



Effects of LED light spectra on the endocrine regulation and sex change in the teleost

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Chapter 6. Conclusion

Chapter **1**



Introduction

Photic environment

✓ Endocrine regulation

- Mammalian < fish (temperature, salinity, photoperiod..)

Light

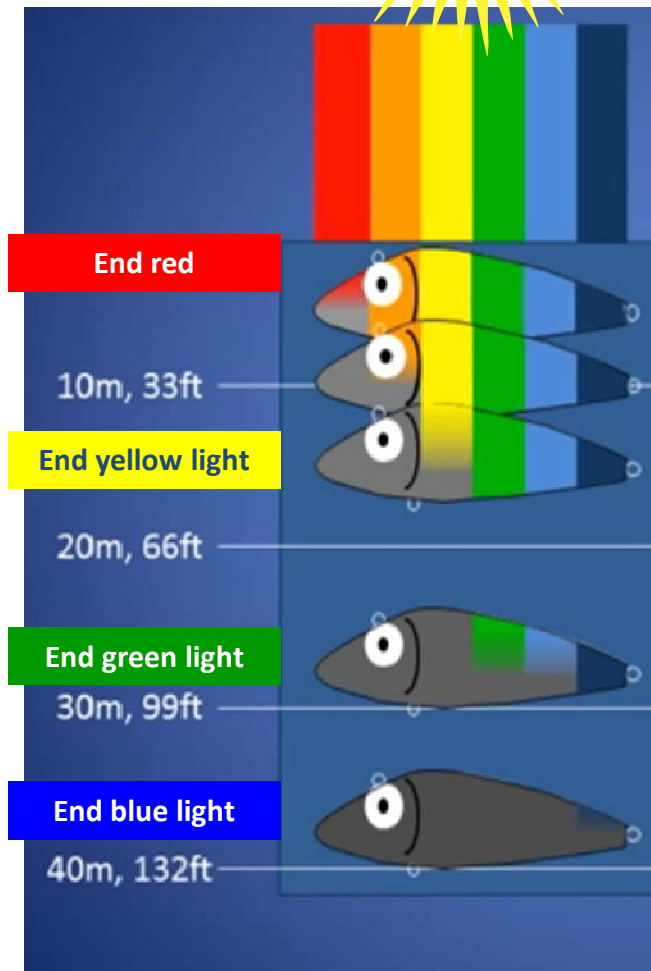
- Light affects many of the **physiological and behavioral changes**
- The central importance of the **day-night light cycle and photoperiod**
- **Light-sensitive circadian clocks** have evolved in most animals

Photic environment

- Fish live in **aquatic environments** with varying light intensity
- They possess **visual pigments** with the absorption spectrum
- Among the factors that influence **circadian rhythms**

★ Light is the most important one ★

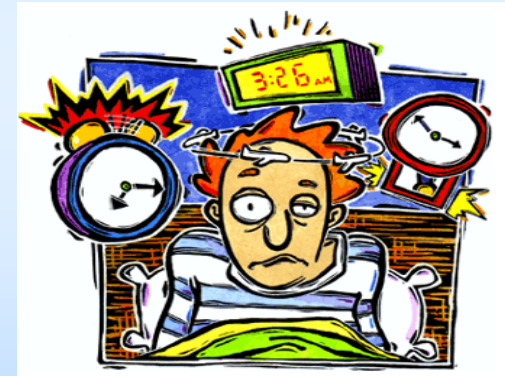
Effects of various wavelengths



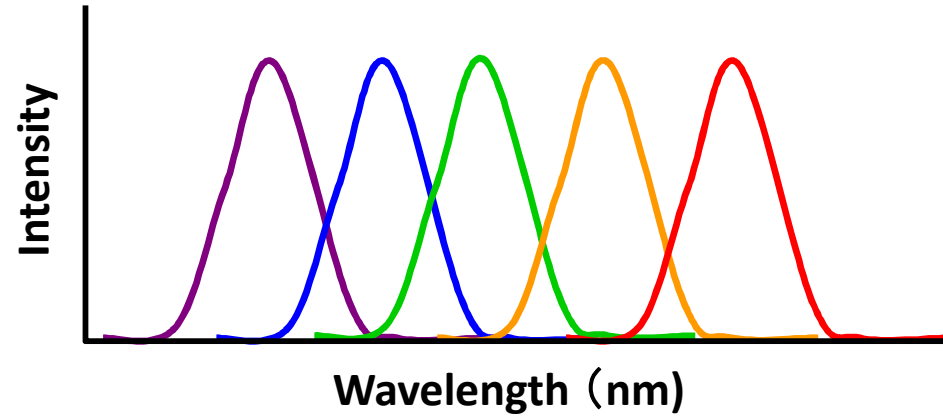
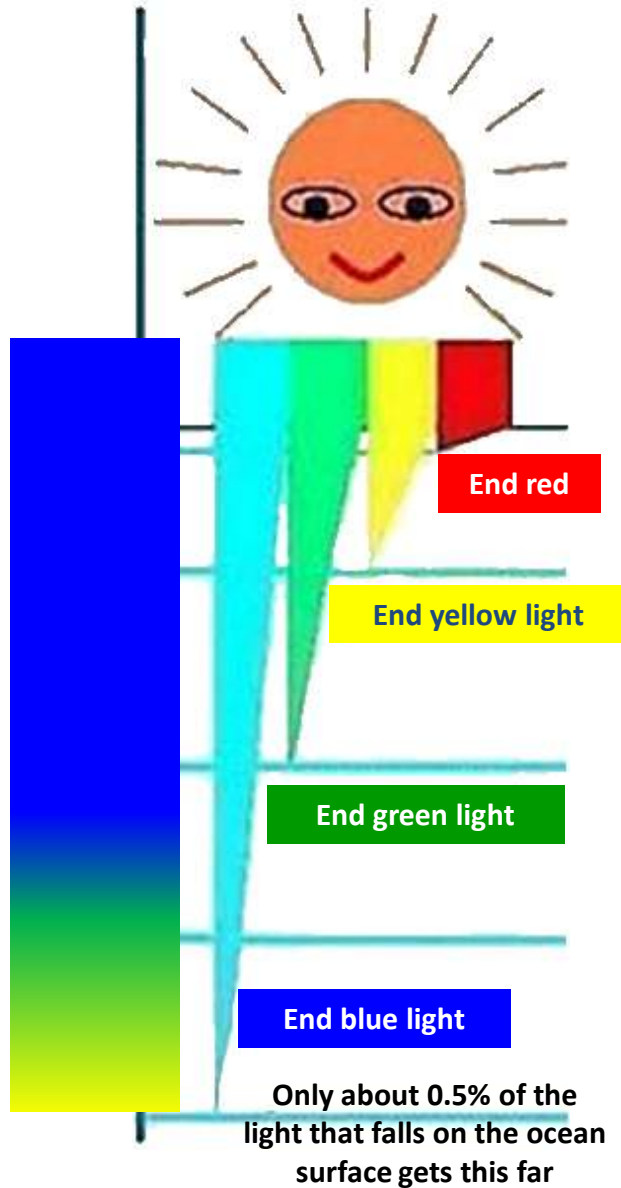
Wavelengths

- The **spectral composition** of incident light : Key properties affecting the physiological response

- **Stress**
- **Circadian rhythm**
- **Growth**
- **Reproduction**

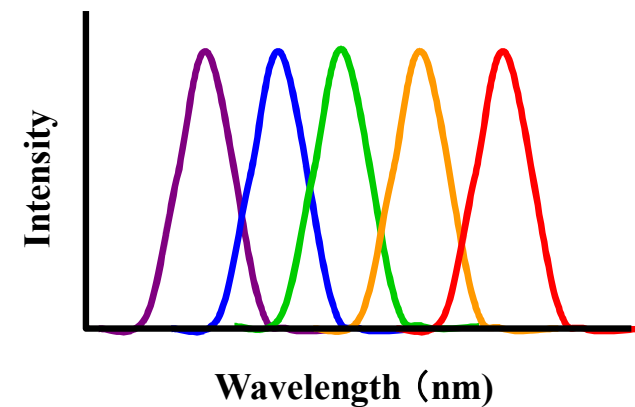


Effects of various wavelengths



Light-emitting diode **LED**

- A new form of lighting technology
- Output specific wavelengths
- Low power requirement
- High efficiency
- Longer life span
- Efficient lighting systems



LED

Indicator lights



LCD panel backlighting

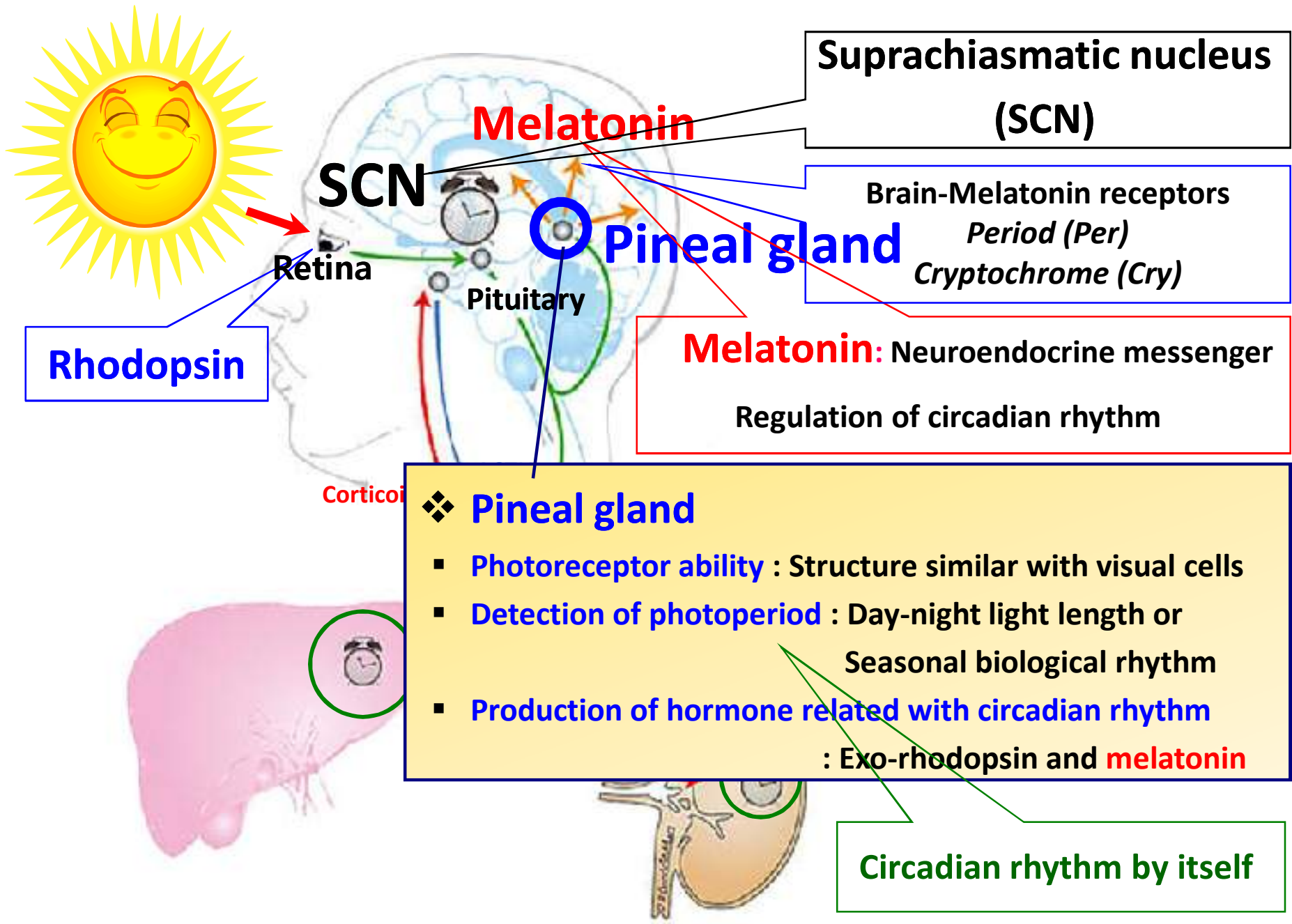


Plant cultivation



Fish-luring light





Suprachiasmatic nucleus (SCN)

Brain-Melatonin receptors
Period (Per)
Cryptochrome (Cry)

Melatonin: Neuroendocrine messenger
 Regulation of circadian rhythm

Rhodopsin

SCN

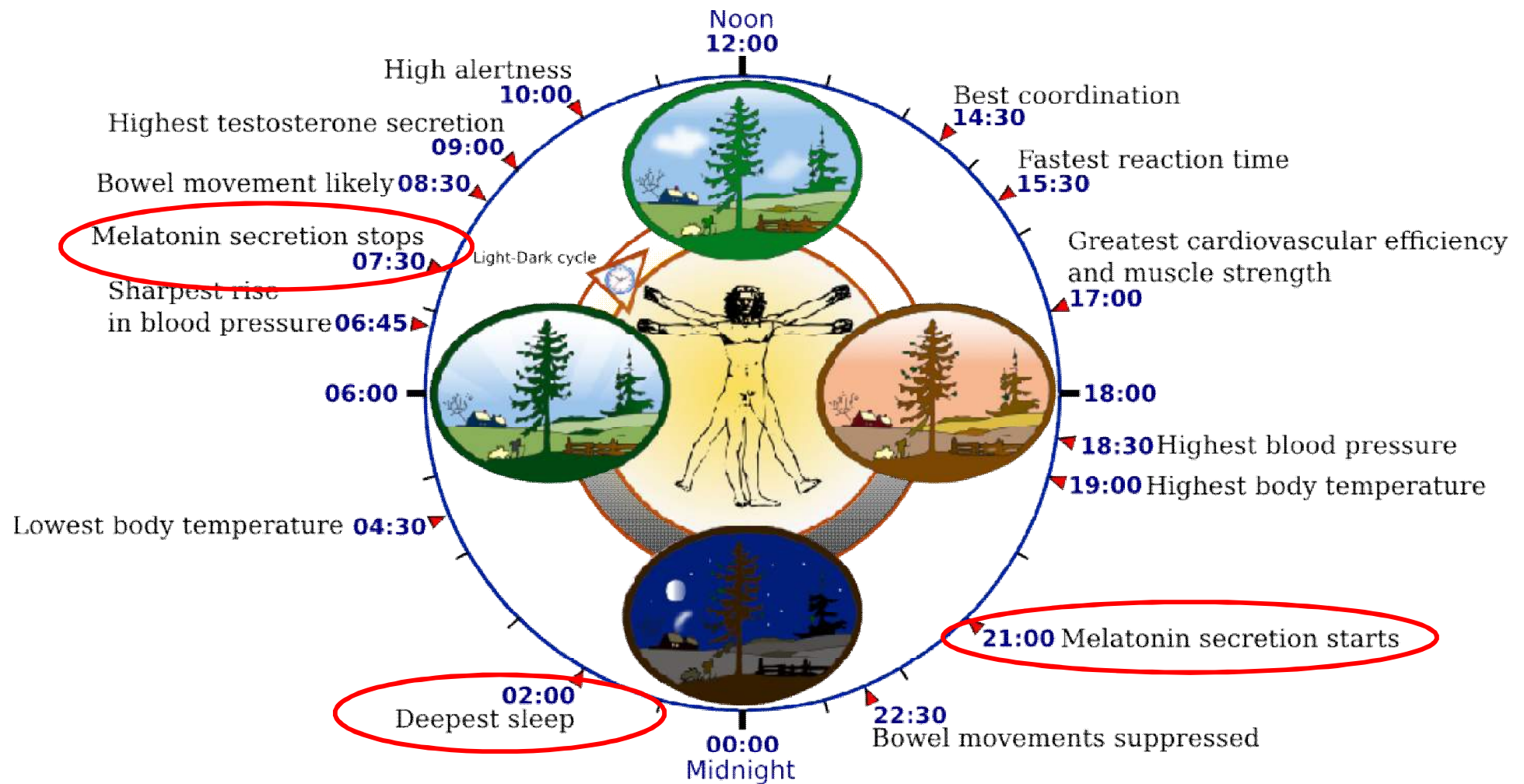
Pineal gland

❖ **Pineal gland**

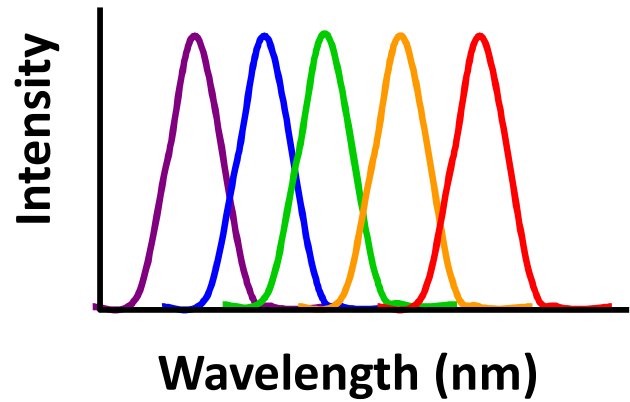
- **Photoreceptor ability** : Structure similar with visual cells
- **Detection of photoperiod** : Day-night light length or Seasonal biological rhythm
- **Production of hormone related with circadian rhythm** : Exo-rhodopsin and **melatonin**

Circadian rhythm by itself

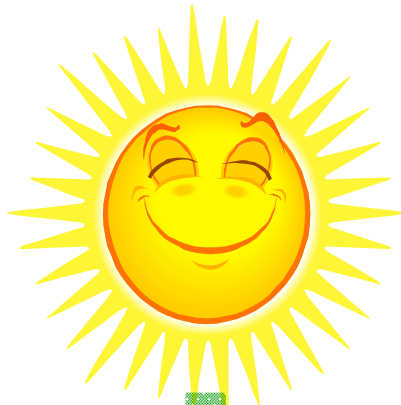
Circadian rhythm



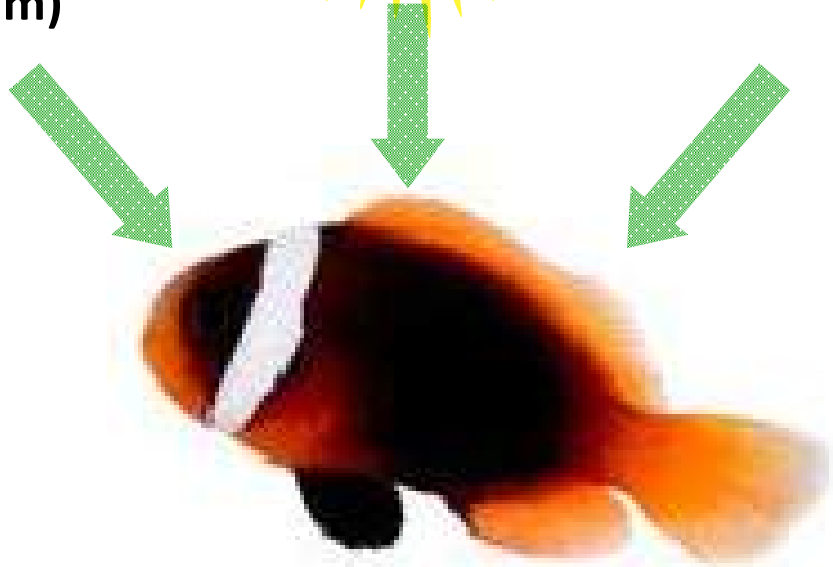
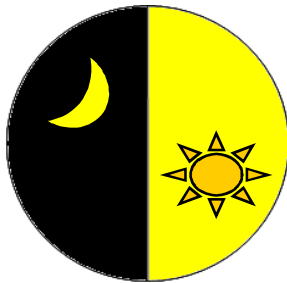
Light spectrums



Light intensity



Light-dark cycle



Endocrine regulation

Circadian rhythm

Sex maturation

Sex change

Chapter 2

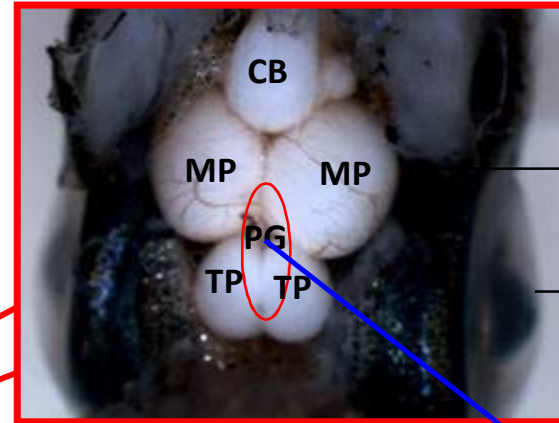
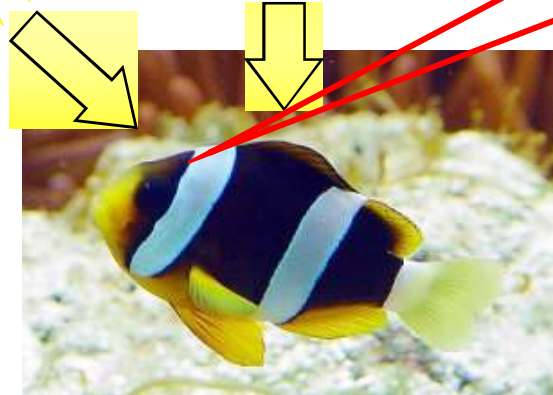
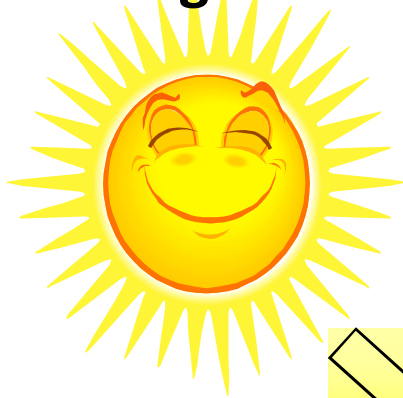


