Individual Differences in Behaviour and Cognition of Eurasian Harvest Mice

Dissertation
der Mathematisch-Naturwissenschaftlichen Fakultät
der Eberhard Karls Universität Tübingen
zur Erlangung des Grades eines
Doktors der Naturwissenschaften
(Dr. rer. nat.)

vorgelegt von
Andrea Claudia Schuster
aus Gräfelfing

Tübingen
2016

Gedruckt mit Genehmigung der Mathematisch-Naturwissenschaftlichen Fakultät der Eberhard Karls Universität Tübingen.

Tag der mündlichen Qualifikation:	07.04.2017
Dekan:	Prof. Dr. Wolfgang Rosenstiel
1. Berichterstatter:	Prof. Dr. Katharina Foerster
2. Berichterstatter:	Prof. Dr. Jana Eccard

Eidesstattliche Erklärung & Eigenanteil

Hiermit erkläre ich an Eides statt, dass ich die folgende Doktorarbeit selbst verfasst habe und sie – abgesehen von der Beratung und Hilfe durch meine Betreuerin – nach Inhalt und Form meine eigene Arbeit darstellt. Andere als die angegebenen Quellen und Hilfsmittel habe ich nicht benutzt und die den benutzten Quellen wörtlich oder inhaltlich entnommenen Stellen habe ich als solche kenntlich gemacht.

Die Verhaltens-Experimente in Kapitel 1 wurden z.T. mit Hilfe von Teresa Carl durchgeführt, die für ihre Bachelorarbeit Verhaltensbeobachtungen an den Zwergmäusen durchführte.

Die Verhaltens-Experimente in Kapitel 2 wurden z.T. mit Hilfe von Uwe Zimmermann und Carina Hauer durchgeführt, die für ihre Bachelorarbeiten jeweils Verhaltensbeobachtungen an den Zwergmäusen durchführten. Zudem hatten ich Unterstützung von studentischen Hilfskräften.

Beim Fang der Zwergmäuse im Freilandgehege hatte ich zeitweise Unterstützung von studentischen Hilfskräften. Die Verhaltens-Experimente in Kapitel 3 wurden z.T. mit Hilfe von Annika Luczak durchgeführt, die für ihre Bachelorarbeit Verhaltensbeobachtungen an den Zwergmäusen durchführte.

Diese Doktorarbeit wurde an keiner anderen Prüfungsbehörde vorgelegt.

Schondorf am Ammersee, 7. Mai 2017

Andrea Schuster

für Opa

Summary

Behavioural traits were assumed to be the most flexible traits in animals (Briffa et al. 2008). However, there is now diverse evidence for constant between-individual behavioural differences in many species, i.e. animal personality (Réale et al. 2007), resulting in less flexible individual behaviour (Evans et al. 2010). Knowledge on animal personality has provided new insights into evolutionary biology and animal ecology, as behavioural types - individuals with a specific combination of two or more behavioural trait levels (Bell 2007) - have been shown to affect fitness (e.g., Dingemanse et al. 2004). Further, those constant behavioural traits (personality traits) form behavioural syndromes, which are characterised by correlations between different behaviours (Sih et al. 2004a). Mechanisms maintaining these correlations could be constrained due to underlying relationships with cognitive traits. There is growing evidence for the non-independence of animal personality and general cognitive abilities, but so far, studies on the direction of the relationship between them revealed contradictory results. Still, it is hypothesised that individuals may exhibit consistent learning and decision styles (Sih and Del Giudice 2012). Within a cognitive syndrome fast behavioural types - consistently bolder and more active individuals - are expected to show also faster learning styles. Slow behavioural types in contrast are assumed to learn slower but more accurately (Sih and Del Giudice 2012). This can be caused by a speed-accuracy trade-off that individuals face during decision making (Chittka et al. 2009). Although, behavioural syndromes were assumed to be stable across time and space, there is growing evidence that these correlations between behaviours appear more flexible (Fischer et al. 2016). Phenotypic correlations can vary between populations of the same species originated from different environments, for instance, laboratory and field populations.

In this thesis, I aim to provide evidence for animal personality in Eurasian harvest mice (*Micromys minutus*). Further, I investigate whether behavioural traits form a behavioural syndrome and how spatial cognitive traits are related to personality traits (cognitive syndrome). Harvest mice have not been in the focus of animal personality research yet. However, this small rodent species is likely to be highly suitable for animal personality and especially spatial cognitive syndrome studies for the following reasons: (I) harvest mice are easy to keep and breed in the laboratory, (II) behavioural and cognitive tests can easily be established based on tasks done with laboratory mice and rats, (III) harvest mice are assumed to be selected for pronounced spatial cognitive traits due to their adaptation to three-dimensional habitat structures (Piechocki 2001), and (IV) harvest mice are a widespread species in the Palaearctic but locally endangered (Darinot 2016), so that more knowledge about their behavioural ecology might provide helpful insights for their conservation.

We tested the assumptions for animal personality and found that exploration, activity and boldness were repeatable in juvenile and adult harvest mice. Spatial recognition was repeatable in adults, as well as spatial learning ability and decision speed. We found no repeatability of decision accuracy. Exploration, activity and boldness were consistent before and after maturation, as well as before and after first sexual contact. Data on spatial recognition provided little evidence for consistency. Harvest mice showed a behavioural syndrome as we observed strong positive correlations between personality traits. The speed-accuracy trade-off was not apparent. Nevertheless, we found weak evidence for a cognitive syndrome as fast behavioural types learned faster than slow types, and shyer harvest mice made decisions quicker than bolder mice. All those experiments were dome in a laboratory population of harvest mice. We compared these results with personality traits and behavioural correlations in harvest mice with a natural genetic background, kept in a semi-natural outdoor enclosure. Exploration, activity and boldness were also repeatable in the field population. However, spatial recognition was not repeatable. Harvest mice of the field population showed also a behavioural syndrome as we observed positive between-individual correlations among personality traits.

With these results, we provide evidence for animal personality in Eurasian harvest mice. In sum, we conclude that harvest mice express repeatable behavioural traits in the laboratory and the field population. Exploration, activity and boldness were also consistent between life-history stages. Although we observed significant differences in the magnitude of phenotypic correlations between the laboratory and the field population, both datasets suggested the same directions of correlations within the syndrome. Hence, we conclude that Eurasian harvest mice in general exhibit a behavioural syndrome along a fast-slow continuum, and that proximate factors are more likely to shape this behavioural syndrome than selection processes. Spatial recognition showed low repeatability and was less consistent across life history stages. We found repeatability of spatial learning ability and decision speed, but not of decision accuracy. Given the rising interest in individual variation in cognitive performance, and in its relationship to animal personality, we suggest that it is important to gather more data on the repeatability and consistency of cognitive traits. The observed relationships between spatial cognitive and personality traits partly support the hypotheses of a cognitive syndrome in harvest mice and will hopefully stimulate more studies on the relationship between (spatial) cognition and personality in animals.

Zusammenfassung

Lange Zeit wurde angenommen, dass Verhaltensmuster die flexibelsten Merkmale von Tieren sind (Briffa et al. 2008). Mittlerweile gibt es allerdings viele Beweise für konstante Verhaltensunterschiede zwischen Individuen einer Population, welche als Tierpersönlichkeit definiert wurde (*animal personality*, Réale et al. 2007). Aus diesen konstanten Verhaltensmustern resultiert, dass Tiere in unterschiedlichen Situationen weniger flexibles Verhalten zeigen (Evans et al. 2010). Das Wissen über Tierpersönlichkeiten ermöglichte neue Einblicke in Bereiche der Evolutionsbiologie und Tierökologie, da unterschiedliche Verhaltenstypen - Individuen mit einer bestimmten Kombination aus mehreren spezifischen Verhaltensweisen, *behavioural types* (Bell 2007) - einen Einfluss auf deren Fitness haben können (z.B. Dingemanse et al. 2004). Konstante Verhaltensweisen (Persönlichkeitsmerkmale) können außerdem sogenannte Verhaltenssyndrome (*behavioural syndromes*) bilden, welche durch Korrelationen zwischen den verschiedenen Verhaltensweisen charakterisiert sind (Sih et al. 2004a). Solche Korrelationen können durch Mechanismen organisiert sein, welche die Beziehungen zwischen Verhaltensweisen und kognitiven Fähigkeiten erzwingen. Es gibt immer mehr Hinweise für einen abhängigen Zusammenhang zwischen Tierpersönlichkeit und kognitiven Fähigkeiten, jedoch zeigen Studien bis jetzt entgegengesetzte Ergebnisse. Unabhängig davon gibt es Hypothesen, die individuell konstante Lern- und Entscheidungsstile vorhersagen (Sih and Del Giudice 2012). Innerhalb eines solchen kognitiven Syndroms (*cognitive syndrome*) wird erwartet, dass schnelle Verhaltenstypen - konstant mutigere und aktivere Tiere - auch schnellere Lernstile haben. Für langsame Verhaltenstypen hingegen wird ein langsameres, jedoch genaueres Lernverhalten erwartet (Sih and Del Giudice 2012). Dies beruht auf der Annahme, dass Individuen während der Entscheidungsfindung einen Kompromiss zwischen der Geschwindigkeit und der Genauigkeit eingehen müssen (*speed-accuracy trade-off*, Chittka et al. 2009). Auf Grund der durch kognitive, physiologische oder genetische Faktoren erzwungenen Zusammenhänge zwischen Verhaltensweisen wurde angenommen, dass Verhaltenssyndrome konstant sind. Es gibt allerdings mehr und mehr Hinweise, dass diese Korrelationen anpassungsfähig und flexibel sind (Fischer et al. 2016). Verhaltenssyndrome können zwischen verschiedenen Populationen der gleichen Art variieren, wenn die Populationen aus unterschiedlichen Umwelten kommen, wie zum Beispiel aus Labor- und Wildpopulationen.

Mit der vorliegenden Doktorarbeit beabsichtige ich, die Existenz von Tierpersönlichkeiten in der Eurasischen Zwergmaus (*Micromys minutus*) zu belegen. Außerdem ist mein Ziel, Zusammenhänge zwischen konstanten Verhaltensweisen (Verhaltenssyndrom) sowie zwischen Verhalten und räumlich-kognitiven Merkmalen zu finden (kognitives Syndrom). Es gibt bislang keine Studien zur Tierpersönlichkeit bei Zwergmäusen. Jedoch eignet sich diese kleine Nagetierart auf Grund folgender Eigenschaften sehr gut für Tierpersönlichkeitsstudien. Dies gilt vor allem in Bezug auf räumlich-kognitive Syndrome. (I) Zwergmäuse sind einfach im Labor zu halten und zu züchten, (II) Verhaltenstests und Tests zu kognitiven Fähigkeiten sind leicht auf der Basis von schon bekannten Versuchsaufbauten bei Labormäusen und -ratten zu etablieren, (III) Zwergmäuse haben vermutlich sehr gut entwickelte räumlich-kognitive Fähigkeiten auf Grund der Anpassung an ein Leben in drei-dimensionalen Lebensräumen (Piechocki 2001) und (IV) Zwergmäuse sind zwar eine relativ weit verbreitete Art, jedoch lokal teils gefährdet (Darinot 2016), sodass mehr Wissen über ihre Verhaltensökologie neue Erkenntnisse zum verbesserten Schutz dieser Art bringen könnte.

Es wurden zunächst die Wiederholbarkeit (*repeatability*) und die Konstanz (*consistency*) von Verhaltensweisen und räumlich-kognitiven Merkmalen untersucht. Dabei zeigte sich, dass Exploration, Aktivität und Mut sowohl in Jungtieren als auch in adulten Zwergmäusen wiederholbar war. Räumliches Erkennen war hingegen nur in erwachsenen Zwergmäusen wiederholbar, außerdem auch die räumliche Lernfähigkeit und die Entscheidungsgeschwindigkeit. Die Genauigkeit der Entscheidungen war nicht wiederholbar. Exploration, Aktivität und Mut waren konstant vor und nach der Geschlechtsreife, sowie vor und nach dem ersten Sexualkontakt. Die Konstanz des räumlichen Erkennens war weniger ausgeprägt. Zwergmäuse zeigten ein Verhaltenssyndrom, da die Verhaltensweisen positiv miteinander korrelierten. Es bestand kein Konflikt zwischen der Entscheidungsgeschwindigkeit und der Genauigkeit (*speed-accuracy trade-off*). Allerdings gab es einige Hinweise für ein kognitives Syndrom, da schnelle Verhaltenstypen auch schneller lernten als langsamere Verhaltenstypen. Außerdem machten schüchtere Zwergmäuse schnellere Entscheidungen als mutige. All diese Untersuchungen wurden mit Zwergmäusen aus einer Labor-Population durchgeführt. Für Zwergmäuse aus einer Gehege-Population mit einem natürlichen genetischen Hintergrund zeigte sich, dass Exploration, Aktivität und Mut auch hier wiederholbar war. Räumliches Erkennen war jedoch nicht wiederholbar. Ein Verhaltenssyndrom mit positiven Korrelationen zwischen den Verhaltensweisen war auch in der Gehege-Population zu finden.

Mit dieser Doktorarbeit konnte zum ersten Mal nachgewiesen werden, dass es auch bei Eurasischen Zwergmäusen Tierpersönlichkeit gibt. Zusammengefasst lässt sich aus den

Ergebnissen schließen, dass Zwergmäuse wiederholbares Verhalten in Labor- und Gehege-Populationen zeigen. Exploration, Aktivität und Mut waren zudem in der Labor-Population über mehrere Lebensabschnitte hinweg konstant. Obwohl Unterschiede in der Stärke der Korrelationen der Verhaltensweisen zwischen der Labor- und der Gehege-Population gefunden wurden, zeigten beide Verhaltenssyndrome gleiche Richtungen an. Daher lässt sich sagen, dass Zwergmäuse generell ein Verhaltenssyndrom entlang eines schnell-langsam-Gradienten aufzeigen. Dieses wird vermutlich eher durch proximate Faktoren als durch Selektionsprozesse gesteuert. Räumliches Erkennen hatte eine geringe Wiederholbarkeit und war weniger konstant über Lebensabschnitte hinweg. Es wurde festgestellt, dass die räumliche Lernfähigkeit und die Entscheidungsgeschwindigkeit wiederholbar waren, jedoch nicht die Entscheidungs-genauigkeit. Auf Grund des steigenden Forschungsinteresses an individueller Variation in kognitiven Fähigkeiten und an dem Zusammenhang mit Tierpersönlichkeit sollten mehr Daten über die Wiederholbarkeit und Konstanz von kognitiven Merkmalen gesammelt werden. Die gezeigten Korrelationen inzwischen Verhaltensweisen und räumlich-kognitiven Merkmalen zeigen zumindest teilweise, dass es ein kognitives Syndrom bei Zwergmäusen geben könnte. Daher werden diese Ergebnisse hoffentlich zu weiteren Studien über die Beziehung von (räumlich-) kognitiven Merkmalen und Tierpersönlichkeit anregen.

Contents

SUMMARY I

ZUSAMMENFASSUNG III

INTRODUCTION 1

PART 1 - INTRODUCTION TO CONSTANT INDIVIDUAL DIFFERENCES IN ANIMAL BEHAVIOUR 1

CONSTANT BETWEEN-INDIVIDUAL DIFFERENCES IN ANIMAL BEHAVIOUR 1

DEFINITIONS AND TERMINOLOGY OF ANIMAL PERSONALITY 3

ASSUMPTIONS OF CONSTANT BETWEEN-INDIVIDUAL BEHAVIOURAL DIFFERENCES 7

I - REPEATABILITY AND CONSISTENCY OF BEHAVIOURAL TRAITS 7

SELECTION FOR CONSTANT BETWEEN-INDIVIDUAL BEHAVIOURAL DIFFERENCES 10

HERITABILITY OF BEHAVIOURAL TRAITS 11

EVOLUTION OF CONSTANT BETWEEN-INDIVIDUAL BEHAVIOURAL DIFFERENCES 13

II - BEHAVIOURAL SYNDROMES: BETWEEN-INDIVIDUAL CORRELATIONS AMONG BEHAVIOURAL TRAITS 14

SELECTION FOR BEHAVIOURAL SYNDROMES 15

GENETICALLY CORRELATED BEHAVIOURAL TRAITS 16

EVOLUTION OF BEHAVIOURAL SYNDROMES 17

PART 2 - INTRODUCTION TO ANIMAL COGNITION 19

STUDYING COGNITION IN ANIMALS 19

SPATIAL COGNITION IN ANIMALS 20

BETWEEN-INDIVIDUAL DIFFERENCES IN COGNITIVE TRAITS 21

SELECTION, HERITABILITY AND EVOLUTION OF INDIVIDUAL DIFFERENCES IN COGNITIVE TRAITS 22

COGNITIVE SYNDROME 24

AIM OF THE THESIS 28

OBJECTIVES OF THIS THESIS 28

CHAPTER 1 33

BEHAVIOURAL REPEATABILITY AND CONSISTENCY IN HARVEST MICE 33

BEHAVIOURAL REPEATABILITY AND CONSISTENCY IN HARVEST MICE 33

REPEATABILITY AND CONSISTENCY OF ANIMAL BEHAVIOURS 34
SEX DIFFERENCES AND LIFE HISTORY TRADE-OFFS 36
DIFFERENT ESTIMATION METHODS 36
STUDY OBJECTIVES AND HYPOTHESES 37
METHODS – REPEATABILITY AND CONSISTENCY IN HARVEST MICE 39
STUDY ANIMALS AND HOUSING CONDITIONS 39
EXPERIMENTAL SET-UP 41
BEHAVIOURAL TESTS 42
DATA ANALYSES 45
RESULTS – REPEATABILITY AND CONSISTENCY IN HARVEST MICE 47
EFFECTS OF CONFOUNDING FACTORS 47
EFFECTS OF SEX AND AGE ON THE MEAN EXPRESSION OF BEHAVIOURS 48
REPEATABILITY OF BEHAVIOURAL TRAITS IN HARVEST MICE 48
CONSISTENCY OF BEHAVIOURAL TRAITS IN HARVEST MICE 49
ESTIMATION METHODS / RELIABILITY 49
DISCUSSION – REPEATABILITY AND CONSISTENCY IN HARVEST MICE 53
EFFECTS OF SEX AND AGE ON THE MEAN EXPRESSION OF BEHAVIOURS 53
REPEATABILITY OF BEHAVIOURAL TRAITS IN HARVEST MICE 54
CONSISTENCY OF BEHAVIOURAL TRAITS IN HARVEST MICE 55
ESTIMATION METHODS AND EFFECT OF THE LITTER 57
CONCLUSION - REPEATABILITY AND CONSISTENCY IN HARVEST MICE 58
REPEATABILITY AND CONSISTENCY IN HARVEST MICE OF THE LABORATORY POPULATION 58

CHAPTER 2 59

BEHAVIOURAL AND COGNITIVE SYNDROMES IN LABORATORY HARVEST MICE 59
BEHAVIOURAL AND COGNITIVE SYNDROMES IN LABORATORY HARVEST MICE 59
THE COGNITIVE SYNDROME 60
STABILITY OF COGNITIVE STYLES 62
SPATIAL ORIENTATION IN COGNITIVE SYNDROMES 62
STUDY OBJECTIVES AND HYPOTHESES 62
MATERIAL AND METHODS – BEHAVIOURAL AND COGNITIVE SYNDROME 63
STUDY ANIMALS AND HOUSING CONDITIONS 63
BEHAVIOURAL TESTS 64

DATA ANALYSES 68

RESULTS – BEHAVIOURAL AND COGNITIVE SYNDROME 70

REPEATABILITY OF PERSONALITY AND COGNITIVE TRAITS IN HARVEST MICE 70

CORRELATIONS BETWEEN TRAITS IN HARVEST MICE 73

DISCUSSION - BEHAVIOURAL AND COGNITIVE SYNDROME 76

REPEATABILITY OF PERSONALITY AND COGNITIVE TRAITS IN HARVEST MICE 76

BEHAVIOURAL SYNDROME IN HARVEST MICE 77

COGNITIVE SYNDROME IN HARVEST MICE 79

CONCLUSION - BEHAVIOURAL AND COGNITIVE SYNDROME IN HARVEST MICE 82

BEHAVIOURAL AND COGNITIVE SYNDROME IN HARVEST MICE OF THE LABORATORY POPULATION 82

CHAPTER 3 83

BEHAVIOURAL SYNDROME IN THE FIELD POPULATION OF HARVEST MICE 83

ANIMAL PERSONALITY AND BEHAVIOURAL SYNDROMES 83

BEHAVIOURAL SYNDROMES IN LABORATORY AND FIELD POPULATIONS 84

STUDY OBJECTIVES AND HYPOTHESES 86

MATERIAL AND METHODS – BEHAVIOURAL SYNDROME IN THE FIELD POPULATION 87

STUDY ANIMALS AND SEMI-NATURAL ENCLOSURE 87

LIFE TRAPPING AND INDIVIDUAL IDENTIFICATION 88

LABORATORY POPULATION 89

HOUSING CONDITIONS AND BEHAVIOURAL TESTS 90

DATA ANALYSES 92

REPEATABILITY 92

CORRELATIONS BETWEEN BEHAVIOURS 93

COMPARISON BETWEEN THE LABORATORY AND THE FIELD POPULATION 94

RESULTS - BEHAVIOURAL SYNDROME IN THE FIELD POPULATION OF HARVEST MICE 94

REPEATABILITY OF BEHAVIOURS IN THE FIELD POPULATION 94

CORRELATIONS BETWEEN BEHAVIOURS IN THE FIELD POPULATION 95

CORRELATIONS BETWEEN BEHAVIOURS IN THE LABORATORY POPULATION 96

COMPARISON BETWEEN LABORATORY AND FIELD POPULATION 97

DISCUSSION - BEHAVIOURAL SYNDROME IN THE FIELD POPULATION OF HARVEST MICE 100

BEHAVIOURAL SYNDROME IN THE FIELD POPULATION OF HARVEST MICE 100

REPEATABILITY OF BEHAVIOURS IN EURASIAN HARVEST MICE 101

BEHAVIOURAL DIFFERENCES BETWEEN THE LABORATORY AND THE FIELD POPULATION 102

CORRELATIONS BETWEEN BEHAVIOURS IN EURASIAN HARVEST MICE 103

CONCLUSION - ANIMAL PERSONALITY IN EURASIAN HARVEST MICE 105

ANIMAL PERSONALITY IN EURASIAN HARVEST MICE 105

DISCUSSION 107

BEHAVIOURAL REPEATABILITY AND CONSISTENCY IN EURASIAN HARVEST MICE 107

BEHAVIOURAL REPEATABILITY IN EURASIAN HARVEST MICE 107

BEHAVIOURAL CONSISTENCY IN EURASIAN HARVEST MICE 108

BEHAVIOURAL AND COGNITIVE SYNDROME IN EURASIAN HARVEST MICE 109

BEHAVIOURAL SYNDROME IN EURASIAN HARVEST MICE 109

COGNITIVE SYNDROME IN EURASIAN HARVEST MICE 110

EVOLUTION OF CONSTANT BETWEEN-INDIVIDUAL DIFFERENCES 111

EVOLUTION OF CONSTANT BETWEEN-INDIVIDUAL DIFFERENCES IN EURASIAN HARVEST MICE 111

CONCLUSION AND OUTLOOK 115

CONSTANT BETWEEN-INDIVIDUAL DIFFERENCES IN EURASIAN HARVEST MICE 115

FUTURE DIRECTIONS 115

DANKSAGUNG XI

APPENDIX XIII

LIST OF FIGURES XXIV

LIST OF TABLES XXVI

LIST OF SUPPLEMENTARY TABLES XXVIII

REFERENCES XXX

Introduction

The evolution of behaviour has always fascinated biologist. In particular, animal behaviour has become a fast growing research area over the last decades, using behaviour as a mechanism to observe how animals (phenotypes and genotypes) interact with their environments (Kappeler 2010). Behavioural ecology thereby aims to understand how and why environmental conditions shape animal behaviours (Lantová and Dall 2016). It is expected that natural and sexual selection favour optimal behaviours of populations in their specific environments (Wilson et al. 1994; Wilson 1998). The individual variation around that optimal mean was assumed to be measurement errors (Dall et al. 2004) or individual phenotypic plasticity (Nussey et al. 2007). However, there is an unstoppable interest in these individual behavioural variation as it has been shown to be an adaptive strategy of individuals to behave constantly different from each other (Bell 2005).

The following introduction consists of two parts: In the beginning, I introduce the research field of animal personality, explaining the importance of constant between-individual differences in behaviours of animals. An overview about the variety of terminology used in this study field is presented. I explain the two assumptions of animal personality (constancy in behaviour and correlation between behaviours). Further, I describe evolutionary processes explaining and maintaining constant individual behavioural differences and correlations between different behaviours in animal populations. In the second part, I introduce the research area of animal cognition, with a focus on spatial cognition. I present the recently hypothesised connection between constant individual behavioural differences and animal cognition. In the end of the introduction, I clarify the aims of this thesis.

Part 1 - Introduction to Constant Individual Differences in Animal Behaviour

Constant Between-Individual Differences in Animal Behaviour

Behavioural traits were assumed to be the most flexible traits in animals (Briffa et al. 2008). However, Dall and colleagues stated that “from an adaptive perspective, it makes sense for individuals to adjust their behaviour according to current conditions (including their internal state), and this can result in individual differences in behaviour if there is between-individual variance in local conditions” (Dall et al. 2004, page 734). Further, there is now various evidence for constant between-individual behavioural differences resulting in less flexible individual behaviour (Evans et al. 2010). Although this was assumed to be improbable because behavioural plasticity (the potential of organisms to produce different, relatively fit phenotypes in different environmental conditions, DeWitt et al. 1998), rather than behavioural constancy seems to be selected for in heterogeneous environments (DeWitt et al. 1998; Dall et al. 2004). Constant between-individual differences in behaviour means that individuals constantly differ in how they behave. Adaptive constant between-individual differences in behaviour were already recognized in, for example, alternative male-mating tactics (Gross 1996). But that animals could generally behave constantly different in various situation and contexts, was totally ignored by behavioural ecologists for a long time (Sih and Bell 2008). Yet, more recently evolutionary biologists "adopt a general set of expectations about adaptive individual differences within single populations that can be applied to all traits" (Wilson 1998, page 199). For instance, in a population are individuals which are constantly bolder and more aggressive than others. Although this phenomenon has been studied in human personality research since 1883 (Galton 1883), studies on the individuality of behaviour in animals has only increased in the last decades (see ***Figure 1***). Behavioural axes - also called behavioural categories - usually studied in animal personality are boldness (risk-taking behaviour), exploration, activity, aggression, and sociability (Réale et al. 2007).

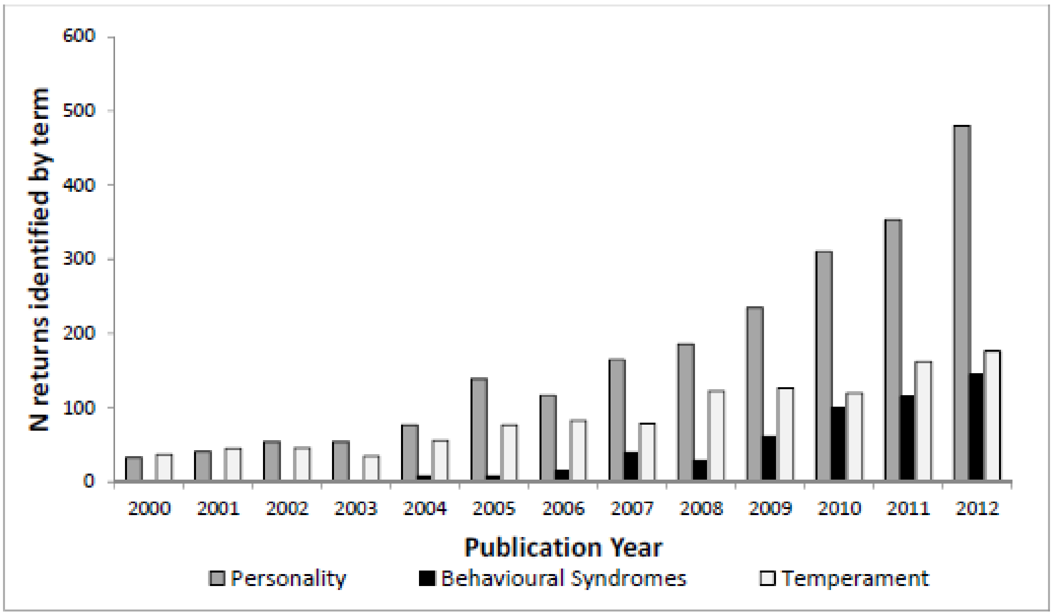

Figure 1 Number of publications per year between 2000 and 2012, dealing with either animal personality, behavioural syndrome or temperament. Graphic is reused from MacKay and Haskell (2015).

There are two possible explanations – the CONSTRAINT HYPOTHESIS and the ADAPTIVE HYPOTHESIS – for constant individual behaviour, and for correlated behavioural traits (see below). The *constraint* *hypothesis* assumes that animals behave constantly because maintaining flexible behaviour is costly (DeWitt et al. 1998). For instant, being bold in a specific situation but reacting cautiously in a different context may be costly. This is because such flexible behaviour may require maintaining a sensitive sensory system and a broad range of physiological processes (Bergmüller 2010). The different situations could in this way specifically perceived and reactions differently validated depending on the situation. Flexibility could also be limited due to fixed coupling of (behavioural) traits (DeWitt et al. 1998). Underlying genetic or physiological constraints could thereby result in fixed individual behaviours even if these behaviours appear to be maladaptive in certain situations or contexts (Carere and Maestripieri 2013). On the other hand, the *adaptive hypothesis* assumes that constant behaving individuals have fitness advantages (Bergmüller 2010). As reviewed by Bergmüller (2010), individual constancy in behaviour may result in fitness benefits of specialists compared to generalists, and may reduce conflicts between individuals. Further, behavioural constancy may also be some kind of commitment because conspecifics can rely on other's previous behavioural pattern (Bergmüller 2010).

Definitions and Terminology of Animal Personality

In the literature, there is a confusing variety of terminology all referring to the phenomenon of constant between-individual behavioural differences in animal populations (David and Dall 2016). The application of (partly) modified expressions used in human personality and psychobiology research, as well as the use of synonymous terms makes it even more puzzling to understand scientific texts in this research area (MacKay and Haskell 2015). In the past, there have been several attempts to provide clear definitions, and conceptual and methodological frameworks in animal science (e.g., Koolhaas et al. 1999; Gosling 2001; Sih et al. 2004a; Bell 2007; Réale et al. 2007). However, we still have no clear picture for the terminology (David and Dall 2016, but see Koski 2014). In general, there are two basic approaches: The first assumes constant behavioural variation between individuals and individual behavioural traits which are repeatable and consistent across time and situations (e.g., temperament and animal personality). The second approach considers constant individual variation in more than one behavioural trait or in more than one context, whereby assuming correlation between these behaviours within populations (e.g., behavioural syndrome and coping style). There are even more terms in the literature describing similar patterns of constant between-individual differences in behaviour (e.g., coping strategies, behavioural profiles, characters, personality dimensions, predispositions, individuality, cf. Uher 2011). However, I will here focus on the following four terms as they are now used most commonly: Animal personality, temperament, behavioural syndrome and coping style.

ANIMAL PERSONALITY is often used as a synonym of temperament (see below) and it is defined as between-individual behavioural differences that are repeatable over time and across situations (Réale et al. 2007). Although the term personality arose from studies in humans, it has been used in animal studies for many years and there has been a vast increase of animal personality publications since the year 2000 (MacKay and Haskell 2015, ***FIGURE 1***). The definition is based on behavioural patterns which are non-random, and it can be used to describe the particular aspect of behavioural phenotypes which are constant over time and differ constantly between individuals within a population (Bergmüller 2010). In animal personality studies, constant individual behaviour is investigated with respect to ecological consequences, and it considers how and why constant between-individual differences have evolved (David and Dall 2016). Animal personality can also be used to describe the more general topic and the research field dealing with constant between-individual behavioural differences in animals. Recently, there is also an integration of animal personality into research fields like community ecology (Modlmeier et al. 2015; Quinn 2015) and animal cognition (Sih and Del Giudice 2012; Morton et al. 2013).

The term *TEMPERAMENT* originates also from human personality and psychology. As reviewed by Réale et al. (2007), there are several definitions, but it seems that even in human research, the definitions are not used uniformly (Gosling 2001). Temperament was defined as inherited behavioural tendencies occurring in early life and providing the basis of personality later in life (Goldsmith et al. 1987). However, temperament is also used as a synonym of (animal) personality (Réale et al. 2007) without the restriction to early life-history phases. Thus, temperament can also be defined as between-individual behavioural differences that are repeatable over time and across situations (Réale et al. 2007). Recently, temperament was found to be mainly used in agricultural research, developmental biology, and veterinary sciences (MacKay and Haskell 2015). In applied ethology, farm animals are also categorised depending on their individual behaviour. Thereby, it is assumed that temperament is preferably used instead of (animal) personality to avoided anthropomorphism (Gosling 2001; MacKay and Haskell 2015). Initially, temperament was considered to be addressed to the individual level, but it can also be used for populations or species (Réale et al. 2007). The use of the term temperament does not restrict to specific species, it is widely used in all animal taxa (e.g., Reale et al. 2000; Brydges et al. 2008; Kobler et al. 2009; Eccard and Rödel 2011; Zambra et al. 2015). MacKay and Haskell stated recently that "temperament occupies a niche which personality and behavioural syndromes cannot fill" (MacKay and Haskell 2015, page 468). They argue that temperament refers to a one-context behavioural variation and is quantifiable in contrast to personality which cannot be quantified and refers to the underlying structure of behavioural variation (MacKay and Haskell 2015). However, their attempt to combine all definitions of temperament, personality and behavioural syndrome (see below) seems to lead to even more confusions.

Sih et al. (2004a) defined *BEHAVIOURAL SYNDROMES* as suits of correlated behaviours across two or more contexts or situations. Context was thereby defined as a behavioural category (e.g., mating or feeding context) and situation was defined as “a given set of conditions at one point in time” (Sih et al. 2004a, page 243). In a population, different BEHAVIOURAL TYPES occur. They are defined by each individual combination of two or more behavioural trait levels (see ***FIGURE 2***, Bell 2007). The correlation between the two or more behaviours was further defined as behavioural syndrome (see ***FIGURE 2***, Bell 2007). Behavioural types could be identified to be either distributed along a shyness-boldness continuum (Wilson et al. 1994), along an avoidance-exploration continuum (Réale et al. 2007), or along a fast-slow continuum (Titulaer et al. 2012). Thereby, behavioural traits along the different continuums further correlated with other behavioural axes. For instance, more exploratory great tits (*Parus major*) were bolder (Verbeek et al. 1994), more aggressive (Carere et al. 2005) and showed more risk-taking behaviours (van Oers et al. 2004b) than less explorative birds. Another definition of a behavioural syndrome was identified to follow a proactive-reactive continuum (see below).

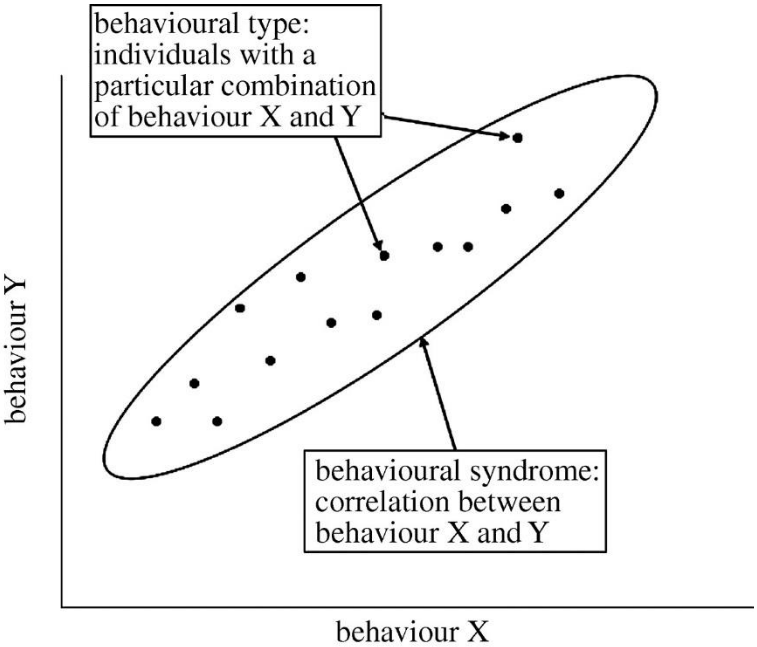

FIGURE 2 Different behavioural types forming a behavioural syndrome between behaviour X and Y. Each dot indicates an individual in a population. With permission (License Number 3879250630192) from Bell (2007).

COPING STYLE was defined as “a coherent set of behavioural and physiological stress responses which is consistent over time and which is characteristic to a certain group of individuals”

(Koolhaas et al. 1999, page 925) meaning that it also defines a syndrome. The term coping style is usually used when physiological parameters (e.g., stress hormones) are investigated in combination with behaviours. Traits or correlations between behavioural and physiological traits are also assumed to be constant over time and characterise populations not individuals (Koolhaas et al. 1999). Within copying styles, two behavioural types (e.g., combination of behavioural trait X and physiological trait Y, cf. ***FIGURE 2***) were defined: The proactive and the reactive type (Koolhaas et al. 1999). Thereby, proactive individuals are expected to respond actively and aggressively towards stress stimuli, while reactive animals behave less active and less aggressive than the proactive type (Koolhaas et al. 1999). Furthermore, both types are assumed to differ in their neuroendocrine characteristics, such as adrenaline or corticosterone levels (Koolhaas et al. 1999), and would be located on the extreme ends of the syndrome (cf. ***FIGURE 2***). Originally, the concept of coping styles was established in laboratory house mice (*Mus musculus*; Benus et al. 1991), but was also shown to apply to a number of other species (e.g., pigs (*Sus scrofa*), tree shrews (*Tupaja belangeri*), and rhesus monkey (*Macaca mulatta*) as reviewed by Koolhaas et al. 1999). Also recently, this concept is widely used, for example in mammals (Monestier et al. 2015), fish (Ferrari et al. 2015), and arthropods (Mesquita et al. 2015).

TABLE 1 Overview of the terminology used in this thesis regarding constant individual behavioural differences. Terms and corresponding definitions are shown, detailed explanations and references are found in the text.

TERM	DEFINITION
ANIMAL PERSONALITY	Individual behavioural differences that are repeatable over time and across situations (Réale et al. 2007)
BEHAVIOURAL TRAIT	A quantifiable behaviour (e.g., activity, boldness, exploration, aggression, docility, sociability)
PERSONALITY TRAIT	A behavioural trait which shows constancy (repeatability and/or consistency) in individuals of a population
BEHAVIOURAL TYPE	An individual with a specific combination of two or more behavioural trait levels (Bell 2007)
REPEATABILITY	The proportion of total variance assigned to between-individual variance from observations within the same contexts or life history phases (Dingemanse et al. 2010)
CONSISTENCY	The proportion of total variance assigned to between-individual variance from observations from different contexts or life history phase
BEHAVIOURAL SYNDROME	Suits of correlated behaviours across two or more contexts or situations (Sih et al. 2004a)

In the following chapters, I refer to *animal personality* when addressing topics dealing with constant between-individual behavioural differences in general. If constancy of behavioural traits (repeatability and/or consistency, see page 7) was proven, behavioural traits will further be referred to as *personality traits*. When speaking about correlations between different personality traits, I will refer to *behavioural syndromes*. All definitions used in the following paragraphs are summarised in ***TABLE 1***.

Assumptions of Constant Between-Individual Behavioural Differences

By definition (of all previously explained terms, see page 3) animal personality is based on the following assumptions: First, between-individual behavioural differences are assumed to be constant through time and across different situations (Réale et al. 2007). Further, individual behaviour is assumed to be constant across context, and phenotypic correlations between different behavioural categories are expected, forming a behavioural syndrome (Sih et al. 2004b).

I - Repeatability and Consistency of Behavioural Traits

Animal personality assumes stability of behavioural traits (Réale et al. 2007). Several terms are used to describe this stability: Test-retest reliability, temporal reliability, repeatability, cross-context repeatability, or consistency. Sometimes, these terms are used to explain similar characteristics of behavioural traits, but sometimes not. Thus, it can be difficult to distinguish between their definitions. In the following paragraph, I explain REPEATABILITY and CONSISTENCY in more detail as those terms are clearly related to the temporal constancy definition of animal personality.

The first assumption of animal personality is that behaviours are repeatable (Réale et al. 2007). In general, the term repeatability is used when assessing the accuracy of specific measurements (Nakagawa and Schielzeth 2010). In animal personality research, repeatability specifies the proportion of between-individual variance relative to the total phenotypic variance in a population for repeated measures of the same behaviour (Dingemanse et al. 2010). It can be interpreted as the expected correlation between repeated measurements of a behaviour of the same individual. The repeatability of a specific behaviour is usually calculated based on repeated observations in the same context and life history phase. When estimating repeatability we assume that environmental factors of each replicated measure are constant (Réale et al. 2007). As defined in ***TABLE 1*** (page 6), I refer to REPEATABILITY as the proportion of between-individual variance from observations within the same life history phase. There have been several estimation methods used to quantify the repeatability of behaviour. When two measures of the behaviour of interest have been taken for each individual, rank correlations between the first and the second measurement can be used to calculate repeatability (e.g., Herde and Eccard 2013; Bajer et al. 2015). When two or more measures of a behavioural trait are available for different individuals, repeatability can further be obtained by running an analysis of variance (ANOVA) with individuals included as a fixed factors (Lessells and Boag 1987). Thereby, the total phenotypic variance is separated into between-individual variance and within-individual variance to estimate repeatability (Lessells and Boag 1987):

$$repeatability = \frac{between\text{-}individual\ variance}{within\text{-}individual\ variance + between\text{-}individual\ variance}$$

More recently, Nakagawa and Schielzeth (2010) recommended the use of linear mixed effect models (LMMs) for the estimation of those variances. Without including any confounding factors, another form of *agreement repeatability* is calculated (Nakagawa and Schielzeth 2010). However, to account for confounding factors, confounding variables through fixed factors and covariates should be included in the model. This form of repeatability was termed *adjusted repeatability* (Nakagawa and Schielzeth 2010) and is commonly used in animal personality studies (e.g., Hall et al. 2015; Small and Schoech 2015). Furthermore, generalised linear mixed effect models (GLMMs) can be applied if models on behavioural traits show non-Gaussian error distributions (Nakagawa and Schielzeth 2010). In cases where varying testing conditions contribute to between-individual variation, the use of LMMs is particularly useful to avoid an overestimation of repeatability (Dingemanse and Dochtermann 2013). Repeatability is calculated as a fraction of total trait variance that can be assigned to between-individual variance due to additive and non-additive genetic effects, as well as due to the permanent environment effect (Falconer et al. 1996; Nakagawa and Schielzeth 2010). Thus, low repeatability for a behavioural trait could be observed if, for example, the conditions chosen for the experimental test do not generate phenotypic variation (i.e. all individuals behave equally) or by high within-individual variation relative to between-individual variation (Réale et al. 2007). The latter could be the case if behavioural observations are done with individuals at different ages, and there is an age effect which is larger than the between-individual effect. Repeatability is a unit-free standardized measure that can readily be compared among populations, species and studies (Réale et al. 2007). However, which estimation method was chosen for the calculation of repeatability can have profound effects on the conclusions, as illustrated by the following example: In birds, Small and Schoech (2015) showed that the stress-induced corticosterone level was repeatable in adult female Florida scrub-jays (*Aphelocoma coerulescens*), but not in males. However, this difference between the sexes was only apparent when agreement repeatability was calculated. When the authors applied LMMs including effects of the study year and the time of day, the resulting values for adjusted repeatability (Nakagawa and Schielzeth 2010) of stress-induced corticosterone level were significant in both sexes (Small and Schoech 2015).

In the vast body of literature of animal personality, repeatable behaviours have been observed in all animal taxa (Bell et al. 2009). Within all animal taxa, Bell et al. (2009) estimated an average behavioural repeatability of 0.37.

