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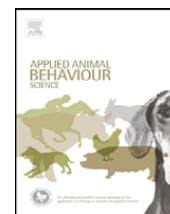
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## Genetic analysis of reactivity to humans in Goettingen minipigs

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## ABSTRACT

Goettingen minipigs are laboratory animals with an increasing demand over the last few years. At the moment, Goettingen minipigs are not selected for a low reactivity to humans and this trait is not included in the breeding programme. However, it is obvious that there is a need for genetically non-responding minipigs during handling to facilitate the treatment and restraint of the animals which is often needed in biomedical experiments. A first testing scheme was developed to evaluate the reactivity of Goettingen minipigs to humans and to analyse whether the trait reactivity to humans can be considered in the breeding programme. In this study temperament scores of this testing scheme for nine different traits from 10,033 animals collected from 2005 to 2008 were analysed. Temperament was subjectively scored on a scale from 1 to 5 while the pig is caught (C), held on the arms (A), standing in a box for weighing (W), standing on a table (T) and walking on the ground (G). The traits were a combination of these situations evaluated at three different ages (2, 4 and 6 months). Genetic parameters were estimated using bivariate models and different possible selection strategies were examined. Heritabilities were low to moderate with a range from 0.09 to 0.22 and phenotypic and genetic correlations between the nine traits were moderate to high with phenotypic correlations between 0.12 (W2 and G4) and 0.64 (W2 and A2) and genetic correlations between 0.44 (A4 and C6) and 1.00 (e.g. W2 and A4). It was shown that the highest genetic progress per year can be obtained when all nine traits are considered in the selection index. Under an economical point of view the selection on the basis of the two arm traits plus the trait W2 should be preferred.

Based on a critical discussion of the explanatory power of the used scoring system a new evaluation scheme was developed. In this scheme the minipigs can be divided into responding and non-responding animals whereas the latter are desired for selection. The suggested scoring system offers better possibilities for statistical analyses. It is planned to include the selection for non-responding Goettingen minipigs in the routine breeding programme.

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## 1. Introduction

Pigs are flight animals by nature. As prey animals they are very sensitive for threatening situations. Usually, even though there is a big variation, they react immediately with warning and flight to a disturbing situation or in non-

escapable situations also with aggression to thread the predator (Hemsworth, 2000). In general, there are two main reactions in a specific situation: Non-response means that the pig is not reacting on the handling. If there is a response we can observe two different behavioural strategies, also called coping styles (Benus et al., 1991). One strategy is active, the so-called fight and flight strategy. Another strategy is the passive strategy which means that the pig is immobile or freezing when it has to cope with a challenging situation. Different endocrine

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mechanisms underlie both strategies (Koolhaas and Bohus, 1989), but both enable the individual to maintain control over the threatening situation.

Non-responding farm and laboratory pigs are desired. A corporation between stockmen and pigs is much easier and handling is not dangerous for the stockperson when the pigs are not responding on humans in the sense of a negative reaction. Non-responding pigs are careful in the relationship to humans but neither aggressive nor fearful. They show a natural curious reaction.

Non-responding pigs can be obtained by close positive contact to humans (Bayne, 2002). The pigs learn that there is no need for a response while humans are present (Grandin and Deesing, 1998). To cope with humans was a needed characteristic for pigs in the process of domestication and the genetic performance plays an important role in this process (Price, 1999). Thus, another way to obtain non-responding pigs is to select animals that do not show any negative reaction at the presence of humans. Even though behaviour traits have only low to moderate heritabilities (McGlone et al., 1998) the selection of non-responding parents will by trend lead to non-responding offspring. Many studies showed the influence of breed effect which is again proof for a specific genetic control of behaviour (Dantzer and Mormede, 1978; Willham et al., 1964).

There are many ways to measure the reactivity of an animal to human handling. In pigs, mainly the human approach test is used (Brown et al., 2009; Scott et al., 2009). In this test the pig enters an arena in which a human is standing or sitting. The time the pig needs to approach the human is measured and a score is given based on the needed time. Additionally, the number of faecal excretion or different other information can be included in the test. Further, there are variations of the standard human approach test (Hemsworth et al., 1981).

