TRANSCRIPTOMICS EXPLORES MOLECULAR FACTORS INVOLVED IN *Penaeus monodon* OVARIAN MATURATION

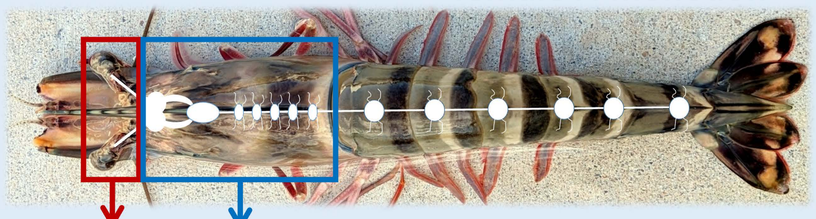
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Background

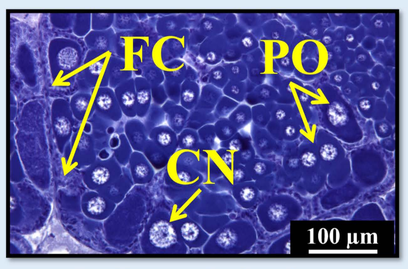
- ✓ The black tiger prawn *P. monodon* is one of the world's most commercially valuable farmed crustaceans [1].
- ✓ Despite advances in its aquaculture, the mechanisms regulating ovarian maturation and spawning in captivity remain poorly understood.
- ✓ Gonad-inhibiting hormone, the key inhibitor of ovarian maturation, is primarily produced in the eyestalk, making eyestalk ablation a common practice in aquaculture.
- ✓ Eyestalk ablation is an invasive procedure, highlighting the need for ablation-free and animal welfare-conscious spawning technologies in *P. monodon* aquaculture [2].

Methodology

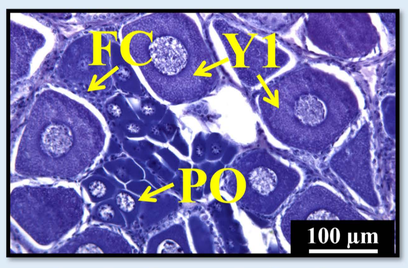


Results

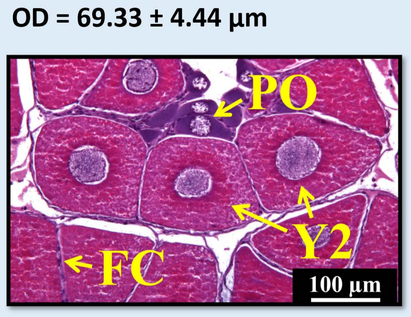
Ovarian stages & female grouping



Previtellogenic (PV)
GSI = 1.57 ± 0.23
OD = 69.33 ± 4.44 µm



Early vitellogenic (EV)
GSI = 3.13 ± 0.69
OD = 149.06 ± 5.07 µm

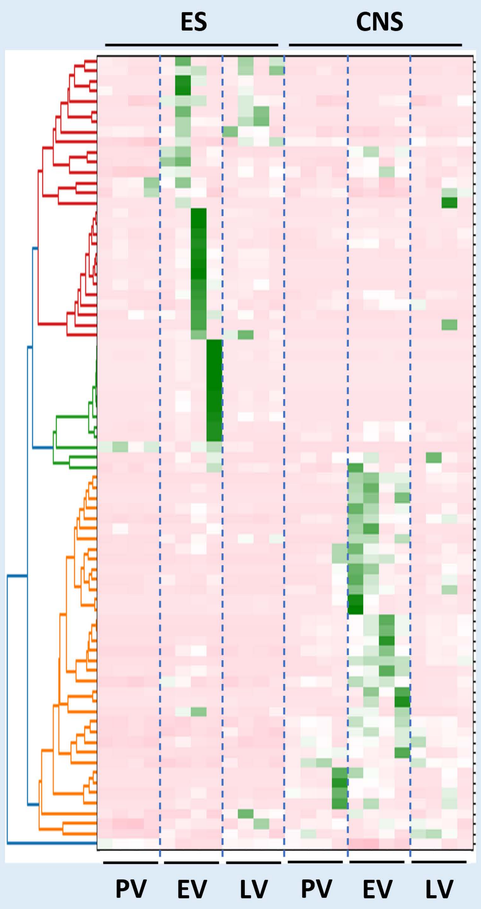


Late vitellogenic (LV)
GSI = 5.52 ± 0.27
OD = 169.50 ± 1.43 µm

PO: perinucleolar oocytes
FC: follicle cells
CN: chromatin nucleolus
Y1: microlecithal oocytes
Y2: yolky oocytes

