Colour pattern modification by coldshock in Lepidoptera

By H. FREDERIK NIJHOUT

Department of Zoology, Duke University, Durham, North Carolina 27706, U.S.A.

SUMMARY

When young pupae of *Vanessa cardui, V. virginiensis* and *Precis coenia* (Lepidoptera: Nymphalidae) are exposed to severe coldshock many develop aberrant adult wing patterns. For each species, a synchronous cohort of experimental animals always develops a broad range of aberrant pattern morphologies but these can always be arranged in a single unbranched morphological series. When such phenotypic series are compared, between species and between wing surfaces within a species (each wing surface usually bears a different colour pattern), many parallel modifications and trends become evident. These parallelisms reveal certain homologies of pattern elements and suggest that a common physiology underlies the development of a considerable diversity of normal and aberrant colour patterns. The case is made that the phenotypic series produced may represent either a series of sequential stages in colour pattern determination or, a series of quantitatively different 'interpretations' of an established gradient system. Colour pattern morphoclines reveal which patterns are developmentally 'adjacent' to one another and may therefore prove useful in elucidating the evolution of patterns.

INTRODUCTION

The colour patterns on the wings of Lepidoptera consist of a simple two-dimensional array of coloured scales. Each scale is the product, or appendage, of one epidermal cell and is, in most cases, of a single homogeneous colour. The overall pattern is therefore a mosaic of monochrome scales comprising a very limited set (usually two to five) of colours (Nijhout, 1980a, 1981). Pattern formation in this system is the process whereby a given scale cell 'selects', from a limited set of possibilities, which pigment it will synthesize in the future. Comparative studies have shown that each species of lepidopteran has a characteristic colour pattern whose component elements can usually be recognized as homologs of similar pattern elements in other species. Furthermore, colour pattern development in individual *wing-cells* (these are areas on a wing bordered by wing-veins) is as a rule independent from that in adjacent wing-cells and the patterns that develop in the wing-cells appear, in fact, to be serially homologous (Schwanwitsch, 1924; Sueffert, 1927; Nijhout, 1978, 1984).

At least a portion of pattern determination in each wing-cell depends on the organizing properties of discrete groups of cells, the foci, which emit a signal that

somehow causes scale-forming cells in their surroundings to become determined to synthesize particular pigments. Diversity in the physiological processes that attend signal propagation and interpretation are responsible for the diversity of patterns observed; yet, the ability to detect clear homologies of pattern elements across a tremendous diversity of patterns suggests that these processes are relatively few in kind and that many of the essential differences between species-specific patterns may be quantitative rather than qualitative (Nijhout, 1978, 1980b, 1984).

Some of the evidence for the proposition that many colour patterns may differ only in quantitative aspects comes from a long tradition (going back to the later decades of the previous century) among lepidopterists of inducing colour pattern aberrations in various species by exposing developing pupae to abnormally high or low temperatures (see Kuehn (1926), Prochnow (1929) and Goldschmidt (1938) for reviews of this early work). Such experiments have led to the observation that many otherwise different-looking patterns became modified in surprisingly similar ways and that the aberrant patterns of many species tended to resemble each other in many features, more so than did their 'wild type' patterns. In a few instances a pattern aberration showed a remarkable resemblance to the colour pattern of a different geographic race of the species. Artificially induced pattern aberrations and the occasional natural occurrences of such patterns have long attracted the casual attention of collectors (indeed, more than a dozen papers appear each year describing such colour pattern aberrations and the most common ones are sometimes given names). Recently, Shapiro (1975, 1981a,b, 1983) has been examining the ecological and evolutionary implications of temperature-induced pattern aberrations, since it is conceivable that under certain circumstances such aberrations could become fixed by genetic assimilation (Waddington, 1953). To date there has been little or no systematic work done to uncover the developmental and physiological basis for these most interesting phenomena. The present paper is a first step towards that end. I will show that temperature-induced pattern aberrations fall into simple morphological series and that there are considerable similarities in the details of pattern modification in three different species of nymphalid butterflies.

MATERIALS AND METHODS

Larvae of *Precis coenia* and *Vanessa cardui* were reared on an artificial diet containing powdered leaves of *Plantago lanceolata* as described by Nijhout (1980a). *Vanessa virginiensis* larvae were reared either on fresh leaves of *Antennaria plantaginifolia* or on the artificial diet of Nijhout (1980a) but containing powdered leaves of *Gnaphalium obtusifolium* and other *Gnaphalium spp.*

Fig. 1. Colour pattern on the dorsal forewing of *Vanessa cardui*. A, dorsal forewing wild type; B, ventral forewing wild type; C, dorsal and D, ventral forewing of coldshocked individuals.

B

1A

2A

B

C

D

All animals were reared at 27 °C under a long-day (16L: 8D) photoperiod. Coldshocks were applied by placing synchronous groups of pupae, aged 3–5 h after pupation, in a freezer regulated to maintain -2 ±1 °C for periods described in the text. After the coldshock animals were immediately returned to 27 °C to continue development. A time table for pupal development and colour pattern development in *Precis coenia* is given in Nijhout (1980*b*).

RESULTS

Vanessa cardui

When pupae of *Vanessa cardui* are exposed to a temperature shock many individuals develop aberrant colour patterns on their wings. The temperature shock must be given early during the pupal stage to be effective in inducing an aberrant pattern. For the experiments reported below, coldshocks were given to animals 3–5 h after pupation. Pupae of this age survive coldshock well (mortality <10 %) and show a larger percentage of pattern aberrations than older pupae subjected to the same coldshock regime. It should be noted that the types of pattern aberrations observed when older (15–24 h) pupae were coldshocked were identical to those described below for 3 h- to 5 h-old pupae.

In order to provide a basis for comparison, Figs 1 and 2 illustrate the normal and selected coldshock-altered colour patterns on the forewing and hindwing, respectively. Coldshock, however, produces quite a broad range of aberrant pattern phenotypes. Perhaps the most remarkable feature of these pattern aberrations is the fact that they can be arranged in a single smooth morphocline of progressive deviation from the 'wild type', as shown in Fig. 3. As one's eye sweeps Fig. 3A → H, one easily detects a progressive decrease in the contrasting pattern elements. Fig. 3 focusses on the pattern in the distal half of the hindwing and, for the discussion and analyses that follow, consideration will be limited to the pattern in each of the four wing-cells that bear small ocelli. In the least affected coldshocked patterns (Fig. 3B), the outer dark ring of the two flanking ocelli has opened up distally and now forms a horseshoe-shaped arc around the remainder of the ocellus (labelled (a) in Fig. 3B). The homolog of this ring is not present in the two central ocelli. In addition one observes the first stages of a line of fusion that becomes established along the wing-cell midline between each ocellus and a thin triangular pattern element (arrow in Fig. 3B) that occurs about halfway between the ocellus and the wing margin. In more affected individuals this line of fusion is broader and the distal pattern element 'moves' closer to and appears to be absorbed by the ocellus (Fig. 3C–E). The ocellus then shrinks to a vaguely defined point and then vanishes (Fig. 3E–F).