For an improved relationship between humans and pigs in commercial pig farms and in breeding and laboratory facilities, it is favourable to use tests in which the human is approaching to the pig and not vice versa. In routine procedures the pigs are vaccinated, transported or sorted by the stockperson. Usually there is no time for the pigs to acclimatize to the handling. Thus, selecting non-responding pigs in ordinary and not in special testing situations is more reasonable.

To study and evaluate the genetic influence on reactivity to humans a well-established breed with complete pedigree information has to be used. Further, the more animals are tested the more accurate are the results of the genetic analyses of reactivity to humans. In this study, the test scores of Goettingen minipig breed were used for analysis.

The Goettingen minipig is a laboratory animal which is mainly used in biomedical research. Compared to laboratory mice, rats or dogs it has the advantage of being anatomically and physiologically very close to humans (Glodek et al., 1977). Therefore, the use of Goettingen minipigs for research in human medicine is increasing in the last few years. Currently, there are four breeding populations of Goettingen minipigs worldwide. Besides the base population in Germany there are two populations

in Denmark which are kept in one breeding centre and one population in the USA. As a laboratory animal the Goettingen minipig has very close contact to humans and is handled much more frequently than its relatives on fattening or breeding pig farms. This handling usually includes the separation from pen mates for the time of handling, quite often a fixation of the pig and a special treatment which normally requires a calm and not struggling minipig.

In the Danish minipig breeding populations a scheme for testing the pigs' reactivity to humans was developed in the past that allowed the collection of a big amount of data while other selection criteria were observed on the pig.

The objectives of this study were to analyse the collected behaviour data with regard to genetic background and practicability. It was assumed that the need for testing more or less all animals in the population for a well-founded genetic analysis may cause a lack of informational value of the tests. Further, special attention was given to the practicability of the actual testing scheme.

## 2. Material and methods

### 2.1. Animals

Goettingen minipigs of the two Danish populations were used in the study. This breed is exclusively used as laboratory animal in medical research. It was developed in the 1960s at the University of Goettingen and consists of 59% Vietnamese Potbelly Pig, 33% Minnesota minipig and 8% German Landrace pig to obtain a completely white minipig (Glodek et al., 1977). The idea was to combine the gentle temperament and body condition of the Minnesota minipig with the high fertility of the Vietnamese potbelly pig. Unfortunately Vietnamese potbelly pigs are very disadvantageous for the use in biomedical research in the sense of aggressive and vigorous behaviour (Bollen et al., 2000).

In Goettingen minipigs this disadvantageous behaviour can also be observed whereas in general this breed is more gentle than the Vietnamese breed. To be able to offer gentle and cooperating minipigs in the future the reactivity to humans was tested in different situations. With the collected data a breeding value estimation shall be conducted for the selection of non-responding Goettingen minipigs.

The minipigs are housed in two different units in one breeding centre. The breeding centre is located on Sealand Island, Denmark and is under genetic control of the University of Goettingen. Concerning management and housing the breeding centre is independently working.

The minipigs stay in the breeding centre until they are sold directly to the customer. Most of the minipigs are sold when they are 2–5 months old (Köhn et al., 2008). However, a small number of pigs stay in the breeding centre until they reach a certain stage of adolescence. Thus, there is a very unbalanced age structure in the breeding units. The minipigs have regular contact to humans due to routine handling procedures. They are ear tagged, notched and have a body check until they are weaned with 4 weeks of age. Between 4 and 8 weeks of age the piglets are gently

stroked regularly. This is done usually by the same staff member per unit by sitting in the pen and stroking the pigs when they are approaching. By stroking the piglets in the sensitive period after weaning the fear of humans should be reduced to prepare the pigs for later human contact in the laboratory facilities.

