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Variability in patterns of intra-specific biting attack in commonly used genetic lines of laboratory mice

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Summary

Bite targets on opponents used by individually housed male, reproductively experienced male and lactating female subjects from outbred TO and Swiss Webster and inbred NZW/Ola, BALB/c, C57BL/10, DBA/2, CBA/Ca and C3H/He lines of mice were assessed. Ten-minute videotaped encounters with anosmic male TO or "congenic" opponents were used for this purpose. In general, the attacks shown by males appeared 'ritualised' with the head and ventral surface of the opponent being avoided. The lactating females showed no such inhibition. The intensity and incidence (and damaging nature of the attacks) was clearly influenced by strain of the resident, its sex, its reproductive experience and (in some cases) the nature of the opponent. This data may be helpful in controlling social stress in mouse studies.

Key words: Aggression, genetic lines of mice, male, female, bite targets

Introduction

The profound deleterious consequences of social stress have been detailed in laboratory rodents in general *(Brain, 1990)* and mice in particular *(Brain, 1989; Brain, 1996)*. One factor influencing the level of the conflict-related 'stress' is the genetic variability in the subjects *(Jones & Brain, 1987)*. This account uses modern videotape technology to assess the incidence and types of biting attack used by a range of commonly employed inbred and outbred lines of laboratory mice of differing sex and in a range of test situations.

Many authors *(e.g. Tinbergen, 1968; Blanchard & Blanchard, 1977)* have distinguished "offensive" and "defensive" forms of intra-specific attack. "Offensive" attack is generally regarded as unprovoked, whilst "defensive" attack is seen as a response to some actual or perceived threat to the individual, such as attack by a predator. In "offensive" attack, animals seem restrained from attacking vulnerable (easily damaged) areas of the opponent's body or from using the deadliest weapons at their disposal, thus limiting the likelihood of serious injury *(e.g. Tinbergen, 1953)*. Blanchard et al (1977) have suggested that "offensive" and "defensive" forms of attack may be distinguished on the basis of bite topography in laboratory rats. Dominant rats deliver fewer bites to the head, thorax, abdomen and genitals than to the backs of intruders, regardless of the relative accessibility of these areas to the attacking male. This preponderance of dorsal bites is also a feature of attacks by feral rats *(Ewer, 1971)*.

Non-random distributions of bites have also been reported for outbred ("TO") laboratory mice *(Childs, 1979)*. This author found that individually housed and reproductively experienced males directed a large proportion of their attack to the back, flanks and rump of opponents, whilst few bites were made on the head or ventral surface. In contrast, lactating females with litters bit the head and ventral surface of opponents almost as frequently as other areas, i.e. showed less "inhibition" of bites to vulnerable areas of the opponent. Childs (1979) concluded that social conflict and maternal aggression in mice were largely "offensive" and "defensive" forms of attack, respectively.

The present study undertook the analysis of bite topographies shown by mice from different strains and treatments in attacks on different types of opponent. The resulting information may be used to ameliorate the social stress associated with general husbandry, breeding and research activities.

Materials and methods

Animal husbandry

Mice employed in this study were bred and housed under controlled conditions in the Animal Facility of University of Wales Swansea. Animals were

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maintained on a reversed light cycle with white fluorescent lights on from 22.30 to 10.30 hours GMT. Ambient temperature, that affects inter-male fighting *(Greenberg, 1972)*, was regulated to 20°C ± 2°C. Handling and extraneous noise are stressful to laboratory rodents and were also carefully controlled. Noise restrictions were imposed on staff and fire alarms were operated at frequencies below the auditory threshold of mice. Handling was restricted to that required for routine maintenance and was avoided in the case of grossly pregnant females, where such disturbance may subsequently affect the physiology and behaviour of offspring. Since both litter size and the presence of sires affects the rate of development in mice *(Egan and Royce, 1973)* litters were culled to six at birth and sires were removed two days after parturition. Outbred subjects were weaned at 20-22 days and inbred mice at 27-29 days, since a high mortality rate was obtained in inbred mice weaned at the younger age. This presumably reflects a slower rate of development in inbred mice. After weaning, mice were randomly assigned to single-sex groups of six and housed on a sawdust substrate in M1-type opaque, polypropylene cages measuring 30cm x 12cm x 11cm (North Kent Plastics, U.K.). The wire lid of each cage contained an ad libitum supply of food (Pillsbury's breeding diet) and water. Inbred mice to be used for breeding purposes were allocated to cages in single-sex groups of littermates to facilitate subsequent brother-sister mating.

