J. Embryol. exp. Morph. **85**, 95–109 (1985)
Printed in Great Britain © *The Company of Biologists Limited 1985*

Pattern regulation in fragments of *Drosophila* wing discs which show variable wound healing

LESLIE DALE* AND MARY BOWNES
Department of Molecular Biology, University of Edinburgh, Kings Buildings, Mayfield Road, Edinburgh EH9 3JR, U.K.

SUMMARY

When complementary fragments of the imaginal wing disc of *Drosophila* are cultured for several days prior to inducing metamorphosis, usually one fragment will regenerate while the second duplicates. It has been proposed that wound healing plays an important part in disc regulation by initiating cell proliferation and determining the mode of regulation (regeneration/duplication). To test the latter proposal 15 types of wing disc fragments were examined for variability both in the mode of wound healing and the mode of pattern regulation. Two modes of wound healing were observed, regular – the two wound edges heal with each other, and irregular – each wound edge heals with itself. When cultured separately fragments that healed regularly regenerated, while fragments that healed irregularly duplicated. This suggests that the mode of wound healing determines the mode of pattern regulation.

INTRODUCTION

Recent models of epimorphic pattern regulation propose that local interactions between cells from formerly disparate positions stimulate intercalation of those intermediary positional values requiring minimal regulation (French, Bryant & Bryant, 1976; Cummings & Prothero, 1978; Russell & Hayes, 1980; Kauffman & Ling, 1981; Lewis, 1981). Evidence in favour of this suggestion is provided by grafting experiments on the larval cockroach limb; the structures regenerated are determined by the normal location of the cells confronted at the host–graft junction (Bohn, 1970*a*,*b*; Bullière, 1971; French, 1976, 1978, 1980). Different host–graft combinations stimulate alternative directions of regeneration in both tissues.

When complementary fragments of *Drosophila* imaginal discs are allowed a period of growth, then generally one fragment will regenerate while the second duplicates (Schubiger, 1971; Bryant, 1971, 1975*a*,*b*; van der Meer & Ouweneel, 1974; Bryant & Hsei, 1977; Littlefield & Bryant, 1979*a*). In these fragments growth is epimorphic, dividing cells being localized at the wound edge (Ulrich, 1971; Postlethwait, Poodry & Schneiderman, 1971; Dale & Bownes, 1980;

* *Present address*: Imperial Cancer Research Fund, Mill Hill Laboratories, Burtonhole Lane, Mill Hill, London NW7 1AD, U.K.
Key words: Wound healing, pattern regulation, imaginal discs, *Drosophila melanogaster*.

Abbott, Karpen & Schubiger, 1981). Pattern regulation is preceded by wound healing which apposes cells from disparate positions in the disc (Reinhardt, Hodgkin & Bryant, 1977; Reinhardt & Bryant, 1981).

We have shown that wound healing is required for the initiation of pattern regulation in the imaginal wing disc of *Drosophila* (Dale & Bownes, 1981). However, it has not been established whether the new structures generated by intercalation are determined by the normal location of apposed cells. If this is the case then variations in the mode of wound healing should lead to variations in the regulative capacity of disc fragments. Pattern regulation in imaginal disc fragments is indeed variable; in many fragments duplication of structures predicted from the fate map is accompanied by regeneration of others (Schubiger, 1971; van der Meer & Ouweneel, 1974; Bryant, 1975*a*; Duranceau, Glenn & Schneiderman, 1980; Kauffman & Ling, 1981; Karlsson & Smith, 1981; Kirby, Bryant & Schneiderman, 1982) and a number of wing disc fragments are capable of alternatively regenerating or duplicating (Karlsson, 1981; Kauffman & Ling, 1981).

Reinhardt *et al.* (1977) have described two modes of wound healing which occur in imaginal disc fragments: regular, in which two wound edges heal with each other and irregular, in which two wound edges heal back upon themselves. If the structures produced in pattern regulation depend upon precisely which cells are apposed then these two modes of wound healing should result in the intercalation of different intermediary structures. We have examined 15 wing disc fragments for variations in both wound healing and pattern regulation and we show that regeneration and duplication in these fragments are associated with different modes of wound healing.

MATERIALS AND METHODS

Wild-type larvae and adults of the Barton strain of *Drosophila melanogaster* were used both as donors and hosts. Stocks were maintained at 25 °C under sterile conditions on autoclaved David's medium (Sang, 1978) containing antibiotics (Penicillin, 50 μg/ml; streptomycin, 30 μg/ml; Tetracyclin 50 μg/ml; Kanamycin 50 μg/ml; only two antibiotics were added at any time, but they were rotated regularly).

Wing discs were dissected from mature third instar larvae, 120±4 h after oviposition, into a Ringer solution (Chan & Gehring, 1971) containing antibiotics (Penicillin, 60 μg/ml; streptomycin, 100 μg/ml; Gentamycin, 40 μg/ml; Kanamycin, 50 μg/ml; only two antibiotics were added at any one time). They were then cut into the required fragments (Fig. 2) using tungsten needles. Fragments were either injected directly into mature third instar larvae to induce metamorphosis immediately, or cultured in the abdomen of adult female hosts for 6 days prior to inducing metamorphosis by reimplanting the fragment into a larval host (Usprung, 1967). This allowed the regulative capabilities of these fragments to be established. All fragments (Fig. 2) cultured in adult female hosts were isolated after two days of culture and examined under the dissecting microscope to determine the mode of wound healing. Each disc was then implanted into a second adult female host and cultured for a further 4 days. All operations were performed under sterile conditions. Implants were dissected from newly eclosed flies in 70 % alcohol and mounted between coverslips in Gurr's water mounting medium.

RESULTS

For the 15 types of fragment examined (Fig. 2) three classes of wound healing were identified: (i) regular, the two wound edges healed together (Fig. 3A); (ii) irregular, each wound edge healed back upon itself (fig. 3B); (iii) indistinguishable, the precise mode of wound healing could not be distinguished. Regular and irregular healing modes had previously been suggested by Reinhardt *et al.* (1977), however to our knowledge this is the first report in which irregular healing has been observed. Furthermore Reinhardt *et al.* (1977) suggested that irregular wound healing would be transient, the regular healing mode being quickly established. Close examination of fragment A after successive days of culture and fragments B and C after 6 days of culture shows that in these fragments irregular healing is not transient but permanent.

