



# UK-Canada doctoral exchange scheme: Placement Proposal

### 1. Student statement of interest

This scheme will provide me with a unique, exciting, and important opportunity to up-skill my competencies as a researcher and to establish personal international networks, all in association with a team of scientists producing world-class research within my field of 'insect acoustics'. The 12-week Canada placement amongst this group will immerse me in skills, research pursuits and colleague-relationships that are distinct but highly complimentary to my UK-based doctoral training. The UK base where I work is helping lead the way at the biomimetic interface of the field, and as such I serve as one of the few biologists in what is largely an engineering environment. Biology is the first link in the biomimicry chain and therefore it is of incalculable value that I seize opportunities to experience more of the biology side of my field and develop into the best biological researcher I can be. Nowhere could provide this biological perspective on insect acoustics better than the laboratory of Professor Andrew Mason of the University of Toronto Scarborough.

Prof. Mason's focus is on acoustic and vibrational communication in insects and spiders, with an internationally recognised expertise on the auditory processing of the fly *Ormia ochracea*, possibly the flagship organism of my field. The placement laboratory boasts a suite of expertise and equipment not available at my UK institution, including the means of precise behavioural measurements of insects using a 'trackball'. It is striking that, prior to learning of this scheme, I had anticipated the need in my doctoral research for collaboration to learn how to build and use one of these trackballs. Moreover, it is notable that the *O. ochracea* fly, the subject of the following proposal, is the natural parasitoid of my PhD subject-organism, the field cricket. As such, this placement project will enhance my UK-based research in both practice and theory.

# 2. Placement proposal

### 2.1 Background

Tachinid flies are parasitoids<sup>1</sup>, and many tachinids 'eavesdrop' on the mating song of a male host and use the signal to phonotactically orientate towards the sound source<sup>1,2</sup>. In North America, *Ormia ochracea* parasitises various cricket species with diverse songs, across geographically separated fly populations. Certain populations are known to target the predominant host species in its locality, thus exhibiting 'host specialisation'<sup>3</sup>. Field data indicates local specialisation is a preference behaviour; flies choose the song of their population's natural host cricket<sup>4</sup>.

Conceivably, these differences in behavioural preferences could be due to populations functioning as 'cryptic species'<sup>5</sup>. In fact, fly populations belong to a single genetically homogenous species, indicating adaptive preference is an inherent potential within all *O. ochracea* flies. Cross populational differences in host preference may therefore be due to an innate scope for behavioural flexibility combined with an ability to learn the song of the predominant local host species<sup>3</sup>.

Song learning has been demonstrated in a *O. ochracea* population to be the main if not exclusive factor in determining host preference<sup>6</sup>, and as such might account for all in-field host preferences across populations<sup>4</sup>. However, learning data exists only for one population<sup>6</sup>, and therefore we do not know the cross-populational role of song learning.

Song learning operating within a common potential for behavioural adaptation would necessitate a single broad 'acoustical template'. Since the degree of song preference

independent of learning opportunity has not been investigated across populations, the boundaries of such a template remain undetermined.

Moreover, evidence suggest a degree of population specific temporal tuning to certain songs. For example, Florida flies almost entirely prefer the host *Gryllus rubens*, the dominant species of the region, the song of which is highly divergent in its pulse rate to that of the secondary host *G. firmus*<sup>3</sup>. Modulating *G. rubens* pulse rate considerably reduces phonotactic success<sup>7</sup>. Yet, in contrast, *Teleogryllus oceanicus* shares a similar pulse rate to the largely rejected *G. rubens*<sup>3</sup>, but in Hawaii receives considerable preference<sup>4</sup>. Such seemingly acute differences might suggest the operation of multiple acoustical templates across populations, not just one.

Previous host-preference data have been collected in the field<sup>4</sup>, whereas this project will be the first to investigate cross-populational host preferences in laboratory reared flies. Phonotactic steering accuracy will be recorded on a trackball<sup>8,9</sup> in response to the songs.

Acoustical template boundaries will be explored by modulating pulse rates<sup>7</sup> under otherwise the same experimental conditions as above.

The relative role of song learning across populations will be ascertained by exposing each treatment of laboratory reared flies exclusively to one of the host songs within a five-day period immediately preceding phonotactic steering trials<sup>6</sup>.

In addition: Observations of adult and even larval flies have suggested the possibility of differential rates of activity in flies across populations, including in the absence of an immediate host song stimulus (Andrew Mason, personal communication). Larval activity rates between populations will be measured by kinematic analysis of video recordings of gravid female dissections. From these larvae, adult activity rates will be established by recordings on the trackball of 'forward velocity' and 'total distance'<sup>9</sup> before during and after song exposure.

### 2.2 Objectives

**Research question 1** What is the role of learning-independent temporal preferences in determining host preferences across *Ormia ochracea* populations?