Fig. 2. Colour pattern on the hindwing of *V. cardui*. A and B, dorsal and ventral surface, respectively, of wild type. C and D, dorsal and ventral surface, respectively, of coldshocked individuals.
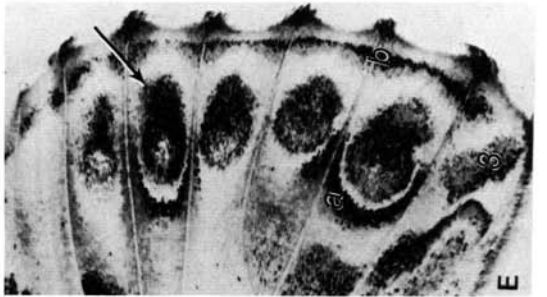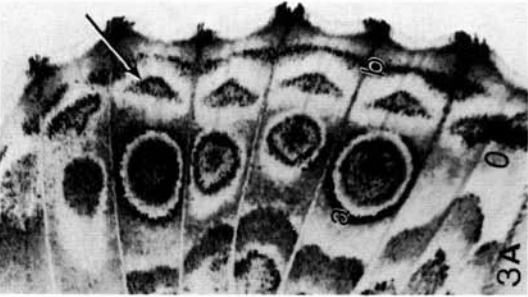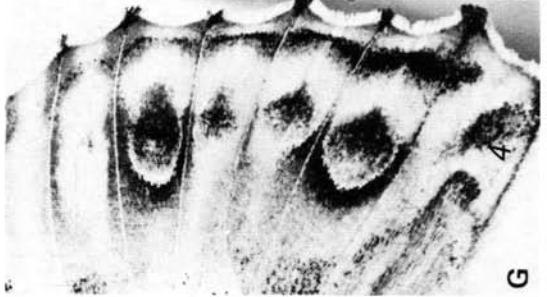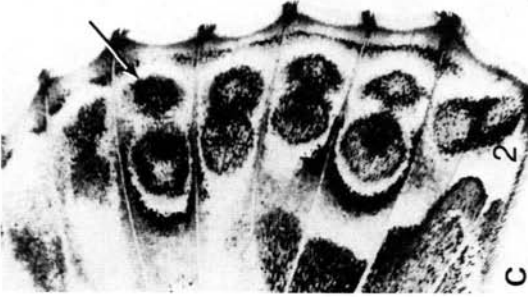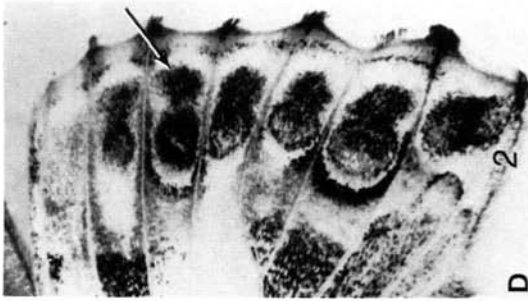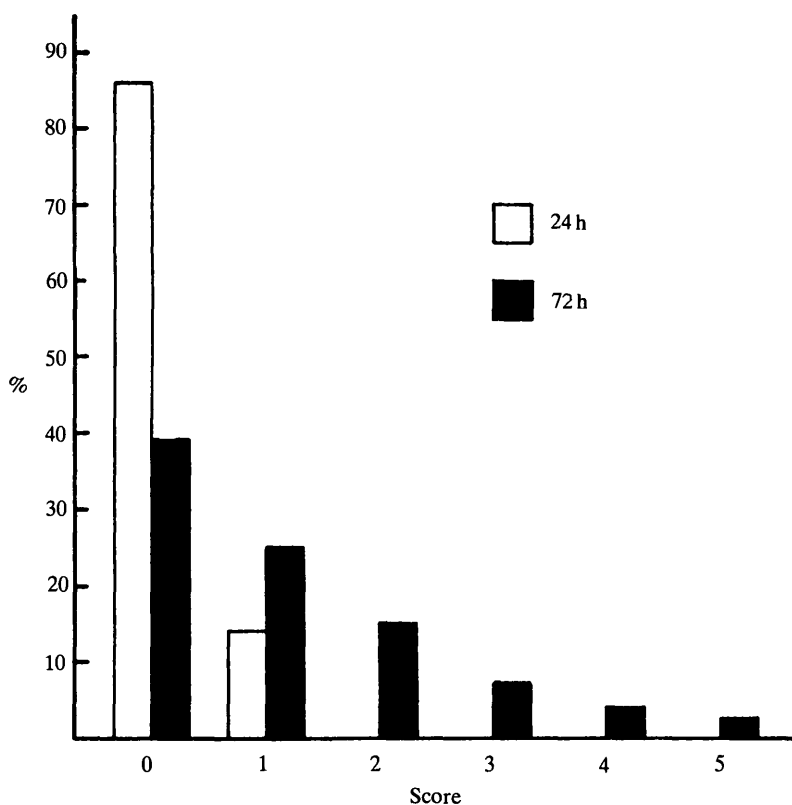
Fig. 4. Frequency-distribution of colour pattern aberrations on the ventral hindwing of *V. cardui* after a 24 h (n = 63) and a 72 h (n = 78) coldshock.

In the later stages of this sequence (Fig. 3G–H) there is also a broadening of the dark submarginal band (labelled (b) in Figs 3B and 3H) that lies close to and parallel to the wing margin. The pattern in each of the four wing-cells follows an identical sequence of aberration, although the changes are not always synchronous (Fig. 3).

Fig. 3. Morphocline of coldshock-induced colour pattern modifications on the ventral hindwing of *V. cardui*. A, wild-type pattern. B–H, pattern aberrations induced by coldshock (72 h at −2 °C) beginning 3–5 h after pupation. Numbers in lower centre of each panel are scoring system for response used in construction of Fig. 4. Animals are assigned to score category of closest resemblance. Because adjacent wing cells do not always show perfectly 'synchronous' pattern modifications and because no two individuals are identical some score categories (e.g. 2 and 3) are illustrated by two patterns which represent the extremes of the range of patterns assigned that score. Arrows indicate homologous pattern elements (the parafocal elements, see Discussion). Here and in Figs 5, 6, 10 and 11 arrows point to homologous pattern elements in homologous wing-cells. Labels refer to pattern elements discussed in text: *a*, peripheral ring of one of the flanking ocelli; *b*, submarginal band.

To quantitate the response to coldshock, the various degrees of pattern aberration were given numerical scores. The scoring system, scaled from 0 (no effect) to 5 (most highly aberrant pattern encountered) is shown superimposed on the corresponding photographs in Fig. 3. The use of such a simple numerical scoring system can be justified by the fact that all pattern aberrations encountered in these and other experiments, over a period of more than 2 years and involving more than 250 individual animals, can be arranged in a single morphological series. The frequency-distribution of the various pattern aberrations induced by a 24 h and a 72 h coldshock are shown in Fig. 4. It is evident that a 24 h coldshock is fairly ineffective in inducing pattern aberrations. Only 14 % of animals are affected and all resemble the pattern given a score of 1 in Fig. 3. A 72 h coldshock, by contrast, induces a broad range of pattern aberrations in about 69 % of the animals and all morphological categories are represented. Both frequency-distributions resemble Poisson distribution (χ^2 test: 24 h pulse, $P > 0.7$; 72 h pulse, $P > 0.1$)

While the response to coldshock is obviously quite variable from individual to individual it should be noted that in these experiments, and in all that follow, the colour pattern on the homologous wing surfaces of any individual (left and right ventral hindwing, for instance) always showed *identical* pattern aberrations. Fig. 5 shows that the colour pattern on the dorsal hindwing develops a similar series of pattern aberrations following coldshock as the ventral hindwing (cf. Fig. 3), though in this case the pattern is significantly simplified because the dorsal pattern is much less elaborate than that on the ventral surface of the hindwing.