Repeatability estimates from observations across contexts or life history phases have also been reported as ‘repeatability’, but the alternative terms cross-context repeatability (e.g., Laskowski and Bell 2013) or consistency (e.g., Wuerz and Krüger 2015) allow us to better specify the different information content of these estimates. The proportion of between-individual variance among measurements from different contexts or life history phases characterises the stability of individual behaviour across different behavioural categories or across a longer time interval assuming probable changes in the environment. Rank correlations of individual behaviour across contexts can also indicate consistent individual behaviour measured in different situations (Brommer and Class 2015). However, while rank correlations report stable or changing rank orders of mean individual behavioural performance within a population, they lack information on the relative contribution of repeatable individual performances to the total phenotypic variance in a population. Given that, I refer to CONSISTENCY of behavioural traits as the proportion of between-individual variance from observations at different ontogenetic stages (see ***TABLE 1***, page 6). There is a large body of literature providing evidence for consistency of behavioural traits in many animal species. However, consistency is not necessarily evident for all kinds of behaviours. For instance, in common voles (*Microtus arvalis*) activity, exploration and boldness were repeatable, but only activity parameters were also consistent when tested over a longer time interval (Herde and Eccard 2013). This might be due to variation in life history parameters, for example, expected life span (Herde and Eccard 2013) or seasonality and environmental changes (Eccard and Herde 2013). In Siberian dwarf hamsters (*Phodopus sungorus*), Kanda et al. (2012) showed that the individual activity level was consistent between all age classes. However, boldness, reactivity and orientation showed less consistency. In great tits (*Parus major*), exploration was a consistent trait over a time period of on average 150 days (Dingemanse et al. 2002). David et al. (2012) showed that zebra finches (*Taeniopygia guttata*) also explored consistently over a seven month interval, which represented about one quarter of the life span of this species. Given our current knowledge, it seems difficult to predict which behaviours may show long-term consistency in a specific species.

Selection for Constant Between-Individual Behavioural Differences

Measuring selection requires an estimation of fitness parameters for each individual (Morand-Ferron et al. 2015). As reviewed by McDougall et al. (2006), strong evidence for selection on personality traits in captivity was shown by associations between personality traits and fitness (e.g., in cheetahs, *Acinonyx jubatus*, Wielebnowski 1999; and in black rhinoceroses, *Diceros bicornis*, Carlstead et al. 1999). In wild populations, constant between-behavioural differences could also be linked to differences in reproductive success (Reale et al. 2000; Both et al. 2005), parental care (Budaev et al. 1999) and survival (Dingemanse et al. 2004; Boon et al. 2007). A meta-analysis by Smith and Blumstein (2008) on laboratory and wild populations further revealed that bolder and more aggressive individuals had on average a higher reproductive success, but more exploratory individuals survived longer.

Different forms of natural selection have been identified to as potential driving forces that can maintain constant between-individual behavioural differences in animal populations. *Balancing selection* is assumed to be one mechanism maintaining diverse behavioural types in a population (Bergmüller 2010). Thereby, *frequency-dependent selection* is assumed to lead to the coexistence of these behavioural types within a population (Dall et al. 2004). In evolutionary game theory, fitness payoffs depending on the frequencies of the competing strategies (here behavioural types) are well known (Smith 1982) and this may also explain between-individual behavioural differences. Namely, negative frequency-dependent selection could be observed if there are two behavioural types occurring in a population and the rare behavioural type has fitness advantages compared to the frequent behavioural type. This may then result in a shift of the frequencies: The rare behavioural type becomes more and more frequent and the frequent behavioural type becomes rarer and thus gains fitness advantages again. It was shown that frequency-dependent selection acted on the frequency distribution of explorative behavioural types in breeding pairs of great tits (*Parus major*, Dingemanse et al. 2004; Both et al. 2005). Thus, under negative frequency dependent selection, one would expect that a certain combination of intermediate frequencies of alternative behavioural types would yield a high average fitness in the population. In three spider species of the genus *Anelosimus*, Lichtenstein and Pruitt (2015) indeed illustrated that a low frequency of the aggressive behavioural type within a colony can favour the fitness (measured as mass gain) of all behavioural types compared to colonies where aggressive individuals were either absent, or more frequent.

Spatio-temporal environmental heterogeneity (Roff 1997) was shown to be a factor that favours the coexistence of different behavioural types (Carere and Maestripieri 2013). During certain environmental conditions one behavioural type has fitness advantages, but when environmental condition changes a different behavioural types is favoured, resulting in the coexistence of both types (Dingemanse et al. 2004). In this case, the direction of selection is not stable and this *temporally fluctuating selection* has been shown to be an important mechanism maintaining individual behavioural differences (e.g., Dingemanse et al. 2004; Boon et al. 2007; Quinn et al. 2009). One recent example in Siberian chipmunks (*Tamias sibiricus*) showed that bolder individuals had higher annual reproductive success than shy animals but only in years with low food availability. When food availability was high, the annual reproductive success was lower in bolder chipmunks than in shyer animals (Le Coeur et al. 2015). Furthermore, spatial variation in selection was observed in Western bluebirds (*Siala mexicana*), where more aggressive birds survived longer in some populations but not in others (Duckworth 2008). Alternatively, spatial or temporal variation of population sizes can cause *density-dependent selection* (Wilson et al. 1994) on behavioural types. In common lizards (*Zootoca vivipara*), sociable animals survived better in high density environments than unsociable individuals, but this was reversed in low density environments (Cote et al. 2008). In the same species, Le Galliard et al. (2015) recently concluded that density-dependent selection is important in maintaining behavioural types, however, they found heterogeneous selection patterns depending on the specific behavioural traits that were analysed. Another possible mechanism resulting in variability of behavioural types might be *disruptive selection* (Wilson 1998). When behavioural types show a bi- or multi-modal frequency distribution, it is likely that these behavioural types were selected for by disruptive selection (Bergmüller 2010). Life history trade-offs (Wolf et al. 2007), as well as sexual selection (Schuett et al. 2010) were shown to be further mechanisms acting on the presence and frequency of specific behavioural types.

Heritability of Behavioural Traits

Only if behavioural traits have a genetic component, we would expect a response to selection on that trait and hence, evolution of behavioural types. To identify the genetic architecture of personality traits seems to be one of the key goals for the understanding of the origin of animal personality, its evolution, and its maintenance (van Oers and Mueller 2010). Thereby, heritability is an important estimate to understand, whether evolutionary processes shape personality traits (Bergmüller 2010).

To calculate the heritability of behaviours, the phenotypic variance (V_P) is partitioned into the additive genetic variance (V_A), variance due to maternal effects (V_M), permanent environmental variance (V_{PE}), and residual variance (V_R) using the following model (Dochtermann et al. 2015):

$$V_P = V_A + V_M + V_{PE} + V_R$$

The narrow sense heritability (h^2) of the behaviour can then be calculated by dividing the additive genetic variance by the total phenotypic variance (Falconer et al. 1996):

$$h^2 = \frac{V_A}{V_A + V_M + V_{PE} + V_R}$$

There is growing evidence that the variability of constant individual behaviours has a genetic basis (van Oers et al. 2005). Several studies observed heritable variance in behavioural traits. An average heritability of 0.14 was estimated across 70 behaviours (Dochtermann et al. 2015). For instance, boldness showed significant heritability of 0.27 in dogs (*Canis domesticus*, Strandberg et al. 2005), boldness and docility were significantly heritable ($h^2 > 0.21$) in bighorn sheep (*Ovis canadensis*, Réale et al. 2009; Poissant et al. 2013), exploration showed significant heritability in great tits (*Parus major*, Dingemanse 2002; Drent 2003), and also risk-taking behaviour was significantly heritable in this species (van Oers et al. 2004b). In rodents, evidence so far suggested a moderate heritability of locomotion ability ($h^2 = 0.21$) but very low heritability of vigilance ($h^2 = 0.08$) in yellow-bellied marmots (*Marmota flaviventris*, Blumstein et al. 2010). The heritability of behavioural traits was shown to be significantly smaller than the heritability of morphological traits (Stirling et al. 2002). These low heritabilities suggest that either strong selection has depleted genetic variation (low V_A) or the traits are very sensitive to environmental influences (high V_{PE}) (Bell 2005).

Recently, the heritability of personality (h^2P) was described by Dochtermann et al. (2015). The "heritability of personality refers strictly to the proportion of personality variation attributable to additive genetic variance" (Dochtermann et al. 2015, page 2). Mathematically, the additive genetic variance (V_A) is divided by the variance attributed to the individual (V_{ind}) as shown in the following equation (Dochtermann et al. 2015):

$$h^2P = \frac{V_A}{V_{ind}},$$

where V_{ind} is defined as the sum of the additive genetic variance (V_A), the variance due to maternal effects (V_M) and the permanent environmental variance (V_{PE}). As the heritability of personality can also be defined as the heritability of a behavioural trait divided by the repeatability of the same behavioural trait, it can be estimated whenever repeatability and heritability is available for the same data set (Dochtermann et al. 2015). Given this relationship, the heritability of personality traits was shown to be higher even if the behaviour itself was only weakly heritable (Dochtermann et al. 2015).

Evolution of Constant Between-Individual Behavioural Differences

Given that selection acts on the constancy of between-individual behavioural differences, and that individual behaviours are heritable, we expect the possibility of a response to selection, i.e. evolution. The response to selection was shown in laboratory experiments using selection lines of different behavioural types. Strong responses to the artificial selection were for instance shown in laboratory rodents: Van Oortmerssen and Bakker (1981) selected male house mice (*Mus musculus domesticus*) successfully for short and long attack latencies (SAL and LAL lines) over eleven generations. These selection lines were continued and showed strong differentiation in the selected attack latency response after 36 generations (Benus et al. 1991). Similar selection lines (e.g., for the active avoidance in a shuttle box) could be created in laboratory rats (see Benus et al. 1987 for review). Studies on wild populations showing a response to selection, however, are scarce. Drent et al. (2003) succeeded to breed selection lines of slow and fast explorers from wild-caught great tits (*Parus major*). The bi-directional artificial selection experiment showed strong effects after four generations, resulting in a mean score change of exploratory behaviour from 1.78 to -1.31 units per generation (Drent et al. 2003).

More recently, evidence increased that specific gene regions are involved in the maintenance of animal personalities (van Oers and Mueller 2010). Homologues of the dopamine receptor D4 gene (DRD4) were for example associated to exploratory behaviour in several species (e.g., great tits, *Parus major*, Fidler et al. 2007; zebra fish, *Danio rerio*, Boehmler et al. 2007; dog breeds, *Canis familiaris*, ITO et al. 2004). Van Oers and Mueller stated that “it is essential for our understanding of the causes and consequences of personality diversity to link phenotypic variation in personality traits with polymorphisms in genomic regions that code for this trait variation” (van Oers and Mueller 2010, page 3991). This is because phenotypic approaches, which only measure selection on personality traits, cannot reveal the evolutionary trajectories of those traits but evolutionary genomics can (van Oers and Mueller 2010).

II - Behavioural Syndromes: Between-Individual Correlations among Behavioural Traits

The second assumption in animal personality research is that different behaviours correlate with each other. As mentioned before, Sih et al. (2004a) defined behavioural syndromes as suits of correlated behaviours across two or more contexts or situations. Contexts were thereby defined as behavioural categories and a situation was defined as a specific set of conditions at a given time (Sih et al. 2004a). Sih et al. (2004a) specified that behavioural syndromes can involve correlations in (a) the same context, but different situations, in (b) different contexts within the same situation, or in (c) different contexts in different situations. Phenotypic correlations (behavioural correlations) can be partitioned into within-individual and between-individual correlations, and the latter actually define a behavioural syndrome (Dingemanse and Réale 2013). It has been shown in many (if not all) animal taxa that different behaviours form such behavioural syndromes. As already explained before, two main approaches can be found in the animal personality literature (***FIGURE 3***): In rodents, Koolhaas et al. (1999) termed a behavioural syndrome the PROACTIVE-REACTIVE SYNDROME, where proactive individuals were more active and more aggressive than reactive animals. In birds, the relation between different behavioural traits followed a fast-slow exploration continuum (FAST-SLOW SYNDROME, Titulaer et al. 2012). More exploratory great tits (*Parus major*) were for example bolder (Verbeek et al. 1994), more aggressive (Carere et al. 2005) and showed more risk-taking behaviours (van Oers et al. 2004b) than less explorative birds. Other terms such as *aggression syndrome*, *activity syndrome*, *shyness-boldness syndrome* can also been found in the literature (reviewed by Sih et al. 2004a).

Several mechanism may explain such phenotypic correlations. The CONSTRAINT HYPOTHESIS (see above) assumes that behavioural syndromes are difficult to break apart because of shared mechanisms underlying the different behaviours (Bell 2005). Behavioural traits can be connected to morphological traits which are relatively fixed. A common physiological or neuroendocrine basis may further result in behavioural correlations (Sih et al. 2004a). In house mice (*Mus musculus*), for instance, different behavioural types (coping styles to respond towards stress) also showed differences in their neuroendocrine profiles (Koolhaas et al. 1999). Other proximate mechanisms explaining behavioural syndromes are genetic effects. If behavioural traits are controlled by common genes, these behaviours cannot evolve independently (see below). Such constraints can result from pleiotropy and genetic linkage if behavioural correlations are in fact, at least in part, caused by underlying genetic correlations (Bergmüller 2010). Regardless of the proximate mechanism behind the correlations between behaviours, behavioural syndromes can result in maladaptive behaviour and conflicts, and they can generate trade-offs (Sih et al. 2004a). If prey species, for example, are active and bold, this may result in higher feeding rates compared to inactive and shy individuals in the absence of predators. When predators are present, however, less active and shy individual should be advantaged compared to active and bold animals because the latter face higher predation risks in such situations (Sih et al. 2004a).

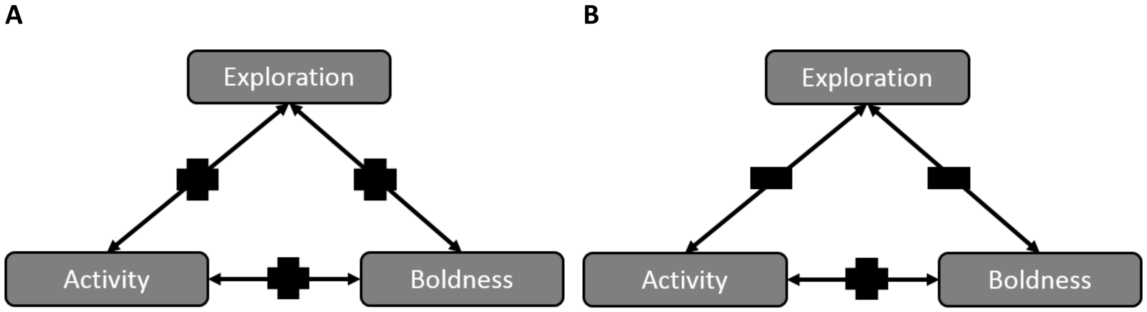

FIGURE 3 Two main approaches of behavioural syndromes in animal species. (A) The FAST-SLOW SYNDROME (Titulaer et al. 2012) defined by positive correlations between behaviours. (B) The PROACTIVE-REACTIVE SYNDROME (Koolhaas et al. 1999) defined by a positive correlation between activity and boldness, but negative correlations between exploration and activity, as well as between exploration and boldness.

Behavioural syndromes further result in restricted behavioural plasticity due to behavioural carryovers which induce less than optimal plasticity (Sih et al. 2004a, but see Bell and Stamps 2004). Limited plasticity is likely to be advantaged in variable environments because animals do unavoidable errors due to unpredictable environmental changes, and thus, exhibiting optimal plastic behaviour may be too costly compared to less plastic behaviour (Sih et al. 2004a). The ADAPTIVE HYPOTHESIS, as mentioned before, assumes that correlational selection favours certain combinations of behavioural traits and that behavioural syndromes are adaptive (Sih et al. 2004a; Bergmüller 2010). In contrast to the *constraint hypothesis* which assumes behavioural syndromes to be a general characteristic of a specific species due to fixed underlying proximate mechanisms, the *adaptive hypothesis* does not necessarily expect the same syndrome structures across different populations of that species (Bell 2005). This is because different behavioural correlations might be favoured in individuals of different selective environments (Bell 2005).

Selection for Behavioural Syndromes

Due to this adaptive reasoning, it has been argued that “when correlations between behaviours exist, they might do so because a particular combination of traits works well together” (Bell 2005, page 465). In this context, correlations between behaviours (behavioural syndrome) are hypothesised to be the result of selection, not because of a constraint on it (Bell 2005). *Correlational selection* could be a possible selective mechanism because it favours those certain combinations of behavioural traits, acting directly on the correlation between traits (Bergmüller 2010). That means that specific combinations of two or more behavioural traits covary with fitness (Bergmüller 2010). However, only one study could identify correlational selection in a field population of garter snakes (*Thamnophis ordinoides*). In this species, correlational selection acted on the relationship between colour morphs and antipredator behaviour (Brodie 1992). In the research field of behavioural syndromes, no correlational selection has been identified in animals yet (Bergmüller 2010, but see Eaves et al. (1990) for correlational selection detected in human personality).

The PHENOTYPIC SELECTION APPROACH developed by Lande and Arnold (1983) provides a framework to study the evolution of complex phenotypes such as personality traits and behavioural syndromes by measuring the covariance between fitness proxies and behaviours (Bergmüller 2010). Using this approach, several forms of selection could be identified as driving forces in the maintenance of animal personality traits and behavioural syndromes: *directional selection*, *disruptive selection,* and *stabilizing selection*. Empirical evidence, however, showed that heterogeneous forms of selection, i.e. *balancing selection* or *antagonistic selection*, are most likely the key processes maintaining animal personality and behavioural syndromes (Dingemanse and Réale 2013). As reviewed by Dingemanse and Réale (2013) temporal and spatial variation in selection pressures were detected to act on behaviours in different animal taxa (e.g., mammals, birds, and insects). Recently, a study on great tits (*Parus major*) for instance found, that "temporal variation in local density represented the primary factor explaining personality-related variation in viability selection" (Nicolaus et al. 2016, page 478). The interplay between fluctuating selection and the lack of adaptive behavioural plasticity in personality traits maintained animal personality variation in wild populations (Nicolaus et al. 2016). Further, the social environment (e.g., population density) was also shown to maintain animal personality and behavioural syndromes (Cote et al. 2008).

Genetically Correlated Behavioural Traits

As mentioned before, phenotypic correlations (behavioural correlations) can be partitioned into within-individual and between-individual correlations, and between-individual correlations are themselves affected by genetic and environmental effects (Dingemanse and Dochtermann 2013). Selection for behavioural types, however, will only result in the maintenance of these types in the population, if the between-individual correlations that underlie the behavioural syndrome are in fact genetic relationships. A genetic correlation (r_A) describes how much genetic components (e.g., genes) two or more (behavioural) traits share (Roff 1996). If there is a genetic correlation between traits ($r_A > 0$), this means that these traits are influenced by common genes. Genetic correlations between behavioural traits result in between-individual correlations because of gene pleiotropy or linkage disequilibrium, which means that single genes regulate the expression of multiple behaviours, or that genes affecting one behaviour are correlated with genes affecting another behavioural trait, respectively (Falconer et al. 1996; Han and Dingemanse 2015). Genetic correlations are independent of the heritability of each behavioural trait (Plomin et al. 2013). Thus, theoretically even if behavioural traits are not heritable, genetic correlations between behavioural traits can be observed. The genetic correlations are fundamental to understand the evolution of behavioural syndromes, because the evolutionary changes are constrained as selection on one trait influences also the response to selection of a genetically correlated trait (van Oers and Mueller 2010). Although, the existence of genetic correlations between behavioural traits can be a possible factor involved in the occurrence of a behavioural syndrome, evidence for such genetic correlations is still scarce in animal species (van Oers and Mueller 2010). Significant genetic correlations were, for instance, observed in great tits (*Parus major*) between risk-taking behaviour and exploration ($r_A = 0.84$, van Oers et al. 2004a), in three-spined sticklebacks (*Gasterosteus aculeatus*) between aggression and boldness ($r_A = 0.84$, Bell 2005); and in yellow-bellied marmots (*Marmota flaviventris*) between activity in an open field test and sociability ($r_A = 0.64$, Petelle et al. 2015).

Evolution of Behavioural Syndromes

An evolutionary consequence of the correlation between behaviours is that behavioural traits cannot evolve independently (Sih et al. 2004b). That means that selection favouring one behavioural trait, also leads to changes in a different behaviour (Stamps 1991). Given that selection acts on correlations between different constant individual behaviours, and that these relationships are genetically based, we would expect a response to selection (see above). Thus, understanding the evolution of correlated behavioural traits requires to estimate the underlying genetic correlations and to reveal the underlying genetic architecture (Karlsson Green et al. 2016). But although the genetic architecture of behavioural syndromes has gained more attention recently, the evolution of behavioural syndromes has been relatively little investigated in natural environments (Karlsson Green et al. 2016).

Results of empirical studies provided evidence for different evolutionary responses towards different selection pressures in different populations and environments. Evidence for the evolution of behavioural syndromes was for instance shown in different population of three-spined sticklebacks (*Gasterosteus aculeatus*): Phenotypic and genetic correlations between behaviours were found in one population of sticklebacks but not in another (Bell 2005). A correlation between aggression and boldness was only found in the population with high predation pressure, but this correlation was not present in the population with low predation risk (Bell 2005). Similar results were found by Dingemanse et al. (2007), who investigated behavioural syndromes in twelve stickleback populations in the United Kingdom. They observed that "the often-documented syndrome between aggressiveness, activity and exploratory behaviour existed only in large ponds where piscivorous predators were present. In small ponds where predators were absent, these behaviours were not (or only weakly) associated." (Dingemanse et al. 2007, page 1128). The authors concluded that behavioural syndromes found in sticklebacks did not act as evolutionary constraints, but that rather natural selection favoured the evolution of behavioural correlations in certain environments depending on the predation pressure in sticklebacks (Bell 2005; Dingemanse et al. 2007). More recently, Fischer et al. (2016) compared behavioural syndromes in different populations of Trinidadian guppies (*Poecilia reticulate*) from low- and high-predation environments. They observed that behavioural variance increased when fish were reared under non-native environmental conditions compared to fish reared in native conditions. Further, behavioural correlations differed between the groups depending on the rearing condition and genetic background. The evolvability (i.e. the capacity for adaptive evolution) was found to increase under non-native environmental conditions (Fischer et al. 2016). The authors concluded that the behavioural syndrome found in Trinidadian guppies is plastic and can evolve differently between populations from different environments (Fischer et al. 2016). A rapid response to selection was also observed by Karlsson Green et al. (2016) in freshwater isopods (*Asellus aquaticus*). In this species, a novel ecotype has diverged after the colonization of a new habitat in Swedish lakes (Karlsson Green et al. 2016). In a common garden experiment, the authors found that genetic correlations among behaviours differed between the two ecotypes (Karlsson Green et al. 2016). They concluded that "genetic correlations behind behavioural traits have become decoupled and differentiated during a very short ecological and evolutionary time frame, corresponding to 40-50 isopod generations" (Karlsson Green et al. 2016, page 150).

Part 2 - Introduction to Animal Cognition

In the second part of the introduction, I introduce to animal cognition: What is animal cognition? Why are we interested in cognitive abilities of animals? I will further give an overview about spatial cognition in animals followed by an introduction to individual differences in cognitive traits. In the last paragraph, the theoretical concept of cognitive syndromes will be explained providing a connection to the first part of the introduction - animal personality.

In general, COGNITION can be defined as mechanisms how individuals (humans and animals) acquire, process, store, and use information from the environment (Shettleworth 2010). On the one hand, cognition includes all physiological and neuronal processes involved in the processing and storage of perceived information. This can include mechanisms of perception in the sensory cells, the release of neurotransmitters and further processing of information in the brain (Shettleworth 2010). On the other hand, the use of information also results in a behavioural reaction reflecting these cognitive processes. That means that cognition results in behaviour and natural selection acts on cognition indirectly through the medium behaviour (Shettleworth 2010). Four domains can be separated within the field of cognition: perception, learning, memory, and decision-making (Shettleworth 2010).

Studying Cognition in Animals

The research field of ANIMAL COGNITION has not been recognized until the 1970s (Shettleworth 2010). It is often also referred to as COMPARATIVE COGNITION because human psychologist in the beginning usually compared the mental characteristics of humans to those of non-human species (Shettleworth 2010). Studying animal cognition implies the one fundamental problem that we cannot investigate consciousness in animals. This is because consciousness is a subjective state and animals cannot tell us whether they experience something consciously or not (Shettleworth 2010). Thus, investigation on animal cognitive abilities rely on changes in the behaviour which can be interpreted as learning or memory. For instant, if we observe the behaviour of an animal in a Y Maze, we would expect that all three arms of this arena are equally visited. However, if we block one of the three arms of the Y Maze during the first of two trials, animals usually spend more time in that specific arm once it is opened in the second trial. This arm is new to them, as they were not able to explore it before. This behavioural reaction of the animals is generally interpreted as spatial recognition of an unknown environment (e.g., Dellu et al. 1992). Thus, we measure a cognitive trait as the change in the expression of an observable behavioural trait.

Spatial Cognition in Animals

Spatial orientation is a very important cognitive trait for all mobile animals as they face the fundamental challenge of not getting lost when they move around to look for food, homes or mating partners. SPATIAL COGNITION can be defined as how animals acquire, process, store, and use *spatial* information from the environment (Shettleworth 2010). This means that individuals should be able to put their own position in relation to the position of others or of cues in their environment. Spatial cognitive processes are assumed to rely on different computational requirements than other types of learning (e.g., associative learning, Dyer 1998). Several mechanisms were identified to be used by different animal species for spatial orientation. For instance, DEAD RECKONING, also known as PATH INTEGRATION, can be defined as an internal sense of distance and direction of a known place compared to the own current position (Shettleworth 2010). In gerbils (*Meriones unguiculatus*) for example, Mittelstaedt and Mittelstaedt (1980) discovered that mother gerbils returned straight to the original location of their nest after searching for a hidden pup, even when the position of the nest had been changed while the female was picking up her offspring. As the experiment was conducted in darkness, no visual cues were available. The authors concluded that females homed by “integrating azimuthal information from rotatory accelerometers over an idiothetic variable which is proportional to the progress of the animal along its path” (Mittelstaedt and Mittelstaedt 1980, page 566). In golden hamsters (*Mesocricetus auratus*), Etienne and Jeffery (2004) likewise discovered that animals used path integration during homing (movement of retouring to their nest/home). Hamsters did hoarding excursions while the experimental room was illuminated by infrared light. At food patches they filled their pouches and then returned directly to their starting point (Etienne and Jeffery 2004). These mechanisms involve egocentric spatial localization which means that objects in the environment (e.g., nests or food patches) are localized with respect to the animal itself (Shettleworth 2010). It was shown that the vestibular system plays an important role for path integration (Wallace et al. 2002). The vestibular system is part of the sensory system in mammals and provides information for balance and spatial orientation. It is located in the inner ear of mammals. As movements consist of rotations and translations, the vestibular system comprises two components: Rotational movements and linear accelerations. This is especially important for nocturnal species which cannot as much rely on visual cues as diurnal species do (Shettleworth 2010).

The opposite of egocentric spatial localization are *allocentric* mechanisms which use environmental geometry or cues in the environment as references (Shettleworth 2010). Two types of cues can be distinguished - BEACONS and LANDMARKS. Let us clarify the differences of these cue types by means of a fundamental cognitive task used in animal cognition. This task is the *Morris Water Maze* (*MWM*) established by Morris (1981). During the task animals (mostly rats and mice) are supposed to learn the location of a platform in a water tank. Animals are released into the water and have to swim searching for the platform where they can stand on and rest. Usually several trials are conducted until the animal has learned the position of this platform. Spatial memory can also be tested by repeating trials with delay of minutes or hours. Very different modifications of the MWM have been used (reviewed by D'Hooge and De Deyn 2001). In all applications, the platform in the MWM function as a BEACON - also called proximal cue - when it is visible for the animal. In this kind of set-up, animals learn that the visible platform is the position of the save area in the water, where they can rest from swimming. In contrast, the platform can also be under the water surface and usually the water is made opaque so that animals have to learn the position of the platform (safe resting area) via symbols attached to the wall of the swimming pool. These symbols serve as LANDMARKS - also called distal cues - when the platform is not visible for the animals. Landmarks are defined as fixed objects with fixed location (cues) that guide animals to their goal position (Shettleworth 2010). Spatial orientation relates on the position of the animal in relation to the landmarks in these cases. It was shown that spatial learning based on beacons involves a different cognitive processing than spatial learning based on landmarks (White and McDonald 2002). The hippocampus is the important brain region of vertebrates involved in the processing of spatial information about the environment in general (Yaskin 2011). It is a well-defined brain region which is c-shaped in most mammals and located around the thalamus (Dukas 2009). Prominent examples in food-storing and non-storing birds showed that the hippocampus volume was larger in species which did store food (Biegler et al. 2001). Further, lesions of the hippocampus induced the loss of spatial orientation ability and spatial memory function in rats (e.g., Stubley-Weatherly et al. 1996). Taken together, this indicates the importance of the hippocampus in spatial cognition in animals.

Between-Individual Differences in Cognitive Traits

Coming back to the Morris Water Maze described before, the MWM is a famous example and commonly used in cognitive, medical, and may also be suitable for behavioural ecology studies. The research field of animal cognition deals with central questions on processes that allow animals to solve a specific task (e.g., how they find the platform). Medical studies focus on the effects of drugs or diseases on the learning or memory capacity by comparing behaviour between treatment and control groups (e.g., under drug treatment, the platform is not found anymore). Among behavioural ecologists, however, there is growing interest in the individuality of task performance. As reviewed by Thornton and Lukas (2012), individual cognitive variation was observed in many animal cognition studies. However, this variation was often ignored or interpreted as noise around the mean of a population (Thornton and Lukas 2012). This history is in common with constant between-individual behavioural variation described in the first part of the introduction. First evidence for individual differences in cognitive traits was explicitly documented in biomedical studies which quantified cognitive variation between genetic strains of rodents (e.g., Walker and Mason 2011; Sweis et al. 2013). Recently, there is growing evidence for these between-individual differences in cognitive traits, and quantifying individual cognitive variation is one of the recent challenges in behavioural ecology (Croston et al. 2015). Individual differences in cognitive traits were shown, for instance, in birds (e.g., Guillette et al. 2015), mammals (e.g., Matzel et al. 2003; Guenther et al. 2014a), and fish (e.g., DePasquale et al. 2014; Mamuneas et al. 2015). Specifically, there is also growing evidence for between-individual differences in spatial cognitive traits (e.g., Carazo et al. 2014).

However, only a handful of studies investigated the repeatability or consistency of individual cognitive differences (Griffin et al. 2015). For instance, Wang et al. (2015) showed that zebrafish (*Danio rerio*) decided repeatably in a colour discrimination task. In Carib grackles (*Quiscalus lugubris*), birds performed consistently well over two problem-solving tasks (Ducatez et al. 2015). Also, black-capped chickadees (*Poecile atricapillus*) reached a learning criterion consistently fast over two different set-ups in a colour association task (Guillette et al. 2015).

Selection, Heritability and Evolution of Individual Differences in Cognitive Traits

As the interest in and evidence for individual variation in cognitive traits has just recently increased (Thornton and Lukas 2012), there are no theories explaining this variation yet. We may hypothesize that mechanisms maintaining the individual variation in cognitive traits are similar to the ones explaining between-individual behavioural variation (see Part 1). Cognitive traits can be the basis for constant between-individual differences in behaviour (Sih et al. 2004a). This connection may result in non-independent evolution of behaviours and related cognitive traits. Further, cognitive traits can be connected to morphological or physiological traits which are relatively fixed. Larger hippocampus sizes were, for instance, correlated with better spatial memory in birds (Pravosudov and Clayton 2002). If this is the case, the *constraint hypothesis* assumes that correlations between those traits and cognitive traits are difficult to break apart (Bell 2005). Thus, indirect selection may maintain variation in cognitive traits. On the other hand, it was assumed that natural selection acts on cognitive traits which produces changes in specific associated brain regions resulting in the evolution of cognitive traits and neuronal structures (Croston et al. 2015). However, “direct evidence for the role of natural selection in driving the evolution of cognitive traits and their neural mechanisms remains elusive” (Croston et al. 2015, page 1447). Nevertheless, the selection mechanisms resulting in variation in cognitive traits may be similar to the ones resulting in variation in behavioural traits (selective mechanisms explained in details in Part 1).

Studying natural selection of animal cognition relies on the fundamental condition that cognitive trait variation is associated with differences in individual fitness (Dukas 2004). Different fitness proxies were recently shown to correlate with individual variation in cognitive traits in wild populations (see review by Morand-Ferron et al. 2015). In grasshoppers (*Schistocera americana*), for example, individuals which learned an association between spatial cues and a food reward, had a significant higher growth rate than animals without this learning experience (Dukas and Bernays 2000). However, no study has directly related individual cognitive variation to “reproductive fitness” (Thornton and Lukas 2012). There are no clear predictions whether or not cognition correlates with fitness because cognition is no unitary trait or usually not clearly defined (Rowe and Healy 2014). It is not expected to find fitness benefits for “smarter” individuals as there is probably more than one way to be smart (Rowe and Healy 2014). Defining the “optimal” or “best” cognitive ability for a specific species in specific environmental conditions would be the critical prerequisite for further investigations into the effect of cognitive variation on fitness. For example, three-spined sticklebacks (*Gasterosteus aculeatus*) from fast-flowing rivers and ponds were trained to locate a hidden reward in a T-maze (Odling-Smee and Braithwaite 2003). Fish from the pond populations appeared to rely more on visual landmarks than fish from the river populations (Odling-Smee and Braithwaite 2003). The authors concluded that “landmarks may be reliable indicators of location only in stable pond habitats. In rivers, turbulence and flow may continually disrupt the visual landscape such that river fish may benefit from learning orientation routes only if learning is constrained so that unreliable visual cues are ignored.” (Odling-Smee and Braithwaite 2003, page 701). Another study showed adaptive variation of problem-solving behaviour in great tits (*Parus major*): Cole and Quinn (2012) observed that problem-solving was negatively correlated with the ability to compete over food resources, which may indicate an indirect measure of fitness. They hypothesised that poor competitors may compensate their disadvantage through better cognitive abilities (e.g., problem-solving) as a different strategy (*cognitive styles*, see below, Cole and Quinn 2012).

Cognitive traits - like all phenotypic traits- can only evolve if there is genetically based variation in traits among individuals, and if traits are heritable (Dukas 2004). General cognitive ability - suits of correlated cognitive abilities measured in different cognitive tasks (Plomin 1999) - was shown to be highly heritable in humans (Croston et al. 2015). There is also growing evidence for a genetic contribution to individual cognitive variation in non-human animals (Dukas 2004). As reviewed by Croston et al. (2015), the general learning ability had a heritable component of 40 % and 53 % in house mice (*Mus musculus*) and chimpanzees (*Pan trogolodytes*), respectively.

Using artificial selection lines, it was shown in different animal species that selection resulted in divergent changes in learning abilities of the different selection lines (e.g., spatial learning in rats, reviewed by Dukas 2004), or associative learning in fruit flies (*Drosophila melanogaster*, Lofdahl et al. 1992; Mery and Kawecki 2002). However, examples for evolved cognitive abilities in the wild are scarce. One famous example is the difference in spatial memory between food-storing and non-storing birds: After, it has been shown that food-storing birds have a larger hippocampus than non-food-storing species (Krebs et al. 1989), it was also observed that food-storers (coal tits, *Parus ater*) were able to remember locations longer that non-storers (great tits, *Parus major*). However, these two species did not differ in memory resolution and memory capacity (Biegler et al. 2001). Individual differences in spatial memory was found by a within-species comparison among different populations of black-capped chickadees (*Poecile atricapilla*, Pravosudov and Clayton 2002), giving evidence for different selection responses of the spatial memory function in different populations depending on the environmental conditions.

Recently, Morand-Ferron et al. (2015) provided a framework to study the evolutionary ecology of cognitive traits. They suggested a six-step approach (c.f. Figure 2 in Morand-Ferron et al. 2015) to link cognitive traits of interest to fitness proxies while identifying confounding effects. Thereby, multivariate analyses should be used to test for selection. Further, the repeatability and heritability of cognitive traits should be assessed. In the end, the overall response to selection, i.e. evolution of the cognitive traits, can be identified based on the current selection pressure and trait variation (Morand-Ferron et al. 2015).

Cognitive Syndrome

Sih and Del Giudice (2012) introduced the term COGNITIVE SYNDROME based on the hypothesis that behavioural types are linked to cognition. They assume that cognitive styles can evolve, if adaptive cognitive behaviour can be achieved through different strategies, or if animals face constant trade-offs when solving cognitive tasks. A cognitive syndrome is defined as consistent between-individual differences in cognitive styles which are related to behavioural types within a population (Sih and Del Giudice 2012). *COGNITIVE STYLES* are thereby defined as strategies *how* individuals acquire, process, store and remember information and which are constant across time and contexts (Gruszka et al. 2010). Consistent cognitive styles are for example expected to arise due to a speed-accuracy trade-off individuals face during decision-making, which can be involved in any cognitive context (e.g., learning and memory). When an individual has to make a decision it may face the fundamental problem of speed-accuracy trade-offs (Chittka et al. 2009). Individuals can either decide fast at the cost of an accurate decision or take their time and decide more accurately (Burns and Rodd 2008). Erroneously, accuracy has been interpreted in the past as a limit to the cognitive ability of an individual (Chittka et al. 2009), but recent data showed that both cognitive styles (fast vs. accurate) can occur in a population side by side (Wang et al. 2015), indicating that both styles might be similar adaptive in specific environmental conditions. Within a cognitive syndrome fast behavioural types (consistently bolder, more aggressive and more active individuals) are expected to show faster decision or learning styles. Slow behavioural types in contrast are assumed to decide or learn less fast but more accurately (Sih and Del Giudice 2012). This is due to a risk-reward trade-off, where fast animals take higher risks while being rewarded faster and slow animals decide or learn more accurate but less fast and take less risks (***FIGURE 4***). Given this, risk-reward trade-offs are expected to provide one potential link between behavioural types and cognitive styles (Sih and Del Giudice 2012, ***FIGURE 4***).

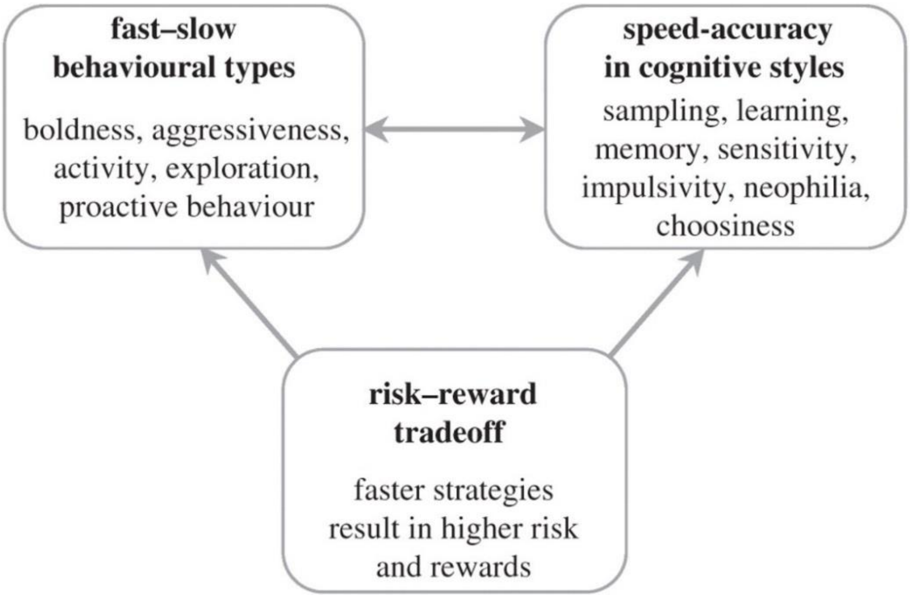

FIGURE 4 The concept of cognitive syndromes proving a possible link between behavioural types and cognitive styles through a risk-reward trade-off. With permission (License Number 3845950396542) from Sih and Del Giudice (2012).

Like behavioural types, cognitive styles are assumed to show temporal repeatability and cross-context consistency (Sih and Del Giudice 2012). This is because behavioural and cognitive syndromes rely on the assumption that individuals constantly differ in their behavioural trait expressions. The rank order of individuals is assumed to be stable between situations, even if the mean behavioural trait expression might differ between tests (Sih et al. 2004b). Based on this, several assumptions should be tested: As for personality traits, constant between-individual differences are assumed for cognitive traits (see above). Thus, repeatability and consistency of those traits should be investigated. Further, as the cognitive syndrome is expected to arise from a risk-reward trade-off (Sih and Del Giudice 2012), cognitive styles could be based on a speed-accuracy trade-off during decision making. Experimental set-ups should thus test for a speed-accuracy trade-off within individuals.

Although, there is growing evidence that behavioural types are related to cognitive styles, studies demonstrating the prerequisite of cognitive syndromes - cognitive traits are constant across time or contexts - are less evident (Griffin et al. 2015). For instance, Wang et al. (2015) showed that zebrafish (*Danio rerio*) decided repeatably in a colour discrimination task. In Carib grackles (*Quiscalus lugubris*), birds performed consistently over two problem-solving tasks (Ducatez et al. 2015). Also, black-capped chickadees (*Poecile atricapillus*) reached a learning criterion consistently over two different set-ups in a colour association task (Guillette et al. 2015). Furthermore, recent studies so far indicated contradictory results: Experimental work in black-capped chickadees (*Poecile atricapillus*) showed a cognitive syndrome, as slow exploring individuals showed higher accuracy levels during learning trials (Guillette et al. 2015). Guenther et al. (2014a) also showed that boldness, activity and aggressiveness were positively correlated to fast association learning in wild cavies (*Cavia aperea*). On the contrary, Bousquet et al. (2015) could not identify any effect of exploration on the accuracy in a spatial learning task in mallards (*Anas platyrhynchos*). Further, Mamuneas et al. (2015) could also not show any differences in the accuracy of decision making in shy and bold three-spined sticklebacks (*Gasterosteus aculeatus*). In guppies, bold females learned faster and also more accurately (Trompf and Brown 2014). Hence, this species did not seem to underlie a speed-accuracy trade-off.

Although repeatability and consistency have been studied thoroughly for personality traits (Bell et al. 2009; Brommer and Class 2015), so far only few tests for those assumptions of cognitive traits have been published. Given the lack of deeper theoretical theories and the lack of available data from empirical studies, more investigations are needed to better understand the pattern of cognitive syndromes in animal species. Especially, evolutionary studies would be important linking cognitive styles to fitness proxies, and investigating the response to selection, i.e. evolution of the cognitive styles (Morand-Ferron et al. 2015). Thereby, genetic components and heritability of cognitive traits should be assessed.

Aim of the Thesis

Objectives of this Thesis

The aim of my dissertation was to provide evidence for constant between-individual differences in behaviour and spatial cognition of Eurasian harvest mice (*Micromys minutus*; Pallas, 1771). I further intended to identify a possible relationship between spatial cognitive abilities and personality traits in this species.

The Eurasian harvest mouse (see ***Figure 5***) is a small rodent species occupying a very different ecological niche compared to other small rodents in Europe. It lives mainly above the ground in high grass vegetation (Piechocki 2001).

Figure 5 Picture of my study species – the Eurasian harvest mouse – in the laboratory (A) and in the semi-natural outdoor enclosure of the Eberhard Karls University Tübingen (B). Photos by Andrea C. Schuster

Harvest mice have not been in the focus of animal personality research yet. However, this small rodent species is likely to be highly suitable for animal personality and especially spatial cognitive syndrome studies for the following reasons: (I) Harvest mice are easy to keep and breed in the laboratory, (II) behavioural and cognitive tests can easily be established based on tasks done with laboratory mice and rats, (III) harvest mice are assumed to be selected for pronounced spatial cognitive traits due to their adaptation to three-dimensional habitat structures (Piechocki 2001), and (IV) harvest mice are a widespread species in the Palaearctic but locally endangered (Darinot 2016), so that more knowledge about their behavioural ecology might provide helpful insights for their conservation. Therefore, the harvest mouse was a perfect study organism for my research questions.

I tested the two assumptions of animal personality as described before - repeatability and consistency, as well as correlations between behavioural traits. Further, I investigated if spatial cognitive abilities are part of a cognitive syndrome in harvest mice. Behavioural tests were conducted on two study populations: The laboratory population and the field population living in a semi-natural outdoor enclosure. Since 2010, harvest mice have been kept in the laboratory at the Eberhard Karls University Tübingen. This laboratory population originated from animals that were obtained from breeders. Further, I captured wild harvest mice in Potsdam in 2013 and released them into a semi-natural outdoor enclosure in Tübingen. The population was able to freely reproduce and was exposed to avian predation. I monitored the population between 2013 and 2016 by regular trapping and nest surveys.