For the analyses in total 10,033 Goettingen minipigs were used (4681 male and 5352 female).

## 2.2. Testing scheme

The basic idea was to collect data of the reactivity to humans for the estimation of genetic parameters and breeding values. The aim was to include the breeding values for reactivity to humans in the total merit index and to select for non-responding and cooperating Goettingen minipigs. Thus, handling procedures in medical experiments should be facilitated. Currently, the total merit index consists of the traits body weight reduction and number of piglets born alive. The index is constructed as a restricted index where the trait body weight reduction is improved and the trait number of piglets born alive is held on a constant level because of a positive correlation between the two traits (i.e. improvement in body weight reduction causes less number of born piglets) (Köhn et al., 2008).

The used testing scheme in the breeding units of the two Danish populations was developed without any influence of the authors and also the data collection was conducted by the staff members themselves. As far as the University of Goettingen is responsible for the genetic management of the Danish populations the collected data was checked in this study for a possible inclusion of the “trait” reactivity to humans in the total merit index.

The data collection started in 2005 and was integrated in routine assessments of a pig's anatomy and morphology. In these routine assessments different criteria were examined on the pig, e.g. existence of black hairs or spots, number of teats, anomalies, shape of back and belly. The reactivity tests were conducted at three different ages with 2 months of age, 4 months of age and 6 months of age. These were the ages at which all minipigs in the populations were evaluated on the basis of anatomical and morphological characteristics. In combination with the breeding values for body weight and litter size the scores of the anatomical evaluation are the basis for the selection decision of breeding animals. A scoring system with a scale from 1 to 5 was used for the reactivity test. The explanation of the scores can be found in Table 1.

The reactivity to humans and to challenging situations was scored while the pig is caught (C), held on the arms (A), standing in a box for weighing (W), standing on a table (T) and walking on the ground (G). From the combination of

**Table 1**  
Explanation of the used scores.

Score	Explanation
1	Whole time struggling, screaming, aggressive
2	Mostly struggling and screaming
3	Partly struggling and screaming, partly gentle
4	Mostly gentle
5	Whole time relaxed, quiet, gentle

**Table 2**

Current scheme of the evaluation of the reactivity to human handling, values represent the number of minipigs tested in the different behavioural tests according at the different ages.

Situations	Age of the piglets (in month)		
	2	4	6
Catching (C)		1,559	1,173
Holding on arms (A)	9,166	1,510	
Weighing (W)	9,166		
Standing on table (T)		1,557	1,116
Walking on ground (G)		1,556	1,174

situation and age nine different tests were scored. The complete testing scheme and the number of scored minipigs per test is displayed in Table 2. In dependence on the anatomical evaluation of each minipig the reactivity to humans had to be easy and fast to score. Thus, no standardized test for each pig at different ages was developed. For the routine assessment of a pig's anatomy it had to be caught while a score was given for the reaction of this handling. Number of teats, number of black hairs and spots and check for anomalies was carried out while the pig was held in the arms. Again a score for the reaction was given. While the pig was standing on a table or walking on the ground anomalies in the legs and the complete body shape can be evaluated. Finally, the pig had to stand in a box for weighing. Again scores for these three situations were given. Even though the testing situations walking on the ground, standing on a table and weighing do not reflect only the reactivity to humans, but also the reactivity to a novel environment and thus are typical situations involving close human contact we included them in the analyses. We were able to make a complete analysis over all collected data that were referred to behavioural responses.

The tests were conducted always by the same specially trained staff members in each unit which were familiar to the pigs.

A main problem that occurred during analyses was the high number of missing scores in the data set. The highest proportion of minipigs is sold to the customers when they are still young and these animals are no longer available to be scored in older ages. Most of the animals in the data set (91.36%) were scored for reactivity to humans when they were 2 months old (W2, A2,) but only 15.05% were scored when they were 4 months old (C4, T4, A4, G4) and 11.12% when they were 6 months old (C6, T6, G6). Only 6.74% of the animals had scores for all nine tests (Table 3).