- **Objective 1** To record phonotactic steering accuracy on a trackball in response to different song signals across population treatments of laboratory reared flies.

**Research question 2** What are the minimally effective song parameters for eliciting song learning across populations of *O. ochracea*?

**Objective 2** To record phonotactic steering accuracy on a trackball in response to different song signals, *and to altered song pulse rates*.

**Research question 3** What is the role of song learning in determining host preferences across *O. ochracea* populations?

**Objective 3** To expose each of the populations-derived treatments to five-days of exclusive exposure to one host song, immediately prior to recording phonotactic steering accuracy on a trackball in response to that same signal.

**Research question 4** How do adult and larval activity rates compare across populations?

- **Objective 4** To measure larval activity rates across populations by kinematic analysis of video recordings of gravid female dissections.
  - **Sub-objective 4B** In adults from these larvae, to record trackball 'forward velocity' and 'total distance' before during and after song exposure.

#### 2.3 Significance

The auditory apparatus of Ormia flies enables sound source localisation at a precision comparable to that of humans and unsurpassed among invertebrates<sup>10</sup>. This novel mechanism has been the inspiration of numerous patented engineering designs of 'MEMS' microphones<sup>11</sup>, the type found inside iPhones. With the continued miniaturisation of technologies in this electronic

age, these sensors are only going to grow in their relevance to both industry and society. However, such microphones are of course constrained by size and therefore limited in the acoustic parameters they can receive, especially at low frequencies<sup>12</sup>. By investigating the extent of Ormia directionality to a diversity of acoustic stimuli, this research may help elucidate what sensory range might be possible in the micro-scale sensors of tomorrow.

Month 1				Month 2				Month 3			
W1	W2	W3	W4	W5	W6	W7	W8	W9	W10	W11	W12
Βι	uild trackba	ll									
l	Refine meth	nodology									
			Obj	. 1 – Hos	t preferen	ces					
					0	Obj. 3 – Song learning					
		Obj. 4	I – Larva	l activity r	ates						
			Obj. 4	4B – Adu	It activity	rates					
							Data a	nalysis			
								'Write-up'			

## 2.4 Timeline

### 2.5 Cited literature

**1.** Cade, W. (1975). Acoustically orienting parasitoids: fly phonotaxis to cricket song. *Science*, *190*(4221), 1312-1313.

Köhler, U., & Lakes-Harlan, R. (2001). Auditory behaviour of a parasitoid fly (Emblemasoma auditrix, Sarcophagidae, Diptera). *Journal of Comparative Physiology A*, *187*(8), 581-587.
Gray, D. A., Kunerth, H. D., Zuk, M., Cade, W. H., & Balenger, S. L. (2019). Molecular biogeography and host relations of a parasitoid fly. *Ecology and evolution*, *9*(19), 11476-11493.
Gray, D. A., Banuelos, C., Walker, S. E., Cade, W. H., & Zuk, M. (2007). Behavioural specialization among populations of the acoustically orienting parasitoid fly Ormia ochracea

specialization among populations of the acoustically orienting parasitoid fly Ormia ochracea utilizing different cricket species as hosts. *Animal Behaviour*, 73(1), 99-104.

**5.** Smith, M. A., Wood, D. M., Janzen, D. H., Hallwachs, W., & Hebert, P. D. (2007). DNA barcodes affirm that 16 species of apparently generalist tropical parasitoid flies (Diptera, Tachinidae) are not all generalists. *Proceedings of the national academy of sciences*, *104*(12), 4967-4972.

**6.** Paur, J., & Gray, D. A. (2011). Individual consistency, learning and memory in a parasitoid fly, Ormia ochracea. *Animal behaviour*, *8*2(4), 825-830.

**7.** Walker, T. J. (1993). Phonotaxis in female Ormia ochracea (Diptera: Tachinidae), a parasitoid of field crickets. *Journal of insect behavior*, *6*(3), 389-410.

**8**. Lott, G. K., Rosen, M. J., & Hoy, R. R. (2007). An inexpensive sub-millisecond system for walking measurements of small animals based on optical computer mouse technology. *Journal of neuroscience methods*, *161*(1), 55-61.

**9.** Lee, N., Kirtley, A. T., Pressman, I. S., Jirik, K. J., Koucoulas, D., & Mason, A. C. (2019). Developing a phonotaxis performance index to uncover signal selectivity in walking phonotaxis. *Frontiers in Ecology and Evolution*, 334.

**10.** Mason, A. C., Oshinsky, M. L., & Hoy, R. R. (2001). Hyperacute directional hearing in a microscale auditory system. *Nature*, *410*(6829), 686-690.

