Student Code: _____

20th INTERNATIONAL BIOLOGY OLYMPIAD

第 20 屆國際生物奧林匹亞競賽

12th – 19th July, 2009

Tsukuba, JAPAN

PRACTICAL TEST 3

GENETICS

遺傳學

Total Points: 98

Duration: 90 minutes

Dear Participants,

- This test includes the following 5 tasks:

Task 1: Phenotypic observation of mutant flies	(9 points)
Task 2: Inheritance of white eye mutation	(33 points)
Task 3: Separation of eye pigments	(18 points)
Task 4: Reading chromatography	(14 points)
Task 5: Analysis of White Protein	(24 points)

親愛的參賽者

本試驗須完成下列五項工作：

工作 **1**： 突變果蠅的外表型觀察 **(9** 分**)**

工作 **2**： 白眼突變的遺傳 **(33** 分**)**

工作 **3**： 眼色素分離 **(18** 分**)**

工作 **4**：色層分析判讀**(14** 分**)**

工作 **5**：白眼蛋白質分析 **(24** 分**)**

- **You must 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 and equipment listed for each task. If any of these items are missing, please raise your hand.
- At the end of the test paper, put the Answer Sheet and Question Paper in the envelope. The supervisor will collect this envelope.
- This series of practicals are time consuming. You will need to be well organized and work quickly to complete five tasks.

答案必須寫在 「答案卷」上，否則不予計分。

確實清點所有材料及設備，如有缺少，請立刻舉手。

實驗做完，將題目及答案卷放入信封；監考人員會來收信封。

本實驗極爲耗時，注意控制時間以完成這五項工作。

Good Luck!!

Task 1 (9 points)

Phenotypic observation of mutant flies

Materials and Equipment	Quantity
1. Petri dishes numbered (1)-(4) containing live fruit flies	1 set
2. Stand loupe (magnifying glass)	1

工作 1：突變果蠅的外表型觀察 (9 分)

材料與設備

1. 培養皿(1)-(4)內有活果蠅	1 組
2. 鏡座放大鏡(放大鏡)	1 檯

Introduction

Fruit flies are commonly used materials in genetics studies. Petri dish (1) contains the wild type, and each of the Petri dishes (2)-(4) contains different mutant flies. Observe the flies carefully by using the loupe (magnifying glass), but do not open the lid of the dishes. You may adjust the height and angle of the loupe for your observations.

簡介

果蠅是遺傳上常用的材料，培養皿(1)內有野生型果蠅，培養皿(2) - (4) 內則有不同突變型的果蠅。用放大鏡小心觀察但不可打開培養皿的上蓋，可調整放大鏡架的高度及角度以便觀察。

Q.1.1. (9 points) In the case of each mutant, what kind of trait differs from the wild type?

Choose the characteristic phenotype of the mutant trait from the following list.

A. eye color	B. eye shape	C. wing shape	D. bristle length
E. antenna shape	F. bristle shape	G. leg shape	H. proboscis shape
I. body color	J. abdomen length		

Q.1.1. (9 分) 每種突變型果蠅的哪一種性狀與野生型不同？由下列選項中選出突變型表現出的性狀特徵。

A. 眼睛顏色	B. 眼睛形狀	C. 翅形狀	D. 剛毛長度
E. 觸角形狀	F. 剛毛形狀	G. 腿形狀	H. 吻部形狀
I. 身體顏色	J. 腹部長度		

Task 2 (35 points)

Inheritance of white eye mutation

Materials and Equipment	Quantity
1. 1.5 ml tubes containing anesthetized fruit flies labeled (5a) and (5b), (6a) and (6b), and (7)	1 set
2. Empty Petri dishes	5
3. White cardboard (place under the Petri dishes for easy observation)	1
4. Forceps	2
5. Stand loupe (magnifying glass) (used in Task 1)	1
6. 1.5 ml tube rack	1

工作 2：白眼突變的遺傳 (35 分)

材料與設備

1. 1.5 ml 試管標示(5a)及(5b)、(6a)及(6b)、(7)，內有被麻醉的果蠅	1 組
2. 空的培養皿	5 個
3. 白色板 (置於培養皿下方，爲方便觀察用)	1 個
4. 鑷子	2 支
5. 鏡座放大鏡(工作 1 中用的)	1 檯
6. 1.5 ml 試管架	1 支

Introduction

Wild type fruit flies (WT) have red eyes, while the mutant flies (*w*) have white eyes. *w* is a recessive mutation and located on the X chromosome. Each of tubes (5a) and (5b) or (6a) and (6b) separately contains male and female flies obtained from two different crossings. Tube (7) contains flies from another crossing. Note that flies can be sexed by their patterns of the posterior dorsal abdomen, which is uniformly black in males.