## 2.3. Genetic analyses

Genetic parameters of the collected scores from the nine tests were estimated using VCE-5 (Kovac et al., 2002). Estimation was carried out with bivariate linear models for all possible combinations of the 9 tests, e.g. the first model included the scores of tests A2 and W2, the second model the scores of tests A2 and A4 and so on. For each test 8 models were calculated and thus, 8 different heritability estimates resulted from the analyses. These heritability estimates were averaged for each test afterwards. At the same time phenotypic and genetic correlations were

**Table 3**

Mean responses (and standard deviations) of the minipigs in different behavioural test (A = holding in the arm, W = weighing, T = standing on a table, C = catching, G = walking on the ground) at the age of 2, 4, and 6 months.

	Trait								
	A2	W2	A4	T4	C4	G4	T6	C6	G6
Mean	3.37	3.34	3.25	3.08	3.02	3.14	3.22	3.08	3.18
Standard deviation	0.74	0.66	0.63	0.72	0.64	0.60	0.70	0.71	0.60
n records	9,166	9,166	1,510	1,557	1,559	1,556	1,116	1,173	1,174

obtained from the bivariate analyses. We chose bivariate models for estimation to obtain uncorrected pair wise correlations for each combination of two tests.

The bivariate animal model was as follows:

$$y_{ijklmno} = S_i + U_j + BM_k + BY_l + l_m + a_n + e_{ijklmno},$$

where  $y_{ijklmno}$  is the reactivity score of trait  $o$  of animal  $n$  within sex  $i$ , unit  $j$ , birth month  $k$ , birth year  $l$  and litter  $m$ ,  $S_i$  is the fixed effect of sex,  $U_j$  is the fixed effect of unit,  $BM_k$  is the fixed effect of birth month,  $BY_l$  is the fixed effect of birth year,  $l_m$  is the random effect of common litter environment,  $a_n$  is the random animal effect, and  $e_{ijklmno}$  is the random measurement error. For the random effects the distributions for  $a \sim N(0, \mathbf{A}\sigma_a^2)$ ,  $l \sim N(0, \mathbf{I}\sigma_l^2)$  and  $e \sim N(0, \mathbf{I}\sigma_e^2)$  were assumed.

Standard errors for heritabilities and genetic correlations are provided to facilitate the interpretation of the results. In the standard error the number of records on which the particular estimate is based is considered.

Using the results from the estimation of genetic parameters, different selection scenarios were considered and the genetic progress for the selected tests of concern was estimated based on information from the animal only. Four selection scenarios were used that differed in considered tests in the selection index and the breeding goal. After estimation the best selection scenario with regard to practicability and genetic progress can be selected for the later use in the breeding routine.

The four applied scenarios are displayed in Table 4. In scenario 1 (S1) all nine tests were in the breeding goal and all nine tests were considered in the selection index. In scenario 2 (S2) only the tests W2, A2 and A4 were

considered for selection but again for all nine tests a genetic progress was calculated. In scenario 3 (S3) the tests A2 and A4 were considered in the breeding goal and scores from the tests W2, A2 and A4 were used for selection whereas in scenario 4 (S4) only scores from the tests A2 and A4 were used for both breeding goal and selection basis. The tests A2 and A4 were chosen as main tests for selection basis in scenarios S2, S3 and S4 because holding a pig on the arms is a typical situation in medical research and it is therefore important that the pig is particularly in this situation calm and relaxed. The trait W2 was included in scenario 2 and 3 to obtain a better accuracy due to a big amount of data for 2-month-old pigs. For the calculation of the genetic progress per year a generation interval for male minipigs of 1.674 and for female minipigs of 2.124 and a selection intensity of 2.2 for male and of 1.7 for female minipigs was assumed (Köhn et al., 2008). Finally, the estimated genetic progresses were weighted with the proportion of available records in each age, i.e. estimates of the tests W2 and A2 were multiplied with 0.91, estimates of tests A4, T4, C4 and G4 were multiplied with 0.15 and estimates of tests T6, C6 and G6 were multiplied with 0.11. With the weighting we were able to account for the different amount of data in each evaluation age.

