



## Workaholic ferrets: Does a two-chamber consumer demand study give insight in the preferences of laboratory ferrets (*Mustela putorius furo*)?



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### ABSTRACT

Although provision of environmental enrichment is an effective tool to refine laboratory animal experiments, it is currently unknown which enrichments ferrets prefer. This study aimed to assess the suitability of a closed economy, two-chamber consumer demand set-up to determine ferrets' preferences for selected enrichments. Twelve female ferrets were housed in a set-up consisting of a home and enrichment chamber (EC) connected by a weighted door. The maximum weights the ferrets pushed for food ( $MPP_{\text{food}}$ ) and an empty chamber ( $MPP_{\text{empty}}$ ) were determined to evaluate the maximum push capacity of the animals and as a control. Although the ferrets pushed significantly more for food ( $1325 \pm 213$  g) than for the empty chamber ( $1169 \pm 193$  g), the weight difference was minor ( $MPP_{\text{empty}}$  was  $89 \pm 13\%$  of  $MPP_{\text{food}}$ ). To evaluate the ferrets' underlying motivation to push for the empty chamber, a second study was performed in which  $MPP_{\text{empty}}$  was tested in seven alternative set-ups. The first three set-ups included adapted versions of the standard design (set-up A<sub>1</sub>, A<sub>2</sub> and A<sub>3</sub>), intended to determine the functional value of the empty chamber. The four other set-ups (set-up B<sub>0</sub>, B<sub>1</sub>, B<sub>3</sub>, B<sub>4</sub>) aimed to evaluate the attractiveness of the door elements by allowing the ferrets to choose whether or not to use the weighted door to enter EC. Results demonstrated no significant differences in  $MPP_{\text{empty}}$  between the A-set-ups, indicating that the value of the empty chamber could not be reduced by adapting the set-up.  $MPP_{\text{empty}}$  reduced when allowing the ferrets free access to EC, demonstrating that the empty chamber had reinforcing properties. Nevertheless, the ferrets were still motivated to use the weighted door despite being granted free access to EC, indicating that the door also has reinforcing properties. The ferrets decreased the use of the weighted door most when, in a set-up with free access to EC, the nest box in the home cage ( $53 \pm 22\%$  of  $MPP_{\text{food}}$ ) was replaced by a manipulable plastic bucket ( $26 \pm 13\%$  of  $MPP_{\text{food}}$ ). These results indicate that availability of items in the home chamber may influence the results, which should be taken into account when designing motivation studies similar to the one performed in this study. The lack of differences between  $MPP_{\text{food}}$  and  $MPP_{\text{empty}}$  furthermore demonstrates that the two-chamber set-up is not suitable for evaluating the ferrets' motivation for enrichments, thus necessitating other alternatives, such as a three- or multi-chamber consumer demand study, to be explored.

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### 1. Introduction

Ferrets (*Mustela putorius furo*) are commonly used for research purposes (e.g. influenza research) (Boyce et al., 2001). To ensure humane use of laboratory animals, the principles of reduction, replacement and refinement (the three R's) are employed (Russell et al., 1959). Refinement includes the optimization of the animal's housing conditions, e.g. by providing environmental enrichment

(Russell et al., 1959). Environmental enrichment can improve animal welfare at least to a certain degree by providing some of the appropriate stimuli needed to perform species-specific behaviour (Newberry, 1995).

Traditionally, the preference for alternative resources is assessed in preference tests, in which the animal is given a two-way unweighted choice between alternative resources. One preference test with ferrets indicated that these animals prefer an enriched over a barren cage, but did not specify which enrichments are preferred (Cruden, 2011). Unfortunately, these preference tests only address the relative rather than the actual value of the enrichment (Kirkden and Pajor, 2006). In other words, they assess whether animals 'like' (affective consequence) a specific enrichment item and not whether they 'want' (are motivated to access) this item (Berridge and Robinson, 2003).

To address the actual value of a specific enrichment item, so-called consumer demand studies may be used. Such studies have been used extensively in various laboratory and production animals, including mice (Sherwin, 1996), rats (Manser et al., 1996, 1998; Patterson-Kane et al., 2002), silver foxes (Hovland et al., 2006) and mink (Mason et al., 2001). A consumer demand study comprises a set-up in which increasing costs are imposed on the animal in order for it to gain access to a specific resource.

The value of the enrichment can be expressed using various indices, including the price elasticity of demand index, the consumer surplus index and the maximum price paid index (Kirkden et al., 2003). The maximum price paid (MPP) index indicates the 'breakpoint' at which the animal is no longer willing to pay the price for the resource. This index has several advantages over the other indices, including a) its relative insensitivity to external cues (Warburton and Mason, 2003); b) its usefulness for the testing of 'all-or-none' goods (Olsson et al., 2002); and c) the possibility to use qualitative rather than quantitative increases in price, thereby omitting the need to make assumptions about subjective value of a task (Cooper, 2004). As a result, the MPP index is generally preferred for analyzing the value of resources.