簡介

野生型果蠅(WT)爲紅眼，突變型果蠅(*w*)具有白眼， *w* 爲位於 X 染色體上的隱性突變。(5a)及(5b)、(6a)及(6b)分別裝有由二不同雜交所得之雄蠅及雌蠅；第(7)管裝有另一雜交所得的雄與雌蠅。注意果蠅的雌雄可用腹部背面色帶的型式來區別。

Female

Male

Q.2.1. (8 points) Remove the flies from tubes (5a) and (5b) into different Petri dishes, and observe them by using the loupe (magnifying glass). Examine sex and eye color, and complete the table with the numbers of the flies including zero.

Q.2.1. (8 分) 將試管(5a) 及(5b)中的果蠅移到不同的培養皿中，用放大鏡觀察，檢查其性別及眼睛顏色，在答案卷之表格中記錄果蠅的數目，包括 0 也要記在表中。

Q.2.2. (8 points) Remove the flies from tubes (6a) and (6b) into different Petri dishes, and observe them by using the loupe (magnifying glass). Examine sex and eye color, and complete the table with the numbers of the flies including zero.

Q.2.2. (8 分) 將試管(6a) 及(6b)中的果蠅移到不同的培養皿中，用放大鏡觀察，檢查其性別及眼睛顏色，將果蠅的數目分別記錄在答案卷表格中，包括 0 也要記在表中。

Q.2.3. (8 points) Remove the flies from tube (7) into a Petri dish, and observe them by using the loupe (magnifying glass). Examine sex and eye color, and complete the table with the numbers of the flies including zero.

Q.2.3. (8 分) 將試管(7)中的果蠅移到不同的培養皿中，用放大鏡觀察，檢查其性別及眼睛顏色，將果蠅的數目分別記錄在表格中，包括 0 也要記在表中。

本題刪除 ~~**Q.2.4. (1 分)** As for the flies from tubes (6a) and (6b), choose the correct combination of eye color and sex from the followings. Answer with symbols.~~

~~A. Both females and males have red eyes.~~

~~B. Both females and males have white eyes.~~

~~C. Females have white eyes and males have red eyes.~~

~~D. Females have red eyes and males have white eyes.~~

本題刪除 ~~**Q.2.4. (1 point)** 觀察試管 (6a) 及(6b) 中的果蠅，選擇眼睛顏色與性別的正確組合並填入字母代號。.~~

~~A. 雌雄果蠅均爲紅眼~~

~~B. 雌雄果蠅均爲白眼~~

~~C. 雌蠅爲白眼、雄蠅爲紅眼~~

~~D. 雌蠅爲紅眼、雄蠅爲白眼~~

本題刪除 ~~**Q.2.5. (1 point)** What is the ratio of red-eyed and white-eyed flies in males of tube (7)? Choose the correct ratio from the followings. Answer with symbols.~~

~~A. 1:0 B. 0:1 C. 3:1 D. 1:3 E. 1:1~~

本題刪除 ~~**Q.2.5. (1 point)** 試管(7)的雄性果蠅中，紅眼對白眼的比例爲何？選擇正確比例並填入字母代號。~~

~~A. 1:0 B. 0:1 C. 3:1 D. 1:3 E. 1:1~~

Q.2.6. (9 points) Which of the following crossings produce the flies of tubes (5a) and (5b), (6a) and (6b), and (7)? Choose all possible cases and answer with symbols.

A. Homozygous red-eyed females and hemizygous red-eyed males

B. Homozygous white-eyed females and hemizygous white-eyed males

C. Homozygous red-eyed females and hemizygous white-eyed males

D. Homozygous white-eyed females and hemizygous red-eyed males

E. Heterozygous females and hemizygous red-eyed males

F. Heterozygous females and hemizygous white-eyed males

Q.2.6. (9 分) Which of the following crossings produce the flies of tubes (5a) and (5b), (6a) and (6b), and (7)? 。選擇所有可能情況並填入字母代號。

A. 同基因合子紅眼雌蠅與半基因合子紅眼雄蠅

B. 同基因合子白眼雌蠅與半基因合子白眼雄蠅

C. 同基因合子紅眼雌蠅與半基因合子白眼雄蠅

D. 同基因合子白眼雌蠅與半基因合子紅眼雄蠅

E. 異基因合子雌蠅與半基因合子紅眼雄蠅

F. 異基因合子雌蠅與半基因合子白眼雄蠅

Task 3 (18 points)

Separation of eye pigments

Materials and Equipment Quantity

In addition to the materials and equipment used in Task 2, you will use the following set of equipment in this task.

