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STOWAWAY TRANSPORT RATES OF HOUSE MICE (*MUS DOMESTICUS*) AND DEERMICE (*PEROMYSCUS MANICULATUS*)*

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ABSTRACT: Stowaway transport rates were obtained from behavioral observations of 14 house mice (*Mus domesticus*) and 14 deermice (*Peromyscus maniculatus*) during commercial transport. One house mouse escaped during unloading and flaking of 2,500 kg 2-year old oat hay. Three house mice and two deermice were handcaught on trucks during unloading of 51,110 kg transported barley straw and grass hay. One house mouse was handcaught after being buried alive in grain. Two house mice escaped during unloading of a semitruck holding dog food. House mice (one live, six recently-dead) and deermice (two recently-dead) were taken from a feedmill screen over which had passed 940,313 kg grain. Ten deermice were trapped in a pickup truck cab. In conclusion, multiplying these transport rates, ($7(10^{15})$ house mice per transported kg hay and $7(10^6)$ house mice per transported kg grain, by the worldwide volume of transported hay and grain implies thousands of stowaway house mice occur. Deermice have comparable transport rates. Experiments (N=82) were done to determine if a particular sex or age predominates among stowaways. For each experiment, I put one to five hand-caught or trapped house mice in a haybale, let them remain in the haybale for 1 to 13 days, put the haybale in a wheelbarrow, pushed the wheelbarrow for 15 minutes, and then searched for transported mice. Stowaways, mice remaining during transport, included 74 of 115 mice (64 %), which were primarily lower weight classes, characterizing house mice coming into reproduction. In conclusion, live stowaways should arrive in > 50 % infested loads.

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INTRODUCTION

Behavioral Ecology Of Stowaways

Historical transport of mouse-infested feed and hay is the widely-accepted cause of the worldwide distribution of commensal house mice *Mus domesticus* (Auffray et al. 1990; Navajas y Navarro and Britton Davidian 1989). House mice meet criteria of successful invaders, including abundance and commensalism, wide diet and habitat breadth, short generation time, high genetic variability; and include pregnant colonists (Ehrlich 1987). However, no data on passively transported mice could be found in volumes 1972 to 1992 of BIOLOGICAL ABSTRACTS or ZOOLOGICAL RECORD (MAMMALIA). By chance, I found Suzuki's (1980) reporting 36 and 28 trapped "Mus" stowaways on two overseas cargo ships. Stowaway house mice transported in garbage probably started an isolated population of house mice at an Ontario dump, which was surrounded by deermouse (*Peromyscus maniculatus*) populations (personal communication, Brock Fenton). Though little behavioral data on stowaways exist, an extensive literature implies stowaways have a genetic impact.

"Foreign" Genetic Markers Of Stowaways

Chance events or selection causes a variety of genetic markers to have restricted geographic distributions in commensal house mouse populations (Sage 1981; Sage et al. 1986a, b; Boursot et al. 1984, 1993; Britton-Davidian 1990; Potter et al. 1986; Berry and Corti 1990; Vanlerberghe et al. 1986, 1988a, 1988b). Examples of known "foreign" genetic markers, which are outside their established ranges, include 19 enzyme loci (total populations surveyed at origin and destination -9), 19 Robertsonian chromosomal translocations (33

populations), 2 mitochondrial DNA (4 populations), 3 Y chromosomal DNA (5 populations), genetic resistance to a retrovirus (2 populations), 5 histocompatibility haplotypes (7 populations), 3 t haplotypes (6 populations), 2 homogeneously-staining regions of chromosome 1 (8 populations), and 1 morphometric marker (2 populations) (Baker, Submitted). Robertsonian translocations occur when two acrocentric chromosomes fuse at their centromeres. Haplotypes designate a chromosome region where several genetic loci have related functions and are inherited together. Stowaways breeding with residents produce progeny carrying these "foreign" genetic markers, thus spreading identical genetic markers in mouse populations in cities connected by commerce. Documenting the survival and spread of unique "foreign" genetic markers provides insight to the possible origin of mice in specific populations and an understanding of their current geographic distributions.

Four explanations for the presence of "foreign" genetic markers follow: 1) identical by descent, 2) identical by kind, 3) identical by descent from escaped laboratory mice, and 4) inaccurate original geographic range.

