



## Playful handling as social enrichment for individually- and group-housed laboratory rats

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### ABSTRACT

Social housing is recommended for laboratory rats because they are highly social mammals but research constraints or medical issues often demand individual housing and, when social housing is practiced, it typically involves housing with only one or two conspecifics. We hypothesized that playful social contact with humans (i.e. tickling), mimicking the dorsal contacts and pins characteristic of rat rough-and-tumble play, could act as a partial substitute for, or supplement to, conspecific social contact in situations when laboratory rats are housed individually or in pairs or triplets. Furthermore, we hypothesized that the beneficial effects of regular tickling when young would persist following discontinuation of tickling. Accordingly, we investigated the responses of juvenile male rats to handling conditions (minimally handled vs. tickled) and group size (singletons, pairs, triplets). We measured (a) production of 22- and 50-kHz ultrasonic vocalizations (USVs) during a 1-min period before handling (interpreted as evidence for negative and positive affective states, respectively), (b) corticosterone levels from faecal pellets collected in the home cage, and pre- and post-treatment body weight (as measures of physiological stress), and (c) behaviour in an Open Field test (to assess anxiety). After 3 weeks of tickling for 2 min/day, individually-housed rats produced more 50-kHz USVs in anticipation of handling than their minimally-handled counterparts ( $P < 0.0001$ ). This effect persisted for at least 4 weeks after discontinuation of the tickling programme ( $P < 0.0001$ ), when all rats were transferred to individual housing. Tickling experience also reduced anxiety-related behaviour of individually-housed rats in the Open Field test ( $P < 0.05$ ). Faecal corticosterone levels and body weight were not affected by tickling experience or group size, although both corticosterone levels and anxiety-related behaviour were elevated following re-housing ( $P < 0.05$ ). No significant differences were detected between rats housed as pairs or triplets for any of the measures investigated ( $P > 0.05$ ). Both the Open Field and USV data affirm that tickling is beneficial for rats, especially when housed individually, and provide evidence that specific experiences derived from playful contact contribute to the well-being of group-housed animals. We conclude that tickling is an appropriate cross-species social enrichment manoeuvre that can promote the well-being of laboratory rats and rats kept as companion animals.

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### 1. Introduction

Laboratory rats are highly social mammals, showing strong motivation to retain social contact with conspecifics (Hurst et al., 1997, 1998), and preference for social contact over larger or physically enriched cages

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(Patterson-Kane et al., 1999, 2001, 2002). Nevertheless, even though social housing is recommended as a standard practice for laboratory rats (e.g. National Research Council, 2011), individual housing may be used due to research constraints (e.g. food/fluid intake monitoring) or medical issues. Yet, individual housing can increase physiological stress responses to common procedures such as cage cleaning and injections, alter social skills in adulthood, and affect anxiety-related behaviour and activity in behavioural tests (Duke et al., 2001; Krohn et al., 2006; Olsson and Westlund, 2007), suggesting that it compromises rat welfare and could reduce the external and face validity of data gained from these animals (Sherwin, 2004). In contrast, in their review, Krohn et al. (2006) propose that individual housing may not be a major biasing variable, and that adverse effects on rat welfare could be reduced, and possibly even eliminated, by the provision of appropriate periodic social enrichment.

Pair housing is the most common form of social housing used for laboratory rats after weaning, considering the need for replication of experimental treatments, limited space availability, and cost constraints. However, pair-housed rats have been reported to show intermediate responses relative to individually-housed rats and those housed in groups of three or more in behavioural tests of anxiety including the Elevated Plus maze and Open Field tests (Botelho et al., 2007; Patterson-Kane et al., 1999), and the Hebb–Williams maze test assessing memory (Patterson-Kane et al., 1999). They were also reported to have heart rate and mean arterial blood pressure more similar to individually-housed rats than those housed in groups of four (Sharp et al., 2002). Furthermore, rats in one study preferred to spend time in groups of three, four and five animals rather than in pairs or alone (Talling et al., 2002), and another study showed group size preference to peak at 5.9 animals (Patterson-Kane et al., 2004). Consequently, housing in pairs may not be ideal as the social housing standard against which effects of individual housing are compared.