**11.** Zhang, Y., Reid, A., & Windmill, J. F. C. (2018). Insect-inspired acoustic microsensors. *Current opinion in insect science*, *30*, 33-38.

**12.** Shah, M. A., Shah, I. A., Lee, D. G., & Hur, S. (2019). Design approaches of MEMS microphones for enhanced performance. *Journal of sensors*, *2019*.

### 3. Benefits to the UK and Canada

### 3.1 Strengthening Canada's research and innovation capacity

My project will focus on North American *Ormia ochracea* populations, mostly in the USA. The bulk of today's impact on this research area is likely arising from US institutions; this project will help towards maintaining Canada's competitiveness in this stream of enquiry. My unique background may facilitate the transfer of new insights to the team in Canada that I will be working with. This background includes: 1) my training as both a biologist but also as a visualiser (the processing of imaging data, such as micro-CT, to develop 3D anatomical models); 2) my experience of a world leading research laboratory working on the biomimetic side of insect acoustics. Lastly, I have identified in my project, to my knowledge, ideas never explored, which should be of benefit to my hosts in Toronto, including: 1) the application of a cross-populational methodology to a laboratory setting; 2) and a song-learning protocol to more than one *O. ochracea* population; 3) as well as the use of video kinematics on larvae activity rates across populations.

### 3.2 Strengthening the UK's research and innovation capacity

In the UK research group led by my home supervisor Professor James Windmill, I help serve as a conduit between biology and engineering, to assist in providing the source of biological insight necessary for the development of novel insect-inspired acoustic sensors. Via the placement project in Canada, I will receive a thorough introduction to the more biological side of the field, which should bring back to our UK team an improved grounding for these sources of biological inspiration. In turn, this should sharpen the UK's cutting-edge in insect-inspired biomimetics.

Prof. Mason's laboratory in Canada is well-established on the biology side of insect acoustics. The laboratory co-founded the Integrative Behaviour & Neuroscience (IBN) Group in 2001 at what is Canada's number one research-intensive university. The IBN is a diverse community of biological scientists. By pursuing my project in the context of the IBN, insights into 'how things are done' will inevitably be obtained, and some insights might be transferred to my UK institution.

The laboratory in Canada in which I will be working boasts a suite of skills and infrastructure that may well be of inspiration to Prof. Windmill's research group at home, and these include: 1) the means to take careful measurements of insect acoustic behaviour – within these skills I intend to learn how to build a phonotaxis trackball which I will later set up in the UK laboratory; 2) the capabilities to make neurophysiological recordings in insects; 3) and lastly, a year-round in house invertebrate rearing facility with a range of organisms that could be of biomimetic interest, from *O. ochracea* flies to black widow spiders and their webs.

#### 3.2 Collaborations between participating researchers

**3.2.1 Does this project build on an existing international collaboration?** Yes No No

**3.2.2 Does this project create potential for future collaborations?** Yes \_\_\_\_\_ No \_\_\_\_\_

### 3.2.3 Please describe briefly the existing, planned or future collaboration.

Professors Mason and Windmill and their respective research groups are already well acquainted in a professional capacity. However, to my knowledge biology personnel have never been exchanged. Over a 12-week period this project will strengthen the existing link which will mutually transfer skills and insights. In turn this could encourage longer periods of future collaboration.

#### 4. Interaction

The project that I will be undertaking at the University of Toronto Scarborough (UTSC) will be one that is very much in line with Professor Mason's existing personal research. As such, Prof Mason will be extremely involved in this project and supervisor-intern interactions will be conducted on a weekly if not daily basis. These interactions will be two-fold, both mentorship – receiving feedback on how to conduct the experiments – and training. The training will be one-to-one under Prof Mason's supervision and will generate skills associated with obtaining data on sensory-guided behaviour in the *Ormia ochracea* fly, specifically: 1) video kinematics, 2) phonotaxis on a trackball (spherical treadmill), and 3) techniques in sensory neurophysiological recordings.

The 12 weeks of research will take place at the host university of UTSC for 100% of the allotted period. This research will be conducted in the laboratory of Prof Mason which is part of the Department of Biological Sciences at the UTSC. The Scarborough campus (UTSC) is one of three integrated campuses that form the University of Toronto. The buildings on this campus that I will be associated with are 'SW 566' and 'SW421-H', and the address is: *Dr. Andrew C Mason, Integrative Behaviour and Neuroscience, University of Toronto Scarborough, 1265 Military Trail, Scarborough ON, M1C 1A4 Canada.* 

In addition, I may have interactions with my host supervisor's colleagues (and their respective students): Prof *Maydianne Andrade*, Associate Prof *Kenneth Welch*, Prof *Nathan Lovejoy*, Assistant Prof *Ina Anreiter*, Assistant Prof *Tod R. Theile*, and Assistant Prof *Minoru Koyama*.