### 3. Results

A big amount of Goettingen minipigs was scored for reactivity to humans and handling. Especially in the tests A2, A4, C4 and C6 the reactivity to humans can be expressed the best because the pig has direct contact with the human and is restrained. Thus, the given scores have a high impact for further interpretation. Regarding the scoring scale most of the pigs had score 3 (55.76%) or 4 (30.56%). The lowest mean score was calculated for both catching tests (3.02 for C4 and 3.08 for C6) and for the test T4 (3.08). The highest scores were evaluated for the tests conducted with 2 months of age (3.37 for W2 and 3.34 for A2).

Estimated heritabilities were in a range from 0.09 for C6 to 0.22 for T4 (Table 5). The estimated correlations showed a high discrepancy between phenotypic and genetic correlations (Table 5). Phenotypic correlations were low to moderate with values from 0.10 (correlation between A2 and C4) to 0.64 (correlation between W2 and A2). Genetic correlations were in general higher with values in a range from 0.44 (correlation between A4 and C6) to 1.00 (different pairs of traits). From the high genetic correlations it can be seen that most of the tests induce reactions that are under the same genetic control.

**Table 4**

Selection scenarios studied, tests used in the selection index and as breeding goal are marked.

Scenario	Traits								
	W2	A2	A4	T4	C4	G4	T6	C6	G6
S1									
Selection index	x	x	x	x	x	x	x	x	x
Breeding goal	x	x	x	x	x	x	x	x	x
S2									
Selection index	x	x	x						
Breeding goal	x	x	x	x	x	x	x	x	x
S3									
Selection index	x	x	x						
Breeding goal		x	x						
S4									
Selection index		x	x						
Breeding goal		x	x						



Table 5

Heritabilities (diagonal), genetic (above diagonal) and phenotypic correlations (below diagonal) with standard errors in parenthesis.

Trait	W2	A2	A4	T4	C4	G4	T6	C6	G6
W2	<b>0.147 (0.04)</b>	0.900 (0.03)	1.000 (0.00)	0.911 (0.05)	0.801 (0.11)	0.897 (0.13)	1.000 (0.00)	1.000 (0.00)	1.000 (0.01)
A2	0.642	<b>0.140 (0.04)</b>	1.000 (0.00)	0.735 (0.11)	0.740 (0.13)	0.672 (0.17)	1.000 (0.00)	1.000 (0.04)	0.988 (0.26)
A4	0.166	0.133	<b>0.120 (0.04)</b>	0.814 (0.07)	0.684 (0.07)	0.762 (0.13)	0.913 (0.16)	0.444 (0.30)	0.473 (0.17)
T4	0.337	0.166	0.480	<b>0.216 (0.04)</b>	0.878 (0.07)	1.000 (0.00)	0.655 (0.15)	0.817 (0.2)	0.840 (0.14)
C4	0.172	0.102	0.498	0.449	<b>0.173 (0.04)</b>	1.000 (0.02)	1.000 (0.01)	1.000 (0.00)	0.478 (0.15)
G4	0.117	0.106	0.445	0.461	0.487	<b>0.105 (0.05)</b>	1.000 (0.00)	0.821 (0.29)	0.765 (0.19)
T6	0.395	0.247	0.304	0.581	0.308	0.277	<b>0.109 (0.05)</b>	0.999 (0.04)	1.000 (0.00)
C6	0.186	0.144	0.309	0.315	0.490	0.251	0.462	<b>0.089 (0.04)</b>	0.560 (0.17)
G6	0.119	0.124	0.331	0.390	0.378	0.365	0.507	0.525	<b>0.132 (0.04)</b>

The standard errors for heritabilities (Table 5) were in an acceptable range (0.04–0.05) but a bit higher than expected. The standard errors for genetic correlations were low for estimates based on a high number of scored animals. As it was expected they increased with decreasing number of scored animals in the particular tests considered for the correlation with a highest value of 0.30 (correlation between A4 and C6). Standard errors for phenotypic correlations are not provided by VCE-5.

