

## Short Note

### Addition of Mesenteries during Asexual Reproduction of the Polyglyphic Sea Anemone *Anthopleura dixoniana* (Anthozoa: Actiniaria)<sup>1</sup>

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**James Lin, Chang-Po Chen and I-Ming Chen (1993)** Addition of mesenteries during asexual reproduction of the polyglyphic sea anemone *Anthopleura dixoniana* (Anthozoa: Actiniaria). *Bull. Inst. Zool., Academia Sinica* 32(2): 140-147. The mesenterial arrangement of the polyglyphic sea anemone *Anthopleura dixoniana* (Haddon & Shackleton) during asexual reproduction was studied from November, 1987 through December, 1988. Individuals were collected from the upper intertidal zone at Wanlitung (120°41'E, 22°01'N) in southern Taiwan. In exocoels, two mesenteries were added in exocoels and developed into either directive or non-directive mesenteries. In a directive endocoel, two mesenteries with retractor muscles facing one another were added, resulting in two adjacent pairs of directive mesenteries; each pair consists of one new and one old member. In a non-directive endocoel, four mesenteries were added, resulting in a pair of directive mesenteries flanked by two pairs of non-directives; each pair of non-directive mesenteries consists of one new and one old member. *Anthopleura dixoniana* maintained a constant ratio between the number of non-directive and directive mesenteries among dividing, divided, and non-dividing individuals. After division, two daughter *A. dixoniana* did not differ significantly in their numbers of mesenteries.

**Key words:** Asexual reproduction, Mesentery, *Anthopleura dixoniana*, Actiniaria, Anthozoa.

Asexual reproduction in sea anemones has been proposed to be a kind of growth meant to circumvent environmental restrictions on body size (Sebens 1979 1982, Lin *et al.* 1992). Many aspects of asexual reproduction have been studied, including periodicity, regulative factors, and influences on genotypic structure of populations (Minasian 1976 1979 1982, Smith and Lenhoff 1976, Minasian and Mariscal 1979, Sebens

1980, Ayre 1983 1984, Hunter 1984, Bucklin 1987a 1987b, Lin *et al.* 1992). However, little information is available on the numerical variation of mesenteries associated with asexual reproduction, **except** for *Haliplanella lineata* (Verrill) (= *Sagartia luciae* = *H. luciae*) (Davis 1919, Atoda 1976) and *Anthopleura elegantissima* (Brandt) (Sebens 1983).

Mesenteries contain most of the important discrete musculature of sea anemones. Each of the several

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mesenterial parts functions differently in gametogenesis, digestion, or water circulation (Stephenson 1928, Manuel 1981). It is assumed that sea anemones add mesenteries in exocoels (Manuel 1981). However, only Davis (1919) has shown that a species (*Sagartia luciae* Verrill) added mesenteries in exocoels as either directive or non-directive mesenteries.

*Anthopleura dixoniana* (Haddon and Shackleton), which reproduces asexually (Lin *et al.* 1992), varies in its numbers of both siphonoglyphs and paired directive mesenteries (England 1987). Here we report patterns of the addition of mesenteries and mesenterial arrangement during *A. dixonian*'s longitudinal fission. We conclude that new mesenteries are added either by making a pair of new mesenteries, or by pairing a new mesentery with an existing one; we also conclude that numbers of mesentery pairs do not differ significantly between two daughter anemones after division.

**Materials and Methods**—*Anthopleura dixoniana* (Haddon and Shackleton) individuals were collected from the upper intertidal zone at Wanlitung (120°41'E; 22°01'N) in southern Taiwan from November, 1987 to December, 1988. From our preliminary study, a dividing *Anthopleura dixoniana* possesses an elongated base and an elliptical oral disc with an orally folded margin (Fig. 1). The length of the longest dimension of the pedal disc of a dividing individual is several times that of the shortest, whereas the pedal disc of a non-dividing individual is approximately circular. By contrast, when a sea anemone moves or inserts itself into a rock crevice, its base may be elongated in the direction of its movement; however, the basal outline is seldom far from being circular. Division proceeds orally from the pedal disc, with one anemone finally separating into

two more-or-less equal parts. The division process takes a few hours to complete. The severed edges of each daughter sea anemone roll inward and then fuse, leaving a conspicuous yellowish-white scar that persists for 2-3 weeks under laboratory conditions. Thus, sea anemones with elongated bases are inferred to be dividing, sea anemones with division scars are inferred to have recently divided, and other anemone (with circular bases and without division scars) are inferred to be non-dividing.