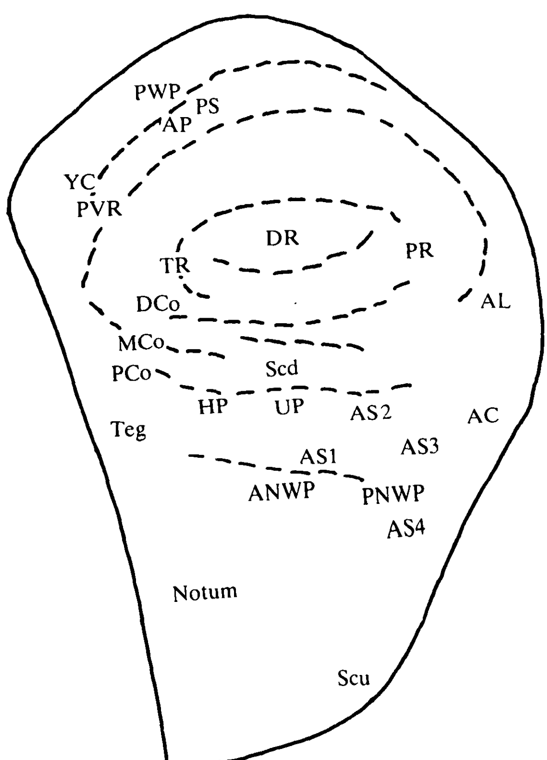

Fig. 1. Schematic diagram of the late third instar wing disc indicating the position of presumptive adult cuticular structures (modified from Bryant, 1975*a*). Abbreviations: Scu, scutellum; ANWP, anterior notal wing process; PNWP, posterior notal wing process; UP, unnamed plate; AS1–4, axillary sclerites 1–4; Scd, dorsal sensilla companiformia; HP, humeral plate; Teg, tegula; PCo, proximal costa; MCo, medial costa; DCo, distal costa; TR, triple row of bristles; DR, double row of bristles; PR, posterior row of bristles; AL, alar lobe; AC, axillary cord; YC, yellow club; PVR, proximal ventral radius; PWP, pleural wing process; PS, pleural sclerite; AP, axillary pouch.

The frequency of the three classes of wound healing for each of the 15 types of fragment used in this study is presented in Table 1. For three of these fragments (A, B and C; Fig. 2) each wound healing class was subsequently cultured separately. The results of scoring the metamorphosed implants of fragments A, B and C following regular and irregular healing are presented in Table 2. It is clear from these results that the mode of pattern regulation in these fragments is dependent upon the mode of wound healing. In these three fragments regular wound healing (Figs 4B and E) results in the regeneration of those structures removed by fragmentation. The only exception was one implant of fragment A which in addition to single copies of HP, PCo and M/DCo (Fig. 1), structures almost always deleted by fragmentation (Table 2), also differentiated duplicated copies of the AS1 and AS2 (Fig. 1). Irregular wound healing resulted in variable pattern regulation but the frequency of duplication was greater than that found following regular wound healing (Table 2). The frequency of duplication in

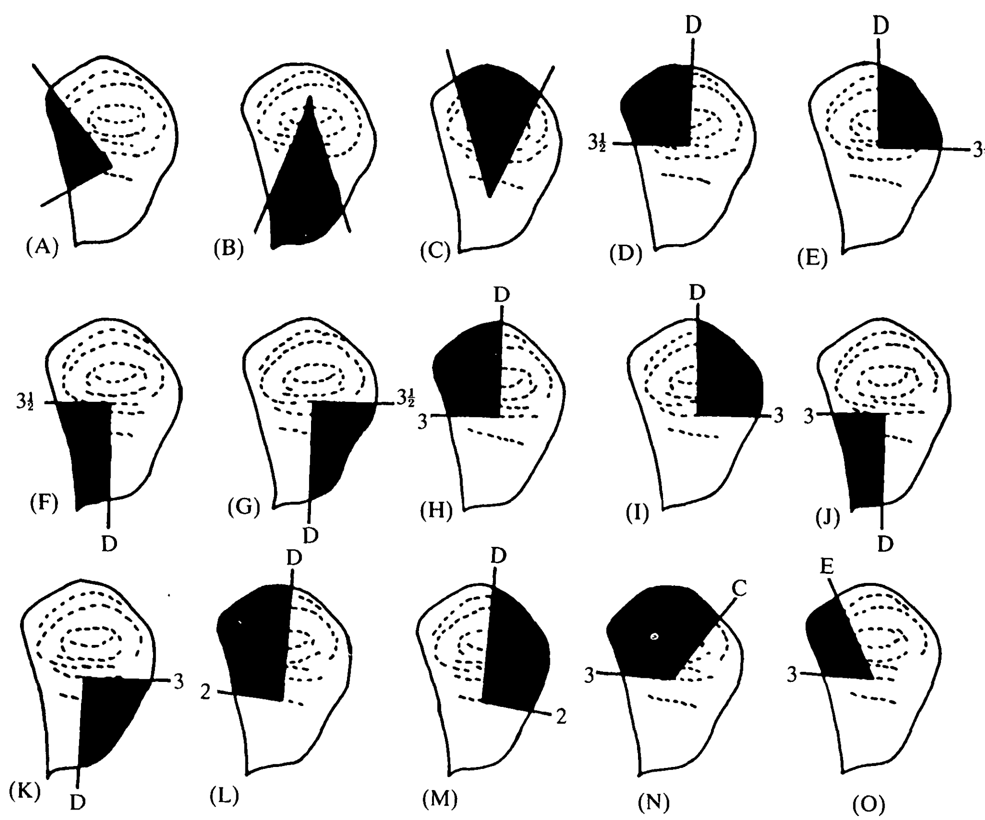

Fig. 2. Schematic diagram of the late third instar wing disc indicating the position of cuts (solid lines) used to generate the fragments used in this report. Nomenclature according to Bryant (1975*a*). Shaded regions were discarded after cutting leaving the experimental fragments. In the text and tables fragments are named after the figure letter.