Effect of **LED** light spectra on **circadian rhythm**

Melatonin

Light

Light-dark cycle



DP (under MP)

Eye

Melatonin secretion

Pineal gland



Circadian rhythm genes

- **Melatonin receptor (MT)**

: Detected in the SCN, where the master circadian clock system

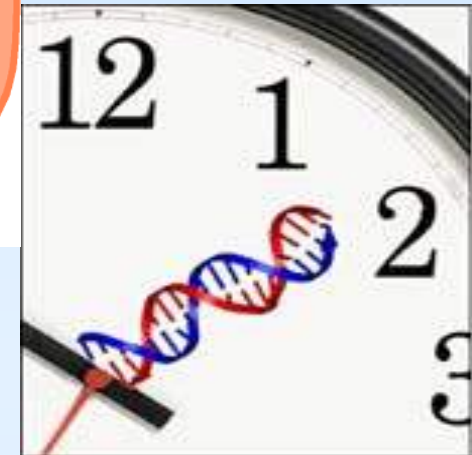
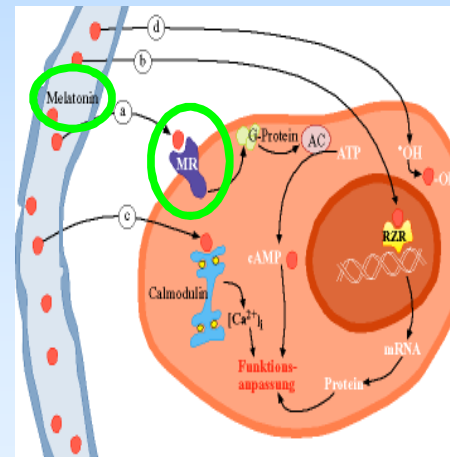
- ***Period (Per)***

: A circadian oscillator

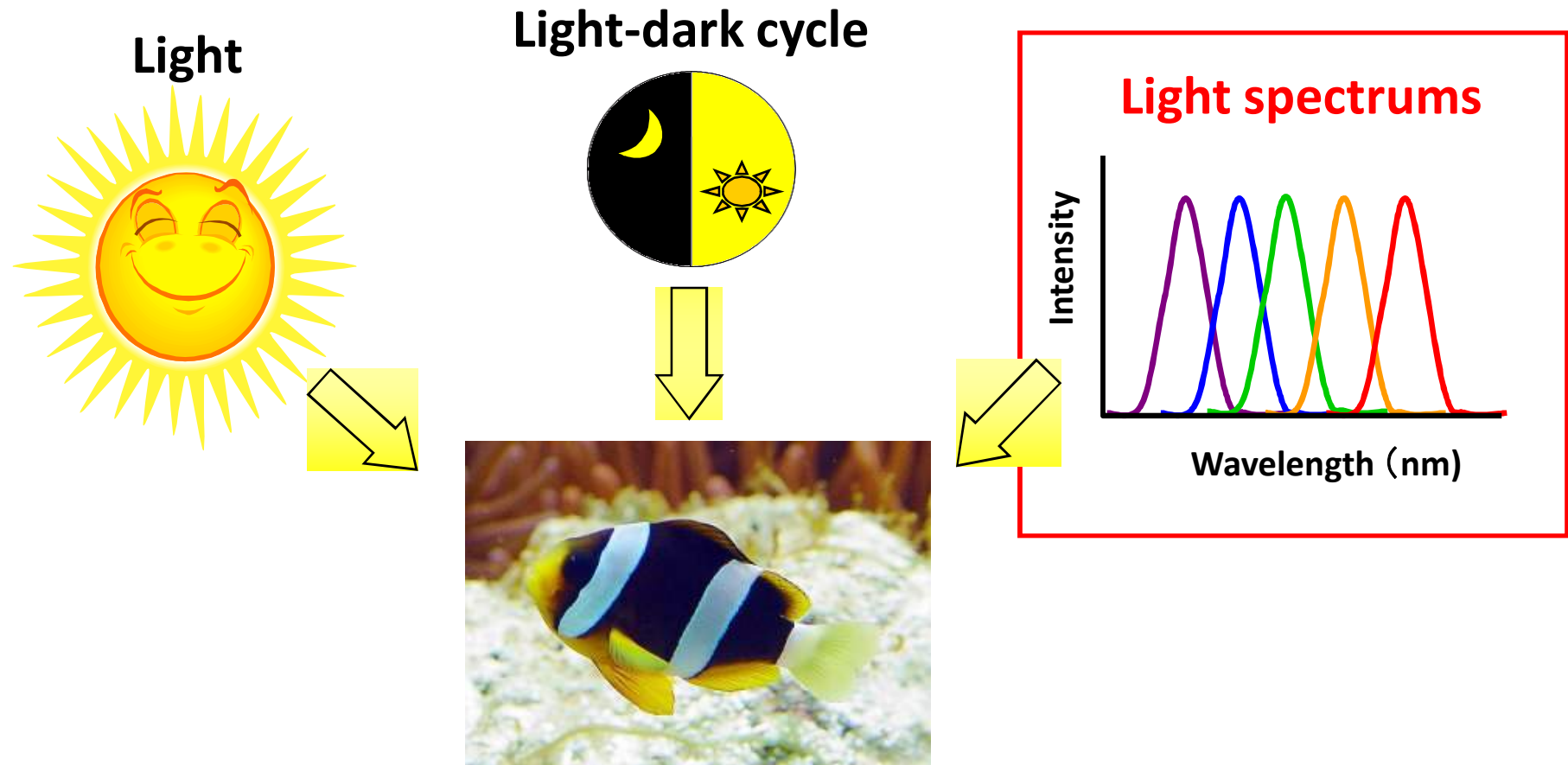
Light-dependent clock resetting

- ***Cryptochrome (Cry)***

: Regulates gluconeogenesis in the liver and the biological clock



Purpose



Effects of light spectra and melatonin on **circadian rhythms**

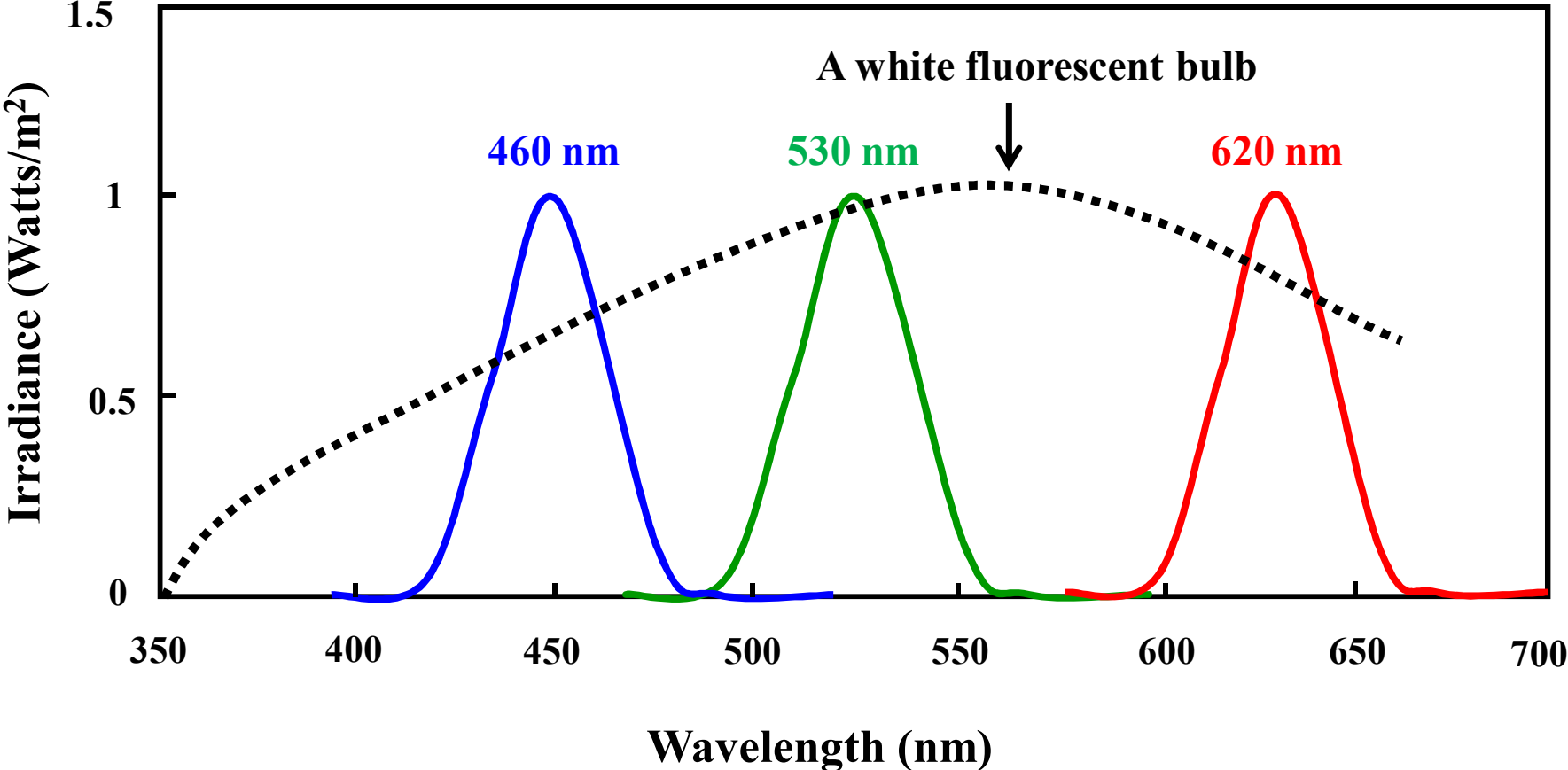
using **LEDs (red, green, and blue)**

Yellowtail clownfish



- *Amphiprion clarkii*
- Order: Perciformes
- Family: Pomacentridae
- It is the most widely distributed anemonefish in Indo West Pacific
- Protrandous hermaphrodite
- A high-value ornamental fish

Spectral profiles of LED used in this experiment

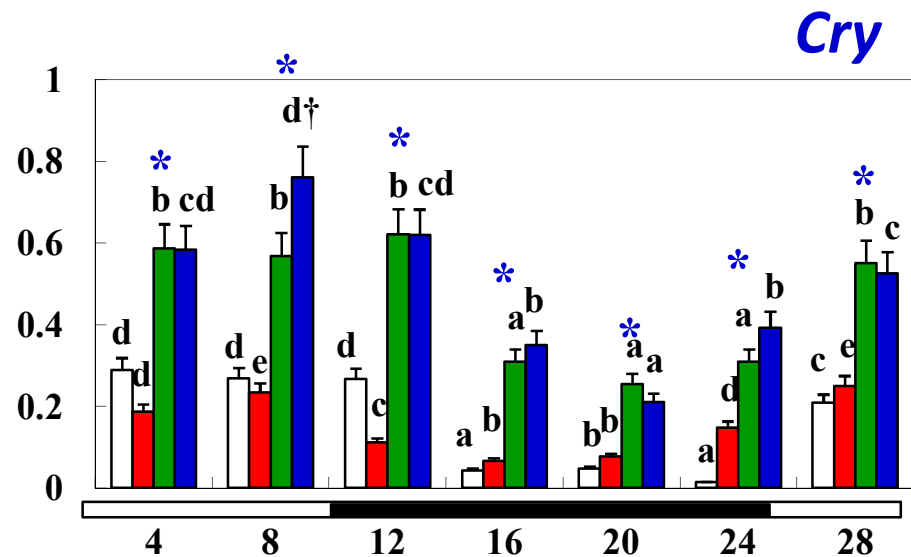
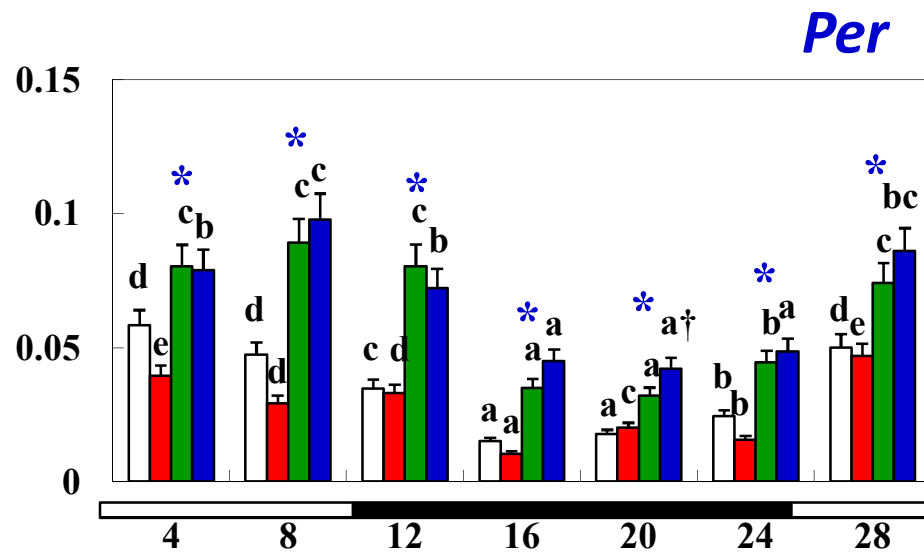
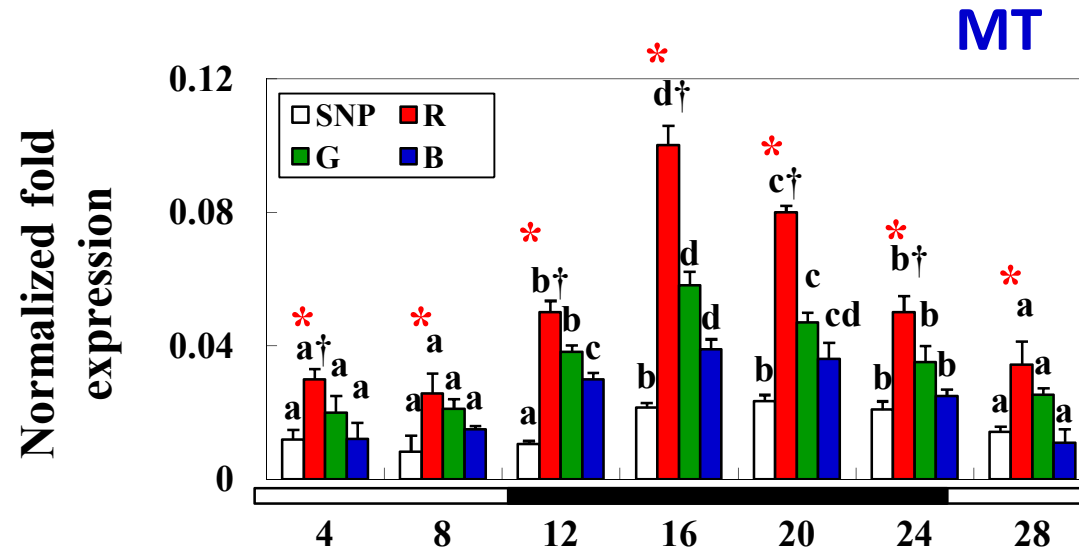


LED aquarium in Korea Maritime and Ocean University



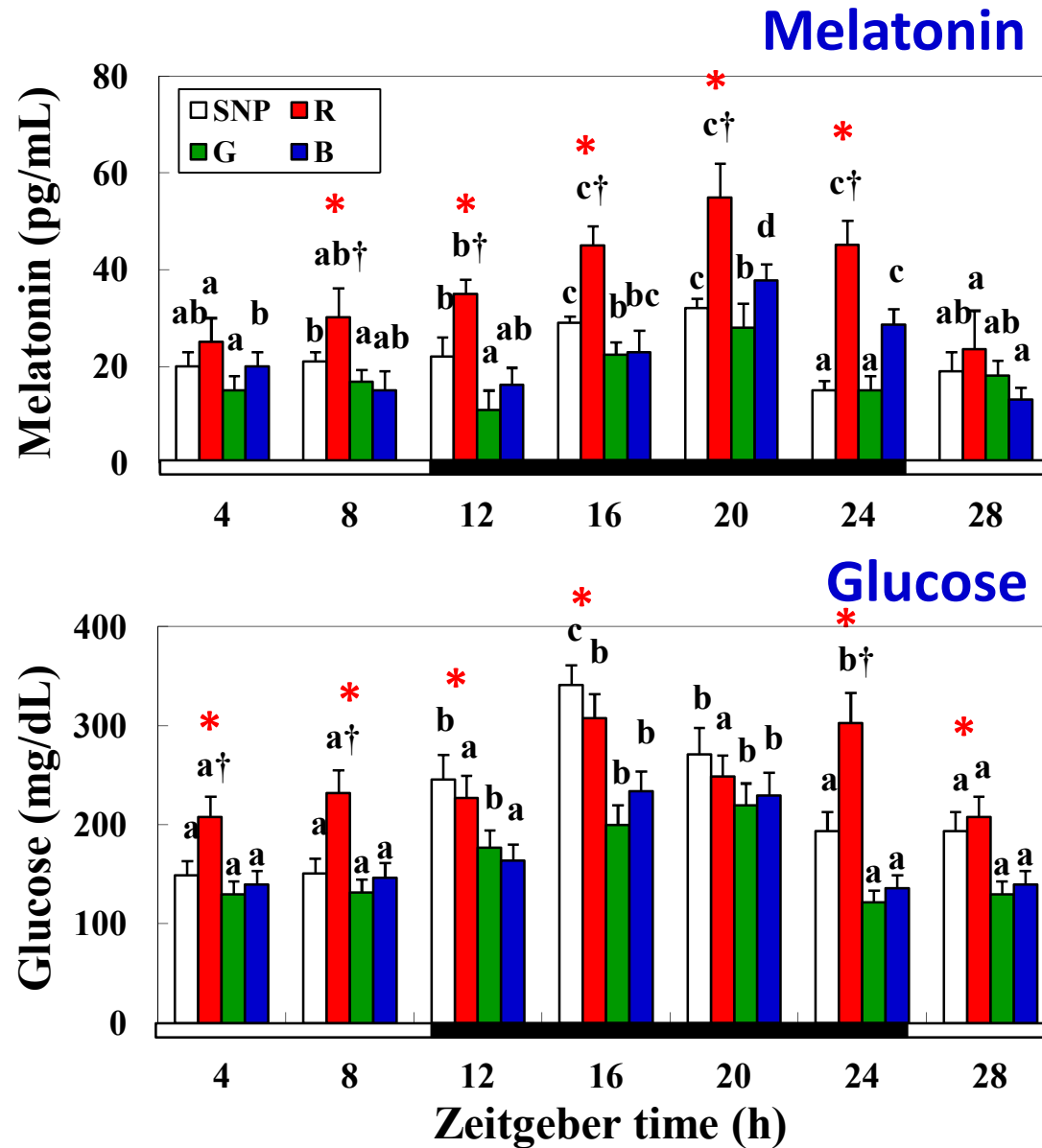
LED controller

Expression levels of *MT*, *Per*, and *Cry* mRNAs

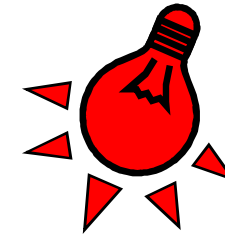
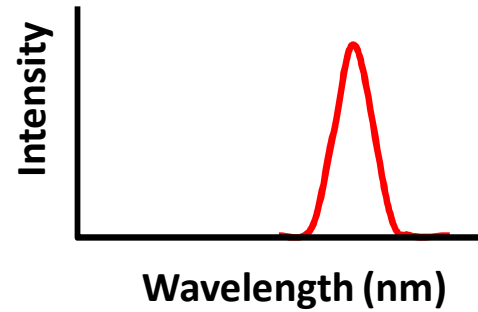


Zeitgeber time (h)