To infer information on the actual value of a resource, MPP is tested consecutively for various resources, including food (MPP<sub>food</sub>, which is considered to reflect the maximum amount of weight an animal is able to push: the maximum push capacity), an empty chamber (MPP<sub>empty</sub>, which serves as a control) and the different enrichment items (e.g. Asher et al., 2009). Most commonly, testing takes place in a closed economy two-chamber set-up, i.e. the animals are housed permanently in the experimental set-up. This prevents the animals from becoming less motivated to work for a resource because of its (free and/or unlimited) access to the item outside of the experimental set-up (Jensen and Pedersen, 2008; Ladewig et al., 2002). Animals have also been tested in three- or multiple-chamber designs, in which they can simultaneously gain access to one or multiple resources and a control (e.g. Hovland et al., 2006; Mason et al., 2001; Seaman et al., 2008). These set-ups, however, introduce an extra variable, as the animal only has limited income (i.e. the time and energy available per day) which it then needs to divide between the different resources, thereby potentially yielding lower MPP values for resources that are less important.

As no consumer demand studies have been performed in ferrets thus far, the current study focused on establishing the functionality of a two-chamber consumer demand study in ferrets using the MPP index. Similar to the consumer demand study with mink (Mason et al., 2001), a weighted door was used. To be considered suitable for testing the ferrets' motivation for resources, the experimental set-up needs to meet three prerequisites: (1) MPP<sub>food</sub> should reflect the maximum push capacity (MPC); (2) the task should be perceived as strenuous and (3) MPP<sub>empty</sub> should be low and sufficiently distinctive from MPP<sub>food</sub>. To assess whether these prerequisites were

met, MPP<sub>food</sub>, MPP<sub>empty</sub> and duration and number of visits to the chamber with food and to an empty chamber were measured. As ferrets were found to push excessively for an empty room (i.e. the third prerequisite was not met), two subsequent studies were performed to assess how and which features of the design may have affected MPP<sub>empty</sub>.

## 2. Animals, materials and methods

### 2.1. Ethical approval

This study was ethically approved by the Institutional Animal Care and Use Committee of Intravacc (DEC 201300057) and Utrecht University (DEC 2013.1.09.073).

### 2.2. Animals

For study 1, 12 female neutered ferrets from Schimmel B.V. were used that weighed  $1.0 \pm 0.2$  kg (665–1145 g). Six ferrets were 4 years old and chemically neutered using a hormonal implant (Suprelorin<sup>®</sup>, Virbac, The Netherlands); the other six ferrets were 5 months old and surgically neutered (ovariectomized). For study 2, five of the 4-year-old female ferrets from study 1 were used. These ferrets weighed  $838 \pm 113$  g (665–938 g).

### 2.3. Housing and nutrition

Ferrets were housed indoors in a room that was kept at a temperature between 18 °C and 22 °C. They were exposed to a 8:16 h light:dark schedule using artificial lighting (light bulbs) that switched on at 9:00 h and off at 17:00 h. In addition, auditory stimulation was available in the form of a radio, which automatically switched on and off concurrent with the light phase. The ferrets were provided water and food (Hill's M/D<sup>®</sup> for the 4-year old ferrets, Hope Farms<sup>®</sup> ferret balance pellets for 5-month-old ferrets) ad libitum. Refreshing of the food and water, as well as cleaning of the cages took place daily at 10:00 h. The ferrets' health and overall condition were monitored prior to and throughout the study.

### 2.4. Experimental housing

Throughout the experiments, the ferrets were individually housed in a closed economy two-chamber set-up consisting of a phenolic faced plywood floor pen with solid floors and walls that measured 1.6 m<sup>2</sup> (Fig. 1a). The pens were divided in two equal spaces by means of a 70 cm high, 6 mm thick phenolic faced plywood divider equipped with a non-transparent one-way cat flap and a one-way horizontal swinging weighted door (Tecnilab-BMI, Someren, The Netherlands; Fig. 1b). Under the door, a wire mesh strip was mounted to provide a traction surface for the ferrets to facilitate them to apply force to the door. The door allowed the ferrets to move from a home chamber (HC) equipped with a resting area (i.e. nest box or plastic sleeping bucket), food (except when testing MPP<sub>food</sub>) and water (provided via a nipple) to the enrichment chamber (EC) where the resource to be tested (e.g. food, enrichment) was placed. The one-way unweighted cat flap could subsequently be used by the ferrets to return to HC.

### 2.5. Task

Similar to mink (Cooper and Mason, 2001), ferrets had to push a weighted door, which is considered as a naturalistic task for ferrets that requires little training and is less prone to operant-reinforcer biases than unnatural tasks (Dawkins, 1990). To open the unweighted door, ferrets needed to exert a force of 200 g. Similar to mink, the force needed to push open the door was gradually

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