Sensory structure roles in odour-gated rheotaxis during navigation by the nudibranch mollusc *Tritonia diomedea*

Russell C. Wyeth[1] and Greg B. McCullagh
Biology, St. Francis Xavier University, Antigonish NS Canada | [1]rwyeth@stfx.ca;

Introduction

Our interest is the neuroethology of navigation, a behaviour performed by all motile animals. Many aquatic animals navigate using a combination of flow direction and odours to travel upstream (positive rheotaxis) or downstream (negative rheotaxis) depending respectively upon cues from either attractive or aversive navigational goals (1,2). This behaviour, odour-gated rheotaxis (OGR), is the optimal strategy because flow transports the odours from the odour source, and thus finding or avoiding the odour source is best accomplished by responding to the flow.

Tritonia diomedea has a number of characteristics that make them amenable for the neuroethological study of navigation (5). The sea slugs use OGR to navigate upstream in the presence of prey odour and downstream in the presence of predator odour (3,4). Odours are detected by the rhinophores. Upstream turns in the absence of odours depend on flow detection by the oral veil (4,6). However, the sense organs responsible for flow detection during OGR has not been tested.

Primary sense organs

Rhinophores
- **detect odours**
- **detect flow?**

Oral veil
- **detects flow?**

Goal: test roles of rhinophores and oral veil in flow detection during odour-gated rheotaxis.

►we are testing three possibilities (see Hypotheses and Predictions)

Methods

We applied localized streams of odour stimuli to one rhinophore and subsequently measured both turn angles and final headings of the animals from digital video of the slugs' responses. A total of 12 treatments were chosen to give different predicted results for each of our three hypotheses for how *Tritonia* navigates. In flowing water, slugs were tested only if they were crawling across the flow. Treatment orders were randomized, and both applied and analyzed blind to the odour treatment.

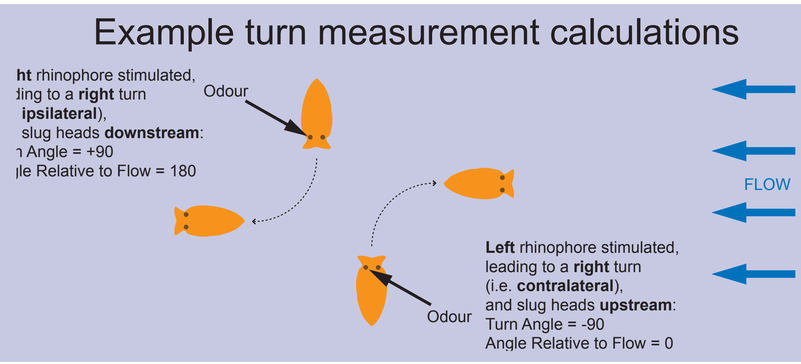
Treatments (2 x 2 x 3 = 12 combinations):

►2 bulk flow conditions: still or flowing water
►2 stimulus directions: odour applied medially or laterally (always to downstream rhinophore in flowing water)
►3 odour types: control seawater, prey, and predator)
Slugs (n = 18) were each tested under all treatment combinations .

Turn Measurements:

Turns were calculated from headings taken at the start and end of the odour application. Each turn was measured in two different ways:
1. Turn angles
   ipsilateral (positive values: 0° to +180°) or
   contralateral (negative values: 0° to -180°)
2. Angles relative to flow (measured at the end of the turn):
   positive values between upstream (0°) and downstream (180°)

Example turn measurement calculations



Hypotheses and Predictions

Three different hypotheses for how *Tritonia* navigates have different predicted turn responses to odour and flow stimulation.

1. Lateralized reflexive turns based on odour type

►strategy: turn towards prey odours and turn away from predator odours
►rhinophores detect odour; no flow detection
►turn direction depends on which rhinophore is stimulated
►turn responses similar in still and flowing water

2. Rheotactic turns based on odour flow and odour type

►strategy: turn upstream in presence of prey odours and downstream in presence of predator odours
►rhinophores detect flow and odours
►turn direction depends on direction rhinophore is stimulated
►turn responses similar in still and flowing water\

3. Rheotactic turns based on bulk flow and odour type

►strategy: turn upstream in presence of prey odours and downstream in presence of predator odours;
►oral veil detects bulk flow; rhinophores detect odours
►turn direction depends on bulk flow direction
►turn responses different in still and flowing water

