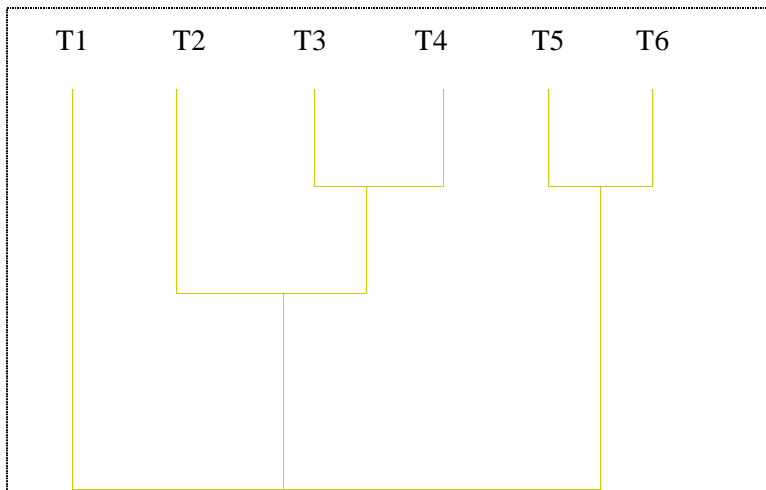


Q7.2. (2 points)

Difference Matrix 2

| D_{ij} or $D_k(ij)$ | T1 | T2 | T(3,4) | T(5,6) |
|-----------------------|----------|----|--------|--------|
| T1 | - | - | - | - |
| T2 | 4 | - | - | - |
| T(3,4) | 7 | 3 | - | - |
| T(5,6) | 6 | 4 | 5 | - |

Tree 2

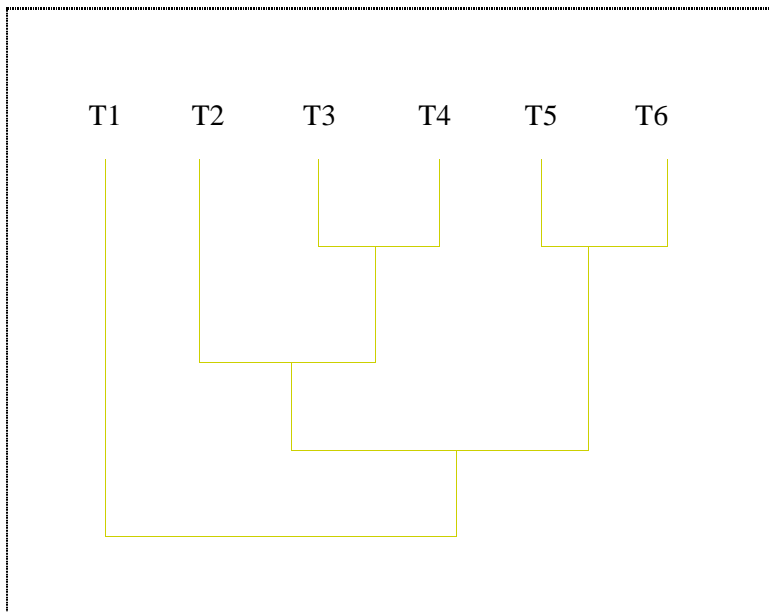


Q7.3. (2 points)

Difference Matrix 3:

| <i>Dij</i> or <i>Dk(ij)</i> | T1 | T(2(3,4)) | T(5,6) |
|-----------------------------|-----|-----------|--------|
| T1 | - | - | - |
| T(2(3,4)) | 5.5 | - | - |
| T(5,6) | 6 | 4.5 | - |

Tree 3

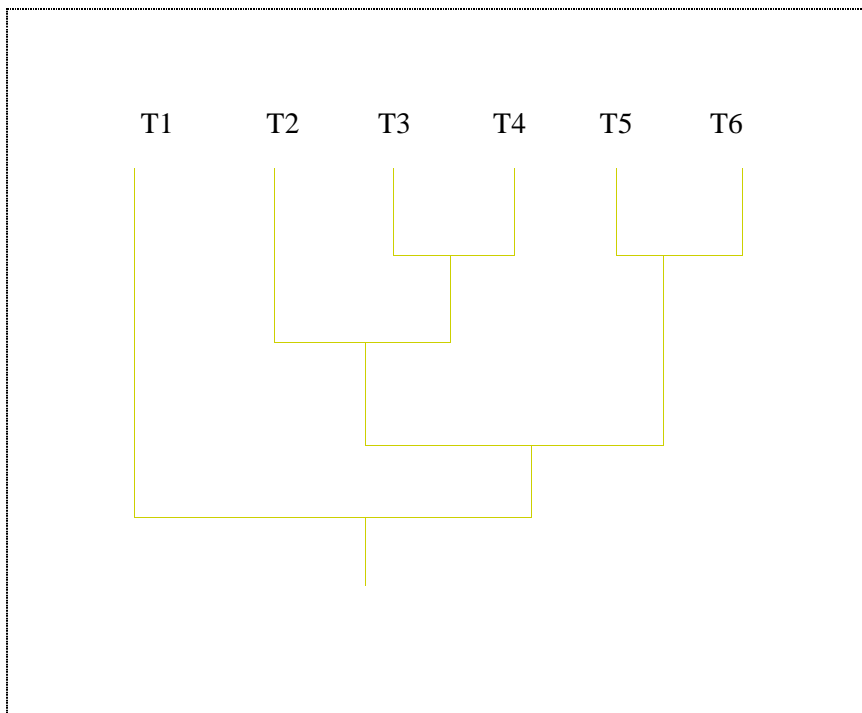


Q7.4. (2 points)

Difference Matrix 4

| D_{ij} or $D_k(ij)$ | T1 | T(2(3,4),(5,6)) |
|-----------------------|------|-----------------|
| T1 | - | - |
| T(2(3,4),(5,6)) | 5.75 | - |

Tree 4



Task III. (7 points)

Q8. (3 points)

| Insect Species | Plant Species |
|----------------|---------------|
| T1 | F |
| T2 | E |
| T3 | D |
| T4 | C |
| T5 | A |
| T6 | B |

Q9. (2 points)

| Insect species | Plant species |
|----------------|---------------|
| T2 | E |

Q10. (2 points)

| A | B | C | D | E |
|---|---|---|---|---|
| √ | | | √ | |