I applied my research questions in both populations and compared the results. To do so, suitable behavioural tests needed to be established for my study species. Behavioural tests thereby should fulfil the two following prerequisites: First, it was important to observe as much behavioural variation as possible in the tested populations. This was because the estimation of repeatability and consistency relies on the within-individual behavioural variance compared to the between-individual behavioural variance in a population (see page 7). Secondly, behavioural tests should be relative fast as it was important to test many individuals to achieve large sample sizes and therefore sufficient statistical power. Standard behavioural tests, originally established to test emotionality in laboratory strains of mice and rats, were modified and adapted to the smaller body size of my study species. I conducted a modified Open Field test (OF, Archer 1973), a Novel Object test (NO, Chitty and Shorten 1946), and a Y Maze (YM, Montgomery 1955) to phenotype activity, boldness, exploration, and spatial orientation abilities (see page 42 for detailed test descriptions).

As Eurasian harvest mice occupy this very different ecological niche compared to other rodent species commonly studied in animal personality, I further established behavioural tests which fitted to their ecology. I designed a Novel Environment (NE) test based on the idea of Verbeek et al. (1994) who analysed exploration in great tits (*Parus major*). Harvest mice were able to climb within the set-up of the designed Novel Environment test (see test description on page 64) which encouraged their behavioural repertoire. The Spatial Orientation Task (SOT) was also established (by Uwe Zimmermann under my supervision of his Bachelor thesis) based on the idea that harvest mice prefer climbing instead of walking on plain plastic. To estimate spatial orientation and spatial learning abilities of harvest mice, animals were released in an arena with an elevated six arm maze and they were supposed to learn the location of a target box in which mice could hide (see test description on page 66).

TABLE 2 Chronology of behavioural experiments conducted in a laboratory and an enclosure population of harvest mice. Behavioural test: Y Maze (YM), Open Field (OF), Novel Object (NO), Scare Test (ST), Novel Environment (NE), Spatial Orientation Task (SOT). * indicates that behavioural tests were conducted with the help of student helpers and Bachelor candidates. Focus of the analysis and referring chapters within the thesis are mentioned.

Month/year	Population	Behavioural test	Analysis	Chapter
06-08/2013	Laboratory	YM, OF, NO	Repeatability, behavioural and cognitive syndrome	2, 3
06/2013 - 05/2014	Laboratory *	ST	Repeatability, behavioural and cognitive syndrome	2, 3
08-10/2013	Enclosure	YM, OF, NO	Repeatability, behavioural and cognitive syndrome	3
08/2013 - 11/2014	Laboratory *	YM, OF, NO	Repeatability and consistency, behavioural and cognitive syndrome	1, 2, 3
03-11/2014	Enclosure	YM, OF, NO	Repeatability, behavioural and cognitive syndrome	3
04/2014 - 02/2015	Laboratory *	SOT	Repeatability, behavioural and cognitive syndrome	2
09-12/2014	Laboratory *	NE	Repeatability, behavioural and cognitive syndrome	2, 3
04-10/2015	Enclosure*	YM, OF, NO, NE	Repeatability, behavioural and cognitive syndrome	3

CHAPTER 1 is based on a manuscript by **Andrea C. Schuster**, Teresa Carl & Katharina Foerster entitled "Repeatability and consistency of individual behaviour in juvenile and adult Eurasian harvest mice" (May 2017: published; *Andrea C. Schuster, Teresa Carl & Katharina Foerster in The Science of Nature (2017) 104:10*). ACS designed the experimental set-up and did the statistical analyses. ACS and TC conducted the behavioural tests. ACS and KF wrote the manuscript. In CHAPTER 1, we aimed to identify repeatable and consistent behavioural and cognitive traits. For this purpose, harvest mice were behaviourally phenotyped (using three different behavioural tests, see ***TABLE 2***) at four different time points during their life. Two observations were done

during the juvenile phase and two behavioural tests were conducted during their adulthood. Thus, it was possible to analyse the repeatability of behavioural traits (two observations within one life-history phase) and the consistency of behavioural traits (two observation among different life-history phases). We expected to find repeatable behaviour in juveniles, as well as in adult harvest mice. Since activity was the most consistent behaviour in other rodents, we expected activity to be consistent across life history stages in our study species. In contrast, we expected that boldness was not consistent across life history stages. We also expected that exploration was not consistent as shown in previous studies of rodent personality. Furthermore, no consistency in spatial recognition was assumed, as the motivation for and the experience in spatial orientation was likely to change over lifetime.

CHAPTER 2 is based on a manuscript by **Andrea C. Schuster**, Uwe Zimmermann, Carina Hauer & Katharina Foerster entitled "A behavioural syndrome, but less evidence for a cognitive syndrome in a spatial orientation context" (May 2017: published; *Andrea C. Schuster, Uwe Zimmermann, Carina Hauer & Katharina Foerster in Frontiers in Zoology (2017) 14:19*). ACS designed the experiments and did the statistical analyses. ACS, UZ and CH conducted the behavioural tests. ACS and KF wrote the manuscript. In CHAPTER 2, we investigated if behavioural traits (activity, boldness, exploration, spatial recognition, spatial learning ability, decision speed, and decision accuracy) tested in adult laboratory harvest mice correlate with each other. As defined by behavioural and cognitive syndromes, we expected correlations between the traits measured in six behavioural tests (see ***TABLE 2***). Further, we analysed if harvest mice faced a speed-accuracy trade-off as assumed in the concept of a cognitive syndrome (see ***FIGURE 4***). We expected that all behavioural traits were repeatable in adult harvest mice as we already could show this before (cf. Chapter 1, page 33). Further, we hypothesised that boldness, activity and exploration were positively correlated and form a *fast-slow-behavioural syndrome* (Titulaer et al. 2012). We assumed that mice with better spatial recognition would learn an orientation task quicker due to improved spatial cognitive abilities. We further predicted that spatial recognition and spatial learning ability were correlated to personality traits. As our cognitive test design was based on active exploration, we assumed a positive relationship between active personality types and spatial learning ability (Sih and Del Giudice 2012). Finally, we expected a speed-accuracy trade-off as suggested by Sih and Del Giudice (2012) and hypothesised that bolder, more active and more explorative individuals decided faster but less accurately in a spatial learning task.

CHAPTER 3 is based on a manuscript by **Andrea C. Schuster**, Annika Luczak & Katharina Foerster entitled "Constant individual behaviours and phenotypic correlations in the field – Comparison between laboratory and field population of Eurasian harvest mice (*Micromys minutus*)" (May 2017: not submitted yet). ACS designed the experimental set-up and did the statistical analyses. ACS and AL conducted the behavioural tests. ACS and KF wrote the manuscript. In CHAPTER 3, we compared the patterns of constant between-individual differences between a captive and a field population of harvest mice. All behavioural test used in Chapter 1 and 2 were also conducted with the animals of the field population living in a semi-natural enclosure. We first tested for the repeatability of three personality and one cognitive trait in the field population. As we could show repeatable and consistent behaviours in harvest mice from the laboratory population before (Chapter 1), we expected behavioural traits to be repeatable also in the field individuals. We further investigated if tested traits (boldness, activity, exploration and spatial recognition) correlated positively with each other forming a behavioural syndrome (Sih et al. 2004b) in the field population, and we compared observed correlations to the results of our study previously done in the laboratory population of harvest mice (Chapter 2). Thereby, we aimed to confirm positive relationships between behaviours in the field population as shown before in the laboratory population (Chapter 2), and as assumed in a *fast-slow-behavioural syndrome* (Titulaer et al. 2012).

Chapter 1

In Chapter 1, we provide evidence concerning the first assumption of animal personality – repeatability and consistency of behaviours – tested in Eurasian harvest mice of a laboratory population:

Knowledge on animal personality has provided new insights into evolutionary biology and animal ecology, as behavioural types have been shown to affect fitness. Animal personality is characterized by repeatable and consistent between-individual behavioural differences throughout time and across different situations. Repeatability is often interpreted as a guarantor of consistency, and both properties of animal personality are assumed to be independent of sex and age. However, recent data has shown that males and females in some species may differ in the repeatability of behavioural traits, as well as in their consistency between different life history stages. We measured the repeatability and consistency of three behavioural and one cognitive traits in juvenile and adult Eurasian harvest mice (*Micromys minutus*). We found that exploration, activity and boldness were repeatable in juveniles and adults. Spatial recognition measured in a Y Maze was only repeatable in adult mice. Exploration, activity and boldness were consistent before and after maturation, as well as before and after first sexual contact. Data on spatial recognition provided little evidence for consistency. Further, we found some evidence for a litter effect on behaviours by comparing different linear mixed models. We concluded that harvest mice express repeatable and consistent personality traits. The tested cognitive trait showed low repeatability and was less consistent across life history stages. Given the rising interest in individual variation in cognitive performance, and in its relationship to animal personality, we suggest that it is important to gather more data on the repeatability and consistency of cognitive traits.

Behavioural Repeatability and Consistency in Harvest Mice

Behavioural Repeatability and Consistency in Harvest Mice

Over the last decades, the interest in individual variation in behaviour within animal species has increased rapidly. Constant between-individual differences in behaviours, known as animal personality, coping styles, or behavioural syndromes (Koolhaas et al. 1999; Sih et al. 2004a; Réale et al. 2007), have been analysed in all main animal taxa until today. This is due to the relevance of animal personality as an important component of animal ecology, and its effects on fitness (Thomas et al. 2016). In wild populations, animal personality could be linked to differences in reproductive success (Réale et al. 2000; Both et al. 2005), parental care (Budaev et al. 1999), survival (Dingemanse et al. 2004; Boon et al. 2007), and even to different life history strategies (e.g., Vetter et al. 2016). Animal personality is specified as constant between-individual behavioural differences throughout time and across situations (Réale et al. 2007). In many species, individuals behave repeatably when tested in the same behavioural test more than once (reviewed by Bell et al. 2009).

Repeatability and Consistency of Animal Behaviours

In general, the term repeatability is used when assessing the accuracy of specific measurements (Nakagawa and Schielzeth 2010). In animal personality research, repeatability specifies the proportion of between-individual variance relative to the total phenotypic variance in a population for repeated measures of the same behaviour (Dingemanse et al. 2010). It can be interpreted as the expected correlation between repeated measurements of a behaviour of the same individual. The repeatability of a specific behaviour is usually calculated based on repeated observations in the same context and life history phase. Repeatability estimates from observations across contexts or life history-phases have also been reported as 'repeatability', but the alternative terms cross-context repeatability (e.g., Laskowski and Bell 2013) or consistency (e.g., Wuerz and Krüger 2015) allow us to better specify the different information content of these estimates. The proportion of between-individual variance among measurements from different contexts or life history phases characterises the stability of individual behaviour across a longer time interval or across changes in the environment. Rank correlations of individual behaviour across contexts can further indicate consistent individual behaviour measured in different situations (Brommer and Class 2015). However, while rank correlations report stable or changing rank orders of mean individual behavioural performance within a population, they lack information on the relative contribution of repeatable individual performances to the total population phenotypic variance. We here restrict the use of the term *repeatability* to estimates of the proportion of between-individual variance from observations within the same life history phase. We define *consistency* of behavioural traits as the proportion of between-individual variance from observations at different ontogenetic stages. We use the term personality trait for behavioural traits that are repeatable and consistent and can hence contribute to animal personality. Behavioural types are characterized by each individual combination of several behavioural trait levels (Bell 2007).

There is growing evidence that repeatable personality traits and behavioural syndromes can be less stable across lifetimes than previously assumed (Wuerz and Krüger 2015; Fischer et al. 2016, but see Gyuris et al. 2012). Developmental changes could reveal underlying mechanisms of animal personalities (Stamps and Groothuis 2010b; Trillmich and Hudson 2011) as different experiences during development can shape behavioural types. Also, the hormonal constitution undergoes considerable change during the phase of vertebrate maturation, and this is likely to influence individual behavioural consistency (Stamps and Groothuis 2010a). Bell et al. (2009) already suggested to analyse repeatable behaviours between different age classes within species to investigate the potential impact of age and individual (sexual) experience on the repeatability and consistency of behavioural traits. Although their meta-analysis could not detect any differences in the repeatability of behaviours between juveniles and adults (Bell et al. 2009), a recent review by Brommer and Class (2015) reported evidence for lower behavioural repeatability in juveniles. Changes in behavioural repeatability and consistency across the life history do not contradict the existence of a behavioural syndrome. They rather highlight the necessity to investigate personality traits across different life history phases, to fully understand when and how animal personality arises and how long it persists. Recently, experiments have in fact shown that animal personality (based on repeatable behaviours) can arise during ontogeny (Polverino et al. 2016), but that it can also undergo a senescent decline in the wild (Class and Brommer 2016). Thus, understanding how repeatability (within life history stages) and consistency (between life history stages) differ between different developmental stages within a species is an important first step towards the definition and investigation of a behavioural syndrome.

Behavioural traits usually investigated in personality research are boldness (the tendency of an individual to take risks), activity (the general activity level of an individual), and exploration (an individual's reaction to a new situation, for instance, novel objects or novel environments; see also Réale et al. 2007). Exploration was a consistent trait over a significant part of an individual's lifetime in great tits (*Parus major*, Dingemanse et al. 2002) and zebra finches (*Taeniopygia guttata*, David et al. 2012). But while boldness, exploration and activity were repeatable behavioural traits over short periods of time in small rodents (e.g., Koolhaas et al. 1999; Boon et al. 2007; Lantová et al. 2011; Petelle et al. 2013), only activity seemed to be consistent across life history stages in that group (e.g., Kanda et al. 2012; Herde and Eccard 2013, but see Guenther et al. 2014).

Behavioural repeatability within, but no consistency across life history phases can arise from developmental changes (see above). However, a simulation study showed that life history trade-offs can promote the evolution of behavioural types that are specific to particular life history phases (Wolf et al. 2007). Empirical data provided evidence that the level of boldness can vary between individuals depending on expected future reproductive success: In grey mouse lemurs

(*Microcebus murinus*), young males had low current but high expected future reproductive success, while the opposite was true in older males (Dammhahn 2012). Hence, the trade-off between the investment into current and future reproduction differed between age classes. Dammhahn (2012) found that young male mouse lemurs were shyer than older males. She hypothesized that selection favoured young mouse lemurs that exhibited less risky behaviour because of the expected future fitness payoff, while older mouse lemurs benefitted most from (also risky) investments into the current reproductive effort. A trade-off between reproductive states may thus maintain animal personality variation in this species (Dammhahn 2012).

Sex Differences and Life History Trade-Offs

The repeatability and consistency of behavioural traits can also differ between the sexes (Schuett et al. 2010). In the above mentioned grey mouse lemurs, males were on average bolder than females and boldness was more repeatable in males than in females (Dammhahn 2012). Schuett et al. (2010) suggested that females prefer males that express consistent behavioural traits as reliable signals of quality. If so, the higher repeatability of boldness in male mouse lemurs could have resulted from sexual selection through female choice (Dammhahn 2012). Sex-specific natural selection can also cause sex differences in behavioural consistency. Male adult field crickets (*Gryllus integer*) behaved more shyly than juvenile males, while females showed consistent boldness across life history stages (Hedrick and Kortet 2012). Due to courtship callings male crickets face a higher predation risk after metamorphosis than females (Hedrick and Kortet 2012). The sex difference in behavioural consistency seemed to result from differences in costs and benefits of risk-taking behaviour between young and adult individuals (Hedrick and Kortet 2012). Overall, males tended to show more repeatable behaviours than females in a meta-analysis including a variety of behaviours in many different animal species, but this conclusion may be biased due to the generally low repeatability of mate preference behaviours in females (Bell et al. 2009).

Different Estimation Methods

As already reviewed by Bell et al. (2009) several statistical methods have been used to estimate repeatability and consistency of individual behaviour. Reported repeatability measurements of various species appeared to be quite variable and this might not only be due to differences between the species, but rather due to the diverse estimation methods used in those studies (Bell et al. 2009). Some estimations were based on rank correlations between the first and the second measurement of each animal. Other studies applied the comparison of within-individual variance with the variance among individuals (Lessells and Boag 1987), based on ANOVA. More recently, Nakagawa and Schielzeth (2010) recommended the use of linear mixed effect models (LMMs) to account for confounding variables through fixed factors and covariates in the model. Furthermore, generalised linear mixed effect models (GLMMs) can be applied if behavioural traits show non-Gaussian error distributions (Nakagawa and Schielzeth 2010). In cases where varying testing conditions contribute to between-individual variation, the use of LMMs is particularly useful to avoid an overestimation of repeatability (Dingemanse and Dochtermann 2013). Which estimation method was chosen for the calculation of repeatability can have profound effects on the conclusions, as illustrated by the following example: In birds, Small and Schoech (2015) showed that the stress-induced corticosterone level was repeatable in adult female Florida scrub-jays (*Aphelocoma coerulescens*), but not in males. However, this difference between the sexes was only apparent when *agreement repeatability* was calculated: A measure of repeatability that ignores any confounding factors (Nakagawa and Schielzeth 2010). When the authors applied LMMs including effects of the study year and the time of day, the resulting values for *adjusted repeatability* (Nakagawa and Schielzeth 2010) of stress-induced corticosterone level were significant in both sexes (Small and Schoech 2015).

Study Objectives and Hypotheses

We investigated the repeatability and consistency of three behavioural traits and one cognitive trait in the Eurasian harvest mouse (*Micromys minutus*; Pallas, 1771), in males and in females, as well as in various life history phases. We measured the standard personality traits activity, boldness and exploration, as we expected them to differ constantly between individuals in our study organism like in other rodent species. Being a small prey species, levels of activity, boldness and exploration are likely to affect the survival of harvest mice critically and may show different adaptive optima depending on the constitution of an individual and on its specific environment. Thus, constant between-individual differences in these behaviours may be expected in harvest mice. We calculated the repeatability of these traits within juveniles and within adults to investigate if individuals already differ constantly before maturation, or if constant between-individual behavioural differences arise later in life. Given the general theoretical prediction of a relationship between animal personality and cognition (Sih and Del Giudice 2012), we also investigated the repeatability of a spatial cognition trait. Spatial cognition can be defined as how animals acquire, process, store, and use spatial information from the environment (Shettleworth 2010). This type of information can have a direct impact on the individual expression of the personality traits activity and exploration. As a first step towards the exploration of a potential link between personality and spatial cognition, we investigated constant between-individual differences in spatial cognition in harvest mice. We assumed that spatial cognition is an important cognitive ability in this species, as harvest mice, in their natural habitat, use a dense three-dimensional grid of various tussock and reeds. Within this compact vegetation, each individual uses several sleeping and breeding nests spread across different heights, and these nests are rebuilt repeatedly at new locations. Harvest mice thus have to acquire and store precise information about the location of their nests, and this information has to be updated frequently. The size of the hippocampus, a brain region involved in processing spatial information about the environment (Yaskin 2011), suggests indeed that the brain structure of harvest mice is specifically adapted to spatial orientation: It occupies 16.2 % of the telencephalon, this is 4.6 % more than in laboratory house mice (Mus musculus, 11.6 % hippocampal volume of the telencephalon, West 1990). We chose to measure object based spatial recognition of a novel arm in a Y Maze and investigated the repeatability of this cognitive trait. Furthermore, we analysed the consistency of all studied traits across life history stages (before and after maturation, as well as before and after the first sexual contact). We considered it likely that developmental processes (e.g., hormonal changes during maturation), or in a natural environment expected changes (e.g., population density) differentially affect behaviours of harvest mice at different life history stages and may thus affect the consistency of individual behaviour.

We applied ANOVA and linear mixed effect models (LMM, following Nakagawa and Schielzeth 2010) to estimate the repeatability and consistency of the behaviours. We thereby chose two approaches for the LMM based estimations: One approach included an additional random factor for the litter identity, the other approach was without this factor in the LMMs. Personality may or may not have a genetic basis, but is probably always also (and in some cases entirely) shaped by environmental influences (e.g., Nicolaus et al. 2016). Theory (e.g., Stamps and Groothuis 2010a; Stamps and Groothuis 2010b; Trillmich and Hudson 2011) and recent experimental evidence (e.g., Polverino et al. 2016) show that personality can arise during ontogeny. Therefore, genetic effects, maternal effects, common environmental effects and interactions between siblings all play a contributing role in shaping personality in adult animals. Repeatable behaviour is a basic prerequisite for animal personality, and it can also arise from both, genetic and environmental effects on behaviour expression. This means animals can behave repeatedly because their alleles affect behaviour to a significant extent, or because their past experience within their environment causes a certain behavioural expression. The comparison between models accounting for a litter effect and models without that litter effect allowed us to obtain a first indication on whether genetic or maternal effects contribute to behavioural repeatability.

We expected to find repeatable behaviour in juveniles, as well as in adult Eurasian harvest mice. Since activity was the most consistent behaviour in other rodents, we expected activity to be consistent across life history stages in our study species. We hypothesized that the expected future reproductive success differs between individuals that have not yet had sexual contact, and those that already experienced sexual contact, given the very short lifespan of harvest mice under natural conditions. Thus, risky behaviour may be less adaptive before the first mating than after the first mating. And we expected that boldness would not be consistent across life history stages. We also expected that exploration would not be consistent as shown in previous studies of rodent personality. Dispersal patterns are not well understood in harvest mice, but we presumed differences in the dispersal probability at different life history stages. Furthermore, we assumed no consistency in spatial recognition, as the motivation for, and the experience in spatial orientation are likely to change over lifetime. We further predicted that juvenile harvest mice are shyer than adult harvest mice due to high predation risk for all ages, but higher expected future reproductive success in young harvest mice. Little is known about mate choice in harvest mice, however, assuming that females are the choosy sex in harvest mice and prefer males that express consistent behavioural traits as reliable signals of quality (Schuett et al. 2010), we expected less repeatable behaviour in female adult mice compared to males. We expected no repeatability differences between the sexes in juveniles. We had no predictions for the difference estimation models and for the random litter effect, as there is no information so far on genetic or maternal effects on behaviour in Eurasian harvest mice. Nevertheless, if we observe reduced estimates of repeatability with a litter effect in the model, compared to a model without that effect, this would mean that the observed repeatability is (partly) due to either genetic effects, direct maternal effects or other environmental effects that are common to litter mates.

Methods – Repeatability and Consistency in Harvest Mice

Study Animals and Housing Conditions

The Eurasian harvest mouse is one of the smallest rodents in Europe with an average body mass of 7 grams (Piechocki 2001). Both sexes show home range overlap (Padilla 1999), but can exhibit fierce aggressive behaviours upon encounter, in particular females with dependent pups (Frank 1957; Piechocki 2001). Females are larger than males (Bohme 1969). The species is widely distributed in Europe and Asia, mostly between 100 and 300 meters above sea level, but harvest mice also occur in montane regions above 1200 meters (Krystufek and Kovacic 1984; Spitzenberger 1986). Harvest mice inhabit wetland areas with reed and sedge zones. In some regions they also successfully populate grain fields and ruderal areas (Spitzenberger 1986;

Feldmann 1997). Feldmann (1984) stated that high grass vegetation and meadows of tall forbs are essential parameters of suitable habitats for harvest mice. Within the Muridae, *Micromys minutus* is quite special as it is an excellent climber and lives mainly above the ground, where it feeds on seeds and insects (Piechocki 2001; Okutsu et al. 2012). The small body size and a long tail that can firmly cling to the vegetation are morphological adaptations to its preferred habitat (Frank 1957). Males and females build several spherical sleeping nests, mainly above ground in dense vegetation (Piechocki 2001). Females raise young alone in a breeding nest. Harvest mice live in unstable environmental conditions caused by seasonal changes of abiotic and biotic factors: Rain, wind and drought affect the grassy habitat structure and food availability, and as in other rodent species, population sizes increase during the summer and show high peaks in autumn, followed by a strong population decline over winter (Piechocki 2001). Breeding occurs mainly during August and September with three to six offspring per litter (Trout 1978b). In good conditions, the inter-litter interval can be as short as 17 days (Trout 1978a; Feldmann 1997). The age of sexual maturation depends on environmental conditions (Frank 1957), but has been recorded between the age of 40 to 50 days (Kubik 1952; Braun and Dieterlen 2005). There is no reliable data on the life span of harvest mice in the wild. Estimates for the average life expectancy range from two to 18 months (Kubik 1952; Piechocki 2001). While harvest mice can live up to 25 months in captivity (Schuster, personal observations), the maximum life span observed in natural or semi-natural populations was 11 and 14 months, respectively (Padilla 1999; Schuster, personal observations).

In total, we tested 41 male and 42 female harvest mice from 34 litters (one to six offspring per litter). All animals stemmed from our laboratory population whose founding individuals (N = 26) originated from four different zoo populations. The mean inbreeding coefficient of the observed individuals was 0.15 (range: 0 to 0.31).

Mice were housed in polycarbonate cages of 60 cm length, 40 cm width and 58 cm height (see ***FIGURE 6***). Individuals were either kept separately or in pairs of equal sex with water and food (hay, grain seeds, fresh fruit and vegetables) *ad libitum*. The back wall of the cage was covered by a coco coir mat for climbing. For environmental enrichment, cages were additionally equipped with an artificial nest, a running wheel, a paper tube, a wooden branch and a sheaf of wheat, oat and spelt. All mice were kept at constant temperature (22°C, range: 21.0°C to 23.5°C) and light-dark cycle (LD 12:12 h). Animal husbandry and behavioural tests (see below) were permitted by the Regierungspräsidium Tübingen – Referat 35, reference number ZO 2/11.

FIGURE 6 Polycarbonate cage in which harvest mice were housed in (separately or in pairs of equal sex). Artificial nest, running wheel, paper tube, wooden branch and sheaf of grain for environmental enrichment. Photo by Andrea C. Schuster

Experimental Set-up

We tested harvest mice in four different age classes (see ***TABLE 3***) in three different behavioural tests. The number of tested animals differed between tests due to some exclusions (see behavioural test descriptions). In total, 83 animals were tested at the age of 6 weeks (mean age = 44.5 days). 39 of these animals were tested at the age of 7 weeks (mean age = 51.7 days). We assumed that in these two age classes, animals were not fully mature yet, but developmental and hormonal changes may have already occurred due to starting maturation. For the third age class, we tested 52 of the initial 83 mice at the age of 12 weeks (mean age = 86.2 days). At this point all animals were assumed to be mature, but none of them has experienced sexual contact yet. Finally, 47 individuals were tested at the age of 24 weeks (mean age = 174.9 days). Some of these (N = 16) were allowed to have sexual contact between week 12 and week 24. We measured the repeatability of behaviours in juvenile harvest mice before maturation using the data obtained in weeks 6 and 7 (subset repeatability, ***TABLE 3***), and the repeatability of behaviours in adult mice after maturation using the data from weeks 12 and 24. Hereby, we analysed individuals which had no sexual experience (subset repeatability, ***TABLE 3***). To test if sexual experience influences the consistency of the behavioural traits in adult harvest mice, we analysed the consistency of adult individuals (subset consistency, week 12 vs. week 24, see ***TABLE 3***), which had sexual experience between tests. During the sexual experience phase, one female and one male spent two weeks together in a home cage giving them the opportunity to mate before being separated again. To measure consistency across different life stages (before and after maturation), we compared the age classes week 6 and week 12 (subset consistency, ***TABLE 3***). Additionally, we compared mean levels of behaviours between males and females in young harvest mice (6 weeks), and in adults (12 weeks). Furthermore, we compared mean levels of behavioural traits between juvenile and adult harvest mice (week 6 vs. week 12).

TABLE 3 The number of tested animals in each of the three behavioural tests for four different age classes (week 6, 7, 12 and 24)

		age class				
	behavioural test	week 6	week 7	week 12	week 24	
number of animals	*Open Field*	83	38	52	31	16
	Novel Object	77	39	52	31	16
	Y Maze	81	39	51	31	16
sexual contact		no	no	no	no	Yes
subsets repeatability		juveniles		adults		
subsets consistency		across maturation		across sexual contact		

Behavioural Tests

Standard behavioural tests, originally established to test emotionality in laboratory strains of mice and rats, were modified and adapted to our study species. It was not possible to record data blind because our study involved focal animal observations, however we accounted for potential confounding factors due to an observer bias (see Data Analysis).

Open Field (OF)

We used a modified OF test (Archer 1973) to measure activity and boldness. In the OF test, rodents are expected to spend more time next to the wall of the arena due to their predisposition to avoid open space and the risk of avian predation (Archer 1973). The OF test is hence supposed to be a suitable set-up to measure boldness in rodents (Herde and Eccard 2013). Mice were released in the middle of a round arena (see ***FIGURE 7***). We used a standard mortar bucket (diameter 51 cm), which we painted with white colour for efficient tracking results. Using automated video tracking (EthoVisionXT, Version 5, Noldus) we measured the total distance moved (in cm) during 4.5

minutes and the time (in seconds) spent at the inner part of the arena (unsafe area, diameter 31 cm) as parameters for activity and boldness, respectively. Tracking started when the animal reached the outer zone of the arena next to the wall for the first time (safe area, 10 cm wide). We excluded one record from the analysis (see ***TABLE 3***, week 7), because one mouse did not move during the OF experiment and spent the whole time in the middle of the arena.

FIGURE 7 Open Field arena (diameter 51 cm) used to quantify activity and boldness of harvest mice. Mortar bucket painted with white colour for efficient automated video tracking results. Photo by Andrea C. Schuster

Novel Object (NO)

To quantify exploratory behaviour we used a NO test (Chitty and Shorten 1946). This was conducted directly after the OF test to reduce further stress as animals were already habituated to the arena setting. We used different objects for each trial: A small plant pot, a plastic box, a glass bowl and LEGO duplo bricks (***FIGURE 8***). Each object was about the same size of the mice and animals were able to explore it from all sides and also from the top by climbing on it. We recorded exploratory behaviour manually for 5 minutes after the first contact with the novel object as the time (in seconds) the animal spent in physical contact with the novel object (touching the object with its head, sniffing at the object from a maximal distance of 1 cm, or climbing onto the object). Accidental contacts, e.g. with the tail, were not quantified as exploration. Since this test was initiated only after all other tests had been established, only 77 animals ran through the NO test at the age of 6 weeks (see ***TABLE 3***).

FIGURE 8 Items that served as novel objects during Novel Object tests to quantify exploration behaviour of harvest mice. Objects were about the same size of the animals. Photos by Andrea C. Schuster

Y Maze (YM)

We quantified spatial recognition in an adapted YM arena (Montgomery 1955). This test relies on the assumption that rodents explore novel environments more than already known environments (Hughes 1968). Thereby, known environments can be recognized through object cues. In our set-up spatial recognition was only based on objects placed around the arena. The YM was made of transparent plastic, each of the three arms was 29 cm long, 3.5 cm wide and 10 cm high (see ***FIGURE 9***). We used a set of extra maze cues which the animals knew from their home cages: Small plant pots, stands of a running wheel, paper tubes, wooden sticks, clothes-pegs, small glass bowls, pieces of coco coir mat, and stems of straw. Each arm of the YM was surrounded by plain cardboard such that the animal could only see the object cues placed around the arm it was in. Visual cues were randomized between arms and animals, as was the position of the start arm. The YM test consisted of two trials. During the first trial, one of the arms was locked (unknown arm) and the animal could explore only two arms (start and known arm) for 15 minutes. After one hour the mouse was released again into the YM at the same start arm. In this second trial all three arms were accessible. In the cognition literature, the behaviour of spending more time in the unknown arm of a Y Maze in the second trial is commonly defined as spatial recognition in the sense that an animal recognized that there is an unknown environment (e.g., Dellu et al. 1992). We video tracked animals using EthoVisionXT software and measured the total distance moved (in cm) during the first five minutes of the second trial as parameter for activity. The time (in seconds) spent in the unknown arm during the first five minutes of the second trial was recorded as parameter for spatial recognition. Two mice sat motionless in the start arm during the entire trial, and one animal ran straight to the end of the known arm and sat there motionless during the entire trial. These observations confer no information about whether the animal recognised the new arm, and were deleted from the YM dataset (see ***TABLE 3***).

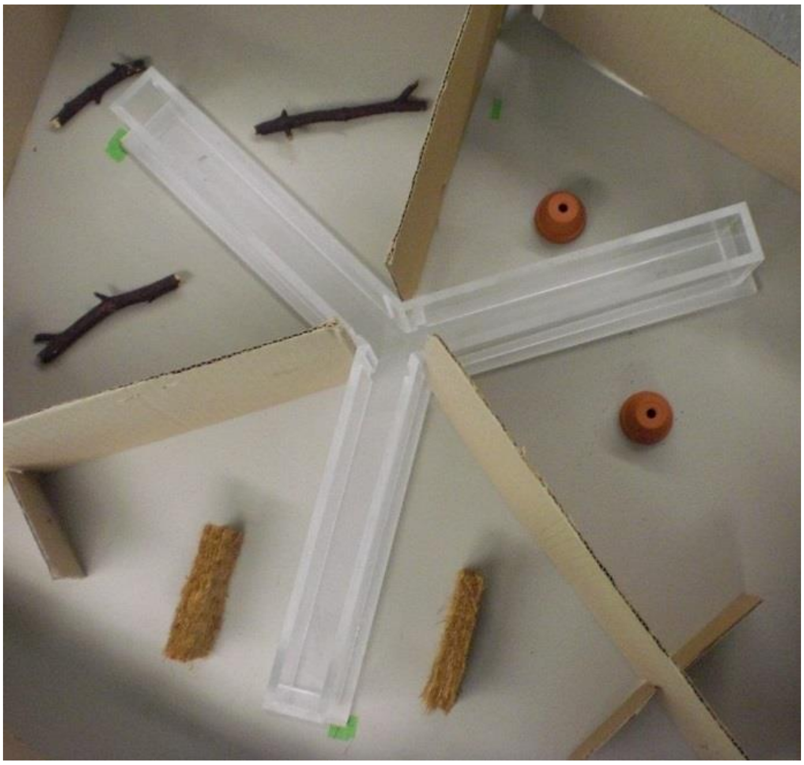

FIGURE 9 Y Maze used to quantify spatial recognition and activity in harvest mice. Each arm surrounded by plain cardboard such that animals could only see object cues placed around the arm it was in. Photo by Andrea C. Schuster

Data Analyses

We performed all statistical analyses using R software, version 3.0.1 (R Core Team 2013). The dependent variables were the measured behaviours: Distance moved in OF and YM (activity), time spent in unsafe area of OF (boldness), time exploring NO (exploration), and time spent in unknown arm of the YM (spatial recognition). We tested for confounding factors of the experimental set-up and of individual characteristics on the measured behaviours by fitting linear mixed models using the "nlme" package (Pinheiro and Bates 1996) in R. For each behavioural trait we included the following confounding factors into the full model: Individual ID as random factor; sex, trial number (the number of test trials of each mouse) and housing condition (whether the mouse was kept individually or in a group of two in the home cage) as fixed factors; body mass, test date and test time as fixed covariates. As males and females might differ in their activity rhythms, we included the interaction sex*test time. Further, females harvest mice are larger than males (Piechocki 2001) and this dimorphism might influence individual behaviours of sexes differently. We therefore also included the interaction sex*body mass in the full model. As the NO test was recorded manually by one of two observers, we also included the ID of the observer to linear models fitting exploratory behaviour. We applied backward stepwise reduction of the full models by excluding non-significant interactions first, followed by non-significant fixed effects if p > 0.05. The random factor of the individual ID remained in all models to account for repeated measures of individuals. All deleted confounding factors were added to the final (reduced) model again one by one, to avoid that a significant effect was missed due to the order in which factors were deleted from the model. However, none of the deleted confounding factors had a significant effect if added again to the final model. We conducted this analysis separately for the four subsets of the data, as those subsets were then used to calculate repeatability or consistency (see Experimental Set-up, ***TABLE 3***). All dependent variables and all covariates were centred and scaled to ensure model convergence. We checked all models for normal distribution of model residuals. We present final models with the remaining significant effects, which we retained for subsequent repeatability analyses. As we were interested in behaviour differences between males and females, and between juvenile and adult mice, we compared mean behaviours between those groups. The respective effect sizes from the confounding factors sex and trial number provided information on significant differences. We present group means ± standard errors of behavioural traits in the text (untransformed values).

The repeatability of behavioural traits was analysed using the package “rptR” (Nakagawa and Schielzeth 2010) in R. We estimated *adjusted repeatability* and we included significant confounding factors as identified from LMMs before (see above). We used untransformed dependent variables for these estimations. We estimated LMM based repeatability without litter ID in the model applying non-parametric bootstrapping. We display asymptotic 95% confidence intervals (CIs) for parameter estimates based on 1000 bootstrapping runs and 1000 permutations. We used Bonferroni correction to account for multiple comparisons. Our significance threshold was then p ≤ 0.008, as we calculated six different repeatability values per behavioural trait (two age classes times three estimation methods) and six different consistency measurements per behavioural trait (two experimental groups times three estimation methods). When using LMM based repeatability estimation, the package “rptR” uses algorisms that do not converge, if R is close to zero or negative. In this case, CIs and *p* values are shown as NA, and we concluded that R is zero. We display results of two further estimation methods: ANOVA based repeatability (Lessells and Boag 1987) implemented in “rpt.aov” function in R using 1000 permutations, and LMM based repeatability with litter ID as a random factor. As tested individuals originated from 34 different litters (one to six offspring per litter), this allowed us to investigate whether differences between litters due to genetic variation and/or direct maternal effects contributed to the repeatability estimation of the tested behaviours. To evaluate how the variance within and between different litters might influence repeatability estimates of behavioural traits, we compared the outcome of

LMMs with, and without the litter ID in the random model, using 84% CIs. Non-overlapping 84% CIs indicated significant differences between repeatability estimates (Small and Schoech 2015). All CIs are given in squared brackets.

Results – Repeatability and Consistency in Harvest Mice

Effects of Confounding Factors

Univariate LMMs revealed some significant fixed effects (***Table 4***) which we then included in the repeatability and consistency estimations (see below). The trial number had an effect on activity and on boldness in juvenile harvest mice (***Table 4***). In the second trial (week 7) juveniles were less active in the OF and in the YM and they were bolder in the OF, than in the first trial (week 6).

Table 4 Results of final LMMs with confounding effects on personality traits. SE = standard error

	personality trait	significant effect	estimate ± SE	*p*
juveniles (week 6 vs. 7)	activity OF	trial number	-0.30±0.15	0.049
		test date	+0.34±0.11	0.005
	boldness	trial number	+0.53±0.15	< 0.001
	activity YM	trial number	-0.46±0.18	0.017
	recognition	none		
	exploration	observer	+0.51±0.20	0.016
adults (week 12 vs. 24)	activity OF	housing condition	+0.99±0.22	< 0.001
		sex	-0.55±0.22	0.021
		trial number	-0.47±0.15	0.005
	boldness	sex	+0.85±0.21	< 0.001
	activity YM	none		
	recognition	none		
	exploration	none		
before and after maturation (week 6 vs. 12)	activity OF	date	+0.33±0.12	0.008
		sex	-0.38±0.18	0.042
		trial number	-0.27±0.12	0.025
	Boldness	trial number	+0.76±0.13	< 0.001
	Activity YM	trial number	-0.31±0.14	0.032
	Recognition	none		
	Exploration	none		
before and after sexual contact (week 12 vs. 24)	activity OF	none		
	boldness	sex	+0.69±0.25	0.01
		trial number	-0.50±0.21	0.03
	activity YM	none		
	recognition	none		
	exploration	none		

There was an effect of the observer identity on juvenile behaviour in the NO test, and the housing condition affected the behaviour of adult mice in that test: mice kept in pairs were more explorative than individually housed animals (see ***Table 4***). We found no significant confounding effects on spatial recognition.

Effects of Sex and Age on the Mean Expression of Behaviours

In juvenile harvest mice, males and females did not differ in their behaviour. Among adult harvest mice, females were bolder ($\bar{x}$ = 77.3 ± 6.0 s) and less active in the OF ($\bar{x}$ = 3506.9 ± 295.5 cm) than males (boldness: $\bar{x}$ = 45.6 ± 4.4 s, activity: $\bar{x}$ = 4374.5 ± 369.2 cm). This sex difference in boldness was also significant in adult mice tested before and after their first sexual contact (females: $\bar{x}$ = 64.6 ± 7.0 s, males: $\bar{x}$ = 42.2 ± 5.2 s). Furthermore, mice were significantly bolder before their first sexual contact than after ($\bar{x}$ = 56.0 ± 5.2 s and 42.2 ± 8.6 s, respectively). We also found that the level of boldness was lower in juveniles that were tested before maturation (week 6) than in adult mice tested after maturation (week 12; $\bar{x}$ = 30.4 ± 2.8 s and 56.0 ± 5.2 s, respectively). However, juveniles were more active in the OF and YM than adults (OF juveniles: $\bar{x}$ = 4115.5 ± 247.6 cm and OF adults: $\bar{x}$ = 4172.6 ± 318.1 cm; YM juveniles: $\bar{x}$ = 1765.8 ± 91.4 cm and YM adults: $\bar{x}$ = 1540.1 ± 143.2 cm). We did not find any differences between juveniles and adults in their exploratory behaviour or in spatial recognition.

Repeatability of Behavioural Traits in Harvest Mice

Eurasian harvest mice showed repeatable behaviours in both age classes (***Table 5***, LMM without litter effect). Activity in the OF test was highly repeatable in juveniles and adults (R = 0.54 ± 0.10 and R = 0.49 ± 0.13, respectively). Activity in the YM was repeatable in both age classes, adult behaviour was more repeatable than juvenile behaviour, shown by non-overlapping 84% CIs (R = 0.69 ± 0.09 [0.57, 0.80] and R = 0.16 ± 0.14 [0, 0.37], respectively). Boldness was also repeatable in juvenile and adult harvest mice (R = 0.57 ± 0.11 and R = 0.20 ± 0.16, respectively). The higher R value for juvenile mice did not differ significantly from the lower value for adults (84% CIs: [0.41, 0.70] and [0, 0.42], respectively). Similarly, exploration seemed more repeatable in juvenile mice than in adults, but the difference was not significant (R = 0.40 ± 0.12 [0.26, 0.59] and R = 0.18 ± 0.15 [0, 0.42], respectively). Recognition of the novel arm in the YM was not repeatable in juveniles (R = 0.00), but adult mice behaved repeatably in this test (LMM without litter effect: R = 0.20 ± 0.15, *p* = 0.004). In juveniles, the activity in the YM was only repeatable in males, not in females (R = 0.64 ± 0.13 and R = 0.00, respectively; data not shown). Secondly, in adult harvest mice, boldness was only repeatable in females, not in males (R = 0.58 ± 0.19 and R = 0.00, respectively; data not shown). Other repeatability estimates did not differ between the sexes.

Consistency of Behavioural Traits in Harvest Mice

Eurasian harvest mice behaved consistently before and after maturation, as well as before and after their first sexual contact (***Table 5***, LMM without litter effect). Harvest mice tested before (week 6) and after (week 12) maturation were consistently active, bold and explorative (all R > 0.39, see ***Table 6***). Only the recognition of the novel arm in the YM was not significantly consistent after Bonferroni correction (R = 0.11 ± 0.11, *p* = 0.010). Adult harvest mice also behaved highly consistently before (week 12) and after (week 24) their first sexual experience (***Table 6***, LMM without litter effect). Activity in the OF and YM, as well as boldness were highly consistent (all R > 0.53). Exploration was also significantly consistent between the tests, although this R value was smaller (R = 0.18 ± 0.19). The only clearly inconsistent behaviour shown before and after first sexual contact was the recognition of the novel arm in the YM (R = 0.00 ± 0.15). None of the consistency estimates differed between the sexes (data not shown).

Estimation Methods / Reliability

When we included a random effect for the litter, we observed, in most cases, reduced values for repeatability and consistency (see ***Table 5*** and ***Table 6***). The most pronounced reduction in R occurred in the models for activity in the YM. For this trait, the repeatability estimate in adults and both consistency estimates differed between the LMMs without and with the litter effect (84% CIs: repeatability in adults [0.57, 0.80] and [0, 0.27], consistency before and after maturation [0.26, 0.59] and [0, 0.23], consistency before and after sexual contact [0.43, 0.82] and [0, 0.13], respectively). Other 84% CIs did not differ significantly between LMMs. However, with litter as random effect (and thus partly excluding potential genetic effects and non-genetic maternal effects from the estimation of R), activity in the OF was repeatable and consistent, boldness was consistent but its repeatability estimates did not reach significance, and exploration was repeatable only in juvenile mice.