The estimates for the random litter effect were highest for the tests scored with 2 months of age (W2 and A2) with values of around 0.16 as expected (Fig. 1). For the other tests lower values were estimated with a range from 0.034 (T4) to 0.073 (A4). The test G4 was an exception with a similar value (0.16) as estimated for the 2 months-tests.

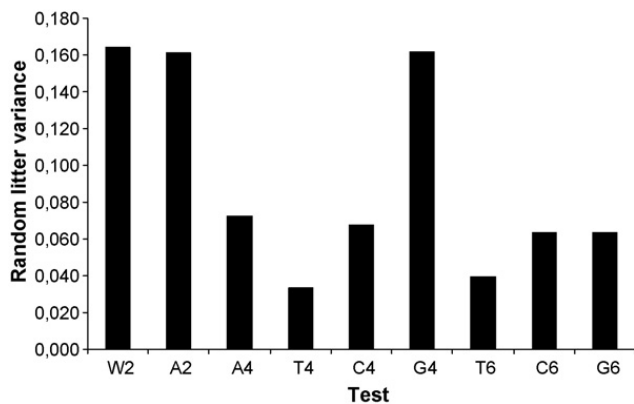


Fig. 1. Random litter variance for the nine different tests.

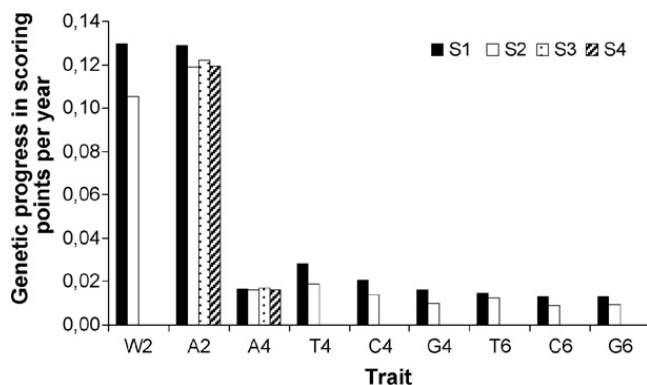


Fig. 2. Expected genetic improvement of the reactivity to humans calculated from four different selection strategies.

The examination of four different selection scenarios showed different genetic progresses (Fig. 2). The selection on the basis of all tests (S1) resulted in a significantly ( $p < 0.001$ ) higher genetic progress per year of in average 0.008 score points for all nine tests compared to S2. Only considering the genetic progress for the tests A2 and A4 no significant differences were found between the selection scenarios. However, some tendencies were observed. For S3 the genetic progress for A2 and A4 is slightly lower compared to S1 but higher compared to S2. When using A2 and A4 both as breeding goal and as tests in the selection index (S4) more or less the same genetic progress was calculated for A2 and A4 compared to the selection based on W2, A2 and A4 but with all tests in the index (S2). These results implicate that a selection on the basis of A2 and A4 is not favourable when the arm tests are in the breeding goal but that an additional inclusion of the test W2 in the selection index increases the genetic progress for the arm tests. Further, the high expected genetic progresses for the tests evaluated with 2 months (W2 and A2) displays the importance of the tests that are scored early in a pig's life.

#### 4. Discussion

Since 2005 scores for the reactivity to humans from Goettingen minipigs of the Danish populations were collected. For a planned inclusion of this behavioural trait into the current breeding programme the collected data had to be analysed and the phenotypic and genetic characteristics had to be examined.