Identity by descent. The strongest examples of the genetic impact of passively transported mice involve those having several "foreign" markers because it is unlikely that all would mutate simultaneously to be the same as those in the putative population of origin. Six examples of mice having more than one "foreign" genetic marker follow: 1) Binasco and Bergamo, Italy and Tubingen, Germany populations have the same Robertsonian translocation and rare transferrin allele (Britton Davidian et al. 1989); 2) Zadar, Yugoslavia and Palermo, Sicily mice share three Robertsonian translocations (Winking et al. 1988); 3) Eday and Faray Island mice share three Robertsonian translocations (Bauchau 1990); 4) Orobic and Molise, Italy mice share two Robertsonian

*In memory of J.S. Williams, statistician, quantitative geneticist, friend.

chromosomes and Y chromosomal DNA (Tucker et al. 1989; Tucker et al. 1992); 5) mice sharing histocompatibility (H2) and t haplotypes were trapped in the following pairs of sites: Temuco, Chile and New York City (histocompatibility haplotype: H2 w31; t haplotype: tw5), Ann Arbor, Michigan and Haifa, Israel (H2 w2, twSL), and Paris, France and Clinton, Montana (H2 w28, tw12) (Nizetic et al. 1982); and 6) mice from Sofia Bulgaria, which is within the range of *M. musculus*, carried 5 of 6 *M. domesticus* genetic markers (Francois Bonhomme unpublished data). Confirming the genetic contributions of stowaways requires sampling more markers and more geographic sites (after Baker et al. 1989; Corbet 1990).

Identity by kind. Independent evolution might cause two genetic markers to look the same. For example, the origin of the same Robertsonian translocation in different populations may be by independent evolution (Britton Davidian et al. 1989; Nachman et al. 1994) or by stowaways (Bauchau 1990). Independent evolution is supported by isolated populations sharing the same Robertsonian translocation but different mitochondrial DNA. However, recombination could cause stowaways to retain their Robertsonian translocations, but acquire different mitochondrial DNA (Nachman et al. 1994); >2 mice per location may be necessary to sample all mitochondrial DNA (Ritte et al. 1992). Putatively identical bands on electrophoretic gels can be checked using other screening protocols that separate alleles by charge or size (Bonhomme and Selander 1978). Alternatively, genes can be sequenced or subject to high resolution electrophoresis. Environment may change the phenotype. For example, rearing temperature influences tail length (Thorington 1970).

Escaped laboratory mice. Escaped lab mice must be distinguished from stowaways (Berry et al. 1981). Screening genetic markers distinguishing among lab strains may help discriminate progeny of stowaways from escaped lab mice. However, recombination over generations decreases the number of strain-specific genetic markers, which is the main diagnostic used to distinguish lab strains.

Inaccurate original geographic range. Geographic boundaries of genetic markers are rarely known in detail. For example, Milinichkov (et al. 1990) reported 74 of 78 mice trapped within the range of *M. musculus* had *M. domesticus* genetic markers. Retrapping and screening genetic markers of mice trapped at the same sites showed Milinichkov was working in a hybrid zone (Francois Bonhomme, personal communication). Some other sites where Bonhomme et al. reported "foreign" genetic markers are near hybrid zones. These sites warrant further trapping to determine their distances from hybrid zones.

Exploratory Study

The following study reports low transport rates, such as 7(10⁻⁴) mice per transported kg grain and 7 (10⁻⁵) mice per transported kg hay. Multiplying these low transport rates by the huge volume of transported grain and hay results in thousands of stowaways transported worldwide. Infestations that keep occurring despite pest control may be caused by stowaways. Experimental data imply a

> 50 % chance of stowaways being transported in infested loads.

MATERIALS AND METHODS

Anecdotes

To learn more about stowaway movements for subsequent quantitative study, I interviewed transport workers (12 hay haulers; 5 garbage haulers; 10 farm workers) and others. Transported infestations occurred in goods that had been stored for several months at sites where mice occurred, such as farms, barns, garages, warehouses, and homes. Stowaways were transported in garbage, hay, feed, grain, household goods, dog food, a mattress, and tires. Stowaway mice infested garbage that had been stored for ca. > 6 months until a designated "free day," when garbage could be unloaded at the dump without charge. Hay denotes the stalk of pasture grass, alfalfa, clover, oat, millet, sorghum-sudan grass, or wheat that is mown in bloom, allowed to dry, and baled for animal feed. Straw denotes the stalk of barley, oat, or wheat that is left after grain removal, allowed to dry, and baled for animal bedding. Straw retains some grain. Transported grain is usually moved by elevators or by augurs (Archimedes screws). Mice may suffocate when buried alive in grain or may be cut when going through an augur. However, live mice arriving with incoming grain were seen routinely by flour mill workers. Evidence supporting these stowaways arriving with incoming grain rather than being mice living in the mill and falling into the grain includes an intensive trapping program inside the mill and on the surrounding grounds. Only one mouse was trapped inside the mill within a 12-month period (personal communication, Lori R. Armstrong).