Pattern aberrations on the forewing are similar in many ways to those described above for the hindwing. Pattern development, however, appears to be somewhat more canalized (i.e. less susceptible to modification), and, since the pattern in each wing-cell is often quite different from that in adjacent cells, the pattern aberrations give the appearance of being more diverse. Analysis of the forewing pattern will be presented elsewhere in the context of the comparative morphology of colour patterns (Nijhout, in preparation).

Vanessa virginiensis

The colour pattern of this species appears to be very stable, or canalized (see also Shapiro, 1981a), and not easily affected by coldshock. Out of 52 animals exposed to a 72 h coldshock only 6 developed relatively minor pattern aberrations. Fig. 6 shows the wild-type pattern and two of the most affected patterns found, on the ventral wing surface of this species (cf. Figs 2B, D and 3 for homologous areas in *V. cardui*). The sequence of pattern modification shown in Fig. 6 reveals homology between the triangular pattern distal to the focus in *V. cardui* and one of the submarginal pigment bands of *V. virginiensis* (cf. arrows in Figs 3 and 6). The hypothesis of homology is further supported by the fact that both pattern elements are greyish blue in colour, in contrast to the brown hues of the rest of the local pattern. The patterns in Fig. 6 clearly show the early stages
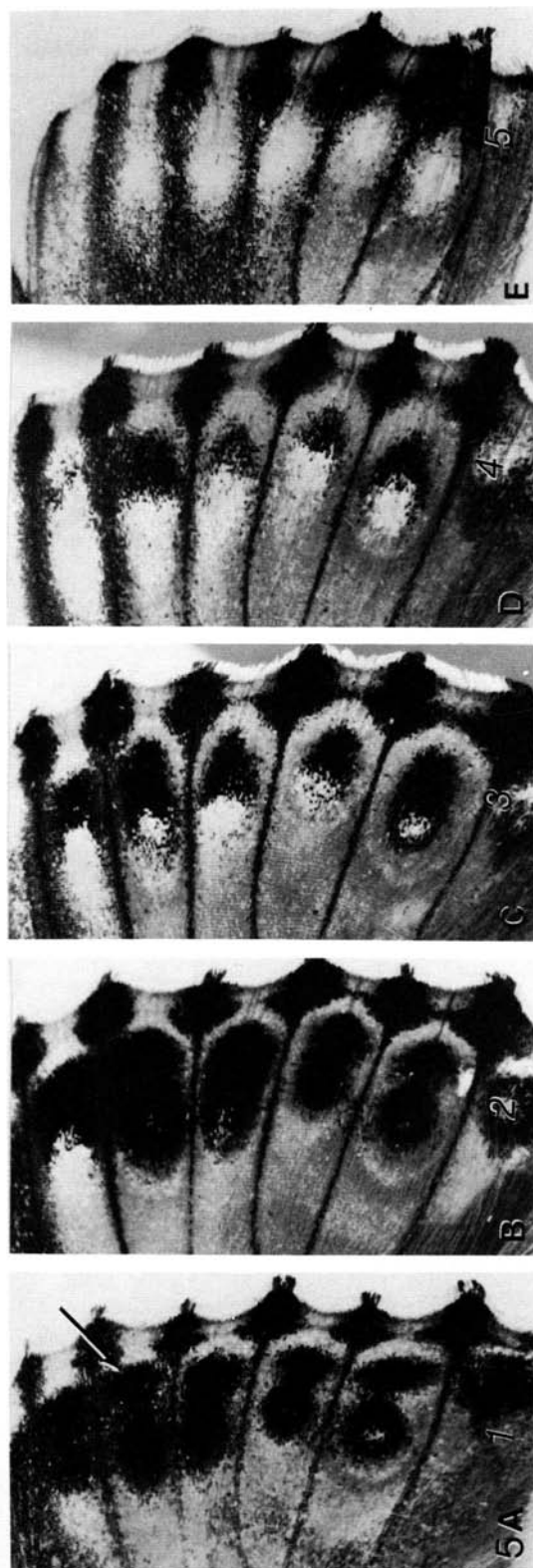
Fig. 5. Morphocline of coldshock-induced pattern modifications on the dorsal hindwing of *V. cardui*. Normal (wild-type) pattern is not illustrated, but see Fig. 2A. Scoring system is used to indicate match to pattern modifications of ventral surface shown in Fig. 3.
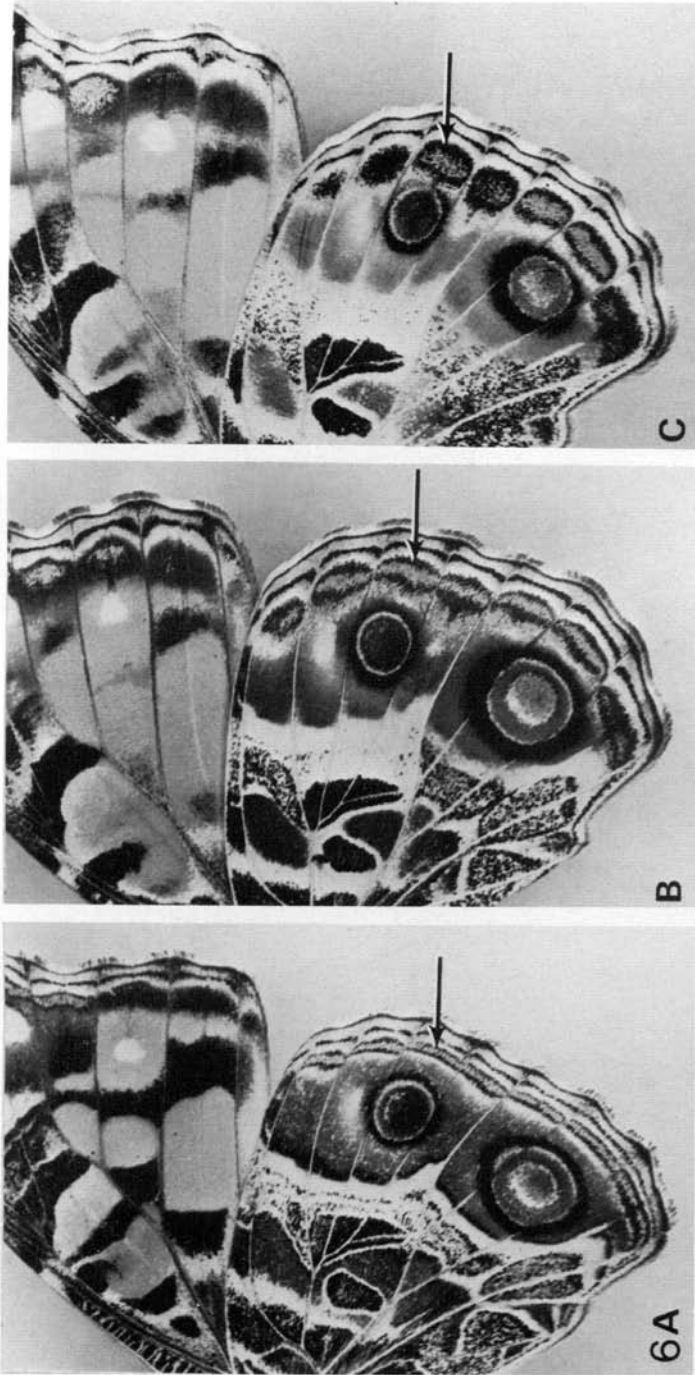
Fig. 6. Coldshock-induced pattern modifications on the ventral wing surface of *Vanessa virginiensis*. A, normal (wild-type) pattern. B, most common pattern modification. C, most extreme pattern modification found. Arrows indicate homologous pattern elements to those shown in Fig. 3.

in the fusion between ocelli and the more distal pattern element also observed in Fig. 3B. Note also that *V. virginiensis* lacks the two central ocelli of *V. cardui*, yet the extension of the distal element along the wing-cell midline towards the presumptive focus is identical to that observed in wing-cell that do bear ocelli.