fragment A was 42 % (8/19), only 16 % (3/19) regenerated and two of these also duplicated ventral hinge structures (PWP, PS, AP). Following irregular wound healing in fragment B equal frequencies of regeneration and duplication were observed, 24 % (4/17), both often occurring in the same fragment, 35 % (6/17). Similarly in fragment C 20 % (4/20) of implants had regenerated either one or two dorsal hinge structures (UP, Scd), 30 % (6/20) duplicated (AS3, AC, Teg, YC, PVR) and a further 30 % both duplicated (AS3, AC, AL) and regenerated (UP, Scd, DR, PWP, PS, AP). Pattern regulation in all these fragments was usually extensive, many structures far from the wound edge being regenerated or duplicated. Indistinguishable wound healing in these three fragments usually results in regeneration although duplication is also frequent (data not shown; Dale, 1983). These results suggest that variations in the mode of wound healing are responsible for the variable pattern regulation observed in fragments A, B and C.

The results of culturing the remaining 12 types of fragment are consistent with this conclusion. Table 1 shows that for six of these fragments (D, E, G, I, J, K;

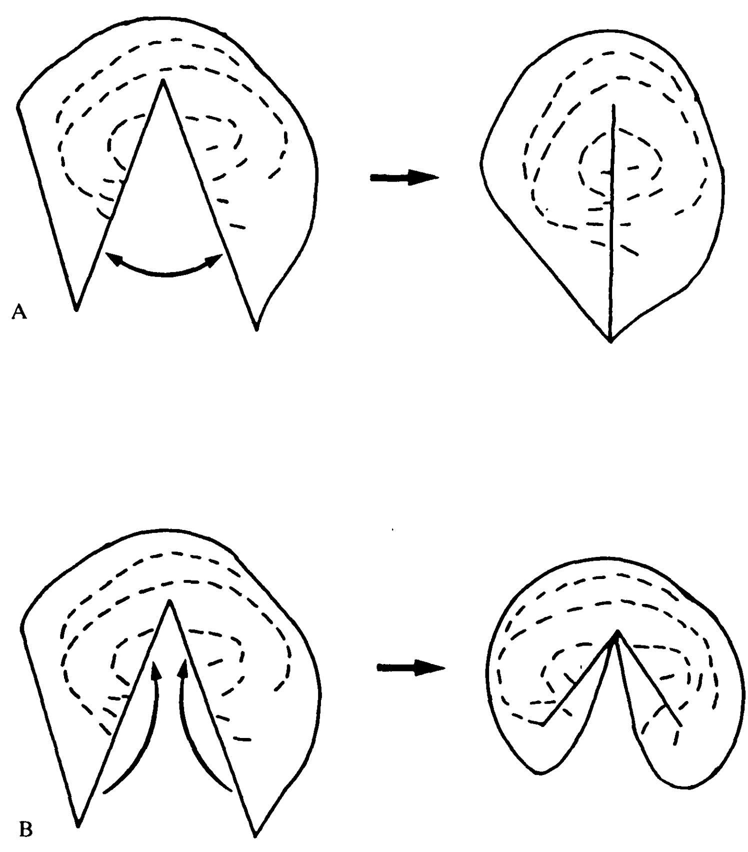

Fig. 3. Schematic diagram of fragment B (Fig. 2B) demonstrating (A) regular wound healing and (B) irregular wound healing.

Table 1. *Frequency of regular (Reg), irregular (Irreg) and indistinguishable (Ind) wound healing in fragments of the wing disc*

Fragment	Reg*	Irreg*	Ind*	n
A	30	31	39	413
B	19	35	47	101
C	9	51	40	198
D	88	0	12	32
E	86	0	14	29
F	16	11	73	44
G	76	4	20	25
H	14	11	75	36
I	80	0	20	30
J	81	0	19	29
K	82	0	18	28
L	6	13	81	31
M	4	14	82	28
N	0	100	0	22
O	29	24	47	34

n = number of fragments scored.
* = frequency in each class expressed as a percentage.

Fig. 2) regular healing (Fig. 5B) was the most common mode of wound healing. All six of these fragments regenerated extensively following culture, duplication being very rare (Tables 3 and 4). For one type of fragment (N) the mode of wound healing was identified as irregular in all cases and duplication was observed in all those implants that regulated (Table 5). Only two examples of regeneration were observed in this last fragment (M/DCo, TR) and both also duplicated extensively (Not, Scut, Teg, HP, UP, AS1, AS2, AS3, AL, Ac). The only other fragment in which wound healing could be clearly identified in a large number of cases was fragment 0. Regular and irregular healing were observed with approximately equal frequency in this fragment (Table 1) and following culture equal frequencies of both regeneration and duplication were observed (Table 5). A further three implants regenerated extensively and duplicated a small number of structures. For the remaining four types of fragment (F, H, L, M), the mode of wound healing could not be clearly distinguished in the majority of cases. All four classes of fragment are capable of both regeneration and duplication, three of these predominantly duplicate (L, H, F) while one predominantly regenerates (M; Tables 3–5).