Pallets And Forklifts

House mice can live among and within stacked goods and pallets on which goods are transported and stored. Progressively larger volumes of goods transported by forklift and semitruck increase the risk that stowaways move undetected. Despite intensive rodent control, mouse infestations associated with pallets of dog food reoccurred at four California branch stores receiving semitruck loads of dog food from the same Nevada warehouse. Stores adjacent to these four branch stores had no rodent infestations implying effective rodent control. Two house mouse stowaways were seen escaping from a pallet holding dog food being forklifted from a semitruck. After control of the rodent infestation in the Nevada warehouse, minimal mouse infestations occurred at these four branch stores (personal communication, Randall G. Blair; Table 1).

Intentional Transport

Though the focus of this work is stowaways being transported accidentally by people, three manufacturers of live-traps encourage transport of live mice. Manufacturers urge discarding trapped live mice in garbage or releasing them outdoors (trap M007 from Woodstream Corporation, trap MFD from Trap-Ease Incorporated, Trip-Trap from North West Plastics Ltd.) (Rex Marsh and Vincent Bauchau, personal communication).

Questions Answered By This Study

Anecdotes show how infestations are transported. However, quantitative data on transport rates can help in allocating control measures and in estimating the potential genetic impact of stowaways.

What is the transport rate of mice infesting commercially-transported hay and grain? Stowaways were found using the following methods. Workers unloading transported animal feed, hay, or straw, hand-caught live mice and donated their carcasses, which were scored for weight (an approximation of age for field-trapped mice) and breeding status (Table 1). Observations were made of unloading hay bales (14 h). A feedmill grain-cleaner was inspected for carcasses on 17 days. Unsuccessful methods follow. Observations were made of unloading garbage (12 h). USDA Federal Grain Inspection Service records of 369 railroad cars holding hard red winter wheat were examined for rodent contamination because some handlers clean grain prior to railroad transport; intensive inspection occurs for wheat and other products consumed by humans; and the proportion of infested transported loads can be approximated. A Teamsters newsletter publicized a request for cooperators seeing mice on trucks.

What is the transport rate of mice infesting experimental hay bales? Behavioral observations during commercial transport fail to distinguish if mice of a particular sex-age class are more likely to be transported and underestimate the total number transported. Therefore experiments were done to simulate conditions mice face during loading and transport. These experiments determined the proportion of each age-sex class remaining during transport of infested loads. Most mice jumped off the transport vehicle during loading and immediately thereafter. This implies that once a mouse is on a transport vehicle, the vehicle size and transport duration have little influence on whether a mouse will jump off.

In 82 experiments, house mice were hand-caught from swine-feeders or trapped in poultry houses, weighed, marked for individual recognition if later recaptured, visually inspected for pregnancy and lactation, and put in an opaque 210 L metal drum that held a hay bale. The hay bale included a chickenwire frame enclosing oat (*Avena barbata*) hay. The hay was supplemented with swine or steer feed because the bale was reused. No mice were dissected, which means that females scored as nonvisibly pregnant include nonpregnant and early-pregnant mice. Age and breeding status of stowaways influence their potential breeding rate with residents (Endler 1979). The number of mice in a hay bale ranged from 1 to 5 (mode 1; median: 2). Table 2 lists weight class and breeding status of the experimental house mice.

After 1 to 13 days (mode: 2, median: 5), the hay bale was put in a wheelbarrow that was pushed for 15 minutes, after which the hay bale was thoroughly searched for mice. During the 15-minute transport, house mice were classified as jumping from (nonstowaways) or riding with the hay bale (stowaways). An additional 22 experiments were excluded from analyses because all mice were on the floor of the metal drum. These mice could have been on the floor before or during the removal of the hay bale.