In the ANOVA analyses, we ignored the confounding factors trial number, sex, test date and housing condition. Depending on the personality trait, this led to a more or less pronounced reduction in the estimated values for repeatability and consistency (see ***Table 5*** and ***Table 6***). This reduction was not consistent within personality traits across tests of different life history stages. It rather appeared to depend on the particular data composition within each model. Overall, out of 16 ANOVA analyses (we exclude the models on recognition here, as they mostly displayed R values close to zero), seven suggested (partly considerably) lower R values than the corresponding LMMs without the litter effect. In some more cases, R values resembled those obtained from LMMs, but more conservative p values did not warrant the same conclusions as from the LMM analyses (see ***TABLE 5*** and ***TABLE 6***). In one case (repeatability of boldness in adult mice), the R value increased compared to LMM method when using the ANOVA calculation. According to the ANOVA analyses, only activity in the OF was a repeatable and consistent behaviour, and exploration tended to be repeatable in juvenile mice and was performed consistently before and after maturation.

TABLE 5 Results of repeatability analyses showing R values calculated for each personality trait per experimental group (juveniles and adults). Sample sizes are given: first trial/second trial. Bold p values were significant after Bonferroni correction ($p < 0.008$). OF: Open Field test, YM: Y Maze test. NA: model did not converge because of too small/negative value of R

				LMM without litter effect			LMM with random litter effect			ANOVA		
experimental group	personality trait	fixed effects in LMM	N	R±SE	95% CI	*p*	R±SE	95% CI	*p*	R±SE	95% CI	*p*
juveniles (week 6 vs. 7)	activity OF	trial number + test date	83/38	0.54±0.10	[0.35, 0.73]	< 0.001	0.45±0.13	[0.20, 0.70]	< 0.001	0.54±0.11	[0.33, 0.76]	0.001
	boldness	trial number	83/38	0.57±0.11	[0.32, 0.73]	< 0.001	0.27±0.13	[0, 0.53]	0.038	0.42±0.13	[0.16, 0.68]	0.013
	activity YM	trial number	81/39	0.16±0.14	[0, 0.47]	< 0.001	0.05±0.11	[0, 0.37]	0.359	0.06±0.18	[-0.31, 0.42]	0.409
	recognition		81/39	0.00	NA	NA	0.00	NA	NA	-0.50±0.22	[-0.94, 0.07]	0.970
	exploration	observer	77/39	0.40±0.12	[0.19, 0.66]	< 0.001	0.39±0.14	[0.17, 0.56]	0.004	0.39±0.14	[0.12, 0.66]	0.015
adults (week 12 vs. 24)	activity OF	trial number + sex + housing condition	52/31	0.49±0.13	[0.21, 0.72]	< 0.001	0.49±0.17	[0.01, 0.69]	0.006	0.53±0.12	[0.26, 0.78]	0.002
	boldness	sex	52/31	0.20±0.16	[0, 0.53]	< 0.001	0.20±0.15	[0, 0.49]	0.161	0.40±0.15	[0.10, 0.70]	0.030
	activity YM		51/31	0.69±0.09	[0.49, 0.82]	< 0.001	0.11±0.10	[0, 0.34]	0.101	0.69±0.09	[0.51, 0.87]	0.001
	recognition		51/31	0.20±0.15	[0, 0.53]	0.004	0.00	NA	NA	0.16±0.19	[-0.21, 0.53]	0.228
	exploration		52/31	0.18±0.15	[0, 0.49]	0.006	0.00±0.11	[0, 0.36]	1.000	0.15±0.19	[-0.23, 0.53]	0.259

TABLE 6 Results of consistency analyses showing R values calculated for each personality trait per experimental group (before and after maturation, before and after sexual contact). Sample sizes are given: first trial/second trial. Bold p values were significant after Bonferroni correction (p < 0.008). OF: Open Field test, YM: Y Maze test. NA: model did not converge because of too small/negative value of R

				LMM without litter effect			LMM with random litter effect			ANOVA		
experimental group	personality trait	fixed effects in LMM	N	R±SE	95% CI	*p*	R±SE	95% CI	*p*	R±SE	95% CI	*p*
before and after maturation (week 6 vs. 12)	activity OF	trial number + sex + test date	83/52	0.64±0.07	[0.49, 0.77]	< 0.001	0.47±0.11	[0.27, 0.70]	< 0.001	0.67±0.07	[0.53, 0.82]	0.001
	boldness	trial number	83/52	0.39±0.11	[0.17, 0.59]	< 0.001	0.36±0.13	[0.10, 0.58]	0.002	0.17±0.14	[-0.11, 0.46]	0.163
	activity YM	trial number	81/51	0.42±0.11	[0.22, 0.63]	< 0.001	0.07±0.09	[0, 0.31]	0.237	0.40±0.12	[0.17, 0.64]	0.006
	recognition		81/51	0.11±0.11	[0, 0.36]	0.010	0.00	NA	NA	0.04±0.16	[-0.27, 0.35]	0.399
	exploration		77/52	0.41±0.12	[0.13, 0.59]	< 0.001	0.13±0.11	[0, 0.39]	0.180	0.46±0.11	[0.24, 0.68]	0.002
before and after sexual contact (week 12 vs. 24)	activity OF	sex + trial number	52/16	0.84±0.06	[0.70, 0.92]	< 0.001	0.77±0.12	[0.46, 0.92]	< 0.001	0.84±0.06	[0.71, 0.96]	0.001
	boldness		52/16	0.53±0.15	[0.24, 0.82]	< 0.001	0.53±0.18	[0.04, 0.78]	0.008	0.39±0.21	[-0.04, 0.81]	0.164
	activity YM		51/16	0.70±0.15	[0.28, 0.87]	< 0.001	0.00±0.06	[0, 0.19]	0.500	0.70±0.12	[0.47, 0.93]	0.018
	recognition		51/16	0.00±0.15	[0, 0.51]	0.009	0.00	NA	NA	0.13±0.29	[-0.45, 0.72]	0.403
	exploration		52/16	0.18±0.19	[0, 0.60]	0.006	0.00±0.11	[0, 0.44]	0.500	0.09±0.29	[-0.50, 0.67]	0.393

Discussion – Repeatability and Consistency in Harvest Mice

All tested behavioural traits were repeatable and consistent, with the only exception of spatial recognition. Activity, boldness and exploration hence fulfil the assumptions for an animal personality trait and could be components of a behavioural syndrome in Eurasian harvest mice, as it was also found in other rodents (e.g., Koolhaas et al. 1999; Boon et al. 2007; Boyer et al. 2010; Kanda et al. 2012; Herde and Eccard 2013). Spatial recognition was repeatable only in adult mice, not in juveniles. Further, we found less evidence that mice recognized the new arm of the Y Maze consistently before and after maturation, as well as before and after the first sexual contact.

Effects of Sex and Age on the Mean Expression of Behaviours

We found evidence that adult females were less active than males. This difference was significant in the OF test, but not in the YM test. In turn, adult males were on average shyer than females. We detected no sex differences in exploration or spatial recognition. There is little evidence for consistent sex differences in animal personality traits in rodents. In general, sex differences in behaviour of rodents are contradictory and depend on species and behavioural trait (common voles, *Microtus arvalis*, Herde and Eccard 2013, but see Lantová et al. 2011; meadow voles, *Microtus pennsylvanicus,* Halliday et al. 2014; wild house mice, *Mus domesticus*, Auclair et al. 2013; laboratory house mice, *Mus musculus*, Montiglio et al. 2010). Frank (1957) described the social behaviour of Eurasian harvest mice and found females to be the more aggressive sex. We did not measure aggression in this study, and we only rarely observed aggressive encounters, and if so, mainly in groups of males (Schuster, personal observations). Given our currently scarce knowledge on individual behaviour of our study species, conclusions on the selection pressures that may maintain sex differences in activity and boldness remain speculative.

Wolf et al. (2007) hypothesized that animals with high expected future reproductive success should be more risk-averse than individuals with low expected future reproductive success. As a consequence, the individual state of an animal in the ongoing trade-off between current and future reproductive success is expected to affect the strength of selection for more risk-averse behaviour early in life, and for more risky behaviour late in life. In our study, young harvest mice were indeed shyer than older mice, but juveniles were more active than older mice. Younger individuals have more to lose, as they did not yet have the opportunity to reproduce. A more cautious behaviour is likely to increase survival chances in the wild, where terrestrial and aerial predators pose a considerable risk to harvest mice. After maturation, selection may favour males that invest more in actively finding receptive females, and it may favour females that invest more in finding the best food sources and potential nest sites within their home range. It should be noted that juveniles behaved more boldly and were less active in the second trial than in the first (week 7 and 6, respectively). This suggests a habituation effect that may have contributed to the behaviour differences between juveniles and adults in our experiments. However, in the wild, habituation is very likely to contribute to behavioural changes across life history stages as well, as individuals get habituated to the specific environment in their home range and as a benefit from that, can show more risky behaviour as soon as the exact location of food sources and hiding places are known. Thus, the here observed changes in boldness and activity across life stages could be the result of life stage specific selection on risk-averse behaviour. However, we also observed that harvest mice behaved more shyly after the first sexual contact, and this observation does not support our hypothesis.

Repeatability of Behavioural Traits in Harvest Mice

As expected, juvenile and adult harvest mouse behaviour was significantly repeatable (***TABLE 5***). Adult mice did not generally behave more repeatably than juveniles. Only activity in the YM, as well as spatial recognition (also measured in YM), were more repeatable in adults than in juveniles. However, our test for spatial recognition might not be adequate for juvenile harvest mice, as we observed that juveniles often climbed the walls of the YM trying to escape. Thus, this climbing activity contributed to both measurements taken in the YM: duration in the novel arm (spatial recognition) and distanced moved in total YM (activity). And this contribution may depend more on the motivation to climb the walls during the test, rather than intrinsic differences in activity and spatial recognition. This leads us to a cautious interpretation of the outcome of the spatial recognition test in juveniles. It might be preferable to design a test where mice cannot climb as much as in the YM. However, as activity in the OF test was highly repeatable in juveniles, we have no doubt that activity is a repeatable behaviour also in juvenile harvest mice.

Our findings confirm the large body of evidence for the repeatable nature of these personality traits in many taxa (e.g., rodents: Koolhaas et al. 1999; Montiglio et al. 2012; Herde and Eccard 2013, birds: Dingemanse et al. 2002; David et al. 2012, reptiles: Carter et al. 2012; Bajer et al. 2015). Activity, boldness and exploration are particularly well studied animal personality traits, and there is growing evidence about potential changes in repeatability across life history stages (Brommer and Class 2015). Bajer et al. (2015) showed that juvenile European green lizards (*Lacerta viridis*) behaved significantly more repeatably than adults in risk-taking and exploratory behaviour. In our study, the repeatability of boldness and exploration were not influenced by age. However, there was an age effect on the repeatability of activity in the YM, but not so in the OF.

This is in concordance with findings in zebra finches (*Taeniopygia guttata*), where activity was repeatable in subadults but not in young and mature birds (Wuerz and Krüger 2015). We found some differences between the repeatability values of males and females. In adult harvest mice, boldness was only repeatable in females. This was in contrast to our expectation, as we assumed that males would behave more repeatably due to the sex role of the chosen sex. However, this result should be interpreted with care as we have to acknowledge the smaller sample sizes (N between 15 and 21 animals per group) compared to other data subsets, which may have influenced these results.

In sum, we found that Eurasian harvest mice already express repeatable behaviours as juveniles. Furthermore, all tested personality traits and a spatial cognition trait were repeatable in adults. Behavioural repeatability did not seem to change across life history stages, which suggests that behavioural types, if existent in this species, may be a stable phenomenon. As this is the first study on individual behaviours in this species, more investigations are warranted to fully understand which factors maintain the repeatability of behaviours here.

Consistency of Behavioural Traits in Harvest Mice

Eurasian harvest mice showed consistency in activity, boldness and exploration (***TABLE 6***). This was in contrast to our expectations. We assumed that differences in the expected future reproductive success and in dispersal patterns at different life history stages would result in life-history-specific behavioural patterns. However, although the mean behavioural expression changed (adult mice were more bold, but less active than juveniles, see above), the behavioural differences between individuals remained constant across maturation and across the first sexual experience. During maturation and the first sexual contact, the relative magnitude of attained reproductive success and expected future reproductive success changes depending on individual experiences. The dynamics of this trade-off may result in a change of selection pressures on animal personality traits (Bell and Stamps 2004). However, these events did not seem to have an influence on the consistency of the studied personality traits in harvest mice. We tested adult mice at the age of three and six month, which probably represents the entire mean life span of this species in the wild (Kubik 1952; Piechocki 2001). Hence, Eurasian harvest mice - at least under laboratory conditions - seem to behave consistently throughout their entire life span, and the short-term consistency of these behaviours was independent of maturational effects and sexual experience. Although, we acknowledge that the sample size to test for the influence of sexual experience was smaller compared to other subsets.

Bell and Stamps (2004) stated that individual behavioural consistency can vary across lifetime. In particular, they suggested a decline in consistency during sexual maturation, because correlations between behavioural traits may be restructured and become instable due to hormonal effects during this life history phase. Contradictory to our findings, some case studies support this idea through data on inconsistent behaviour across individual maturation. In guinea pigs (*Cavia aperea*), Guenther et al. (2014b) showed that the personality structure changed over different life history stages: While fearlessness and boldness were consistent over maturation, exploration was not, and correlations between behavioural traits changed during ontogeny. In common voles (*Microtus arvalis*, Herde and Eccard 2013) and Siberian dwarf hamsters (*Phodopus sungorus*, Kanda et al. 2012), activity was a consistent behavioural trait across different life history stages. Boldness, exploration, reactivity, and orientation were not consistent. In contrast, activity, boldness and exploration were consistent in some studied birds and insects (Dingemanse et al. 2002; David et al. 2012; Fisher et al. 2015). A study on firebugs (*Pyrrhocoris apterus*) showed that the personality structure can also be consistent across life history stages: Gyuris et al. (2012) found that firebugs behaved consistently and showed consistent trait correlations across larval and adult stages, even past final ecdysis. Thus, even major changes between life history stages are not necessary connected to inconsistent individual behaviour. To date, no comprehensive theory has been developed to predict which factors would favour or disfavour consistent behaviour in specific species. This is in part due to the fact that consistent behaviours can arise from two sources: Natural selection for stable behavioural types, or physiological constraints that prevent flexible behaviour. The relative contribution of these to the expression of personality traits is currently largely unknown. It could be that in harvest mice, consistently behaving animals were favoured by selection. In a frequently changing environment, short lived animals may save energy and avoid erroneous reactions if they restrain from fast behavioural changes in response to environmental cues. Adopting an individual behavioural tactic that suits best the individual constitution may be advantageous. Alternatively, or in addition, physiological constraints may maintain consistent behaviours across the short lifespan.

We also tested if harvest mice behaved repeatably and consistently when they explored a novel arm in a Y Maze. This set-up measured an individual's ability to recognize a new spatial environment based on object cues. Unlike in the personality traits discussed above, we observed less evidence for consistency in spatial recognition. While the performance in the Y Maze was repeatable among adult harvest mice, it was not consistent before and after the first sexual contact. Since there was no mean difference in the performance before and after sexual experience, we hypothesize that sexual experience affected the ability or the motivation of individual mice for this cognitive task. Spatial orientation and recognition can rely on the spatial arrangement of specific cues. We offered object cues in our test trials and excluded olfactory cues. In nature, harvest mice can certainly also use olfactory cues, or environmental geometry for orientation. Individual harvest mice may also differ in which cues they prefer to rely on, and furthermore, specific experiences (like maturation or first sexual contact) may change the individual priority of used cues. Thus, if individual cue perception and cue use changes during the life of the mice (in our experiment: Between the two test trials), this could have contributed to the observed inconsistency in spatial recognition in our test set-up. However, it should be noted that the low sample size of adult mice tested after their first sexual experience (N = 16) only permits preliminary conclusions.

For a long time, the main interest in animal cognition research focussed on the comparative evaluation of cognitive abilities between species. Therefore, little is known about individual variation, repeatability and consistency of cognitive traits within species. We here provided a first evidence for the repeatability (in adults) and consistency (over maturation) of spatial recognition through object cues in harvest mice. Typical animal personality traits such as activity and exploration may correlate with particular cognitive styles (repeatable cognitive behaviour) to form a cognitive syndrome (Griffin et al. 2015; Sih and Del Giudice 2012). An assumption to this is repeatability and consistency of cognitive traits. Spatial recognition as tested in our set-up meets this assumption and may thus be used for a test of correlations between personality and spatial cognition in harvest mice.

Estimation Methods and Effect of the Litter

Repeatability of a behavioural trait is the fraction of total trait variance that occurs between individuals, due to additive and non-additive genetic effects, the permanent environment effect, as well as other effects that cause differences between individuals, such as direct maternal effects (Falconer et al. 1996; Nakagawa and Schielzeth 2010). When we included the grouping factor litter as a random factor in the LMM analyses, the between-individual variance that is due to differences between litters was attributed to that random effect and could hence not contribute to the estimate of R. Accordingly, all models with the litter effect, except two (OF activity and boldness in adults), estimated lower R values than the corresponding models without the litter effect. While those R values present a biased estimate, as genetic and early environmental contributions to behavioural repeatability are partly excluded, they can give an indication on whether heritability or direct maternal effects contribute to the repeatability of a trait. Reduced estimates of repeatability (or consistency) with a litter effect in the model, compared to a model without that effect, indicate that the observed R value is (partly) due to either genetic effects, direct maternal effects or other environmental effects that are common to litter mates and thus cause differences between litters. In this study, we found particular strong evidence for those effects on activity in the Y Maze, as R values for this behaviour decreased significantly when the litter effect was included. Further investigations using the full pedigree of all tested animals are needed to quantify genetic and non-genetic maternal effects on activity in harvest mice.In the ANOVA estimation of R, we ignored any confounding effects and thus calculated *agreement repeatability* (Nakagawa and Schielzeth 2010). These R values were generally smaller than those from the original LMM analyses (except for boldness in adult mice). Although we expected that the bias here would depend on the magnitude of the confounding effects, we did not find congruent reductions of R in models of the same dependent trait. We conclude that the bias of R due to un-modelled confounding effects is not well predictable, as it seems to depend on the specific data structure within each model. As suggested by Nakagawa and Schielzeth (2010) we base our conclusions on LMM analysis of *adjusted repeatability*.

Conclusion - Repeatability and Consistency in Harvest Mice

Repeatability and Consistency in Harvest Mice of the Laboratory Population

In sum, our results show that activity, boldness and exploration were repeatable in juvenile and adult Eurasian harvest mice. Furthermore, these behaviours were expressed consistently independent of age, maturation and individual sexual experience. Our findings show that behaviours in harvest mice meet the two essential assumptions in animal personality research - repeatable and consistent between-individual differences (Réale et al. 2007). Further, we tested spatial recognition based on object cues in a Y Maze. This trait was repeatable in adult mice, but not in juveniles, and it showed little evidence for consistency across life history phases. More investigations are needed to better understand the repeatability and consistency of cognitive traits as this would be a prerequisite for further studies on cognitive syndromes (Sih and Del Giudice 2012).

Chapter 2

In Chapter 2, we provide evidence concerning the second assumption of animal personality – correlations between different behaviours – tested in adult Eurasian harvest mice. Further, we show how behavioural types are related to cognitive styles in this species:

Animals show constant between-individual behavioural differences in many species. Further, personality traits form behavioural syndromes, characterised by correlations between different behaviours. Mechanisms maintaining these correlations could be constrained due to underlying relationships with cognitive traits. There is growing evidence for the non-independence of animal personality and general cognitive abilities in animals, but so far, studies on the direction of the relationship between them revealed contradictory results. Still, it is hypothesised that individuals may exhibit consistent learning and decision styles. Within a cognitive syndrome fast behavioural types (consistently bolder and more active individuals) are expected to show also faster learning styles. Slow behavioural types in contrast are assumed to learn slower but more accurately. This can be caused by a speed-accuracy trade-off that individuals face during decision making. We measured the repeatability of three personality and four spatial cognitive traits in adult Eurasian harvest mice (*Micromys minutus*). We analysed correlations among personality traits (behavioural syndrome) and investigated the relationship between personality and spatial cognitive traits (cognitive syndrome). Our results showed that exploration, activity and boldness were repeatable in adult mice. Spatial recognition measured in a Y Maze was also significantly repeatable, as well as spatial learning ability and decision speed. We found no repeatability of decision accuracy. Harvest mice showed a behavioural syndrome as we observed strong positive correlations between personality traits. The speed-accuracy trade-off was not apparent within, nor between individuals. Nevertheless, we found weak evidence for a cognitive syndrome as fast behavioural types learned the Spatial Orientation Task faster than slow types, and shyer harvest mice made decisions quicker than bolder mice. Given these relationships, our data partly support the hypotheses of a cognitive syndrome in harvest mice and will hopefully stimulate more studies on the relationship between cognition and personality in animals.

Behavioural and Cognitive Syndromes in Laboratory Harvest Mice

Behavioural and Cognitive Syndromes in Laboratory Harvest Mice

Constant between-individual differences in behaviour are known as animal personality (Wilson 1998; Réale et al. 2007). Further, correlations between constant behavioural traits – also called personality traits – are defined as behavioural syndrome (Sih et al. 2004b). In birds, the relation between different personality traits followed a fast-slow continuum (Titulaer et al. 2012). For instance, more exploratory great tits (*Parus major*) were bolder (Verbeek et al. 1994), more aggressive (Carere et al. 2005) and showed more risk-taking behaviours (van Oers et al. 2004b) than less explorative birds. In rodents, Koolhaas et al. (1999) termed a similar behavioural syndrome the proactive-reactive syndrome, where proactive individuals were more active and more aggressive than reactive mice (*Mus musculus*). The behaviour pattern that an individual expresses was defined as a behavioural type, representing the characteristics of an individual's personality (Bell 2007). Selection for proactive (fast) behavioural types is likely in stable environments, where risky behaviours confer advantages in competitive situations. Reactive (slow) behavioural types, on the other hand, would have advantages in unstable environments which favour behavioural flexibility (Koolhaas et al. 1999).

The concept of behavioural types can also be applied to cognitive behaviour (Sih and Del Giudice 2012). If adaptive cognitive behaviour can be achieved through different strategies, or if animals face consistent trade-offs when solving cognitive tasks, we would expect to observe cognitive styles. Cognitive traits in general provide the basis of any other behaviour as they refer to the capacity of individuals to acquire, process, store and remember information (Shettleworth 2010). Cognitive styles were defined as strategies *how* individuals do this, and these strategies are expected to be consistent across time and contexts (Sih and Del Giudice 2012). Consistent cognitive styles could arise due to constant decision-making behaviour. When an individual has to take a decision it faces the fundamental problem of a speed-accuracy trade-off (Chittka et al. 2009). Individuals can either decide fast at the potential cost of an accurate decision or take their time and decide more accurately (Burns and Rodd 2008). Accuracy has been interpreted in the past as a limit to the cognitive ability of an individual (Chittka et al. 2009), making strong selection for accuracy unlikely. However, recent data showed that both cognitive styles (fast vs. accurate) can occur in a population side by side (Wang et al. 2015), indicating that both styles might be similar adaptive under specific environmental conditions.

The Cognitive Syndrome

Sih and Del Giudice (2012) provided a clear theoretical framework for a link between behavioural types and cognitive styles based on a risk-reward trade-off. Within a cognitive syndrome, fast behavioural types (consistently bolder, more aggressive and more active individuals) are expected to take higher risks while being rewarded faster (fast learning styles). Slow behavioural types in contrast are assumed to decide more accurately but less fast, and to take less risks (Sih and Del

Giudice 2012). Recent tests of this hypothesis showed contradictory results: Experimental work on black-capped chickadees (*Poecile atricapillus*) confirmed the hypothesis of a cognitive syndrome, since slow exploring individuals showed higher accuracy levels than fast exploring birds during learning trials of an instrumental discrimination task (Guillette et al. 2015). Bousquet et al. (2015) on the contrary could not identify any correlation between exploration and decision accuracy in a spatial learning task in mallards (*Anas platyrhynchos*). Further, Mamuneas et al. (2015) did not find any differences in decision making accuracy between shy and bold three-spined sticklebacks (*Gasterosteus aculeatus*). In guppies, bold females learned faster and also more accurately (Trompf and Brown 2014). Hence, this species did not seem to underlie a speed-accuracy trade-off, but still expressed a cognitive syndrome, as the personality trait boldness correlated with the cognitive style accuracy.

As opposed to the relationship between personality and cognitive styles, we currently lack further theoretical predictions for the relationship between personality and cognitive ability in animals (Sih and Del Giudice 2012; Griffin et al. 2015). Like for fast behavioural types, stable environments may also select for cognitive abilities which involve routine formations (Guillette et al. 2011). This is because learning a task fast and then form a routine in how to solve it, has more advantages when the environmental conditions are constant. Indeed, proactive mice (*Mus musculus*) formed routines and reacted less to changes in a maze structure than reactive animals (Benus et al. 1990). Furthermore, in cases where environmental cues can be used for spatial learning, reactive individuals should react more to these cues, while proactive individuals should be less sensitive to external cues (Benus et al. 1987) and are expected to orientate internally driven (Mamuneas et al. 2015). This would result in a better ability of reactive individuals to solve learning tasks which rely on such environmental cues. Alternatively, since proactive (fast) individuals are more active and explore novel environments faster, they might also be able to learn spatial tasks faster. For instance, Guenther et al. (2014a) showed that boldness, activity and aggressiveness correlated positively with association learning ability in wild cavies (*Cavia aperea*). In starlings (*Sturnus vulgaris*), fast explorers were also faster in learning to obtain a food reward (Boogert et al. 2006). There is a growing interest in the individuality of cognitive abilities within animal species and indications for a relationship between personality and general cognitive ability accumulate, however, it seems hard to predict the direction of these relationships in specific species (Griffin et al. 2015).

Stability of Cognitive Styles

Like behavioural types, cognitive styles are assumed to show temporal repeatability and cross-context consistency (Sih and Del Giudice 2012). This is because behavioural and cognitive syndromes rely on the assumption that individuals constantly differ in their behavioural trait expressions. The rank order of individuals is assumed to be stable between situations, even if the mean behavioural trait expression might differ between tests (Sih et al. 2004b). These properties have been studied thoroughly for personality traits (Bell et al. 2009; Brommer and Class 2015), but so far, only few tests for repeatability and consistency of cognitive traits have been published (Ducatez et al. 2015; Griffin et al. 2015; Guillette et al. 2015; Wang et al. 2015).

Spatial Orientation in Cognitive Syndromes

Spatial orientation skills can be essential for a maximal individual fitness gain and are assumed to be favoured differently depending on each species' ecology (Shettleworth and Hampton 1998). Spatial learning refers to processes through which animals can encode information about the environment, can navigate through their habitat, and can recall locations of important stimuli within space (Floresco 2015). Like any other learning process it can be influenced by constant individual behaviour, as new situations are differently encountered, perceived, and assessed by different behavioural types, which can result in different learning outcomes (Sih and Del Giudice 2012). Furthermore, any learning event itself is part of the individual experience, which further regulates individuality of behaviour (Stamps and Groothuis 2010a). Therefore, spatial learning is one of the suitable candidates for studying cognitive syndromes.

Study Objectives and Hypotheses

Here, we investigated spatial cognitive traits in a laboratory population of the Eurasian harvest mouse (*Micromys minutus*; Pallas, 1771). We add to the scarce knowledge about repeatability of cognitive traits in animals focusing on spatial orientation abilities. We further present new evidence for behavioural and cognitive syndromes in a spatial orientation context.

The Eurasian harvest mouse lives in high grass vegetation of wetland areas, but may also occur in grain fields and ruderal areas (Feldmann 1984; Spitzenberger 1986; Feldmann 1997). It thus occupies a very different ecological niche compared to other European rodents (Piechocki 2001). We assume that this species is exposed to a high selection pressure on spatial orientation abilities due to its complex three-dimensional use of habitats. The hippocampus of mammals is involved in the processing of spatial information about the environment (Yaskin 2011). Interestingly, the hippocampus of harvest mice occupied 16.2 % of the telencephalon, which is, e.g., 4.6 % more than in laboratory mice (*Mus musculus*; 11.6 % hippocampal volume of the telencephalon, West 1990). This may indicate that the brain structure of harvest mice is specifically adapted to spatial orientation. We investigated individual differences in spatial recognition, spatial learning ability and decision style (speed vs. accuracy) in Eurasian harvest mice.

We used the previously described personality traits boldness, activity, and exploration (see Chapter 1) to test for a behavioural syndrome. We further established three new behavioural tests (Novel Environment, Scare Test, and Spatial Orientation Task). All personality and cognitive traits were tested for their repeatability in adult harvest mice. We then measured the correlations between personality traits, and between personality and spatial cognitive traits to test for a relationship between them. As variable environments may favour different behavioural types and cognitive styles in populations, it is likely that these also occur in our population because harvest mice inhabit unstable environments (Piechocki 2001). We expected that the personality traits boldness, activity and exploration correlate positively and form a fast-slow-behavioural syndrome (Titulaer et al. 2012). We assumed that mice with better spatial recognition would learn an orientation task faster due to improved spatial cognitive abilities. We further predicted that spatial recognition and spatial learning ability correlate with personality traits. As our cognitive test design was based on active exploration, we assumed a positive relationship between active personality types and spatial learning ability (Sih and Del Giudice 2012). Finally, we expected a speed-accuracy trade-off as suggested by Sih and Del Giudice (2012) and hypothesised that bolder, more active and more explorative individuals decide faster but less accurately in a spatial learning task.

Material and Methods – Behavioural and Cognitive Syndrome

Study Animals and Housing Conditions

The harvest mice tested in this study stemmed from our laboratory population whose founding individuals (N = 26) originated from four different zoo populations. All individuals were born in Tübingen between 2011 and 2014. Mice were housed in polycarbonate cages of 60 cm length, 40 cm width and 58 cm height. Individuals were either kept separately or in pairs of equal sex with water and food (hay, grain seeds, fresh fruit and vegetables) ad libitum. The back wall of the cage was covered by a coco coir mat for climbing. For environmental enrichment, cages were additionally equipped with an artificial nest, a running wheel, a paper tube, a wooden branch and a sheaf of wheat, oat and spelt. All mice were kept at constant temperature (22°C, range: 21.0°C to 23.5°C) and light-dark cycle (LD 12:12 h). Animal husbandry and behavioural tests (see below) were permitted by the Regierungspräsidium Tübingen – Referat 35, reference number ZO 2/11.

Behavioural Tests

Standard behavioural tests, originally established to test emotionality in laboratory strains of mice and rats, were modified and adapted to our study species. It was not possible to record data blind because our study involved focal animal observations, however, we included the identity of the observer as a confounding factor in the data analyses (see below).

We tested male and female adult harvest mice (age range: 57 to 743 days). We previously presented repeatability and consistency of activity, boldness and exploration in young and adult harvest mice of the same population (see Chapter 1). Since young mice expressed less repeatable spatial recognition behaviour measured in a Y Maze, we now analysed only the data from adult mice. 28 individuals were added to the previous data set and we established three new behavioural tests (Novel Environment, Scare Test, and Spatial Orientation Task) to measure more traits.

Open Field (OF)

We used a modified OF test (Archer 1973) to analyse activity and boldness as previously described in Chapter 1. Using automated video tracking (EthoVisionXT, Version 5, Noldus), we measured the total distance moved (in cm) during 4.5 min in a round arena and the time (in seconds) individuals spent at the inner part of the arena (unsafe area) as parameters for activity and boldness, respectively. We tested 96 adult harvest mice once and 90 of them also a second time after 28 to 101 days (mean = 72.5 days).

Novel Object (NO)

To analyse exploratory behaviour we used a NO test (Chitty and Shorten 1946). This test was conducted directly after the OF test to reduce further stress as animals were already habituated to the arena setting. We recorded exploratory behaviour manually for 5 min after the first contact with the novel object. We quantified the time (in seconds) animals spent in physical contact with the novel object. For further details see Chapter 1. We tested 69 mice once and 62 of them were tested a second time after 85 to 101 days (mean = 89.9 days).

Novel Environment (NE)

We established a NE test to quantify spatial exploration in harvest mice. Originally, the NE test was used by Verbeek et al. (1994) to analyse exploration in great tits (*Parus major*). We here simulated a novel environment by using a polycarbonate cage (***FIGURE 10***). This arena was similar to the home cages of the mice with a size of 58.5 cm length, 39.0 cm width and 69.5 cm height. An intermediate floor made of opaque polycarbonate was inserted at 36.0 cm height. Bottom and intermediate floor were connected by five tubes (4 cm in diameter). Each tube had an entrance (2 x 2 cm) at the bottom and was equipped with straws to ensure that mice could climb up and down through the tubes (see ***FIGURE 10***). Straws were replaced and all surfaces of the NE were cleaned with 70% ethanol after each test. We used two different arrangements of the tubes for the two trials. A trial lasted until animals visited all five tubes, but at least 10 min and at maximum 30 min. Mice were observed directly. We defined that a mouse had visited a tube if it climbed at least one body length up or down within a tube. Because during 24 of 119 observations (20.2 %) the mouse did not visit all five tubes, we used the latency (in seconds) until any four of the five tubes were visited by the mouse as a parameter for exploration. Eight individuals did not visit four tubes during 30 min and were scored with 1800 seconds. We tested 60 adult harvest mice once and 59 of them were tested a second time after 23 to 35 days (mean = 29.9 days).

FIGURE 10 Test arena of the Novel Environment showing the five tubes connecting bottom and intermediate floor which harvest mice could explore by climbing. Photo by Christoph Jäckle

Scare Test (ST)

We used the Scare Test (established by da Luz Correia 2013) to analyse the reaction towards disturbances in a familiar environment. We used the individual home cages (see ***FIGURE 6***) as testing arenas and animals were observed directly. During an active phase of the mouse (e.g., the mouse was moving around in its cage), the observer fully opened the sliding door of the cage and closed it again immediately. The animals instantly hid under the hay or behind the coco coir mat in the back of the cage. We measured the latency (in seconds) until they returned from their hiding places as a parameter for boldness. If a mouse did not return from the hiding place within 15 min, we aborted the trial and scored the animal with 900 seconds (N = 9 tests). 56 adult harvest mice were tested once and 38 of them were tested a second time after 2 to 218 days (mean = 78.3 days).

Y Maze (YM)

We quantified spatial recognition in an adapted YM arena (Montgomery 1955), which relies on the assumption that rodents explore novel environments more than already known environments (Hughes 1968). Known environments can be recognised through association with known object cues. In the YM arena, objects that were only visible from one arm of the arena enabled the animals to recognise whether they had previously visited a specific arm, based on spatial association. The YM test consisted of two trials. During the first trial, one of the arms was locked (unknown arm), in the second trial all three arms were accessible. For a more detailed description of the test see Chapter 1. We video tracked animals using EthoVisionXT software and measured the total distance moved (in cm) during the first 5 min of the second trial as a parameter for activity. The time (in seconds) spent in the unknown arm during the first 5 min of the second trial was recorded as a parameter for spatial recognition. We excluded one animal from further analyses as it ran straight into the known arm at the beginning of the trial and did not leave this arm again. We tested 96 adult harvest mice once and 89 of them were tested a second time after 40 to 99 days (mean = 76.1 days).

Spatial Orientation Task (SOT)

To evaluate individual spatial learning ability, as well as decision speed and accuracy, we established the *Spatial Orientation Task* (Zimmermann 2014). We used a round arena, 51 cm in diameter. An elevated six arm maze was placed inside the arena, 20 cm above the bottom. Mice reached the maze by climbing through a centrally placed tube (4 cm in diameter, see ***FIGURE 11***). We placed a green plastic box (5 x 5 x 5 cm) filled with hay at the outer end of each arm. Mice could enter only the target box via a 2 cm hole in the lid. The other five boxes had closed but perforated lids, so that olfactory cues were the same for each box. As boxes were opaque, mice could not see the holes in the lid while walking on the arms of the maze. The SOT consisted of 10 or 20 learning runs (run number) conducted on two consecutive days (test day; run 1-10 on the first day, run 11-20 on the second day). Each learning run ended 1 min after the mouse had entered the target box and left again (maximum: 10 min). We did not catch the animals in close vicinity to the target box to avoid any negative associations. During the runs mice were supposed to learn the location of a target box (***FIGURE 11***) which offered a hide. If mice did not reach the learning criterion (see below) within 10 runs on the first test day, we conducted further runs (11-20) on the next day. Between the learning runs, all surfaces were cleaned with 70% ethanol to remove olfactory cues. Optical cues attached to the wall of the arena (***FIGURE 11***) served as landmarks that the animals could use for orientation, and for association between the position of the target box and both cues on either side of the box. The position of the target box was the same for all mice tested in the first trial, but we changed cue positions randomly. The animals were filmed from above, while the observer recorded individual behaviour on a monitor placed out of sight of the mice.

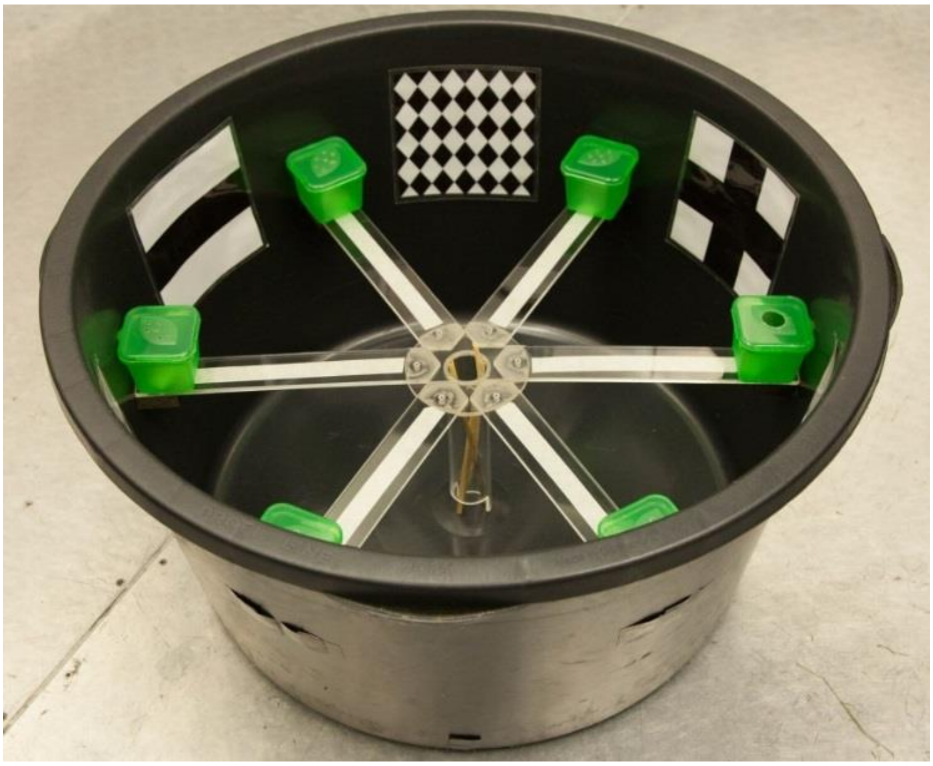

FIGURE 11 Experimental set-up of the Spatial Orientation Task. Six-arm maze elevated 20 cm above the ground with target box on the right and optical cues for spatial orientation between the arms. Photo by Uwe Zimmermann

We scored spatial learning ability as the number of learning runs needed to fulfil the learning criterion. We defined the learning criterion as less than seven non-target boxes visited in total, within four consecutive learning runs. This criterion was defined during a pilot study (Zimmermann 2014). If an individual did not reach the learning criterion, it was scored with 20 learning runs. After the learning criterion was fulfilled, all following runs (six to twenty runs per individual) were used to record decision speed and accuracy. We measured decision speed as the time (in seconds) between the moment when the mouse had reached the platform, and when it visited any box (all four paws on box/platform), multiplied by -1. If the animal visited the target box first, we scored an accurate decision. If any other box was visited first, we scored an inaccurate decision. The proportion of accurate first decisions within all learning runs *after* the learning criterion was fulfilled was defined as decision accuracy. During the second trial of the SOT (to estimate repeatability of spatial learning ability, decision speed and decision accuracy) we changed the set-up by changing the test room and the position of the target box within the six-arm maze. Further, we used six different optical cues than during the first trial. We tested 57 adult harvest mice once and 53 of them were tested a second time after 45 to 491 days (mean = 139.0 days).

Data Analyses

We performed all statistical analyses using *R* software, version 3.0.1 (R Core Team 2013) and ASReml, version 4.1 (Gilmour et al. 2014). We measured the effects of potential confounding factors due to the experimental set-up, and due to individual characteristics of the mice, by fitting linear mixed models (LMM) using the "nlme" package (Pinheiro and Bates 1996) in *R*. Potential confounding factors caused by the experimental set-up, that were regarded in the analyses, were the identity of the observer (observer), the date of the test performance (test date), and the time of the test performance (test time). Potential confounding individual factors were the sex of the mouse (sex), whether the mouse completed a specific test for the first or the second time (trial number), the body mass of the mouse measured directly before the test performance (body mass), the age in days of the mouse at the test date (age), the lab generation the mouse belonged to (our lab population consisted of seven generations), and whether the mouse was housed alone or in a pair (housing condition). We thus included the following confounding factors into each full model: The mouse identity (ID) as random factor (to account for repeated measures); sex, trial number, observer and housing condition as fixed factors; body mass, age, test date, test time and generation (quadratic function) as fixed covariates. As males and females might differ in their activity rhythms, we included the interaction sex*test time. Further, females harvest mice are larger than males (Piechocki 2001) and this dimorphism might influence individual behaviours of sexes differently. We therefore included the interaction sex*body mass in the full model. Additionally, we included the run number and the test day (one or two) to the linear models fitting decision speed and accuracy of the SOT. We used backward stepwise reduction of the full model by excluding non-significant interactions first, followed by non-significant main effects ($p < 0.05$). We applied square root (activity in OF and YM, exploration in NO, boldness in OF and spatial recognition) or log (boldness in ST, exploration in NE and spatial learning ability) transformations to the dependent variables to ensure a normal distribution of the model residuals. For boldness in ST, exploration in NE, and spatial learning ability we multiplied the transformed values by -1 to simplify correct interpretations of the directions of correlation coefficients (i.e. larger values correspond to a faster exploration, faster learning, and bolder individuals). We present final models with the remaining confounding factors in the **SUPPLEMENTARY TABLE 1**. Confounding factors with significant effects were retained in the models for subsequent repeatability and correlation analyses.

Repeatability of Personality and Cognitive Traits

The narrow sense repeatability of personality and cognitive traits was analysed using the package "rptR" (Nakagawa and Schielzeth 2010) in *R*. We calculated *adjusted repeatability* (R_A) (Nakagawa and Schielzeth 2010) by including confounding factors identified from LMMs before (see above). We estimated LMM based repeatability, including individual ID as random factor, based on 1000 bootstrapping runs and 1000 permutations. We display R_A values with standard errors and asymptotic 95% confidence intervals (CIs), and permutation based p values.

Correlations between Traits

We fitted multivariate LMMs using restricted maximum likelihood (REML) in ASReml4 (Gilmour et al. 2014) to estimate phenotypic variances for each personality and cognitive trait, as well as the phenotypic covariances between those. We partitioned the within-individual variances and covariances from the between-individual variances and covariances as recommended by Dingemanse and Dochtermann (2013). We then used these variances and covariances to calculate phenotypic correlations between the traits within individuals (r_W) and between individuals (r_B). We included all previously defined significant confounding factors in the fixed models. The animal ID was included as random factor to all models.

We fitted the first multivariate model for the six personality traits to test for a behavioural syndrome. We used a stepwise approach to build up the final model. First, we applied a diagonal variance model, which estimated only variances, but no covariances. Then, we applied an unstructured variance model where all between-individual and within-individual covariances were first fixed to zero. We then estimated the between-individual and within-individual covariances one by one, using estimates from respective bivariate models as starting values. We present correlations based on the final six-trait model with a fully unstructured between-individual variance-covariance matrix. The within-individual covariance matrix was modelled between activity in the OF test, activity in the YM, boldness in the OF test, and exploration in the

NO test. For boldness in the ST and exploration in the NE, we were not able to model within-individual covariances, as those behaviours were measured at different time points. Thus, our modelling approach is a combination of scenario 3 and 4 described in Table 2 by Dingemanse and Dochtermann (2013).

We extended the six-trait model by adding spatial recognition and spatial learning ability to test for a relationship between personality and spatial cognitive traits. We also calculated correlations based on the final eight-trait model with a fully unstructured between-individual variance-covariance matrix. Within-individual covariances with spatial learning ability in the SOT could not be estimated, as this behaviour was measured at a different time point.