Values are mean ± SEM
N = 4/stage or group

NPs expression profile



78 NPs were expressed in both tissues across three stages. Of those,

- **21 were upregulated in ES**
- **24 were upregulated in CNS**

Higher expressions in ES

- Crustacean hyperglycemic hormones
- Moult-inhibiting hormone – 1
- Pigment-dispersing hormones
- Crustacean female sex hormones

Higher expressions in CNS

- Allatostatins
- Bursicons
- Calcitonins
- FLRFamide
- Glycoprotein beta – 5
- Neuropeptide – F1
- WxxxRamide/nalisin

Higher expressions in both tissues

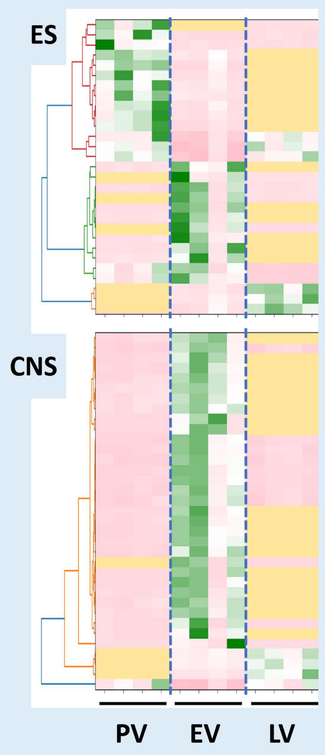
- Allatostatin – C
- Red pigment-concentrating hormone
- HIGSLYamide-partial
- Glycoprotein alpha – 2

Note for heatmaps
ES: eyestalk
CNS: central nervous system
Expression across each row
Lower — Higher
-2 -1 0 1 2 3 4
Individual across each column
N = 4/stage; Not detected

Red – inhibitory
Green – stimulatory
According to literature [3 – 10]

- ✓ **Several NPs, both stimulatory & inhibitory, upregulated during early vitellogenesis.**
- ✓ **The known stimulatory and inhibitory NPs showed similar expression patterns in the CNS.**

DEGs expression profile



Heatmaps visualising annotated DEGs; 29 in ES & 34 in CNS

The following genes, most consistent across the samples, were associated with synapse formation, neuromuscular junction, terminal bouton, molecular binding, and structure formation GO terms.

Upregulated in CNS at EV

- Peripheral-type benzodiazepine receptor-associated protein 1
- Plasma membrane calcium-transporting ATPase 3
- Synapse-associated protein
- Ras-related protein Rab6
- RIMS-binding protein 2
- Synapsin
- Fasciclin-2
- Complexin
- Synaptogyrin
- Synaptotagmin 1
- Proteoglycan cow

Upregulated in ES

- Protein hu-li tai shao **at PV**
- Glutaminase kidney isoform, mitochondrial **at PV & LV**

Upregulated in CNS at LV

- Ribosomal protein
- Glutaminase kidney isoform, mitochondrial

Insights from this study

1. **Glycoprotein alpha – 2, Pigment-dispersing hormone – 1C, Glycoprotein beta – 5, and Calcitonin, upregulated during early and late vitellogenesis, might have a role in ovarian maturation.**
2. **DEGs upregulated during early vitellogenesis could have a role in overcoming the inhibition of vitellogenesis in eyestalk-intact *P. monodon*, and may support NP and neurotransmitter release and neuronal signal transmission.**
3. **Early vitellogenesis in *P. monodon* may involve enhanced neuronal connectivity, synaptic signalling, and NPs secretion, supporting a broader neuroendocrine regulatory network controlling ovarian maturation.**
4. **Therefore, research to overcome the inhibition of vitellogenesis should consider multiple NPs and neuroendocrine factors, rather than focusing only on gonad/vitellogenesis-inhibiting hormone.**

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Project funding bodies
FRDC
AUSTRALIAN Prawn Farmers ASSOCIATION

Industry partner
tassal

Australian Government

World Aquaculture Singapore 2026 June 2 - 5