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Food neophobia in wild and laboratory rats (multi-strain comparison)

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ABSTRACT

Although empirical studies comparing neophobia in wild and laboratory rats have been conducted in the past, a few decades have passed since most of them were completed. This is a substantial period of time in the case of fast-breeding animals such as rats. Equally important are the inconsistencies in research findings with respect to comparisons between wild and laboratory rats, and within domesticated strains. As well as having the aim of updating knowledge of neophobia among different types of rats, the present experiment was also an attempt to isolate a specific fear of a new food from a general fear of a novel object.

The procedure was that rats accustomed to one type of food served in a specific location and in a familiar container were given a different type of food. Test trials were preceded by food deprivation. The following variables were measured: feeding latency, the pace of eating, the number of approaches to the container, and the number of times food was sampled in each trial. The amount of food consumed in each trial was weighed and also taken into account. Grooming time served as the measure of stress among the rats in the experiment.

The results of the experiment did not confirm the assertion of some authors that wild rats avoid eating unfamiliar foods. All groups demonstrated only a temporary decrease in the amount of food consumed, the magnitude of which was similar in all strains. No evidence of particularly low neophobia in albino rats was found. However, the behavioral symptoms indicated higher levels of stress in wild rats compared to the other groups.

1. Introduction

Caution is a natural response of rats to unfamiliar objects (Hebb, 1946). This is also the case when they are confronted with physical changes to the environment and variations in feeding routines (Barnett, 1963, 2009). It is claimed that many rats will not eat unfamiliar food. This strategy may persist for several days at a time, and may be more pronounced in an unfamiliar environment (Burritt and Provenza, 1997; Chapple et al., 1987; Cheney and Miller, 1997). When encountering a novel food, an animal does not know whether it is edible or not. It has to suppress its initial neophobia, and then evaluate the consequences of eating the new food (Barnett, 1963, 2009). The term coined for the avoidance of unfamiliar food is food neophobia (Barnett, 1958, 1963, 2009). It is present in many species (Addessi et al., 2004; Bryan, 1987; Inglis et al., 1996; Kronenberger and Médioni, 1985; Launchbaugh et al., 1997), including rats (Barnett, 1963, 2009; Caroll et al., 1975; Cowan, 1977; Inglis et al., 1996; Mitchell, 1976). The need to distinguish between edible and inedible food is particularly relevant to omnivores, which face the so-called generalist's dilemma (Rozin, 2000).

Rats respond to changes of the location where food appears, changes of the container in which the food is provided, as well as changes of the feed itself (Barnett, 1963, 2009; Caroll et al., 1975; Cowan, 1977; Inglis et al., 1996; Mitchell, 1976). Their response to these novel conditions results from the interaction of behaviors motivated by curiosity about the novel object's potential value and fear of its possible toxicity. This behavior is typified by initial avoidance of a new food, followed by gradual sampling in regular time intervals (Barnett, 1963, 2009). If the new food does not become associated with adverse body symptoms, its intake increases (Barnett, 1963, 2009). The hungrier the rat, the quicker it starts to eat unfamiliar food (Barnett, 1963, 2009). Rats develop an aversion to foods which cause adverse effects within a couple to a dozen hours (Hankins et al., 1973; Revusky and Bedarf, 1967). It has been hypothesized that rats display caution when first encountering new food because they have no gag reflex and, as a result, are unable to eliminate toxic substances from the stomach (Barnett, 1956). It is often claimed that food neophobia is an innate phenomenon (Moron and Gallo, 2007) and that it persists in genetically

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wild rats, even those reared in a laboratory (Barnett, 1958; Galef and Whiskin, 2003). However, some researchers have suggested that food neophobia may be primarily a response learned in the process of socialization (Barnett, 1956, 2005; Taylor and Thomas, 1989).

The container in which new food is served appears to play an important part. Some researchers have asserted that rats prefer food served in a familiar container (Mitchell et al., 2005) and that fear of a new container is significantly stronger than food neophobia itself (Inglis et al., 1996).

There is a lot of empirical data on anatomical and behavioral differences between wild and laboratory rats (Barnett et al., 1979; Blanchard et al., 1986; Huck and Price, 1975; Himmler et al., 2013, 2015; Lockard, 1968; Price, 1999; Stryjek et al., 2012a,b, 2013). It has also been suggested that laboratory rats are characterized by lower neophobia than their wild counterparts (Barnett, 1958; Calhoun, 1963; Cowan, 1977; Mitchell, 1976; Tanaś and Pisula, 2011). The underlying cause for the development of this trait may be the lack of predatory pressure in laboratory conditions, combined with low environment variability. The absence of natural pressures may have significantly diminished the natural constraints of stimulus-seeking behavior in laboratory rats, an activity which plays an important role in adapting to a natural environment (Pisula, 2007). In addition, the changeability of wild rat's habitats may have led to the development of avoidance responses of varying intensity towards a number of environmental changes. Some researchers have suggested that attempts by humans to eradicate rats in their environment may have contributed to the development of food neophobia (Barnett, 1956, 2005; Inglis et al., 1996; Taylor and Thomas, 1989). Rat species not subjected to population suppression through the use of rat poison due to their human-independent diet do not demonstrate neophobic responses to novel foods (Barnett, 1956, 2005; Cowan, 1977; Inglis et al., 1996). Furthermore, food neophobia seems to be absent in Norway rats inhabiting landfills, given their constantly changing environment and the ubiquity of novelty (Barnett, 1963, 2009; Boice, 1971). Absence of food neophobia has also been reported in a group of *Rattus norvegicus* which lived for over a century on an island isolated from human activity (Taylor and Thomas, 1989). Another possibility is that the ancestors of laboratory rats were captured precisely because they exhibited lower neophobia (Mitchell, 1976). These rats may have been less cautious when they encountered bait placed in the trap.