DISCUSSION

The results presented in this paper demonstrate that in the imaginal wing disc of *Drosophila* different modes of wound healing result in the formation of different structures during pattern regulation. This is most clearly demonstrated by fragment A in which the two modes of wound healing described by Reinhardt *et al.*

Table 2. *Percentage of structures formed by fragments A, B and C following regular (Reg) and irregular (Irreg) wound healing*

Structures scored	Fragment/Mode of wound healing								
	A			B			C		
		Reg	Irreg		Reg	Irreg		Reg	Irreg
	Culture period (days)								
	0	6	6	0	6	6	0	6	6
Notum*	100	100	100(21)	0	89	53	100	100	100
Scut	84	94	79(21)	74	78	53	100	100	100
ANWP	41	76	16	0	78	53	14	100	30
PNWP	73	82	37	0	78	53	45	100	45
UP	49	76	5	0	89	59	0	100	50
AS1	80	82(6)†	74(21)	0	89	59	36	100	65
AS2	73	82(6)	11	0	89	59	18	100	35
AS3	94	94	74(26)	100	100	94(47)	95	100	75(60)
AS4	80	76	58(21)	94	67	53(24)	95	67	60(20)
Scd	39	59	21(5)	0	89	53	0	100	20
HP	4	53	5	0	67	53	14	100	40
Teg	22	65	26	100	100	71(6)	91	100	95(20)
PCo	0	59	16	100	78	88	82	100	65
M/DCo	6	41	0	100	78	94	64	100	40
TR	82	88	74(5)	100	100	71	50	67	45
DR	92	59	84(5)	44	56	53	0	100	5
PR	90	65	32	100	78	76	50	67	35
AL	94	59	32(11)	89	78	71(12)	95	100	50(10)
AC	78	79	63	100	89	94(18)	100	100	60(20)
Wing	100	100	100	100	100	100	100	100	80
YC	61	50	32(16)	100	78	88(18)	45	100	35(10)
PVR	67	67	32(16)	100	78	88(18)	45	100	35(10)
PWP	90	94	84(42)	100	89	94(18)	0	67	30
PS	96	94	84(42)	100	89	94(18)	0	33	20
AP	96	94	84(42)	100	78	94(18)	0	33	20
No. of implants	51	17	19	18	9	17	22	3	20

* abbreviations as in Fig. 1.
† figures in brackets frequency of duplication.

(1977) can easily be distinguished. Reinhardt *et al.* have suggested that these two modes of wound healing, in accordance with recent models of epimorphic pattern regulation (French *et al.* 1976; Kauffman & Ling, 1981; Lewis, 1981) should result in the formation of different structures during intercalation. An examination of Table 2 confirms that this is the case; regular healing results in regeneration of those structures removed by fragmentation, while irregular healing results in duplication of those structures present in the fate map of the fragment. It is clear that fragment A is capable of forming alternative structures during intercalation and that it is the mode of wound healing which determines which ones are formed.

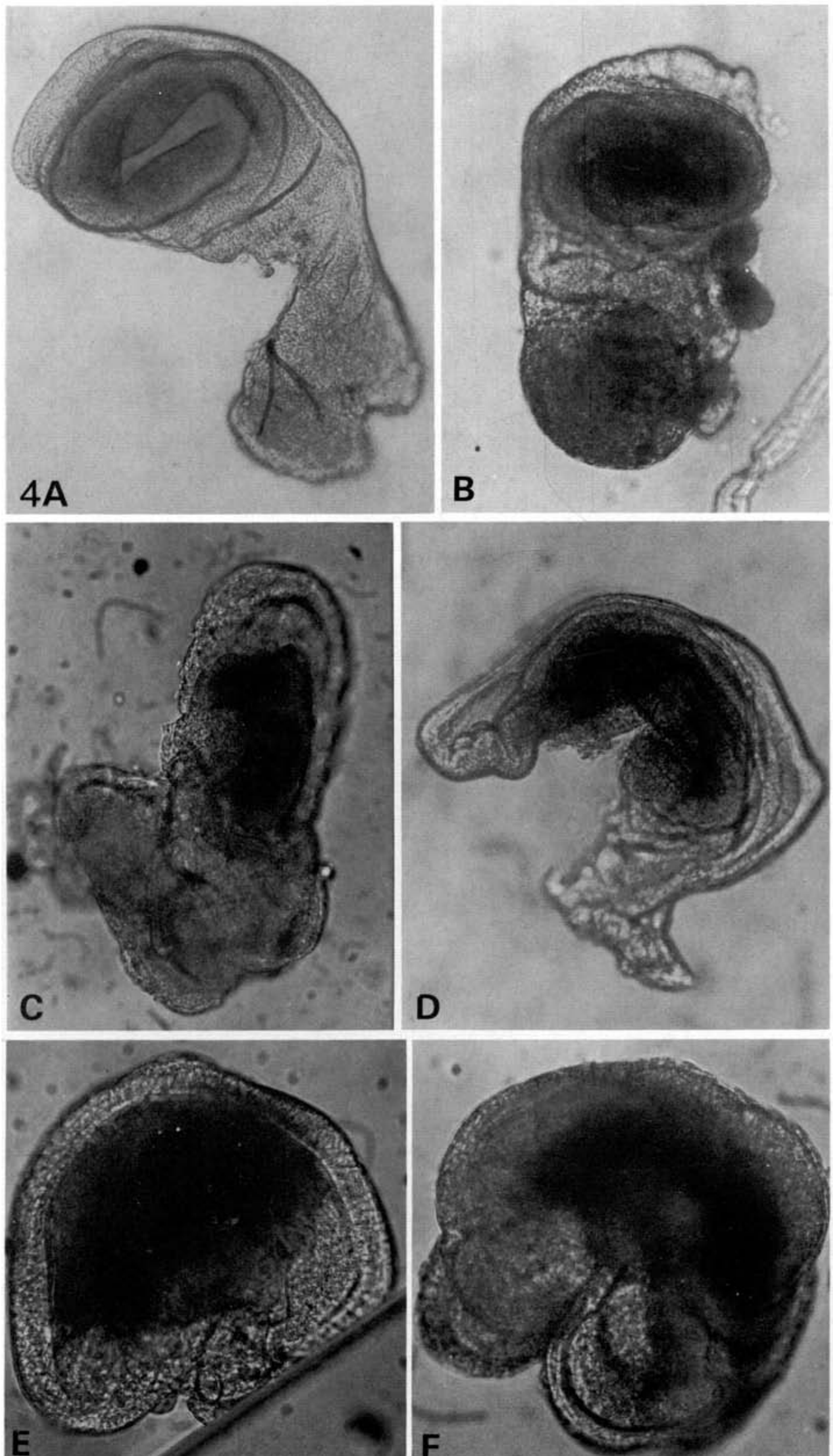

Fig. 4.