For histological study, individuals were narcotized in either 7% MgCl<sub>2</sub> fresh water solution or seawater containing a few drops of saturated chlorobutanol seawater solution. After relaxation, they were fixed in Bouin's seawater solution for at least 24 h, then dehydrated in alcohol, cleared in xylene, and embedded in paraffin; 5  $\mu$ m serial sections were cut and stained with basic fuchsin and indigo carmine (Clark 1980).

Some anemones were maintained in a seawater (33-35 ppt salinity) aquarium for several months. After division, daughter anemones were fixed and sectioned to ascertain differences in mesenteries between individuals.

The terminology used this study follows Stephenson (1928): non-directive mesenteries are those in which the retractor muscles of the two members of a pair face each other; directive mesenteries are those in which the retractor muscles face away from each other; perfect mesenteries are those in which the coelenteric edge of a mesentery connects with the actinopharynx; imperfect mesenteries fail to join the actinopharynx; exocoels are the spaces between two adjacent pairs of mesenteries; and endocoels are the spaces enclosed by each pair of mesenteries.

From our preliminary study, all imperfect *An-*



Fig. 1. Longitudinal fission of *Anthopleura dixoniana*. a) A dividing individual. Notice the elongated base and elliptical oral disc. b) Division starts at the aboral end. Scale bars = 3 mm.

*thopleura dixoniana* mesenteries ultimately grow to connect with the actinopharynx, thereby becoming perfect. Thus, imperfect mesenteries appearing in our studied sections were assumed to be new.

One pair of directive mesenteries between adjacent pairs of non-directive mesenteries was termed a "single pair of directives" (Fig. 2a); two, three, and four sequential pairs of directives were termed "double, triple, and quadruple pairs of directives" respectively (Fig. 2b, c, d).

Mesenterial data that differed significantly in one-way ANOVA tests were subjected to SNK tests to compare differences among dividing, divided, and non-

dividing sea anemones (Sokal and Rohlf 1981). Observed differences in mesenterial numbers between two divided daughter sea anemones were compared according to a paired *t*-test.

**Results**—A total of 162 *Anthopleura dixoniana* individuals were collected and sectioned. All mesenteries were paired and perfect with the exception of newly-added mesenteries. All directive mesenteries were connected to siphonoglyphs. All individuals had the same number of mesentery pairs both orally and pedally, except for individuals with new mesenteries; those first appeared in the pedal portion. The total number