Precis coenia

The normal colour pattern and selected coldshock-induced pattern aberrations of the forewing of *Precis coenia* are shown in Fig. 7, while Fig. 8 illustrates the hindwing patterns. The pattern of *P. coenia* appears to be somewhat more susceptible to modification by coldshock than that of either of the *Vanessa's*, as evidenced by the significantly higher percentage of affected individuals (Fig. 9). The affected patterns once more form morphological series which are illustrated for the ventral forewing (Fig. 10, which also gives the scoring system used in the construction of Fig. 9) and ventral hindwing (Fig. 11). The frequency-distributions of the aberrant phenotypes of the ventral forewing pattern after coldshocks of 24 h and 72 h duration (Fig. 9) once more resemble Poisson distributions (χ^2 test: 24 h pulse, $P > 0.4$; 72 h pulse $P > .55$).

As in the case of *V. cardui*, the series of pattern modifications represent a progressive reduction of the pattern. The most important features of the series shown in Fig. 10 are as follows. 1) In the least affected individuals the small anterior eyespot, between veins M_1 and M_2 is reduced (Fig. 10B; cf. Figs 8B and 10A) but the remainder of the pattern is little affected. 2) In more affected individuals the small eyespot either disappears or is reduced to a dark point and one observes a progressive reduction of the large eyespot between veins Cu_1 and Cu_2 (Fig. 10C–F). 3) An equally important modification is found in the scalloped (or W-shaped) band that runs about halfway between the focus of the eyespot and the wing margin (arrows in Fig. 10). This band undergoes a change that is very similar to that seen before in the triangular pattern element on the ventral hindwing of *V. cardui* (arrows in Fig. 3).

Fig. 11 illustrates the morphocline for the ventral hindwing pattern of *P. coenia*. The pattern on this wing surface is particularly susceptible to coldshock-induced modification so that the most affected patterns (e.g. Fig. 11E, F) are already found in animals with a ventral forewing score of 3 (Fig. 10). The colour pattern on the dorsal hindwing undergoes a series of quite unusual and dramatic

Fig. 7. Colour pattern on the forewing of *Precis coenia*. A and B, dorsal and ventral surface, respectively, of wild type. C and D, dorsal and ventral surface, respectively, of coldshocked individuals.