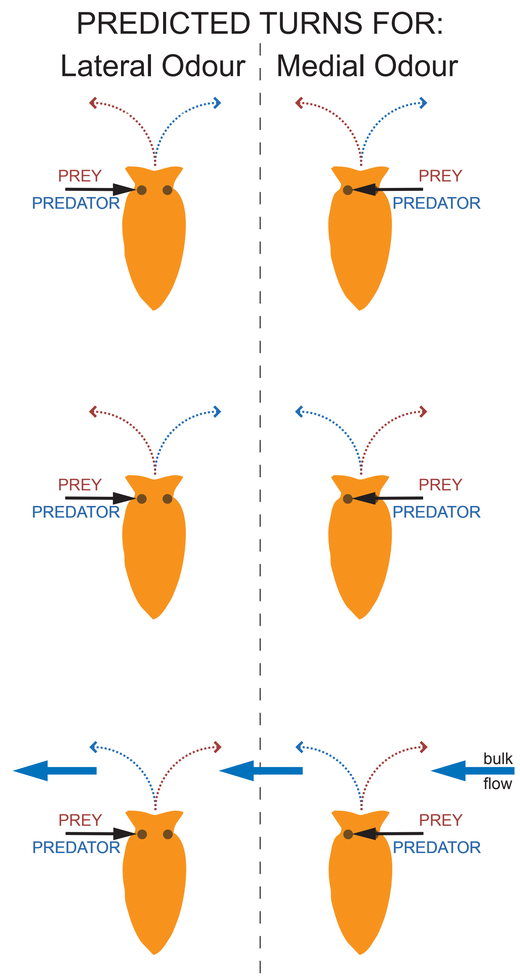


Fig 1. Turn Directions (Still Water)

Results

Turn responses were not significantly different between any of the odour stimuli in any of the treatments

In still water, slugs showed no consistent turn direction in response to odour stimulation (Fig 1).

In flowing water, slugs tended to turn contralaterally in all treatments, including controls (Fig 2).

In flowing water, slugs tended to have final headings facing upstream in all treatments, except medial stimulation with predator odour (Fig 3).

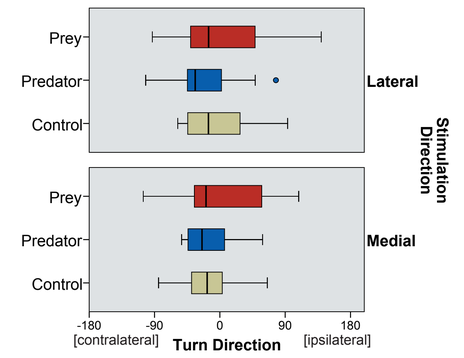


Fig 2.Turn Directions (Flowing Water)

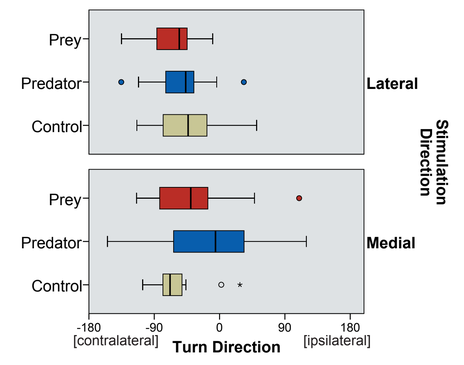


Fig 3. Final Heading (Flowing Water)

Conclusions

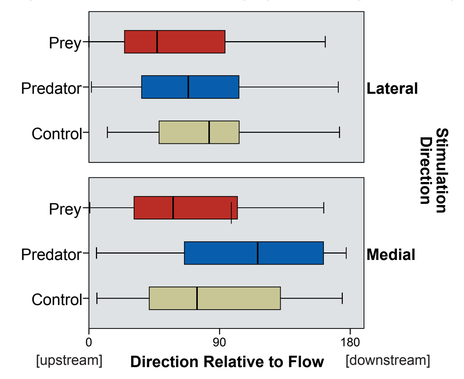
We are unable to distinguish between two possible explanations for our results:

I. Slugs primarily responded to flow rather than odours.

►In the absence of odours, *Tritonia* is positively rheotactic. Our odour stimuli may not have been strong enough to overcome this default response.

II. *Tritonia* integrates odour and flow information from both rhinophores and the oral veil.

►Stimulation of one rhinophore or conflicting flow stimuli between a rhinophore and the oral veil resulted in erratic behaviours.



Future Work

I. Detailed analysis of head movements and paths to determine if finer movements showed significant trends.

II. Test responses of slugs with just one rhinophore to odour plumes.

References

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