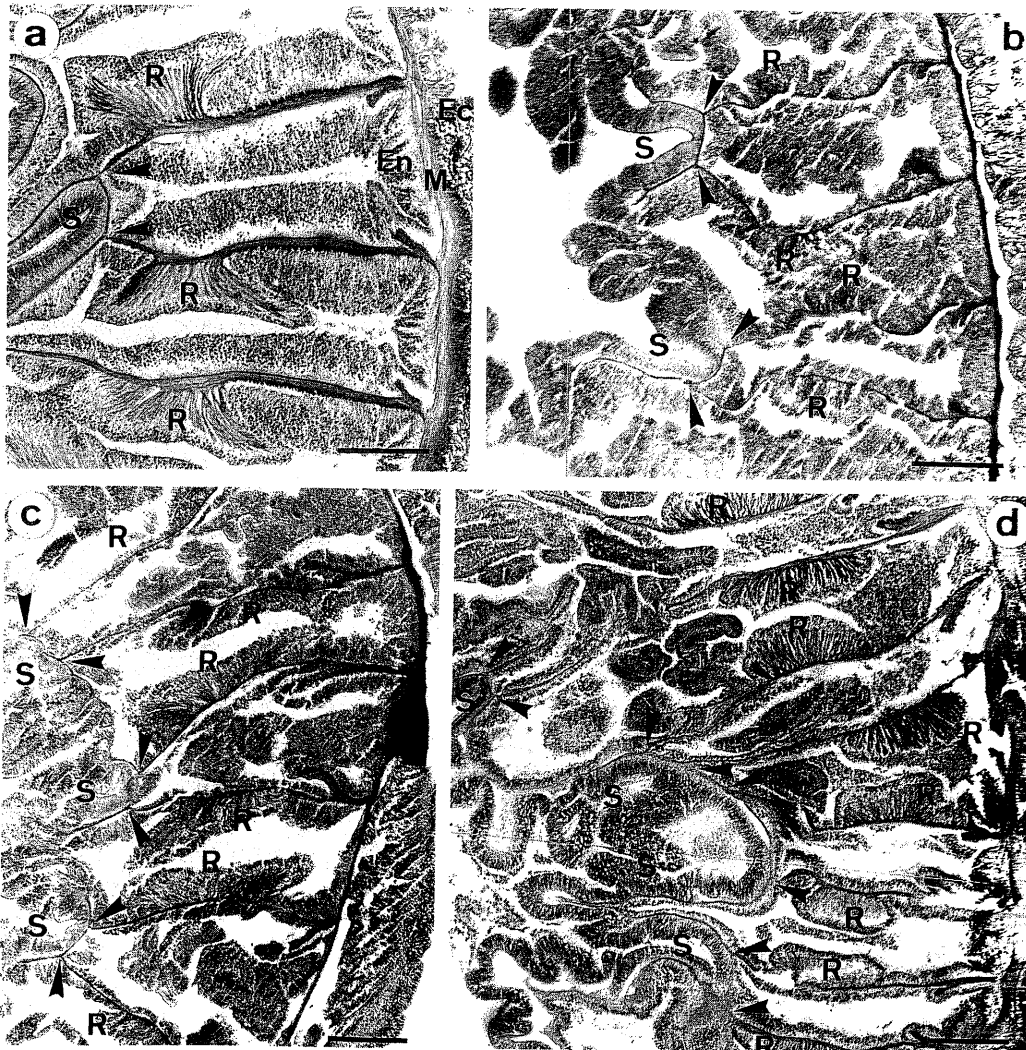


Fig. 2. Cross sections through upper columns and actinopharynxes of *Anthopleura dixoniana*. a) A single pair of directives. b) A double pair of directives. c) A triple pair of directives. d) A quadruple pair of directives. Each pair is connected to one siphonoglyph (S). R: retractor muscle; En: endoderm; M: mesoglea; Ec: ectoderm. Arrows indicate the joints of siphonoglyphs and mesenteries. Scale bars = 0.5 mm.

of mesenteries equalled the number of tentacles.

The number of directive mesentery pairs varied from two to fifteen in the 162 sea anemones that were sectioned. Single pairs of directives appeared in all sectioned sea anemones, while double pairs appeared in sixty-four individuals, triple pairs appeared in ten individuals, and quadruple pairs appeared in five individuals.

New mesenteries were formed in exocoels flanked by either directive or non-directive mesenteries, or in endocoels of directive or non-directive mesenteries. When compared with existing mesenteries, new mesenteries had thinner layers of mesoglea, weaker or no retractor or parieto-basilar muscles, and no filaments (Fig. 3). Forty-seven of the 162 sea anemone individuals sectioned revealed 82 instances of mesenterial addition, resulting in 101 pairs of new mesenteries (Table 1). In each exocoel, only two new mesenteries were added to form one pair of non-directive or directive mesenteries (Figs. 3). In the endocoel of non-directive and directive mesenteries, new mesenteries were added to form at least one pair

of directive mesenteries (Table 1).

