Ph.D. Thesis

Investigation of structural and pigment-based colouration in the collared flycatcher

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1. Introduction

There are numerous studies that support the possibility that secondary sexual characters could reflect honestly the bearer’s individual quality, hence these traits could be the subject of mate choice, and could thus have an effect on fitness. In birds, one kind of secondary sexual character is the colouration of plumage. In countless cases, individuals of the same species present different colours, such as in the great tit (*Parus major*) or the collared flycatcher (*Ficedula albicollis*). The colour production of these birds and other animals can be fundamentally determined by three distinct mechanisms, and these could appear simultaneously, and could exert their effects together (e.g. Rutowski *et al*., 2005): 1) structural colours are created by the special microstructure of a given tissue, for example due to selective light reflection, 2) pigment-based ornaments are produced by differential absorption of light, while 3) in bioluminescence, light emission during chemical reactions causes the perceived colour.

Colour ornaments may have different production and maintenance costs. Melanin pigments are synthesized *de novo* by birds, and this process is energy demanding, furthermore often requires an elevated testosterone level that could suppress the immune system (Jawor & Breitwisch, 2003). Melanin synthesis may also require metal ions the availability of which in nature is limited (McGraw, 2003). Most structural colours have been investigated extensively from the viewpoint of their costs and information content (Shawkey *et al*., 2003; Griggio *et al.*, 2009), but white colours are an exception, being but scarcely studied (e.g. McGlothlin *et al*., 2007).

There is a serious caveat in several investigations of colours, namely these studies do not make any distinction between different colour producing mechanisms (e.g. carotenoid-structure) even within the same trait (but see e.g. Jacot *et al*., 2010). It is also a common fault that in the case of animals with different colour types, researchers generally investigate only one trait (e.g. Delhey *et al*., 2006), or in the case of studying multiple traits, usually treat these traits independently (with regard to both statistics and biological interpretation) (e.g. Doucet *et al*., 2005), regardless of the potential relationships among them, based on the assumption that different colour ornament types convey different information on their bearer (e.g. McGraw *et al*., 2002). Additionally, besides the above, the investigation of females in relation to secondary sexual traits is scarce although the sexual selection can be strong in this sex too (e.g. Clutton-Brock, 2007).

2. Aims

In this work, I investigated the plumage signal system of the female and male collared flycatcher which display different colours (melanin based and structural). In relation to plumage ornaments, this species, particularly the male, is primarily investigated by the size of the white forehead patch and wing patch (Gustaffson *et al*., 1995; Hegyi *et al*., 2010), while the features of feather reflectance have been ignored. Accordingly, the aims of my investigations could be summarised in the following questions:

1. Are there any relationships among the colour features of different plumage areas within individuals? What is the pattern of the relationships?

2. What is the mating pattern between the individual females and males with respect to colour? Does the mating pattern differ characteristically between distinct, area-level colour features on one hand and plumage-level, composite colour variables (which contains colour variables of different plumage areas) on the other hand?

3. Do the composite colour variables reflect certain aspects of individual quality (i.e. condition and reproductive effort in the past and the present)?

3. Data collection, field and statistical methods

Data were collected in the Pilis-Visegrád mountains near Pilisszentlászló, between 2006 and 2012.

The collared flycatcher is a long-distance migratory, insectivorous species, which presents strong sexual dichromatism (Cramp & Perrins, 1993). The individuals arrive at the mating sites between mid-April and early May, the adult males usually the earliest. This species is generally socially monogamous, although we often detect extra-pair young in the nests (Rosivall *et al.*, 2009), and there is occasional social polygyny (Garamszegi *et al*., 2004). Females nest only once in a mating season, and lay four to eight eggs. The female incubates alone, but both parents feed the offspring.

In spring, males wear nuptial plumage, namely their crown and back is black, while the collar around the neck is white. The belly and the breast are basically white, but sometimes there are a few lightly pigmented feathers in these areas too. One-year-old males have brown primaries, secondaries, coverts and tail, whereas in contrast to this, all of these feathers of adult males ares black. Both yearlings and adults display white forehead patch and wing patch, albeit the size of the latter is half as big in yearlings. In the case of females, the crown, forehead, back, primaries, secondaries and coverts are brownish grey, the wing patch is white, while the belly and the breast are white dappled with brownish feathers (Svensson, 1992).

The birds were captured in their nestbox, when the nestlings were eight- to twelve-days-old. We ringed the individuals, determined their age class (yearling or adult), measured the size of forehead patch, wing patch and tarsus using a sliding caliper, and weighed the body mass using a Pesola spring balance.