1. 1.5 ml tubes (8) and (9) containing eye-pigments extraction solution	1 set (1 spare set)
2. Empty 1.5 ml tubes (10) and (11)	1 set (1 spare set)
3. Micropestles (in 15 ml tube)	2 (1 spare)
4. Centrifuge	1
5. Micropipette (P20)	1
6. Pipette tips (for P200 and P20)	1 pack
7. Empty 1.5 ml tubes (no numbers written on the lid)	2 (2 spares)
8. Cellulose/plastic sheet	1 (1 spare)
9. Micropipette (P2)	1
10. Pipette tips (P2)	1 pack
11. 50 ml tube containing solvents	1
12. Tube rack for the 50 ml tube	1

工作 3： 眼色素分離 (18 分)

材料與設備

1. 1.5 ml 試管(8)及(9)，內含有果蠅眼色素的抽取液	1 組 (1 備用)
2. 空的 1.5 ml 試管(10)及(11)	1 組 (1 備用)
3. 研磨均質器 (在 15 ml 試管中)	2 支 (1 備用)
4. 離心機	1 檯
5. 微量吸管(P20)	1 支
6. 微量吸管頭(P200 及 P20 用)	1 盒
7. 空的 1.5 ml 試管(蓋子上無標示)	2 個 (2 備用)
8. 纖維/塑膠片	1 片 (1 備用)
9. 微量吸管(P2)	1 支
10. 微量吸管頭(P2 用)	1 盒
11. 50 ml 試管內含溶劑	1 支
12. 50 ml 試管的試管架	1 支

Procedure 步驟

1. Select five red-eyed and five white-eyed flies classified in Task 2 (either females or males), and remove their heads from the bodies using two pairs of forceps.

   *** Be sure not to crush eyes and abdomen of the flies.**

1. 選取工作 2 中的紅眼果蠅及白眼果蠅各 5 隻(雌或雄均可），用 2 支鑷子取下其頭部。

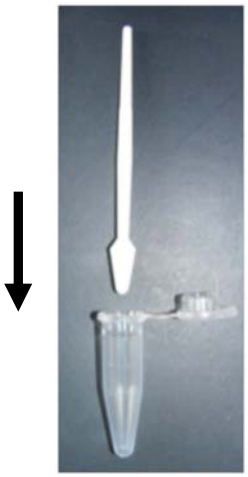
   *** 注意千萬不要壓壞了果蠅眼睛及腹部。**

2. By using forceps transfer the heads of red-eyed flies into tube (8), the heads of white-eyed flies into tube (9), the bodies of red-eyed flies into tube (10), and the bodies of white-eyed flies into tube (11). Tubes (10) and (11) will be used in Task 5.

2. 用鑷子將紅眼果蠅頭部放置於試管(8），將白眼果蠅頭部置於試管(9），紅眼果蠅身體置於試管(10），白眼果蠅身體置於試管(11）。試管(10)及 (11)將會在工作 5 中使用。

3. Insert a micropestle in each of tubes (8) and (9) and grind fly heads by revolving and pressing the pestle against the bottom of the tube with your hand. Use different pestles for different samples.

3. 在試管(8)及(9)中各插 1 支研磨均質器，用均質器以反覆旋轉並壓擠方式研磨果蠅頭部，不同樣本應用不同均質器研磨。



4. Centrifuge tubes (8) and (9) at 14,000 rpm for 3 min **(see the "Instruction for the centrifuge" at the end of this test, pages 18-19, and ask the supervisor for assistance if required).**

4. 將試管(8)及(9) 以 14,000 rpm 離心 3 分鐘**(看本試卷最後所附「離心機使用規則」，若需要幫助時，可問助教。)**

5. Transfer 5 µl of supernatant from tubes (8) and (9) into new tubes.

5. 將試管(8)及(9)上清液 5 µl 移入新試管中。

6. Look at the cellulose/plastic sheet. The shorter sides of the cellulose/plastic sheet are the top and the bottom, and the non-glazy surface is the cellulose surface, which is used in this experiment. Write your student code with pencil at the top of the cellulose surface.