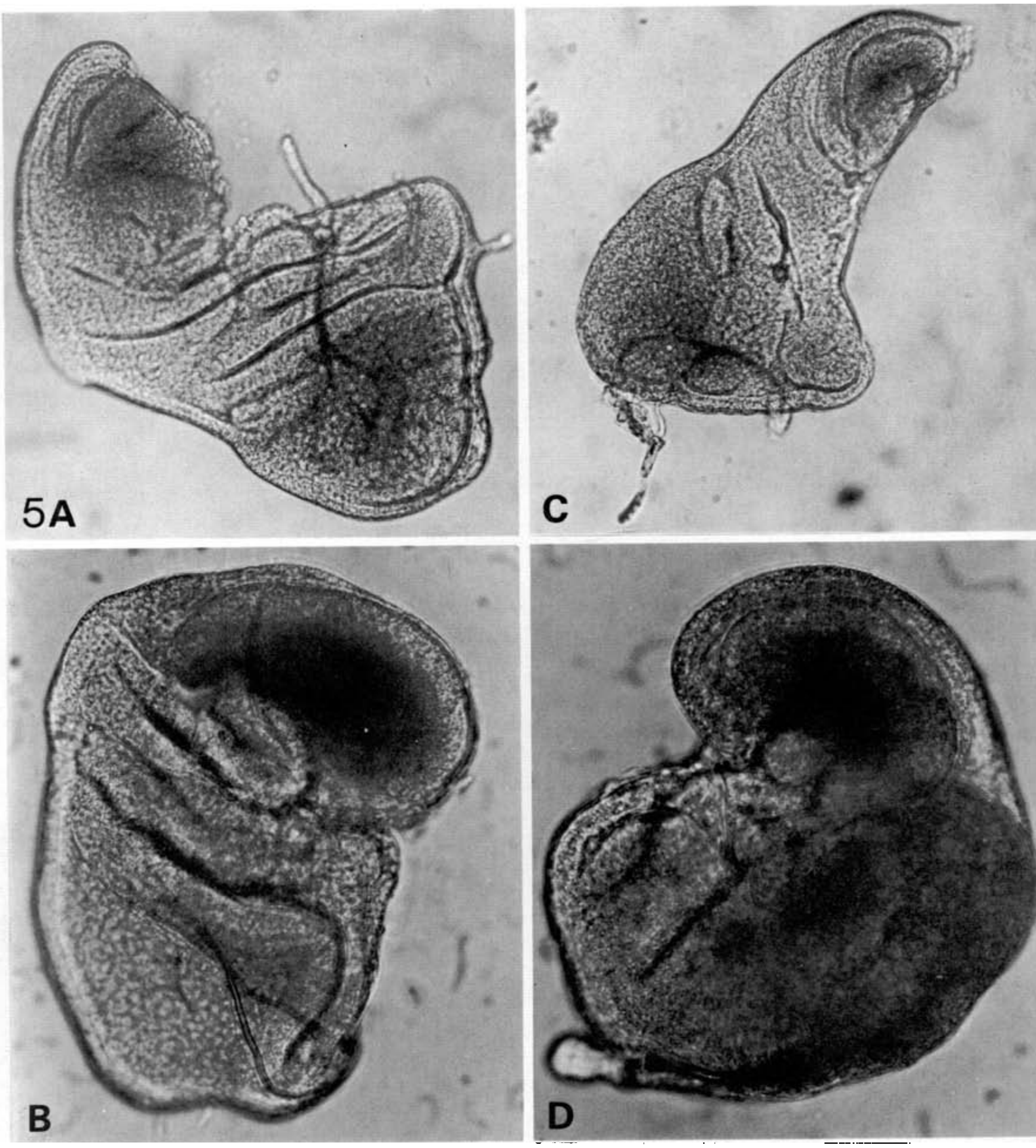

Fig. 5. Wound healing in fragments D and N. (A) Fragment D uncultured. (B) Fragment D after two days of culture demonstrating regular healing. (C) Fragment N uncultured. (D) Fragment N after two days of culture demonstrating irregular healing.

Fig. 4. Wound healing in Fragments A and B. (A) Fragment A uncultured. (B and C) Fragment A after two days of culture; (B) regular wound healing, (C) irregular wound healing. (D) Fragment B uncultured. (E and F) Fragment B after two days of culture; (E) regular healing, (F) irregular healing.

Similar results were obtained with fragments B and C. Although the frequency of regeneration following irregular wound healing in these fragments is high, this is not entirely unexpected. Recent models of epimorphic pattern regulation suggest that following irregular wound healing the two wound edges should regulate independently. The mode of pattern regulation, i.e. regeneration or duplication, would be dependent upon those positional values which are apposed by wound healing. If, as suggested by Karlsson (1981), positional values are unevenly distributed in the wing disc then it is conceivable that irregular wound healing could create positional discrepancies along one of the wound edges sufficient for regeneration. Indeed fragments with wound edges similar to either the anterior or posterior wound edge in fragments B and C, have been found to both regenerate and duplicate (Bryant, 1975*a*; Karlsson, 1981; Karlsson & Smith, 1981; Kauffman & Ling, 1981; Dale, 1983). On the other hand it is inconceivable that wing disc

Table 3. *Percentage of structures formed by fragments D, E, F & G*

	Fragment							
	D		E		F		G	
Structures scored	Culture period (days) 0	6	0	6	0	6	0	6
Notum*	100	100	100	100	100	100	100	100
Scut	100	96	100	92	100	74(4)	0	39
ANWP	72	83	92	92	0	22	86	56
PNWP	83	78	92	69	90	65	0	72
UP	94	74	85	92	15	35	23	67
AS1	94	78	85	92	95	70(22)	32	72
AS2	94	78	85	100	90	61(17)	5	61
AS3	94	87	92	96(4)†	100	83(7)	0	56
AS4	72	74	77	85	100	57	0	53
Scd	94	78	77	96	20	39	18	67
HP	72	74	77	88	0	22(4)	86	72
Teg	83	91	92	100	0	39(4)	100	100
PCo	83	74	85	84	5	26	86	67
M/DCo	0	39	92	76	95	78(7)	95	83
TR	0	61	100	92	100	96(9)	100	100
DR	56	52	31	48	100	65	100	72
PR	94	74	0	56	100	52	88	61
AL	78	65	0	32	90	48	64	56
AC	78	83	8	72	85	61(4)	23	56
Wing	100	100	100	100	100	100	100	100
YC	0	52	85	92(4)	95	91(26)	100	94
PVR	0	43	85	88	95	91(35)	100	100
PWP	0	52	85	92(4)	100	91(17)	100	100
PS	0	52	85	92	100	87(17)	100	100
AP	0	48	85	92	100	91(17)	100	100
No. of implants	18	23	13	25	20	23	22	18