Plasma melatonin and glucose levels under LEDs



Result



Red : MT ↑

Blue-Green : *Per, Cry* ↑



Circadian rhythm

- **Red** light could not be detected by the visual system of the fish
- **Red** wavelengths **LED** inhibits the circadian rhythms
- **Red** wavelengths **LED** is induced the stress

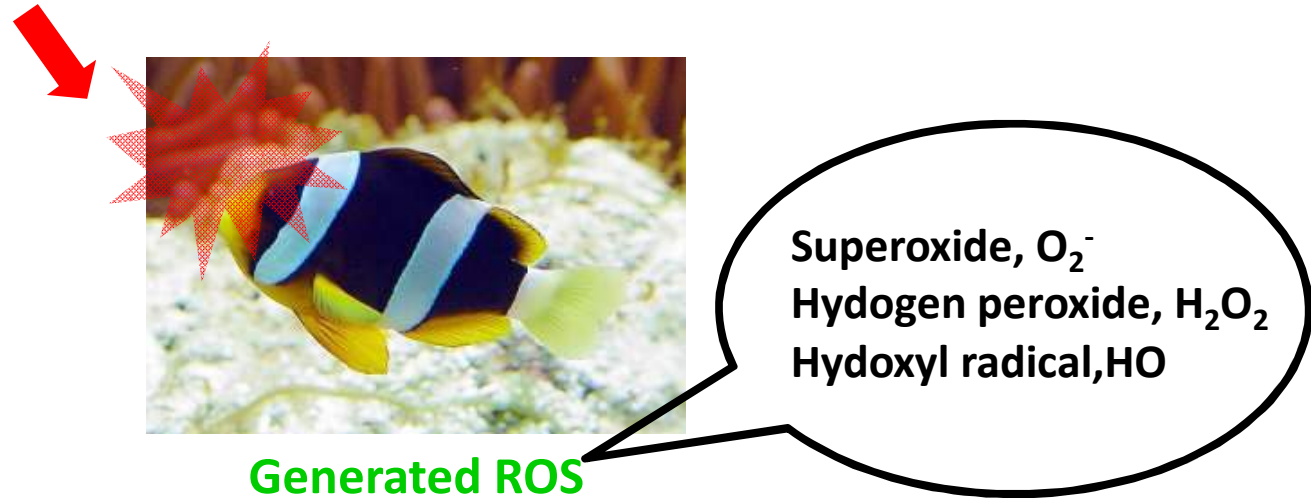
Chapter 3



**Effect of LED light spectra
on stress and immunity**

Oxidative stress

Various environmental factors



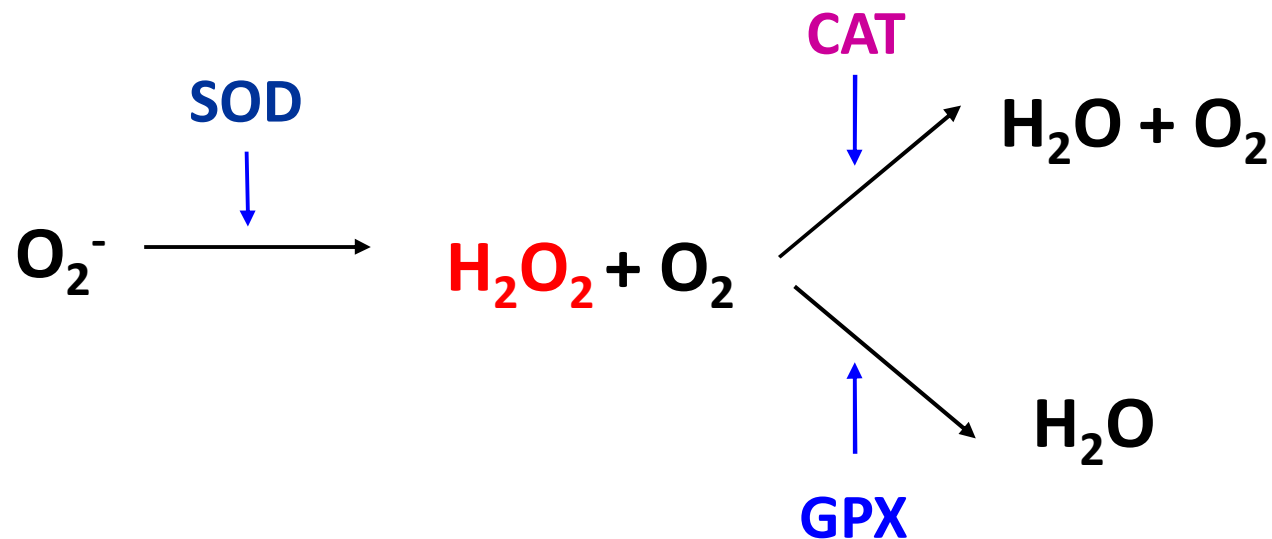
↓
Induced oxidative stress

↓
Strand breaks in DNA
oxidation of polyunsaturated fatty acids in lipids

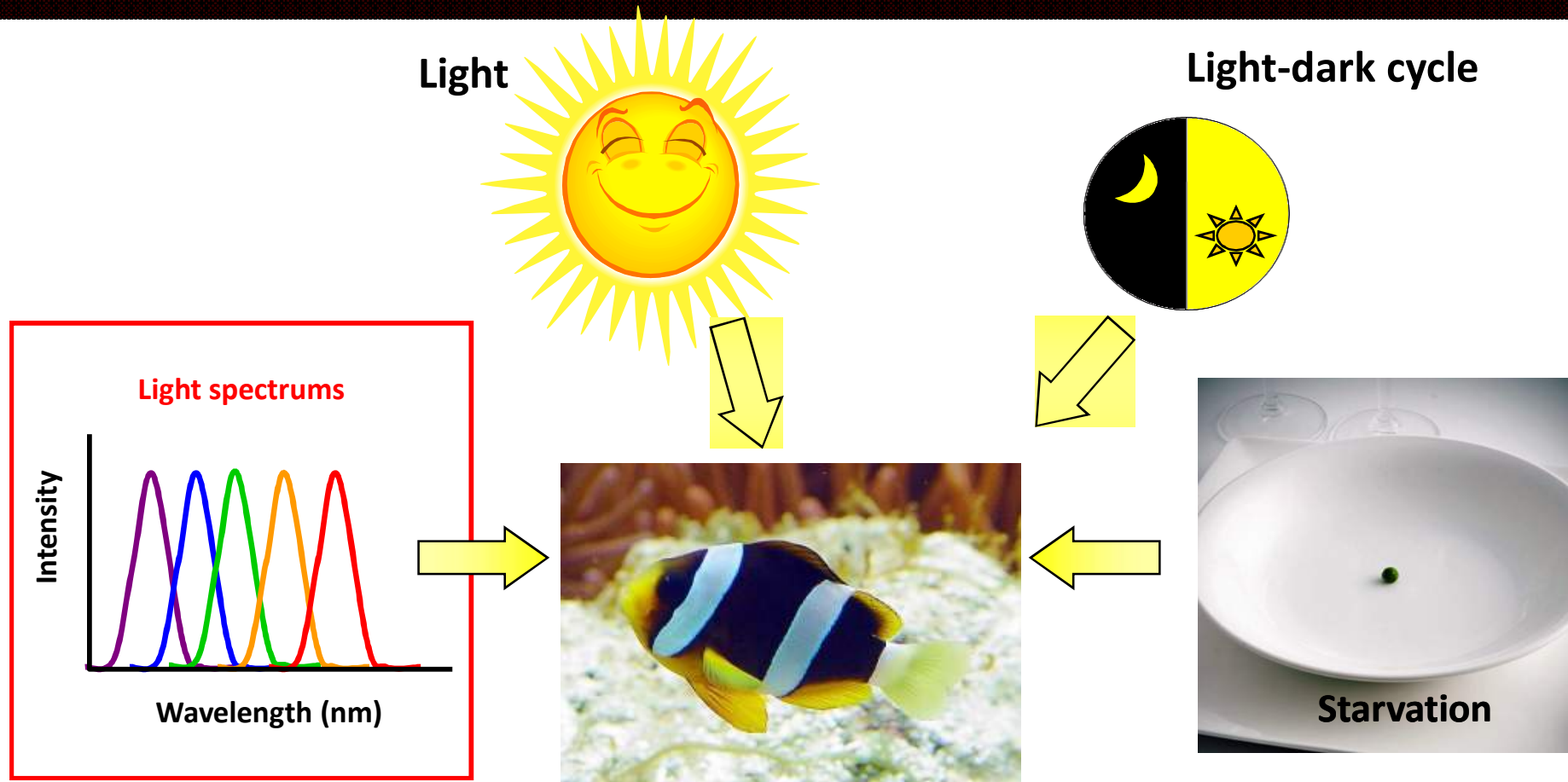
Reduced disease resistance and reproductive ability

Antioxidant defense system

- ❖ To protect themselves against oxidative stress, organisms have evolved complex antioxidant defense system
 - ✓ Superoxide dismutase (SOD), catalase (CAT),
 - ✓ Glutathione peroxidase (GPX), glutathione-S-transferase (GST),
 - ✓ Glutathione (GSH), metallothionein (MT), ascorbic acid, vitamin E



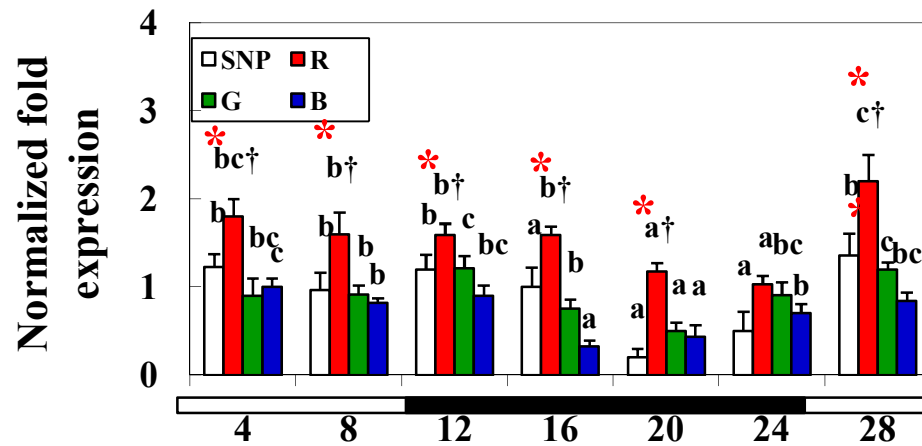
Purpose



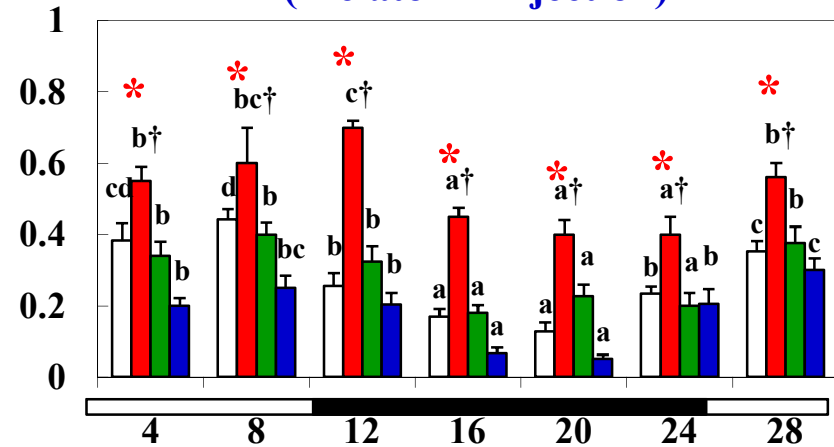
Effects of light spectra and starvation on **oxidative stress** and **immunity** using **LEDs** (**red**, **green**, and **blue**)

Expression of SOD and CAT under LEDs

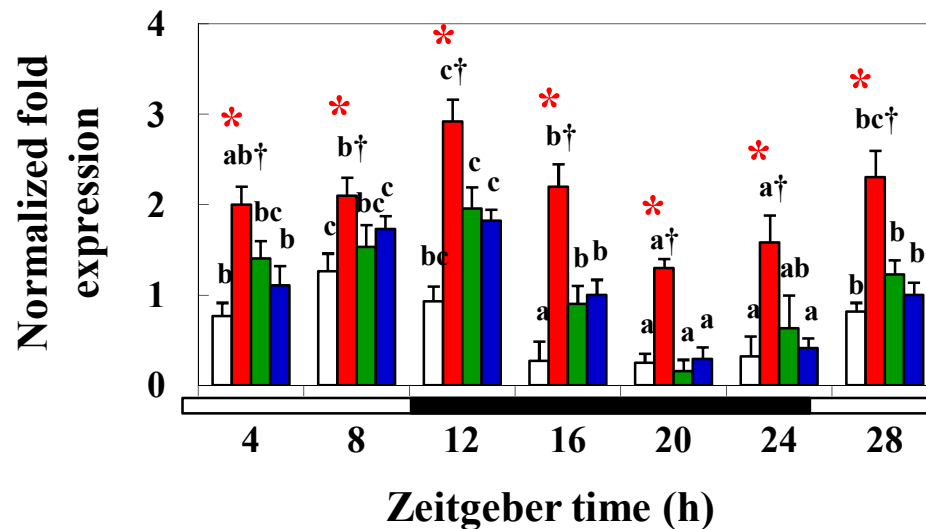
SOD mRNA expression in liver



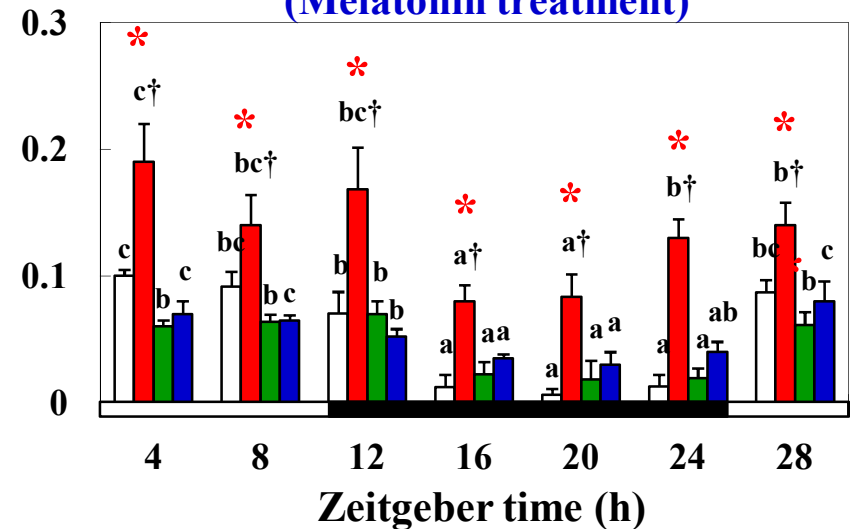
SOD mRNA expression in liver (Melatonin injection)



CAT mRNA expression in liver

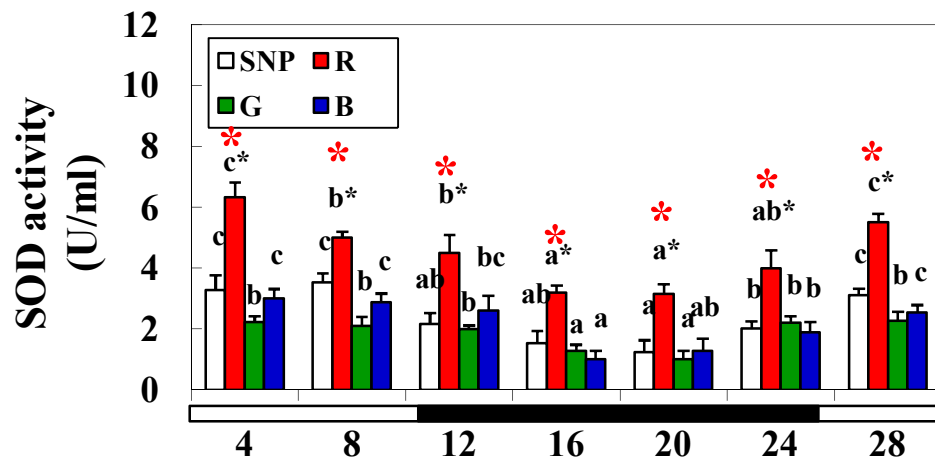


CAT mRNA expression in liver (Melatonin treatment)

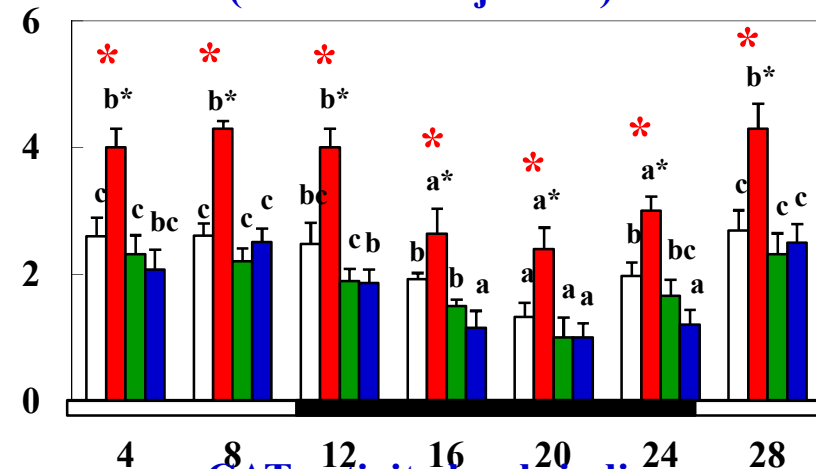


Activity of SOD and CAT under LEDs

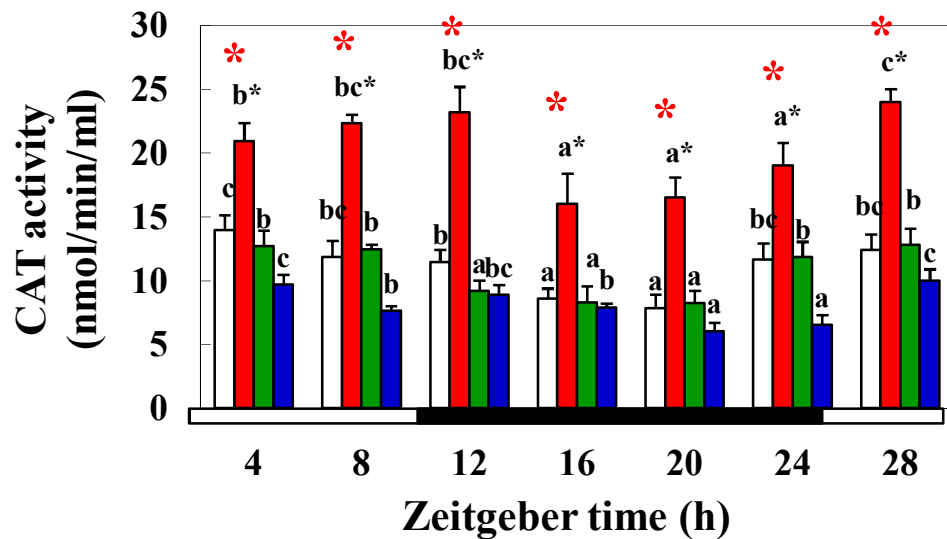
SOD activity levels in liver



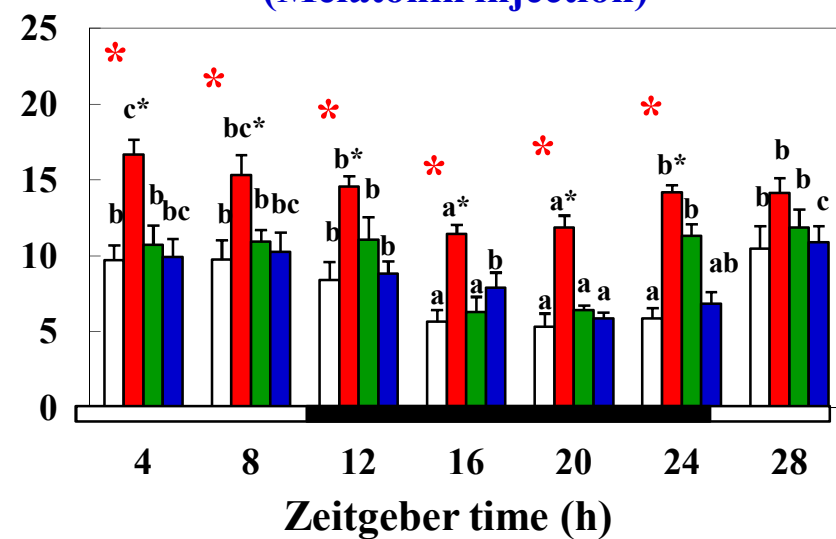
SOD activity levels in liver (Melatonin injection)