6. 注意纖維/塑膠片的正反面，較短的邊爲頂端與底端，用塗有纖維素的毛面作實驗，以鉛筆在纖維面的頂端寫下你的學生編號。

7. First, spot 1µl of the red-eyed heads extract at 1/3 from the left side and about 2 cm from the bottom of the sheet. Do not draw a line using a pencil or a marker pen, which may scratch the cellulose coating.

7. 先用 1µl 紅眼果蠅頭部萃取液，點在左邊的 1/3、約距離膠片底端 2 cm 處，千萬不要用筆在纖維素那面畫線，那會破壞纖維素的膜。

8. Then, spot 1µl of the white-eyed heads extract at 1/3 from the right side and about 2 cm from the bottom of the sheet.

8. 然後用 1µl 白眼果蠅頭部萃取液，點在右邊 1/3、約距離膠片底端 2 cm 處。

9. When the spots dry, set the sheet into the 50 ml tube so that the bottom of the sheet touches the solvent, and close the cap tightly. Make sure the spots are not touched by the solvent. Open and close the cap of the tube quickly to minimize the leak of vapor.

9. 當墨點一乾即將膠片放進 50 ml 試管中，讓膠片的底部能接觸到溶劑，但千萬不要讓墨點接觸到溶劑，將蓋子栓緊。開關蓋子要快以減少氣體的揮發。

10. Keep the tube straight on the tube rack to start solvent development. You can continue with task 4 and 5 in the test and come back to this section. **Please read part 11 below before you continue.**

10. 將試管放在試管架上保持直立，等待溶劑的展開，等待的時間可以先作其他部分，**但繼續之前你應先讀步驟 11**。

11. When the solvent front on the sheet reaches the 30 ml graduation mark of the tube, take the sheet out from the tube, let it dry on a piece of paper towel and close the cap of the tube. Raise your hand once the cellulose sheet is dry. (Your assistant will collect your sheet to evaluate the result.) **(18 points)**

11. 當溶劑前緣開展到試管上 30 ml的刻度時，將膠片取出放在擦手紙上等待被風乾，栓緊管蓋。膠片乾時請舉手(叫助教收膠片以評估結果) **(18 分)**

Task 4 (14 points)

Reading chromatography

工作 4：色層分析判讀 (14 分)

Introduction

Although some of the eye pigments involved in the compound eyes of fruit flies are invisible to our eyes, they can be visualized under UV lamp. Figure 1 shows an example of eye pigment spots resolved by chromatography and recorded under UV light. Note that the samples include not only WT (wild type) and *w* (white eyes) but also *se* (sepia eyes), *bw* (brown eyes), and *cn* (cinnabar eyes).

簡介

果蠅複眼中的某些色素無法以人眼看見，但若在 UV 燈下就可被辨識出來。圖 1 的例子顯示眼色素經色層分析後在 UV 燈照下的記錄。注意樣本不只包括 WT (野生型) 及 *w* (白眼），也包括 *se* (黃眼）、 *bw* (棕眼)及 *cn* (朱紅色眼）。

There are two pathways of eye pigment production in fruit flies, ommochrome pathway and pteridin pathway. The wild type eye color is formed if all pigments produced in both of the pathways are normally transferred to the compound eyes. Eyes are white if both the ommochrome and the pteridin pigments are absent. Of the pigments and their intermediate compounds involved in the two pathways, only those of the pteridin pathway can be separated by chromatography of this experiment.

果蠅眼色素的產生有兩條路徑，ommochrome 及 pteridin 路徑。若所有二路徑形成的色素均能正常轉移到複眼中，即造成野生型眼睛顏色。若 ommochrome 及 pteridin 的色素均缺乏則爲白眼。兩路徑中只有 pteridin 路徑的色素及其中間產物能以本實驗中的色層分析分開。

The migration of each pigment during chromatography is determined by the chemical nature of the compound, the solubility of the compound to the solvent, and the migration distance of the solvent. The migration distance of a given pigment depends on the developing time of chromatography, but the Rf value is constant for each pigment, which is calculated by the following equation.

$$Rf = \frac{\text{Distance from the base line to the center of the spot}}{\text{Distance from the baseline to the solvent front}}$$

色層分析時每種色素的移動決定於其化學特性、對溶劑中的溶解度及溶劑移動的距離。一特定色素移動的距離，決定於色層分析展開的時間，但每種色素的 Rf 值是一定的，可由下列公式計算：

$$Rf = \frac{\text{由基線到墨點中心的距離}}{\text{由基線到溶劑前緣的距離}}$$

Table 1 summarizes color under UV lamp and Rf value of each pigment separated from the compound eyes of fruit flies.