Fig. 8. Colour pattern on the hindwing of *P. coenia*. A and B, dorsal and ventral surface, respectively, of wild type. C and D, dorsal and ventral surface, respectively, of coldshocked individuals.
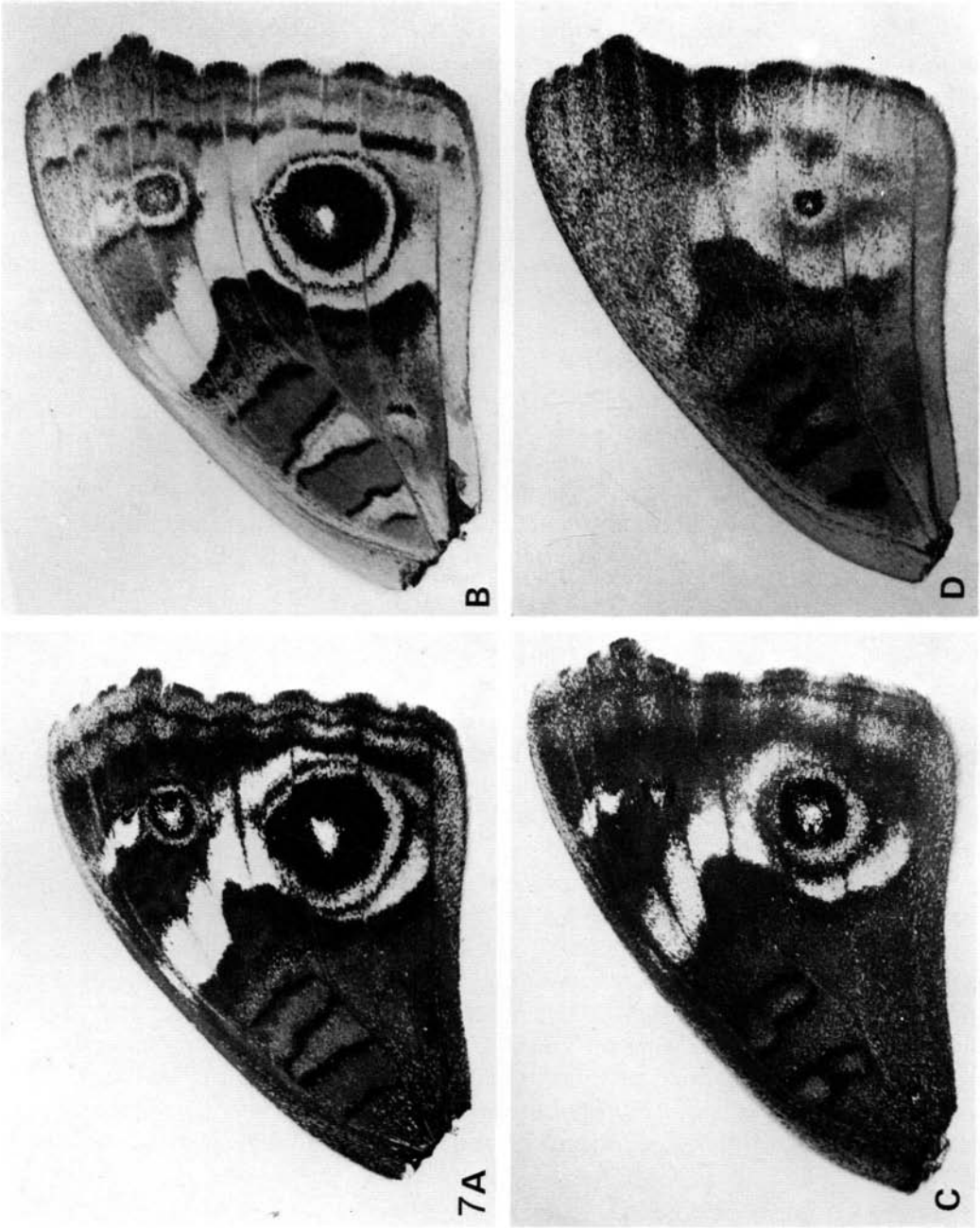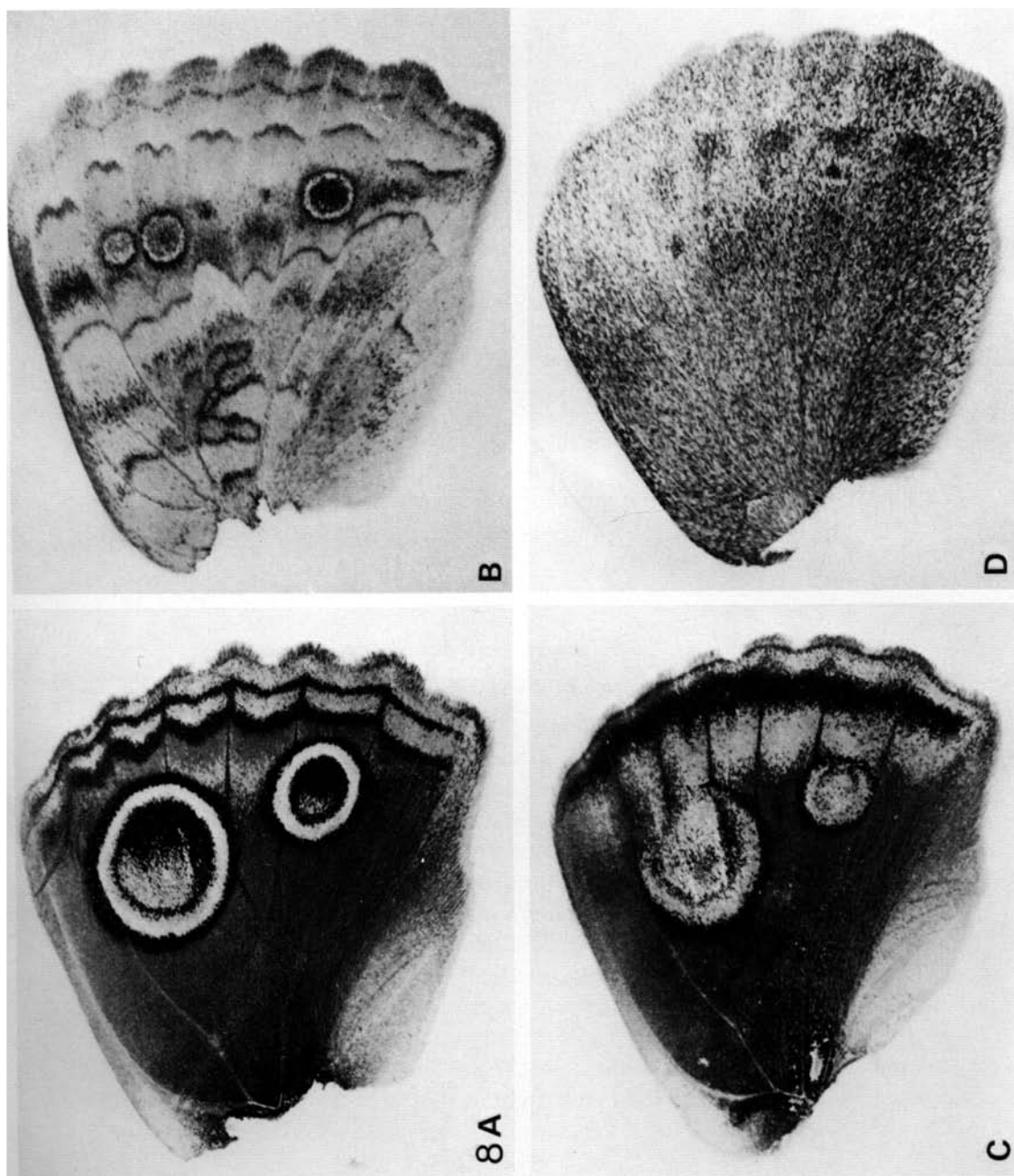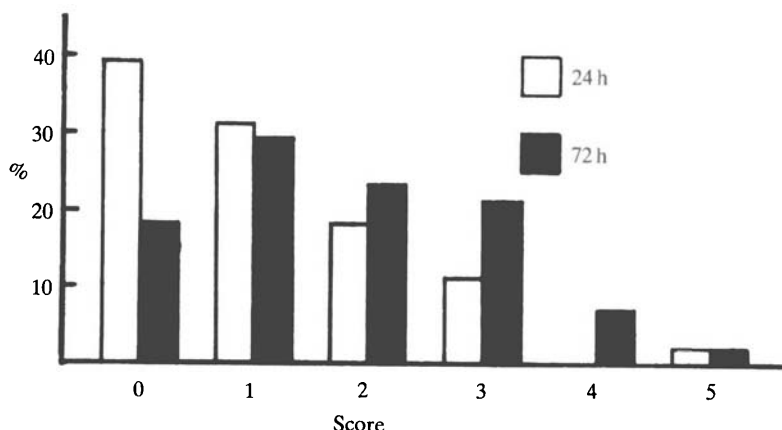
Fig. 8 for legend see p. 295

Fig. 9. Frequency distribution of colour pattern aberrations on the ventral forewing of *P. coenia* after a 24 h (n = 56) and a 72 h (n = 90) coldshock. Scoring system used to construct this figure is shown in Fig. 10.

modifications. These reveal a most interesting process at work in the development of the dorsal wing pattern and will be the subject of a separate communication (Nijhout, in preparation).

DISCUSSION

General features of the response to coldshock

The most striking features of the coldshock-induced pattern aberrations described above, and that apply to all three species studied, are the following. 1) Individuals of a synchronous cohort are affected to very different degrees. 2) For the pattern elements under consideration, the pattern aberrations induced by coldshock can be arranged in a continuous unbranched series. This finding suggests that the difference between adjoining patterns in a series is a quantitative one and implies the operation of a relatively simple process (simple in the sense of involving few and interdependent variables). 3) The frequency distribution of individuals within a series resembles a Poisson distribution, indicating that a rare, stochastic event is instrumental in generating the pattern aberrations. This stochastic nature of the pattern aberrations applies only to comparisons between individuals. 4) Within an individual the response of equivalent patterns on left and right wings is identical. Furthermore, homologous patterns that have the same morphology in the normal wing