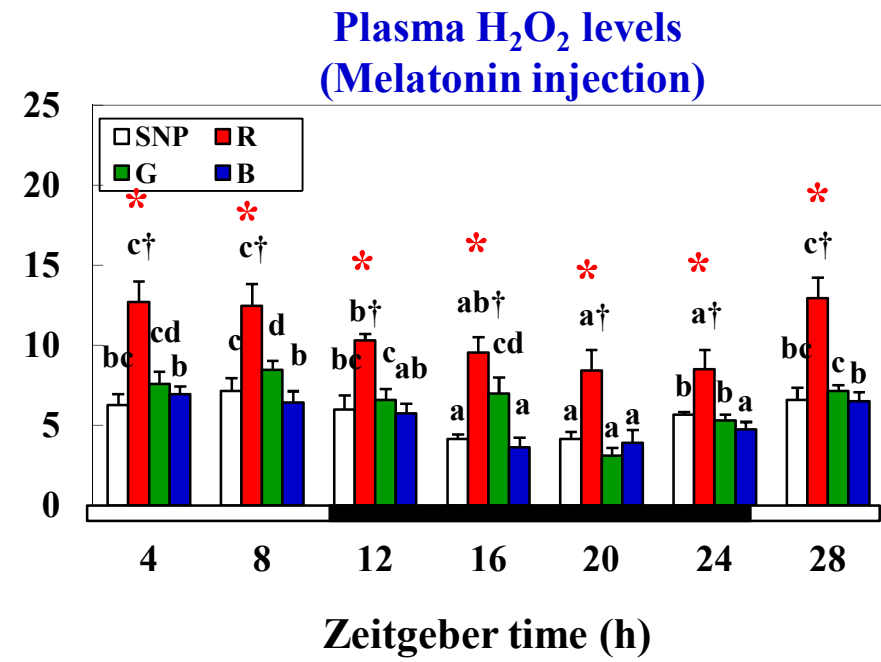
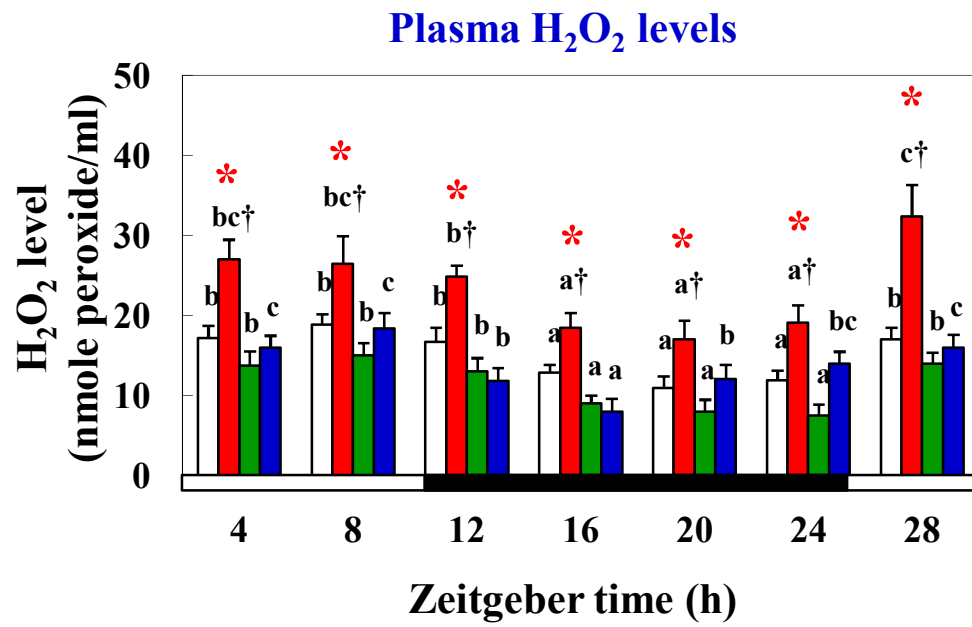
CAT activity levels in liver



CAT activity levels in liver (Melatonin injection)

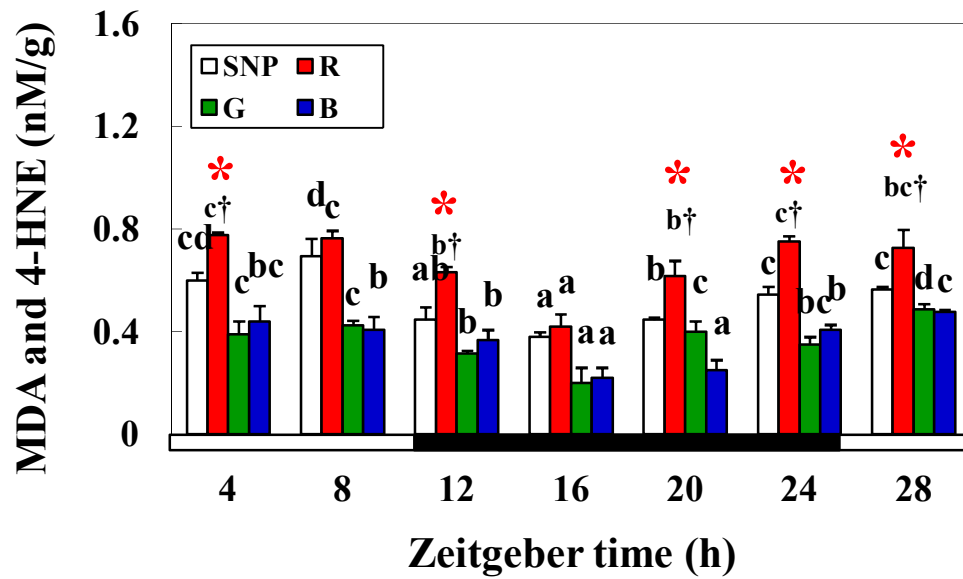


Plasma H₂O₂ levels under LEDs

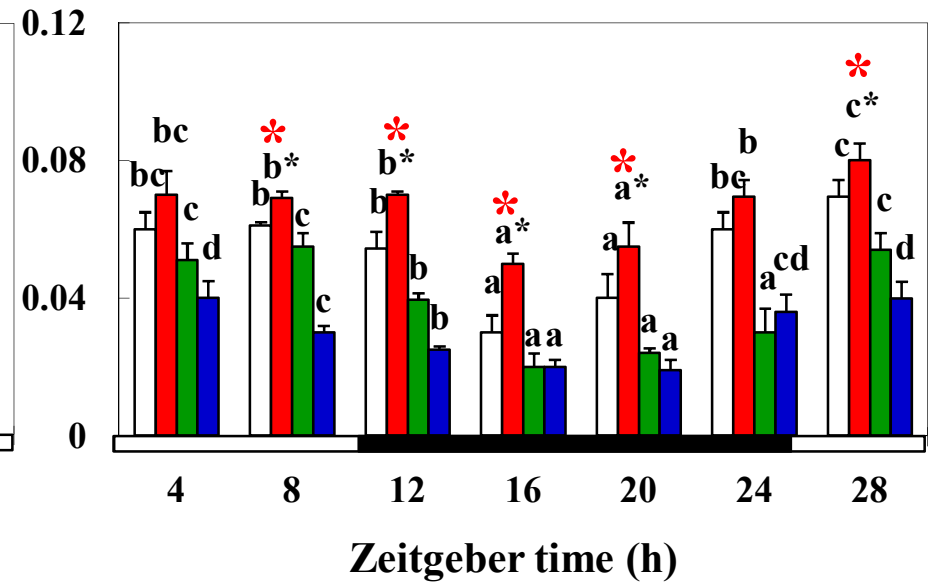


LPO levels under LEDs

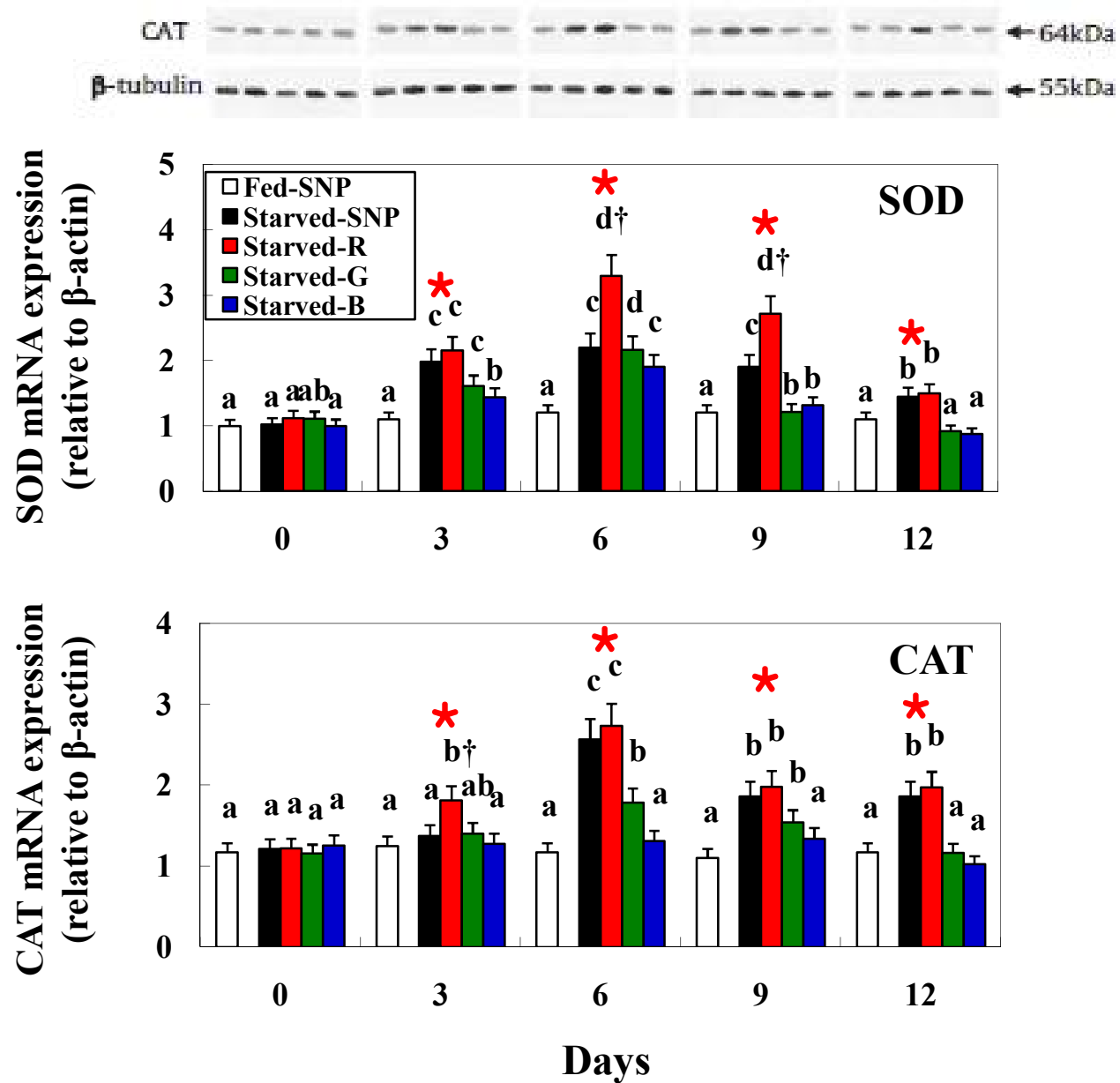
LPO levels



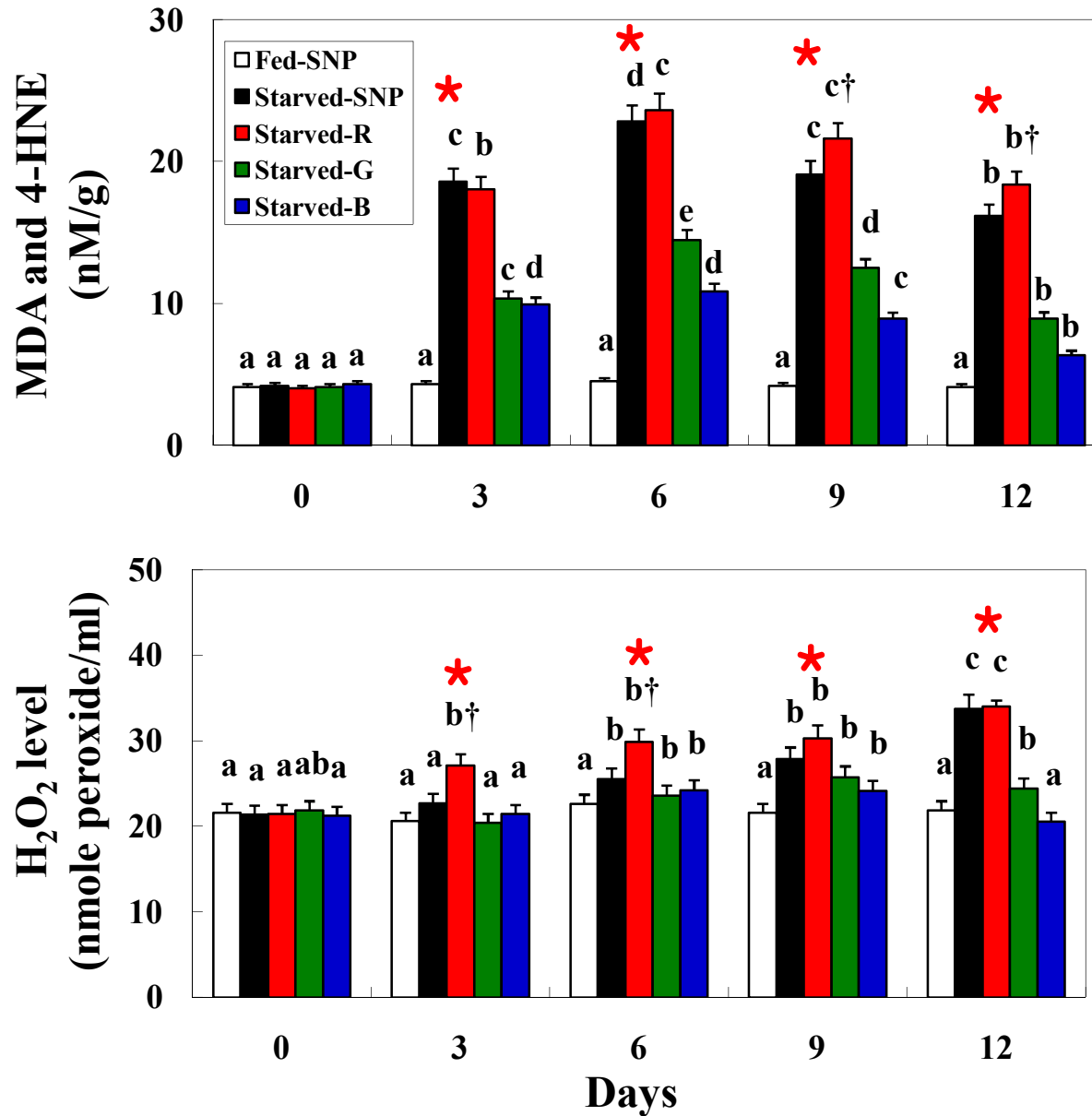
LPO levels (Melatonin injection)



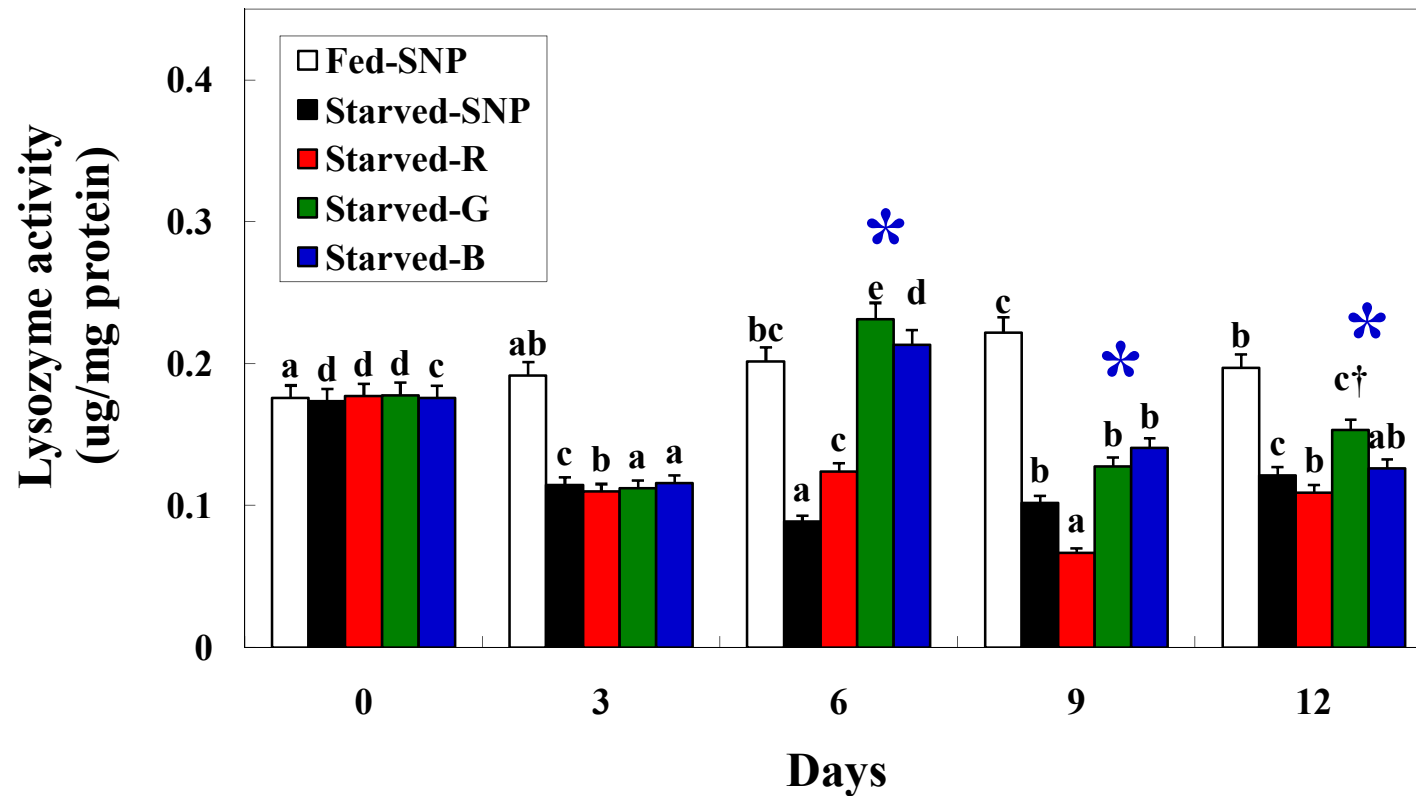
Expression of SOD and CAT during starvation under LEDs



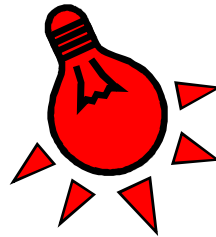
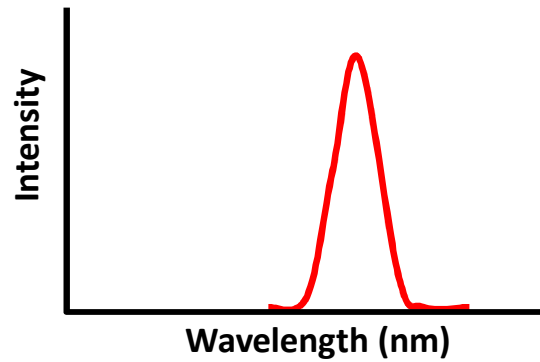
Levels of LPO and H₂O₂ during starvation under LEDs