表 1 總結果蠅複眼中各色素在 UV 燈下的顏色及 Rf 值。

Table 1 Characters of pteridin pigments in compound eyes of fruit flies

Code 代碼	**Name 名稱**	**Color under UV lamp UV 燈下的顏色**	**Rf value Rf 值**
A	2-amino-4-hydroxypteridin	blue	0.57
B	biopterin	blue	0.61
C	drosopterin	orange	0.21
D	sepiapterin	yellow	0.52
E	isoxanthopterin	yellow	0.69
F	xanthopterin	green-blue	0.38
G	isosepiapterin	violet-blue	0.25

Q.4.1. (5 points) Choose the pigment from Table 1 that corresponds to each of the spots separated in the Figure 1 chromatography. Answer with the code in the table. How are the compositions of the pteridin eye pigments of the mutants different from that of the wild type? Estimate the approximate amount of each pigment deduced from the Figure 1 chromatography. Write "++" if there is a lot more of the pigment as compared with the wild type, "+" if the pigment is present in similar amounts as in wild type, and "-" if the pigment is not present.

Q.4.1. (5 分) 由 表 1 中選出對應於圖 1 色層分析中各墨點的色素，將代碼寫在答案卷的表中。突變型 pteridin 眼色素 之組成與野生型有何不同？ 由圖 1 色層分析估計各色素的大約含量多寡寫在答案卷表中，如果比野生型多出非常多的色素，用 "++" 表示， 若色素含量與野生型相似，則用"+" 表示；色素不存在者，用 "-"表示。

Q.4.2. (9 分) Given the eye color and the results of chromatography shown in Figure 1, which of the following abnormalities do *se* (sepia eyes), *bw* (brown eyes), and *cn* (cinnabar eyes) have? Write the corresponding alphabet.

A. Ommochrome pigments must be absent.

B. All pteridin pigments are absent but ommochrome pigments must be present.

C. Both ommochrome and pteridin pigments are absent.

D. Constituent of pteridin pigments differs from the wild type.

Q.4.2. (9 points) 由圖 1 眼睛顏色及色層分析的結果判斷，*se* (黃眼）、 *bw* (棕眼)及 *cn* (朱紅色眼）各有何異常？將代碼寫在答案卷的表中。