Genetic lines of mice

Mice of the following lines and coat colours were employed.

Outbred strains:	Tuck Ordinary (TO)	- albino
	Swiss Webster	- albino
Inbred strains:	NZW/Ola	- albino
	BALB/c	- albino
	C57BL/10	- black
	DBA/2	- grey
	CBA/Ca	- agouti
	C3H/He	- agouti

With the exception of NZW/Ola mice, which were obtained directly from suppliers at weaning (21-23 days), all lines were bred in the Animal Facility for at least three generations prior to behavioural testing. Strains were chosen mainly on the basis of differences in intermale aggression but breeding performance (reviewed by *Festing, 1979*) was also taken into consideration since it often deteriorates with inbreeding. Five of the inbred strains used (i.e. excluding NZW/Ola) are among the 10 most popular in research *(Festing, 1976)* thus optimising the relevance of this study.

As far as possible, inbred strains, particularly those of like coat colour, were housed in separate rooms to minimise the risk of genetic contamination by accidental inter-line mating.

Types of opponents

In these studies, encounters took place between two adult mice of the same or opposite sex. Such encounters employed a "Resident-Intruder" design in which one animal (the "opponent" or "intruder") was introduced for 10 minutes into the home cage of, or the "neutral arena" already occupied by, the other animal (the "resident"). Behavioural observations were concentrated mainly on "residents". The tests were terminated immediately if any individual was subjected to serious risk of injury.

Intruders were of two types namely

(i) Group-housed, anosmic TO strain males

(ii) Group-housed, anosmic males of the same line as the resident ("congenic" opponents)

All male intruders were housed in single-sex groups of six from weaning until used in behavioural tests. TO strain male intruders were 6-10 weeks old while the age of congenic opponents closely approximated that of their respective residents, both intruders and residents being generated by the same breeding cycle. Young adult TO males were chosen as intruders to minimise weight discrepancies between them and the lighter, inbred residents, since there is evidence *(White et al., 1969)* that such discrepancies may inhibit the appearance of aggression. Similar considerations resulted in rejection of any obviously underweight animals.

Care was taken to keep the cages of intruders and their corresponding residents separate in order to minimise any variation in subsequent interactions as a result of odour-mediated familiarisation *(Brain et al., 1982)*. Efforts were also made to ensure that the cages of residents from different strains or treatments occupied comparable shelf positions since this feature of housing has been reported to affect the behaviour of mice *(Hegmann & Possidente,* *1981)*. The shelf position of intruders was also controlled.

Although group housing generates relatively non-aggressive males, male intruders employed in the present study were rendered temporarily anosmic to further discourage attacks on residents. Peripheral anosmia may be temporarily induced in rodents by perfusion of the nasal cavities with a dilute solution of zinc sulphate *(Thor & Flannelly, 1977)*. Anosmia induced in this manner is thought to be the result of changes in the nasal epithelium, involving extensive necrosis of sensory and supporting cellular structures *(Takagi, 1971)*. Zinc sulphate-induced anosmia abolishes all aggressive acts normally directed by rats against unfamiliar conspecifics while anosmic intruders evoke the same aggressive response as untreated ones *(Flannelly & Thor, 1976)*. There is also evidence that anosmic mice do not initiate attack and rarely retaliate, even when severely attacked. Careful note was made of any retaliation observed in the present study (these were rare).

One can only really attempt to assess the 'aggressiveness' of subjects by using opponents that a) can behave freely and b) will not confuse the issue by contributing to fights. Use of temporary anosmia was thought to be preferable to using castrates (which do not generate the appropriate olfactory cues) or repeatedly defeated subjects (which may be severely stressed and rather variable). There is less fighting than if both mice are intact.

All male intruders were subjected to the following procedure both 72 hours and 24 hours prior to behavioural testing. Four- percent zinc sulphate solution was applied to the nares of the lightly anaesthetised (ether) animal using a 1-ml syringe with a carefully blunted needle. The animal was then held head downward, until fully conscious, with its nose in contact with absorbent paper towel. This reduces the likelihood of the toxic solution being ingested. Zinc sulphate-induced anosmia, as expressed by the failure of animals to distinguish between distilled water and urine from like-strain females, has been found (in this laboratory) to persist for up to 6 days.