Activity of lysozyme during starvation under LEDs



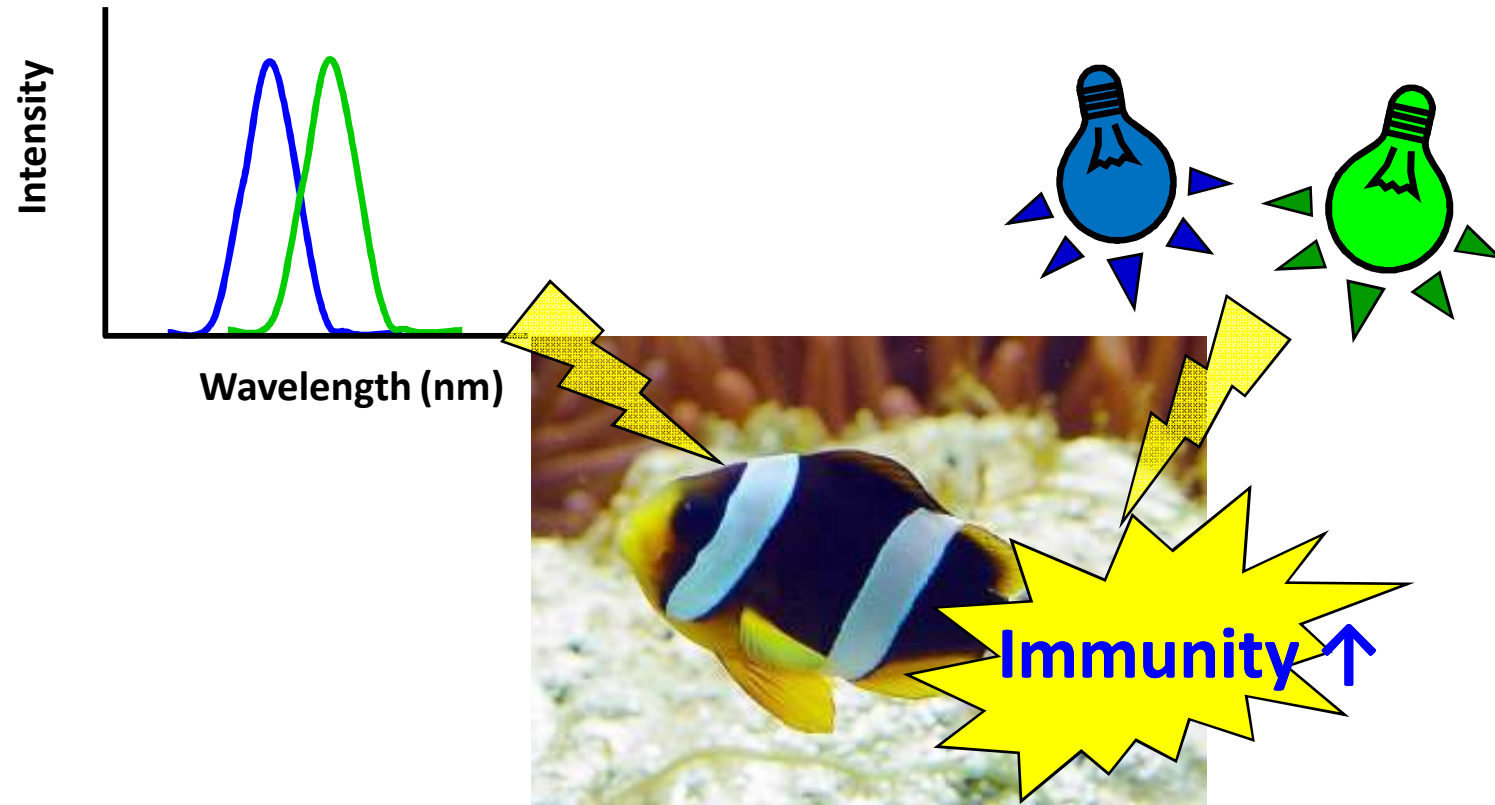
Negative results



Oxidative stress ↑

- **Red** wavelengths **LED** is a factor in inducing oxidative stress
- Light is known as a factor that affects various physiological changes in fish

Positive results



- Short wavelength light inhibit starvation-induced oxidative stress
- **Green** and **blue** wavelengths effectively enhance **immune function**

Chapter 4



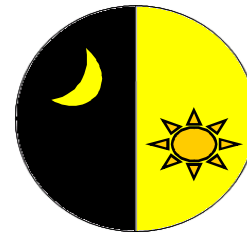
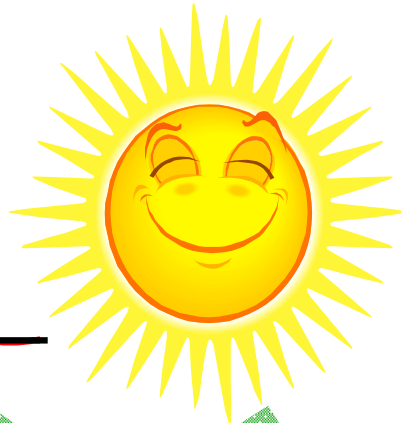
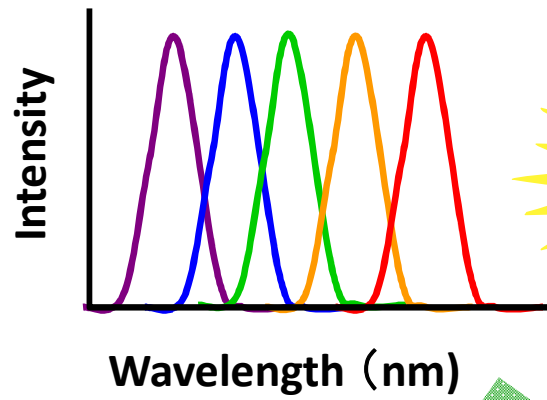
Effect of **LED** light spectra on **sex maturation**

Light spectrums

Light intensity

Light-dark cycle

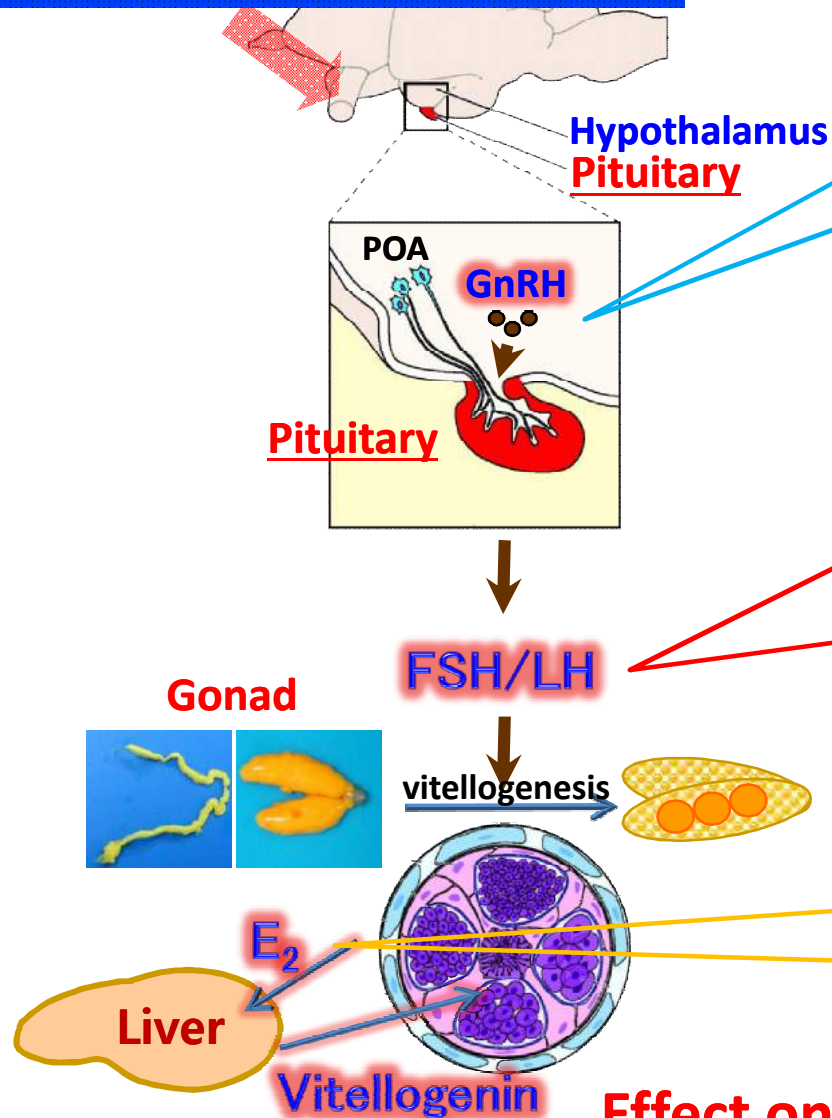
Temperature



Sex maturation

Hypothalamus-pituitary-gonad (HPG) axis

Various environmental factor



Gonadotropin-releasing hormone (GnRH)

- 15 GnRH isoforms
- Stimulate the gonadotropin (GTH) secretion in pituitary

Gonadotropin (GTH)

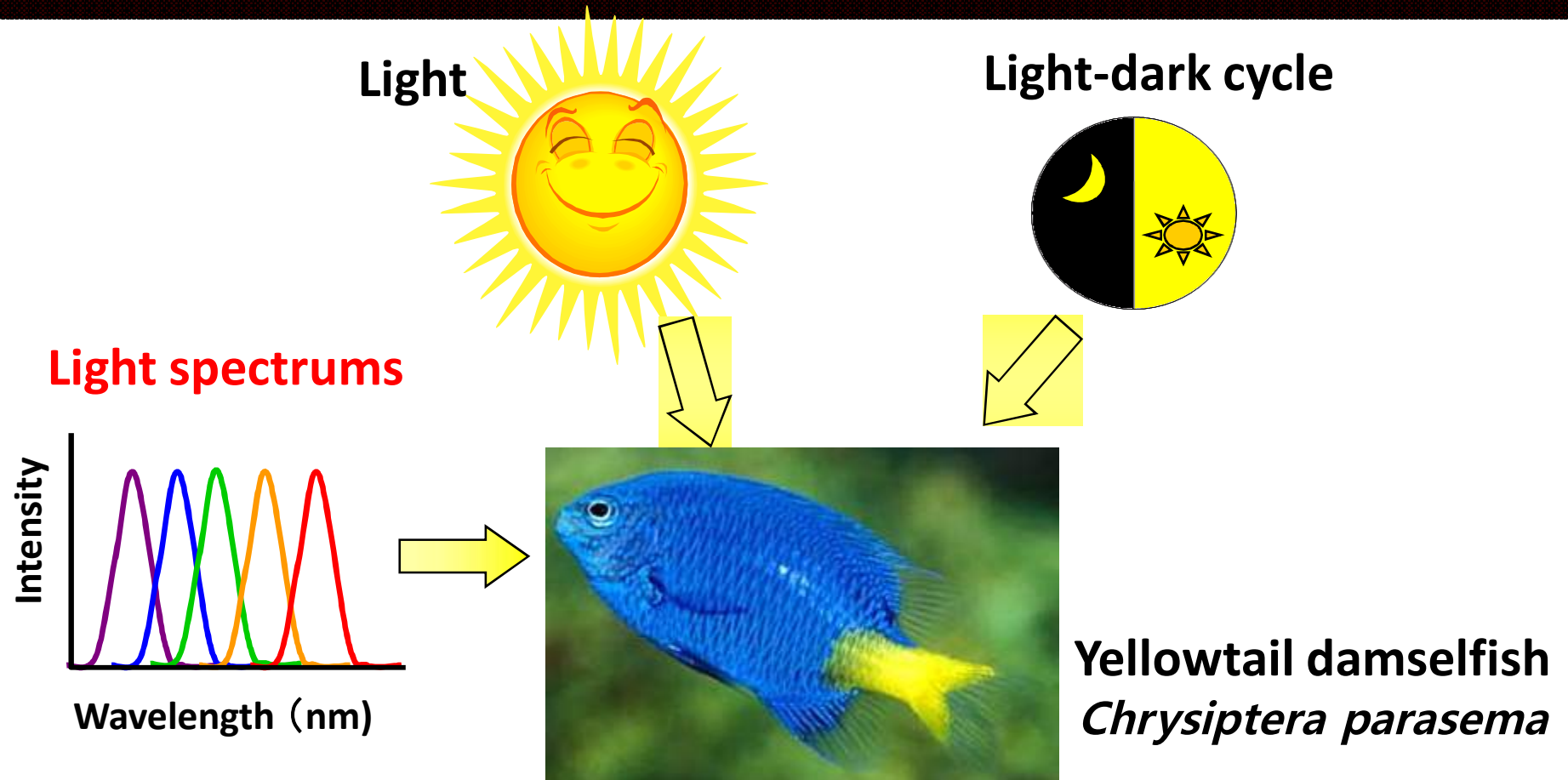
- α and β subunit
- Follicle stimulating hormone (FSH)
- Luteinizing hormone (LH)
- Stimulate gonadal steroid hormone (estrogen, progesterone, testosterone)
- Gametogenesis, ovulation, spawning

Estrogen receptor (ER)

- ER α , ER β 1 and ER β 2
- Bind to 17 β -estradiol (E₂)
- Regulate gonadal maturity

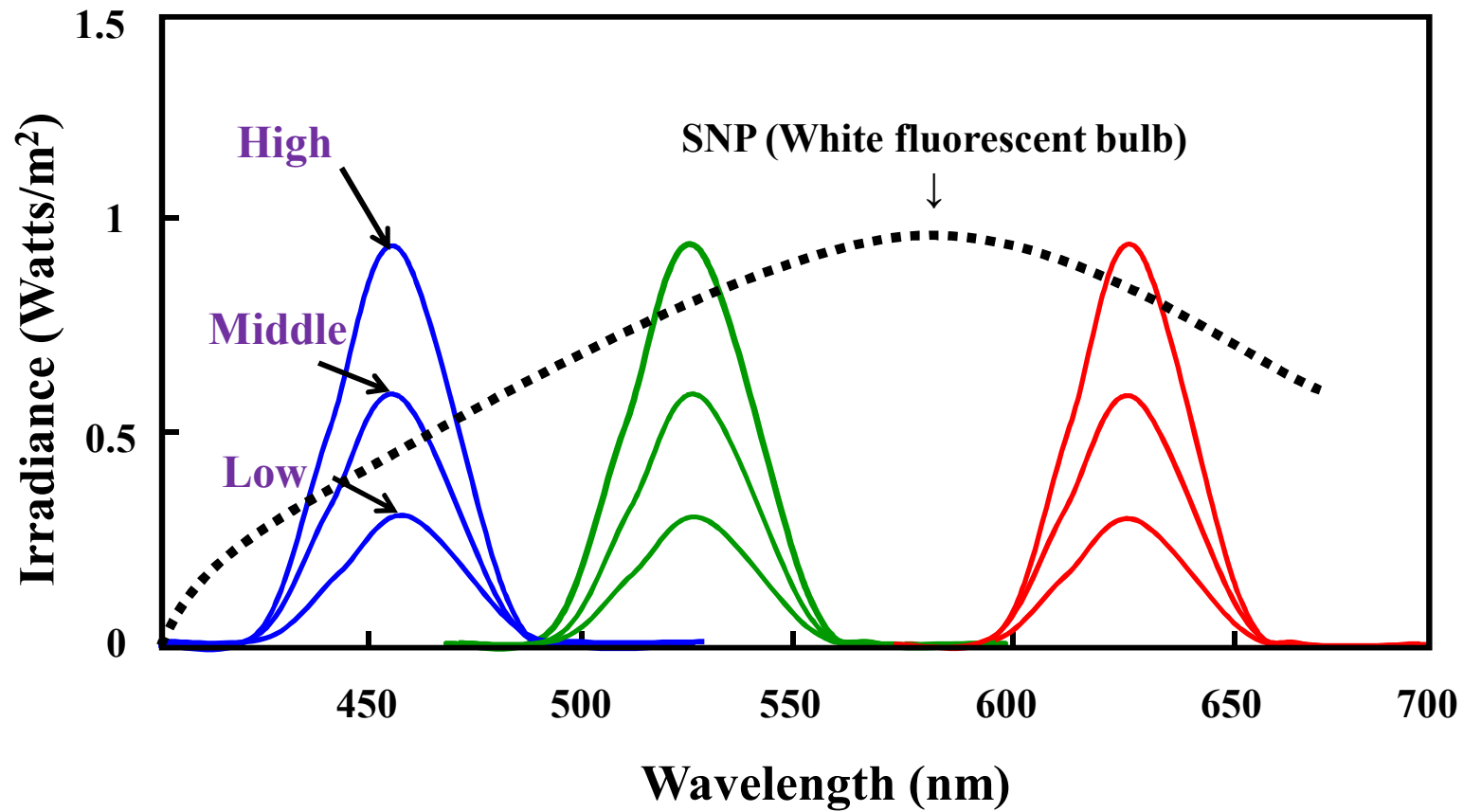
Effect on gonadal development and maturation

Purpose

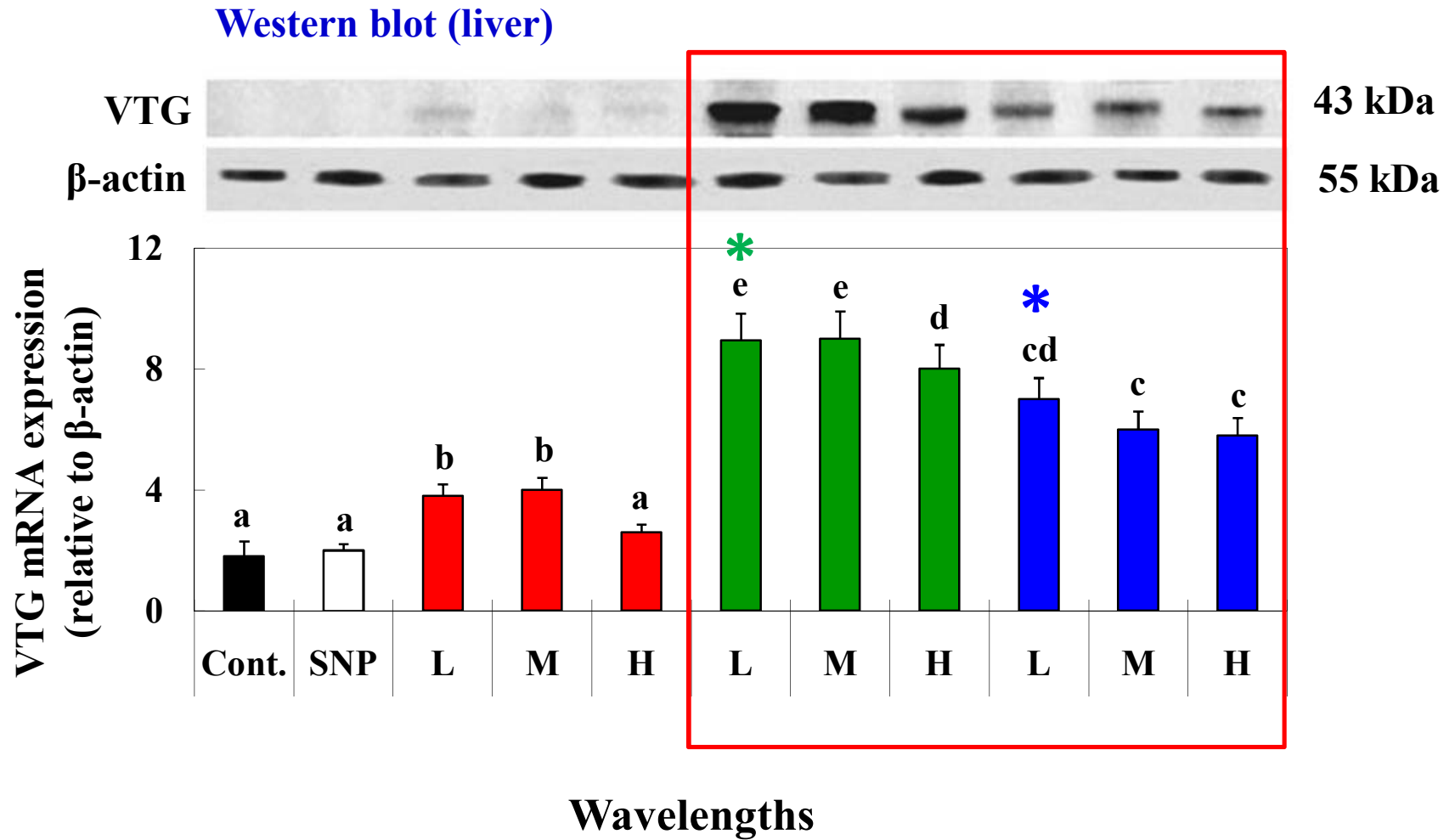


Effects of exposure to different **LED** spectra and
Intensities on **ovarian maturation**