A. Ommochrome 色素絕不存在

B. 所有 pteridin 色素均不存在、但 ommochrome 色素一定存在

C. ommochrome 及 pteridin 色素均不存在.

D. pteridin 色素的組成與野生型的不同.

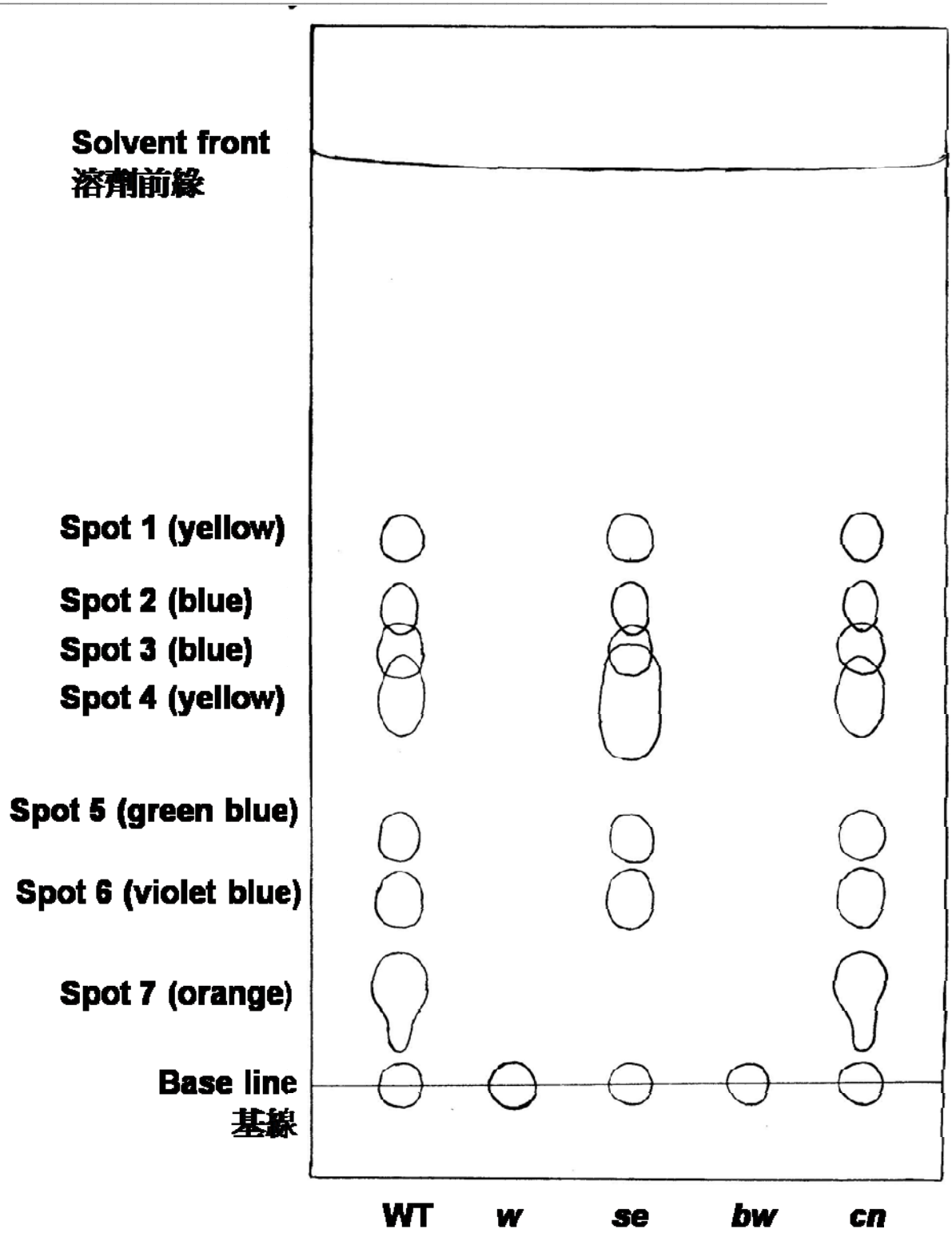


Figure 1. Chromatography of eye pigments from wild type and mutant flies

野生型與突變型果蠅眼色素的色層分析

Task 5 (24 points)

Analysis of White Protein

工作 5：白眼蛋白質分析 (24 分)

Materials and Equipment 材料與設備	Quantity 數量
1. 1.5 ml tube A: Protein extraction buffer 1.5 ml 試管 A: 蛋白質萃取液	1
2. 1.5 ml tubes (two are (10) and (11) of Task 3) 1.5 ml 試管 (從工作 3 (10) 與 (11) 來源，各 2)	4
3. Micropestles (in 15 ml tube) 研磨均質器 (在 1.5ml 試管中)	2 (1 spare)
4. Electrophoresis apparatus with percast gel 電泳設備	1
5. Micropipetter (P200) 微量吸管 (P200)	1
6. Micropipetter (P20) 微量吸管 (P20)	1
7. Pipette tips (for both P200 and P20) 微量吸管頭 (P200 與 P20 用)	1 pack
8. 1.5 ml tube rack 1.5 ml 試管架	1
9. 1.5 ml tube C: Protein electrophoresis marker 1.5 ml 試管 C：蛋白質電泳標記	1

Protein extraction and electrophoresis

蛋白質萃取與電泳

1. Add 50 µl protein extraction buffer (tube A) in the tube (10) (bodies of red-eyed flies) and (11) (bodies of white-eyed flies) prepared in Task 3. Crush the flies with the micropestle. Use different micropestles for wild type and mutant samples.
   分別將 50µl 蛋白質萃取液 (試管 A) 添加到來自工作 3 的(10)號試管(紅眼果蠅的身體)及 (11) 號試管 (白眼果蠅身體)中。用研磨均質器磨碎果蠅身體，但野生型與突變型不可混用。

2. Centrifuge tubes (10) and (11) at 14,000 rpm for 3 min, and then transfer supernatant into fresh 1.5 ml tube **(see the "Instruction for the centrifuge" at the end of this test, pages 18-19, and ask the supervisor for assistance if required)**
   於 14,000 rpm 下 離心 3 分鐘，將上清液移到新的 1.5 ml 試管中。(使用方法參考本試題最後面的離心機使用法或請助教協助)