Both male and female intruders were visually distinguished from residents by application of methyl violet fur dye to the backs of the former. C57BL/10 intruders were tail-marked with non-toxic white paint since no available fur dyes were visible against their black coats. Intruders were used once only in encounters with residents.

N.B. in the text of this paper the term "congenic" is used to denote anosmic, group-housed male intruders of the same strain as the resident and not in the strict genetic sense of two strains of mice differing at a single locus (*Festing 1979*, p. 24).

Videotape recording of intraspecific encounters

All intraspecific encounters took place under dim red lighting during the dark phase of the reversed light cycle and were recorded using a black-and-white camera sensitive to light intensities as low as 0.3 lux (National Panasonic Model 1350A with a 12.5-75mm, f 1.8-16 lens, Fuji Photo Optical Co., Japan). During taping, the test cage was fitted with a Perspex lid having ventilation holes along both its longer edges. The camera was mounted vertically, approximately 5 feet (1,5m) above the cage, and was connected to a monitor and videotape recorder ("U"-matic, Model CR6060E, Victor Company of Japan Ltd.). A timer (Model VTG33, For-A Company Ltd., Japan) was also incorporated into the circuit and superimposed a digital time display on the videotape record (Sony KCA60 videocassettes). A "frame advance" facility of the recorder allowed events 20 milliseconds apart to be distinguished during playback.

A camera capable of operating in dim red light has important advantages. It allows tests to be conducted under conditions approximating the nocturnal habit of this species, since red light is beyond the visual spectrum of mice *(Thiessen & Lindzey, 1968)*. Moreover, there is evidence to suggest that many of the behavioural differences observed between albino and pigmented strains of mice (e.g. *Thiessen et al, 1970*) are a function of differential responses to test illumination (Creel, 1980). Under high intensity illumination, albino mice differ from pigmented ones in "fear" responses (Henderson, 1979) and show suppression of general activity (Creel op. cit.) and intermale aggression *(Klein et al, 1970)*. Low intensity illumination therefore minimises differences in behaviour due to varying sensitivity to light. At this point it seems appropriate to note that the agouti C3H/He strain carries the gene for retinal degeneration (rd) and shows total loss of rod photoreceptors by the age of 7 weeks. The cones, however, survive for up to 18 months in a functional state and probably mediate some kinds of visually guided behaviour *(Carter-Dawson et al. 1978).*

Experimental procedures

This section describes the housing conditions of "residents" and the situations in which animals were tested for aggressive behaviour.

Aggression induced by individual housing

Individual housing has proved a popular method of inducing fighting in mice since it requires no sophisticated manipulations, such as surgery, and produces a behavioural change that is relatively chronic *(Valzelli, 1969)*. Individual housing is often referred to as "isolation", a more succinct but less accurate term since "isolated" mice are usually in olfactory and auditory communication with conspecifics *(Brain & Benton, 1983).* Fighting induced by this means has been considered as both "inter-male" and "territorial" by Moyer (1968) and has been variously interpreted as a pathological response to "social deprivation" *(Valzelli, 1973)* or the result of removing an aggression-inhibiting effect provided by other members of a social group *(O'Donnell et al., 1981)*.

The amount of fighting generated is affected both by the age at, and duration of, "isolation" *(Goldsmith et al, 1976)*. In the present study these factors were kept constant across strains to facilitate comparisons. Mice were individually housed 7 days after weaning and were tested after a period of 28 days. This treatment is designed to minimise variation in the fighting experience of residents by separating them before inter-male fighting develops (as early as day 29 in some strains) while allowing aggression tests to be conducted on adults. Mice from all strains and both sexes were subjected to this treatment.

Aggression tests consisted of 10-minute encounters conducted (and recorded) under dim red light in the resident's home cage. To control for potential circadian variation in fighting activity, all animals in these studies were tested between 10.30 and 15.30 hours. Home cages were cleaned 24 hours prior to testing to reduce the effect on aggression of variation in cage odour *(Brain et al, 1982)*.

Kessler et al, (1975) showed that the intensity of intermale fighting might depend on the strain of the opponent. To enable the effect of opponent strain on the behaviour of isolated males to be assessed in greater detail, male residents were tested (on the 29th or 30th day of isolation) against either an anosmic, group-housed TO male (TO intruder) or an anosmic, 'congenic' intruder.

