Mole National Park, Ghana, December 5, 2021

Review report on the PhD thesis of Mgr. Jana Vokurkova

In this report, I evaluate the PhD thesis by Mgr. Jana Vokurkova, entitled "Application of bioacoustics approaches in investigation of bird species interactions and community species richness".

The thesis presents original research results in the area of bioacoustic approach applied to study between-species interactions and bird assemblage richness. Specifically, by using two closely related species of nightingales as a study model, it indicates that the heterospecific song convergence in a secondary contact zone results from cultural transmission rather than from interspecific hybridisation and/or gene introgression. The thesis reports also on the effectiveness of passive acoustic monitoring in providing reliable data on bird species richness in comparison to a traditional counting method by a human observer. In addition, the dissertation shows the effect of rainfall on year-round vocal activity patterns in tropical bird communities.

The core of the thesis is comprised of three papers published in peer-reviewed journals. In two (in *Plos One* and in *Journal of Tropical Ecology*), Mgr. Jana Vokurkova is the first author and in one (*Ostrich*) she is listed as the second. According to co-author's statements, her involvement with each study was significant what makes publications that arose from the thesis research Mgr. Vokurkova's contribution to knowledge in a scientific area.

The first article of the PhD thesis of Mgr. Jana Vokurkova combined bioacoustics and genetics to show that Common Nightingale *Luscinia megarhynchos* and Trush Nightingale *L. luscinia* in their secondary contact zone converge vocally what may result from cultural transmission rather than gene flow. Since the article has already been peer-reviewed, I have only minor comments/questions on it:

1. How many of the birds were first recorded and then mist-netted and how one can be entirely sure that these were the same individuals? In addition, six Common Nightingales were recorded at night what limits their identification based on colour ring combination.
2. Please, correct me if I am wrong, but could it be still possible that some of the songs assigned to 'LL' category were actually mixed songs having elements typical for Common Nightingale, but not found in the reference catalogue, although having LM's structure?

The fact is noteworthy that the PhD thesis by Mgr. Jana Vokurkova is supported by three appendices. Two of these articles were also co-authored by Mgr. Vokurkova and were published in well-known behavioural journals like *Behavioral Ecology* and *Animal Behaviour*. Although Mgr. Jana Vokurkova has not been a leading author, she also contributed to the papers that emerged as a continuation of findings of *Plos One*'s article.

The second article included in the thesis compared results of traditional point counts by a human observer with data derived from post-hoc listening to acoustic recordings that were made at the same time as point counts. In general, similarity of species communities based on both methods was relatively high, but in details, the results could be more prone to errors due to a methodological approach applied. Specifically, my concerns are related to a minimum distance between neighbouring points and radius categories of a sampled area (less than 50 m and more than 50 m).

1. A distance of 150 m between counting points does not seem to be long enough to avoid both counting and recording of the same individuals twice (200 m distance was applied in the third article of the PhD thesis). Unsurprisingly, bird species differ to a great extent in their vocal amplitudes, thus at least for some of them, an acoustic overlapping between points sampled could be an issue. Accordingly, songs of some species could still be well-heard on recordings even if produced at a distance longer than 50 m. This makes assigning given vocalisation/record susceptible to bias.
2. Under an assumption that point counts cannot overlap in sampling, a radius of 75 m to correctly estimate a distance of a calling, singing or seen bird, appears to be large in a tropical forest that is frequently densely vegetated.
3. A test showing no differences between the observers would be appreciated.

Nevertheless, the study was the first to apply passive acoustic monitoring for investigating bird assemblages in generally understudied Africa and I appreciate the effort on conducting such comparisons.

The third article that emerged from the PhD study of Mgr. Jana Vokurkova described how bird species richness and vocal activity patterns vary throughout the year in a lowland rain forest. Primarily, the study showed a profound effect of rainfall on the bird community and it was the first one to investigate year-round vocal activity using autonomous recording units. Its importance results from the fact that it documented an annual cycle of avian acoustic performance in an area which can be viewed as highly seasonal, at least when comparing to other tropical regions.

1. Although I find the results and patterns conclusive, the study is limited in a context of the sample size with only one device operating throughout the year continuously. I hope that my concerns could be justified by clear differences in number of species recorded by different automatic recorders in time they covered.

2. Exclusion from the analysis acoustic samples taken during rain could result in failure to find species-specific details on acoustic behaviour. For instance, this is the case of Tambourine Dove *Turtur tympanistria* found in mountain forests of NW region of Cameroon. Apart from being the most vocally active at the beginning of the dry season, the dove shows the second peak of singing output in July (a month with one of the highest rainfall during the year). In this context, the study lacks separation of type vocalisations and their proportions which can be misleading.

In summary, I have no doubts that the PhD thesis of Mgr. Jana Vokurkova presents original and important research results and my recommendation is that the candidate should be awarded with the doctoral degree.

Sincerely,

Paweł Szymański