Finally, we extended the six-trait model by adding decision speed and decision accuracy to test for a speed-accuracy trade-off and for a cognitive syndrome. We again calculated correlations based on the final eight-trait model with a fully unstructured between-individual variance-covariance matrix. The within-individual covariance matrix was modelled between activity in the OF test, activity in the YM, boldness in the OF test, and exploration in the NO test. From these eight-trait models we only display between-individual correlation coefficients with $p < 0.1$, the full information is given in the Appendix.

In the above described analyses, our measure for decision accuracy summarized accuracy over all learning runs after the learning criterion was fulfilled, because we were not able to conduct multivariate generalized LMMs. However, since ASReml4 allows bivariate models with one binominal response variate, we conducted an additional test for the speed-accuracy trade-off. In this analysis, decision accuracy was scored as a binary response (correct decision or not) for each learning run and was modelled with a logit link. We then calculated between- and within-individual correlations from fully unstructured between- and within-individual variance-covariance matrices. Potential confounding fixed factors (as described above) were identified before by fitting a generalized LMM using the “lme4” package (Bates et al. 2014) in *R*. The trial number (estimate ± SE = -0.242 ± 0.123, $p = 0.049$) and the test day (-0.282 ± 0.124, $p = 0.023$) significantly influenced the decision accuracy and remained in the generalized LMM.

Results – Behavioural and Cognitive Syndrome

Repeatability of Personality and Cognitive Traits in Harvest Mice

All personality traits were significantly repeatable (**TABLE 7**). R_A values ranged between 0.221 and 0.598, whereby activity was the behaviour with the highest repeatability. Most cognitive traits were also significantly repeatable: Spatial recognition, spatial learning ability, and decision speed showed repeatabilities between 0.127 and 0.263 (**TABLE 7**). However, decision accuracy was not significantly repeatable. The results of the model reductions can be found in the **SUPPLEMENTARY TABLE 1**. Significant confounding factors were retained in the fixed model for all further analyses and are listed in **TABLE 7**.

In the Spatial Orientation Task most harvest mice were able to learn the position of the target box. 53.6 % of the animals reached the learning criterion already within the first five runs. However, 5.5 % of the individuals did not reach the learning criterion within the 20 learning runs.

TABLE 7 Repeatability (R_A) with 95% confidence interval (CI) for each personality and cognitive trait. Generation was fitted as a quadratic function. Sample sizes (N) are given as observations/individuals. Significant repeatability values are marked bold.

personality/cognitive trait	**behavioural test**	**confounding factors**	**N**	**R_A**	**95% CI**	**p value**
activity	Open Field	generation + generation^2	186/96	**0.404±0.086**	[0.226, 0.552]	< 0.001
	Y Maze	-	184/96	**0.598±0.064**	[0.456, 0.705]	< 0.001
boldness	Scare Test	-	94/56	**0.256±0.145**	[0.000, 0.534]	< 0.001
	Open Field	trial number + observer	186/96	**0.319±0.097**	[0.115, 0.502]	< 0.001
exploration	Novel Object	housing condition	131/69	**0.328±0.113**	[0.097, 0.531]	< 0.001
	Novel Environment	trial number	119/60	**0.221±0.119**	[0.000, 0.432]	< 0.001
spatial recognition	Y Maze	-	184/96	**0.200±0.104**	[0.000, 0.429]	0.002
spatial learning ability	Spatial Orientation Task	trial number	110/57	**0.127±0.116**	[0.000, 0.382]	0.042
decision accuracy	Spatial Orientation Task	-	102/56	0.125±0.122	[0.000, 0.411]	1.000
decision speed	Spatial Orientation Task	observer + run number	1156/56	**0.263±0.027**	[0.211, 0.313]	< 0.001

Correlations between Traits in Harvest Mice

We observed four significant positive between-individual correlations among personality traits, three more relationships showed a tendency ($p < 0.1$) with high positive between-individual correlation coefficients (**FIGURE 12**). The two measures of activity showed a significant positive between-individual correlation ($r_B = 0.541 \pm 0.146$, $p < 0.001$). Both activity measures correlated significantly with boldness measured in the Scare Test, but not with boldness measured in the Open Field test. However, exploration in the Novel Environment correlated significantly with boldness in the Open Field test. There were two further relationships between boldness and exploration, which tended to correlate positively (see **FIGURE 12**). Finally, exploration in the Novel Environment tended to correlate positively with activity in the Open Field test. The two measures of exploration showed no significant between-individual relationship, nor did the two measures of boldness.

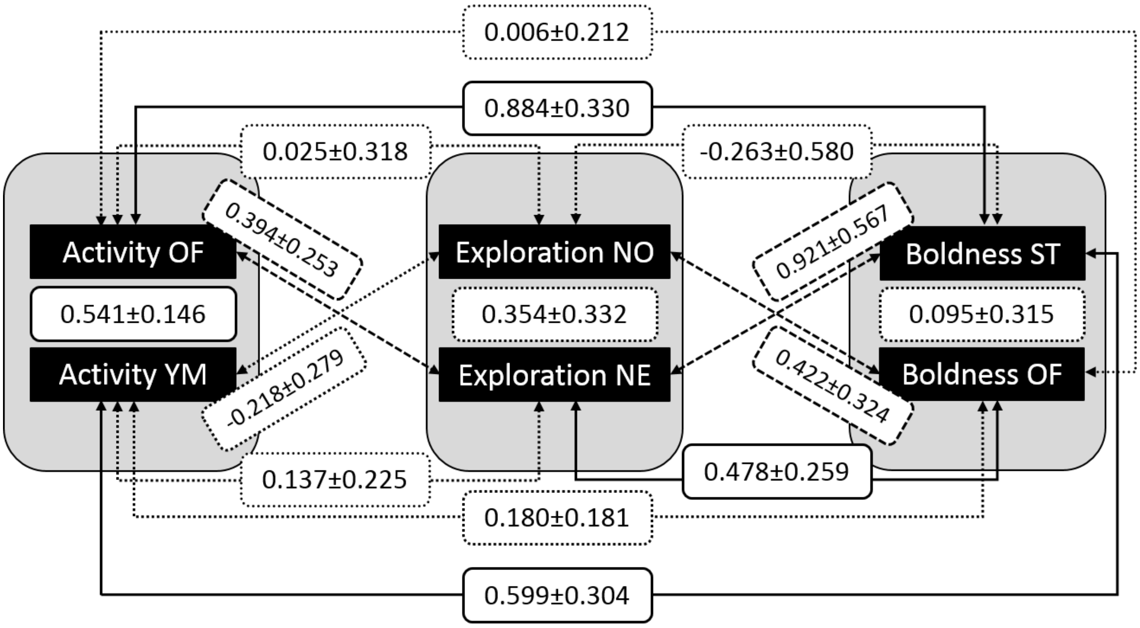

FIGURE 12 Behavioural syndrome: Between-individual correlations (± standard errors) between personality traits calculated from a multivariate mixed model. Solid lines indicate significant between-individual correlations (p < 0.05), dashed lines indicate tendencies (p < 0.1), and dotted lines indicate non-significant between-individual correlations. OF: Open Field, YM: Y Maze, NO: Novel Object, NE: Novel Environment, ST: Scare Test.

We observed only one significant within-individual correlation between the activity in the Open Field test and activity in the Y Maze ($r_W = 0.257 \pm 0.108$, $p = 0.009$). All other modelled within-individual correlation coefficients were very small and not significant (see **TABLE 8**).

TABLE 8 Behavioural Syndrome: Within-individual correlations between personality traits calculated from a multivariate mixed model. Within-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. Significant correlation is marked in bold. NA: within-individual correlation not modelled due to missing data. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	activity YM	boldness OF	boldness ST	exploration NO	exploration NE
activity OF		**0.257±0.108**	-0.006±0.106	NA	0.082±0.126	NA
activity YM	0.009		-0.040±0.116	NA	0.065±0.125	NA
boldness OF	0.477	0.364		NA	0.062±0.125	NA
boldness ST	NA	NA	NA		NA	NA
exploration NO	0.256	0.301	0.311	NA		NA
exploration NE	NA	NA	NA	NA	NA	

When we extended this six-trait model by spatial recognition and spatial learning ability, our conclusions from the between-individual and within-individual correlations among personality traits did not change (see **SUPPLEMENTARY TABLE 2** and **SUPPLEMENTARY TABLE 3**). In the eight-trait model, we identified a strong and significant between-individual correlation among activity in the Open Field and spatial learning ability (rB = 0.793, see **FIGURE 13**) and a tendency for a strong and positive between-individual correlation among spatial learning ability and boldness in the Scare Test (rB = 1.696, see **FIGURE 13**). Further non-significant between-individual and within-individual correlation coefficients are not displayed in **FIGURE 13**, but all results of the multivariate mixed model can be found in the **SUPPLEMENTARY TABLE 2** and **SUPPLEMENTARY TABLE 3**. There was no significant between-individual correlation, nor any within-individual correlation between spatial recognition and any personality trait. The between-individual correlation coefficient between spatial learning ability and spatial recognition was large and positive (rB = 0.562 ± 0.594), but not significant (p = 0.172).

When we extended the six-trait model by decision speed and decision accuracy, between-individual and within-individual correlations among personality traits did also not change meaningfully (see **SUPPLEMENTARY TABLE 4** and **SUPPLEMENTARY TABLE 5**). In this eight-trait model, we identified a significant negative between-individual correlation between boldness in the Open Field and decision speed (r_B = -0.412 ± 0.179, see **FIGURE 14**). Further non-significant between-individual and within-individual correlation coefficients are not displayed in Figure 5, but all results of the multivariate mixed model can be found in the **SUPPLEMENTARY TABLE 4** and **SUPPLEMENTARY**

TABLE 5. There was no significant between-individual correlation among decision speed and decision accuracy ($r_B = 0.207 \pm 0.425$, $p = 0.313$).

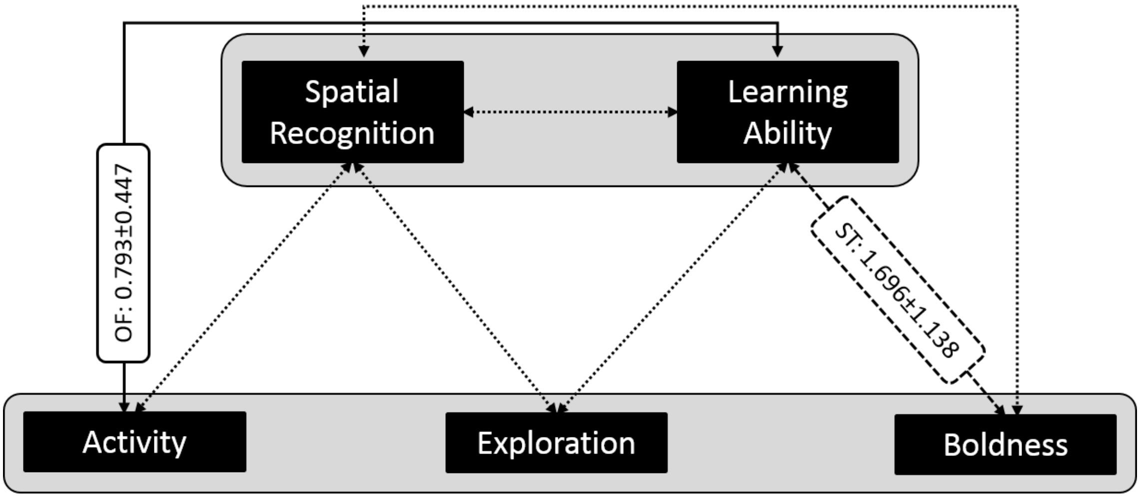

FIGURE 13 Relationship between personality and spatial cognition: Between-individual correlations (± standard errors) between personality traits and cognitive traits calculated from an eight-trait multivariate mixed model, but only five traits are displayed (see explanation in the text). The solid line indicates the significant correlation ($p < 0.05$) between activity in the Open Field (OF) and learning ability in the Spatial Orientation Task, the dashed line indicates the tendency ($p < 0.1$) between boldness in the Scare Test (ST) and learning ability in the Spatial Orientation Task, and dotted lines indicate non-significant correlations between the other traits.

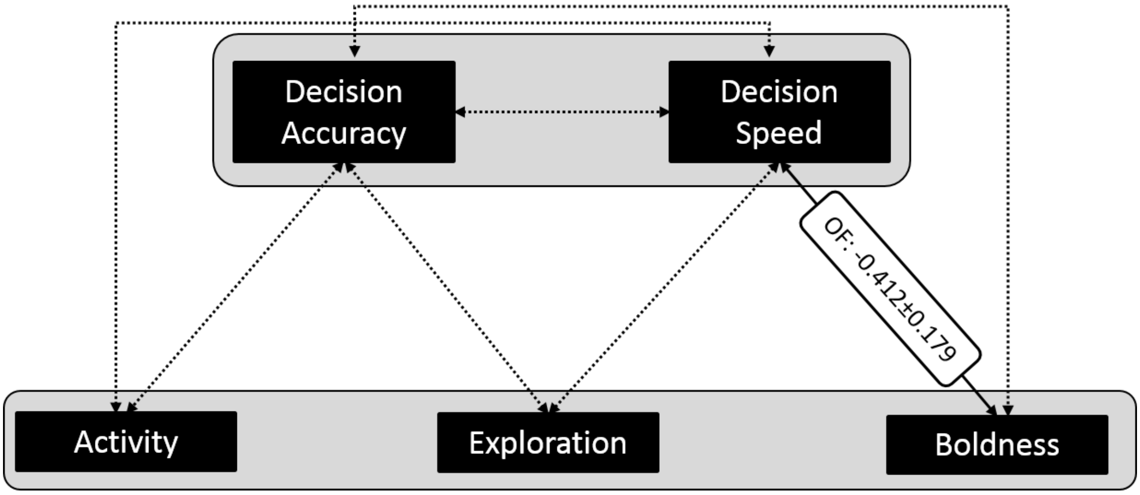

FIGURE 14 Test for a cognitive syndrome based on a speed-accuracy trade-off: Between-individual correlations (± standard errors) between personality traits, and decision speed and accuracy calculated from an eight-trait multivariate mixed model, but only five traits are displayed (see explanation in the text). The solid line indicates the significant correlation ($p < 0.05$) between activity in the Open Field (OF) and decision speed in the Spatial Orientation Task, dotted lines indicate non-significant correlations between the other traits.

The bivariate generalized LMM with decision accuracy as binary response and decision speed with normal distribution resulted in a within-individual correlation coefficient of $r_W = 0.023 \pm 0.020$ (p = 0.125) and a between-individual correlation coefficient of $r_B = 0.018 \pm 0.248$ (p = 0.471).

Discussion - Behavioural and Cognitive Syndrome

We found strong evidence for a behavioural syndrome in Eurasian harvest mice. As we expected, activity, boldness and exploration showed positive correlations and formed a fast-slow-behavioural syndrome (Titulaer et al. 2012). Further, we found some indication for a relationship between spatial learning ability and personality types. More active harvest mice reached the learning criterion in the Spatial Orientation Task earlier than less active individuals. The speed-accuracy trade-off was not apparent within, nor between individuals. Nevertheless, we found weak evidence for a cognitive syndrome as shyer harvest mice made decisions faster than bolder mice. Thus, our data partly support the hypothesis that behavioural types correlate with individual cognitive ability (Sih and Del Giudice 2012).

Repeatability of Personality and Cognitive Traits in Harvest Mice

One basic assumption of animal personality is the repeatability of behaviours (Réale et al. 2007). We here confirmed this assumption for all personality traits tested in adult Eurasian harvest mice. R_A values were between 0.221 and 0.598, which lies within the range of usually observed repeatabilities of personality traits (Bell et al. 2009; Garamszegi et al. 2013). We previously reported similar repeatabilities for a smaller dataset on juvenile and adult harvest mice from the same laboratory population (see Chapter 1).

Spatial recognition and spatial learning ability were also significantly repeatable in adult harvest mice. Lantová et al. (2011) already observed that common voles (*Microtus arvalis*) performed repeatably in an eight-arm radial arm maze. Voles thereby showed repeatable maze-exploring tactics and also the maze exploration activity was significantly repeatable (Lantová et al. 2011). Currently, there is only scarce evidence for repeatability of spatial recognition and general learning ability in animals (Griffin et al. 2015). However, it was shown that individuals behaved consistently in different types of learning tasks and solved these tasks consistently well (e.g., Ducatez et al. 2015; Guillette et al. 2015). Repeatability and consistency of cognitive traits results in less flexible behaviour of individuals in situations when cognitive abilities are needed. This may cause a constraint to the expression of adaptive behaviour. It should be noted, however, that studies on within-species variation in cognitive abilities (including our study) generally focus on few test situations for specific cognitive tasks. Individuals with low performance in those tasks may show stronger cognitive abilities in other tasks. It remains to be shown, which trade-offs cause repeatable reduced cognitive ability in a specific test.

Among the two traits that characterise cognitive styles (Sih and Del Giudice 2012), decision speed was repeatable in harvest mice, whereas decision accuracy was not. The only other published study on repeatability of decision speed and accuracy was conducted on zebrafish (*Danio rerio*), where Wang et al. (2015) showed that both, speed and accuracy, were significantly repeatable in a spatial colour discrimination task. Harvest mice may indeed not behave as repeatably during decisions as zebrafish did, or we may have failed to measure decision accuracy adequately. Our test design was based on the assumption that the mice have a high motivation to inspect the target box (their hide in the test arena) as soon as they were released into the arena. However, after a few runs, some mice may not have been motivated any more to visit the hide, but rather preferred to explore the remainder of the test arena. As there is no other evidence available so far, more experimental work is needed to better understand the repeatability of decision accuracy and speed in animals in general. If relationships between personality types and cognitive styles continue to support the idea of cognitive syndromes (Sih and Del Giudice 2012), the test of the basic assumptions of repeatability and consistency of the involved behaviours should become routine ingredients to studies in this field.

Behavioural Syndrome in Harvest Mice

We found significant positive between-individual correlations or positive tendencies between the three studied personality traits. Activity, exploration and boldness thus form a fast-slow-behavioural syndrome (Titulaer et al. 2012) in our study species. This is in line with a large body of personality literature in rodents: Starting with the observations in laboratory house mice (Koolhaas et al. 1999), more and more studies reported fast-slow-behavioural syndromes (e.g., in Belding's ground squirrels, *Urocitellus beldingi*, Dosmann et al. 2015; and in cavies, *Cavia aperea,* Guenther et al. 2014a). However, some behavioural correlations were not stable over ontogeny (e.g., Kanda et al. 2012; Guenther et al. 2014b), and in some species, no behavioural syndrome was detected (e.g., in yellow-bellied marmots, *Marmota flaviventris*, Petelle et al. 2013).

In our study, a number of tested relationships between personality traits showed reasonable between-individual correlation coefficients, but large standard errors. This suggests that our current sample size (56 to 96 individuals per trait) might be too small to fully describe the relationship between all measured behaviours. Alternatively, within-individual covariance might, if not estimated, mask or inflate between-individual covariance. Only between-individual covariances are indicative for a behavioural syndrome (Dingemanse and Dochtermann 2013). Since we did not measure all traits at the same time point, we were not able to model all within-individual covariances. However, among those relationships, where we did estimate within-individual covariance, it was significant only between the two measures of activity. In that case, an alternative model excluding within-individual covariances did not lead to different conclusions about the relationships between the traits (data not shown). We thus conclude that, in our test situation, within-individual covariances did not largely influence phenotypic correlations, and that higher sample sizes might allow to estimate correlations with higher accuracy. Further, the lack of significant correlations between the two measures of explorative behaviour (Novel Object and Novel Environment), and between the two measures of boldness (Scare Test and Open Field) may indicate a suboptimal behavioural test selection. These tests may indeed measure different behaviours. Open Field and Novel Object tests are frequently used to measure either exploration, activity or boldness. Different experimenters thereby apply different methods to quantify the measured behaviours (e.g., boldness as the latency to reach novel objects, Guenther et al. 2014a; or boldness as the latency to enter the middle of an Open Field, Herde and Eccard 2013). However, since behavioural syndromes assume relationships between similar behavioural traits in different situations, we would still expect correlations between slightly different measures of boldness and exploration. We cannot exclude that larger sample sizes may reveal correlations between these measures. Currently, less individuals were tested in the Novel Environment and in the Scare Test, compared to the Novel Object and the Open Field, respectively.

We detected positive correlations between activity, exploration and boldness. These relationships could arise from constraints in proximate mechanisms, such as physiological pathways, maintaining these behaviours (Sih et al. 2004a). Alternatively, selection for alternative types may maintain fast and slow behavioural types in a population. This type of disruptive selection can be caused by variation in selective agents in the environment (e.g., fluctuation of food availability in great tits, *Parus major*, Dingemanse et al. 2004), or by alternative behavioural optima during different life-history stages (e.g., life-history trade-off between current and future reproduction in grey mouse lemurs, *Microcebus murinus*, Dammhahn 2012). Harvest mice live in a very variable environment where it is likely that fitness advantages for different behavioural types change during the year. Like in other rodent species, population sizes of harvest mice increase dramatically during the summer and show high peaks in autumn, followed by a marked reduction in population size over winter (Piechocki 2001). More active, bolder and more explorative individuals may have higher fitness in the more competitive situations during high population density. Then, the fast behavioural type may gain better access to suitable nest sites, food and mating partners. Slow (reactive) harvest mice on the other hand may have fitness advantages during winter and spring, when population density is lower. During this time, the slow behavioural type may save energy through lower levels of activity and exploration, and potentially fewer interactions with conspecifics. However, selection for behavioural types will only result in the maintenance of these types in the population if the correlations that underlie the behavioural syndrome are in fact genetic relationships. We here present phenotypic trait correlations, and it remains to be tested if these relationships are also genetically based.

Cognitive Syndrome in Harvest Mice

In line with our expectation, we found some indication for a relationship between spatial learning ability and personality types. Harvest mice that were more active in the Open Field reached the learning criterion in the Spatial Orientation Task earlier than less active individuals. Further, a non-significant tendency suggested a positive relationship between boldness in the Scare Test and spatial learning ability. Finally, the between-individual correlation coefficient between spatial learning ability and boldness in the Open Field test showed a large correlation coefficient, with large standard error (**SUPPLEMENTARY TABLE 4**).

We expected that mice which learned the orientation task faster, would also show better spatial recognition. Although the between-individual correlation coefficient between spatial recognition and learning ability was large and positive, it did not reach significance due to the large standard error (**SUPPLEMENTARY TABLE 4**). This large error variance may occur because individuals differed in their motivation to solve the orientation task. If some mice were indeed more interested to explore the six arm maze rather than to inspect the target box, we have to assume a large error in our measure of learning ability. The challenge remains to design a spatial learning task that ensures high task solving motivation in all individuals without restricting the test situation to a feeding context.

We did not observe any of the expected correlations between personality traits and spatial recognition in the Y Maze. The time animals spent in the unknown arm of the Y Maze (a measure of the association between unknown landmarks and a new environment) was not related to activity, exploration or boldness of the individuals, as measured in other behavioural tests. Mamuneas et al. (2015) could also not identify any differences in the spatial recognition between shy and bold three-spined sticklebacks (*Gasterosteus aculeatus*) tested in a T Maze where landmarks could be associated with food rewards in one of the arms. Even if behavioural types do not differ in their spatial recognition, they may still use different, equally successful strategies to recognize known areas or to build associations with specific optical cues. In our test for spatial recognition, mice could recognize the unknown arm based on landmarks that we placed around the arms. Thereby, the animals' view was blocked to the side of the Y Maze, but they could have used cues above them (e.g., the camera, or structures in the ceiling of the room) to define a known position in the room. However, mice may also have recognized the unknown arm based on path integration – by keeping track of their own location in relation to a known position in their environment (Shettleworth 2010). We were not able to test if mice of different personality types employed different orientation strategies. There is indeed evidence for this in common carp (*Cyprinus carpio*) where different personality types solved an orientation task using different types of information to get access to a food reward (Mesquita et al. 2015). Reactive (slow) individuals followed a light cue that was associated with food, while proactive (fast) animals formed fixed movement routines to reach the location of the food reward (Mesquita et al. 2015).

Taken together, we found some weak indication that mice of different personality types may differ in their ability to learn. Some recent studies actually suggested such a relationship: Fast behavioural types demonstrated better conditioning learning in Panamanian bishop fish (*Brachyrhaphis episcopi*) and better association learning in cavies (*Cavia aperea*, DePasquale et al. 2014; Guenther et al. 2014a). The difference in (spatial) learning ability is assumed to result from differences in how individuals assess and attend to the learning situation (Sih and Del Giudice 2012). However, no such relationship was reported in Eastern water skinks (*Eulamprus quoyii*) and in common carp (*Cyprinus carpio*), where bold as well as shy animals learned spatial association tasks equally successful (Carazo et al. 2014; Mesquita et al. 2015).

A cognitive syndrome could arise if individuals face a trade-off between fast and accurate decisions (Sih and Del Giudice 2012). In this study, we did not observe significant correlations between decision speed and decision accuracy. Thus, harvest mice that decided more quickly, did not made more or less accurate decisions than slow deciding individuals, indicating that harvest mice did not face a speed-accuracy trade-off in the Spatial Orientation Task. We will need to design more specific tests for decision speed and accuracy, to measure the correlation between them in further contexts. Decision speed and accuracy did not correlate significantly with any personality trait, with one exception: Shy harvest mice which spent less time in the unsafe part (middle) of the Open Field decided faster in the Spatial Orientation Task. This finding is in contrast to the cognitive syndrome hypothesis (Sih and Del Giudice 2012), where slow (and shy) behavioural types are expected to make slow, but accurate decisions. In our specific set-up, shy mice (that spent less time in the middle of the Open Field) might have preferred to leave the exposed position in the middle of the Spatial Orientation Task arena faster, than bold individuals, and thus expressed a faster decision behaviour in that test. Under a speed-accuracy trade-off, this behaviour could indeed lead to a lower spatial learning ability due to more frequent inaccurate decisions. Our data did not exclude the possibility that less active individuals may actually have decided less accurately: The two relevant correlation coefficients between decision accuracy and activity were large and positive, albeit not significant due to high variation in the data (reflected in large standard errors, see **SUPPLEMENTARY TABLE 4**). However, overall, we found little support for the hypothesis that harvest mice show a relationship between behavioural types and cognitive styles.

So far, few studies have investigated speed-accuracy trade-offs and cognitive syndromes, and results are contradictory. Speed-accuracy trade-offs were identified in wild-caught Carib grackles (*Quiscalus lugubris*), in wild-caught black-capped chickadees (*Poecile atricapillus*), and in wild Florida scrub-jays (*Aphelocoma coerulescens*, Ducatez et al. 2015; Guillette et al. 2015; Bebus et al. 2016). On the other hand, Bousquet et al. (2015) could not identify any effect of exploration on the accuracy in a spatial learning task in captive mallards (*Anas platyrhynchos*). Speed-accuracy trade-offs were also not consistently identified in fish (three-spined sticklebacks, *Gasterosteus aculeatus* and guppies, *Poecilia reticulata*, Trompf and Brown 2014; Mamuneas et al. 2015). While Burns and Rodd (2008) observed such a trade-off in guppy populations that were exposed to high predation pressure, Trompf and Brown (2014) could not confirm the trade-off in guppies in the laboratory, where fish may have habituated to the absence of predators (Trompf and Brown 2014). This might also be the case in our study. The tested harvest mice originated from laboratory populations, which did not experience any natural predation for several generations. It would thus be interesting to measure decision speed and accuracy in harvest mice from wild populations. Further, the experimental design of the Spatial Orientation Task may not imitate predation risk for harvest mice sufficiently, so that they were actually not forced to decide between speed and accuracy. If there is no need to hide (escape to the target box) because of missing perceived predation risk, mice might have known the position of the target box but may not have been motivated to instantly seek shelter. Future experiments may use a simulation of a predator, appearing above the Spatial Orientation Task arena, to imitate predation risk and thus motivate animals to reach the target box.

Conclusion - Behavioural and Cognitive Syndrome in Harvest Mice

Behavioural and Cognitive Syndrome in Harvest Mice of the Laboratory Population

Our results showed that adult Eurasian harvest mice behaved repeatably and that they expressed a behavioural syndrome with strong positive correlations between the three personality traits activity, exploration, and boldness. Further, we provide new insights into the relationship between personality traits and spatial learning abilities. More active and bolder individuals (the fast behavioural type) were faster in learning the Spatial Orientation Task than mice expressing the slow behavioural type. Thus, we could show that a specific cognitive ability correlated with behavioural types, which supports the hypothesis by Sih and Del Giudice (2012). If fast behavioural types in general outperform slow individuals in cognitive tasks, we should wonder why both types still persist in animal populations. Although slow learning harvest mice may not have any fitness advantages, shy and less active harvest mice may indeed be the more adapted behavioural type during some parts of the year. Thus, the observed relationship may cause non-independent trait evolution and maintain slow learners in the population. Spatial recognition, however, did not correlate with any personality trait. In addition, we only found weak indications for a cognitive syndrome: Shyer mice decided more quickly in the Spatial Orientation Task, but no other between-individual correlations among personality traits and decision styles were significant (despite some large correlation coefficients). We also found no support for a speed-accuracy trade-off. This may indicate that our test situation did not reveal these relationships sufficiently, and that any underlying trade-offs would appear more clearly in an improved test situation (e.g., under time constraint, or with mice from a population under predator pressure). On the other hand, if harvest mice do indeed not express a cognitive syndrome, decision styles and behavioural types could evolve independently from each other. As current results (including this study) are contradictory, more investigations are needed to further investigate the presence and the evolutionary significance of cognitive syndromes in animals.

Chapter 3

In Chapter 3 we tested parts of the described assumptions I and II of animal personality (behavioural repeatability and correlations between behavioural traits) in a field population of harvest mice. Further, we compared the results with what we have observed in the laboratory population of harvest mice (Chapter 1 and 2).

Animals show constant behavioural types in many species. Further, personality traits form behavioural syndromes which means that different behaviours correlate within populations. Behavioural syndromes were assumed to be stable across time and space, as the correlations between traits can be constrained by hormonal, physiological, or genetic factors. But there is growing evidence that these correlations between behaviours appear more flexible. Phenotypic correlations can vary between populations of the same species originated from different environments, for instance, laboratory and field populations. We described a behavioural syndrome in a laboratory population of Eurasian harvest mice (*Micromys minutus*) in Chapter 1. Now, we investigated whether three personality traits, as well as one spatial cognition trait also correlate in a population with a natural genetic background, kept in a semi-natural outdoor enclosure. We further compared behavioural correlations within the field population with observations of harvest mice bred in the laboratory. Our results showed that exploration, activity and boldness were repeatable in the field population of harvest mice. However, spatial recognition measured in a Y Maze was not repeatable. Harvest mice of the field population showed a behavioural syndrome as we observed positive between-individual correlations among the three behaviours activity, exploration and boldness. Although we observed significant differences in the magnitude of phenotypic correlations between the laboratory and the field population, both datasets suggested the same directions of correlations within the syndrome. Hence, we conclude that Eurasian harvest mice in general exhibit a behavioural syndrome along a fast-slow continuum, and that proximate factors are more likely to shape this behavioural syndrome than selection processes.

Behavioural Syndrome in the Field Population of Harvest Mice

Animal Personality and Behavioural Syndromes

Constant between-individual differences in behaviour are known as animal personality, coping styles or temperament (Wilson et al. 1994; Koolhaas et al. 1999; Gosling 2001; Réale et al. 2007).

Correlations between these constant behavioural traits (personality traits) were defined as behavioural syndrome (Sih et al. 2004b).

Behavioural Syndromes in Laboratory and Field Populations

Behavioural syndromes were assumed to be stable across time and space (Fischer et al. 2016) as the correlations between traits can be constrained by hormonal, physiological, or genetic factors (Bergmüller 2010). But there is growing evidence that these correlations between behaviours appear more flexible (Fischer et al. 2016). For instance, phenotypic correlations can vary between populations of the same species originated from different environments (Bell 2005; Dingemanse et al. 2007). Behavioural syndromes can also evolve differently between populations (Fischer et al. 2016). This was shown in different populations of Trinidadian guppies (*Poecilia reticulate*), originating from low- and high-predation environments. Behavioural variance increased when fish were reared under non-native environmental conditions compared to fish reared in native conditions. Further, behavioural correlations differed between the groups depending on the rearing condition and genetic background (Fischer et al. 2016). The authors concluded that the behavioural syndrome found in Trinidadian guppies is plastic and can evolve differently between populations of different environments (Fischer et al. 2016). Different magnitudes of correlations or even opposing relationships between personality traits were found within the same species. For example, a strong and positive relationship between exploration and aggression was observed in several populations of great tits (*Parus major*, Verbeek et al. 1996; Dingemanse and de Goede 2004, but not in others Carere et al. 2005). Correlations between activity, exploration, aggression and risk-taking behaviour were detected in several populations of three-spined sticklebacks (*Gasterosteus aculeatus*) depending on the predation pressure (Bell 2005; Dingemanse et al. 2007), but in a study on different stickleback populations, Brydges et al. (2008) did not detect these patterns. The structure of the phenotypic correlations between personality traits can also change over lifetime as shown by Wuerz and Krüger (2015) in zebra finches (*Taeniopygia guttata*). Ontogenetic changes and developmental aspects could thereby be mechanisms underlying behavioural syndromes (Wuerz and Krüger 2015). In basal tarantulas (*Brachypelma smithi*), for example, correlations between behaviours only occurred in spiders which were reared under naturalistic conditions and not in animals reared in a restricted housing treatment (Bengston et al. 2014). Thus, the environmental conditions during development may be an important factor shaping the occurrence of phenotypic correlations later in life.

There is an enormous body of literature on constant individual behavioural differences in captive bred or laboratory animal populations, and also the number of studies on wild populations has increased rapidly in the last decade. Investigations of animal personality in wild populations are indispensable to understand fitness consequences of behavioural types and the role of individual behaviour in local adaptation (Archard and Braithwaite 2010). In wild populations, constant behavioural differences were already linked to differences in reproductive success (Reale et al. 2000; Both et al. 2005) and survival (Dingemanse et al. 2004; Boon et al. 2007). A meta-analysis by Smith and Blumstein (2008) on laboratory and wild populations further revealed that bolder and more aggressive individuals had on average a higher reproductive success, but more exploratory individuals survived longer. Analyses of animal personality in captive populations have many limitations due to the effects of habituation and domestication, and because natural selection processes are relaxed in laboratory populations (Archard and Braithwaite 2010). In a population that lives under relaxed natural selection over several generations, individual behaviour might change, such as might behavioural differences between individuals and correlations between behavioural traits (Archard and Braithwaite 2010). For instance, in pumpkinseed sunfish (*Lepomis gibbosus*), wild-caught animals showed individual differences in boldness, but these differences disappeared after around 20 days post capture when kept in the laboratory under social isolation (Wilson et al. 1993). In old-field mice (*Peromyscus polionotus subgriseus*), McPhee (2003) showed that predator avoidance decreased with generations in captivity, because the captive population did no longer experience predation. The variance in predator-response behaviour increased with generations in captivity (McPhee 2003). This may influence the repeatability of this behaviour which is calculated as the proportion of between-individual variance relative to the total phenotypic variance in a population (Dingemanse et al. 2010). In a comparative study by Brust and Guenther (2015), guinea pigs (*Cavia porcellus*) were less aggressive and less bold than their relatives, the wild cavies (*Cavia aperea*). This domestication effects in guinea pigs was also found in a cognitive trait as domesticated guinea pigs learned an association task faster than wild cavies (Brust and Guenther 2015). In wild cavies, there were significant positive relationships between boldness, aggression and association learning, but these correlations did not appear in the domesticated guinea pigs (Brust and Guenther 2015).

Despite the increasing evidence for the *adaptive hypothesis* shaping behavioural syndromes (indicated by the differences in correlations between behaviours observed in different selective environments), there are still contradictory results which may also show that behavioural syndromes are difficult to break apart because of proximate mechanisms underlying the different behaviours (*constraint hypothesis*, Bell 2005). Whether behavioural correlations are a general characteristic of a species, and ubiquitous across different populations of this species, or whether selection might favour the coupling between the behaviours still seems difficult to predict even within taxa. Especially, studies comparing laboratory and field populations of the same species, using the same experimental set-ups are scarce. Such comparisons would provide more insights into whether or not laboratory populations, and behavioural syndromes observed in those, represent what is actually found in natural populations in the field.

Study Objectives and Hypotheses

We studied constant between-individual differences in behaviours of Eurasian harvest mice (*Micromys minutus*; Pallas, 1771) in a laboratory population (founded by zoo animals and kept in captivity for more than eight generations) and in a population in a semi-natural outdoor enclosure (founded from wild-caught animals). We previously analysed personality traits in the laboratory population and documented repeatable and consistent behaviour in juveniles and adults, as well as a behavioural syndrome in adult laboratory harvest mice (Chapter 1 and 2, respectively). Now, we investigated the same personality traits in wild-caught individuals and their offspring born in the semi-natural outdoor enclosure (in the following referred to as field population). This allowed us two compare patterns of constant between-individual differences in behaviours between standardized laboratory conditions and a semi-natural environment within two genetically different populations of the same species. We first measured the repeatability of three personality traits and one cognitive trait in the field population. We then investigated if the tested traits (boldness, activity, and exploration) correlated positively with each other forming a behavioural syndrome (Sih et al. 2004b). We expected that behaviours are repeatable and assumed the presence of a *fast-slow-behavioural syndrome* (Titulaer et al. 2012) as this was already found in the laboratory population of this species (Chapter 2). We also measured spatial recognition in a Y Maze. We tested the repeatability of this trait and its correlations with the personality traits. As spatial recognition was quantified from an active exploration behaviour in the Y Maze, we assumed a positive relationship between fast personality types and spatial recognition (Sih and Del Giudice 2012).

It is likely that different rearing environments (field enclosure versus laboratory conditions) affect individual behaviours and correlations between behavioural traits. We thus compared the mean behaviours and the observed correlations between laboratory and field population of harvest mice. We assumed a more natural behavioural expression induced by the semi-natural habitat in the enclosure. Because of this, and since mice of the field population occupy larger home ranges than laboratory mice, we hypothesized that the field population expresses more active and more explorative behaviour. However, we expected mice of the field population to be shyer than laboratory individuals, because we assumed that laboratory animals were more habituated to laboratory test situations and handling of observers. If the behavioural syndrome is maintained by natural selection on specific trait correlations, then it might get lost in populations with a long history of relaxed selection pressure (Archard and Braithwaite 2010). Under this scenario, we would expect stronger phenotypic relationships between personality traits in the field population, than in the laboratory population, as we assumed more relaxed selection in the laboratory than in the field. However, if the behavioural syndrome, observed in the laboratory population of harvest mice, is caused by genetic, morphological or physiological constraints, we would expect similar correlations between behaviours also in the field population of harvest mice.

Material and Methods – Behavioural Syndrome in the Field Population

Study Animals and Semi-natural Enclosure

Harvest mice tested in this study stemmed from our field population whose 29 funding individuals were trapped in August 2013 and May 2014 at the field station of the University of Potsdam in Potsdam-Bornim, Germany (N 52°26'22", E 13°0'44"). Harvest mice were captured under permission of the Landesumweltamt Brandenburg (reference number 24.01.01.18) and released into our semi-natural outdoor enclosure (***Figure 15***) in Tübingen, Germany (N 48°32'20", E 9°01'46").

Figure 15 The semi-natural outdoor enclosure of the harvest mouse population located near the Eberhard Karls University in Tübingen, Germany. Photo by Andrea C. Schuster

The outdoor enclosure was 33 m x 18 m large and surrounded by concrete walls (1.2 to 1.9 m in height), which had a metal rail on top and were embedded 0.5 m into the ground (see ***Figure 15***). Thus, no terrestrial predators were able to enter the enclosure, but avian predation was possible. The area was planted with high grass vegetation mainly consisting of bunch grasses. Species with highest abundances were *Sorghastrum nutans*, *Molinia arundinacea*, *Miscanthus sinensis*, *Panicum virgatum*, and *Spodiopogon sibiricus*. The study population was monitored by regular trapping events (see below) and was allowed to reproduce freely. During the winter months, we provided additional food by scattering sunflower seeds and oat flakes once per month within the enclosure. Additionally, we placed six wooden huts (54.0 x 77.0 x 66.5 cm) filled with hay and straw into the enclosure for shelter during winter time.

Life Trapping and Individual Identification

Between September 2013 and November 2015 we trapped mice at least once per month except for the winter periods (see ***TABLE 9***).

TABLE 9 Number of harvest mice captured the first time in the outdoor enclosure (unmarked individuals) in each month (roman numerals) of the three study years (2013, 2014, 2015). * indicates wild-caught animals from Potsdam, which were released to the enclosure. - are months without trapping events.

Year	**month**												**sum**
	I	**II**	**III**	**IV**	**V**	**VI**	**VII**	**VIII**	**IX**	**X**	**XI**	**XII**	
2013	-	-	-	-	-	-	-	24*	-	1	-	-	25
2014	-	-	9	6	0	5*	11	0	0	5	-	-	36
2015	-	-	0	3	5	5	0	13	8	0	0	-	34
all years	-	-	9	9	5	10	11	37	8	6	0	-	95

We used metal life traps (Ugglan special No2, Grahnab, Sweden). Traps were baited with oat flakes, peanut flips and apples to provide sufficient food and water resources. Traps were checked at least twice a day, around 8 am in the morning and between 6 and 8 pm in the evening. In the summer months, we check traps every three hours as young offspring cannot survive without maternal care and harvest mice reproduced mostly between May and September. Pregnant and lactating females were released directly at the trapping location. All other animals were brought to the laboratory for behavioural phenotyping. In 2013 and 2014 mice were individually tagged using *passive integrated transponders* (PIT). Individuals were narcotised by inhalation of isoflurane and we subcutaneously implanted a 9.0 mm x 1.4 mm transponder (ISO FDX-B, Planet ID, Essen, Germany) dorsal between the shoulders. In 2015, we marked animals by individual combinations of fur cuts and ear marks. Individual marking was always done after the first two phenotyping trials (in 2013 after the first trial) and animals spent at least one day in the laboratory after the PIT implantation before we released them again into the enclosure. PIT tag injection and ear marking were done under permission of the Regierungspräsidium Tübingen (reference numbers: ZO 2/11 and Anzeige vom 21.7.2014).

TABLE 10 summarises the number of behavioural observations collected between 2013 and 2015 in the field population of harvest mice.

TABLE 10 Number of the behavioural observations on the field population. Mice were tested one to four times in each of the five tests.

behavioural test	tested once	tested twice	tested three times	tested four times	# total observations
Open Field	11	52	19	9	208
Novel Object	11	49	23	9	192
Novel Environment	1	25	10	1	85
Y Maze	13	47	22	9	209
Scare Test	16	7	13	0	69

Laboratory Population

Harvest mice have been housed in our laboratory in Tübingen since November 2010. The harvest mice tested in this study stemmed from our laboratory population whose funding individuals (N = 26) originated from four different zoo populations. We recently showed that individuals from this captive population behaved repeatably and consistently (Chapter 1) and that behaviours correlated positively in adult harvest mice (Chapter 2). As recommended by Dingemanse and Dochtermann (2013) more observations per individual increase the power to statistically detect multivariate between- and within-individual correlations. For this reason we decided to reanalyse the previously published data while increasing our sample size (number of observations per individual) by combining all observations of juvenile and adult mice. We also included three further individuals to the data set. All individuals were born in our laboratory between October 2010 and September 2014, except for the three here added animals which were founding individuals that arrived in Tübingen in July 2011. In total, we tested 61 male and 53 female harvest mice of the laboratory population up to four times for each behavioural test (***TABLE 11***).

TABLE 11 Number of the behavioural observations of the laboratory population. Mice were tested one to four times in each of the five tests

behavioural test	tested once	tested twice	tested three times	tested four times	# total observations
Open Field	8	38	33	33	315
Novel Object	2	9	33	33	251
Novel Environment	1	59	0	0	119
Y Maze	7	37	33	33	312
Scare Test	18	41	0	0	100

Housing Conditions and Behavioural Tests

Mice of the field population were housed in polycarbonate cages of 60 cm length, 30 cm width and 35 cm height. Individuals were kept separately with water and food (hay, grain seeds and fresh fruits) *ad libitum*. The housing room had a natural light cycle and the room temperature varied between 16°C and 26°C, depending on the air temperature outside. Housing conditions of the laboratory animals were similar to the housing conditions of field population. A detailed description of the laboratory housing conditions and behavioural tests on laboratory mice can also be found in Chapters 1 and 2. Behavioural tests done on the field population are described below. Mice were tested up to four times, sample sizes for each behavioural test are shown in ***TABLE 10***. We used standard behavioural tests, originally established to test emotionality in laboratory strains of mice and rats. Tests were modified and adapted to our study species.