Fig. 10. Morphocline of coldshock-induced pattern modifications on the ventral forewing of *P. coenia*. A, wild-type pattern. B–F, pattern aberrations induced by coldshock (72 h at −2 °C) starting 3–5 h after pupation. Numbers at bottom centre of each panel are scoring system used in the construction of Fig. 9. Arrows indicate pattern elements homologous to those shown in Figs 3 and 6.
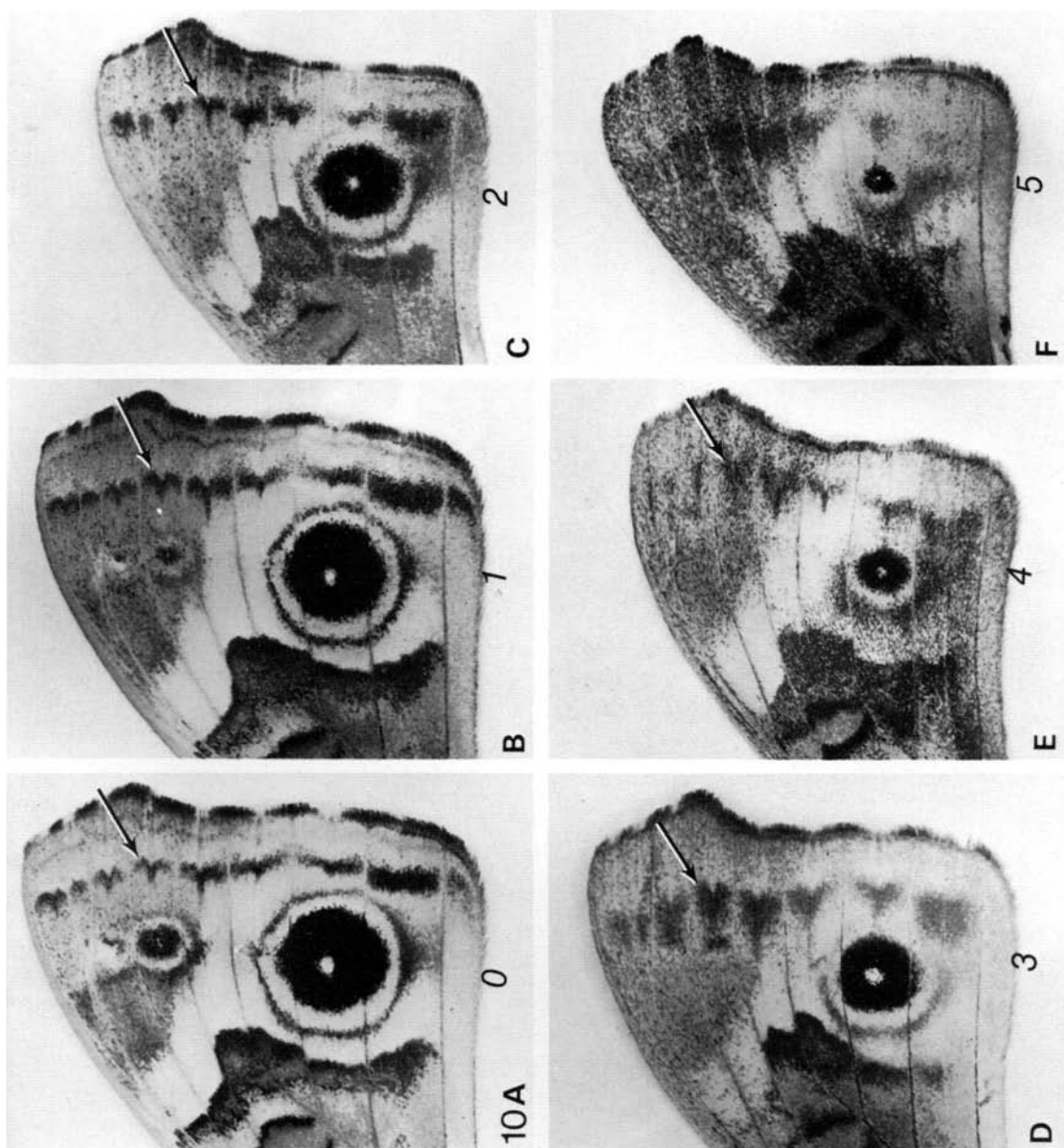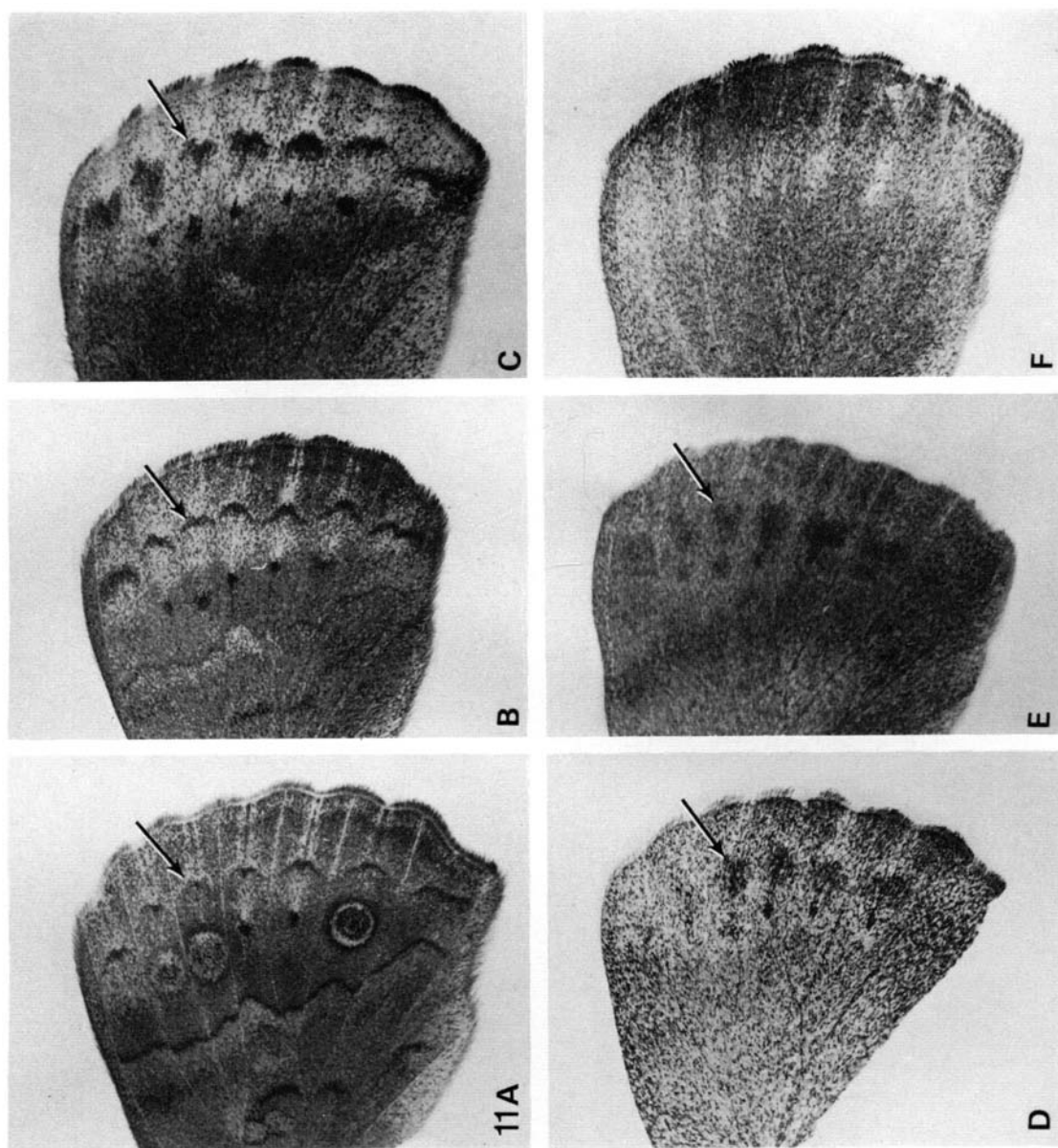
Fig. 10

Fig. 11

(e.g. similar patterns in adjacent wing-cells) become modified in the same way. This finding suggests either that the response to coldshock is mediated by a systemic effect, or that homologous developmental fields within an individual are very similar in their response to coldshock. 5) The percentage of animals affected and, among those, the degree of the effect, are positively correlated with the duration of the exposure to cold. 6) Cold-induced aberrations generally involve a diminution of the pattern though the morphology of this diminution appears to be quite complex. 7) There is an overall darkening (melanization?) of the pattern and a loss of definition, not only because pattern and background become more similar in colour but also because colour bands and spots tend to become broader and their outlines more diffuse in progressively more affected patterns. One or another of these responses to temperature-shock have previously been reported for these and several other species by Kuehn (1926), Kuehn & von Engelhardt (1936) and Shapiro (1975, 1981*a,b*, 1983). For a review of the very early literature on temperature-induced colour pattern aberrations see Prochnow (1929) and Goldschmidt (1938), and for illustrations of many wild caught aberrations, most of which are probably temperature induced and many of which show similarities to the patterns illustrated in the present paper, see Russwurm (1978).

