

goes up only on time lag. The water-sprayed, open cane (control) showed sharp bands in all samplings, pointing to poor protection from SAI inversion. On the other hand, the water-sprayed, covered cane and those sprayed with electrolysed water were protected in the initial phase of the study, but were found to show comparatively weaker SAI expression recorded 144 h after harvest. However, an appreciable suppression of SAI expression was observed over the entire study period of 192 h (=8 days), especially in response to the BKC + SMS combination treatment, evident from the fact that no bands were observed with respect to SAI in any of the lanes corresponding to the T-4 treatment.

Reducing sugars in juice are considered an important indicator of cane deterioration. Solomon *et al.*<sup>8,9,19</sup> have also reported higher levels of reducing sugars in juice on storage of harvested cane. Also, CCS is the major quality determining factor which is considered while studying the deterioration. Thus, alongside the expression analysis, reducing sugars and CCS% were also estimated in juice samples to ascertain the deterioration in cane quality. As shown in Figure 3, at the time of harvest, reducing sugars were recorded as 76.65/100 Brix which, over the period of 192 h, increased by 2.09 fold in control cane (T-1). However, in T-2, T-3 and T-4 canes it increased by 1.63, 1.42 and 1.35 fold respectively. CCS% at the time of harvest was calculated as 14.01 which decreased by 4.91, 4.29, 2.59 and 2.23 in T-1, T-2, T-3 and T-4 canes respectively, in the span of 192 h post-harvest.

These results reassert the efficacy of chemical treatments with a synergistic anti-bacterial and anti-inversion effect, in

minimizing post-harvest sucrose loss, especially biochemical inversion. However, since the inversion of sucrose into glucose and fructose is the major cause of significant loss of sucrose in stale cane, future research must be targeted towards increasing sucrose yield by careful down-regulation of the enzymes, especially SAI, involved in sucrose inversion. With the recent advancements in genomics anti-sense technology developed to reduce invertase activity soon after harvest of the cane crop, would be useful in minimizing the post-harvest sucrose losses. Such control can be realized by employing the RNAi approach to tune the level of SAI at suitable locations, soon after harvest, which will have far-reaching impact on the sugar industry vis-à-vis export policy of the Government.

1. Solomon, S., *Sugar Tech.*, 2009, **11**, 109–123.
2. Solomon, S., Shahi, H. N., Suman, A., Gaur, A., Deb, S. and Singh, I., *Proc. Int. Soc. Sugar Cane Technol.*, 2001, **24**, 380–381.
3. Solomon, S., *Sugar Tech.*, 2000, **2**, 1–18.
4. Agrawal, M., Ojha, S. K. and Jha, S. P., *Ganna Kheti*, 1976, 1–3.
5. Magdum, D. N. and Kadam, S. K., *Bharatiya Sugar*, 1996, 45–52.
6. Solomon, S. and Singh, P., *Sugar Tech.*, 2009, **11**, 228–230.
7. Solomon, S. *et al.*, *Sugar Tech.*, 2006, **8**, 74–78.
8. Solomon, S., Shrivastava, A. K. and Yadav, R. L., In Proceedings of the Annual Convention of Sugarcane Technologist Association of India, 2007, vol. 68, pp. 112–121.
9. Solomon, S., Shrivastava, A. K., Singh, P., Singh, I., Sawnani, A. and Prajapati, C. P., *Int. Sugar J.*, 2008, **110**, 236–241.

10. Solomon, S., Srivastava, K. K., Bhatnagar, S. and Madan, V. K., *Indian Sugar*, 1990, **39**, 895–899.
11. Batta, S. K. and Singh, R., *Bharatiya Sugar*, 1991, **16**, 49–50.
12. Uppal, S. K., Bhatia, S. and Thind, K. S., *Sugar Tech.*, 2008, **10**, 346–349.
13. Mao, L., Que, F. and Wang, G., *Food Chem.*, 2006, **98**, 338–342.
14. Lontom, W., Kosittrakun, M. and Lingle, S. E., *Thai J. Agric. Sci.*, 2008, **41**, 143–151.
15. Chandra, A., Jain, R., Rai, R. K. and Solomon, S., *Natl. Acad. Sci. Lett.*, 2010, **33**, 355–359.
16. Chandra, A., Roopendra, K., Sharma, A., Jain, R. and Solomon, S., *Natl. Acad. Sci. Lett.*, 2014 (in press).
17. Nelson, N., *Biol. Chem.*, 1944, **153**, 375–387.
18. Bakshi Ram, Sahi, B. K., Kumar, S., Sharma, V. P. and Chathurvedi, B. K., *Indian J. Sugarcane Technol.*, 2001, **16**, 36–43.
19. Solomon, S., Shrivastava, A. K., Srivastava, B. L. and Madan, V. K., *Technical Bulletin No.37. Indian Institute of Sugarcane Research, Lucknow*, 1997, pp. 1–217.