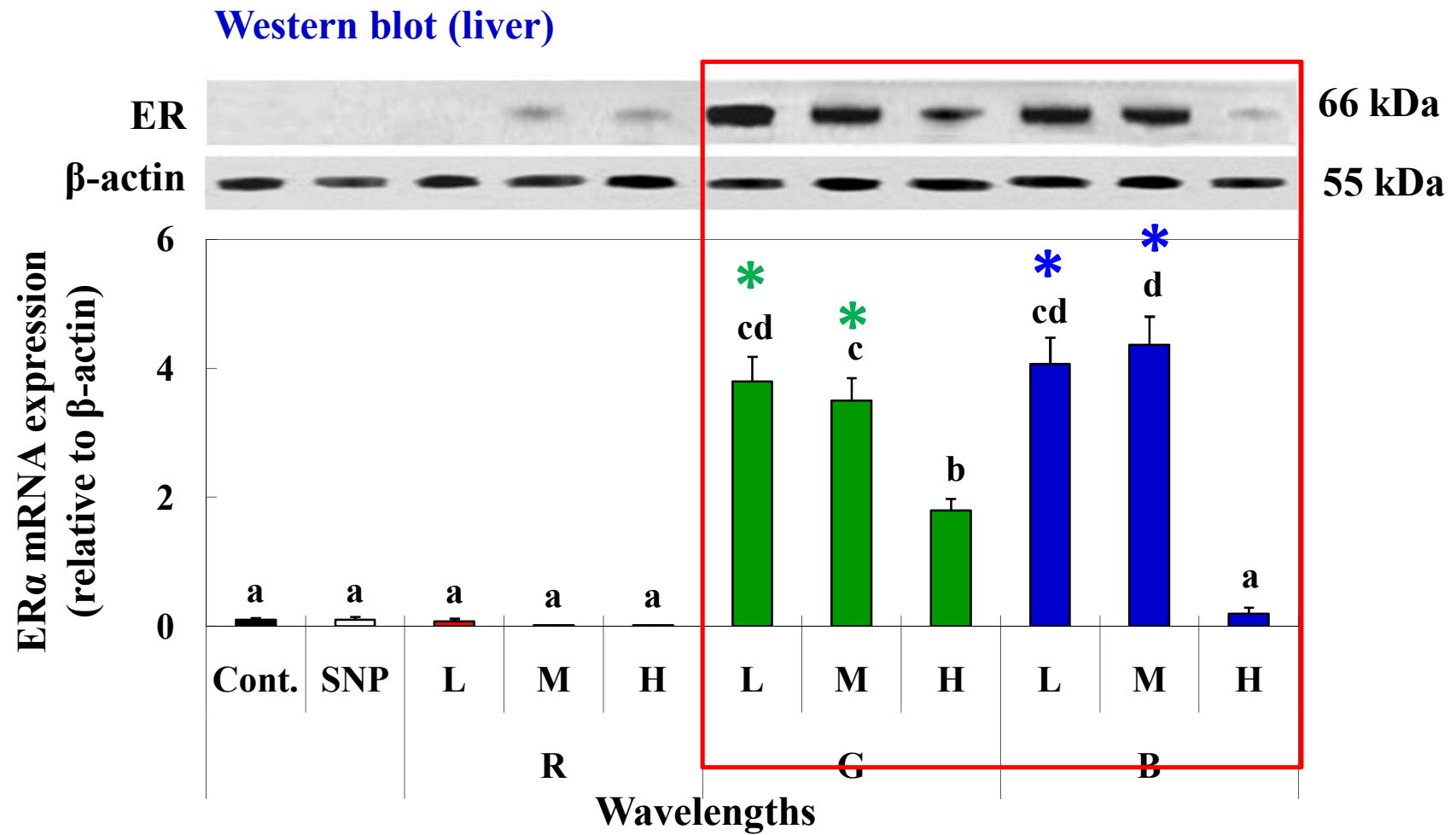
Spectral profiles of LED used in this experiment



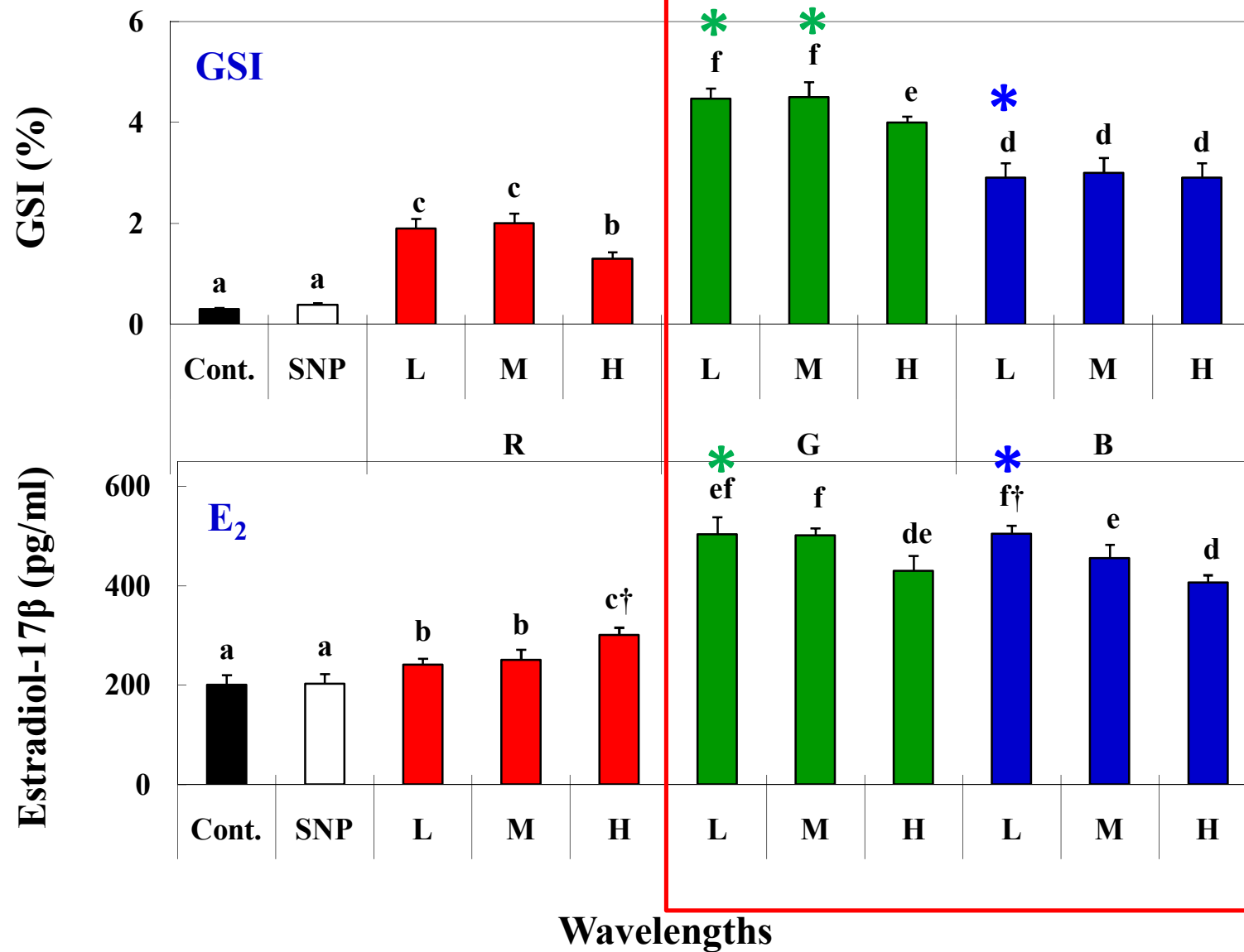
Vitellogenin expression under LEDs



Estrogen receptor expression under LEDs

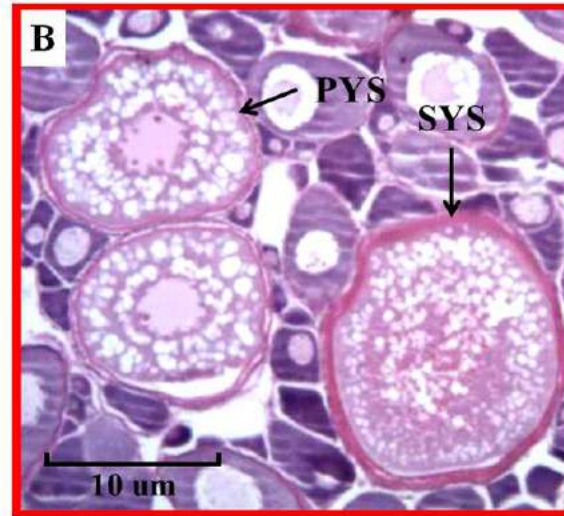
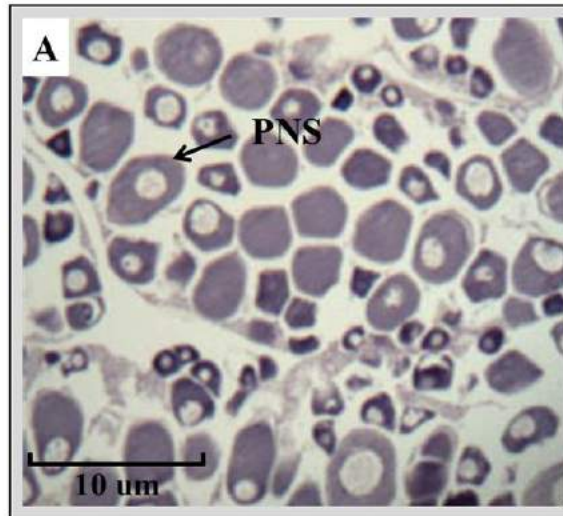


Changes in the GSI and E₂ levels under LEDs



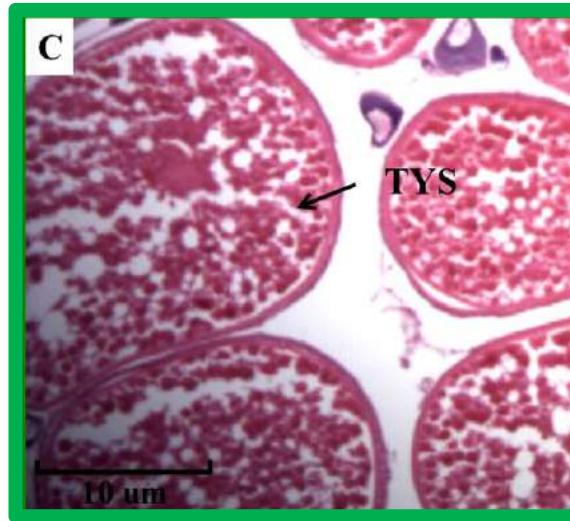
Ovary histology under LEDs

Control

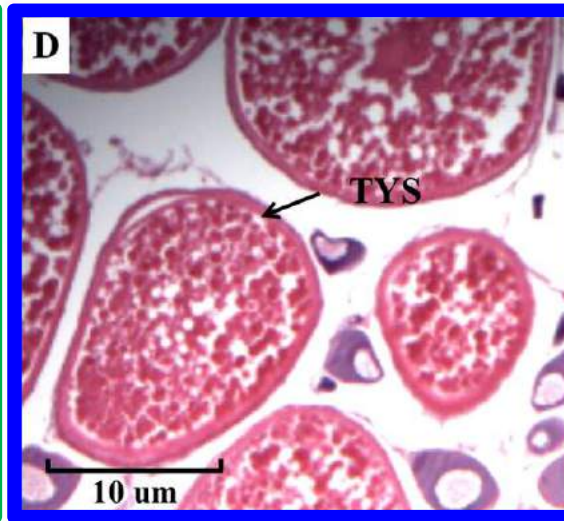


Red LED

 Green LED

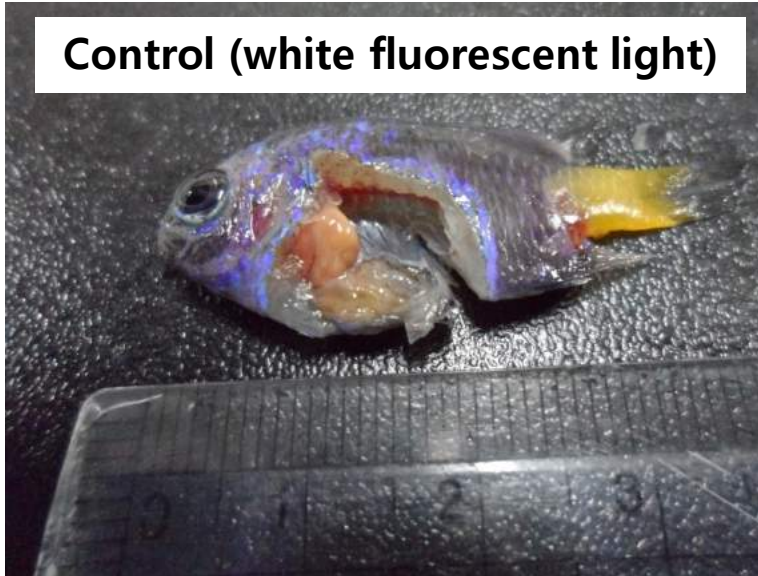


 Blue LED

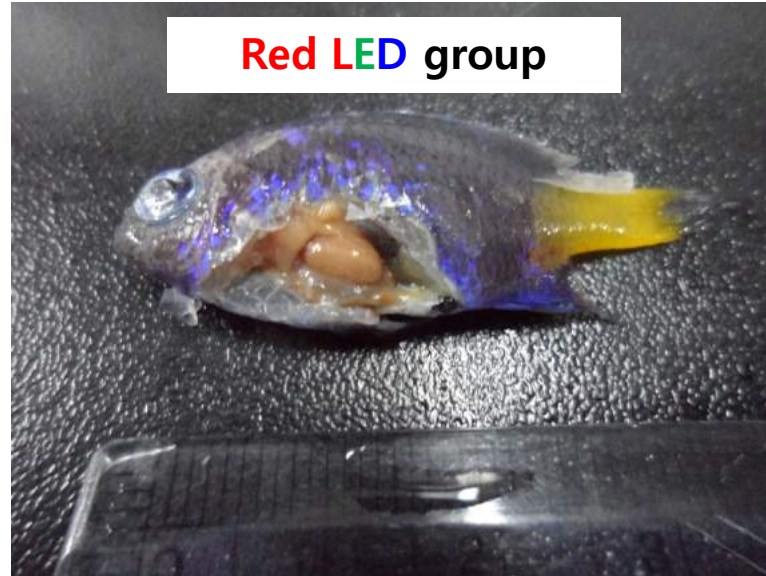


Ovary maturation under LEDs

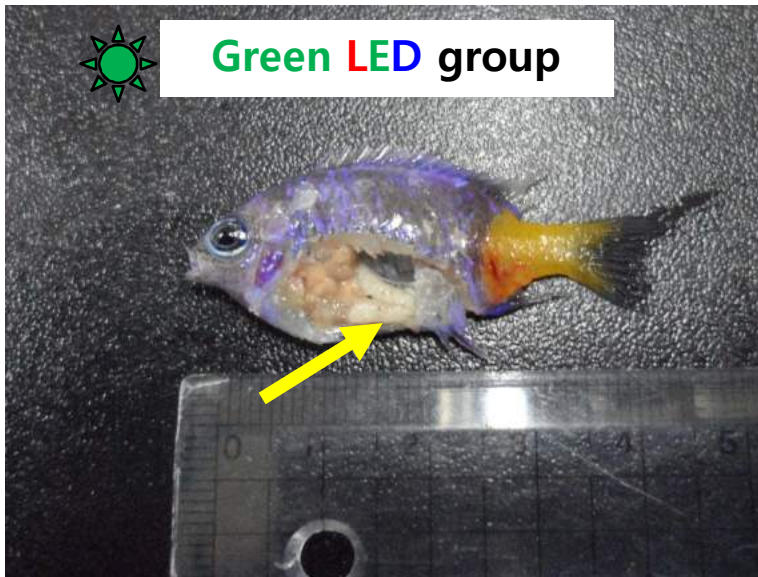
Control (white fluorescent light)



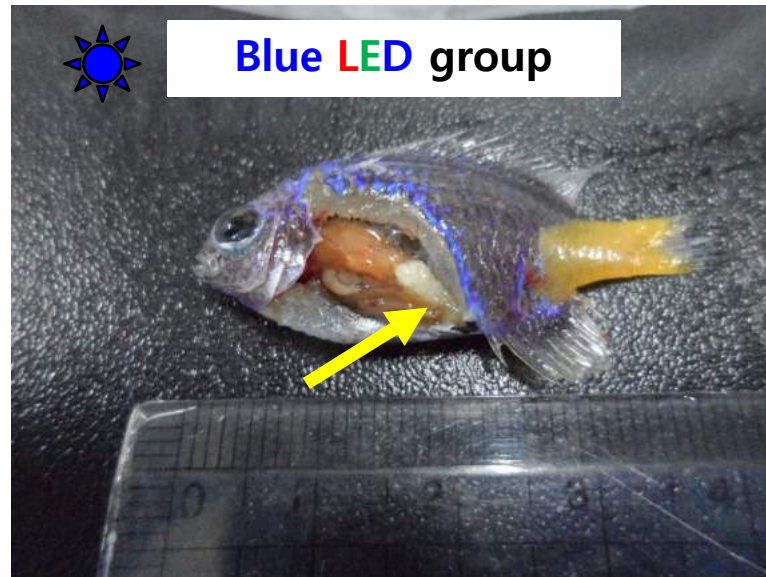
Red LED group



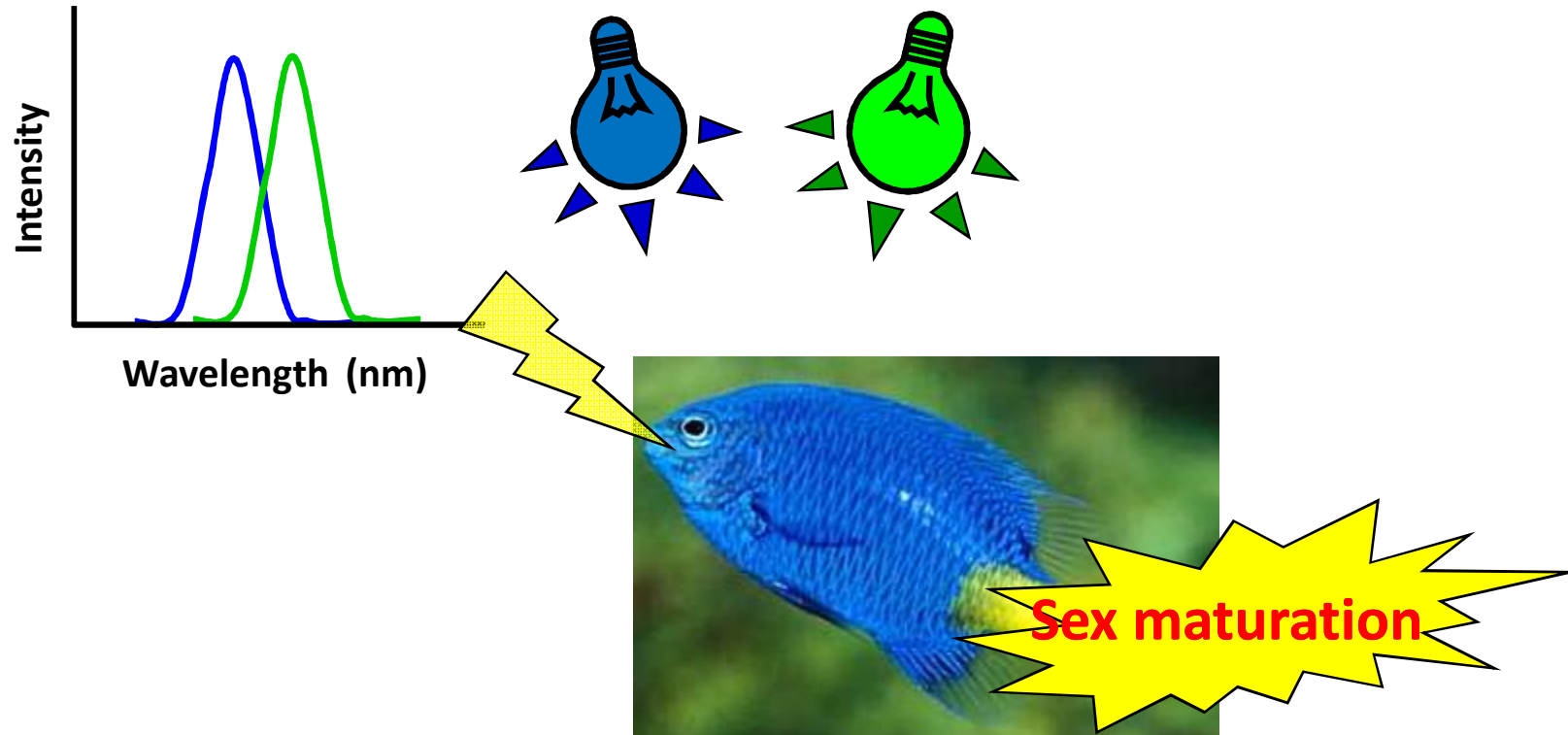
Green LED group



Blue LED group



Result



- Short wavelength light enhance gonadal development
- Low intensity **LEDs** lighting significantly increased maturation
- **Green** and **blue** wavelengths **LEDs** would be valuable by reproductive ability