The parafocal pattern elements

A common feature in the morphoclines of both *Precis* and *Vanessa* is the 'fate' of a band-like pattern element roughly halfway between the focus (or ocellus) and the wing margin (arrows in Figs 3, 4, 10 and 11). In both species this band becomes elongated along the cell midline towards the focus and, in the most affected individuals, this entire pattern element shrinks in size and appears to be 'absorbed' into the focus or, when present, into the ocellus that develops around the focus (Figs 3B–G, 4, 10C–F, 11C–F). A similar fusion of an ocellus with such a distal pattern element following temperature shock has been documented in *Argynnis paphia* by Kuehn (1926). Russwurm (1978) has illustrated a large number of virtually identical pattern aberrations from nature in other species of Nymphalidae and also in a number of species of the Satyridae and Lycaenidae. Evidently this particular aberration is easily induced in the butterflies. The above findings and observations strongly suggest that the distal pattern elements of *Vanessa* and *Precis*, while morphologically dissimilar, are homologous. In fact, these distal pattern elements, called parafocal pattern elements (Nijhout, 1984),

---

Fig. 11. Morphocline of coldshock-induced pattern modification on the ventral hindwing of *P. coenia*. A, normal pattern. B–F, pattern aberrations induced by coldshock (72 h at −2 °C) starting 3–5 h after pupation. There is no correspondence to similarly labelled panels in Fig. 10. Patterns A and B are found in animals with a forewing score of 1, C and D in those with a forewing score of 2, E and F in those with a forewing score of 3 or higher. Arrows indicate parafocal elements (see Discussion) homologous to those indicated in Figs 3, 5, 6 and 10.

belong to a category of patterns that is extremely widespread among the butter-flies. They are diverse in the details of their morphology, but have in common a position roughly halfway between focus and wing margin and a bilateral sym-metry of shape with the axis of symmetry running along the wing-cell midline.

A simple hypothesis to account for the observation that parafocal pattern elements tend to 'fuse' with a focus, or with the pattern generated around a focus (an ocellus), is that the focus is somehow involved in the induction of the parafocal element much in the way it is involved in the induction of perifocal patterns such as ocelli (Nijhout, 1980a). But this is certainly not the whole story because, while focal cautery can abolish development of an ocellus, it has no effect on the morphology of the adjoining parafocal element (Nijhout, 1980a). By contrast, cauteries at the wing margin do cause an abnormal development of the nearest parafocal element (Nijhout, unpublished), and indicate the presence of a pattern-organizing function in that region of the wing-cell. It is unlikely, however, that the morphological continuity of the parafocal element with a focus is accidental since it is observed repeatedly, even in unrelated species. The functional relations of the parafocal elements to the focus and to the presumptive marginal signalling source are currently under investigation.

Field boundaries revealed?

The various elements of the colour pattern of *Vanessa* and *Precis* differ in their response to coldshock. In *Vanessa*, for instance, much of the pattern on the proximal half of the wing remains constant even in specimens where other aspects of the colour pattern are highly affected. In *Precis*, likewise, elements like the red bars in the discal cell of the forewing are quite stable and normal in appearance even in cases when the remainder of the pattern is virtually obliterated. Such stability is probably due to the fact that these pattern elements have already been determined by the time the temperature shock is applied (Kuehn, 1926; Kuehn & von Engelhardt, 1933, 1936; Nijhout, 1980a,b).

A particularly interesting partial stability of pattern is seen on the ventral hindwing of *Vanessa* (Fig. 3) in which the outer pigment ring of the two lateral eyespots first becomes modified into a horseshoe shape, becomes slightly broader, but then maintains a stable morphology while the remainder of the eyespot becomes progressively modified and eventually vanishes. The narrow pigment band that runs parallel and close to the wing margin undergoes a similar modification. It does not vanish, as do the ocellus and adjacent parafocal element, but it broadens considerably and changes shape so that in the most affected pattern (Fig. 3H) its proximal outline coincides precisely with what is 'normally' the distal outline of the parafocal element. Together, the horseshoe-shaped band and marginal bar form a perfect outline of the field in which ocellus and parafocal element normally develop (cf. Figs 3A and H). Evidently these highly aberrant patterns are obeying the same set of boundaries from without, as the normal pattern obeys from within. Equivalent pattern 'outlines' are not

expressed on the dorsal hindwing nor on the wings of *Precis coenia*. Other species in the genus *Precis*, however, have distinctly different pigments proximal and distal to their parafocal pattern element, suggesting that there too the parafocal element lies on or adjacent to a boundary between two regions with different prospective fates for pigment synthesis.

Significance of the pattern morphoclines

Perhaps the most remarkable feature of the pattern morphoclines illustrated in Figs 3, 4, 6, 10 and 11 is that they are linear. Not only are the series unbranched but if one were to construct a morphocline based only on part of the pattern it would be congruous with the one based on the pattern as a whole. This observation is non-trivial because it implies that a given morphocline results from variation in a single determinant of pattern. If more than one determinant had been affected independently by the coldshock the morphoclines of pattern aberrations would be expected either to branch or to have non-congruent or discordant elements. Of course, it is possible that more than one process is affected by the coldshock, but the linearity of the morphological response indicates that if multiple determinants of pattern were affected, they varied in a perfectly co-ordinated manner and thus behaved as a single variable.

I have shown elsewhere (Nijhout, 1978, 1980a, 1981) that the theory of positional information developed by Wolpert (1971) provides a particularly useful conceptual framework for the study and experimental analysis of colour patterns and their development. It may be useful, therefore, to phrase an interpretation of the findings reported here in terms of positional information. Two alternative interpretations are possible.

We know that in *Precis coenia* (Nijhout, 1980a) and in *Ephestia kuehniella* (Kuehn & von Engelhardt, 1933) pattern determination depends on the emission of signals from discrete sources. In *Precis* this signal propagates from cell to cell with a dynamics that resembles diffusion (Nijhout, in preparation). Such signals provide positional cues and positional value would be a function of signal strength (Nijhout, 1978, 1984). Coldshock could simply affect the dynamics of synthesis and/or propagation of one of the positional signals, yielding an 'aberrant' gradient. Pattern morphoclines could therefore represent the succession of patterns that is specified when a quantitative parameter of one of the positional signals is systematically altered. The alternative interpretation is that the positional signals remain unaltered, but that coldshock somehow affects the biochemical process whereby positional signals provoke their cellular effect. If this were the case, morphoclines could represent a series of progressively different interpretations of the same positional information. It might be possible to differentiate between these two possible mechanisms by studying the effects of coldshock in genetic mosaics. In Lepidoptera we have, as yet, not the suitable means of providing such a demonstration.