Open Field (OF)

We used a modified OF test (Archer 1973) to analyse activity and boldness as described before (Chapter 1). We videotaped field individuals from above using a digital camcorder (Handycam HDR-CX730E, SONY). We used the tool FFmpeg (www.ffmpeg.org) to convert and normalise all our recorded videos. After normalisation all videos had the same scale and video format, and were then used for automated video tracking (EthoVisionXT, Version 5, Noldus). We measured the total distance moved (in cm) during 4.5 min in a round arena and the time (in seconds) individuals spent at the inner part of the arena (unsafe area) as parameters for activity and boldness, respectively.

Novel Object (NO)

To analyse exploratory behaviour we used the NO test (Chitty and Shorten 1946). The NO was conducted directly after the OF test to reduce further stress as animals were already habituated to the arena setting. We recorded exploratory behaviour manually for five minutes after the first contact with the novel object as the time (in seconds) animals spent in physical contact with the novel object. For further details see Chapter 1.

Novel Environment (NE)

We used the Novel Environment test to further phenotype exploratory behaviour in harvest mice. Originally, the NE test was used by Verbeek et al. (1994) to analyse exploration in great tits (*Parus major*). We here simulated a novel environment by using a polycarbonate cage. An intermediate floor made of opaque polycarbonate was inserted. The bottom and intermediate floor were connected by five tubes. For a detailed description of the test see Chapter 2. Mice were observed directly. The latency (in seconds) until any four of the five tubes had been visited by the mouse was used as a parameter for exploration. We scored exploration in the NE only for field animals that were tested in 2015.

Scare Test (ST)

We used the Scare Test (Da Luz Correia 2013) to analyse the reaction towards disturbances in a familiar environment (see Chapter 2 for more details). The test took place in the individual home cages. During an active phase of the mouse (e.g., the animal was moving around in its cage), the metal lid of the cage was lifted and directly closed again. Animals immediately fled to hide under the hay or in the paper tube located at the back end of the cage. We measured the latency (in seconds) until mice returned from their hiding places as a parameter for boldness. If a mouse did not return from the hiding place within 15 min, we aborted the trial and scored the animal with 900 seconds (N = 9 observations).

Y Maze (YM)

We quantified spatial recognition in an adapted YM arena (Montgomery 1955), which relies on the assumption that rodents explore novel environments more than already known environments (Hughes 1968). The YM test consisted of two trials. During the first trial, one of the arms was locked (unknown arm) and in the second trial all three arms were accessible. For a more detailed description of the test see Chapter 1. We videotaped test individuals from above using a digital camcorder (Handycam HDR-CX730E, SONY). We used the tool FFmpeg (www.ffmpeg.org) to convert and normalise all our recorded videos. After normalisation all videos had the same scale and video format and were then used for automated video tracking (EthoVisionXT, Version 5, Noldus). We measured the total distance moved (in cm) during the first five minutes of the second trial as a parameter for activity. The time (in seconds) spent in the unknown arm during the first five minutes of the second trial was recorded as a parameter for spatial recognition. If an animal did not move during the second trial, we excluded individuals from further analyses as these observations confer no information about whether the animal recognised the new arm. Thus, we excluded eight observations from further analyses as the mouse did not leave the start arm of the YM.

Data Analyses

We performed all statistical analyses using *R* software, version 3.0.1 (R Core Team 2013) and ASReml, version 4.1 (Gilmour et al. 2014). As described in Chapter 1 and 2 we measured the effects of potential confounding factors on behaviours due to the experimental set-up, and due to individual characteristics of the mice, by fitting linear mixed models using the "nlme" package (Pinheiro and Bates 1996) in *R*. We applied square root transformations to the dependent variables (activity in OF and YM, exploration in NO, boldness in OF and spatial recognition) or log (boldness in ST, exploration in NE) to ensure a normal distribution of the model residuals. Exploration in NE and boldness in ST were multiplied by -1 for easier interpretation of the effect sizes and the correlation coefficients such that larger values correspond to a faster exploration and bolder individuals. For the dataset of the field population, we included the following confounding factors into each full model: the mouse identity (ID) as random factor (to account for repeated measures); sex, trial number, year (cohort: caught first time in 2013, 2014 or 2015), observer, and birth place (wild-caught in Potsdam or enclosure-born in Tübingen) as fixed factors; body mass, test date, and test time as covariates. For the dataset of the laboratory mice, we included the mouse identity (ID) as random factor; sex, trial number, year (tested in 2012, 2013 or 2014), whether the mouse was housed alone or in a pair (housing condition), and the observer as fixed factors; body mass, the lab generation the mouse belonged to as quadratic function (our lab population consisted of seven generations), test date, and test time as covariates. We applied backward stepwise reduction of the full model by successively excluding non-significant effects ($p < 0.05$). When confounding factors had more than two levels (e.g., trial number), we applied post-hoc tests (Tukey-test) and give estimates and test statistic for each contrast. We present final models with the remaining confounding factors in the ***Supplementary Table 6*** and ***Supplementary Table 7***. Confounding factors with significant effects were retained in the models for subsequent repeatability and correlation analyses.

Repeatability

Narrow sense repeatability of personality and cognitive traits was analysed using the package "rptR" (Nakagawa and Schielzeth 2010) in *R*. We estimated *adjusted repeatability* (R_A) (Nakagawa and Schielzeth 2010) by including confounding factors that we identified in the LMMs from the former analyses (see above). We estimated LMM based repeatability, including individual ID as random factor, based on 1000 bootstrapping runs and 1000 permutations. We display R_A values of untransformed variables with standard errors and asymptotic 95% confidence intervals (CIs), and permutation based p values.

Correlations between Behaviours

We fitted multivariate LMMs using restricted maximum likelihood (REML) in ASReml4 (Gilmour et al. 2014) to estimate phenotypic variances for each personality and cognitive trait, as well as the phenotypic covariances between these traits. We partitioned the within-individual variances and covariances from the between-individual variances and covariances as recommended by Dingemanse and Dochtermann (2013). We then used these variances and covariances to calculate phenotypic correlations between the traits within individuals (r_W) and between individuals (r_B). We included all previously defined significant confounding factors in the fixed model. The animal ID was included as random factor to all models. As spatial recognition was not repeatable in harvest mice of the field population (see results), we did not include this variable here.

We fitted the first multivariate model for the six personality traits to test for a behavioural syndrome in the field population (N = 95 individuals). We used a stepwise approach to build up the final model. First, we applied a diagonal variance model, which estimated only variances, but no covariances. Then, we applied an unstructured variance model where all between-individual and within-individual covariances were first fixed to zero. We then estimated the between-individual and within-individual covariances one by one, using estimates from respective bivariate models as starting values. Since we did not test mice in the NE in 2013 and 2014 and no mice were phenotyped in the ST in 2015, we could not model any covariance among these two traits for the field population. The between-individual and within-individual covariance among these traits was set to zero in the final model. We present between-individual correlations based on the final six-trait model. Within-individual correlation coefficients can be found in the appendix (***SUPPLEMENTARY TABLE 8***).

We fitted the second multivariate model for the six personality traits to estimate the correlations in the behavioural syndrome in the laboratory population (N = 112 individuals). We used the same stepwise approach to build up the final model as for the field data. We here present correlations based on the final six-trait model with fully unstructured between-individual variance-covariance matrix. The within-individual covariance matrix was modelled between activity in the OF test, activity in the YM, boldness in the OF test, and exploration in the NO test. For boldness in the ST and exploration in the NE, we were not able to model within-individual covariances, as those behaviours were measured at different time points. We present between-individual correlations based on the final six-trait model with described between-individual variance-covariance matrix. Within-individual correlations coefficients can be found in the appendix (***SUPPLEMENTARY TABLE 9***).

Comparison between the Laboratory and the Field Population

We compared mean levels of behaviours between the laboratory and the field population using the effect sizes of the fixed factor origin (laboratory and field) from linear mixed models. We fitted these models by combining the data sets of the laboratory and the field population and included the mouse identity (ID) as random factor and confounding factors with significant effects from previous models (see above). Further, we tested for homogeneity of variances between the two study populations using Levene's tests.

We calculated Z-tests (unpaired) to test for differences of correlation coefficients between laboratory and field population. We used Bonferroni correction to account for multiple comparisons. Our significance threshold was then $p \leq 0.01$, as each observed behavioural variable contributed to five correlation coefficients.

Results - Behavioural Syndrome in the Field Population of Harvest Mice

Repeatability of Behaviours in the Field Population

All tested personality traits were significantly repeatable in the field population. R_A values ranged between 0.038 and 0.517 (see ***TABLE 12***), where the two activity traits measured in the Open Field and in the Y Maze had the highest repeatability values. Exploration in the Novel Object test showed a modest but significant repeatability (***TABLE 12***). The cognitive trait spatial recognition was not repeatable in this population.

TABLE 12 Repeatability (R_A) with 95% confidence interval (CI) for personality traits and spatial recognition tested in the field population of harvest mice. Sample sizes (N) are given as observations/individuals. Significant repeatability values are marked bold.

trait	behavioural test	confounding effects	N	R_A	95% CI	p value
activity	Open Field	trial number	208/95	**0.517±0.069**	[0.370, 0.634]	< 0.001
	Y Maze	test time	209/94	**0.332±0.083**	[0.170, 0.498]	< 0.001
boldness	Scare Test	-	69/39	**0.214±0.148**	[0.000, 0.540]	< 0.001
	Open Field	trial number	208/95	**0.246±0.090**	[0.055, 0.414]	< 0.001
exploration	Novel Object	trial number + year	192/83	**0.038±0.062**	[0.000, 0.200]	< 0.001
	Novel Environment	trial number	85/37	**0.246±0.128**	[0.000, 0.508]	< 0.001
spatial recognition	Y Maze	-	209/94	0.00	NA	NA

Correlations between Behaviours in the Field Population

We observed three significant positive between-individual correlations among personality traits (r_B values between 0.421 and 0.807, ***FIGURE 16***). The two activity variables, as well as the two boldness variables, correlated significantly. However, the between-individual correlation between exploration in the Novel Environment test and in the Novel Object test was not significant. The third significant relationship confirmed our expectation of a behavioural syndrome: more active harvest mice were bolder in the Open Field test, than less active mice. The relationship between activity in the Y Maze and boldness in the Scare Test, as well as boldness in the Open Field was also strong and positive, but not significant ($p = 0.082$ and 0.069, respectively). All other between-individual correlations were not significant, but mostly positive. Unexpectedly, we observed two negative between-individual correlation coefficients. Both of them had large standard errors and were not significant (***FIGURE 16***).

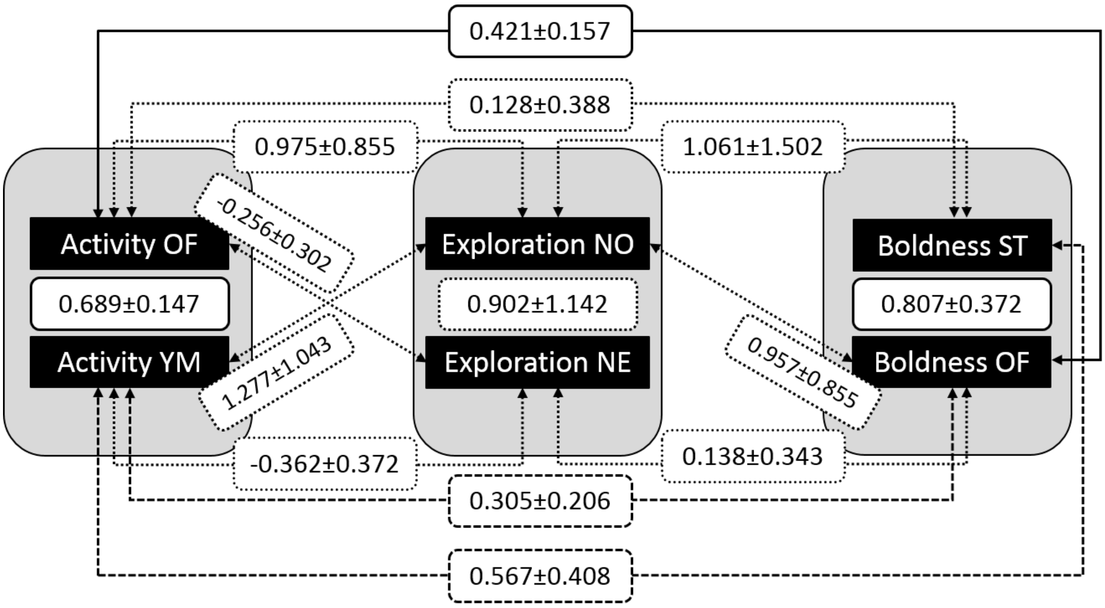

FIGURE 16 Behavioural syndrome in the field population of Eurasian harvest mice: Between-individual correlations (± standard errors) among personality traits calculated from a multivariate mixed model. Solid lines indicate significant correlations ($p < 0.05$), dashed lines indicate tendencies ($p < 0.1$), and dotted lines indicate non-significant correlations. OF: Open Field, YM: Y Maze, NO: Novel Object, NE: Novel Environment, ST: Scare Test. The covariance between exploration NE and boldness ST was not modelled due to missing data.

We observed two significant within-individual correlations out of the 14 modelled relationships. Activity in the Open Field and exploration in the Novel Environment were positively correlated (r_W = 0.222 ± 0.131, $p = 0.045$). The relationship between boldness in the Scare Test and exploration in the Novel Object test showed a significant negative within-individual correlation coefficient (r_W = -0.637 ± 0.127, $p < 0.001$). Results of the non-significant within-individual correlations can be found in the Appendix.

Correlations between Behaviours in the Laboratory Population

We observed eight significant positive between-individual correlations among personality traits in the laboratory population (r_B values between 0.408 and 0.797, ***FIGURE 17***). The two activity variables, as well as the two exploration variables, correlated significantly. However, the between-individual correlation coefficient between boldness in the Scare Test and in the Open Field test was small and not significant. The significant relationships between activity in the Open Field and exploration in the Novel Environment, as well as between both activity measures and boldness in the Scare Test, confirmed our expectation of a behavioural syndrome. Further, we observed strong positive between-individual correlations among both exploration and both boldness measures (see ***FIGURE 17***). All other between-individual correlations were not significant, but mostly positive. Unexpectedly, we observed two negative between-individual correlation coefficients. Both of them had large standard errors and were not significant (***Figure 17***).

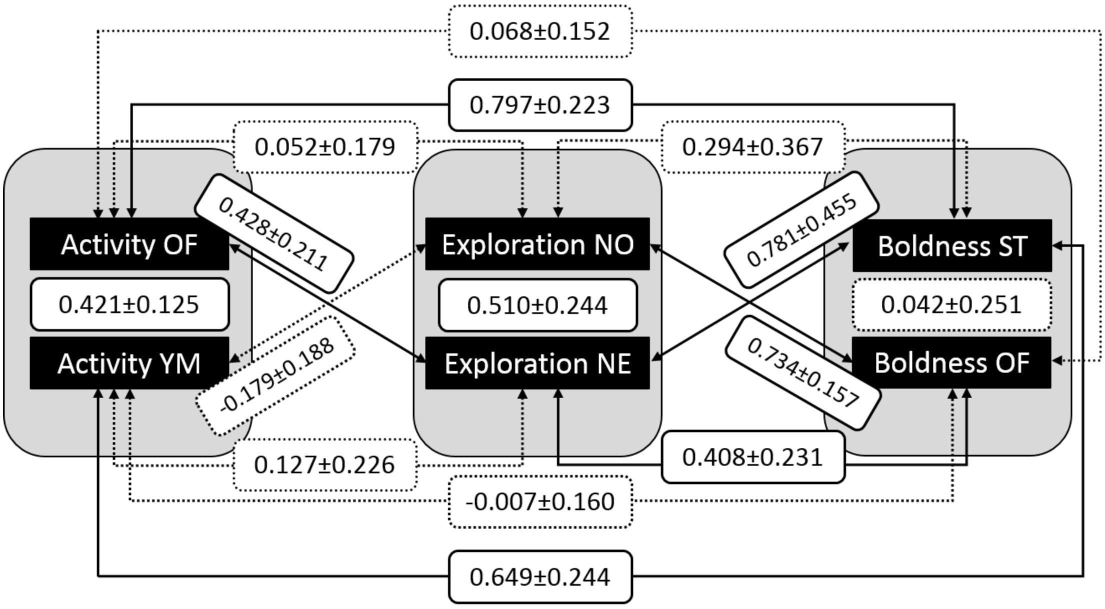

Figure 17 Behavioural syndrome in the laboratory population of Eurasian harvest mice: Between-individual correlations (± standard errors) among personality traits calculated from a multivariate mixed model. Solid lines indicate significant correlations ($p < 0.05$), and dotted lines indicate non-significant correlations. OF: Open Field, YM: Y Maze, NO: Novel Object, NE: Novel Environment, ST: Scare Test.

We observed two significant within-individual correlations out of the six modelled relationships. Both activity measures were positively correlated ($r_W = 0.239 \pm 0.067$, $p < 0.001$). The relationship between activity in the Open Field and exploration in the Novel Object test showed also a significant positive within-individual correlation coefficient ($r_W = 0.187 \pm 0.075$, $p = 0.006$). Two more within-individual correlations tended to be significant: activity in the Y Maze and boldness in the Open Field, as well as activity in the Y Maze and exploration in the Novel Object test ($p = 0.090$ and 0.089, respectively). Results of the non-significant within-individual correlations can be found in the Appendix.

Comparison between Laboratory and Field Population

We did observe some mean differences in behaviours between the laboratory and the field population: Activity and spatial recognition both tested in the Y Maze differed between populations, as well as exploration in the Novel Environment (***Table 13***). Harvest mice of the field population were more active in the Y Maze arena and spent more time in the unknown arm of the Y Maze compared to laboratory animals. Mice of the laboratory population, however, were more explorative in the Novel Environment than mice of the field population.

The Levene's test for homogeneity of variance across the two study populations revealed that there was less variance in spatial recognition in the Y Maze and exploration in the Novel Environment in laboratory animals than in field animals (***Table 13***). However, activity in the Open Field and exploration in the Novel Object showed more variance in the mice of the laboratory than of the field population (***Table 13***).

TABLE 13 Final LMMs with significant confounding factors and the effects of origin (field versus laboratory population) on personality and cognitive traits in harvest mice Significant differences between factor levels and contrasts which tended do differ are shown by the results of Tukey post-hoc tests (estimates ± standard error, z values and p values). Sample sizes (N) are given as observations/individuals. SE = standard error. Test statistic of Levene`s tests are displayed in the last columns.

										Levene's Test	
personality/cognitive trait	**behavioural test**	**N**	**confounding factors**	**F value**	**p value**	**contrast**	**estimate ± SE**	**z value**	**p value**	**F value**	**p value**
activity	Open Field	523/207	trial number	8.019	< 0.001	2 - 1	-0.33 ± 0.07	-4.606	< 0.001		
						3 - 2	-0.30 ± 0.09	-0.333	0.006		
			origin	0.186	0.667		0.05 ± 0.12	0.431	0.667	**39.821**	< 0.001
	Y Maze	521/206	trial number	2.659	0.048	2 - 1	-0.23 ± 0.08	-2.777	0.027		
			test time	12.140	< 0.001		-0.14 ± 0.04	-3.484	< 0.001		
			origin	4.912	0.028		**-0.29 ± 0.11**	-2.581	0.011	3.284	0.071
boldness	Scare Test	169/98	origin	0.000	0.995		0.00 ± 0.18	0.006	0.995	1.652	0.201
	Open Field	523/207	trial number	33.633	< 0.001	2 - 1	0.67 ± 0.08	8.677	< 0.001		
						3 - 1	0.41 ± 0.10	4.119	< 0.001		
						4 - 1	1.04 ± 0.14	7.523	< 0.001		
						3 - 2	-0.26 ± 0.10	-2.613	0.042		
						4 - 2	0.37 ± 0.14	2.687	0.034		
						4 - 3	0.63 ± 0.14	4.369	< 0.001		
			year	6.665	< 0.001	2013 - 2012	-0.63 ± 0.19	-3.326	0.005		
						2014 - 2012	-0.77 ± 0.18	-4.221	< 0.001		
						2015 - 2012	-0.87 ± 0.24	-3.606	0.002		
			origin	0.321	0.571		-0.08 ± 0.13	-0.567	0.571	1.619	0.204
exploration	Novel Object	523/207	trial number	7.979	< 0.001	2 - 1	0.50 ± 0.10	5.011	< 0.001		
						3 - 1	0.37 ± 0.12	3.219	0.007		
			year	5.778	0.004	2014 - 2013	0.41 ± 0.14	2.959	0.008		
						2015 - 2013	0.60 ± 0.20	3.031	0.007		
			origin	0.530	0.468		0.10 ± 0.13	0.728	0.468	**4.885**	0.028
	Novel Environment	204/97	trial number	10.946	< 0.001	2 - 1	0.57 ± 0.11	5.161	< 0.001		
						4 - 1	2.14 ± 0.83	2.563	0.040		
						4 - 3	1.95 ± 0.86	2.280	0.082		
			origin	26.389	< 0.001		**0.75 ± 0.15**	5.137	< 0.001	**24.356**	< 0.001
spatial recognition	Y Maze	521/206	trial number	2.798	0.040	2 - 1	-0.25 ± 0.10	-2.573	0.047		
			origin	34.383	< 0.001		**-0.51 ± 0.09**	-5.864	< 0.001	**18.479**	< 0.001

We observed significant between-individual and within-individual correlations in both populations (see above). But while in the laboratory population eight between-individual correlations were significant, only two appeared to be significant in the field population. Only the two activity measures correlated significantly in both populations, the other between-individual correlations did not correspond in their significance between both populations (see ***FIGURE 16*** and ***FIGURE 17***). Further, we observed two significant within-individual correlations in each of the populations. However, these significant within-individual correlations differed between laboratory and field population (***SUPPLEMENTARY TABLE 8*** and ***SUPPLEMENTARY TABLE 9***).

Z-tests showed that 85.7 % (12 out of 14) of the between-individual correlation coefficients differed significantly between the two study populations (***TABLE 14***). Nine of those were larger in the field population compared to the laboratory population. Between-individual correlation coefficients between activity in the Y Maze and boldness in the Scare Test, as well as between boldness in the Open Field and exploration in the Novel Environment did not differ between laboratory and field population. However, the p value was 0.021 for the latter comparison (no significance after Bonferroni correction). In all cases, where we observed differences between correlation coefficients, the correlation was not significantly different from zero in at least one of the populations, except for the correlation between activity in the Open Field and in the Y Maze.

TABLE 14 Comparison of between-individual correlations from the field and the laboratory population. The difference between correlation coefficients of the field and the laboratory population (effect size) are displayed below the diagonal. Z values are shown above the diagonal and are marked in bold when significant after Bonferroni correction. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	**activity YM**	**boldness OF**	**boldness ST**	**exploration NO**	**exploration NE**
activity OF		**4.41**	**4.24**	**6.03**	**22.09**	**4.99**
activity YM	**0.268**		**3.57**	0.82	**41.23**	**3.51**
boldness OF	**0.353**	**0.312**		**6.75**	**10.06**	2.04
boldness ST	**- 0.669**	- 0.082	**0.765**		**21.92**	NA
exploration NO	**0.923**	**1.456**	**0.223**	**0.767**		**6.38**
exploration NE	**- 0.684**	**- 0.489**	- 0.270	NA	**0.392**	

Discussion - Behavioural Syndrome in the Field Population of Harvest Mice

Behavioural Syndrome in the Field Population of Harvest Mice

We could show for the first time that Eurasian harvest mice from a semi-natural field population behaved repeatably. Activity, boldness and exploration, quantified in two different behavioural tests each, were significantly repeatable in harvest mice of a natural genetic background. We observed large and positive between-individual correlation coefficients between behaviours. As three correlations between these behaviours were significant, we concluded that harvest mice of the field population expressed a fast-slow behavioural syndrome.

Repeatability of Behaviours in Eurasian Harvest Mice

All behaviours were significantly repeatable in harvest mice from the field population. The mean R_A value was 0.27 (range between 0.04 and 0.52) which is within the range of the general average for personality traits (Bell et al. 2009). As already shown in laboratory harvest mice (Chapter 1 and 2), the two activity variables had the highest repeatability estimates. In the meta-analysis by Bell et al. (2009), behavioural repeatability was higher in natural populations than in laboratory populations. We did not observe such a pattern in harvest mice. Repeatability estimates of same behaviours tested in the laboratory population ranged between 0.16 and 0.57 in juvenile harvest mice (Chapter 1) and between 0.22 and 0.60 in adults (Chapter 2). We acknowledge that this was not true for exploration in the Novel Object test, where we observed a repeatability of 0.33 in the laboratory population (Chapter 2), but less evidence for repeatable exploration in the field population (R_A = 0.04, this study). Harvest mice of the field population may react more differentiated and therefore less repeatably towards the different novel objects we used during the distinct trials. Nevertheless, we conclude that Eurasian harvest mice with a wild population background and kept under semi-natural conditions show equal repeatability of behaviours, as laboratory animals. Activity, boldness and exploration thus seem to fulfil an essential assumption of a behavioural syndrome in Eurasian harvest mice (in wild and captive populations).

Spatial orientation is assumed to be a very important trait for harvest mice as they live in a complex three-dimensional habitat. We tested spatial recognition in a Y Maze. Before, we identified significant repeatability of spatial recognition in adult harvest mice of the laboratory population ($R_A = 0.20 \pm 0.10$, Chapter 2), but not in juveniles (Chapter 1). Harvest mice of the field population did also not perform repeatably in the Y Maze. Laboratory environments are more stable than natural conditions (Archard and Braithwaite 2010). Thus, constant spatial recognition might be favoured in captive populations but not in wild populations. This might be the case because natural environments are too unstable, so that flexible spatial orientation skills are probably more adaptive for wild harvest mice than in laboratory populations. This is because in such environments orientation based on changing visual cues is not advantageous. Harvest mice may instead use olfactory cues for orientation as they might be more reliable. Another explanation for our finding could be the age of the animals from the field population. We could not identify the exact age of the individuals in the field population, but as we tested unmarked animals when they were caught the first time in the enclosure, the average age of the mice tested may be younger compared to the average individual from the laboratory population. We previously found that among laboratory animals, young individuals (age = 35 - 53 days) did not show repeatable spatial recognition, while adult mice (age = 81 - 181 days) did behave repeatably in the Y Maze (Chapter 1). If we tested mainly young mice in the field, this may explain the lack of repeatability in this behaviour.

Behavioural Differences between the Laboratory and the Field Population

Our hypotheses that individuals from the field are more active, more explorative, but shyer than the laboratory individuals were partly confirmed. Harvest mice of the field population were more active in the Y Maze arena compared to laboratory animals. But this difference was not found in the Open Field. In contrast to our expectations, animal of the laboratory population were more explorative in the Novel Environment than mice of the field population. However, this was not the case in the Novel Object test. We observed higher levels of spatial recognition in the field population than in the laboratory population of harvest mice. Field animals spent on average more time in the unknown arm of the Y Maze than laboratory individuals which supports the hypothesis that wild populations express better (but not constant) cognitive abilities than captive ones (Brust and Guenther 2015). All other behaviours did not differ between laboratory and field animals. Compared to the standard laboratory housing of rodents, we provide the harvest mice with a more natural habitat through environmental enrichment, to facilitate physical activity, explorative behaviour, and natural climbing behaviour. The same applied to the captive populations where we received the founder individuals from. This may have contributed to the maintenance of a natural behavioural expression even after many generations in captivity.

However, we did observe differences in the variance of behavioural expression between laboratory animals and the field population. Interestingly, harvest mice from the laboratory showed less variance in spatial recognition in the Y Maze and exploration in the Novel Environment, than mice from the field population. Those were the two behavioural test, which mostly involved spatial orientation skills. Thus, as mentioned before, flexible and more variable spatial orientation skills may be more adaptive for field harvest mice than for laboratory animals. In contrast, activity in the Open Field and exploration in the Novel Object test showed more variance in the laboratory than in the field population.

Correlations between Behaviours in Eurasian Harvest Mice

We identified strong positive relationships between behaviours in the field population, but only three between-individual correlations were statistically significant. Standard errors were relatively large for most correlations, which may be due to the limited sample size. In a previous study we also observed strong positive correlations between behaviours in adult harvest mice of the laboratory population (Chapter 2), and these results were confirmed by the analyses of the enlarged laboratory data set in this study. We had more observations from the laboratory than from the field (100 - 315 per trait, and 69 - 209 per trait, respectively). The larger sample size might be the reason for the fact that we observed more significant between-individual correlations in the laboratory population. Small sample sizes can lead to large standard errors and may result in unrepresentative conclusions. They may also cause some of the commonly observed differences between laboratory and wild population of the same species. In a meta-analysis, the mean effect size of phenotypic correlations between personality traits was 0.198 which was statistically different from zero (Garamszegi et al. 2012). Applying a power analysis, Garamszegi et al. (2012) estimated that at least 198 individuals were required to statistically confirm phenotypic relationships of such an effect size (Garamszegi et al. 2012). However, the mean sample size in the studies used in their meta-analysis was only 45 individuals (Garamszegi et al. 2012), and only one of the studies had access to the supposedly required sample sizes: The work by Cote et al. (2010) on mosquitofish (*Gambusia affinis*). Thus, erroneous conclusions about the lack of phenotypic correlations may be widespread. Still, in some cases, strong significant correlations between behaviours were observed despite low sample sizes (e.g., N = 21 in wild cavies, Guenther et al. 2014a; Brust and Guenther 2015). In our analysis of the field population, we calculated all correlations with data from less than 200 individuals, and we assume that, with larger sample sizes, some of the observed large correlation coefficients would indeed prove to be significantly different from zero.

As we used the same experimental set-ups for the field and the laboratory population, the results can be directly compared. We observed that the correlation coefficients differed in 85.7 % between the laboratory and the field population. However, these comparisons might be influenced by the different sample sizes. Nine correlation coefficients were larger in the field population compared to the laboratory population. Three correlation coefficients were larger in the laboratory population compared to the field population. This was in line with our expectations, as we assumed stronger relationships between behaviours due to stronger selection pressure in the field population. We observed differences in the sign of the between-individual correlations, but none of the three negative correlation coefficients was significantly smaller than zero. As all other between-individual correlations indicated the same direction of relationship between behaviours in laboratory and in field harvest mice, we conclude that Eurasian harvest mice in general exhibit a fast-slow behavioural syndrome (Titulaer et al. 2012).

Recently, Fischer et al. (2016) showed that behavioural syndromes can be plastic. Correlations between behaviours differed among populations of Trinidadian guppies (*Poecilia reticulate*) depending on their genetic background and rearing environment (Fischer et al. 2016). In spiders differences in the emergence of phenotypic correlations were observed: For example, between restricted compared to environmental enriched rearing conditions (Bengston et al. 2014), or between field-rear compared to laboratory reared (Sweeney et al. 2013). In contrast, the different environmental conditions in our two study populations did not seem to influence the occurrence of the behavioural syndrome in harvest mice. The housing conditions of our laboratory population might reflect near natural conditions and thus helped to maintain a natural behavioural expression. However, selection is relaxed in the laboratory, thus our finding of strong correlations in the laboratory population is also a sign for the fact that the observed behavioural syndrome may not be actively maintained by natural selection in harvest mice. The syndrome might rather be fixed through proximate mechanisms (e.g., genetic correlations or physiological trade-offs). There is also evidence from other species that behavioural correlations can be stable over time and under different selective environments: In rock pool prawns (*Palaemon elegans*), for example, it was shown that the behavioural syndrome structure was consistent over a period of 2 weeks (Chapman et al. 2013). This was also observed by Gyuris et al. (2012) for a longer time span, as the correlation structure among personality traits in firebugs (*Pyrrhocoris apterus*) remained constant across ontogeny. In mustard leaf beetles (*Phaedon cochleariae*), it was shown that the clustering of behaviours was stable throughout the entire lifetime (Müller and Müller 2015). In damselflies (*Lestes congener*), it was found that “larval behaviour in one environment may carry over metamorphosis into adult behaviour in a completely different environment” (Brodin 2009, page 35). Hermit crabs (*Pagurus bernhardus*) exhibited a consistent behavioural syndrome structure between two different ecological conditions (low versus high predation risk situations, Mowles et al. 2012). Stable behavioural correlations were also found in fish species (e.g., wild brown trout, *Salmo trutta*, Adriaenssens and Johnsson 2013; and sheepshead swordtails, *Xiphophorus birchmanni*, Boulton et al. 2014). And in the fast-slow selections lines of wild great tits (*Parus major*) and laboratory house mice (*Mus musculus*), behavioural correlations were still apparent after many generations (Koolhaas et al. 1999; Carere et al. 2005).

Given the inconsistent results among studies of behavioural syndromes, with stable behavioural correlations on the one hand, but flexible behavioural syndrome structures on the other hand (see above), it seems still had to predict why and when traits are expected to correlate and what mechanisms are maintaining the behavioural syndromes (Favati et al. 2016). In our study species, a fixed behavioural syndrome, constrained by a proximate basis, may lead to maladaptive behaviour in some environments. In competitive environments (e.g., high population density) more active and explorative individuals may have advantages if they are also bold. However, this behavioural type may be less advantageous under high predation risks, because bolder individuals may face higher predation than shy individuals. In such a situation, more active mice should also be shy to express more caution towards their predators. However, a flexible behavioural syndrome (i.e. different relationships between behaviours in different environments) may be prevented by the constraints maintaining it, even if flexible relationships would be advantageous. This might be because evolution cannot attain the maintenance: For instance, a more flexible behavioural syndrome might require a change in the proximate mechanism underlying the behavioural correlations. However, that change is not likely to emerge, because the evolution of a more flexible behavioural syndrome might be "prevented by crossing of an adaptive valley of the fitness landscape" (Carere and Maestripieri 2013, page 263). To further understand the architecture of behavioural correlations in harvest mice, more investigations are needed to reveal whether the observed behavioural syndrome is adaptive or not in different selective environments, and what kind of proximate mechanisms maintain those relationships between behaviours.

Conclusion - Animal Personality in Eurasian Harvest Mice

Animal Personality in Eurasian Harvest Mice

In sum, we conclude that Eurasian harvest mice showed constant between-individual differences in behaviours, i.e. animal personality. We identified behavioural correlations in the laboratory and in a field population of harvest mice along a fast-slow continuum. As different selective environments (laboratory versus field) did not result in different behavioural syndromes in the two study populations, our data suggest that the fast-slow behavioural syndrome in Eurasian harvest mice seems to be evolutionarily stable and fixed by underlying proximate mechanisms (*constraint hypothesis*). We suggest further investigations of those mechanisms underlying the behavioural syndrome, using for example pedigree information to analyse whether similar genetic correlations form the behavioural syndromes found in the laboratory and in the field population of harvest mice.

Discussion

Our results provide strong evidence for the existence of constant between-individual differences in behavioural traits in Eurasian harvest mice (*Micromys minutus*).

As harvest mice have not been subjects in animal personality research yet, this was the first time that constant between-individual behavioural differences were identified in this species. Moreover, this thesis provides initial evidence for constant between-individual differences in spatial cognitive traits, and for a relationship between personality and spatial cognition in Eurasian harvest mice.

Behavioural Repeatability and Consistency in Eurasian Harvest Mice

Activity, boldness and exploration were repeatable in juvenile and adult harvest mice of the laboratory population (Chapter 1 and 2). Furthermore, those behaviours were expressed consistently independent of age, maturation and individual sexual experience (Chapter 1). Harvest mice of the field population behaved also repeatably (Chapter 3).

Further, we tested spatial recognition based on object cues in a Y Maze. This trait was repeatable in adult mice, but not in juveniles, and it showed little evidence for consistency across life history phases (Chapter 1). Spatial recognition was repeatable in adult harvest mice of the laboratory population (Chapter 1 and 3), but not in individuals of the field population (Chapter 3). Spatial learning ability and decision speed were additional spatial cognitive traits showing repeatability in adult harvest mice, but we found no repeatability of decision accuracy (Chapter 2).

Behavioural Repeatability in Eurasian Harvest Mice

Behavioural repeatability estimates in the laboratory population ranged between 0.16 and 0.57 in juvenile harvest mice (Chapter 1) and between 0.18 and 0.69 in adults (Chapter 1 and 2). In the field population the behavioural repeatability ranged between 0.04 and 0.52 (Chapter 3). Repeatability estimates (mean R_A value = 0.35) thus were within the range of usually observed repeatabilities of personality traits (Bell et al. 2009; Garamszegi et al. 2013).

In a meta-analysis by Bell et al. (2009), behavioural repeatability was higher in wild populations than in laboratory populations. Such a pattern was not observed in harvest mice (despite for exploration in the Novel Object test). As behavioural repeatabilities were similar in different selective environments (laboratory and field population), it is likely that proximate constraints rather than selection maintain personality traits in harvest mice (see below).

Although the meta-analysis by Bell et al. (2009) could not detect any differences in the repeatability of behaviours between juveniles and adults, a recent review by Brommer and Class (2015) reported evidence for lower behavioural repeatability in juveniles. We did not observe this differences between juveniles and adults. Eurasian harvest mice already expressed repeatable behaviours as juveniles (Chapter 1). Likewise, behavioural repeatability did not seem to change across life history stages, which further suggests that behavioural types in this species may be a stable phenomenon which is maintained by proximate mechanisms.

Repeatability of spatial cognitive traits ranged between 0.13 and 0.26. However, decision speed in the Spatial Orientation Task was not repeatable (Chapter 2). Another published study on repeatability of decision speed and accuracy was conducted on zebrafish (*Danio rerio*), where Wang et al. (2015) showed that both traits were significantly repeatable in a spatial colour discrimination task. However, there is currently only scarce evidence for repeatability of spatial recognition and general learning ability in animals (Griffin et al. 2015).

We found that spatial recognition in the Y Maze was repeatable in adult harvest mice of the laboratory population, but not in individuals of the field population (Chapter 3). This might be due to different selective processes in the two environments rather than constraints (in contrast to personality traits). However, more data is needed to investigate if constant between-individual differences in spatial cognitive traits of harvest mice are maintained by selection.

Behavioural Consistency in Eurasian Harvest Mice

Behavioural consistency estimates in the laboratory population ranged between 0.18 and 0.84 in harvest mice (Chapter 1). Activity, boldness and exploration were consistent over maturation, and when mice were tested before and after their first sexual experience. This shows that even major changes between life-history stages (e.g., maturation or first sexual contact) are not necessarily connected to inconsistent individual behaviours. During maturation and the first sexual contact, the relative magnitude of attained reproductive success and expected future reproductive success changes depending on individual experiences. The dynamics of this trade-off may result in a change of selection pressures on animal personality traits (Bell and Stamps 2004). However, these events did not seem to have an influence on the consistency of the studied personality traits in harvest mice.

We tested adult mice at the age of three and six month, which probably represents the entire mean life span of this species in the wild (Kubik 1952; Piechocki 2001). Hence, Eurasian harvest mice - at least under laboratory conditions - seem to behave consistently throughout their entire life span, and the short-term consistency of these behaviours was independent of maturational effects and sexual experience. Our results are in line with previous findings in animal personality research: Exploration was a consistent trait over a significant part of an individual's lifetime in great tits (*Parus major*, Dingemanse et al. 2002) and zebra finches (*Taeniopygia guttata*, David et al. 2012). In small rodents, the three commonly tested personality traits boldness, exploration and activity were repeatable over short periods of time (e.g., Koolhaas et al. 1999; Boon et al. 2007; Lantová et al. 2011; Petelle et al. 2013). However, only activity seemed to be consistent across life history stages in that group (e.g., Kanda et al. 2012; Herde and Eccard 2013, but see Guenther et al. 2014).

To date, no comprehensive theory has been developed to predict which factors would favour or disfavour consistent behaviour in specific species. This is in part due to the fact that consistent behaviours can arise from two sources: Natural selection for stable behavioural types, or physiological constraints that prevent flexible behaviour. More investigations (e.g., consistency analyses in different selective environments) are needed to reveal which factors maintain behavioural consistency in harvest mice.

Behavioural and Cognitive Syndrome in Eurasian Harvest Mice

We found that harvest mice of the laboratory population expressed a behavioural syndrome along a fast-slow continuum (Chapter 2 and 3), and those behavioural relationships were also found in the field population (Chapter 3).

Our results further indicated weak evidence for a cognitive syndrome as fast behavioural types learned the spatial orientation task faster than slow types, and shyer harvest mice made decisions quicker than bolder mice (Chapter 2).

Behavioural Syndrome in Eurasian Harvest Mice

Based on our results from the laboratory and the field population, we conclude that Eurasian harvest mice in general exhibit a behavioural syndrome along a fast-slow continuum (Titulaer et al. 2012). Significant correlation coefficients ranged between 0.41 and 0.88 (Chapter 2 and 3). Such a fast-slow syndrome is in line with a large body of personality literature in rodents: Starting with the observations on laboratory house mice (Koolhaas et al. 1999), further studies reported fast-slow-behavioural syndromes in recent years, for instance, in Belding's ground squirrels (*Urocitellus beldingi*, Dosmann et al. 2015), and in cavies (Cavia aperea, Guenther et al. 2014a).

However, studies comparing laboratory and field populations of the same species, and using the same experimental set-up are still scarce. But those comparisons can provide more insights into whether or not laboratory populations, and behavioural syndromes observed in those, represent what is actually found in natural populations in the field. In harvest mice, nine of twelve correlation coefficients were larger in the field population compared to the laboratory population. Only three correlation coefficients were larger in the laboratory population than in the field population. This corresponded to our expectations, as we assumed stronger relationships between behaviours due to stronger selection pressure in the field population.

As selection is relaxed in the laboratory, our finding of strong correlations in the laboratory population of harvest mice is a sign for the fact that the observed behavioural syndrome may not be actively maintained by natural selection in harvest mice. The behavioural syndrome in harvest mice might rather be evolutionarily stable and fixed by underlying proximate mechanisms (*constraint hypothesis*). However, whether behavioural syndromes are based on proximate constraints or maintained by selection still remains not predictable for different animal species. Studies found evidence for both mechanisms in different taxa (Yuen et al. 2016). We suggest further investigations of the mechanisms underlying the behavioural syndrome in harvest mice (e.g., cognitive or genetic constraints, see below). A fixed behavioural syndrome, constrained by a proximate basis, may lead to maladaptive behaviour of behavioural types in some environments. In competitive environments (e.g., high population density) more active and explorative harvest mice may have advantages if they are also bold. But those individuals may be less advantaged under high predation risk, because bolder mice may face higher predation than shy animals. In such a situation, more active individuals should also be shy to express more caution towards their predators. But regardless of the mechanism behind the behavioural syndrome in harvest mice, selection for behavioural types will only result in the maintenance of these types in the population if the correlations that underlie the behavioural syndrome are in fact genetic relationships. This has not been investigated in harvest mice yet, but will give more insights to the understanding of behavioural syndromes in this species (see below).

Cognitive Syndrome in Eurasian Harvest Mice

We found initial indications for a relationship between spatial learning ability and personality types in harvest mice. More active individuals reached the learning criterion in the Spatial

Orientation Task earlier than less active animals (r_B = 0.79 ± 0.45). Further, a non-significant tendency suggested a positive relationship between boldness in the Scare Test and spatial learning ability. Finally, the between-individual correlation coefficient between spatial learning ability and boldness in the Open Field test showed a large correlation coefficient, with large standard error. The speed-accuracy trade-off was not apparent within, nor between individuals. Nevertheless, we found weak evidence for a cognitive syndrome as shyer harvest mice made decisions faster than bolder mice (r_B = -0.41 ± 0.18). Thus, our data partly support the hypothesis that behavioural types correlate with individual cognitive ability (Sih and Del Giudice 2012), and our results are to some extent in line with findings in other animal species (e.g., in starlings, *Sturnus vulgaris*, Boogert et al. 2006; and in cavies, *Cavia aperea*, Guenther et al. 2014a).