RESULTS

A total of 28 stowaways (14 house mice, 14 deermice) were found during commercial transport (Table 1), an underestimation because some mice escape detection. The 14 house mice included 3 handcaught during unloading of grass hay and barley straw from trucks; 1 observed but not caught during unloading of oat hay from a front-end loader; 2 observed but not caught during unloading of pallets of dogfood from a semitruck; 1 handcaught after going through an augur; and 7(1 alive; 6 freshly-dead) taken from a feedmill grain cleaner. Freshly-dead carcasses, which are soft and supple, differ from flattened, dried-out carcasses characteristic of mice that died months before transport. Mice dying just before or during transport are indistinguishable. Flaking (breaking the bales into sections) and a two-year period when bales were at risk of infestation contributed to one observation of a stowaway juvenile house mouse when 2,500 kg (90 bales) of two-year old oat hay were transported by front-end loader. In contrast, 10 adult house mice ran for cover into the remaining stack as the workers threw bales from the stack; and one adult that stayed on a bale immediately jumped out of the front-end loader. The 14 deermice included 2 handcaught during unloading of hay trucks; 2 freshly-dead taken from a feedmill graincleaner; and 10 trapped in a truck cab. The truck was parked in tall weeds, which were probably climbed by these deermice. Adult stowaways (> 10 g) predominated (10 of 12 house mice; all 14 deermice). Of mice whose sex was determined, house mice included 2 males and 3 females; deermice, 1 male and 13 females.

Mice were transported 0.1 and 0.5 km, within the range of mouse dispersal distances, and > 5 km, which is greater than mouse dispersal distances. The minimum distance of grain hauled to a feedmill (5 km) is the radius between the feedmill and the nearest agricultural fields where grains were grown, whereas the maximum distance is unknown. Transport rates for house mice were within one order of magnitude ranging from $7(10^6)$ mice per transported kg grain to $7(10^{n5})$ mice per transported kg hay (Table 1), where the total transported hay and straw was 59,310 kg. Deermice had similar transport rates.

When little is known about the variability of stowaway rates, failures are expected. None of the 369 railroad cars were condemned for rodent infestation, confounding an attempt to determine the proportion of commercial loads infested with mice. No mice were seen during 12 h observing garbage unloading (primarily dead trees, wood products) nor during most of the 10 h observing unloading of hay baled two to three months earlier, which included 256 alfalfa bales (ca. 7,000 kg) and 43 oat hay bales (ca. 1,200 kg). No Teamsters contacted me. The lack of flaking and the brief period when these bales could be infested, probably worked against observing stowaways. Mice may be less attracted to wood products than to garbage.

Most "jumpers" jumped from the bale within the first minute (mode and median: 1 min; range: 1 sec - 13 min). Some mice walked on the outside of the bale, walked on the wheelbarrow, and returned to the cover of the bale. Stowaways, mice remaining in the bale for 15 min, included 64% of all mice, significantly more lower weight classes and more nonvisibly pregnant females (Table 2).

Table 1. Sex¹, species², weight³, and number of live stowaway mice in transported products, distance transported (km), and transport rates (number of transported mice/kg transported product). House mice transport rates varied from 7 (10⁶) mice per transported kg grain to 7 (10⁶) mice per transported kg hay (59,310 kg, total transported hay and straw). Deermice had comparable transport rates.

<u>PRODUCT</u>					<u>RATE</u>	<u>COMMENT</u>
Stowaway Mice					#/kg product	
Sp	Sex	G	No.	Km		
<u>POULTRY FEED</u>						
Md	m	21	1	0.1	-	Handcaught after going unharmed through augur and buried alive
<u>PALLET OF DOG FOOD</u>						
Md	-	a	2	280	-	Seen escaping from pallet holding gnawed-open dog food bags, during unloading of semitruck (Randall Blair pers. comm.)
<u>FEEDMILL GRAIN CLEANER</u>						
Md	np	7	1	≥5	1 live	Handcaught in corn in grain; does recently-dead imply alive during transport? (transport rate includes 1 live and 6 dead)
Md	-	a	6	≥5	6 recently-dead carcasses in 940,313 kg = 7 (10 ⁶) mice/transported kg	
Pm	np	a	2	≥5	2 recently-dead carcasses in 940,313 kg = 2 (10 ⁶) mice/transported kg	In grain
<u>BALES</u>						
Md	np	13	3	13	3/35,000 kg = 8 (10 ⁻⁵) mice/transported kg	Barley straw; handcaught on truck between bales of one truckload (1,800 kg)
Md	np	15				
Md	m	12				2-year old oat hay bale flaked at destination; 10 adults on haystack not transported
Md	-	j	1	0.5	1/2 500 kg = 4 (10 ⁻⁴) mice/transported kg	
Pm	np	14,15	2	16	2 mice/13,610 kg/3y = 1.5 (10 ⁻⁴) mice/transported kg	Handcaught in grass hay on truck between bales of one load (900 kg)
<u>TRUCK CAB</u>						
Pm	p	28	1	52		18-20 May 90
Pm	m	18,22	2			
Pm	np	14,17	4			6-13 Nov 90
		16,16				
Pm	np	16	1			26 Nov 90
Pm	np	14	1			22 Dec 90
Pm	np	13	1			19 Jan 91