We visited the nestbox plots multiple times a week in order to determine exactly the date of the onset of egg laying.

I quantified the plumage colouration using a USB2000 spectrophotometer. In the process of this, I sampled five plumage areas: forehead, crown, wing patch, wing coverts and breast. I took two spectral readings in each sampling area, and based on this established that the repeatability of the measurements was high, that is, the method was reliable. Using the raw reflectance data, I created two objective spectral variables which are independent of the hypothesized features of the sensory system of birds (Cuthill *et al.*, 1999): brightness and UV chroma (e.g. Woodcock *et al.*, 2005).

For the analyses, I used the following statistical methods: Kaiser–Meyer–Olkin-index, common principal component analysis (CPC), principal component analysis (PCA), Kolmogorov–Smirnov and Lilliefors test for normality, Pearson and Spearman correlation, Student t test, and general linear models with backward stepwise model selection.

4. Brief summary of results and discussion

– *Sexual dichromatism*: I found differences in colouration not only in the human visible part of the spectrum, but also in the near ultraviolet range. Based on this, the plumage areas that seem similar for human observers between the sexes (breast, wing patch) differed appreciably concerning brightness and UV chroma.

– *Correlation structure of area-level spectral variables*: There were strong correlations among several colour variables, but the integration was moderate between brightness variables and UV chroma variables. The correlation structure among the plumage areas moderately differed between the two intervals of examinations (2006, 2008, 2009 vs. 2008-2012). Based on the very strong sexual dichromatism, I supposed that the correlation structure (the structure of integration) between the sexes might also differ (Vincent & Lailvaux, 2008), but the patterns of parallel colour change across the plumage were similar in males and females.

– *Integration of white patches and spectral features*: The size of forehead patch and wing patch was largely unrelated to the composite, integrated colour variables (i.e. the PCs). This and the above mentioned results suggest that the different attributes of plumage, namely brightness, UV chroma and patch sizes change independent of each other among the individuals, and it is therefore possible that these plumage features reflect different aspects of individual quality (Badyaev *et al*., 2001; Candolin, 2003).

– *Mating pattern in relation to integrated colour variables*: I detected strong assortative mating patterns regarding integrated colour variables, namely females with more pronounced colour saturation (UV chroma) across the whole plumage mated with more saturated males. I found similar relationships in the case of pigment-based and structural-based component of plumage brightness. In numerous species which display multiple similar or different colours, it is not known whether only the individual traits function as sexually selected characters and convey information on individual quality, or also the integrated composite traits. My results supported the latter scenario.

– *Analysis of functional integration with respect to mating pattern*: In the case of integrated spectral variables and the area-level spectral variables belonging to the same PC I found strong mating patterns, at the same time the patterns were very weak in relation to area-level spectral variables belonging to different PCs. This result supports the hypothesis that the plumage-level, integrated colour features could play a substantial role in mating decisions.

– *Information content of female colouration*: Those females that had been in better condition in a given mating season, developed darker feathers in the pigmented plumage areas in the subsequent year. The pigment-based component of brightness related positively to the laying date in the given year, that is, darker females started to lay the eggs earlier.

– *Information content of male colouration*: Males that were in better condition in a given breeding season displayed darker pigmented plumage areas in the same season. The structural-based component of male brightness related negatively to the laying date of the first egg in his nest in the same year, and the UV chroma of white plumage areas related positively to the clutch size in the given season.

In summary, the results suggest that some area-level plumage colour traits may covary and provide information on individual quality together, and this plumage-level spectral variation could play a role in mating decisions. Expression of pigmentation was related to condition and egg laying date, while the structural component of plumage-level colouration was related to certain aspects of reproductive effort. Hence my work underlines that future investigations of sexual selection should analyse potential developmental and functional integration among apparently distinct traits, as receivers of the signals do not necessarily treat different plumage parts separately, but may also assess them as an integrated unit when evaluating the signaller's quality.

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6. References

Badyaev, A.V., Hill, G. E., Dunn, P.O., Glen, J.C. 2001. Plumage color as a composite trait: Developmental and functional integration of sexual ornamentation. The American Naturalist, 158, 221-235.

Candolin, U. 2003. The use of multiple cues in mate choice. Biological Reviews, 78, 575-595.

Clutton-Brock, T. H. 2007. Sexual selection in males and females. Science, 318, 1882-1885.