* abbreviations as in Fig. 1.
† figures in brackets frequency of duplication.

fragments as large as B and C would duplicate following regular wound healing. Given this the critical observation in these fragments is that regular wound healing results in regeneration alone, while irregular wound healing results in duplication as well as regeneration (Table 2). Following irregular wound healing in these two fragments 60 % of implants duplicated, most of these extensively including many structures far from the wound edge. Along with the permanency of irregular wound healing, the frequency and extent of duplication in these fragments precludes an explanation based on small-scale regulative events involving transient wound healing (Reinhardt *et al.* 1977).

The results obtained after culturing the remaining 12 types of fragment (Tables 3–5) are consistent with those obtained for fragments A, B and C. Frequent and extensive duplication is only observed in those fragments in which wound healing is either clearly irregular or indistinguishable. Those fragments in which wound

Table 4. *Percentage of structures formed by fragments H, I, J & K*

Structures scored	H 0	H 6	I 0	I 6	J 0	J 6	K 0	K 6
Notum*	100	100(30)†	100	100	100	100	100	95
Scut	95	70(5)	100	100	100	81	0	16
ANWP	91	55(20)	78	89	17	54	80	74
PNWP	91	75(30)	94	89	92	81	0	42
UP	91	75(30)	94	89	17	42	44	68(5)
AS1	95	75(30)	94	95	100	58	52	89(5)
AS2	95	75(15)	94	89	83	58(4)	20	84(5)
AS3	86	95(20)	39	84(5)	100	81(4)	0	47
AS4	76	70(10)	61	84	75	81	0	16
Scd	67	70(10)	39	89	75	65	88	79
HP	43	30	83	89	25	46	92	84
Teg	91	85(20)	89	95	0	58	96	95
PCo	0	10	94	95	92	54	100	74(5)
M/DCo	0	5	78	95	83	65	100	84(5)
TR	0	0	94	95	92	88	100	95
DR	67	40	78	58	100	69	92	63
PR	76	70	0	58	100	58	100	63
AL	95	50(5)	0	58	100	54	92	37
AC	90	100(5)	6	58(5)	100	42(4)	68	47(5)
Wing	100	100	100	100	100	100	100	100
YC	0	10	94	95(5)	92	77(4)	96	79(5)
PVR	0	15	94	89(5)	92	73	96	84(5)
PWP	0	25	100	89(5)	92	85	96	89(5)
PS	0	30(5)	100	89(5)	92	81	96	89
AP	0	25(5)	100	89	92	81	96	84
No. of implants	21	20	18	19	12	26	25	19

Fragment; Culture period (days).

* abbreviations as in Fig. 1.
† figures in brackets frequency of duplication.

healing is clearly regular predominantly regenerate, duplication being rare and restricted to a small number of structures.

Variations in the mode of wound healing cannot explain all the variable pattern regulation observed in imaginal disc fragments. Recently it has been shown that many wing disc fragments which initially duplicate are also capable of regenerating if the culture period is extended (Duranceau *et al.* 1980; Kauffman & Ling, 1981; Kirby *et al.* 1982). Similar results have been obtained in the genital disc (Lüönd, 1961). It is unlikely that the mode of wound healing could explain this phenomenon, additional explanations are required.

During wound healing in imaginal discs heterotypic contacts are established between the wound edges of the epithelium proper and those of the peripodial membrane lying directly beneath them (Reinhardt *et al.* 1977; Reinhardt & Bryant, 1981), it is unlikely that this mode of wound healing would be variable.

Table 5. *Percentage of structures formed by fragments L, M, N & O*

Structures scored	Fragment L, Culture period (days) 0	L 6	M 0	M 6	N 0	N 6	O 0	O 6
Notum*	100	100(55)†	100	100	100	100(45)	100	100(6)
Scut	100	95(40)	100	84	100	77(27)	100	89
ANWP	54	10	85	84(11)	95	82(5)	100	72
PNWP	77	75(50)	69	53(11)	95	95(54)	79	72(6)
UP	8	25	54	58(5)	89	86(36)	93	72(11)
AS1	69	40(5)	46	79(16)	95	95(59)	93	100(17)
AS2	92	30	15	47(11)	89	95(54)	93	83(6)
AS3	92	90(50)	0	47	95	86(45)	100	100(6)
AS4	69	85(30)	61	47	84	82(27)	100	67(6)
Scd	15	20	69	68	26	64(23)	43	89(6)
HP	0	0	92	68	74	27(9)	64	61(6)
Teg	46	20	100	95	95	82(36)	100	72(22)
PCo	0	0	100	79	21	14	0	44
M/DCo	0	5	100	84	0	4	0	44
TR	0	5	92	89	0	9	43	89
DR	77	20	15	26	21	23	86	83
PR	85	45	0	21	84	64	100	72
AL	77	45(10)	0	26	79	68(18)	100	83
AC	85	60(15)	0	11	79	73(18)	93	72
Wing	100	100	100	100	100	100	100	100
YC	0	0	92	95(5)	0	0	0	50(6)
PVR	0	5	100	95(5)	0	0	0	56(5)
PWP	0	15	100	95(11)	0	0	100	83(17)
PS	0	15	100	95(11)	0	0	100	83(17)
AP	0	5	100	95	0	0	100	83(6)
No. of implants	13	20	13	19	19	22	14	18

* abbreviations as in Fig. 1.
† figures in brackets frequency of duplication.