Even though several studies comparing neophobia levels in wild and laboratory rats were conducted in the past, many date back to the 1950s (Barnett, 1958; Cowan, 1977; Calhoun, 1963; Mitchell, 1976). During the long period of laboratory breeding since that time, more changes may have developed in the behavior of laboratory rats. There are also significant inconsistencies in conclusions drawn from comparisons of different lines. The frequently referenced paper by (Barnett, 1958) claimed that laboratory rats demonstrated no food neophobia, in contrast to highly food-neophobic wild rats. Other researchers have suggested that both wild and laboratory rats are food-neophobic (Mitchell et al., 1973), the only difference being that in wild rats food neophobia is stronger (Mitchell, 1976). Mitchell suggested that these differences may have resulted from distinct causes of behavior. He claims that wild rats are afraid of a novel food, while laboratory rats are curious about it. This statement is in accordance with our earlier findings (Pisula et al., 2012), which showed a clear positive response towards novel objects in laboratory rats, but not in wild subjects. This categorization, however, is not consistent with the conclusions of other researchers (Rozin, 2000), who have pointed out that omnivores demonstrate both tendencies at the same time. A more plausible explanation is that fear, as a response to novel food, dominates in wild rats that have adapted to a threatening environment. Furthermore, (Mitchell, 1976) claimed that wild rats were much more averse to eating from an unfamiliar food container than hooded laboratory rats, with albino rats demonstrating the lowest aversion. In his studies, all strains exhibited increased feeding latency when an unfamiliar food container was introduced, but only albino rats showed a decrease in food intake. Still, all rats initially demonstrated aversion to the new container, which means that differences are in the intensity of neophobia rather than in its presence or absence.

It therefore seems necessary to conduct further experiments aimed at systematising knowledge on food neophobia. It is particularly important to control the potential effect of a novel container introduced with novel food, to test multiple strains of laboratory rats (both pigmented and albino strains), and to update the information obtained in studies conducted many decades ago. Furthermore, the experimental procedure described below was designed to reduce the amount of stress experienced by tested animals through shorter testing times and limited length of food deprivation. The novelty of the testing environment was reduced by conducting the experiment in a cage as identical as possible to the ones the rats lived in. Additionally, simple and non-invasive tests associated with the observation of food neophobia in animals may measure their levels of fear. This would be particularly useful in the context of growing interest in emotional processes in animals.

2. Methods

2.1. Ethics statement

All procedures described in this paper were approved by the 4th Local Ethics Commission on Animal Experimentation, Warsaw, Poland. All rats prior to the experiment were cared for in accordance with the Regulation of the Polish Minister of Agriculture and Rural Development of 10 March 2006 on laboratory animal care.

2.2. Animals

The sample consisted of 51 adult rats *Rattus norvegicus*. Experimental groups included 12 Long Evans laboratory rats (6 females and 6 males), 14 Brown Norway laboratory rats (6 females and 8 males), 14 Sprague-Dawley laboratory rats (7 females and 7 males) and 11 wild WWCPS rats (6 females and 5 males).

The WWCPS (Warsaw Wild Captive Pisula Stryjek) rats were derived in 2006 from a sample originating from 5 independent colonies of feral rats (Stryjek and Pisula, 2008). The experiment used the third generation (F3) of laboratory-reared WWCPS wild rats. In order to prevent the development of domestication features in the breeding colony, we systematically include wild rats (freshly caught in a variety of locations) in the breeding scheme. As a result, we have fourth generation laboratory-reared animals at most.

The Brown Norway and Sprague-Dawley rats were sourced from the Mossakowski Medical Research Centre at the Polish Academy of Sciences, while the Wistar rats were taken from the Experimental Medicine Centre at the Medical University of Bialystok, Poland.

Prior to the experiment all rats were housed in groups of 3–5 in Eurostandard type IV cages with ad libitum access to water and standard laboratory fodder. The day/night cycle was set at 12/12 h.

Prior to testing, the rats were weighed and the results were recorded in grams. Females were lighter than males in three lines (WWCPS – $M_f = 214(SD = 23.3)$, $M_m = 272(SD = 20.5)$, $t(9) = 4.308$, $p < 0.01$; Sprague-Dawley – $M_f = 292(SD = 9.5)$, $M_m = 370(SD = 21.7)$, $t(12) = 8.679$, $p < 0.001$; Brown Norway – $M_f = 190(SD = 36.3)$, $M_m = 382(SD = 25.3)$, $t(12) = 11.692$, $p < 0.001$). There were no sex differences in the weights of the Long Evans rats ($M_f = 280(SD = 24.12)$, $M_m = 296(SD = 14.2)$, $p > 0.05$).