Aggression induced in males by reproductive experience

Housing with females has been found to augment conspecific attack in both male rats *(Barnett et al, 1968)* and mice *(Brain et al, 1978)*. This phenomenon is usually interpreted as suggesting an effect of copulatory behaviour on male hormones that then act to increase aggression, although the mechanism of fighting inhibition release (as applied to "isolation"-induced fighting) may also be invoked since housing with females does not involve male-to-male contact.

As in the case of aggression induced by individual housing, attack by males from cohabitation with females may be classed as "inter-male" *(Moyer, 1968)*. These two forms of aggression show similar responses to certain physiological and situational manipulations and consequently they have been classed by Brain (1981) as "social conflict".

In the present study, males and females of the same strain were paired when 9 weeks old. Males were removed to separate cages on the day of their partner's parturition (as judged on the basis of twice-daily checks of breeding pairs), and tested 24 hours later in their individual cages with either an anosmic TO intruder or an anosmic intruder of the same strain (congenic opponents).

N.B. Since all males from cohabitation with females successfully sired a litter, they are referred to in the text as "reproductively-experienced males". It is possible that, in addition to experience of copulation and the removal of "fighting inhibition", contact with their own offspring may be a factor in promoting aggression in these males.

Maternal aggression

Some female rodents become highly aggressive shortly after the delivery of their young (parturition), a response referred to as "maternal" or "post-partum" aggression *(Svare, 1981a)*. Since this behaviour occurs spontaneously during lactation, a reproductive state in which wild females spend the majority of their adult life *(Brown, 1953)*, it seems to provide an ethologically relevant model for research on female aggressive behaviour that has, in comparison with male aggression, received scant attention. Maternal aggression, which ostensibly serves to protect the young offspring, is induced by suckling stimulation and reaches peak intensity in mice between the 3rd and 8th days of lactation, being completely suppressed by removal of the litter *(Svare & Gandelman, 1973)*.

The lack of attack stimulus specificity shown by lactating females, together with the implication of hypothalamic involvement in its control, have led Svare (1981a) to class maternal aggression as a form of "irritable aggression". Moyer (1968) has classed "maternal aggression" as a separate behavioural category. Brain (1979), in contrast, terms it "parental defence"; implying that protection of the young may be shown by parents of either sex. The term "maternal aggression" is preferred here since male parents were not tested for aggression in the presence of their offspring.

The primiparous female mates of animals described in the previous section were tested for maternal aggression in their home cage, and in the presence of their litter (culled to six at birth) on the 5th day of lactation, the day of parturition being considered the first day of lactation. Each female encountered either a TO or congenic male intruder. Attacks on female intruders have also been demonstrated *(Svare & Gandelman, 1973)* but with less consistency between studies than for males which were therefore used in preference here. As in all tests of intraspecific aggression employed in this study, each encounter was of 10 minutes duration. Shredded paper was provided as nesting material but nests were not constructed by all strains. To avoid possible disruption of lactation by handling of females, the routine cleaning of home cages 24 hours prior to behavioural testing was not performed in the case of maternal aggression. It was not possible to complete all the tests in these lines when using congenic opponents because of a high incidence of attempted cannibalism.

Results

The bite topographies shown by mice from various treatments and lines are presented in Tables 1 to 6 as medians (and ranges) of percentages of bites delivered by individual subjects to each of five body areas, namely the flanks, back, rump, head and ventral surface of the opponent. In addition, the totalled numbers of bites per line and the incidences of attacking individuals are given. Statistical comparisons attempted to assess variations between the lines of mice, the different test situations and with different types of opponent. Any differences are broadly supported by other measures of attack behaviour (see *Jones and Brain, 1987)*.

Bite topographies shown by individually-housed males

Individually housed males generally deliver bites to the flanks and back of "TO" opponents and, to a lesser extent, the rump (see Tables 1 and 2). In comparison, the head and ventral surface are rarely bitten. Strain differences in bite topography are most obvious in connection with dorsal bites, which are significantly higher in C57BL/10, and lower in BALB/c mice, in comparisons with all other strains (Table 1). Using congenic opponents (Table 2) did not produce such clear evidence of differences between the genetic lines and the pattern of attack was broadly that seen with TO opponents. To summarise, the bite topography of individually housed males shows some variation with respect to the genetic line of resident and intruder but may nevertheless be interpreted as "offensive" attack.

There was great variability in the incidence and intensity of biting in the different strains with TO and Swiss mice showing greater responses than many other lines whereas C3H/He animals were very docile. Using different types of opponent had a profound effect on behaviour of some lines e.g. BALB/c which showed impressive levels of response to TO opponents but no attacks on congenic mice.