Heterotypic wound healing is transient, contact between the two cell types are broken once stable homotypic wound healing has occurred. This latter type of wound healing is clearly variable, resulting in alternative modes of pattern regulation, suggesting that it is stable contact between cells of the epithelium proper that determines which structures are formed during pattern regulation.

The results presented above are consistent with those obtained from grafting experiments in the imaginal discs of *Drosophila* and the larval limbs of cockroaches. Imaginal disc fragments which when cultured alone duplicate, can be induced to regenerate by mixing them with fragments from different positions of either the same or different discs (Haynie & Bryant, 1976; Adler & Bryant, 1977; Wilcox & Smith, 1977; Bryant *et al.* 1978; Adler, 1979; Karlsson, 1979; Littlefield & Bryant, 1979*b*; Haynie & Schubiger, 1979; Haynie, 1982). Following amputation of an insect limb, both proximal and distal stumps regenerate a new set of distal limb structures (Bohn, 1965; Bullière, 1970; Shaw & Bryant, 1975). If epidermal cells from different proximal–distal levels are confronted, wound healing is followed by growth and intercalary regeneration of structures normally intermediate between the host graft junction (Bohn, 1970; Bullière, 1970; French, 1976). Since the intercalary regenerate is derived from both host and graft tissue (Bullière, 1971; Bohn, 1971; French, 1976), cells can form either more proximal or distal structures during intercalation. Growth and intercalary regeneration also occur when strips of cockroach leg epidermis are grafted into different circumferential positions (French, 1978, 1980). Cells confronted at the host–graft junction are able to regenerate in either direction around the circumference (French, 1980); the direction taken is governed by their normal position in the limb, such that intercalary regeneration always occurs via the shortest section of the circumference normally separating the confronted cells. The developmental fate of cells can clearly be modified by cell–cell interactions.

In conclusion the results presented in this paper suggest that pattern regulation in the imaginal wing disc of *Drosophila melanogaster*, is the result of local cell–cell interactions between normally non-adjacent cells apposed by wound healing. These interactions result in localized cell proliferation and the formation of intermediary structures by intercalation.

We thank Graham Brown for photographic assistance and the S.E.R.C. for financial support.

REFERENCES

Abbott, L., Karpen, G. & Schubiger, G. (1981). Compartmental restrictions and blastema formation during pattern regulation in *Drosophila* imaginal leg discs. *Devl Biol.* **87**, 64–75.

Adler, P. N. (1979). Position-specific interaction between cells of the imaginal wing and haltere discs of *Drosophila melanogaster*. *Devl Biol.* **70**, 262–267.

Adler, P. N. & Bryant, P. J. (1977). Participation of lethally irradiated imaginal disc tissue in pattern formation in *Drosophila*. *Devl Biol.* **60**, 293–304.

BOHN, H. (1965). Analyse der Regenerationsfähigkeit der Insekten-extremität durch Amputations- und Transplatationsversuche an Larven der afrikanschen Schabe *Leucophaea maderae* Fabr. (Blattaria). I. Mitt. Regenerationspotenzen. *Wilhelm Roux' Arch. EntwMech. Org.* **156**, 49–74.

BOHN, H. (1970*a*). Interkalare Regeneration und Segmentale Gradienten bei den Extremitäten von *Leucophaea*-Larven (Blattaria). I. Femur und Tibia. *Wilhelm Roux' Arch. EntwMech. Org.* **165**, 303–341.

BOHN, H. (1970*b*). Interkalare Regeneration und Segmentale Gradienten bei den Extremitäten von *Leucophaea*-Larven (Blattaria). II. Coxa und Tarsus. *Devl Biol.* **23**, 355–379.

BOHN, H. (1971). Interkalare Regeneration und Segmentale Gradienten bei den Extremitäten von *Leucophaea*-Larven (Blattaria). III. Die Herkunft des interkalaren Regenerates. *Wilhelm Roux' Arch. EntwMech. Org.* **167**, 209–221.

BRYANT, P. J. (1971). Regeneration and duplication following operations *in situ* on the imaginal discs of *Drosophila melanogaster. Devl Biol.* **26**, 637–651.

BRYANT, P. J. (1975*a*). Pattern formation in the imaginal wing disc of *Drosophila melanogaster*: Fate map, regeneration and duplication. *J. exp. Zool.* **193**, 49–78.

BRYANT, P. J. (1975*b*). Regeneration and duplication in imaginal discs. *Ciba Found. Symp.* **29**, 71–93.

BRYANT, P. J., ADLER, P. N., DURANCEAU, C., FAIN, M. J., GLENN, S., HSEI, B., JAMES, A. A., LITTLEFIELD, C. L., REINHARDT, C. A., STRUB, S. & SCHNEIDERMAN, H. A. (1978). Regulative interactions between cells from different imaginal discs of *Drosophila melanogaster. Science* **201**, 928–930.

BRYANT, P. J. & HSEI, B. W. (1977). Pattern formation in asymmetrical and symmetrical discs of *Drosophila melanogaster. Amer. Zool.* **17**, 595–611.

BULLIÈRE, D. (1970). Interprétation des régénérats multiples chez les insectes. *J. Embryol. exp. Morph.* **23**, 337–357.

BULLIÈRE, D. (1971). Utilisation de la régénération intercalaire pour l'étude de la determination cellulaire au cours de la morphogenese chez *Blabera craniifer* (Insecta Dictyoptere). *Devl Biol.* **25**, 672–709.

CHAN, L. N. & GEHRING, W. (1971). Determination of blastoderm cells in *Drosophila melanogaster. Proc. natn. Acad. Sci., U.S.A.* **68**, 2217–2221.

CUMMINGS, F. W. & PROTHERO, J. W. (1978). A model of pattern formation in multicellular organisms. *Coll. Phenom.* **3**, 41–53.

DALE, L. (1983). Pattern regulation in the imaginal wing disc of *Drosophila.* Ph.D. Thesis, University of Edinburgh.