Cramp, S., Perrins, C. M. (eds.) 1993. The Birds of the Western Palearctic, Vol. VII. Oxford University Press, Oxford.

Cuthill, I. C., Bennett, A. T. D., Partridge, J. C., Maier, E. J. 1999. Plumage reflectance and the objective assessment of avian dichromatism. The American Naturalist, 160, 183-200.

Delhey, K., Peters, A., Johnsen, A., Kempenaers, B. 2006. Seasonal changes in blue tit crown color: do they signal individual quality? Behavioral Ecology, 17, 790-798.

Doucet, S. M., Mennill, D. J., Montgomerie, R., Boag, P. T., Ratcliffe, L. M. 2005 Achromatic plumage reflectance predicts reproductive success in male black-capped chickadees. Behavioral Ecology, 16, 218-222.

Garamszegi, L. Z., Török, J., Michl, G., Møller, A. P. 2004. Female survival, lifetime reproductive success and mating status in a passerine bird. Oecologia, 138, 48-56.

Griggio, M., Serra, L., Licheri, D., Campomori, C., Pilastro, A. 2009. Moult speed affects structural feather ornaments in the blue tit. Journal of Evolutionary Biology, 22, 782-792.

Gustaffson, L., Qvarnström, A., Sheldon, B. C. 1995. Trade-offs between life-history traits and a secondary sexual character in male collared flycatchers. Nature, 375, 311-313.

Hegyi, G., Szöllősi, E., Jenni-Eiermann, S., Török, J., Eens, M., Garamszegi, L. Z. 2010. Nutritional correlates and mate acquisition role of multiple sexual traits in male collared flycatchers. Naturwissenschaften, 97, 567-576.

Jacot, A., Romero-Diaz, C., Tschirren, B., Richner, H., Fitze, P. S. 2010. Dissecting carotenoid from structural components of carotenoid-based coloration: a field experiment with Great Tits (*Parus major*). The American Naturalist, 176, 55-62.

Jawor, J. M., Breitwisch, R. 2003. Perspectives in ornithology. Melanin ornaments, honesty, and sexual selection. The Auk, 120, 249-265.

McGlothlin, J. W., Duffy, D. L., Henry-Freeman, J. L., Ketterson, E. D. 2007. Diet quality affects an attractive white plumage pattern in dark-eyed juncos (*Junco hyemalis*). Behavioral Ecology and Sociobiology, 61, 1391-1399.

McGraw, K. J. 2003. Melanins, metals, and mate quality. Oikos, 102, 402-406.

McGraw, K. J., Mackillop, E. A., Dale, J., Hauber, M. E. 2002. Different colors reveal different information: how nutritional stress affects the expression of melanin- and structurally based ornamental plumage. The Journal of Experimental Biology, 205, 3747-3755.

Rosivall, B., Szöllősi, E., Hasselquist, D., Török, J. 2009. Effects of extra-pair paternity and sex on nestling growth and condition in the collared flycatcher, *Ficedula albicollis*. Animal Behaviour, 77, 611-617.

Rutowski, R. L., Macedonia, J. M., Morehouse, N., Taylor-Taft, L. 2005. Pterin pigments amplify iridescent ultraviolet signal in males of the orange sulphur butterfly, *Colias eurytheme*. Proceedings of the Royal Society of London Series B, 272, 2329-2335.

Shawkey, M. D., Estes, A. M., Siefferman, L., Hill, G. E. 2003. Nanostructure predicts intraspecific variation in ultraviolet-blue plumage colour. Proceedings of the Royal Society of London Series B, 270, 1455-1460.

Svensson, L. 1992. Identification Guide to European Passerines. 4th edn. Stockholm: Märstatryck.

Vincent, S. E., Lailvaux, S. P. 2008. Does phenotypic integration constrain sexual size dimorphism in eastern lubber grasshoppers (*Romalea microptera*)? Journal of Orthoptera Research, 17, 219-225.

Woodcock, E. A., Rathburn, M. K., Ratcliffe, L. M. 2005. Achromatic plumage reflectance, social dominance and female mate preferences in black-capped chickadees (*Poecile atricapillus*). Ethology, 111, 891-900.