Bite topographies shown by reproductively-experienced males

The distribution of bites shown by reproductively experienced males to both TO and congenic opponents (Tables 3 and 4) are also indicative of "offensive" attack. The back, rump and flanks of intruders are frequently bitten while bites to the head and ventral surface are few or absent. In comparison to individually housed counterparts, mated males show higher proportions of dorsal bites. This diffe-

Table 1

Bite topographies shown by individually-housed males (N=12) in attacks on “TO” line opponents

LINE OF RESIDENT	BODY AREA BITTEN					TOTAL NUMBER OF BITES	NUMBER OF ANIMALS BITING IN SAMPLE
	FLANKS	BACK	RUMP	HEAD	VENTRAL SURFACE		
a) TO	54.9 (38.4-66.2)	14.0 (5.6-52.9)	18.9 (5.9-51.2)	0 (0-3.6)	4.5 (0-10.0)	471 a	6
b) SWISS	50.0 (33.3-80.0)	18.8 (0-35.7)	25.0 (0-58.3)	0 (0-4.5)	0 (0-4.3)	276	11 c
c) NZW/01a	31.0 (0-66.7)	35.7 (5.6-100.0)	25.9 (0-65.3)	0 (0)	0 (0-8.3)	232	9
d) BALB/c	43.8 (0-79.0)	2.8^* (0-18.2)	41.7 (21.1-100.0)	0 (0-4.3)	0 (0-5.6)	278 b	7
e) C57BL/10	31.1 (0-50.9)	55.8^* (29.2-100.0)	10.4 (0-22.9)	0 (0-8.3)	1.1 (0-3.8)	296 b	6 r
f) DBA/2	53.3 (34.8-100.0)	18.8 (0-47.8)	20.0 (0-36.4)	0 (0)	0 (0-4.0)	122	9 r
g) CBA/Ca	0	0	0	0	0	0	0 d
h) C3H/He	50.0	50.0	0	0	0	2	1 d

a differs from b; c and f $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
b differs from f $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
c differs from a; d and e $P<0.05$ on Fisher’s exact probability test
d differs from all other lines $P<0.05$ on Fisher’s exact probability test
r differs from reproductively-experienced counterparts $P<0.05$ on Fisher’s exact probability test

Table 2

Bite topographies shown by individually-housed males (N=12) in attacks on ‘congenic’ opponents

LINE OF RESIDENT	BODY AREA BITTEN					TOTAL NUMBER OF BITES	NUMBER OF ANIMALS BITING IN SAMPLE
	FLANKS	BACK	RUMP	HEAD	VENTRAL SURFACE		
a) SWISS	37.5 (20.0-83.6)	17.8 (0-60.0)	28.1 (5.7-67.7)	0 (0-1.9)	0 (0-9.4)	286	10 a
b) NZW/01a	37.5 (16.7-100.0)	6.3 (0-66.7)	38.7 (0-62.5)	0 (0-11.1)	0 (0-3.2)	230	7
c) BALB/c	0	0	0	0	0	0	0 b
d) C57BL/10	19.4 (9.8-25.5)	17.3 (4.9-48.7)	56.1 (25.0-85.5)	0 (-1.3)	1.6 (0-7.5)	524	8
e) DBA/2	22.2 (6.3-37.5)	33.3 (15.6-58.3)	46.9 (22.9-50.0)	0 (0-4.2)	0 (0-3.7)	162	5
f) CBA/Ca	71.4 (50.0-75.0)	14.3 (0-50.0)	0 0-25.0)	0 0-14.3)	0 (0)	33	3
g) C3H/He	36.5 (28.6-44.4)	37.7 (11.1-64.3)	25.8 (7.1-44.4)	0 (0)	0 (0)	37	2 c

a differs from e and f P<0.05 on Fisher’s exact probability test
b differs from all other lines except g P<0.05 on Fisher’s exact probability test
c differs from a; b and d P<0.05 on Fisher’s exact probability test

Table 3

Bite topographies shown by reproductively-experienced males (N=12) in attacks on “TO” line opponents