In a non-directive endocoel, four mesenteries were added. Two mesenteries with retractor muscles facing outward were first added and paired with existing old mesenteries, thus forming two new non-directive pairs (each consisting of one new and one old member). After the addition of these first two mesenteries, one new mesentery—with its retractor muscle facing the existing old mesenteries—was added in each of the two newly-formed non-directive endocoels (Fig. 4). This resulted in one pair of directive mesenteries with one pair of non-directive mesenteries on each side, each non-directive pair consisting of one new and one old member.

In a directive endocoel, two new mesenteries were produced with retractors facing one another. This resulted in two adjacent pairs of directive mesenteries—each pair consisting of one new and one old mesentery. Fig. 5 shows non-directive mesenteries added to the directive endocoel and directive mesenteries added to the two flanking exocoels, thus resulting in a quadruple pair of directive mesenteries.

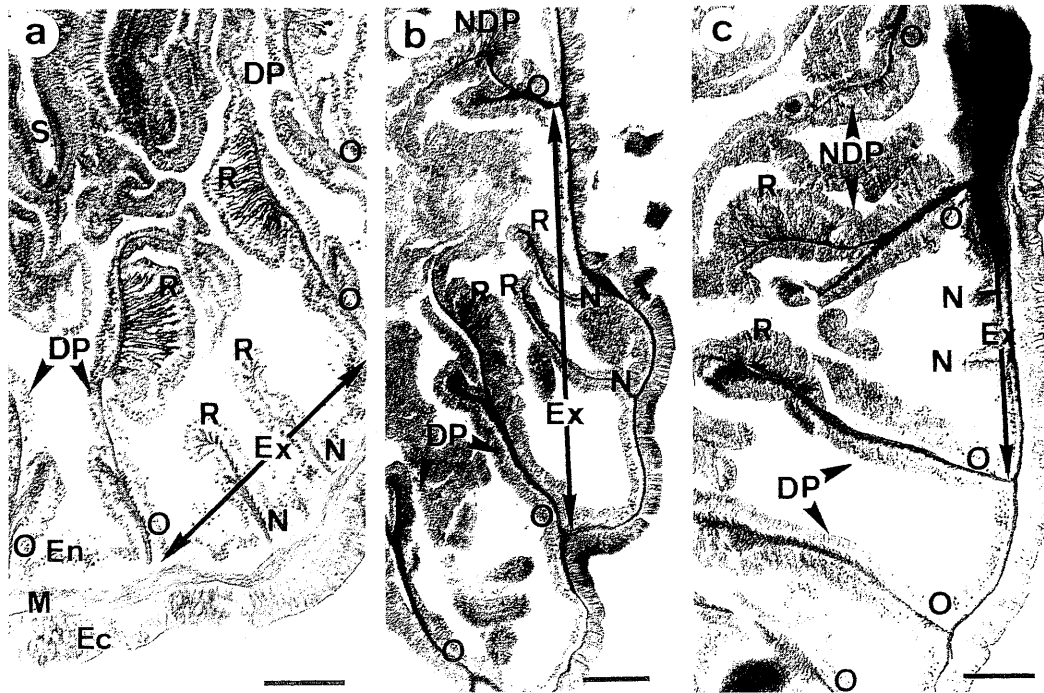


Fig. 3. Cross sections through lower column of *Anthopleura dixoniana* show new mesenteries (N) formed in exocoels (Ex) of *Anthopleura dixoniana*. a) New non-directive mesenteries. b) New directive mesenteries. c) New mesenteries which cannot be identified as being non-directive or directive. DP: directive pair of mesenteries; Ec: ectoderm; En: endocoel; M: mesoglea; N: new mesentery; NDP: non-directive pair of mesenteries; O: old mesentery; R: retractor muscle; S: siphonoglyphs. Scale bars = 0.5 mm.