The heritability estimates (Table 5) for the nine tests were in a normal range for behavioural traits (Grandin and Deesing, 1998; Hemsworth et al., 1990) whereas no direct comparison to estimates of other studies can be made because standardized behaviour tests were used by other authors (e.g. novel object test, human approach test) while a specific evaluation of reactivity to humans was done in our study. Heritabilities were moderate to low but would enable a selection of minipigs with a desired reactivity to humans. The standard errors of heritability estimates were a bit higher than expected. These values implicate the problem of missing data and the resulting low number of records per trait that was used for estimation. However, the analysed number of records was sufficient for reliable heritability estimates for all tests.

Due to the fact that phenotypic correlations consist of the genetic correlation and an environmental correlation the lower phenotypic correlations compared to the genetic

correlations (Table 5) indicate that environmental factors substantially influence the reaction on a handling situation at different ages and situations (Hazel, 1943). These environmental factors can be a different day-time at which the pigs are scored or they can come out of different groups at each scoring which also has an influence of the rank of the animal in the group and therefore on the level of aggression (Bolhuis et al., 2005). The high genetic correlations show that the reactivity to humans is more or less consistent for each pig for all tests and that there is no need to conduct all tests to have a valuable score for the behavioural expression “reactivity to humans”.

We assumed the scores of the two arm tests (A2, A4) to be most relevant for a selection decision. In these tests the minipigs are fixed by a human and are additionally examined for anatomical disorders by a second person. The restraint is much more intensive than in the short time of catching or when no direct human contact exists (weighing, standing on a table, walking on the ground). Further, a sufficient data basis is important for reliable results that exists for the arm test conducted at 2 months of age (A2). Thus, we focused on the two arm tests for an interpretation of the results of the selection scenarios. A genetic progress of 0.12 scoring points per year for test A2 and 0.01 scoring points per year for test A4 can be achieved when selection is only focusing on reactivity to humans, disregarding the already implemented selection traits number of piglets born alive and body weight reduction. There is a high discrepancy in these results due to the weighting by the available number of records for the particular test. An inclusion of the test A4 in the selection scheme does not seem to be useful if the number of available records cannot be increased.

In our study we analysed how the nine conducted tests interact and how they can be included in one selection index. That is the reason for the consideration of different selection scenarios as was already recommended by Hazel (1943).

For the planned development of a new total merit index with the combination of the traits body weight reduction, number of piglets born alive and positive reactivity to humans, many aspects have to be considered. It is known that selection for one trait will always influence other traits, either positive or negative (Hazel, 1943). Thus, each trait that has to be improved should be selected carefully and all effects on other considered traits should be known. In this study we analysed the nine tests but not the interaction of the reactivity to humans to other traits. In a study of Hemsworth et al. (1989) a reduction of levels of fear of humans improved the reproduction performance of sows whereas no significant influence of the fear level on growth performance of male pigs was found by Pearce et al. (1989). Further, the economic aspect has to be taken into account. If the collection of data is inefficient compared to the expected benefit an inclusion of this trait in the planned selection scheme should be reconsidered.

As it was shown in Table 2 there was a high number of missing values especially for the tests at higher age that complicated the analyses. Further, the given scores for the nine testing situations showed that the scale from 1 to 5

was not sufficiently used and most of the pigs had a score of 3 or 4 (Table 3). This indicates that immobile/freezing minipigs are scored as gentle with a score of 3 or higher.

Regarding all results (genetic and phenotypic correlations, heritabilities and genetic progress for the different tests under different selection strategies) we conclude the following aspects.

The nine different tests for reactivity to humans interact genetically and to a lower extent also phenotypically. It is therefore not necessary to score the minipigs' reactivity to humans in all tests. A focus on tests with a high number of scored pigs and a high relevance for the expression of reactivity to humans is sufficient. The estimated heritabilities enable a selection for low reactivity to humans.