LINE OF RESIDENT	BODY AREA BITTEN					TOTAL NUMBER OF BITES	NUMBER OF ANIMALS BITING IN SAMPLE
	FLANKS	BACK	RUMP	HEAD	VENTRAL SURFACE		
a) TO	19.2 (0-53.6)	70.5 (39.3-100.0)	4.7 (0-15.4)	0 (0-5.6)	0 (0-3.5)	366 a	8 f
b) SWISS	19.3 (0-32.9)	50.0 (19.1-86.7)	24.6 (6.7-63.6)	0 (0-2.6)	0 (0-6.8)	624 a	11
c) NZW/01a	53.8 (31.1-100.0)	6.1 (0-66.7)	0 (0-60.7)	0 (0-3.0)	0 (0-16.7)	159	9 f
d) BALB/c	72.7 (44.4-80.0)	9.3 (0-55.6)	13.6 (0-29.2)	0 (0)	0 (0)	52	4
e) C57BL/10	0	0	0	0	0	0	0 b
f) DBA/2	31.6	42.1	26.3	0	0	19	1 c
g) CBA/Ca	0	0	0	0	0	0	0 c
h) C3H/He	0	0	0	0	0	0	0 c

a differs from all other lines $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
b differs from a; b and c $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
c differs from a; b; c and d $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
f differs from female counterparts $P<0.05$ on Fisher’s exact probability test

Table 4

Bite topographies shown by reproductively-experienced males (N=12) in attacks on ‘congenic’ opponents

LINE OF RESIDENT	BODY AREA BITTEN					TOTAL NUMBER OF BITES	NUMBER OF ANIMALS BITING IN SAMPLE
	FLANKS	BACK	RUMP	HEAD	VENTRAL SURFACE		
a) SWISS	43.3 (14.3-65.0)	21.3 (4.0-45.5)	27.5 (0-50.9)	0 (0-14.3)	5.0 (0-20.6)	779 a; i; f	11 b
b) NZW/Ola	34.7 (14.3-37.5)	18.8 (16.7-28.6)	47.0 (40.0-57.1)	0 (0)	0 (0-5.0)	78	4
c) BALB/c	65.9 (48.3-100.0)	0 (0-9.1)	24.2 (0-50.0)	0 (0)	4.6 (0-9.1)	43	4
d) C57/BL/10	36.1 (27.8-44.4)	36.3 (33.3-39.2)	27.6 (22.2-32.9)	0 (0)	0 (0)	124	2
e) DBA/2	0	0	0	0	0	0	0
f) CBA/Ca	0	0	0	0	0	0	0
C3H/He	40.0	40.0	20.0	0	0	10	1

a differs from all other lines except d $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
b differs from all other lines $P<0.05$ on Fisher’s exact probability test
i differs from individually-housed counterparts $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
f differs from female counterparts $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test

rence reaches significance when mice from each treatment are compared irrespective of their line. In the case of reproductively experienced mice, bites aimed at the backs of "Upright defensive" opponents may, due to the rotation of the opponent during the attacker's biting lunge, be delivered to the flanks. In this posture, the rump is relatively inaccessible and less likely to be bitten than the flanks. It may not be a simple coincidence, therefore, that the preponderance of rump bites evident in attacks by DBA/2 and C57BL/10 males on congenic but not on "TO" opponents is associated with a significant increase in the frequency of chasing congenic (compared with "TO") opponents. This suggests both C57BL/10 and DBA/2 intruders have a greater tendency to "Flee" when bitten than do "TO" intruders, which may adopt "Upright defensive" postures more readily. The failure of BALB/c males to bite "TO" intruders dorsally is not so readily explained. Lateral attack (i.e. from a "Sideways offensive" posture) does not predominate (over "Upright offensive") as in other strains and may reflect a reduced tendency to lunge around upright intruders to deliver back bites. The prominence of upright (compared with supine) defensive postures in mice (compared with rats) may explain why sideways "offensive" postures predominate in most lines over upright ones, since lateral attack is more effective in achieving dorsal bites on upright opponents.

Ventral bites, although relatively infrequent, are more common in males of the TO and C57BL/10 strains than in some other lines. Childs (1979) has suggested that ventral bites may result from reduced target discrimination during intense attacks, rather than from a change of "aim". The observation that TO and C57BL/10 males also show relatively high attack intensities (for this treatment) seems consistent with this view, though this explanation may not hold for all strains or treatments. Reproductively experienced Swiss males, for example, show attacks of similar intensity on TO and "congenic" opponents but different incidences of ventral biting (Tables 3 and 4).