Table 1. Locations and type of new mesenteries formed in 47 individuals of *Anthopleura dixoniana*.

Location of new mesenteries	Number of instances	Pairs of mesenteries formed		
		Directive	Non-directive	Indistinguishable
exocoel	71	10	36	25
non-directive endocoel	8	8	16	
directive endocoel	3	6		
Total	82	24	52	25



Fig. 4. Cross sections through lower columns of *Anthopleura dixoniana* show new mesenteries (N) in non-directive endocoels (NEn). a) Early stage: the two new central mesenteries are added with retractors facing outward to make new pairs with existing non-directives; the retractors of the other two new mesenteries are still indistinguishable. b) Later stage: the two new central mesenteries form one pair of directive mesenteries, and the other two new mesenteries make pairs with existing non-directives, thereby forming two pairs of non-directive mesenteries. Each non-directive pair consists of one new (N) and one old (O) mesentery. Scale bars = 0.5 mm.

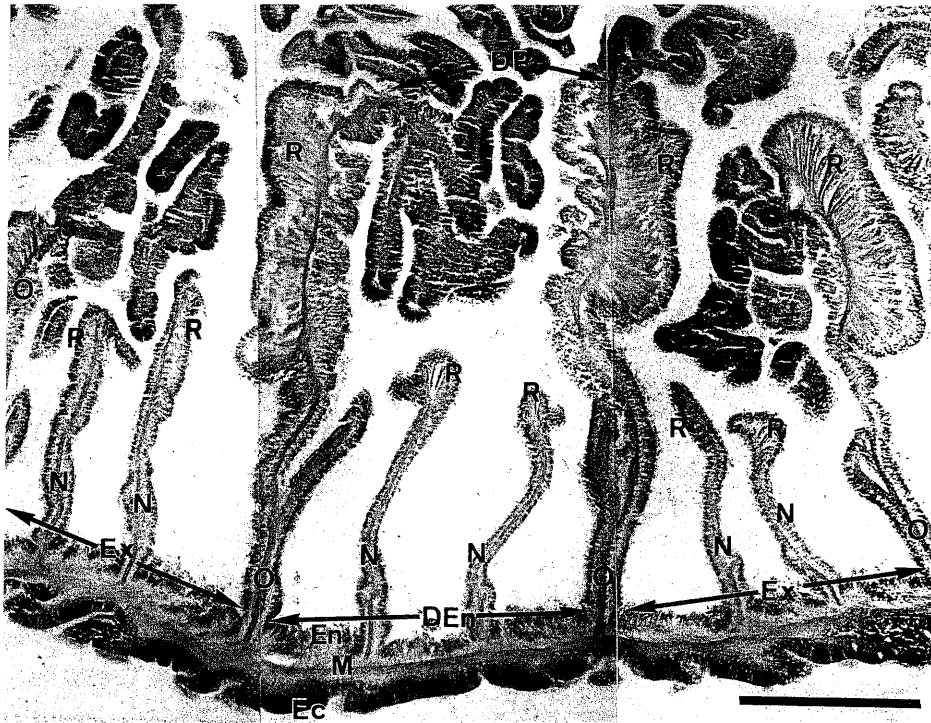


Fig. 5. Cross section through lower column of *Anthopleura dixoniana* shows new mesenteries in the directive endocoel (DEn) and exocoels (Ex), consequently forming a quadruple pair of directive mesenteries. New mesenteries in the directive endocoel form two pairs of directives, of which each pair is a combination of one new (N) and one old (O) mesentery. Scale bars = 0.5 mm.

The total number of mesentery pairs ranged from eight to twenty-nine in dividing, six to nineteen in divided, and four to twenty-nine in non-dividing individuals (Table 2). These differences in total number of mesentery pairs were found to be significant (ANOVA,  $F = 15.66$ ,  $p < 0.01$ ) (Table 2); in addition, our SNK ( $p < 0.05$ ) showed that dividing individuals had the greatest total number of mesentery pairs per individual and that divided individuals had the fewest.