3. The assistant has prepared a gel for you and it is ready for use.
   助教已經幫你把膠體與電泳設備裝置完成。

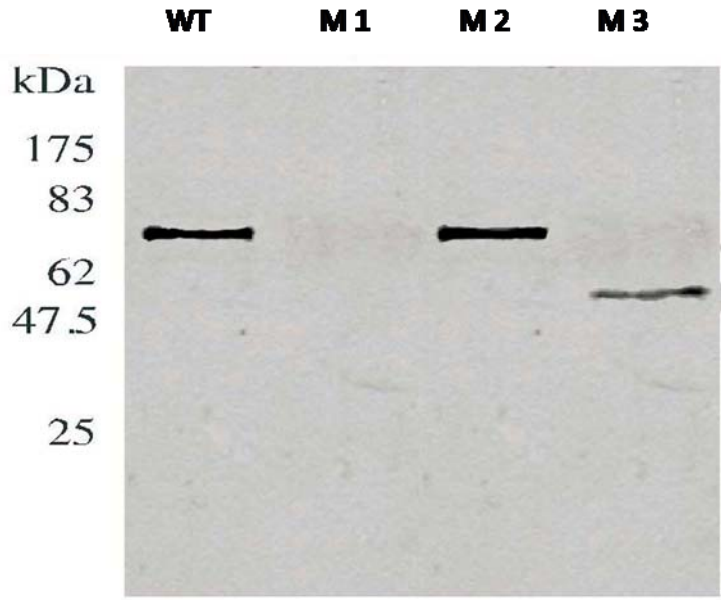
4. Load 5 µl of each sample on the slots in the middle of the gel plate in the order of molecular weight marker, red eye and white eye (from left to right). When you have finished sample loading, raise your hand for the supervisor. Your assistant will take care of the apparatus and start electrophoresis.
   依照分子量標記、紅眼、白眼的順序(由左到右)，在膠體的中間處加入樣本。每個樣本添加 5µl。請在添加樣本完成時舉手。助教會協助你完成裝置並跑電泳。
5. After 5 min, call your assistant by raising your hand. Your assistant will collect the lower part of the apparatus and take a photograph of the gel for evaluation **(18 points).**
   Please check the image on the camera with your assistant
   跑膠至少經 5 分鐘後，再舉手請助教來協助膠體攝影以供評分**(18 分).**。
   請在照相機的視窗下與助教確認影像。

Analysis of protein electrophoresis data

M1, M2 and M3 flies are different mutant lines for the eye pigment genes. After separating proteins of these mutant flies through SDS polyacrylamide gel, proteins were transferred onto a nylon filter to be probed with antibody that specifically recognizes the protein encoded by the *white* gene. The following result was obtained.

蛋白質電泳結果分析

M1, M2 與 M3 果蠅爲不同眼睛顏色的突變種。當電泳完成後，膠體上的蛋白質被轉印到尼龍紙上，並以抗體免疫染色來進行白眼蛋白的辨識。結果如下圖所示：



Q.5.1. (3 points) Which of the following defects of eye pigment genes causes the electrophoresis results of M1, M2 and M3? Choose the corresponding symbols from A, B and C.

A. The mRNA initiation site of the *white* gene is deleted, and the gene is not expressed.

B. A stop codon mutation has occurred in the coding region of the White protein, resulting in failure of translation of carboxyl terminal peptide sequence corresponding molecular weight 20 kDa.

C. Although a normal White protein is synthesized, genes involved in the synthesis of ommochrome pigments are defective.

Q.5.1. (3 分)下列何種眼色素基因的缺陷引起 M1、M2 、M3 電泳結果？在答案卷中填 A、B 或 C。

A. *white* 基因 mRNA 啓動位置被刪除，基因無法表現。

B. 白眼蛋白的編碼區出現終止碼突變，造成 C 端有 20 kDa 的氨基酸序列無法轉譯

C. 雖有正常白眼蛋白合成，但合成 ommochrome 色素的基因有缺陷。

Q.5.2. (3 points) Choose another defect of eye pigment gene from A, B and C that would cause the same phenotypes as M1, M2 and M3.

A. The coding sequence of the *white* gene is fused with the coding sequence of another gene by chromosomal translocation, resulting in a novel sequence encoding a fusion protein that retains antibody reacting sites but exhibits about 30 % lower molecular weight.

B. A single base substitution has occurred in the protein-coding region of the *white* gene changing an amino acid coding sequence into another amino acid coding sequence. However, immunological reactivity of the altered protein for the antibody is not lost.