7. Publications that are parts of the thesis

Laczi, M., Hegyi, G., Herényi, M., Kiss, D., Markó, G., Nagy, G., Rosivall, B., Szöllősi, E., Török, J. 2013. Integrated plumage colour variation in relation to body condition, reproductive investment and laying date in the collared flycatcher. Naturwissenschaften, 100, 983-991. (IF: 1,971)

Laczi, M., Török J., Rosivall B., Hegyi G. 2011. Integration of spectral reflectance across the plumage: Implications for mating patterns. PLoS ONE 6:e23201. (IF: 4,092)

Laczi M., Török, J., Rosivall, B., Hegyi G. 2011. Information content of plumage-level reflectance variation in the collared flycatcher. EOU VIII., Riga.

Laczi M., Török J., Rosivall B., Hegyi G. 2010. A tollazati színek összefüggései és a párbaállás mintázata örvös légykapónál. MET Konferencia XII., Veszprém.

Laczi M., Garamszegi L. Z., Rosivall B., Török J., Hegyi G. 2009. Rearing environment and whole plumage reflectance in male and female collared flycatchers. ESEB XII., Torino.

Laczi, M., Hegyi, G., Cserepes, T. M., Michl, G., Rosivall, B., Török, J. 2007. A depigmentált tollazat lehetséges információtartalma egy hazai énekesmadár fajnál. MET Konferencia X., Göd.

Laczi, M., Hegyi, G., Cserepes, T. M., Michl, G., Rosivall, B., Török, J. 2007. Spectral features of depigmented plumage patches in male collared flycatchers (*Ficedula albicollis*). 8th Behavioral Ecology Meeting, Kolozsvár.

8. Other publications

Vaskuti, É., Zsebők, S., Herczeg, G., Blázi, Gy., **Laczi, M.**, Nagy, G., Török, J., Garamszegi, L. Z. A kulturális evolúció nyomai az örvös légykapó (*Ficedula albicollis*) énekében. Manuscript in decision.

Kötél, D., **Laczi, M.**, Török, J., Hegyi, G. Mutual ornamentation and the parental behaviour of male and female collared flycatchers in the incubation period. Manuscript in decision.

Szöllősi, E., Garamszegi, L. Z., Hegyi, G., **Laczi, M.**, Rosivall, B., Török, J. Haemoparasite infection status of collared flycatcher males changes within a breading season. Manuscript in decision.

Zsebők, S., Herczeg, G., Blázi, Gy., **Laczi, M.**, Nagy, G., Szász, E., Markó, G., Török, J., Garamszegi, L. Z. Signal design and personality in bird song: short-term and long-term repeatability of traits expressed at different hierarchical level. Manuscript.

Zsebők, S., Herczeg, G., Blázi, Gy., **Laczi, M.**, Nagy, G., Török, J., Garamszegi, L. Z. Don't bother with calculating repertoire size of birdsong – usage of minimum spanning tree to estimate the diversity/complexity of communication signals. Manuscript.

Hargitai, R., Nagy, G., Herényi, M., Nyiri, Z., **Laczi, M.**, Hegyi, G., Eke, Zs., Török, J. 2016. Darker eggshell spotting indicates lower yolk antioxidant level and poorer female quality in the Great Tit (*Parus major*). Auk, 133, 131-146. (IF_{2014}: 1,864)

Hegyi, G., **Laczi, M**. 2015. Using full models, stepwise regression and model selection in ecological data sets: Monte Carlo simulations. Annales Zoologici Fennici, 52, 145-159. (IF_{2014}: 0,855)

Hegyi, G., Kötél, D., **Laczi, M.** 2015. Direct benefits of mate choice: a meta-analysis of plumage colour and offspring feeding rates in birds. Naturwissenschaften, 102, 62. (IF_{2014}: 2,10)

Hegyi, G., **Laczi, M.**, Nagy, G., Szász, E., Kötél, D., Török, J. 2015. Stable correlation structure among multiple plumage colour traits: can they work as a single signal? Biological Journal of the Linnean Society, DOI: 10.1111/bij.12412. (IF: 2,264)

Hargitai, R., Hegyi, G., Herényi, M., **Laczi, M.**, Nagy, G., Rosivall, B., Szöllősi, E., Török, J. 2014. Winter body condition in the collared flycatcher: Determinants and carry-over effects on future breeding parameters. Auk, 131, 257-264. (IF: 1,864)

Boross, N., Markó, G., **Laczi, M.**, Garamszegi, L. Z., Hegyi, G., Herényi, M., Kiss, D., Nagy, G., Rosivall, B., Szöllősi, E. and Török, J. 2012: Source of variation in haematocrit in the collared flycatcher (*Ficedula albicollis*). Ornis Hungarica, 20, 64-72.