Ratios of non-directive mesentery pairs to directive mesentery pairs were not significantly different among dividing, divided, and non-dividing individuals ( $F = 1.24$ ,  $p = 0.29$ ) (Table 2).

Some *Anthopleura dixoniana* divided while being maintained under laboratory conditions. A total of twenty-five pairs of newly-divided *Anthopleura dixoniana* were sectioned. The number of observed non-directive mesentery pairs was  $10 \pm 3.14$  (mean  $\pm$  SD,  $n = 50$ ), and the number of observed directive mesentery pairs was  $3.78 \pm 0.83$  ( $n = 50$ ). Results of a paired t-test revealed that no significant differences in numbers of non-directive mesentery pairs existed between indi-

viduals of each pair ( $t = 0.36$ ,  $p > 0.05$ ); the same was true for directive mesentery pairs ( $t = -1.09$ ,  $p > 0.05$ ) and total number of mesentery pairs ( $t = 0.50$ ,  $p > 0.05$ ).

**Discussion**—*Anthopleura dixoniana* forms new mesenteries in a more complex manner than sea anemones have previously been assumed to do (Manuel 1981). *Haliplanella lineata* develops all-new directive or non-directive mesenteries in its exocoels between adjacent pairs of mesenteries (Davis 1919, Atoda 1976). *Anthopleura dixoniana*, however, generates new directive mesenteries in the endocoels of directive or non-directive mesenteries, in addition to adding new mesenteries in its exocoels as do other sea anemones.

Although complex, there is a distinct rule for adding mesenteries: mesenteries are added in even numbers, and form pairs either with themselves or with neighboring mesenteries. In cases of mesenteries added in exocoels, new mesenteries pair up. In cases of mesenteries added in endocoels, new mesenteries pair with existing mesenteries. Thus, the addition of new mesenteries does not alter the pairing arrangement of mesenteries.

Table 2. Numerical characteristics of mesenteries of *Anthopleura dixoniana* among dividing, divided, and non-dividing individuals.

	Dividing	Divided	Non-dividing	F value	p
Individuals	18	24	120		
TN					
Range	8-29	6-19	4-29		
(mean $\pm$ SD)	(19.72 $\pm$ 4.81)	(12.35 $\pm$ 3.42)	(15.75 $\pm$ 4.28)	15.66	<0.01
RND					
Range	1.4-5	1-6	0-6.5		
(mean $\pm$ SD)	(3.09 $\pm$ 0.98)	(3.23 $\pm$ 1.52)	(2.81 $\pm$ 1.29)	1.24	0.29

TN: total number of mesentery pairs.

RND: ratio of the number of non-directive to directive mesenterial pairs in each individual.

England (1987) reported that during the formation of the first double pair of directive mesenteries in *Anthopleura dixoniana* and *A. waridi* (Carlgren), new directive and non-directive mesenteries were added in the new exocoel between the double pair. Thus, the mesenterial arrangement of double, triple, and quadruple pairs of directives that we found to be common in *A. dixoniana* were not found by England in specimens from Singapore, the Torres Strait, Hong Kong, the Addu Atoll, and the Maldives.

The number of directive and non-directive mesentery pairs in *Anthopleura dixoniana* varies greatly. Despite mesenterial irregularities, *A. dixoniana* retains a constant ratio of the number of non-directive mesentery pairs to that of directive pairs among dividing, divided, and non-dividing individuals. Dividing individuals have more mesentery pairs than non-dividing individuals, implying that sea anemones add mesenteries before undergoing asexual reproduction. Thus, both non-directive and directive mesenteries are added, and a constant ratio between them is maintained. The function of creating paired mesenteries and maintaining a certain ratio between non-directive and directive mesenteries is not clear.