Received 1 August 2013; revised accepted 21 May 2014

AMARESH CHANDRA\*  
KRITI ROOPENDRA  
PRIYANKA SINGH  
RADHA JAIN  
C. P. PRAJAPATI  
SUSHIL SOLOMON

*Plant Physiology and Biochemistry*  
Division,  
Indian Institute of Sugarcane Research,  
Lucknow 226 002, India  
\*For correspondence.  
e-mail: amaresh\_chandra@rediffmail.com

## Transformation of colourful pattern of eyespot in peacock wing

An eyespot or ocellus is an eye-like pattern or structure found in various phyla, including butterflies, reptiles, felids, fishes and birds<sup>1–3</sup>. In some species of fishes and butterflies, eyespot is a form of mimicry to draw a predator's attention away from the most vulnerable body parts, or to appear as an inedible or even dangerous animal<sup>2,4–6</sup>. In some butterflies, besides antipredatory function it also plays an

important role in kin recognition and sexual selection<sup>5,6</sup>. In birds like the peacock, eyespot is present in the tail feather with a function of intraspecific communication and courtship. The communication is mediated by the fan of the tail feather, which is composed of 170 eye feathers bordered by the 30 T-shaped feathers which do not contain an eyespot<sup>7</sup>. All the eye feathers are arranged in an in-

creasing length so that all the eyespots will be visible when the tail feather is fanned<sup>7</sup>. The eyespot of the peacock is different from other animals as it includes an intricate shape and multiple rings having bright and iridescent colours (colours that change with the viewing angle). The peacock has various feather patterns throughout its body. Feather patterns observed in birds are diverse in

nature and classified into various categories: central pigment patch, hollow central patch with a non-pigmented centre, concentric central patch, bars, chevrons, a circular eyespot or eye, a pair of distal and proximal circular central spots, rows of laterally paired spots, and an array of offset dots<sup>8</sup>. Peacocks possess both bar and eye pattern feathers. The present study deals with the various developmental stages involved in the process of eyespot formation.

Peacock feathers were collected from BITS-Pilani, Pilani campus, Rajasthan, India, during August–September 2013, as this is the shedding season for peacocks. Damaged feathers were discarded during preliminary sampling. Undamaged feathers were further separated as T-shaped feathers and eyespot-shaped feather. T-shaped feathers can be easily identified from their shape and discarded from the final sampling. Feathers depicting various developmental stages of eyespot were selected for further analysis, and used as a platform to study pattern formation. These feathers were then further classified according to their developmental stages that were photographed using a digital camera and these photographs were further analysed using Adobe Photoshop.

The peacock feathers have some attractive features, which include formation of feathered fan and uniform distribution of eyespots across the feather. A developed eyespot has several adjacent iridescent colours, including a dark purple–black centre surrounded by two large concentric regions of blue–green and bronze–gold, as well as a few narrower outer bands of additional colours. Purple–black colour appears at the centre and forms the pupil of the eyespot. Blue appears next to the purple–black colour and sketches the iris for the eyespot. The size

of the eyespot increases with the length of the feather. The size of each layer changes in the right proportion so that the pattern of the eyespot is maintained with the growth of the feather (Figure 1). Let us consider the various stages prior to the complete eyespot pattern formation.

In the first stage the feather appears dark brown in colour (Figure 2 *a*), which later changes to green colour and is classified as the second stage (Figure 2 *b*).

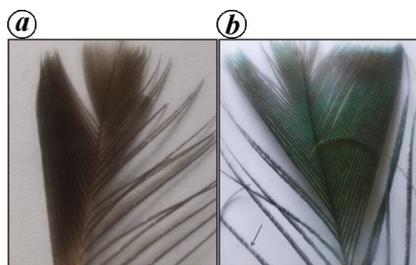
In the third stage eyespot formation begins after the second stage. The feather includes two colours: bronze and green. Bronze colour appears at the centre of the feather in the form of a convex lens which is surrounded by the bright green colour (Figure 3 *a*).

In the fourth stage three different colours appear in the feather. The eyespot of the feather appears in two colours: blue and bronze. Blue colour appears at the centre of the bronze colour with a structural resemblance of a convex lens. The eyespot is further surrounded by green colour at the margin (Figure 3 *b*).

The fifth stage occurs prior to the complete eyespot pattern formation. The eyespot includes an eye pattern, with dark purple, blue, bronze and green colours. The dark purple colour appears as a

dot at the centre of the blue colour of the eyespot (Figure 4). Later the purple colour grows and reaches its fully developed shape in a well-developed eyespot (compare Figures 1 and 4). Away from the eyespot region, the barbules are uniformly green in colour, adding attractiveness to the peacock feather. As observed above, these are the various stages of development of the eyespot on the peacock feathers.