Spatial cognition is assumed to be a possible key target of evolutionary cognition studies, as the hippocampus provides “a credible neurobiological target for selection” (Morand-Ferron et al. 2015, page 370). Our established spatial learning test was suitable to cover between-individual variation in spatial learning ability in harvest mice However, if fast behavioural types would in general outperform slow harvest mice in spatial cognitive tasks, we should wonder why both types still persist in populations. Although slow learning harvest mice may not have any fitness advantages, shy and less active harvest mice may indeed be the more adapted behavioural type during some parts of the year (e.g., in low population density, see above). Thus, the observed relationship may cause non-independent trait evolution and maintain slow learners in the population. However, we have to acknowledge that we only used one spatial learning task. To test if harvest mice really show constant cognitive styles in several situations requires more tasks. Fast cognitive spatial learning styles should solve various tasks equally well (cf. Sih and Del Giudice 2012). This was shown, for example, in Carib grackles (*Quiscalus lugubris*) and in black-capped chickadees (*Poecile atricapillus*). In these species, birds performed consistently well over two different tasks (Ducatez et al. 2015; Guillette et al. 2015). To investigate such relationships between different tasks, we recommend to establish further spatial learning tests for harvest mice (see below).

Evolution of Constant Between-Individual Differences

Evolution of Constant Between-Individual Differences in Eurasian Harvest Mice

Our findings showed for the first time that behaviours in Eurasian harvest mice met an essential assumption in animal personality research - repeatable and consistent between-individual differences (Réale et al. 2007). However, as this is the first study on constant between-individual behavioural differences in this species, more investigations are warranted to fully understand which factors maintain the repeatability and consistency of behaviours. It could be that in harvest mice, repeatably and consistently behaving animals are favoured by selection. Alternatively, or in addition, physiological constraints may maintain consistent behaviours across the short lifespan. Based on our data, we currently would rather assume that proximate constraints maintain the constant between-individual differences in harvest mice, because of the similar observed repeatability values in the laboratory and field population.

We identified behavioural correlations in the laboratory and the field population of harvest mice along a fast-slow continuum (see above). As different selective environments (laboratory versus field) did not result in different behavioural syndromes in the two study populations, our data suggest that the fast-slow behavioural syndrome in Eurasian harvest mice seems to be evolutionarily stable, fixed by underlying proximate mechanisms (*constraint hypothesis*). This further would suggest that the behavioural syndrome is not easily to decouple by natural selection in harvest mice. Fischer et al. (2016) showed that behavioural syndromes can be plastic, as correlations between behaviours differed among populations of Trinidadian guppies (*Poecilia reticulate*) depending on their genetic background and rearing environment. On the other hand, there were several stable behavioural syndrome structures observed (e.g., in firebugs, *Pyrrhocoris apterus*, Gyuris et al. 2012; or in mustard leaf beetles, *Phaedon cochleariae*, Müller and Müller 2015). Whether behavioural syndromes are based on proximate constraints or maintained by selection still remains not predictable for different animal species. We suggest further investigations of those mechanisms underlying the behavioural syndrome, using for example pedigree information to analyse whether similar genetic correlations form the behavioural syndromes found in the laboratory and in the field population of harvest mice.

Analyses of selection on personality or cognitive traits require large sample sizes. Dingemanse and Réale (2005) suggested that observations of at least 100 individuals are necessary to sufficient analyse selection and trustfully exclude Type 2 errors. We had such large sample sizes, however, personality and spatial cognitive traits now need to be correlated to fitness proxies to investigate whether selection favours specific behavioural types or cognitive styles in the semi-natural enclosure. As I took tissue samples of all individuals of the field population, parentage analysis could reveal the pedigree structure and the number of offspring each individual sired in the field population (see below, *Future Directions*, page 115). Harvest mice were regularly life-trapped in the enclosure, this data may be a good measure of individual life span, providing another fitness proxy. Lifetime reproductive success is assumed to be the best estimate for fitness in relation to cognitive abilities, as cost and benefits of cognitive traits may differ between life history phases (Mery and Kawecki 2003). Thus, our data may also reveal if spatial cognitive trait variation is correlated to fitness in harvest mice. Selection on cognitive traits was so far only estimated in a wild population of great tits (*Parus major*, Cole et al. 2012; Morand-Ferron et al. 2015). More investigation on fitness differences between behavioural types and cognitive styles would hence provide a deeper understanding of the evolutionary mechanisms that maintain behavioural and cognitive syndromes in animals.

For this kind of analysis, genetic data would also be necessary. Only if behavioural or (spatial) cognitive traits have a genetic component, we would expect a response to selection on that trait and hence, evolution of constant between-individual differences in those traits. Further, if selection acts on correlations between different constant individual behaviours and/or cognitive traits, and if these relationships are genetically based, we would expect a response to selection on trait correlations. Thus, understanding the evolution of correlated traits (i.e. behavioural types and cognitive styles) requires to estimate the underlying genetic correlations and to reveal the underlying genetic architecture (Karlsson Green et al. 2016). Significant genetic correlations between behaviours were, for instance, observed in great tits (*Parus major*, van Oers et al. 2004a; in three-spined sticklebacks, *Gasterosteus aculeatus*, Bell 2005; and in yellow-bellied marmots, *Marmota flaviventris*, Petelle et al. 2015). The potential presence of genetic correlations within a cognitive syndrome has not been investigated yet, but would deepen our understanding about the evolutionary mechanisms that maintain relationships between personality and cognitive traits.

Conclusion and Outlook

This thesis provides strong evidence for the existence of constant between-individual differences in behaviours and spatial cognitive traits, and some initial evidence for a relationship between personality and spatial cognitive traits in Eurasian harvest mice (*Micromys minutus*).

We were able to establish the following behavioural test settings for our study species: The Open Field test, the Novel Object test, the Scare Test, the Novel Environment test, the Y Maze, and the Spatial Orientation Task. These tests were used to measure activity, boldness, exploration, spatial recognition, spatial learning ability, decision speed, and decision accuracy. The tests fulfil the two prerequisites to investigate animal personality in harvest mice: First, they captured sufficient behavioural variance (within-individual variance and between-individual variance) in test performance, which was necessary for the estimation of repeatability and consistency. Importantly, this variance was also present during second, third and fourth trials, so that repeated observations are possible using these tests. Secondly, the tests were relatively fast so that we were able achieve large sample sizes (N_{total} = 1860 behavioural observations in 207 individuals) and therefore sufficient statistical power for our analyses. This was also possible for the spatial cognitive tasks.

Constant Between-Individual Differences in Eurasian Harvest Mice

Using those behavioural and spatial cognitive tests, we found that Eurasian harvest mice behaved constantly and that personality traits formed a behavioural syndrome in the laboratory and the field population. We also showed that there might be a relationship between personality and spatial cognitive traits in harvest mice forming a cognitive syndrome. Nevertheless, several open questions remain and further investigations will contribute to a deeper understanding in how animal personality and constant individual spatial cognitive traits have evolved and are maintained in harvest mice.

Future Directions

Future investigation should, for instance, address the following questions: (I) Are personality and spatial cognitive traits heritable in harvest mice? (II) Are the behavioural syndromes found in the laboratory and the field population only based on phenotypic relationships, or are they based on genetic correlations? (III) How stable are the behavioural and cognitive syndromes over ontogeny in harvest mice? (IV) Do males and females express the same behavioural and cognitive syndrome structures in harvest mice? (V) Do harvest mice exhibit spatial cognitive styles (in laboratory and field populations)? (VI) What mechanisms maintain constant between-individual differences of behaviours and spatial cognitive traits, as well as the relationships between them in harvest mice?

If behavioural or (spatial) cognitive traits have a heritable component, we could expect a response to selection on those traits. To investigate the heritability of behavioural and (spatial) cognitive traits in harvest mice would require pedigree information of the tested individuals. This data is actually already available: As the laboratory population of harvest mice is bred under controlled conditions, the whole pedigree structure of the laboratory population is known. First preliminary results may indicate evidence for genetic variance in some of the tested traits (data not shown). Further, I took tissue sample of all animals from the field population (data not shown). A parentage analysis will reveal the pedigree structure of the field population, and this can be used for the heritability analysis of personality and spatial cognitive traits in harvest mice.

Selection for behavioural syndromes will only result in the maintenance of these behavioural types in the population, if the between-individual correlations that underlie the behavioural syndromes are in fact genetic relationships. Thus, using the pedigree information will further reveal whether or not behavioural and cognitive syndromes are genetically based in harvest mice.

So far, we do not know which mechanisms actually maintain behavioural and cognitive syndromes in harvest mice. Further insides could be given by analyses about the stability of the syndromes over ontogeny. In other rodents, behavioural correlations appeared to be unstable over ontogeny (e.g., Kanda et al. 2012; Guenther et al. 2014b). To investigate and compare behavioural and cognitive syndromes in juveniles and adults, may reveal how developmental changes influence those syndromes in harvest mice. This would lead to more knowledge about the maintenance and evolution of behavioural and cognitive syndromes in this species.

Further, sexual selection may play a role in the maintenance of animal personality and behavioural syndromes. Different selection pressures and life history trajectories can lead to behavioural divergence between males and females in many animal species (Michelangeli et al. 2016). However, both sexes are rarely analysed within on study, ignoring the very strong possibility that key components of personality may vary between males and females (Michelangeli et al. 2016). Thus, investigating sex differences in the behavioural (and cognitive) syndrome structures might be another important step towards a better understanding of the maintenance of constant between-individual differences in harvest mice.

Measuring individual variation in cognitive traits appears more difficult than assumed because cognitive trait variability should be carefully interpreted (Rowe and Healy 2014). To confirm the existence of a cognitive syndrome in harvest mice will require further spatial cognitive tests which also capture the within-individual and the between-individual variances. This is, because within the concept of a cognitive syndrome, animals should consistently show the same (spatial) cognitive style, and individuals should solve various tasks equally well (Sih and Del Giudice 2012). If harvest mice of the field population also show within-individual and between-individual variance in spatial learning ability is currently not known. Own unpublished observations indicate that harvest mice of the field population show similar trait variability but solve the Spatial Orientation Task on average faster than mice of the laboratory population (data not shown). We recommend further investigations of cognitive syndromes in harvest mice, especially in populations from a natural genetic background, to investigate whether the observed cognitive syndrome of the laboratory population reflects what can also be found in natural populations.

Finally, to better understand what factors influence constant between-individual differences of behaviours and spatial cognitive traits, as well as the relationships between them in harvest mice, more populations should be compared. In comparison to the predator mediated behavioural syndromes in sticklebacks (syndrome structure was only found under high predation, Bell 2005), behavioural repeatability and consistency, as well as behavioural and cognitive syndromes might be influenced by predation risk, or by populations density (and thus intraspecific competition) in harvest mice. The influences of those environmental factors on constant between-individual differences can be investigated in experimentally manipulated captive populations under semi-natural conditions. Alternatively, this question can be tackled by a study across several wild populations that inhabit areas with varying vegetation structure, or varying predation pressure.

Danksagung

An dieser Stelle möchte ich mich bei allen bedanken, die es mir ermöglicht haben diese Doktorarbeit fertigzustellen.

Zuerst möchte ich mich bei Prof. Dr. Katharina Foerster bedanken für die Möglichkeit diese Thesis anfertigen zu dürfen. Liebe Katharina, vielen Dank für deine Ideen und Anregungen, deine Unterstützung und Hilfe, für die vielen Freiräume und dein Vertrauen in mich während der letzten Jahre. Es war eine tolle und spannende Zeit in Tübingen, in der ich viel gelernt und mich weiterentwickelt habe. Danke für Alles!

Ein herzlicher Dank geht an Prof. Dr. Jana Eccard für die Betreuung aus der Ferne und die Begutachtung meiner Arbeit. Liebe Jana, vielen Dank für deine Unterstützung und Ratschläge, für die hilfreichen Diskussionen in Gülpe und für die Möglichkeit „meine“ Zwergmäuse in Potsdam fangen zu können. Danke, dass du mich in Potsdam so gut für die Mäusewelt vorbereitet hast.

Ich bedanke mich bei Prof. Dr. Andreas Nieder und Dr. Katja Heubel für die Betreuung im EVEREST Komitee und die vielen hilfreichen Anregungen und Ratschläge.

Ein großes Dankeschön geht an Anne Cohrs und Maria Thrun für die Hilfe bei bürokratischem Papierkram. Dr. Klaus Eisler danke ich für die Hilfe bei jeglichen EDV- und Technikproblemen. Vielen Dank, Anne und Herr Eisler, für die gemeinsamen Mittagessen und die vielen geteilten Kuchen ☺

Herrn Dieter Georgi danke ich für den Bau der Überwinterungshütten für die Zwergmäuse und der Versuchsarenen für die Verhaltenstests.

Nur dank der Tierpfleger der Uni Tübingen war es möglich so viele Zwergmäuse im Labor zu organisieren. Vor allem bedanke ich mich bei Gisela Wohlbold und Melanie Anzalone für die Hilfe und Unterstützung bei den Zwergen. Bei Dr. Annette Denzinger bedanke ich mich für die Hilfe bei der Versuchsraumorganisation und den Versuchsanträgen. Den Tierärzten des tierärztlichen Dienstes, vor allem Dr. Susanne Gerold, danke ich für die Unterstützung bei den Transpondermarkierungen und für die tierärztliche Versorgung der Zwergmäuse.

Ohne die tatkräftige Unterstützung und Hilfe der vielen Hiwis und Studenten wäre es nicht möglich gewesen so viele Zwergmäuse zu testen und immer wieder zu fangen, sowie das Gehege in Stand zu halten. Vielen Dank an Teresa Carl, Danja Fritscher, Annika Luczak, Mara Wolters, Carina Hauer, Uwe Zimmermann, Philipp Löffler, Jannik Beninde, Mischa Schreiner und Sarah Daller.

Für die Durchführung der genetischen Laborarbeit und die lustige Bürogemeinschaft bedanke ich mich bei Tatjana Stooß und Katrin Hils.

Allen Mitarbeitern der Vergleichenden Zoologie in Tübingen, vor allem Linda Katzmann und Christina Nitzsche, danke ich für die tollen Jahre auf der Morgenstelle.

Danke an die EVEREST Kommilitonen für spannende Diskussionen und entspannte, lustige Retreats, und den EVEREST Koordinatoren für die guten Hilfestellungen.

Ich bedanke mich bei Dr. Melanie Dammhahn für die hilfreichen Kommentare und Anregungen zum zweiten Kapitel. Außerdem danke ich allen Mitarbeitern der Tierökologie in Potsdam für die Einladungen zum Tieröko-Retreat in Gülpe und die hilfreichen Diskussionen.

Für das Korrekturlesen, die Gespräche wissenschaftlicher oder privater Art und für alles, was ich über Mäuseartige gelernt habe, danke ich Dr. Antje Herde. Antje, du bist die Beste!

Ich bin froh über die alltägliche Unterstützung, das Zuhören und das Verständnis, das ich mit Deike Lüdtke und Julia Geue teilen konnte. Deike, danke dir für die gute Ablenkung beim Boxen ☺

Ich danke allen meinen Freunden für die gemeinsamen Erlebnisse, Wochenenden, Urlaube und das ein oder andere Bier gegen die kleinen Anzeichen der Verzweiflung. Danke für eure Motivationsanstöße, ihr lieben „Freisinger“ und „Möllner“ ☺

Natürlich bin ich auch unglaublich dankbar für die uneingeschränkte Unterstützung durch meine Familie, die immer an mich geglaubt hat, auch wenn sie oft nicht verstanden hat, was ich da eigentlich die ganzen Jahre mache ☺ Mama, Papa, Oma, Petra, ich hab euch sehr lieb!

Dr. Dominik Grimm gilt mein Dank für das Korrekturlesen meiner Paper-Drafts und für seine unfassbare Geduld mit mir. Dominik, du hast mich immer unterstützt, an mich geglaubt und mich nicht aufgeben lassen. Du bist ein Schatz und ich bin sehr froh dich an meiner Seite zu wissen!

Mein tiefster Dank gilt den 321 Zwergmäusen, die mich in den letzten Jahren begleiten haben und die (meinst brav) meine Versuche über sich ergehen haben lassen. Ohne sie wäre diese Arbeit nicht möglich gewesen. Danke!

Appendix

Supplementary Table 1 Final LMMs with significant confounding factors on personality and cognitive traits in harvest mice. Sample sizes (N) are given as observations/individuals. Generation was fitted as a quadratic function. SE = standard error.

personality/cognitive trait	behavioural test	N	confounding factors	estimate ± SE	p value
activity	Open Field	186/96	generation generation^2	- 0.848±0.247 +0.118±0.032	< 0.001 < 0.001
	Y Maze	184/96	-		
boldness	Scare Test	94/56	-		
	Open Field	186/96	trial number observer	+0.327±0.117 - 0.377±0.149	0.006 0.013
exploration	Novel Object	131/69	housing condition	+0.544±0.182	0.004
	Novel Environment	119/60	trial number	+0.584±0.148	< 0.001
spatial recognition	Y Maze	184/96	-		
spatial learning ability	Spatial Orientation Task	110/57	trial number	- 0.378±0.175	0.035
decision accuracy	Spatial Orientation Task	102/56	-		
decision speed	Spatial Orientation Task	1156/56	observer run number	- 0.117±0.015 +0.025±0.005	< 0.001 < 0.001

We observed that the trial number had a significant effect on three variables. Boldness increased in the second trial of the Open Field, and exploration also increased in the second trial of the Novel Environment as mice visited four tubes faster than in the first trial. These effects were likely caused by a habituation of the mice to the test arenas. Spatial learning ability decreased in the second trial (mice reached the learning criterion on average later than during the first trial). Here, mice might have been less motivated to reach the target box during the second trial, as they recognized the general test set-up. With increasing run numbers during the Spatial Orientation Task, mice decided faster, which may indicate learning and memory of the target box position. The observer had a significant effect on boldness in the Open Field and on decision speed in the Spatial Orientation Task, which could be due to differences in handling of the animals. Harvest mice housed in pairs were further more explorative in the Novel Object test. We discussed confounding factors in more detail in Chapter 1.

SUPPLEMENTARY TABLE 2 Between-individual correlations among personality and cognitive traits (spatial recognition and spatial learning ability) in harvest mice, calculated from a multivariate mixed model. Between-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. Significant correlations are marked in bold, tendencies are printed in italics. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	**activity YM**	**boldness OF**	**boldness ST**	**exploration NO**	**exploration NE**	**spatial recognition**	**learning ability**
activity OF		**0.514±0.149**	-0.019±0.212	**0.884±0.400**	0.028±0.319	*0.371±0.257*	0.024±0.313	**0.793±0.447**
activity YM	0.000		0.175±0.182	**0.590±0.353**	-0.230±0.280	0.148±0.227	-0.066±0.279	-0.216±0.326
boldness OF	0.465	0.168		0.201±0.345	*0.437±0.325*	**0.453±0.263**	-0.102±0.322	0.508±0.407
boldness ST	0.014	0.047	0.280		-0.209±0.619	*1.106±0.689*	0.208±0.504	*1.696±1.138*
exploration NO	0.465	0.206	0.089	0.368		0.359±0.335	-0.183±0.526	0.265±0.475
exploration NE	0.074	0.257	0.042	0.054	0.142		0.280±0.394	-0.030±0.412
spatial recognition	0.470	0.407	0.376	0.340	0.364	0.239		0.562±0.594
learning ability	0.038	0.254	0.106	0.068	0.288	0.471	0.172	

Supplementary Table 3 Within-individual correlations between personality and cognitive traits (spatial recognition and spatial learning ability) in harvest mice, calculated from a multivariate mixed model. Within-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. The significant correlation is marked in bold. NA: within-individual correlation not modelled due to missing data. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	**activity YM**	**boldness OF**	**boldness ST**	**exploration NO**	**exploration NE**	**spatial recognition**	**learning ability**
activity OF		**0.264±0.107**	-0.007±0.106	NA	0.084±0.126	NA	0.092±0.109	NA
activity YM	0.007		-0.045±0.116	NA	0.073±0.125	NA	0.041±0.106	NA
boldness OF	0.474	0.348		NA	0.059±0.125	NA	-0.086±0.111	NA
boldness ST	NA	NA	NA		NA	NA	NA	NA
exploration NO	0.251	0.278	0.319	NA		NA	0.094±0.119	NA
exploration NE	NA	NA	NA	NA	NA		NA	NA
spatial recognition	0.198	0.348	0.218	NA	0.215	NA		NA
learning ability	NA	NA	NA	NA	NA	NA	NA	

SUPPLEMENTARY TABLE 4 Test for a cognitive syndrome caused by a speed-accuracy trade-off in harvest mice: Between-individual correlations among personality traits and decision styles (decision speed and accuracy) calculated from a multivariate mixed model. Between-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. Significant correlations are marked in bold, tendencies are printed in italics. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test

	activity OF	**activity YM**	**boldness OF**	**boldness ST**	**exploration NO**	**exploration NE**	**decision speed**	**decision accuracy**
activity OF		**0.546±0.145**	0.016±0.212	**0.877±0.351**	0.023±0.316	*0.408±0.254*	0.179±0.190	0.606±0.672
activity YM	0.000		0.177±0.182	**0.593±0.318**	-0.220±0.278	0.140±0.227	0.164±0.173	1.136±0.999
boldness OF	0.470	0.166		0.159±0.325	0.412±0.324	**0.469±0.262**	**-0.412±0.179**	0.166±0.517
boldness ST	0.006	0.031	0.312		-0.402±0.592	*0.928±0.593*	-0.393±0.385	0.190±1.167
exploration NO	0.472	0.214	0.102	0.248		0.342±0.333	0.093±0.243	-0.357±0.680
exploration NE	0.054	0.270	0.037	0.059	0.152		-0.134±0.222	-0.079±0.557
decision speed	0.173	0.172	0.011	0.154	0.351	0.274		0.207±0.425
decision accuracy	0.184	0.128	0.374	0.435	0.300	0.443	0.313	

SUPPLEMENTARY TABLE 5 Test for a cognitive syndrome caused by a speed-accuracy trade-off in harvest mice: Within-individual correlations between personality traits and decision styles (decision speed and accuracy) calculated from a multivariate mixed model. Within-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. The significant correlation is marked in bold. NA: within-individual correlation not modelled due to missing data. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	**activity YM**	**boldness OF**	**boldness ST**	**exploration NO**	**exploration NE**	**decision speed**	**decision accuracy**
activity OF		**0.258±0.108**	-0.011±0.106	NA	0.067±0.125	NA	NA	NA
activity YM	0.008		-0.041±0.116	NA	0.083±0.126	NA	NA	NA
boldness OF	0.461	0.364		NA	0.061±0.125	NA	NA	NA
boldness ST	NA	NA	NA		NA	NA	NA	NA
exploration NO	0.296	0.255	0.313	NA		NA	NA	NA
exploration NE	NA	NA	NA	NA	NA		NA	NA
decision speed	NA	NA	NA	NA	NA	NA		NA
decision accuracy	NA	NA	NA	NA	NA	NA	NA	

Supplementary Table 6 Final LMMs with significant confounding factors (indicated by F values and corresponding p values) on personality and cognitive traits in harvest mice of the field population. Significant differences between factor levels and contrasts which tended do differ are shown by the results of Tukey post-hoc tests (estimates ± standard error, z values and p values). Sample sizes (N) are given as observations/individuals. SE = standard error.

personality/cognitive trait	behavioural test	N	confounding factors	F value	p value	contrast	estimate ± SE	z value	p value
activity	Open Field	208/95	trial number	5.393	0.002	2 - 1	-0.32 ± 0.11	-2.831	0.022
						3 - 2	0.62 ± 0.17	3.645	0.001
	Y Maze	209/94	test time	10.889	0.001		-0.21 ± 0.06	-3.300	0.001
boldness	Scare Test	69/36	none						
	Open Field	208/95	trial number	18.482	< 0.001	2 - 1	0.83 ± 0.11	7.230	< 0.001
						4 - 1	0.81 ± 0.28	2.903	0.017
						3 - 2	-0.54 ± 0.17	-3.164	0.007
exploration	Novel Object	192/83	trial number	7.763	< 0.001	2 - 1	0.78 ± 0-15	5.181	< 0.001
						3 - 2	-0.44 ± 0.19	-2.304	0.089
			year	4.928	0.001	2014 - 2013	0.66 ± 0.26	2.543	0.028
						2015 - 2013	0.82 ± 0.26	3.138	0.004
	Novel Environment	85/37	trial number	5.301	0.003	2 - 1	0.66 ± 0.20	3.376	0.004
						4 - 1	2.21 ± 0.91	2.428	0.060
spatial recognition	Y Maze	209/94	none						

We observed that the trial number had a significant effect on four behavioural variables. Activity in the Open Field decreased from the first to the second observation, but increased again significantly to the third observation. Boldness increased in the second trial of the Open Field (animals spent more time in the middle of the arena), but decreased again to the third observation. Exploration increased in the second trial of the Novel Object test, as well as of the Novel Environment test, where mice visited four tubes faster than in the first trial. These effects were likely caused by a habituation of the mice to the test arenas and to handling situations. The year had a significant effect on exploration in the Novel Object test. Animal explored the novel objects more in 2014 and 2015 compared to 2013. This could be due to the fact that mice tested in 2013 were wild-caught animals and individuals tested in 2014 and 2015 were born in the enclosure. The test time had a significant effect on activity in the Y Maze, where activity decreased later during the day. This could be due to different activity phases of the animals during the day. We discussed confounding factors in more detail in Chapter 1 and 2.

SUPPLEMENTARY TABLE 7 Final LMMs with significant confounding factors (indicated by F values and corresponding p values) on personality and cognitive traits in harvest mice of the laboratory population. Significant differences between factor levels and contrasts which tended do differ are shown by the results of Tukey post-hoc tests (estimates ± standard error, z values and p values). Sample sizes (N) are given as observations/individuals. SE = standard error.

personality/cognitive trait	behavioural test	N	confounding factors	F value	p value	contrast	estimate ± SE	z value	p value
activity	Open Field	315/112	trial number	4.867	0.003	2 - 1	-0.36 ± 0.09	-3.794	< 0.001
						4 - 1	-0.34 ± 0.15	-2.319	0.090
			generation	2.984	0.087		-0.61 ± 0.18	-3.386	0.001
			generation^2	15.052	< 0.001		0.09 ± 0.02	3.880	< 0.001
	Y Maze	312/112	trial number	6.705	< 0.001	2 - 1	-0.41 ± 0.10	-3.991	< 0.001
						3 - 1	-0.46 ± 0.12	-3.777	< 0.001
			generation	4.467	0.004		-0.44 ± 0.18	-2.412	0.018
			generation^2	8.835	< 0.001		0.07 ± 0.02	2.972	0.004
boldness	Scare Test	100/59	none						
	Open Field	315/112	trial number	17.248	< 0.001	2 - 1	0.53 ± 0.10	5.169	< 0.001
						3 - 1	0.44 ± 0.12	3.622	0.002
						4 - 1	1.09 ± 0.16	6.879	< 0.001
						4 - 2	0.57 ± 0.16	3.544	0.002
						4 - 3	0.66 ± 0.17	3.972	< 0.001
			year	10.531	< 0.001	2013 - 2012	-0.67 ± 0.20	-3.445	0.002
						2014 - 2012	-0.84 ± 0.18	-4.553	< 0.001
exploration	Novel Object	251/83	observer	13.618	< 0.001		0.50 ± 0.13	3.690	< 0.001
	Novel Environment	119/60	trial number	15.498	< 0.001		0.58 ± 0.15	3.937	< 0.001
spatial recognition	Y Maze	312/112	trial number	3.102	0.028	4 - 2	0.51 ± 0.20	2.612	0.043

In the data set of the laboratory population we also observed that the trial number had a significant effect on five behavioural variables. Activity in the Open Field decreased from the first to the second observation, and decreased from the first to the fourth observation. Boldness in the Open Field increased with the trial number (animals spent more time in the middle of the arena). Exploration in the Novel Environment test increased in the second trial, where mice visited four tubes faster than in the first trial. Animal also spent more time in the novel arm of the Y Maze during the fourth observation compared to the second. These effects were likely caused by a habituation of the mice to the test arenas and to handling situations. The year had a significant effect on boldness in the Open Field. Animal were bolder in 2012 compared to 2013 and 2014. The observer had an effect on exploration in the Novel Object test. This could be related to the year effect as one observer did all of the observations in 2012. We cannot disentangle those effects in this data set, however, we recommend to account for an observer effect when animal are recorded manually due to different handling and individual perception patterns which may influence behaviour of animals or measurements. We discussed confounding factors in more detail in Chapter 1 and 2. The individuals of the laboratory population were from seven generations. The quadratic function of the generation had an effect on activity in the Open Field and in the Y Maze. Laboratory conditions may influence natural behaviour. Especially, if behaviours are heritable, the effect of the generation animals spent in the laboratory may have an effect on behavioural expressions due to inbreeding. However, further conclusions are not possible, as so far heritability of tested behavioural traits in our study species is not known.

In sum, a habituation effect shown by the confounding factor *trial number* occurred in the laboratory and the field population of harvest mice. Effect sizes were in general in a similar range and the effect was mostly in the same direction. Thus, repeated observations seem to effect harvest mice similarly, independent of the genetic background of the population or the housing condition (laboratory versus field enclosure).

Supplementary Table 8 Within-individual correlations among personality traits in harvest mice of the field population, calculated from a multivariate mixed model. Within-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. Significant correlations are marked in bold, tendencies are printed in italics. NA: within-individual correlation not modelled due to missing data. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	activity YM	boldness OF	boldness ST	exploration NO	exploration NE
activity OF		0.087±0.098	-0.013±0.095	0.185±0.166	*-0.123±0.096*	**0.222±0.131**
activity YM	0.186		-0.118±0.096	-0.029±0.161	-0.015±0.094	0.030±0.137
boldness OF	0.444	0.110		-0.021±0.163	*-0.128±0.095*	-0.039±0.156
boldness ST	0.133	0.429	0.450		**-0.637±0.127**	NA
exploration NO	0.099	0.438	0.089	0.000		-0.084±0.135
exploration NE	0.045	0.415	0.401	NA	0.268	

Supplementary Table 9 Within-individual correlations between personality traits in harvest mice of the laboratory population, calculated from a multivariate mixed model. Within-individual correlation coefficients (± standard errors) are shown above, and p values below the diagonal. Significant correlations are marked in bold, tendencies are printed in italics. NA: within-individual correlation not modelled due to missing data. OF = Open Field, YM = Y Maze, NO = Novel Object, NE = Novel Environment, ST = Scare Test.

	activity OF	activity YM	boldness OF	boldness ST	exploration NO	exploration NE
activity OF		**0.239±0.067**	0.049±0.070	NA	**0.187±0.075**	NA
activity YM	0.000		*0.094±0.070*	NA	*0.103±0.076*	NA
boldness OF	0.241	0.090		NA	0.044±0.078	NA
boldness ST	NA	NA	NA		NA	NA
exploration NO	0.006	0.089	0.288	NA		NA
exploration NE	NA	NA	NA	NA	NA	

List of Figures

FIGURE 1 NUMBER OF PUBLICATIONS PER YEAR BETWEEN 2000 AND 2012, DEALING WITH EITHER ANIMAL PERSONALITY, BEHAVIOURAL SYNDROME OR TEMPERAMENT. GRAPHIC IS REUSED FROM MACKAY AND HASKELL (2015). 2

FIGURE 2 DIFFERENT BEHAVIOURAL TYPES FORMING A BEHAVIOURAL SYNDROME BETWEEN BEHAVIOUR X AND Y. EACH DOT INDICATES AN INDIVIDUAL IN A POPULATION. WITH PERMISSION (LICENSE NUMBER 3879250630192) FROM BELL (2007).. 5

FIGURE 3 TWO MAIN APPROACHES OF BEHAVIOURAL SYNDROMES IN ANIMAL SPECIES. (A) THE FAST-SLOW SYNDROME (TITULAER ET AL. 2012) DEFINED BY POSITIVE CORRELATIONS BETWEEN BEHAVIOURS. (B) THE PROACTIVE-REACTIVE SYNDROME (KOOLHAAS ET AL. 1999) DEFINED BY A POSITIVE CORRELATION BETWEEN ACTIVITY AND BOLDNESS, BUT NEGATIVE CORRELATIONS BETWEEN EXPLORATION AND ACTIVITY, AS WELL AS BETWEEN EXPLORATION AND BOLDNESS. 15

FIGURE 4 THE CONCEPT OF COGNITIVE SYNDROMES PROVING A POSSIBLE LINK BETWEEN BEHAVIOURAL TYPES AND COGNITIVE STYLES THROUGH A RISK-REWARD TRADE-OFF. WITH PERMISSION (LICENSE NUMBER 3845950396542) FROM SIH AND DEL GIUDICE (2012). 25

FIGURE 5 PICTURE OF MY STUDY SPECIES – THE EURASIAN HARVEST MOUSE – IN THE LABORATORY (A) AND IN THE SEMI-NATURAL OUTDOOR ENCLOSURE OF THE EBERHARD KARLS UNIVERSITY TÜBINGEN (B). PHOTOS BY ANDREA C. SCHUSTER 28

FIGURE 6 POLYCARBONATE CAGE IN WHICH HARVEST MICE WERE HOUSED IN (SEPARATELY OR IN PAIRS OF EQUAL SEX). ARTIFICIAL NEST, RUNNING WHEEL, PAPER TUBE, WOODEN BRANCH AND SHEAF OF GRAIN FOR ENVIRONMENTAL ENRICHMENT. PHOTO BY ANDREA C. SCHUSTER 41

FIGURE 7 OPEN FIELD ARENA (DIAMETER 51 CM) USED TO QUANTIFY ACTIVITY AND BOLDNESS OF HARVEST MICE. MORTAR BUCKET PAINTED WITH WHITE COLOUR FOR EFFICIENT AUTOMATED VIDEO TRACKING RESULTS. PHOTO BY ANDREA C. SCHUSTER 43

FIGURE 8 ITEMS THAT SERVED AS NOVEL OBJECTS DURING NOVEL OBJECT TESTS TO QUANTIFY EXPLORATION BEHAVIOUR OF HARVEST MICE. OBJECTS WERE ABOUT THE SAME SIZE OF THE ANIMALS. PHOTOS BY ANDREA C. SCHUSTER 44

FIGURE 9 Y MAZE USED TO QUANTIFY SPATIAL RECOGNITION AND ACTIVITY IN HARVEST MICE. EACH ARM SURROUNDED BY PLAIN CARDBOARD SUCH THAT ANIMALS COULD ONLY SEE OBJECT CUES PLACED AROUND THE ARM IT WAS IN. PHOTO BY ANDREA C. SCHUSTER 45

FIGURE 10 TEST ARENA OF THE NOVEL ENVIRONMENT SHOWING THE FIVE TUBES CONNECTING BOTTOM AND INTERMEDIATE FLOOR WHICH HARVEST MICE COULD EXPLORE BY CLIMBING. PHOTO BY CHRISTOPH JÄCKLE 65

FIGURE 11 EXPERIMENTAL SET-UP OF THE SPATIAL ORIENTATION TASK. SIX-ARM MAZE ELEVATED 20 CM ABOVE THE GROUND WITH TARGET BOX ON THE RIGHT AND OPTICAL CUES FOR SPATIAL ORIENTATION BETWEEN THE ARMS. PHOTO BY UWE ZIMMERMANN.. 67

FIGURE 12 BEHAVIOURAL SYNDROME: BETWEEN-INDIVIDUAL CORRELATIONS (± STANDARD ERRORS) BETWEEN PERSONALITY TRAITS CALCULATED FROM A MULTIVARIATE MIXED MODEL. SOLID LINES INDICATE SIGNIFICANT BETWEEN-INDIVIDUAL

CORRELATIONS (P < 0.05), DASHED LINES INDICATE TENDENCIES (P < 0.1), AND DOTTED LINES INDICATE NON-SIGNIFICANT BETWEEN-INDIVIDUAL CORRELATIONS. OF: OPEN FIELD, YM: Y MAZE, NO: NOVEL OBJECT, NE: NOVEL ENVIRONMENT, ST: SCARE TEST. 73

FIGURE 13 RELATIONSHIP BETWEEN PERSONALITY AND SPATIAL COGNITION: BETWEEN-INDIVIDUAL CORRELATIONS (± STANDARD ERRORS) BETWEEN PERSONALITY TRAITS AND COGNITIVE TRAITS CALCULATED FROM AN EIGHT-TRAIT MULTIVARIATE MIXED MODEL, BUT ONLY FIVE TRAITS ARE DISPLAYED (SEE EXPLANATION IN THE TEXT). THE SOLID LINE INDICATES THE SIGNIFICANT CORRELATION (P < 0.05) BETWEEN ACTIVITY IN THE OPEN FIELD (OF) AND LEARNING ABILITY IN THE SPATIAL ORIENTATION TASK, THE DASHED LINE INDICATES THE TENDENCY (P < 0.1) BETWEEN BOLDNESS IN THE SCARE TEST (ST) AND LEARNING ABILITY IN THE SPATIAL ORIENTATION TASK, AND DOTTED LINES INDICATE NON-SIGNIFICANT CORRELATIONS BETWEEN THE OTHER TRAITS. 75

FIGURE 14 TEST FOR A COGNITIVE SYNDROME BASED ON A SPEED-ACCURACY TRADE-OFF: BETWEEN-INDIVIDUAL CORRELATIONS (± STANDARD ERRORS) BETWEEN PERSONALITY TRAITS, AND DECISION SPEED AND ACCURACY CALCULATED FROM AN EIGHT-TRAIT MULTIVARIATE MIXED MODEL, BUT ONLY FIVE TRAITS ARE DISPLAYED (SEE EXPLANATION IN THE TEXT). THE SOLID LINE INDICATES THE SIGNIFICANT CORRELATION (P < 0.05) BETWEEN ACTIVITY IN THE OPEN FIELD (OF) AND DECISION SPEED IN THE SPATIAL ORIENTATION TASK, DOTTED LINES INDICATE NON-SIGNIFICANT CORRELATIONS BETWEEN THE OTHER TRAITS. 75

FIGURE 15 THE SEMI-NATURAL OUTDOOR ENCLOSURE OF THE HARVEST MOUSE POPULATION LOCATED NEAR THE EBERHARD KARLS UNIVERSITY IN TÜBINGEN, GERMANY. PHOTO BY ANDREA C. SCHUSTER 87

FIGURE 16 BEHAVIOURAL SYNDROME IN THE FIELD POPULATION OF EURASIAN HARVEST MICE: BETWEEN-INDIVIDUAL CORRELATIONS (± STANDARD ERRORS) AMONG PERSONALITY TRAITS CALCULATED FROM A MULTIVARIATE MIXED MODEL. SOLID LINES INDICATE SIGNIFICANT CORRELATIONS (P < 0.05), DASHED LINES INDICATE TENDENCIES (P < 0.1), AND DOTTED LINES INDICATE NON-SIGNIFICANT CORRELATIONS. OF: OPEN FIELD, YM: Y MAZE, NO: NOVEL OBJECT, NE: NOVEL ENVIRONMENT, ST: SCARE TEST. THE COVARIANCE BETWEEN EXPLORATION NE AND BOLDNESS ST WAS NOT MODELLED DUE TO MISSING DATA. 96

FIGURE 17 BEHAVIOURAL SYNDROME IN THE LABORATORY POPULATION OF EURASIAN HARVEST MICE: BETWEEN-INDIVIDUAL CORRELATIONS (± STANDARD ERRORS) AMONG PERSONALITY TRAITS CALCULATED FROM A MULTIVARIATE MIXED MODEL. SOLID LINES INDICATE SIGNIFICANT CORRELATIONS (P < 0.05), AND DOTTED LINES INDICATE NON-SIGNIFICANT CORRELATIONS. OF: OPEN FIELD, YM: Y MAZE, NO: NOVEL OBJECT, NE: NOVEL ENVIRONMENT, ST: SCARE TEST. 97

List of Tables

TABLE 1 OVERVIEW OF THE TERMINOLOGY USED IN THIS THESIS REGARDING CONSTANT INDIVIDUAL BEHAVIOURAL DIFFERENCES. TERMS AND CORRESPONDING DEFINITIONS ARE SHOWN, DETAILED EXPLANATIONS AND REFERENCES ARE FOUND IN THE TEXT. 6

TABLE 2 CHRONOLOGY OF BEHAVIOURAL EXPERIMENTS CONDUCTED IN A LABORATORY AND AN ENCLOSURE POPULATION OF HARVEST MICE. BEHAVIOURAL TEST: Y MAZE (YM), OPEN FIELD (OF), NOVEL OBJECT (NO), SCARE TEST (ST), NOVEL ENVIRONMENT (NE), SPATIAL ORIENTATION TASK (SOT). * INDICATES THAT BEHAVIOURAL TESTS WERE CONDUCTED WITH THE HELP OF STUDENT HELPERS AND BACHELOR CANDIDATES. FOCUS OF THE ANALYSIS AND REFERRING CHAPTERS WITHIN THE THESIS ARE MENTIONED. 30

TABLE 3 THE NUMBER OF TESTED ANIMALS IN EACH OF THE THREE BEHAVIOURAL TESTS FOR FOUR DIFFERENT AGE CLASSES (WEEK 6, 7, 12 AND 24) 42

TABLE 4 RESULTS OF FINAL LMMS WITH CONFOUNDING EFFECTS ON PERSONALITY TRAITS. SE = STANDARD ERROR 47

TABLE 5 RESULTS OF REPEATABILITY ANALYSES SHOWING R VALUES CALCULATED FOR EACH PERSONALITY TRAIT PER EXPERIMENTAL GROUP (JUVENILES AND ADULTS). SAMPLE SIZES ARE GIVEN: FIRST TRIAL/SECOND TRIAL. BOLD P VALUES WERE SIGNIFICANT AFTER BONFERRONI CORRECTION (P < 0.008). OF: OPEN FIELD TEST, YM: Y MAZE TEST. NA: MODEL DID NOT CONVERGE BECAUSE OF TOO SMALL/NEGATIVE VALUE OF R 51

TABLE 6 RESULTS OF CONSISTENCY ANALYSES SHOWING R VALUES CALCULATED FOR EACH PERSONALITY TRAIT PER EXPERIMENTAL GROUP (BEFORE AND AFTER MATURATION, BEFORE AND AFTER SEXUAL CONTACT). SAMPLE SIZES ARE GIVEN: FIRST TRIAL/SECOND TRIAL. BOLD P VALUES WERE SIGNIFICANT AFTER BONFERRONI CORRECTION (P < 0.008). OF: OPEN FIELD TEST, YM: Y MAZE TEST. NA: MODEL DID NOT CONVERGE BECAUSE OF TOO SMALL/NEGATIVE VALUE OF R 52

TABLE 7 REPEATABILITY (R_A) WITH 95% CONFIDENCE INTERVAL (CI) FOR EACH PERSONALITY AND COGNITIVE TRAIT. GENERATION WAS FITTED AS A QUADRATIC FUNCTION. SAMPLE SIZES (N) ARE GIVEN AS OBSERVATIONS/INDIVIDUALS. SIGNIFICANT REPEATABILITY VALUES ARE MARKED BOLD. 72

TABLE 8 BEHAVIOURAL SYNDROME: WITHIN-INDIVIDUAL CORRELATIONS BETWEEN PERSONALITY TRAITS CALCULATED FROM A MULTIVARIATE MIXED MODEL. WITHIN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. SIGNIFICANT CORRELATION IS MARKED IN BOLD. NA: WITHIN-INDIVIDUAL CORRELATION NOT MODELLED DUE TO MISSING DATA. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST. 74

TABLE 9 NUMBER OF HARVEST MICE CAPTURED THE FIRST TIME IN THE OUTDOOR ENCLOSURE (UNMARKED INDIVIDUALS) IN EACH MONTH (ROMAN NUMERALS) OF THE THREE STUDY YEARS (2013, 2014, 2015). * INDICATES WILD-CAUGHT ANIMALS FROM POTSDAM, WHICH WERE RELEASED TO THE ENCLOSURE. - ARE MONTHS WITHOUT TRAPPING EVENTS. 88

TABLE 10 NUMBER OF THE BEHAVIOURAL OBSERVATIONS ON THE FIELD POPULATION. MICE WERE TESTED ONE TO FOUR TIMES IN EACH OF THE FIVE TESTS. 89

TABLE 11 NUMBER OF THE BEHAVIOURAL OBSERVATIONS OF THE LABORATORY POPULATION. MICE WERE TESTED ONE TO FOUR TIMES IN EACH OF THE FIVE TESTS 90

TABLE 12 REPEATABILITY (R_A) WITH 95% CONFIDENCE INTERVAL (CI) FOR PERSONALITY TRAITS AND SPATIAL RECOGNITION TESTED IN THE FIELD POPULATION OF HARVEST MICE. SAMPLE SIZES (N) ARE GIVEN AS OBSERVATIONS/INDIVIDUALS. SIGNIFICANT REPEATABILITY VALUES ARE MARKED BOLD........ 95