¹ m = male, p = pregnant, np = not pregnant

² sp = species, Md = *Mus domesticus*, Pm = *Peromyscus maniculatus*

³ j = ≤10 g, a = >10 g

Table 2. Influence of mouse weight and sex on its jumping from or riding in a haybale that was put in a wheelbarrow and pushed 15 minutes; "unknowns" were omitted from χ^2 (chi-square) analyses. Stowaways, mice remaining in the bale for 15 minutes, included 64% of all mice, significantly more lower weight classes and more nonvisibly pregnant females.

Behavior	Weight (g)						X^2	P<
	4-10	11-15	16-20	21-25	26-35	Unknown		
Jump	4	5	18	5	6	3		
Ride	20	17	15	12	4	6	13.6	0.001

Behavior	Sex				X^2	P<
	Not visibly pregnant	Pregnant or lactating	Male	Unknown		
Jump	4	20	16	1		
Ride	21	21	31	1	7.79	0.025

Thus, mice coming into their maximum breeding potential were most likely to be transported. The probability of a mouse remaining in the bale for 15 mins was not significantly influenced by season, days held in the bale, and number of conspecifics in the bale (data not reported).

DISCUSSION

Stowaway Numbers

Three variates determine the total number of stowaways worldwide: 1) kg of transported goods; 2) % kg of transported goods with mouse infestations; and 3) number of stowaways/kg transported infested loads. The kg of transported goods can be approximated from government indexes. An attempt to document % kg of transported goods with mouse infestations failed when USDA records showed no railroad cars holding hard red winter wheat were condemned for mouse infestations. This study begins to quantify the number of stowaways/kg transported infested load, here 10^4 mice/transported kg grain to 10^5 mice/transported kg hay (Table 1). The main conclusion of this exploratory study is that stowaway house mice have the potential to move rapidly worldwide. Some stowaways breed at their destinations. However, between 1987 and 1988, 5.5 (10^5) kg hay and straw were exported from the USA alone (Table 5 of USDA 1988; see also Anonymous 1986). Multiplying these rates by the huge volume of transported products worldwide implies thousands of stowaway house mice. As more than 50 % of experimental stowaways remained during transport of infested loads (Table 2), some mice are likely to arrive in infested loads. Two reasons to conclude that stowaways breed with residents at their destination follow (Baker, submitted). Stowaways that subsequently bred with residents could cause the putatively identical genetic markers shared by mouse populations in geographically-

isolated cities connected by transport. The genetic impact of stowaways was documented in five successful experimental introductions of genetic markers into populations lacking these markers (Anderson et al. 1964; Baker 1981; Bennett 1978; Berry et al. 1991).

Request For Cooperators In Commerce

The most convincing stowaway data are live mice taken off transport vehicles, but these are the hardest to obtain without cooperators in the transport industry. Interested cooperators in the transport industry are sought. Variability in transport rates will be high because many variates influence the process, such as availability of nearby sources of house mice; storage duration when the product is at risk of infestation; food value of transported product; and method of moving the product onto a vehicle. For example, fewer stowaways are likely to remain when bales are thrown onto a front end loader than when bales rest on a pallet that is forklifted onto a truck. Studies of mice carrying unique genetic markers from location of origin to destination may be logistically simpler and can provide estimates of the genetic impact of stowaways.

Pest Control

This work has implications for pest control. Though the number of stowaways per kg product is low, the huge volume of transported products implies thousands of stowaways worldwide. Stowaways genetically resistant to poisons (Lund 1985; Jackson et al. 1985; Misenheimer et al. 1994) may start infestations that are difficult to control. Knowing that stowaways could fall into products will confound lawyers' attempts to dismiss million dollar lawsuits brought by plaintiffs finding dead mice in manufactured goods. However, location-specific genetic markers are a potentially powerful forensics tool.

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