DALE, L. & BOWNES, M. (1980). Is regeneration in *Drosophila* the result of epimorphic regulation? *Wilhelm Roux' Arch. devl Biol.* **189**, 91–96.

DALE, L. & BOWNES, M. (1981). Wound healing and regeneration in the imaginal wing disc of *Drosophila. Wilhelm Roux' Arch. devl Biol.* **190**, 185–190.

DURANCEAU, C., GLENN, S. L. & SCHNEIDERMAN, H. A. (1980). Positional information as a regulator of growth in the imaginal wing disc of *Drosophila melanogaster.* In *Insect Biology in the Future* (ed. M. Locke & D. Smith), pp. 479–516. New York – London: Academic Press.

FRENCH, V. (1976). Leg regeneration in the cockroach, *Blatella germanica.* I. Regeneration from a congruent tibial graft/host junction. *Wilhelm Roux' Arch. EntwMech. Org.* **179**, 57–76.

FRENCH, V. (1978). Intercalary regeneration around the circumference of the cockroach leg. *J. Embryol. exp. Morph.* **47**, 53–84.

FRENCH, V. (1980). Positional information around the segments of the cockroach leg. *J. Embryol. exp. Morph.* **59**, 281–313.

FRENCH, V., BRYANT, P. J. & BRYANT, S. V. (1976). Pattern regulation in epimorphic fields. *Science* **193**, 969–981.

HAYNIE, J. L. (1982). Homologies of positional information in thoracic imaginal discs of *Drosophila melanogaster. Wilhelm Roux' Arch. devl Biol.* **191**, 293–300.

HAYNIE, J. L. & BRYANT, P. J. (1976). Intercalary regeneration in imaginal wing disc of *Drosophila melanogaster. Nature* **259**, 659–662.

HAYNIE, J. L. & SCHUBIGER, G. (1979). Absence of distal to proximal intercalary regeneration in imaginal wing discs of *Drosophila melanogaster. Devl Biol.* **68**, 157–161.

KARLSSON, J. (1979). A major difference between transdetermination and homeosis. *Nature* **279**, 426–428.

KARLSSON, J. (1981). The distribution of regenerative potential in the wing disc of *Drosophila. J. Embryol. exp. Morph.* **61**, 303–316.

KARLSSON, J. & SMITH, R. J. (1981). Regeneration from duplicating fragments of the *Drosophila* wing disc. *J. Embryol. exp. Morph.* **66**, 117–126.

KAUFFMAN, S. A. & LING, E. (1981). Regeneration by complementary wing disc fragments of *Drosophila melanogaster. Devl Biol.* **82**, 238–257.

KIRBY, B. S., BRYANT, P. J. & SCHNEIDERMAN, H. A. (1982). Regeneration following duplication in imaginal wing disc fragments of *Drosophila melanogaster. Devl Biol.* **90**, 259–271.

LEWIS, J. (1981). Simpler rules for epimorphic regeneration: The polar coordinate model without polar coordinates. *J. theor. Biol.* **88**, 371–392.

LITTLEFIELD, C. L. & BRYANT, P. J. (1979*a*). Prospective fates and regulative capacities of fragments of the female genital disc of *Drosophila melanogaster. Devl Biol.* **70**, 127–148.

LITTLEFIELD, C. L. & BRYANT, P. J. (1979*b*). Sexual homologies and intercalation between parts of the male and female genital discs of *Drosophila melanogaster. Devl Biol.* **70**, 180–194.

LÜÖND, H. (1961). Untersuchungen zur Mustergleiderung in fragmentierten Primordien des männlichen Geschlecht-sapporates von *Drosophila seguyi. Devl Biol.* **3**, 615–656.

POSTLEWAIT, J. H., POODRY, C. A. & SCHNEIDERMAN, H. A. (1971). Cellular dynamics of pattern duplication in imaginal discs of *Drosophila melanogaster. Devl Biol.* **26**, 125–132.

REINHARDT, C. A. & BRYANT, P. J. (1981). Wound healing in the imaginal discs of *Drosophila.* II. Transmission electron microscopy of normal and healing wing discs. *J. exp. Zool.* **216**, 45–61.

REINHARDT, C. A., HODGKIN, N. M. & BRYANT, P. J. (1977). Wound healing in the imaginal discs of *Drosophila.* I. Scanning electron microscopy of normal and healing wing discs. *Devl Biol.* **60**, 238–257.

RUSSELL, M. A. & HAYES, P. (1980). The genetics of pattern formation. In *Insect Biology in the Future* (ed. Locke & Smith), pp. 543–564. New York – London: Academic Press.

SANG, J. H. (1978). The nutritional requirements of *Drosophila*. In *The Genetics and Biology of* Drosophila (ed. Ashburner & Wright), Vol. 2a, pp. 159–192. New York – London: Academic Press.

SCHUBIGER, G. (1971). Regeneration, duplication and transdetermination in fragments of the leg disc of *Drosophila melanogaster. Devl Biol.* **26**, 277–295.

SHAW, V. K. & BRYANT, P. J. (1975). Intercalary leg regeneration in the large milkweed bug *Oncopeltus fasciatus. Devl Biol.* **45**, 187–191.

ULRICH, E. (1971). Cell lineage, determination and regeneration in the female genital disc of *Drosophila melanogaster. Wilhelm Roux' Arch. EntwMech. Org.* **167**, 64–82.

URSPRUNG, H. (1967). In vivo culture of *Drosophila* imaginal discs. In *Methods in Developmental Biology* (ed. Wilt & Wessels), pp. 485–492. New York: Crowell.

VAN DER MEER, J. M. & OUWENEEL, W. J. (1974). Differentiation capacities of the dorsal metathoracic (Haltere) disc of *Drosophila melanogaster.* II. Regeneration and duplication. *Wilhelm Roux' Arch. EntwMech. Org.* **174**, 361–373.

WILCOX, M. & SMITH, R. J. (1977). Regenerative interactions between *Drosophila* imaginal discs of different types. *Devl Biol.* **60**, 287–297.

(*Accepted 12 October 1984*)