TABLE 13 FINAL LMMS WITH SIGNIFICANT CONFOUNDING FACTORS AND THE EFFECTS OF ORIGIN (FIELD VERSUS LABORATORY POPULATION) ON PERSONALITY AND COGNITIVE TRAITS IN HARVEST MICE SIGNIFICANT DIFFERENCES BETWEEN FACTOR LEVELS AND CONTRASTS WHICH TENDED DO DIFFER ARE SHOWN BY THE RESULTS OF TUKEY POST-HOC TESTS (ESTIMATES ± STANDARD ERROR, Z VALUES AND P VALUES). SAMPLE SIZES (N) ARE GIVEN AS OBSERVATIONS/INDIVIDUALS. SE = STANDARD ERROR. TEST STATISTIC OF LEVENE`S TESTS ARE DISPLAYED IN THE LAST COLUMNS........ 99

TABLE 14 COMPARISON OF BETWEEN-INDIVIDUAL CORRELATIONS FROM THE FIELD AND THE LABORATORY POPULATION. THE DIFFERENCE BETWEEN CORRELATION COEFFICIENTS OF THE FIELD AND THE LABORATORY POPULATION (EFFECT SIZE) ARE DISPLAYED BELOW THE DIAGONAL. Z VALUES ARE SHOWN ABOVE THE DIAGONAL AND ARE MARKED IN BOLD WHEN SIGNIFICANT AFTER BONFERRONI CORRECTION. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST. 100

List of Supplementary Tables

SUPPLEMENTARY TABLE 1 FINAL LMMS WITH SIGNIFICANT CONFOUNDING FACTORS ON PERSONALITY AND COGNITIVE TRAITS IN HARVEST MICE. SAMPLE SIZES (N) ARE GIVEN AS OBSERVATIONS/INDIVIDUALS. GENERATION WAS FITTED AS A QUADRATIC FUNCTION. SE = STANDARD ERROR. XIII

SUPPLEMENTARY TABLE 2 BETWEEN-INDIVIDUAL CORRELATIONS AMONG PERSONALITY AND COGNITIVE TRAITS (SPATIAL RECOGNITION AND SPATIAL LEARNING ABILITY) IN HARVEST MICE, CALCULATED FROM A MULTIVARIATE MIXED MODEL. BETWEEN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. SIGNIFICANT CORRELATIONS ARE MARKED IN BOLD, TENDENCIES ARE PRINTED IN ITALICS. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST. XV

SUPPLEMENTARY TABLE 3 WITHIN-INDIVIDUAL CORRELATIONS BETWEEN PERSONALITY AND COGNITIVE TRAITS (SPATIAL RECOGNITION AND SPATIAL LEARNING ABILITY) IN HARVEST MICE, CALCULATED FROM A MULTIVARIATE MIXED MODEL. WITHIN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. THE SIGNIFICANT CORRELATION IS MARKED IN BOLD. NA: WITHIN-INDIVIDUAL CORRELATION NOT MODELLED DUE TO MISSING DATA. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST. XVI

SUPPLEMENTARY TABLE 4 TEST FOR A COGNITIVE SYNDROME CAUSED BY A SPEED-ACCURACY TRADE-OFF IN HARVEST MICE: BETWEEN-INDIVIDUAL CORRELATIONS AMONG PERSONALITY TRAITS AND DECISION STYLES (DECISION SPEED AND ACCURACY) CALCULATED FROM A MULTIVARIATE MIXED MODEL. BETWEEN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. SIGNIFICANT CORRELATIONS ARE MARKED IN BOLD, TENDENCIES ARE PRINTED IN ITALICS. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST XVII

SUPPLEMENTARY TABLE 5 TEST FOR A COGNITIVE SYNDROME CAUSED BY A SPEED-ACCURACY TRADE-OFF IN HARVEST MICE: WITHIN-INDIVIDUAL CORRELATIONS BETWEEN PERSONALITY TRAITS AND DECISION STYLES (DECISION SPEED AND ACCURACY) CALCULATED FROM A MULTIVARIATE MIXED MODEL. WITHIN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. THE SIGNIFICANT CORRELATION IS MARKED IN BOLD. NA: WITHIN-INDIVIDUAL CORRELATION NOT MODELLED DUE TO MISSING DATA. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST. XVIII

SUPPLEMENTARY TABLE 6 FINAL LMMS WITH SIGNIFICANT CONFOUNDING FACTORS (INDICATED BY F VALUES AND CORRESPONDING P VALUES) ON PERSONALITY AND COGNITIVE TRAITS IN HARVEST MICE OF THE FIELD POPULATION. SIGNIFICANT DIFFERENCES BETWEEN FACTOR LEVELS AND CONTRASTS WHICH TENDED DO DIFFER ARE SHOWN BY THE RESULTS OF TUKEY POST-HOC TESTS (ESTIMATES ± STANDARD ERROR, Z VALUES AND P VALUES). SAMPLE SIZES (N) ARE GIVEN AS OBSERVATIONS/INDIVIDUALS. SE = STANDARD ERROR. XIX

SUPPLEMENTARY TABLE 7 FINAL LMMS WITH SIGNIFICANT CONFOUNDING FACTORS (INDICATED BY F VALUES AND CORRESPONDING P VALUES) ON PERSONALITY AND COGNITIVE TRAITS IN HARVEST MICE OF THE LABORATORY POPULATION. SIGNIFICANT DIFFERENCES BETWEEN FACTOR LEVELS AND CONTRASTS WHICH TENDED DO DIFFER ARE

SHOWN BY THE RESULTS OF TUKEY POST-HOC TESTS (ESTIMATES ± STANDARD ERROR, Z VALUES AND P VALUES). SAMPLE SIZES (N) ARE GIVEN AS OBSERVATIONS/INDIVIDUALS. SE = STANDARD ERROR. XXI

SUPPLEMENTARY TABLE 8 WITHIN-INDIVIDUAL CORRELATIONS AMONG PERSONALITY TRAITS IN HARVEST MICE OF THE FIELD POPULATION, CALCULATED FROM A MULTIVARIATE MIXED MODEL. WITHIN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. SIGNIFICANT CORRELATIONS ARE MARKED IN BOLD, TENDENCIES ARE PRINTED IN ITALICS. NA: WITHIN-INDIVIDUAL CORRELATION NOT MODELLED DUE TO MISSING DATA. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST... XXIII

SUPPLEMENTARY TABLE 9 WITHIN-INDIVIDUAL CORRELATIONS BETWEEN PERSONALITY TRAITS IN HARVEST MICE OF THE LABORATORY POPULATION, CALCULATED FROM A MULTIVARIATE MIXED MODEL. WITHIN-INDIVIDUAL CORRELATION COEFFICIENTS (± STANDARD ERRORS) ARE SHOWN ABOVE, AND P VALUES BELOW THE DIAGONAL. SIGNIFICANT CORRELATIONS ARE MARKED IN BOLD, TENDENCIES ARE PRINTED IN ITALICS. NA: WITHIN-INDIVIDUAL CORRELATION NOT MODELLED DUE TO MISSING DATA. OF = OPEN FIELD, YM = Y MAZE, NO = NOVEL OBJECT, NE = NOVEL ENVIRONMENT, ST = SCARE TEST. XXIII

References

A

Adriaenssens B, Johnsson JI (2013) Natural selection, plasticity and the emergence of a behavioural syndrome in the wild. Ecology Letters 16:47-55

Archard GA, Braithwaite VA (2010) The importance of wild populations in studies of animal temperament. Journal of Zoology 281:149-160

Archer J (1973) Tests for emotionality in rats and mice: a review. Animal Behaviour 21:205-235

Auclair Y, Koenig B, Lindholm AK (2013) A Selfish Genetic Element Influencing Longevity Correlates with Reactive Behavioural Traits in Female House Mice (Mus domesticus). PLoS ONE 8

B

Bajer K, Horvath G, Molnar O, Toeroek J, Zsolt Garamszegi L, Herczeg G (2015) European green lizard (Lacerta viridis) personalities: Linking behavioural types to ecologically relevant traits at different ontogenetic stages. Behavioural Processes 111:67-74

Bates D, Mächler M, Bolker B, Walker S (2014) Fitting linear mixed-effects models using lme4. arXiv preprint arXiv:14065823

Bebus SE, Small TW, Jones BC, Elderbrock EK, Schoech SJ (2016) Associative learning is inversely related to reversal learning and varies with nestling corticosterone exposure. Animal Behaviour 111:251-260

Bell AM (2005) Behavioural differences between individuals and two populations of stickleback (Gasterosteus aculeatus). Journal of Evolutionary Biology 18:464-473

Bell AM (2007) Future directions in behavioural syndromes research. Proceedings of the Royal Society of London B: Biological Sciences 274:755-761

Bell AM, Hankison SJ, Laskowski KL (2009) The repeatability of behaviour: a meta-analysis. Animal Behaviour 77:771-783

Bell AM, Stamps JA (2004) Development of behavioural differences between individuals and populations of sticklebacks, Gasterosteus aculeatus. Animal Behaviour 68:1339-1348

Bengston SE, Pruitt JN, Riechert SE (2014) Differences in environmental enrichment generate contrasting behavioural syndromes in a basal spider lineage. Animal Behaviour 93:105-110

Benus RF, Bohus B, Koolhaas JM, Vanoortmerssen GA (1991) Heritable variation for aggression as a reflection of individual coping strategies. Experientia 47:1008-1019

Benus RF, Dendaas S, Koolhaas JM, Vanoortmerssen GA (1990) Routine Formation and Flexibility in Social and Non-Social Behaviour of Aggressive and Non-Aggressive Male Mice. Behaviour 112:176-193

Benus RF, Koolhaas JM, Vanoortmerssen GA (1987) Individual Differences in Behavioural Reaction To a Changing Environment in Mice and Rats. In, pp 105-122

Bergmüller R (2010) Animal personality and behavioural syndromes. In: Animal behaviour: evolution and mechanisms. Springer, pp 587-621

Biegler R, McGregor A, Krebs JR, Healy SD (2001) A larger hippocampus is associated with longer-lasting spatial memory. Proceedings of the National Academy of Sciences of the United States of America 98:6941-6944

Blumstein DT, Lea AJ, Olson LE, Martin JGA (2010) Heritability of anti-predatory traits: vigilance and locomotor performance in marmots. Journal of Evolutionary Biology 23:879-887

Boehmler W, Carr T, Thisse C, Thisse B, Canfield V, Levenson R (2007) D4 Dopamine receptor genes of zebrafish and effects of the antipsychotic clozapine on larval swimming behaviour. Genes, Brain and Behavior 6:155-166

Bohme W (1969) Beitrag zur Kenntnis der Zwergmaus Micromys minutus Pallas, 1778. (Rodentia, Muridae). Faunistisch-Oekologische Mitteilungen 3:247-254

Boogert NJ, Reader SM, Laland KN (2006) The relation between social rank, neophobia and individual learning in starlings. Animal Behaviour 72:1229-1239

Boon AK, Réale D, Boutin S (2007) The interaction between personality, offspring fitness and food abundance in North American red squirrels. Ecology Letters 10:1094-1104

Both C, Dingemanse NJ, Drent PJ, Tinbergen JM (2005) Pairs of extreme avian personalities have highest reproductive success. Journal of Animal Ecology 74:667-674

Boulton K, Grimmer AJ, Rosenthal GG, Walling CA, Wilson AJ (2014) How stable are personalities? A multivariate view of behavioural variation over long and short timescales in the sheepshead swordtail, Xiphophorus birchmanni. Behavioral ecology and sociobiology 68:791-803

Bousquet CAH, Petit O, Arrive M, Robin J-P, Sueur C (2015) Personality tests predict responses to a spatial-learning task in mallards, Anas platyrhynchos. Animal Behaviour 110:145-154

Boyer N, Réale D, Marmet J, Pisanu B, Chapuis JL (2010) Personality, space use and tick load in an introduced population of Siberian chipmunks Tamias sibiricus. Journal of Animal Ecology 79:538-547

Braun M, Dieterlen F (2005) Die Säugetiere Baden-Württembergs 2, vol 2, 1 edn. Ulmer

Briffa M, Rundle SD, Fryer A (2008) Comparing the strength of behavioural plasticity and consistency across situations: animal personalities in the hermit crab Pagurus bernhardus. Proceedings of the Royal Society of London B: Biological Sciences 275:1305-1311

Brodin T (2009) Behavioral syndrome over the boundaries of life - carryovers from larvae to adult damselfly. Behav Ecol 20

Brommer JE, Class B (2015) The importance of genotype-by-age interactions for the development of repeatable behavior and correlated behaviors over lifetime. Front Zool 12:1-13

Brust V, Guenther A (2015) Domestication effects on behavioural traits and learning performance: comparing wild cavies to guinea pigs. Animal Cognition 18:99-109

Brydges NM, Colegrave N, Heathcote RJ, Braithwaite VA (2008) Habitat stability and predation pressure affect temperament behaviours in populations of three-spined sticklebacks. Journal of Animal Ecology 77:229-235

Budaev SV, Zworykin DD, Mochek AD (1999) Individual differences in parental care and behaviour profile in the convict cichlid: a correlation study. Animal Behaviour 58:195-202

Burns JG, Rodd FH (2008) Hastiness, brain size and predation regime affect the performance of wild guppies in a spatial memory task. Animal Behaviour 76:911-922

C

Carazo P, Noble DWA, Chandrasoma D, Whiting MJ (2014) Sex and boldness explain individual differences in spatial learning in a lizard. Proc Biol Sci 281:20133275

Carere C, Drent PJ, Privitera L, Koolhaas JM, Groothuis TGG (2005) Personalities in great tits, Parus major: stability and consistency. Animal Behaviour 70:795-805

Carere C, Maestripieri D (2013) Animal personalities: behavior, physiology, and evolution. University of Chicago Press

Carlstead K, Mellen J, Kleiman DG (1999) Black rhinoceros (Diceros bicornis) in US zoos: I. Individual behavior profiles and their relationship to breeding success. Zoo Biology 18:17-34

Carter A, Goldizen A, Heinsohn R (2012) Personality and plasticity: temporal behavioural reaction norms in a lizard, the Namibian rock agama. Animal Behaviour 84:471-477

Chapman BB, Hegg A, Ljungberg P (2013) Sex and the syndrome: individual and population consistency in behaviour in rock pool prawn Palaemon elegans. PLoS ONE 8:e59437

Chittka L, Skorupski P, Raine NE (2009) Speed-accuracy tradeoffs in animal decision making. Trends in Ecology & Evolution 24:400-407

Chitty D, Shorten M (1946) Techniques for the study of the Norway rat (Rattus norvegicus). Jour Mammal 27:63-78

Class B, Brommer JE (2016) Senescence of personality in a wild bird. Behavioral Ecology and Sociobiology 70:733-744

Cole EF, Morand-Ferron J, Hinks AE, Quinn JL (2012) Cognitive Ability Influences Reproductive Life History Variation in the Wild. Current Biology 22:1808-1812

Cole EF, Quinn JL (2012) Personality and problem-solving performance explain competitive ability in the wild. Proc R Soc B-Biol Sci 279:1168-1175

Cote J, Dreiss A, Clobert J (2008) Social personality trait and fitness. Proceedings of the Royal Society of London B: Biological Sciences 275:2851-2858

Cote J, Fogarty S, Weinersmith K, Brodin T, Sih A (2010) Personality traits and dispersal tendency in the invasive mosquitofish (Gambusia affinis). Proceedings of the Royal Society of London B: Biological Sciences:rspb20092128

Croston R, Branch CL, Kozlovsky DY, Dukas R, Pravosudov VV (2015) Heritability and the evolution of cognitive traits. Behavioral Ecology 26:1447-1459

D

D'Hooge R, De Deyn PP (2001) Applications of the Morris water maze in the study of learning and memory. Brain Research Reviews 36:60-90

da Luz Correia A (2013) Ein Test für das proaktiv-reaktive Verhaltenssyndrom bei Zwergmäusen (Micromys minutus). In. Eberhard Karls Universität Tübingen, Tübingen

Dall SRX, Houston AI, McNamara JM (2004) The behavioural ecology of personality: consistent individual differences from an adaptive perspective. Ecology Letters 7:734-739

Dammhahn M (2012) Are personality differences in a small iteroparous mammal maintained by a life-history trade-off? Proc R Soc B-Biol Sci 279:2645-2651

Darinot F (2016) The harvest mouse (Micromys minutus Pallas, 1771) as prey: a literature review. Folia Zoologica 65:117-135

David M, Auclair Y, Cezilly F (2012) Assessing Short- and Long-Term Repeatability and Stability of Personality in Captive Zebra Finches Using Longitudinal Data. Ethology 118:932-942

David M, Dall SRX (2016) Unravelling the Philosophies Underlying "Animal Personality' Studies: A Brief Re-Appraisal of the Field. Ethology 122:1-9

Dellu F, Mayo W, Cherkaoui J, Le Moal M, Simon H (1992) A two-trial memory task with automated recording: study in young and aged rats. Brain research 588:132-139

DePasquale C, Wagner T, Archard GA, Ferguson B, Braithwaite VA (2014) Learning rate and temperament in a high predation risk environment. Oecologia 176:661-667

DeWitt TJ, Sih A, Wilson DS (1998) Costs and limits of phenotypic plasticity. Trends in Ecology & Evolution 13:77-81

Dingemanse N, Both C, Drent PJ, van Oers K, van Noordwijk A (2002) Repeatability and heritability of exploratory behaviour in great tits from the wild. Animal Behaviour 64:929-938

Dingemanse N, Réale D (2013) What is the evidence for natural selection maintaining animal personality variation? In: Animal personalities: Behavior, physiology, and evolution. University of Chicago Press

Dingemanse NJ, Both C, Drent PJ, Tinbergen JM (2004) Fitness consequences of avian personalities in a fluctuating environment. Proc R Soc B-Biol Sci 271:847-852

Dingemanse NJ, de Goede P (2004) The relation between dominance and exploratory behavior is context-dependent in wild great tits. Behavioral Ecology 15:1023-1030

Dingemanse NJ, Dochtermann NA (2013) Quantifying individual variation in behaviour: mixed-effect modelling approaches. Journal of Animal Ecology 82:39-54

Dingemanse NJ, Kazem AJN, Réale D, Wright J (2010) Behavioural reaction norms: animal personality meets individual plasticity. Trends in Ecology & Evolution 25:81-89

Dingemanse NJ, Réale D (2005) Natural selection and animal personality. Behaviour 142:1159-1184

Dingemanse NJ, Wright J, Kazem AJ, Thomas DK, Hickling R, Dawnay N (2007) Behavioural syndromes differ predictably between 12 populations of three-spined stickleback. Journal of Animal Ecology 76:1128-1138

Dochtermann NA, Schwab T, Sih A (2015) The contribution of additive genetic variation to personality variation: heritability of personality. Proc R Soc B-Biol Sci 282

Dosmann AJ, Brooks KC, Mateo JM (2015) Within-Individual Correlations Reveal Link Between a Behavioral Syndrome, Condition, and Cortisol in Free-Ranging Belding's Ground Squirrels. Ethology 121:125-134

Drent PJ, Oers Kv, Noordwijk AJv (2003) Realized heritability of personalities in the great tit (Parus major). Proceedings of the Royal Society of London Series B: Biological Sciences 270:45-51

Ducatez S, Audet JN, Lefebvre L (2015) Problem-solving and learning in Carib grackles: individuals show a consistent speed-accuracy trade-off. Animal Cognition 18:485-496

Duckworth RA (2008) Adaptive dispersal strategies and the dynamics of a range expansion. the american naturalist 172:S4-S17

Dukas R (2004) Evolutionary biology of animal cognition. Annual Review of Ecology Evolution and Systematics 35:347-374

Dukas R (2009) Learning: mechanisms, ecology, and evolution. Cognitive ecology II:7-26

Dukas R, Bernays EA (2000) Learning improves growth rate in grasshoppers. Proceedings of the National Academy of Sciences of the United States of America 97:2637-2640

Dyer FC (1998) Cognitive ecology of navigation. Cognitive ecology (R Dukas, ed) University of Chicago Press, Chicago, Illinois:201-260

E

Eaves L, Martin N, Heath A, Hewitt J, Neale M (1990) Personality and reproductive fitness. Behavior genetics 20:563-568

Eccard JA, Herde A (2013) Seasonal variation in the behaviour of a short-lived rodent. Bmc Ecology 13

Eccard JA, Rödel HG (2011) Optimizing temperament through litter size in short-lived, iteroparous mammals in seasonal environments. Developmental Psychobiology 53:585-591

Etienne AS, Jeffery KJ (2004) Path integration in mammals. Hippocampus 14:180-192

Evans J, Boudreau K, Hyman J (2010) Behavioural syndromes in urban and rural populations of song sparrows. Ethology 116:588-595

F

Falconer DS, Mackay TF, Frankham R (1996) Introduction to quantitative genetics (4th edn). Trends in Genetics 12:280

Favati A, Zidar J, Thorpe H, Jensen P, Løvlie H (2016) The ontogeny of personality traits in the red junglefowl, Gallus gallus. Behavioral Ecology 27:484-493

Feldmann R (1984) Die Saugetiere Westfalens. Zwergmaus - Micromys minutus (Pallas, 1778). Abhandlungen aus dem Westfaelischen Museum fuer Naturkunde 46:221-230

Feldmann R (1997) Studies on the autoecology and reproductive biology of the harvest mouse Micromys minutus. Abhandlungen aus dem Westfaelischen Museum fuer Naturkunde 59:107-115

Ferrari S, Millot S, Leguay D, Chatain B, Begout M-L (2015) Consistency in European seabass coping styles: A life-history approach. Applied Animal Behaviour Science 167:74-88

Fidler AE, van Oers K, Drent PJ, Kuhn S, Mueller JC, Kempenaers B (2007) Drd4 gene polymorphisms are associated with personality variation in a passerine bird. Proceedings of the Royal Society of London B: Biological Sciences 274:1685-1691

Fischer EK, Ghalambor CK, Hoke KL (2016) Plasticity and evolution in correlated suites of traits. Journal of evolutionary biology

Floresco SB (2015) Spatial learning in animals. Encyclopedia of Psychopharmacology:1620-1623

Frank F (1957) Zucht und Gefangenschafts-Biologie der Zwergmaus (Micromys minutus subobscurus Fritsche). Zeitschrift fuer Saugietierkunde 22:1-44

G

Galton F (1883) Inquiries into the human faculty & its development. JM Dent and Company

Garamszegi LZ, Marko G, Herczeg G (2013) A meta-analysis of correlated behaviors with implications for behavioral syndromes: relationships between particular behavioral traits. Behavioral Ecology 24:1068-1080

Garamszegi LZ, Markó G, Herczeg G (2012) A meta-analysis of correlated behaviours with implications for behavioural syndromes: mean effect size, publication bias, phylogenetic effects and the role of mediator variables. Evolutionary Ecology 26:1213-1235

Gilmour A, Gogel B, Cullis B, Welham S, Thompson R, Butler D, Cherry M, Collins D, Dutkowski G, Harding S (2014) ASReml user guide. Release 4.1 structural specification. VSN International Ltd, Hemel Hempstead, HP1 1ES, UK www vsni co uk

Goldsmith HH, Buss AH, Plomin R, Rothbart MK, Thomas A, Chess S, Hinde RA, McCall RB (1987) Roundtable: What is temperament? Four approaches. Child development:505-529

Gosling SD (2001) From mice to men: What can we learn about personality from animal research? Psychological Bulletin 127:45-86

Griffin AS, Guillette LM, Healy SD (2015) Cognition and personality: an analysis of an emerging field. Trends in Ecology & Evolution 30:207-214

Gross MR (1996) Alternative reproductive strategies and tactics: diversity within sexes. Trends in Ecology & Evolution 11:92-98

Gruszka A, Matthews G, Szymura B (2010) Handbook of Individual Differences in Cognition. Springer

Guenther A, Brust V, Dersen M, Trillmich F (2014a) Learning and Personality Types Are Related in Cavies (Cavia aperea). Journal of Comparative Psychology 128:74-81

Guenther A, Finkemeier MA, Trillmich F (2014b) The ontogeny of personality in the wild guinea pig. Animal Behaviour 90:131-139

Guillette LM, Hahn AH, Hoeschele M, Przyslupski A-M, Sturdy CB (2015) Individual differences in learning speed, performance accuracy and exploratory behaviour in black-capped chickadees. Animal Cognition 18:165-178

Guillette LM, Reddon AR, Hoeschele M, Sturdy CB (2011) Sometimes slower is better: slow-exploring birds are more sensitive to changes in a vocal discrimination task. Proc R Soc B-Biol Sci 278:767-773

Gyuris E, Fero O, Barta Z (2012) Personality traits across ontogeny in firebugs, Pyrrhocoris apterus. Animal Behaviour 84:103-109

H

Hall ML, van Asten T, Katsis AC, Dingemanse NJ, Magrath MJ, Mulder RA (2015) Animal personality and pace-of-life syndromes: do fast-exploring fairy-wrens die young. Frontiers in Ecology and Evolution 3

Halliday WD, Morris DW, Devito JA, Start DM (2014) Male and female voles do not differ in their assessments of predation risk. Ecoscience 21:61-68

Han CS, Dingemanse NJ (2015) Effect of diet on the structure of animal personality. Front Zool 12:1-9

Hedrick AV, Kortet R (2012) Sex differences in the repeatability of boldness over metamorphosis. Behavioral Ecology and Sociobiology 66:407-412

Herde A, Eccard JA (2013) Consistency in boldness, activity and exploration at different stages of life. Bmc Ecology 13

Hughes RN (1968) Behaviour of male and female rats with free choice of 2 environments differing in novelty. Animal Behaviour 16:92-&

I

ITO H, NARA H, INOUE-MURAYAMA M, SHIMADA MK, KOSHIMURA A, UEDA Y, KITAGAWA H, TAKEUCHI Y, MORI Y, MURAYAMA Y (2004) Allele frequency distribution of the canine dopamine receptor D4 gene exon III and I in 23 breeds. Journal of Veterinary Medical Science 66:815-820

K

Kanda LL, Louon L, Straley K (2012) Stability in Activity and Boldness Across Time and Context in Captive Siberian Dwarf Hamsters. Ethology 118:518-533

Kappeler P (2010) Animal behaviour: evolution and mechanisms. Springer Science & Business Media, Berlin

Karlsson Green K, Eroukhmanoff F, Harris S, Pettersson LB, Svensson EI (2016) Rapid changes in genetic architecture of behavioural syndromes following colonization of a novel environment. Journal of Evolutionary Biology 29:144-152

Kobler A, Engelen B, Knaepkens G, Eens M (2009) Temperament in bullheads: do laboratory and field explorative behaviour variables correlate? Naturwissenschaften 96:1229-1233

Koolhaas JM, Korte SM, De Boer SF, Van Der Vegt BJ, Van Reenen CG, Hopster H, De Jong IC, Ruis MAW, Blokhuis HJ (1999) Coping styles in animals: current status in behavior and stress-physiology. Neuroscience & Biobehavioral Reviews 23:925-935

Koski SE (2014) Broader horizons for animal personality research. Front Ecol Evol 2:1-6

Krebs JR, Sherry DF, Healy SD, Perry VH, Vaccarino AL (1989) Hippocampal specialization of food-storing birds. PNAS 86:1388-1392

Krystufek B, Kovacic D (1984) Distribution, habitat requirements and morphometric characteristics of Micromys minutus Pallas, 1771 (Rodentia, Mammalia) in Yugoslavia. Acta Biologica Iugoslavica Serija G Biosystematicka 10:99-112

Kubik J (1952) Micromys minutus Pall. in Bialowiecza National Park. Ann Univ Mariae Curie Skodowska Sect C Biol 7:449-495

L

Lande R, Arnold SJ (1983) The measurement of selection on correlated characters. Evolution:1210-1226

Lantová P, Šíchová K, Sedláček F, Lanta V (2011) Determining Behavioural Syndromes in Voles – The Effects of Social Environment. Ethology 117:124-132

Laskowski KL, Bell AM (2013) Competition avoidance drives individual differences in response to a changing food resource in sticklebacks. Ecology letters 16:746-753

Le Coeur C, Thibault M, Pisanu B, Thibault S, Chapuis J-L, Baudry E (2015) Temporally fluctuating selection on a personality trait in a wild rodent population. Behavioral Ecology 26:1285-1291

Le Galliard JF, Paquet M, Mugabo M (2015) An experimental test of density-dependent selection on temperament traits of activity, boldness and sociability. Journal of Evolutionary Biology 28:1144-1155

Lessells CM, Boag PT (1987) Unrepeatable repeatabilities - a common mistake. Auk 104:116-121

Lichtenstein JLL, Pruitt JN (2015) Similar patterns of frequency-dependent selection on animal personalities emerge in three species of social spiders. Journal of Evolutionary Biology 28:1248-1256

Lofdahl KL, Holliday M, Hirsch J (1992) Selection for conditionability in Drosophila melanogaster. Journal of Comparative Psychology 106:172

M

MacKay JR, Haskell MJ (2015) Consistent individual behavioral variation: the difference between temperament, personality and behavioral syndromes. Animals 5:455-478

Mamuneas D, Spence AJ, Manica A, King AJ (2015) Bolder stickleback fish make faster decisions, but they are not less accurate. Behavioral Ecology 26:91-96

Matzel LD, Han YR, Grossman H, Karnik MS, Patel D, Scott N, Specht SM, Gandhi CC (2003) Individual differences in the expression of a "general" learning ability in mice. Journal of Neuroscience 23:6423-6433

McDougall P, Réale D, Sol D, Reader S (2006) Wildlife conservation and animal temperament: causes and consequences of evolutionary change for captive, reintroduced, and wild populations. Animal Conservation 9:39-48

McPhee ME (2003) Generations in captivity increases behavioral variance: considerations for captive breeding and reintroduction programs. Biol Conserv 115:71-77

Mery F, Kawecki TJ (2002) Experimental evolution of learning ability in fruit flies. PNAS 99:14274-14279

Mery F, Kawecki TJ (2003) A fitness cost of learning ability in Drosophila melanogaster. Proceedings of the Royal Society of London B: Biological Sciences 270:2465-2469

Mesquita FO, Borcato FL, Huntingford FA (2015) Cue-based and algorithmic learning in common carp: A possible link to stress coping style. Behavioural Processes 115:25-29

Michelangeli M, Chapple DG, Wong BB (2016) Are behavioural syndromes sex specific? Personality in a widespread lizard species. Behavioral Ecology and Sociobiology 70:1911-1919

Mittelstaedt M-L, Mittelstaedt H (1980) Homing by path integration in a mammal. Naturwissenschaften 67:566-567

Modlmeier AP, Keiser CN, Wright CM, Lichtenstein JLL, Pruitt JN (2015) Integrating animal personality into insect population and community ecology. Current Opinion in Insect Science 9:77-85

Monestier C, Morellet N, Gaillard J-M, Cargnelutti B, Vanpe C, Hewison AJM (2015) Is a proactive mum a good mum? A mother's coping style influences early fawn survival in roe deer. Behavioral Ecology 26:1395-1403

Montgomery KC (1955) The relation between fear induced by novel stimulation and exploratory drive. Jour Comp and Physiol Psychol 48:254-260

Montiglio P-O, Garant D, Pelletier F, Reale D (2012) Personality differences are related to long-term stress reactivity in a population of wild eastern chipmunks, Tamias striatus. Animal Behaviour 84:1071-1079

Montiglio P-O, Garant D, Thomas D, Reale D (2010) Individual variation in temporal activity patterns in open-field tests. Animal Behaviour 80:905-912

Morand-Ferron J, Cole EF, Quinn JL (2015) Studying the evolutionary ecology of cognition in the wild: a review of practical and conceptual challenges. Biological Reviews

Morris RGM (1981) Spatial localization does not require the presence of local cues. Learning and Motivation 12:239-260

Morton FB, Lee PC, Buchanan-Smith HM (2013) Taking personality selection bias seriously in animal cognition research: a case study in capuchin monkeys (Sapajus apella). Animal Cognition 16:677-684

Mowles SL, Cotton PA, Briffa M (2012) Consistent crustaceans: the identification of stable behavioural syndromes in hermit crabs. Behavioral Ecology and Sociobiology 66:1087-1094

Müller T, Müller C (2015) Behavioural phenotypes over the lifetime of a holometabolous insect. Front Zool 12

N

Nakagawa S, Schielzeth H (2010) Repeatability for Gaussian and non-Gaussian data: a practical guide for biologists. Biological Reviews 85:935-956

Nicolaus M, Tinbergen JM, Ubels R, Both C, Dingemanse NJ (2016) Density fluctuations represent a key process maintaining personality variation in a wild passerine bird. Ecology Letters

Nussey DH, Wilson aJ, Brommer JE (2007) The evolutionary ecology of individual phenotypic plasticity in wild populations. Journal of Evolutionary Biology 20:831-844

O

Odling-Smee L, Braithwaite VA (2003) The influence of habitat stability on landmark use during spatial learning in the three-spined stickleback. Animal Behaviour 65:701-707

Okutsu K, Takatsuki S, Ishiwaka R (2012) Food composition of the harvest mouse (Micromys minutus) in a western suburb of Tokyo, Japan, with reference to frugivory and insectivory. Mammal Study 37:155-158

P

Padilla AVS (1999) Untersuchungen zur Öko-Ethologie der Zwergmaus Micromys minutus (Pallas 1778). Diss. thesis, Technischen Universität Carolo-Wilhelmina, Braunschweig

Petelle MB, Martin JGA, Blumstein DT (2015) Heritability and genetic correlations of personality traits in a wild population of yellow-bellied marmots (Marmota flaviventris). Journal of Evolutionary Biology 28:1840-1848

Petelle MB, McCoy DE, Alejandro V, Martin JGA, Blumstein DT (2013) Development of boldness and docility in yellow-bellied marmots. Animal Behaviour 86:1147-1154

Piechocki R (2001) The harvest mouse. Neue Brehm-Buecherei 222:3-126

Pinheiro JC, Bates DM (1996) Unconstrained parametrizations for variance-covariance matrices. Statistics and Computing 6:289-296

Plomin R (1999) Genetics and general cognitive ability. Nature 402:C25-C29

Plomin R, DeFries JC, Knopik VS, Neiderheiser J (2013) Behavioral genetics. Palgrave Macmillan

Poissant J, Reale D, Martin JGA, Festa-Bianchet M, Coltman DW (2013) A quantitative trait locus analysis of personality in wild bighorn sheep. Ecology and Evolution 3:474-481

Polverino G, Cigliano C, Nakayama S, Mehner T (2016) Emergence and development of personality over the ontogeny of fish in absence of environmental stress factors. Behavioral Ecology and Sociobiology:1-11

Pravosudov VV, Clayton NS (2002) A test of the adaptive specialization hypothesis: population differences in caching, memory, and the hippocampus in black-capped chickadees (Poecile atricapilla). Behavioral neuroscience 116:515

Q

Quinn JL (2015) Animal personality meets community ecology: founder species aggression and the dynamics of spider communities. Journal of Animal Ecology 84:1457-1460

Quinn JL, Patrick SC, Bouwhuis S, Wilkin TA, Sheldon BC (2009) Heterogeneous selection on a heritable temperament trait in a variable environment. Journal of Animal Ecology 78:1203-1215

R

R Core Team (2013) R: A language and environment for statistical computing. In. R Foundation for Statistical Computing, Vienna, Austria.

Reale D, Gallant BY, Leblanc M, Festa-Bianchet M (2000) Consistency of temperament in bighorn ewes and correlates with behaviour and life history. Animal Behaviour 60:589-597

Réale D, Martin J, Coltman D, Poissant J, FESTA-BIANCHET M (2009) Male personality, life-history strategies and reproductive success in a promiscuous mammal. Journal of evolutionary biology 22:1599-1607

Réale D, Reader SM, Sol D, McDougall PT, Dingemanse NJ (2007) Integrating animal temperament within ecology and evolution. Biological Reviews 82:291-318

Roff DA (1996) The Evolution of Genetic Correlations: An Analysis of Patterns. Evolution 50:1392-1403

Roff DA (1997) In: Evolutionary Quantitative Genetics. Springer, New York

Rowe C, Healy SD (2014) Measuring variation in cognition. Behavioral Ecology 25:1287-1292

S

Schuett W, Tregenza T, Dall SRX (2010) Sexual selection and animal personality. Biological Reviews 85:217-246

Shettleworth S (2010) Cognition, evolution, and behavior: Oxford University Press. New York, NY

Shettleworth SJ, Hampton RR (1998) Adaptive specializations of spatial cognition in food storing birds? Approaches to testing a comparative hypothesis. Animal cognition in nature:65-98

Sih A, Bell A, Johnson JC (2004a) Behavioral syndromes: an ecological and evolutionary overview. Trends in Ecology & Evolution 19:372-378

Sih A, Bell AM (2008) Chapter 5 Insights for Behavioral Ecology from Behavioral Syndromes. In. Academic Press, pp 227-281

Sih A, Bell AM, Johnson JC, Ziemba RE (2004b) Behavioral Syndromes: An Integrative Overview. The Quarterly Review of Biology 79:241-277

Sih A, Del Giudice M (2012) Linking behavioural syndromes and cognition: a behavioural ecology perspective. Philosophical Transactions of the Royal Society B-Biological Sciences 367:2762-2772

Small TW, Schoech SJ (2015) Sex differences in the long-term repeatability of the acute stress response in long-lived, free-living Florida scrub-jays (Aphelocoma coerulescens). Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology 185:119-133

Smith BR, Blumstein DT (2008) Fitness consequences of personality: a meta-analysis. Behavioral Ecology 19:448-455

Smith JM (1982) Evolution and the Theory of Games. Cambridge university press

Spitzenberger F (1986) Die Zwergmaus, Micromys minutus Pallas, 1771. Mammalia austriaca 12 (Mamm., Rodentia, Muridae). Mitteilungen der Abteilung fuer Zoologie am Landesmuseum Joanneum:23-40

Stamps J, Groothuis TGG (2010a) The development of animal personality: relevance, concepts and perspectives. Biological Reviews 85:301-325

Stamps JA (1991) Why evolutionary issues are reviving interest in proximate behavioral mechanisms. American Zoologist 31:338-348

Stamps JA, Groothuis TGG (2010b) Developmental perspectives on personality: implications for ecological and evolutionary studies of individual differencese. Philosophical Transactions of the Royal Society B-Biological Sciences 365:4029-4041

Stirling DG, Reale D, Roff DA (2002) Selection, structure and the heritability of behaviour. Journal of Evolutionary Biology 15:277-289

Strandberg E, Jacobsson J, Saetre P (2005) Direct genetic, maternal and litter effects on behaviour in German shepherd dogs in Sweden. Livestock Production Science 93:33-42

Stubley-Weatherly L, Harding JW, Wright JW (1996) Effects of discrete kainic acid-induced hippocampal lesions on spatial and contextual learning and memory in rats. Brain research 716:29-38

Sweeney K, Gadd RDH, Hess ZL, McDermott DR, MacDonald L, Cotter P, Armagost F, Chen JZ, Berning AW, DiRienzo N, Pruitt JN (2013) Assessing the Effects of Rearing Environment, Natural Selection, and Developmental Stage on the Emergence of a Behavioral Syndrome. Ethology 119:436-447

Sweis B, Veverka K, Dhillon E, Urban J, Lucas L (2013) Individual differences in the effects of chronic stress on memory: behavioral and neurochemical correlates of resiliency. Neuroscience 246:142-159

T

Thomas P, Herrel A, Hardy I, Aujard F, Pouydebat E (2016) Exploration Behavior and Morphology are Correlated in Captive Gray Mouse Lemurs (Microcebus murinus). International Journal of Primatology:1-11

Thornton A, Lukas D (2012) Individual variation in cognitive performance: developmental and evolutionary perspectives. Philosophical Transactions of the Royal Society B-Biological Sciences 367:2773-2783

Titulaer M, van Oers K, Naguib M (2012) Personality affects learning performance in difficult tasks in a sex-dependent way. Animal Behaviour 83:723-730

Trillmich F, Hudson R (2011) The emergence of personality in animals: the need for a developmental approach. Developmental psychobiology 53:505-509

Trompf L, Brown C (2014) Personality affects learning and trade-offs between private and social information in guppies, Poecilia reticulata. Animal Behaviour 88:99-106

Trout RC (1978a) Review if studies on captive harvest mice (Micromys minutus (Pallas)). Mammal Review 8:159-175

Trout RC (1978b) Review of studies on populations of wild harvest mice (Micromys minutus (Pallas)). Mammal Review 8:143-158

U

Uher J (2011) Individual behavioral phenotypes: An integrative meta-theoretical framework. Why “behavioral syndromes” are not analogs of “personality”. Developmental psychobiology 53:521-548

V

van Oers K, de Jong G, Drent PJ, van Noordwijk AJ (2004a) A genetic analysis of avian personality traits: Correlated, response to artificial selection. Behavior Genetics 34:611-619

van Oers K, de Jong G, van Noordwijk AJ, Kempenaers B, Drent PJ (2005) Contribution of genetics to the study of animal personalities: a review of case studies. In, pp 1185-1206

van Oers K, Drent PJ, de Goede P, van Noordwijk AJ (2004b) Realized heritability and repeatability of risk-taking behaviour in relation to avian personalities. Proc R Soc B-Biol Sci 271:65-73

van Oers K, Mueller JC (2010) Evolutionary genomics of animal personality. Philosophical Transactions of the Royal Society B: Biological Sciences 365:3991-4000

Van Oortmerssen G, Bakker TC (1981) Artificial selection for short and long attack latencies in wildMus musculus domesticus. Behavior genetics 11:115-126

Verbeek ME, Boon A, Drent PJ (1996) Exploration, aggressive behaviour and dominance in pair-wise confrontations of juvenile male great tits. Behaviour 133:945-963

Verbeek MEM, Drent PJ, Wiepkema PR (1994) Consistent individual differences in early exploratory behaviour of male great tits. Animal Behaviour 48:1113-1121

Vetter SG, Brandstätter C, Macheiner M, Suchentrunk F, Gerritsmann H, Bieber C (2016) Shy is sometimes better: personality and juvenile body mass affect adult reproductive success in wild boars, Sus scrofa. Animal Behaviour 115:193-205

W

Walker MD, Mason G (2011) Female C57BL/6 mice show consistent individual differences in spontaneous interaction with environmental enrichment that are predicted by neophobia. Behavioural brain research 224:207-212

Wallace DG, Hines DJ, Pellis SM, Whishaw IQ (2002) Vestibular information is required for dead reckoning in the rat. The Journal of neuroscience 22:10009-10017

Wang M-Y, Brennan CH, Lachlan RF, Chittka L (2015) Speed-accuracy trade-offs and individually consistent decision making by individuals and dyads of zebrafish in a colour discrimination task. Animal Behaviour 103:277-283

West MJ (1990) Stereological studies of the hippocampus: a comparison of the hippocampal subdivisions of diverse species including hedgehogs, laboratory rodents, wild mice and men. Progress in Brain Research 83:13-36

White NM, McDonald RJ (2002) Multiple parallel memory systems in the brain of the rat. Neurobiology of learning and memory 77:125-184

Wielebnowski NC (1999) Behavioral differences as predictors of breeding status in captive cheetahs. Zoo Biology 18:335-349

Wilson DS (1998) Adaptive individual differences within single populations. Philosophical Transactions of the Royal Society of London Series B-Biological Sciences 353:199-205

Wilson DS, Clark AB, Coleman K, Dearstyne T (1994) Shyness and boldness in humans and other animals. Trends in Ecology & Evolution 9:442-446

Wilson DS, Coleman K, Clark AB, Biederman L (1993) Shy-bold continuum in pumpkinseed sunfish (Lepomis gibbosus): An ecological study of a psychological trait. Journal of Comparative Psychology 107:250

Wolf M, van Doorn GS, Leimar O, Weissing FJ (2007) Life-history trade-offs favour the evolution of animal personalities. Nature 447:581-584

Wuerz Y, Krüger O (2015) Personality over ontogeny in zebra finches: long-term repeatable traits but unstable behavioural syndromes. Front Zool 12:1-14

Y

Yaskin VA (2011) Seasonal changes in hippocampus size and spatial behaviour in mammals and birds. Zhurnal Obshchei Biologii 72:27-39

Yuen C, Pillay N, Heinrichs M, Schoepf I, Schradin C (2016) Personality traits are consistent when measured in the field and in the laboratory in African striped mice (Rhabdomys pumilio). Behavioral Ecology and Sociobiology:1-12

Z

Zambra N, Gimeno D, Blache D, van Lier E (2015) Temperament and its heritability in Corriedale and Merino lambs. Animal 9:373-379

Zimmermann U (2014) Individual differences in the performance of harvest mice (Micromys minutus) in a spatial orientation task. Bachelor Thesis. Eberhard Karls Universiät Tübingen, Tübingen