Thus the bite topographies observed in individually housed males might reflect the attack and defence strategies of particular residents and intruders. Since a preference for dorsal sites has also been demonstrated for mice *(Blanchard et al., 1979)*, which commonly chase fleeing opponents (bites "aimed" at the back of fleeing opponents might fall short and be delivered to their rump). Similarly, the greater duration of cohabitation of these mice with male conspecifics (9 weeks compared with 4 weeks in isolates) may render their attack strategy more "successful" (in terms of delivering bites to a "preferred" dorsal target). These effects presumably result from greater fighting experience prior to tests of "aggression" and may be comparable to the effects of social experience, which improve the effectiveness of defensive strategies in rats.

Apart from a continued tendency for BALB/c males to show few dorsal bites, there are no obvious "strain-characteristic" bite patterns. There is a tendency, however, for Swiss and BALB/c males to show higher proportions of ventral bites in attacks on congenic opponents. This distinction is also evident in lactating females of these strains (Tables 3 and 4) where it reaches significance in the case of Swiss females. This effect does not seem related to the intensity of biting attack but may be related to the defence strategy of congenic opponents (and hence the attack strategy of residents). There is evidence, for example, of higher frequencies of "Fight" and "Opponent-on-back" for Swiss males in encounters with congenic opponents. Thus the opponent may vigorously defend itself (by kicking etc.) and precipitate wrestling attack where the likelihood of ventral bites may be greater.

TO and Swiss mice showed significantly higher incidences and intensities of this form of attack than all other lines used whereas consistently low incidences were obtained in other lines e.g. DBA/2, CBA/Ca and C3H/He. Reproductively experienced Swiss males paired with congenic opponents showed significantly more attack than individually housed males or lactating female counterparts. The incidences of attack in TO and NZW/Ola lines of reproductively experienced males were also higher than in lactating female counterparts (see later).

Bite topographies shown by lactating females

Many features of maternal aggression, including its short latency and the lack of discrimination between male, female and juvenile opponents *(Svare & Gandelman, 1973)*, are consistent with its ostensible function of pup defence *(Svare, 1981b)* and distinguish it from the "offensive" attacks of males.

It is not surprising, therefore, that females show significantly higher proportions of ventral and head bites than male mice from all other treatments (Tables 5 and 6), suggesting that maternal attack is more "defensive".

Lactating females, especially those of the Swiss strain, distinguish between "TO" and congenic opponents by directing high proportions of ventral bites towards the latter. This cannot be explained in terms of attack intensity or "wrestling". Some feature of the congenic opponent (e.g. its behaviour or subtle visual or olfactory cues) may render it a more "threatening" stimulus to females of these lines? The preponderance of rump and flank bites, but lack of dorsal bites, by lactating females might indicate an inefficient attack strategy through lack of fighting experience or that females do not show a "preference" for dorsal sites as do male mice *(Blanchard et al, 1979)*.

Swiss animals showed a significantly higher incidence and intensity of biting than most of the other lines.

Discussion

The bite topographies obtained in the present study suggest that attack by males from all treatments may be classed as "offensive" whilst, in comparison, attack by lactating females (i.e. maternal aggression) may be classed as "defensive". Differences in bite topographies between strains may in part be related to observed differences in the particular attack and defence strategies of residents and intruders, respectively, e.g. chasing of opponents seems to be associated with high proportions of rump bites.

Ventral bites, though of low incidence, are more prominent in mice than rats *(Blanchard et al, 1979)*. This apparent lesser constraint may, these authors have suggested, relate both to the nature of the wounds inflicted by, and the feral social organisation of, these two rodent species. Mice make small punctuate wounds (compared with the tearing bites of rats) which are not associated with high mortality when delivered to the ventrum of opponents, thus making constraints on bites to this area less necessary in terms of injury-limiting strategies. In addition, ventral bites (which in rats may kill or castrate opponents) are not as maladaptive in mice since, being territorial, they are less likely (compared with the colonial rat) to injure a closely related animal. The preference for dorsal sites shown by rats is consequently interpreted by these authors as adaptive as they may serve to disperse younger, closely related males (thus reducing competition for food and females) without rendering the bitten males incapable of reproduction.

The lesser reluctance of mice to bite the ventrum of opponents has been used to explain *(Blanchard & Blanchard, 1981)* the absence of an obvious supine defence posture and the corresponding "On-top-of" attack posture. Although the back is protected in the supine posture, if, as in mice, the attacker is willing to bite the ventrum, then "the dangers involved in this defensive strategy would outweigh the benefits". In the "Upright defensive" posture, however, the mouse can manoeuvre to keep its back relatively inaccessible, while the position of its head does not preclude retaliatory biting, as does the supine position. The high incidence of head bites in attacks by rats and mice on anaesthetised conspecifics *(Blanchard et al 1977; Blanchard et al, 1979)* has been used to confirm the role of retaliatory biting in protecting the head from attack. In the present study, the consistently low proportions of head-bites confirm the "protected" nature of this site.