Chapter 5



**Effect of LED light spectra
on sex change**

Sex differentiation in fish



Protogynous hermaphrodite fish



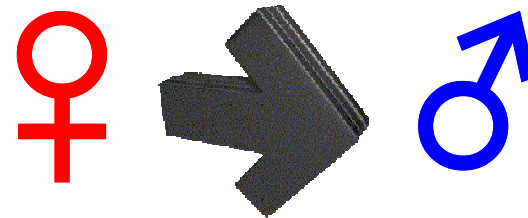
Bluestreak cleaner wrasse



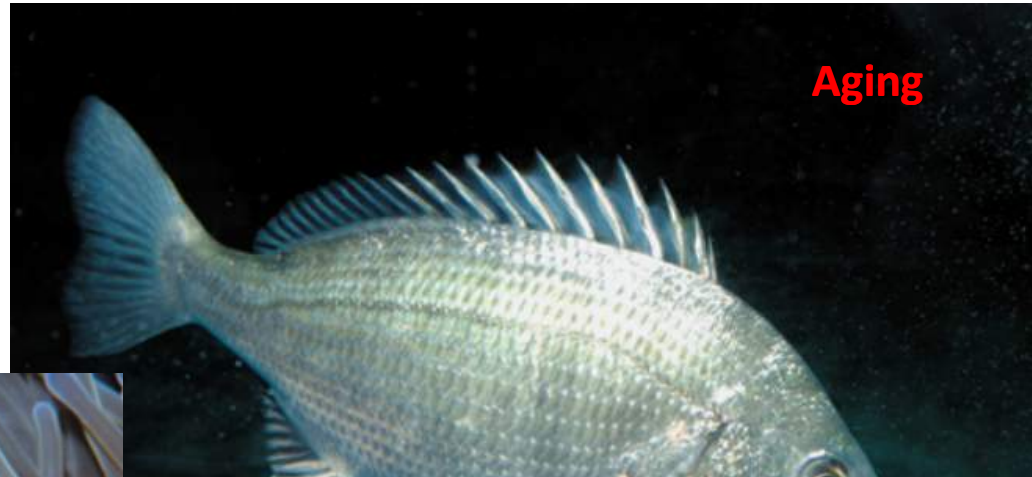
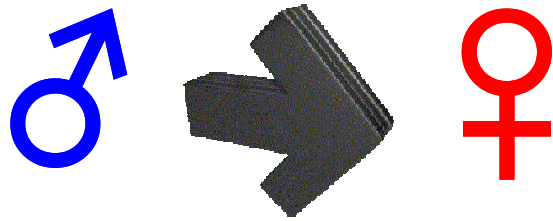
Multicolorfin rainbowfish



Grouper



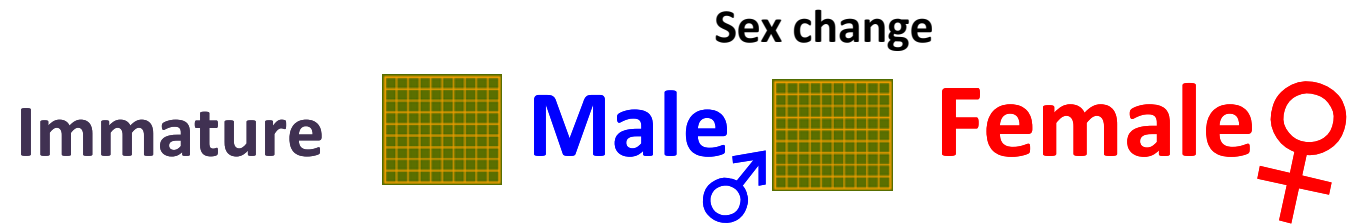
Protandrous hermaphrodites fish



Clownfish

Cinnamon clownfish

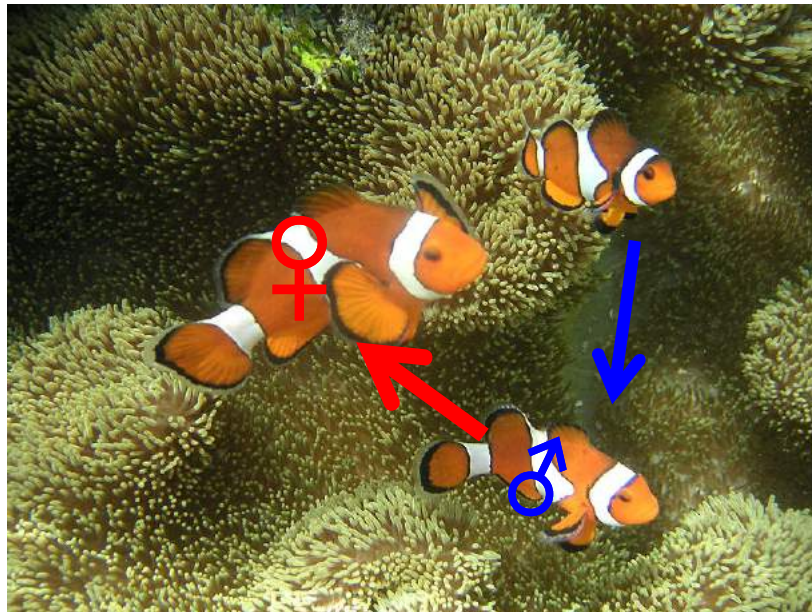
✓ *Protandrous hermaphrodites fish*



- ❖ *Amphiprion melanopus*
- ❖ Order: Perciformes
- ❖ Family: Pomacentridae
- ❖ Popular ornamental seawater fish

Sex differentiation of cinnamon clownfish

Social ranking in the group controls the sex



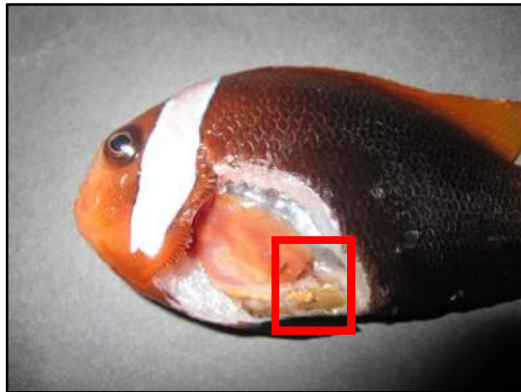
		Ranking
Group	Female ♀	1
	Male ♂	2
	Immature fish	3

- If a dominant female dies or is absent
 - the **male partner** undergoes a sex change to become a female

→ *Good model for studying the mechanism of sex change*

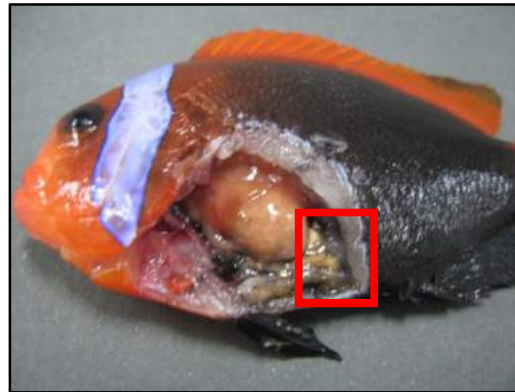
Gonadal development during sex change

Male

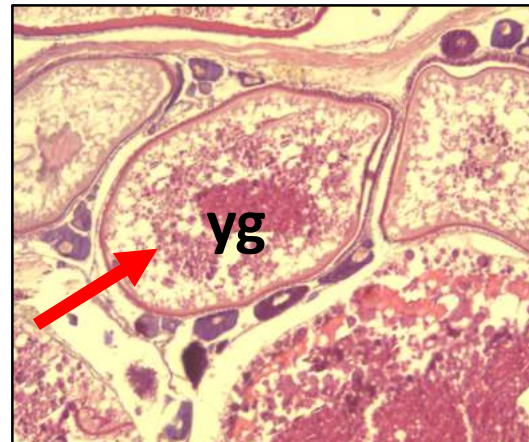
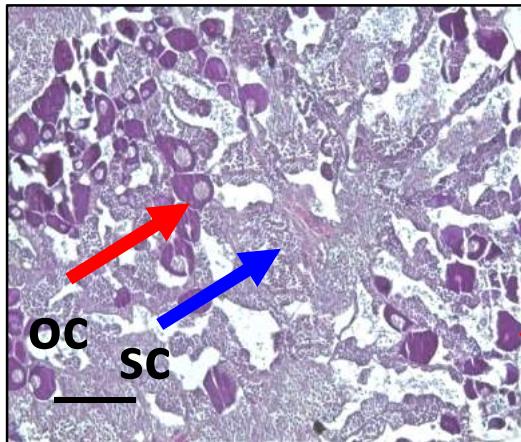
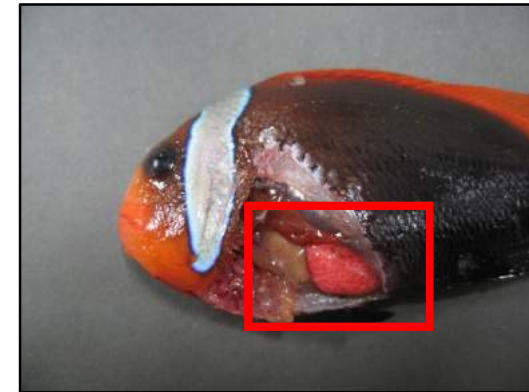


Male

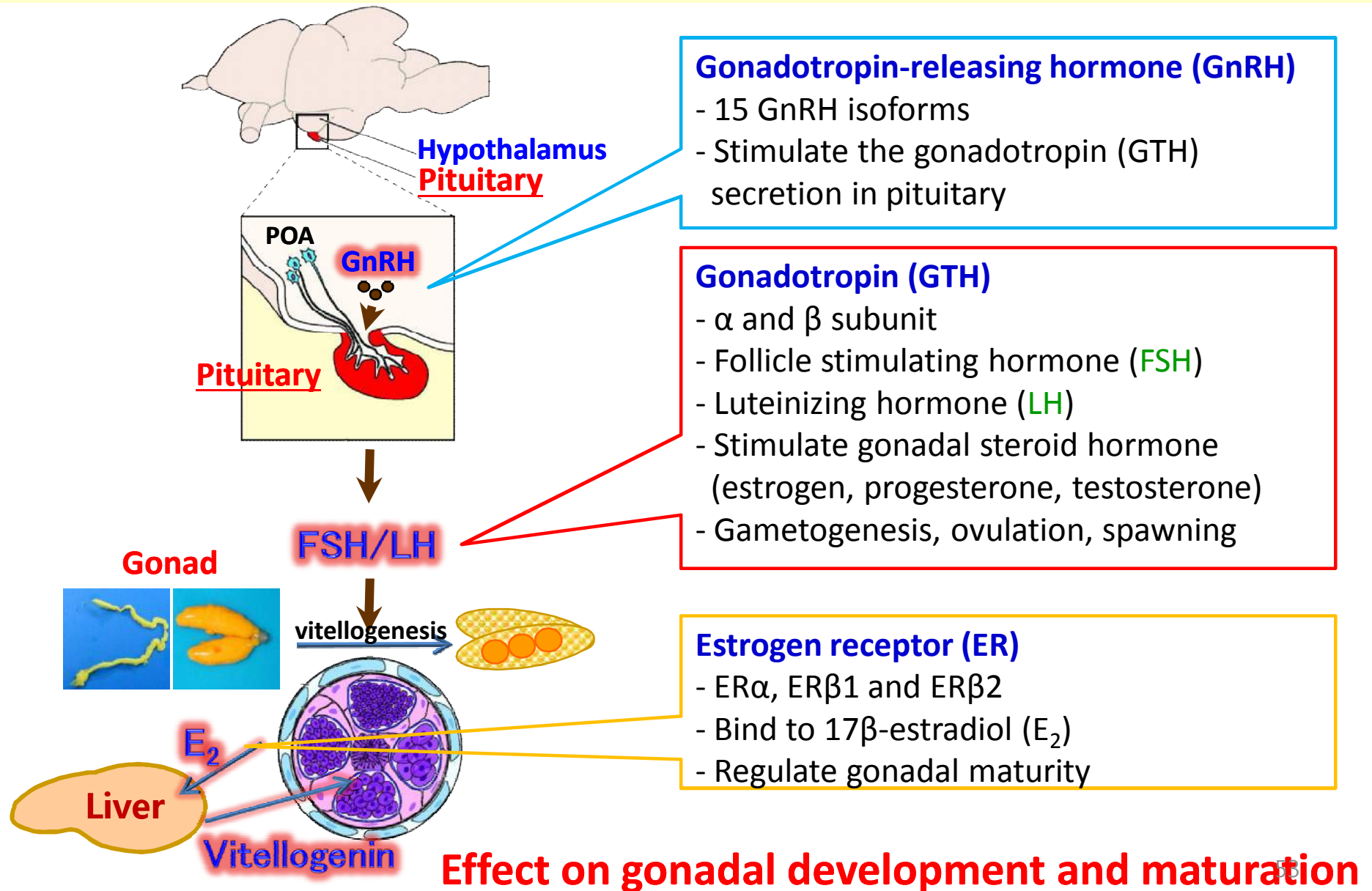
after removal of the female



Female

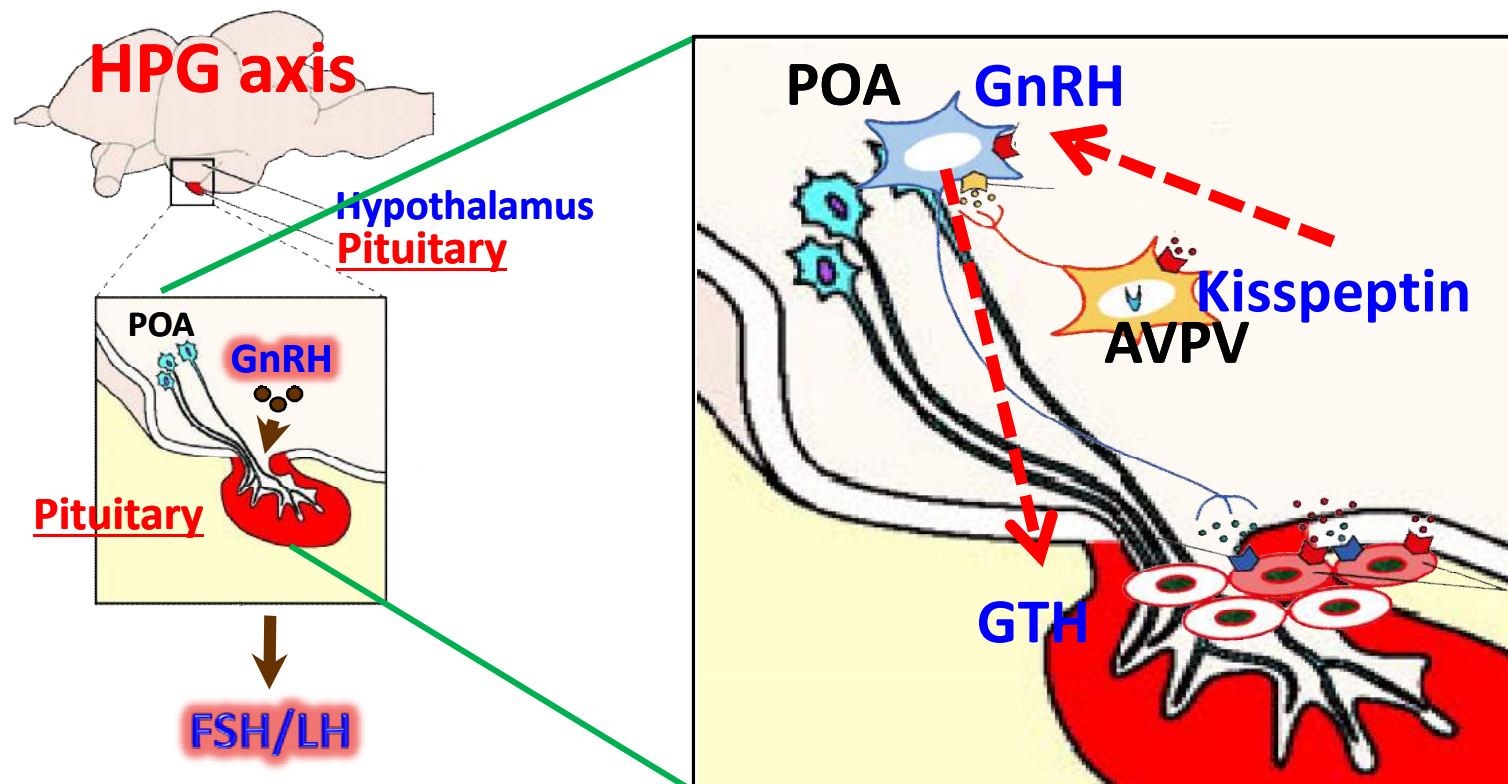


Hypothalamus-pituitary-gonad (HPG) axis



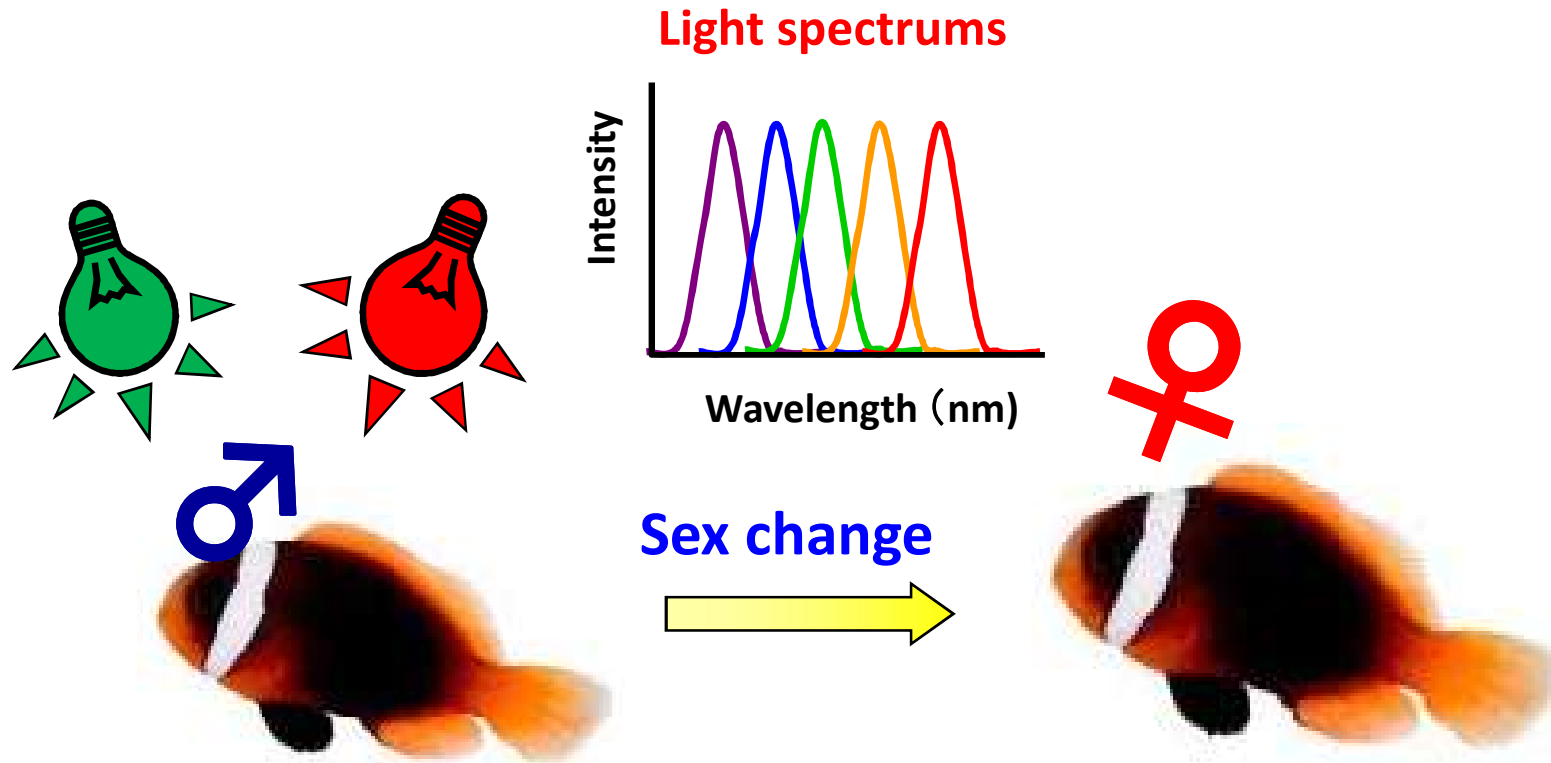
Kisspeptin

- **Neuropeptide**, located in **POA** of the **hypothalamus**
- Regulates **sexual differentiation** and **sex maturation**
- Central regulation of **the HPG axis** regulates in sexual maturation