C. A large deletion exists in the chromosomal region that involves the entire *white* gene.

Q.5.2. (3 分) 在下列的 A、B、C 中，選擇會引起與 M1, M2 及 M3 相同外表型的另一種眼色素基因缺陷，填在答案卷中。

A. 因染色體轉位使 *white* 基因的編碼序列與另一基因的編碼序列融合，造成帶有一異常序列的蛋白，保留了抗體活化位但分子量約低 30 %。

B. 因 *white* 基因的蛋白質編碼區發生單一鹼基取代，造成一個氨基酸的不同，但以改變後蛋白質生成之抗體其免疫活性並未喪失。

C. 染色體中出現包括 *white* 基因的整段刪除。

Instructions for the centrifuge

離心機使用說明

Ask the supervisor for assistance if required

有需要時可向監考人員求助

1. Press the OPEN button at the upper-right of the operation panel (Fig. 1 - 1) to open the centrifuge lid (2).

壓下操作面板右上方之 OPEN 按鍵 (圖 1~1)打開離心機的外蓋(2)。

2. The rotor is covered by a plastic cap (Fig. 2 - 3). To remove the cap, hold the cap with one hand, and unfasten the central black screw (4) anti-clockwise with the other hand.

離心機的轉子上有一塑膠蓋子(圖 2~3)，先用一手固定塑膠蓋子，另一手以逆時鐘方向鬆開中間的黑色旋紐(4)以打開塑膠蓋子。

3. There are 24 holes inside the rotor (Fig. 3). Set the sample tubes in a symmetric position, considering their balance.

離心機的轉子內有 24 個插孔，將樣本以對稱方式放入插孔內，注意樣本間的平衡。

4. Turn the rotor cap screw (4) clockwise to fasten the cap on the rotor.

以順時方向轉動黑色旋紐(4)，將塑膠蓋子鎖緊在離心機的轉子上。

5. Close the centrifuge lid firmly. You should hear a beep that tells complete closure.

輕輕蓋上離心機的外蓋，當外蓋完全蓋上時，你會聽到「嗶」聲。

6. The centrifuge speed (140 x 100 rotation per minute) and time (3 min) is preset. Confirm the set parameters in the windows (5) and (7) by pressing the DISP/CE button (6), and press the START button (8) to start centrifugation.

離心機的轉速(140 x 100 轉/每分鐘)及轉動時間(3 分鐘)已預先設好，按下 DISP/CE 按鍵，在視窗(5)及(7)上確認轉速及轉動時間，確認後按下 START 按鍵開始離心。

7. When centrifugation is finished, the lid (2) is automatically unlocked. Then, open the lid (2) fully and remove the rotor cap by unfastening the screw (4) anti-clockwise while holding the rotor cap with the other hand.

當離心完成後，外蓋(2)會自動解除鎖定，打開外蓋(2)，一手固定塑膠蓋子，另一手以逆時鐘方向鬆開中間的黑色旋紐(4)以打開塑膠蓋子。

8. In order to not disturb the precipitates, take out the sample tubes carefully from the rotor. Leave them on the tube stand.

   避免影響沈澱，小心取出樣本管並放在試管架上。

9. Replace the rotor cap (3) and fasten the screw (4) clockwise, and close the centrifuge lid (2).

   重新蓋上塑膠蓋子(3)，以順時方向鎖緊黑色旋紐(4)，將外蓋蓋上。

Figure 1

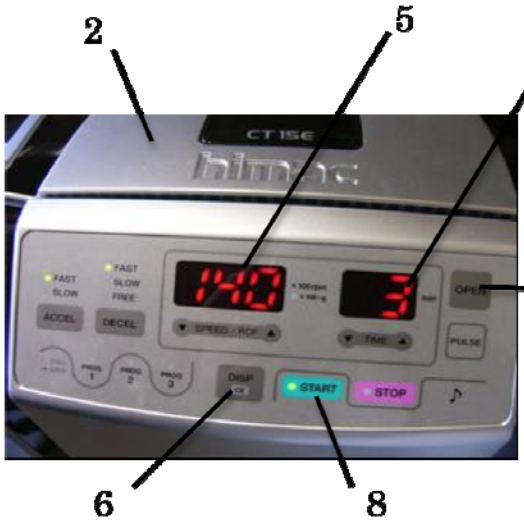


Figure 2

Figure 3