Human caretakers can become integral members of the social environment of laboratory rats as evidenced by similarities in the responses of rats to opportunities for positive interaction with humans and conspecifics (Panksepp and Burgdorf, 2000; Sloan and Latané, 1974; Werner and Latané, 1974), and rat discrimination between familiar and unfamiliar humans (Davis et al., 1997). Positive contact with humans could, therefore, provide a useful means of attenuating adverse effects of social isolation from conspecifics. Handling combining rubbing, petting, stroking and scratching was shown to serve as a partial social surrogate for cage mates in one study (Sloan and Latané, 1974). However, other studies have indicated aversive responses, or absence of effects, when some of these handling techniques were applied individually (e.g. Brudzynski and Ociepa, 1992; Burgdorf and Panksepp, 2001; Cloutier and Newberry, 2008; Werner and Anderson, 1976).

In contrast, playful handling appears to serve as a robustly beneficial form of social contact, especially for young rats. Human handlers can mimic the playful rough-and-tumble behaviour of rats by alternating between tickling the rat's nape (dorsal contact) and ventral surface (pinning) using vigorous, rapid finger movements. This

form of interaction (tickling) induces a positive motivational state in young rats, reduces fear of humans, and is actively solicited by the animals (Burgdorf and Panksepp, 2001; Cloutier et al., 2012; Panksepp and Burgdorf, 2000). Tickling serves as a motivator for learning new operant responses, providing further evidence that tickling is a substantial reward for young rats (Burgdorf and Panksepp, 2001). Tickling by caretakers could, therefore, be useful in a research setting by serving as a form of social enrichment for individually-housed juvenile rats. It could also have carry-over benefits, which would be useful if it was anticipated that the rats would have to spend some time in individual housing in the future.

We hypothesized that playful social contact with humans (i.e., tickling) could act as a partial substitute for, or beneficial supplement to, conspecific social contact, and that beneficial effects of regular tickling when young would persist following discontinuation of tickling. We investigated this hypothesis by comparing responses of rats housed individually, in pairs or in triplets to one of two different methods of handling, namely tickling during a daily 2-min handling period or minimal handling only, which occurred once weekly when all rats were transferred to clean cages. Rats were exposed to the handling and group size treatments during a 4-week period, re-housed individually, and exposed to minimal handling for another 4-week period. Responses were assessed at the start and end of each 4-week period. We predicted that tickling would reduce fear of humans compared with minimal handling, with this effect being stronger for individually- than group-housed rats. The latter effect was predicted on the basis that individually-housed rats are deprived of conspecific play interactions that may be partially compensated for by human tickling, and that they value tickling more than group-housed rats (Panksepp and Burgdorf, 2000). We also anticipated that tickling and social housing would increase body weight and lower corticosterone levels (indicating reduced physiological stress), and dampen anxiety-related responses in an Open Field test. Alternatively, if separating group-housed rats for behavioural testing is stressful (Hall, 1998), then we expected to observe elevated anxiety-related behaviour when testing group-housed rats. We also expected the effects of tickling to be maintained after the end of the tickling period, when rats were temporarily moved from social to individual housing.

## 2. Methods

### 2.1. Animals and husbandry

We obtained 72 male Sprague-Dawley rats from Simonson Laboratory (Gilroy, CA, USA), where they were housed in litters with their dam until weaning and shipping at 21 days of age. We focused on males because they are more commonly used in biomedical research than females (OECD Guideline 486, 1997). On arrival, we housed them in standard clear plastic cages (46 cm *L* × 24 cm *W* × 20 cm *H*) with wood fibre bedding (Biofresh Comfort Bedding Natural Soft Cellulose, Absorption Corp., Ferndale, WA, USA), a paper hut (Shepherd Specialty Paper, Chicago, IL, USA), and a metal lid. We kept them individually, in pairs or in

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