The most striking feature of the peacock is its multicoloured eyespot in the tail feather. The peahens are attracted towards the number and density of the eyespots at the time of selecting their mates<sup>9–11</sup>. Recently Dakin and Montgomerie<sup>3</sup> have reported that it is not the number but the colouration of the eyespot which plays a critical role in the mate selection process. In peacock, the eyespot development is a post-embryonic process, and various colours of the ring appear gradually. Furthermore, the colouration of the eyespot is due to structural colouration which is produced by the development of fine structures called barbules in the barb<sup>12</sup>. Although barbules have been reported in various birds such as hummingbirds, pigeons and kingfishers, the peacock possesses the largest iridescent barbules<sup>13</sup>. The fine structure along with visible light and pigments help in the formation of the multi-coloured eyespot in the wing. Peacock tail possesses melanin as the pigment granule, which gives the barbs a uniform brown colour. The spectacular colours of the eyespot are due to an optical interference phenomenon, which is caused by a



**Figure 2.** Early developmental stages of the eye feather. *a*, First stage of development when the feather appears brown in colour. *b*, Second stage of development when the feather appears green in colour.



**Figure 3.** Intermediate developmental stages of the eye feather. *a*, Bronze colour appears at the centre of the feather. *b*, Appearance of blue colour at the core of the bronze colour.



**Figure 1.** Structure of the eye feather. Multi-coloured large and small feathers with eyespots. The pattern of the eyespot remains the same in both cases.



**Figure 4.** Late stage of the eye feather. Purple colour appears in the middle of the blue colour. The blue colour increases in size leading to the completion of pattern formation in the eye feather.

minute change in the thickness of the keratin layer<sup>12</sup>. How much the thickness varies to bring this beautiful pattern is currently not known. A ultrastructure study of each ring of the eyespot will answer this question. However, during evolution, changes in keratin layer thickness were probably introduced by mutation and genetic information was stored. How genetic changes are encoded and specified during development need further studies. In butterfly, cautery of the hind wing of *Precis* induced the formation of an eyespot-like pattern<sup>14</sup>. It would be interesting to check with a cautery or transplantation experiment at the centre of the eyespot to know further about its development. In birds, the most accepted theory of pattern formation is the reaction–diffusion model proposed by Prum and Williamson<sup>8</sup>. According to this model, the outline of the eyespot can be stimulated with the addition of a second inhibitory signal that inhibits both the activating and primary inhibitory signals<sup>8</sup>. In nature, patterns evolve by distorting or displacing pattern elements, and by selectively suppressing or enlarging specific elements. All these above stated mechanisms also occur during development and help evolve patterns.

Considering the interrelationship between pattern evolution and development as proposed by Haeckel<sup>15</sup>, various stages described here are probably the various patterns that evolved during evolution and were stored as a blueprint. A pattern is made up of semi-independent pattern elements whose identity can be traced from species to species and genera to genera. The pattern elements of butterfly have homology with those of the bones of vertebrate pattern. In this context it would be interesting to study the homologous structure of eyespot pattern of the peacock.

1. Vallin, A., Jakobsson, S. and Wiklund, C., *Behav. Ecol. Sociobiol.*, 2007, **61**, 1419–1424.
2. Kjærnsmo, K. and Merilaita, S., *Proc. Biol. Sci.*, 2012, **279**, 4192–4198.
3. Dakin, R. and Montgomerie, R., *Behav. Ecol.*, 2013, **24**, 1048–1057.
4. Stevens, M., *Biol. Rev.*, 2005, **80**, 573–588.
5. Blest, A. D., *Behaviour*, 1957, **11**, 209–256.
6. Lyytinen, A., Brakefield, P. M. and Maples, J., *Oikos*, 2003, **100**, 372–379.
7. Burgess, S., *J. Creation*, 2001, **15**(2), 96.
8. Prum, R. O. and Williamson, S., *Proc. R. Soc. London, Ser. B*, 2002, **269**, 781–792.

9. Petrie, M., Halliday, T. and Sanders, C., *Anim. Behav.*, 1991, **41**, 323–331.
10. Petrie, M. and Halliday, T., *Behav. Ecol. Sociobiol.*, 1994, **35**, 213–217.
11. Loyau, A., Saint, J. M. and Sorci, G., *Ethology*, 2005, **111**, 810–820.
12. Zi, J. *et al.*, *Proc. Natl. Acad. Sci. USA*, 2003, **100**, 12576–12578.
13. Mason, C. W., *J. Phys. Chem.*, 1923, **27**, 440.
14. Nijhout, H. F., *J. Embryol. Exp. Morphol.*, 1985, **86**, 191–203.
15. Haeckel, E., *Generelle Morphologie der Organismen*, Georg Reimer, Berlin, 1867.

ACKNOWLEDGEMENTS. I thank Ms Chhabi Parida for help in collecting the peacock feathers used in this study, and the anonymous reviewers for their valuable comments that helped improve the manuscript.

Received 14 February 2014; revised accepted 16 May 2014

MONALISA MISHRA

*Department of Life Science,  
NIT Rourkela,  
Rourkela 769 008, India  
e-mail: mishramo@nitrkl.ac.in*