The attack and defence strategies of mice and rats as discussed here apply to the confined laboratory environment and to feral situations where, for some reason, subordinates are forced to live in close proximity to dominant males. Flight, however, makes strategies for the protection of dorsal sites unnecessary, and may be a primary defence in both rats *(Blanchard & Blanchard, 1981)* and mice *(Blanchard et al, 1979)*. Flight remains a prominent defensive behaviour of mice even in inescapable laboratory situations, where it is extremely dangerous, precipitating nearly 60% of the total number of attacks *(Blanchard et al, 1979)*.

Blanchard and Blanchard (1981) have opined that the supine posture of rats is a defence strategy rather than a "submission signal" (cf. *Grant, 1963*). Although the posture does not inhibit the opponent from attempting to attack, it provides some relief from biting, by reducing the attacking animal's access to the dorsal surface.

In animal welfare terms, the present data clearly reveal that potentially damaging biting attacks in social contexts appear at different intensities in dif-

Table 5

Bite topographies shown by lactating females (N=12) in attacks on “TO line opponents

LINE OF RESIDENT	BODY AREA BITTEN					TOTAL NUMBER OF BITES	NUMBER OF ANIMALS BITING IN SAMPLE
	FLANKS	BACK	RUMP	HEAD	VENTRAL SURFACE		
a) TO	10.3 (0-63.2)	0 (0-5.3)	21.0 (0-80.8	0 (0-2.6)	10.5 (6.4-100.0)	98	3
b) SWISS	60.7 (22.4-100.0)	9.6 (0-28.6)	15.4 (0-66.7)	1.3 (0-36.7)	5.5 (0-13.5)	549 a	11 b
c) NZW/Ola	66.7	16.7	0	0	16.7	6	1
d) BALB/c	49.0 (33.3-66.7)	0 (0-12.5)	33.3 (33.3-45.5)	2.0 (0-16.7)	0 (0.16.7)	96	5 c
e) C57BL/10	0	0	0	0	0	0	0
f) DBA/2	0	0	0	0	0	0	0
g) CBA/Ca	0	0	0	0	0	0	0
h) C3H/He	0	0	0	0	0	0	0

a differs from all other lines $P<0.05$ on 2-tailed Mann-Whitney ‘U’ test
b differs from all other lines except d $P<0.05$ on Fisher’s exact probability test
c differs from e; f; g and h $P<0.05$ on Fisher’s exact probability test

Table 6

Bite topographies shown by lactating females (N=12) in attacks on ‘congenic’ opponents

LINE OF RESIDENT	BODY AREA BITTEN					TOTAL NUMBER OF BITES	NUMBER OF ANIMALS BITING IN SAMPLE
	FLANKS	BACK	RUMP	HEAD	VENTRAL SURFACE		
a) SWISS	41.1 (25.0-55.0)	14.8 (0-41.7)	21.7 (5.0-30.0)	5.6 (0-18.2)	17.9^* (2.7-34.3)	284	8 a
b) BALB/c	52.6 (31.3-56.0)	6.0 (5.3-6.3)	36.8 (18.0-56.3)	5.3 (0-10.0)	6.3 (0-10.0)	101	3
c) C3H/He	0	0	0	0	0	0	0

- NB NZW/Ola, C57BL/10, DBA/2 and CBA/Ca lines could not be tested satisfactorily because of pup mortality

a differs from c $P<0.05$ on Fisher’s exact probability test ferent genetic lines of mice (a factor which should be considered in experimental design). The sex and recent reproductive experiences of the animals may also have a profound influence on such behaviours. Indeed, although biting is less common, the activity can be most potentially damaging in the females of some lines as well as being influenced by the nature of the opponent (some lines respond differently to animals of different genetic backgrounds depending on their similarity to the subject). Given the recent 'explosion' in the numbers of created transgenic lines of mice, it seems important to reiterate that the line selected for genetic modification will have an important effect on the generated levels of 'social stress'. All other things being equal, it is better to select lines in which males and females are docile especially if the intention is to group-house the resulting progeny for extended periods of time.

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