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## 荻氏襟疣海葵(*Anthopleura dixoniana*)

### 無性生殖時膈膜的形成

林 義 展      陳 章 波      陳 一 鳴

由 1987 年 11 月至 1988 年 12 月，在萬里桐高潮區採得荻氏襟疣海葵(*Anthopleura dixoniana*)，研究其無性生殖(縱分裂)時膈膜排列之變化。荻氏襟疣海葵除了在膈膜外腔(ecocoel)中形成新膈膜外，也可在膈膜內腔(endocoel)中形成。在定向膈膜內腔(directive endocoel)中，兩片新膈膜與原有之舊膈膜形成兩對定向膈膜(directive mesenteries)，每對包括一片新膈膜和一片舊膈膜；在非定向膈膜內腔(non-directive endocoel)中，四片膈膜與原有之舊膈膜形成一對新定向膈膜及兩對非定向膈膜(non-directive mesenteries)，每對非定向膈膜包括一片新膈膜和一片舊膈膜。縱分裂無性生殖時，在分裂中、分裂後與未分裂的個體中，荻氏襟疣海葵維持一定相對比率的定向膈膜和非定向膈膜數。縱分裂後，兩個子個體的膈膜數無明顯差異。



## Short Note

### Induction of Host Plant Choice for Feeding and Oviposition in Diamondback Moth Larvae (*Plutella xylostella*)

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**How-Jing Lee and Yi-Chien Tseng (1993)** Induction of host plant choice for feeding and oviposition in Diamondback moth larvae (*Plutella xylostella*). *Bull. Inst. Zool., Academia Sinica* 32(2): 148-152. Last larval instar of Diamondback moth (DBM), *Plutella xylostella*, can be induced to prefer the same host plant species that the first three larval instars feed on. Although the DBM used in our experiments showed rather weak feeding induction response in a choice test, a significant preference for its familiar host plant was observed in a no-choice test. However, feeding preferences learned during DBM larval stages were not transferred to adult ovipositional preference. The significance of feeding induction and its effects on ovipositional choice are discussed.

**Key words:** Induction, Feeding, Oviposition, Diamondback moth, *Plutella xylostella*.

Feeding induction (Dethier 1982) is a common phenomenon in phytophagous insects (Jermy *et al.* 1968, Saxena and Schoonhoven 1982, de Boer and Hanson 1984). Food preference induction has also been reported in a variety of polyphagous and oligophagous species (Hsiao 1985). Although feeding preference can be altered by previous experience, the capacity for induction is rather species-specific, and is usually correlated with taxonomic distance between inducing plant pairs (de Boer and Hanson 1984).

Induction has some adaptive advantages in nature (Yamamoto 1974); since leaves and branches of different plants often intermingle in a natural habitat, larvae may come into contact with several plant species during normal feeding. Therefore, preference induction may restrict feeding to a single host species. Since it is metabolically costly to detoxify secondary plant chemicals (Schoonhoven and Meerman 1978, Brattsten 1979), an induction which confines larval feeding to one host species reduces the additional metabolic cost of adapting to the chemistry of a second host plant species.

The Diamondback moth (DBM), *Plutella xylostella*, is a serious pest affecting crucifers (Talekar *et al.* 1985). Its host plants are restricted to a limited number of cruciferous vegetables, and the economic damage it causes can be extensive. Although DBM (as well as *Manduca sexta*) are oligophagous insects, and although the latter can be induced to feed on non-host plants (de Boer and Hanson 1984), a preferred non-cruciferous plant species has yet to be documented for DBM. This study focuses on feeding induction among four host plants and the possibility of transferring feeding experience to ovipositional preference.

**Materials and Methods**—Diamondback moth larvae were collected from the field in areas outside of Taipei, Taiwan. These and all other experimental animals were kept in an environmental chamber under 23° C, L/D 12:12 h conditions. Larvae were fed fresh leaves of Ching-chiang Pai-tecei (CP) (*Brassica chinensis*). After pupation, leaves with attached cocoons were transferred to a 30X30X30 cm plastic container. Honey water was provided for emerging adult moths. Adults in the container were allowed to mate freely, and fresh