Colour pattern modifications in Lepidoptera show many similarities to

phenocopies in *Drosophila*. As is the case with phenocopies, identical colour pattern aberrations can be induced by diverse non-specific traumas such as heat-shock, coldshock and cautery, and the pattern modification that is obtained depends on the timing of the inducing stimulus, not on the nature of the stimulus (Prochnow, 1929; Kuehn & Henke, 1936; Kuehn & von Engelhardt, 1936). In *Ephestia kuehniella*, temperature shock-induced pattern modifications phenotypically resemble certain wing pattern mutants (Kuehn & Henke, 1936). This finding, together with the observation that temperature shock-induced pattern modifications may resemble the normal pattern of certain geographic races of the species in question, led Goldschmidt (1938) to designate these colour pattern modifications as phenocopies, analogous to those found in *Drosophila* (Goldschmidt, 1935*a,b*). Since the pattern morphoclines can be ascribed to variation in a single parameter, they are probably analogous to the variation in penetrance and expressivity that is commonly encountered in *Drosophila* phenocopies (Goldschmidt, 1935*a,b*; Gloor, 1947).

In contrast to the situation in *Drosophila*, the pattern modifications reported here cannot be compared to mutant phenotypes because a genetics of colour pattern is unavailable for any of the species studied. The pattern modifications do, however, show a remarkable resemblance to the *normal* pattern phenotypes in other species of the genera *Vanessa* and *Precis* (see Nijhout (1984) for illustrations). The temperature-induced pattern modifications described above may thus be viewed as interspecific phenocopies. They could, therefore, prove to be useful tools in the discovery of pattern homologies and in elucidating the evolution of colour patterns by revealing what kind of morphologies are developmentally 'adjacent' to one another, and by revealing the kind of variability that is available for genetic assimilation (Waddington, 1953; Rachootin & Thomson, 1981; Shapiro, 1983). An observation that is of interest in this regard is that the phenotypic morphocline of *V. virginiens* (Fig. 6C → A) can be interpreted as an extension (with allowance for a few species specific differences) of the morphocline of *V. cardui* (Fig. 3H → B). This continuity in the morphoclines of two different species (and see Nijhout (1984) for additional examples) reveals not only that one of the submarginal bands of *V. virginiensis* is homologous to the parafocal element of *V. cardui* but also that much of the interspecific diversity of this pattern element may come about through variation in a single determinant of pattern.

REFERENCES

GLOOR, H. (1947). Phaenokopie-Versuche mit Aether an *Drosophila*. *Rev. Suisse Zool.* **54**, 637–712.

GOLDSCHMIDT, R. (1935*a*). Gen und Auszeneigenschaft (Untersuchungen an *Drosophila*. I. *Z. Indukt. Abstamm.-u. VererbLehre* **69**, 38–69.

GOLDSCHMIDT, R. (1935*b*). Gen und Auszeneigenschaft (Untersuchungen and *Drosophila*). II. *Z. Indukt. Abstamm.-u. VererbLehre* **69**, 70–131.

GOLDSCHMIDT, R. (1938). *Physiological Genetics*. New York: McGraw-Hill.

KUEHN, A. (1926). Ueber die Aenderungen des Zeichnungsmusters von Schmetterlingen durch Temperaturreize und das Grundschema der Nymphalidenzeichnung. *Nachr. Gesellsch. Wiss. Goettingen. Math.-Phys.* KI, 120–141.

KUEHN, A. & HENKE, K. (1936). Genetische und Entwicklungsphysiologische Untersuchungen an der Mehlmotte *Ephestia kuehniella* Zeller. *Abh. Gesellsch. Wiss. Goettingen. Math.-Physik KI. N.F.* **15**, 1–272.

KUEHN, A. & VON ENGELHARDT, M. (1933). Ueber die Determination des Symmetriesystems auf dem Vorderfluegel von *Ephestia kuehniella*. *Willhelm Roux Arch. EntwMech. org.* **130**, 660–703.

KUEHN, A. & VON ENGELHARDT, M. (1936). Ueber die Determination des Fluegelmusters bei *Abraxas grossulariata* L. *Ges. Wiss. Nachr. Biol.* **2**, 171–199.

NIJHOUT, H. F. (1978). Wing pattern formation in Lepidoptera: A model. *J. exp. Zool.* **206**, 119–136.

NIJHOUT, H. F. (1980*a*). Pattern formation on lepidopteran wings: determination of an eye-spot. *Devl Biol.* **80**, 267–274.

NIJHOUT, H. F. (1980*b*). Ontogeny of the color pattern on the wings of *Precis coenia* (Lepidoptera: Nymphalidae). *Devl Biol.* **80**, 275–288.

NIJHOUT, H. F. (1981). The color patterns of butterflies and moths. *Sci. Amer.* **245**(5), 145–151.

NIJHOUT, H. F. (1984). The developmental physiology of colour patterns in Lepidoptera. *Adv. Insect Physiol.* (in press).

PROCHNOW, O. (1929). Die Faerbung der Insekten. In *Handbuch der Entomologie*, Vol. II, pp. 430–591, (ed. C. Schroeder). Jena: Fischer Verlag.

RACHOOTIN, S. P. & THOMSON, K. S. (1981). Epigenetics, paleontology and evolution. In *Evolution Today. Proc. 2nd Int. Congr. Syst. Evol. Biol.*, pp. 181–193, (eds G. G. E. Scudder & J. L. Reveal). Pittsburgh: Hunt Institute for Botanical Documentation.

RUSSWURM, A. D. A. (1978). *Aberrations of British Butterflies*. Faringdon: Classey.

SCHWANWITSCH, B. N. (1924). On the groundplan of the wing pattern in Nymphalids and certain other families of Rhopalocera. *Proc. Zool. Soc. London* **34**, 509–528.

SHAPIRO, A. M. (1975). Natural and laboratory occurrences of "*elymi*" phenotypes in *Cynthia cardui* (Lepidoptera: Nymphalidae). *J. Res. Lepid.* **13**, 37–62.

SHAPIRO, A. M. (1981*a*). A new record of *Vanessa virginiensis* "*ab. ahwashtee*" from Northern California (Lepidoptera: Nymphalidae). *J. Res. Lepid.* **20**, 176–178.

SHAPIRO, A. M. (1981*b*). Phenotypic plasticity in temperate and subarctic *Nymphalis antiopa* (Nymphalidae): Evidence for adaptive canalization. *J. Lepid. Soc.* **35**, 124–131.

SHAPIRO, A. M. (1983). Testing visual species recognition in *Precis* (Lepidoptera: Nymphalidae) using a cold-shock phenocopy. *Psyche* **90**, 59–65.

SUEFFERT, F. (1927). Zur vergleichende Analyse der Schmetterlingszeichnung. *Biol. Zentralbl.* **47**, 385–413.

WADDINGTON, C. H. (1971). Genetic assimilation of an acquired character. *Evolution* **7**, 118–126.

WOLPERT, L. (1971). Positional information and pattern formation. *Curr. Top. devl Biol.* **6**, 183–224.