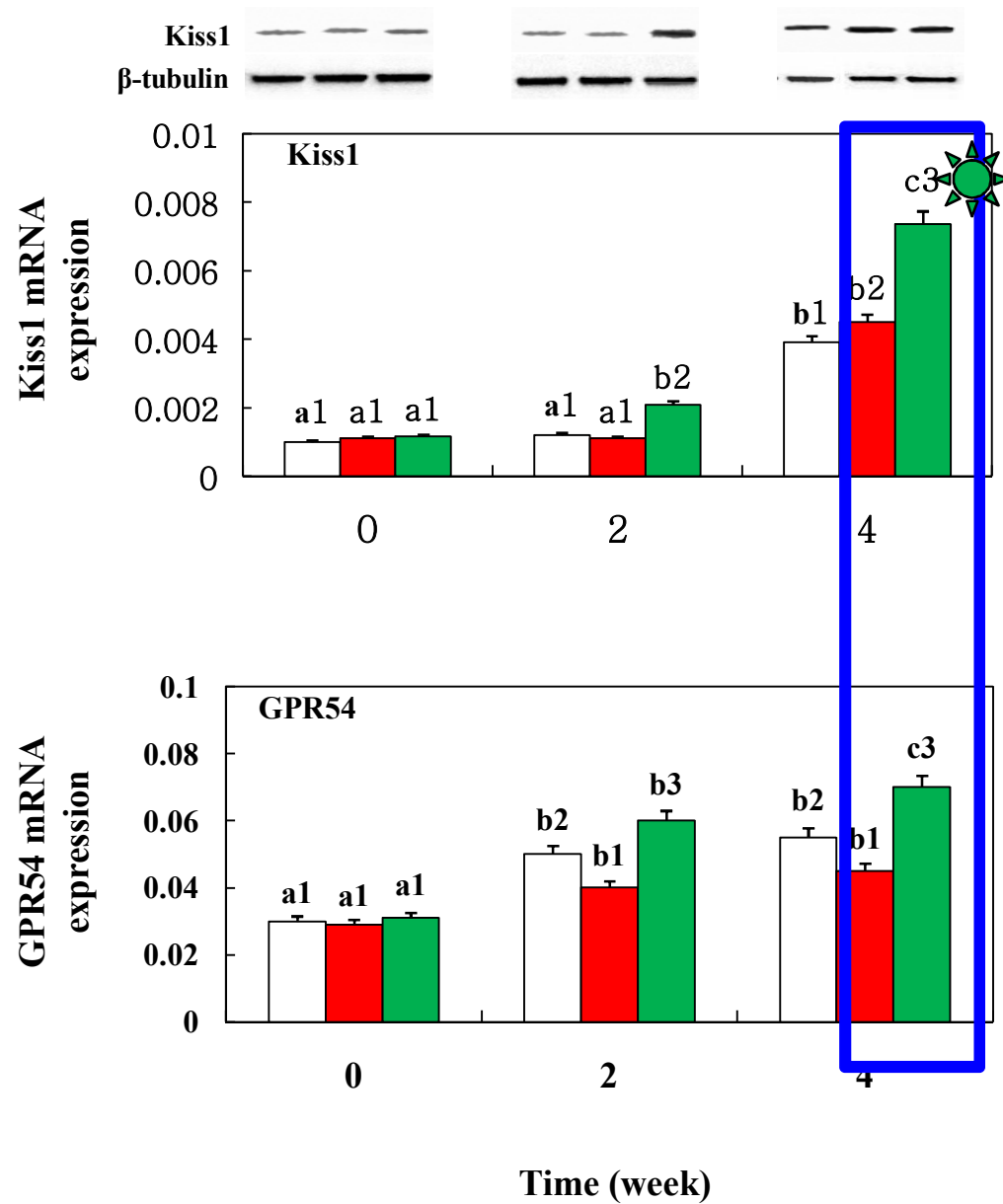
AVPV; anteroventral periventricular nucleus
POA; pre-optic area

Purpose

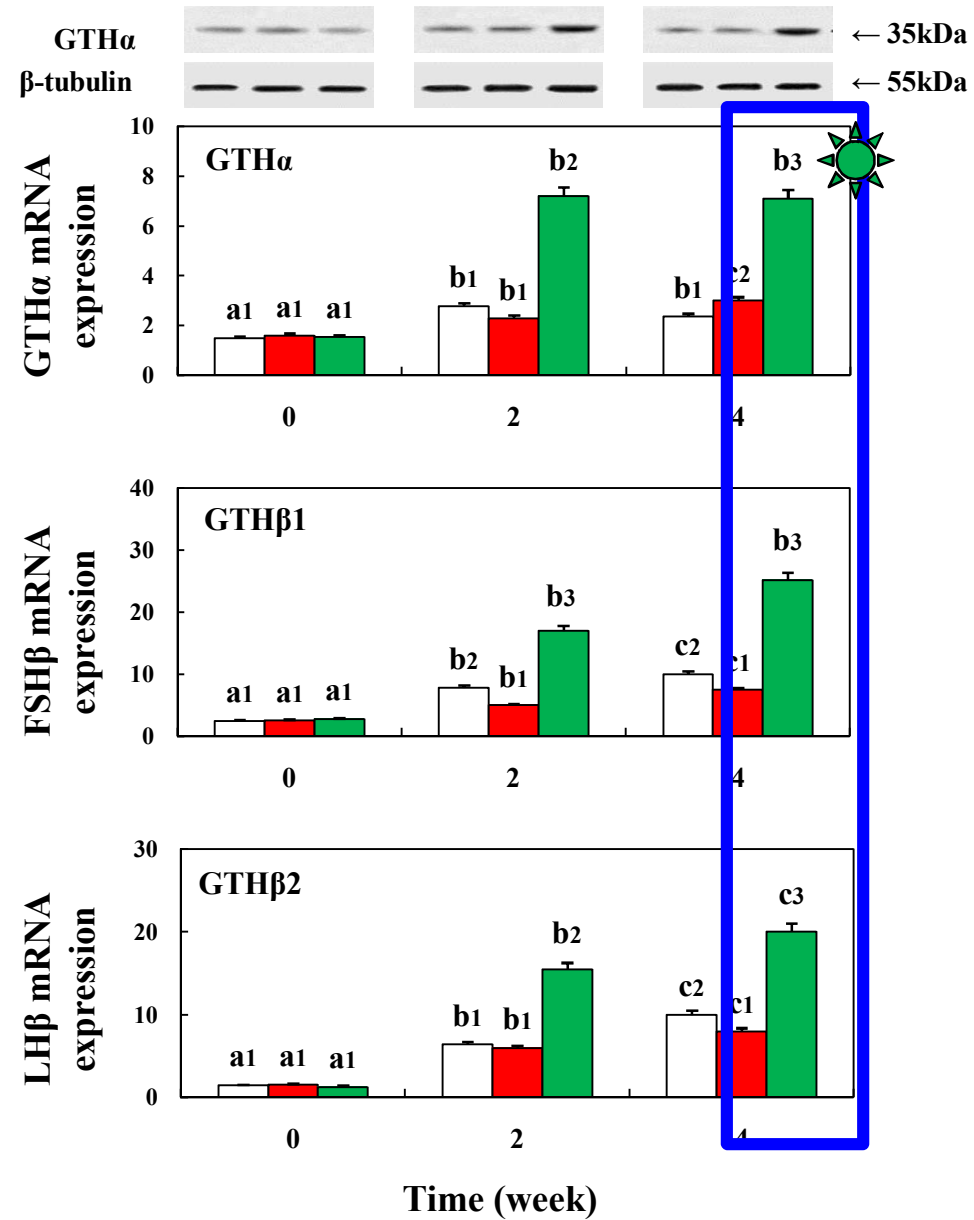


Effects of exposure to **LED** spectra (**red** and **green**)
on **sex change** in cinnamon clownfish

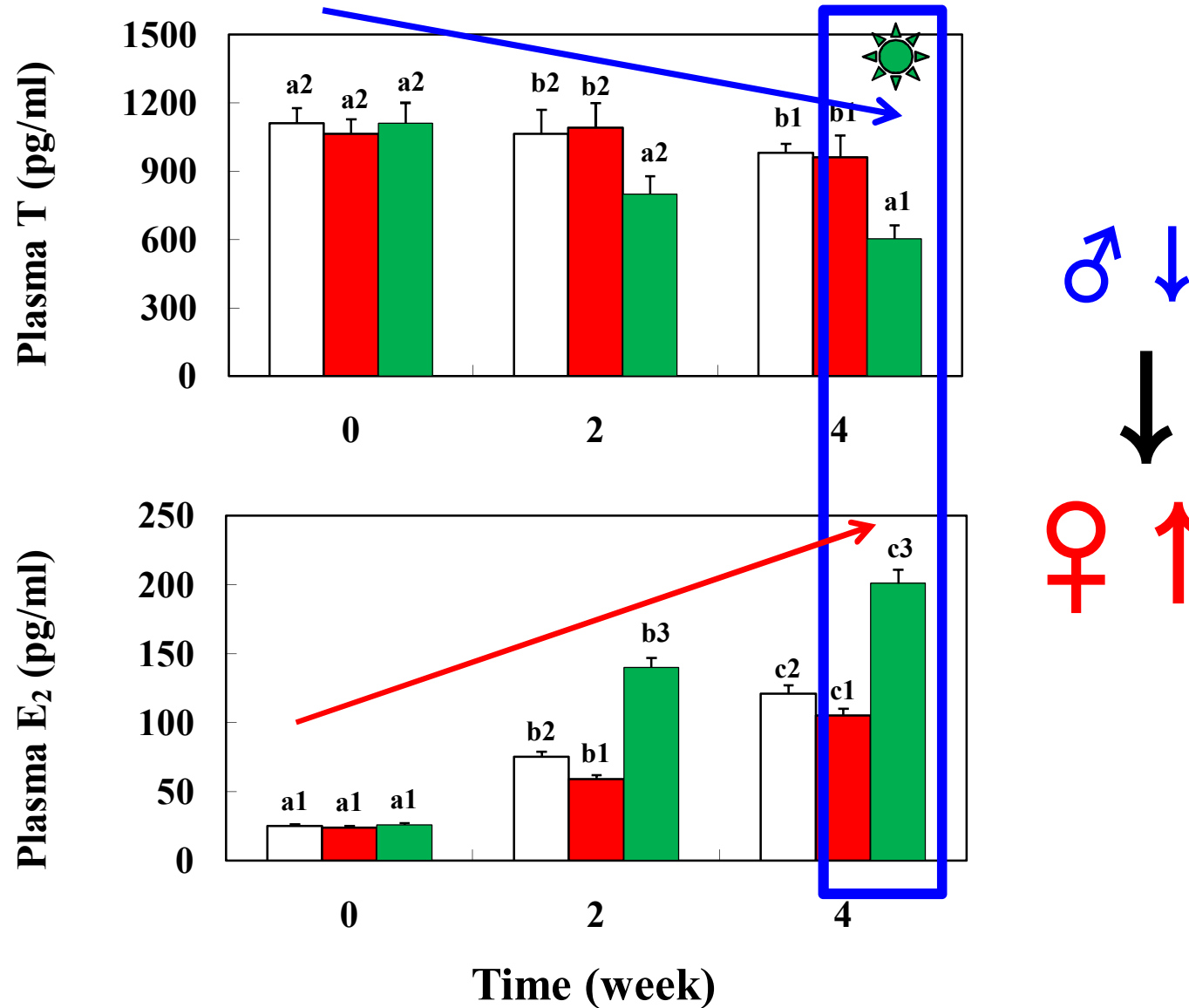
Kiss expression levels under LEDs in male fish



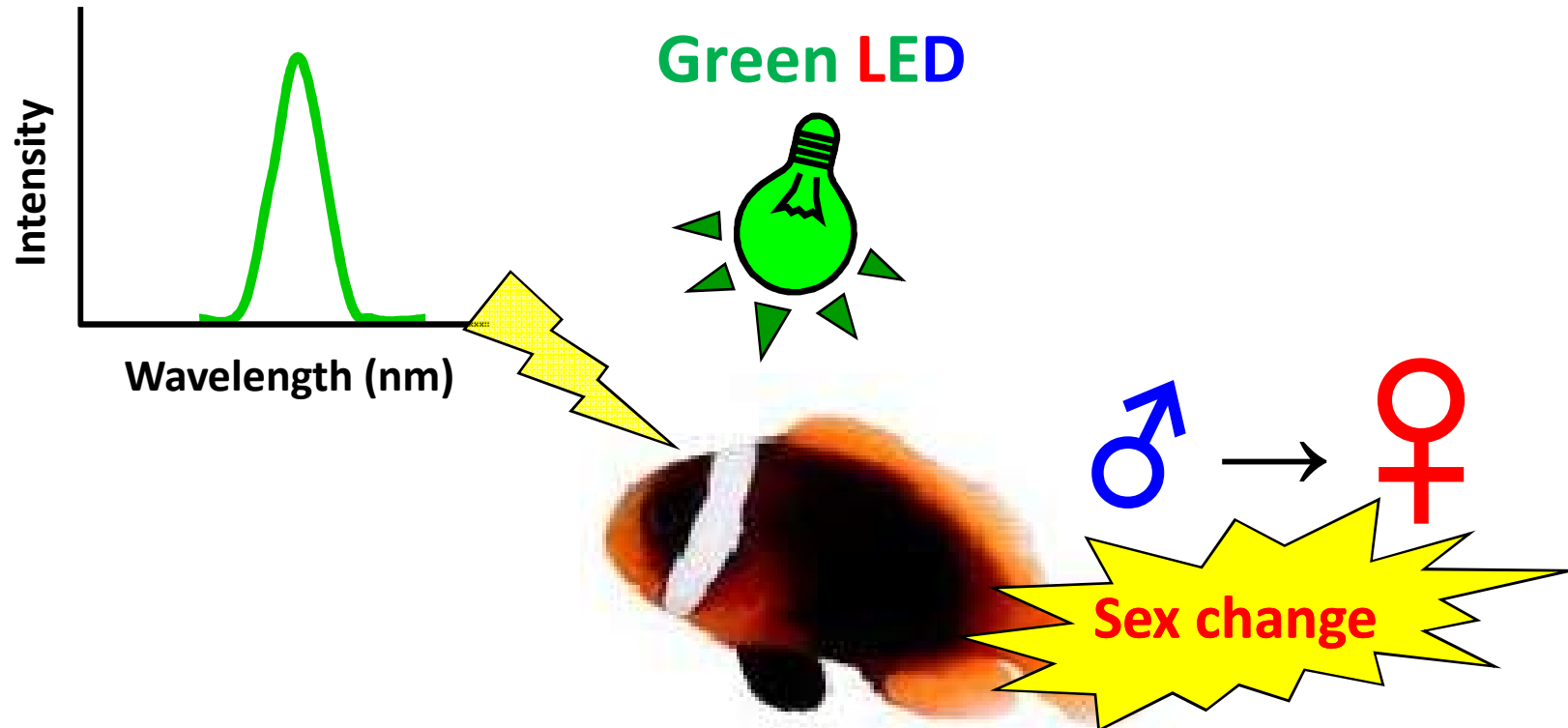
GTH expression levels under LEDs



Plasma T and E₂ levels under LEDs



Result



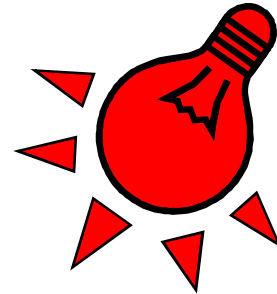
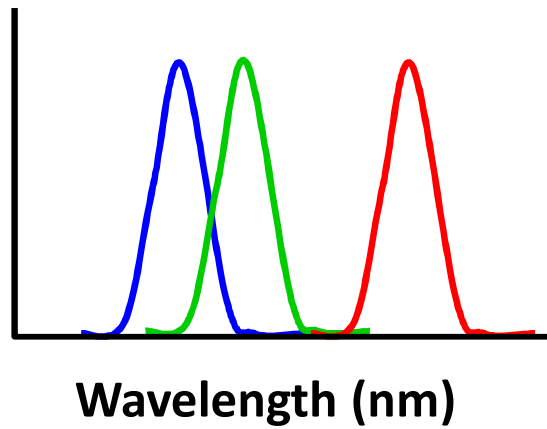
- **Green** wavelengths **LEDs** lighting significantly increased the expressions of Kisspeptin and GTHs, increase plasma levels of E_2
- Short wavelengths **LEDs** would be valuable by Induced gonadal development, sex maturation, and sex change from male to female

Chapter 6



Conclusion

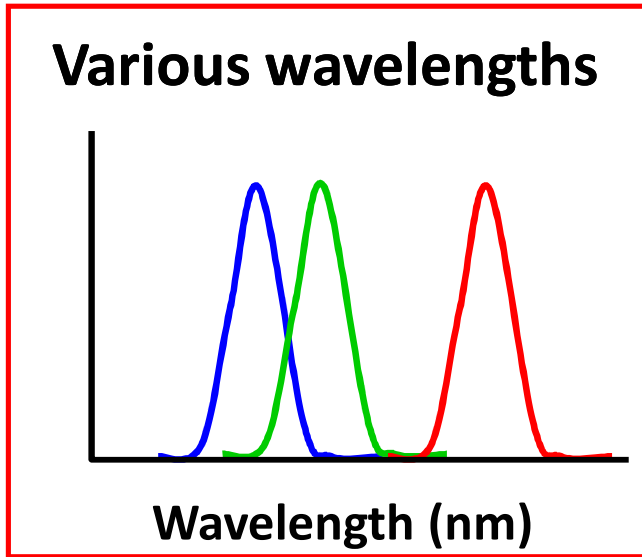
Various wavelengths



Oxidative stress ↑

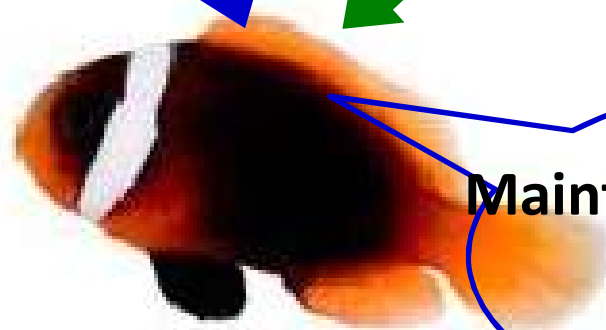
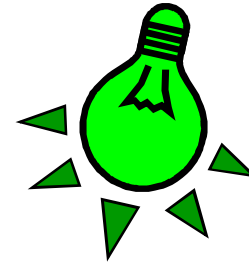
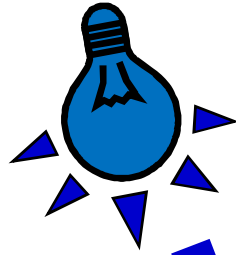
Red LED (long wavelength)

inhibits circadian rhythm and induces oxidative stress

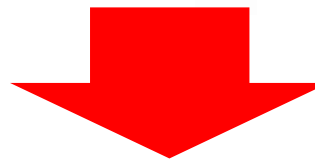


Blue LED

Green LED

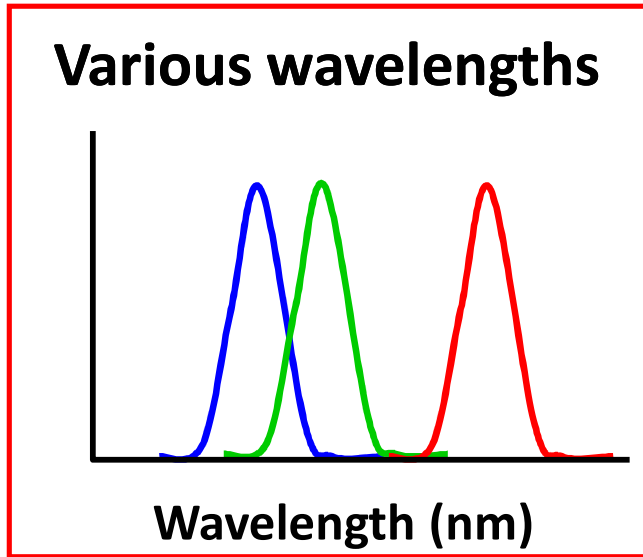


Maintained circadian rhythm
antioxidant ↑
immunity ↑



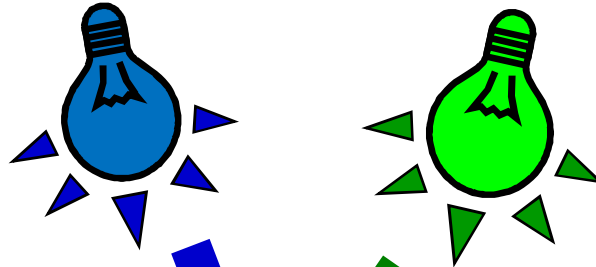
Blue and green LEDs (short wavelengths)

maintain circadian rhythm and immunity

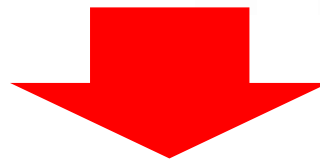


Blue LED

Green LED



Sex maturation ↑
Sex change ↑



Blue and green LEDs (short wavelengths)

enhance sex maturation and induce sex change

Boys can become girls.

**Thanks for your
attention**