Markó, G., Azcárate, M., Hegyi, G., **Laczi, M**., Nagy, G., Senar, J. C., Török, J., Garamszegi, L.Z. 2013. Behavioural responses to handling stress in the great tit: within-individual consistency and the effect of age, sex and body condition. Ornis Hungarica, 21, 12-25.

Bajer, K., Molnár, O. R., Hegyi, G., Herczeg, G., **Laczi, M.**, Török, J. 2009. Zöld gyíkok színezete és morfológiája: jelzések és funkciók. Állattani Közlemények, 94, 167-176.

Hegyi, G., Szigeti, B., **Laczi, M.**, Eens, M., Török, J. 2008. Correlated variation of colour between melanin and carotenoid pigmented plumage areas in great tits *Parus major*. Evolutionary Ecology Research, 10, 559-574. (IF: 1,17)

Laczi, M., Cserepes, T. M., Hegyi, G., Michl, G., Szigeti, B., Török, J. 2006. A tollazat UV-reflektanciája és a táplálkozás kapcsolata vízimadaraknál. Állattani Közlemények, 91, 117-126.

Vaskuti, É., Zsebők, S., Herczeg, G., Blázi, Gy., **Laczi, M.**, Nagy, G., Török, J., Garamszegi, L. Z. 2016. A kulturális evolúció az örvös légykapó (*Ficedula albicollis*) énekében. VI. Szünzoológiai Szimpózium, Budapest.

Vaskuti, É., Zsebők, S., Herczeg, G., Blázi, Gy., **Laczi, M.**, Nagy, G., Török, J., Garamszegi, L. Z. 2015. Kulturális evolúció az örvös légykapó énekében. MET Konferencia XVII., Dobogókő.

Zsebők, S., Herczeg, G., Blázi, Gy., **Laczi**, **M.**, Nagy, G., Török, J., Garamszegi, L. Z. 2015. What does the hierarchical analysis of quantitative and qualitative song traits tell us about signal design and reliability? IBAC XXV., Murnau.

Hegyi, G., Kötél, D., **Laczi, M.** 2014. A „jó szülő" hipotézis metaanalízise madaraknál. MET Konferencia XVI., Tihany.

Kötél, D., **Laczi, M.**, Török, J., Hegyi, G. 2014. Kotlás alatti viselkedés és tollazati díszek összefüggései hím és tojó örvös légykapóknál. MET Konferencia XVI., Tihany.

Zsebők, S., **Laczi, M.**, Nagy, G., Blázi, G., Garamszegi, L. Z. 2013. Cultural evolution of bird song: a long-term study of the Collared Flycatcher. IBAC XXIV., Pirenópolis.

Boross, N., Markó, G., **Laczi, M.,** Garamszegi, L. Z., Hegyi, G., Herényi, M., Kiss, D., Nagy, G., Rosivall, B., Szöllősi, E., Török, J. 2013. A hematokrit szezonális mintázata és kapcsolata az egyedi minőséggel örvös légykapónál (*Ficedula albicollis*). 14. Kolozsvári Biológus Napok, Kolozsvár.

Boross, N., Markó, G., **Laczi, M.**, Garamszegi, L. Z., Hegyi, G., Herényi, M., Kiss, D., Nagy, G., Rosivall, B., Szöllősi, E., Török, J. 2013. A hematokrit repetabilitása és szezonális változása az örvös légykapónál (*Ficedula albicollis*). 5. Szünzoológiai Szimpózium, Vácrátót.

Laczi, M., Cserepes, T. M. 2007. A tollazat UV reflektanciája és a táplálkozás kapcsolata vízimadaraknál. 28. OTDK, Biológia Szekció, Debrecen.

Hegyi, G., Szigeti, B., **Laczi**, M., Török, J. 2007. Több jelzés összhangja: az egyedek közti variáció közös tengelyei széncinegék (*Parus major*) tollazati foltjainak színezetében. MBT, Ökológiai Szakosztály, Budapest.

Laczi, M., Cserepes, T. M., Török, J. 2006. A tollazat UV színezete halevő madaraknál. 7. Magyar Ökológiai Kongresszus, Budapest.

Cserepes, T. M., **Laczi, M**., Michl, G., Török, J. 2006. A tollazat UV reflektanciája és a táplálkozás kapcsolata vízimadaraknál. MBT, Állattani Szakosztály 946. ülése, Budapest.

Laczi, M., Cserepes, T. M. 2005. A tollazat UV reflektanciája és a táplálkozás kapcsolata vízimadaraknál. Biológus TDK, Állatökológia Szekció.