Regarding the structure of the data and after observing the evaluation routine in the breeding centre we hypothesize that the current testing system is very labour and time consuming. The testing situations walking on the ground, standing on a table or weighing are no good indicators for the reactivity to humans. They would offer good results if the minipig had enough time to voluntarily contact the handler (who is usually standing next to the pig) and if the time until first contact was recorded as in the human approach test. Unfortunately, the scores in these testing situations are only by-products of the anatomical evaluation of the minipigs that is already very time-consuming. Further, these testing situations are more related to reactivity to a novel environment (weighing box, table, corridor) than to humans. However, these tests were included in this study to evaluate the current testing scheme and to better analyse the genetic background of behavioural responses in Goettingen minipigs.

A study of Tanida and Nagano (1998) showed that minipigs are able to discriminate between different handlers. It is therefore important that an unfamiliar person is handling the pigs in the tests as is done in the current testing scheme. Reactivity to humans that were feeding or stroking the pigs in the past is not reflecting the desired behavioural response.

We assume that a clear discrimination between responding and non-responding minipigs is not possible with the current testing scheme. The biggest problem is the exact differentiation of an immobile (responding) minipig and a relaxed (non-responding) minipig.

The consequence of our conclusions and hypotheses is the proposal of a new evaluation scheme for the assessment of reactivity to humans in Goettingen minipigs.

As far as the reactivity to humans is one of the most important behavioural traits for the customers of laboratory pigs, a testing situation has to be developed that reflects a possible handling situation in later experiments. Further, one test at one standardized age is sufficient to score a pig's reactivity to humans. Janczak et al. (2003) studied the behaviour of female pigs between 8 and 24 weeks of age in different test situations, e.g. the response to humans. They found positive correlations between measurements at different ages. These results support the theory that fear and anxiety are specific personality traits in pigs and that these traits are not changing basically over different ages. Studies from Erp-v. d. Kooij et al. (2002) and

**Table 6**

Possible new evaluation scale, score 3 = most of the time relaxed/freezing/struggling, score 2 = partly, score 1 = almost not.

	Reaction		
	Relaxed	Freezing	Struggling
Score	3	3	3
	2	2	2
	1	1	1

Hessing et al. (1993) confirm that there is a certain consistency in behaviour over time. Hessing et al. (1993) discovered a high persistency in the reactions of the 218 piglets in the backtest and concluded that each individual has its own strategy to cope with a challenging situation and that these strategies are based on different endocrine mechanisms (Koolhaas and Bohus, 1989).

We propose to include the new test for reactivity to humans in standardized situations like the ear tagging or notching that are conducted before weaning. We ensure with this testing situation that all piglets are still available for testing and thus, the number of missing values is extremely reduced compared to the current testing scheme. To avoid that pain-related reactions influence or even mask the reactivity to humans the scores should be recorded before the ear tagging or notching procedure and should be given for the reactivity from catching up to fixation for the following procedure.

For an easy and meaningful analysis of the new test scores a different scoring system with a clear differentiation of responding and non-responding minipigs is proposed. To avoid statistical artefacts threshold models (Gianola and Foulley, 1983) or the Bayesian approach could be used for analysis (Gianola and Fernando, 1986).

We propose a scoring scale with a range from 3 to 1 that will be used for three different possible reactions to overcome the problem that all calm animals will be classified the same. A possible scale is shown in Table 6. To find the right scoring system needs more scientific studies. It is very unlikely that the three categories relaxed, freezing and struggling we plan for the scoring system are sufficient to reflect the individual responses to a challenging situation. As it was written by Spooler et al. (1996) the behaviour of pigs is consistent but very individual and thus it is almost not possible to generate as many categories as individual reactions.

Additionally to the score it could be marked in the evaluation form if the pig was vocalising or not. In a backtest study with 219 pigs carried out by Erhard et al. (1999) a positive correlation was found between freezing and struggling pigs and their vocalisation.

The Goettingen minipig as a laboratory animal is not only a good animal model for human medicine. The tested reactivity to humans is also very important in commercial pig farms with fattening and breeding pigs. As many studies proof, aggressive and fearful pigs result in numerous problems for the stockperson and even the performance of the pigs can suffer (Hemsworth et al., 1989; Paterson and Pearce, 1992; Pearce et al., 1989). Due to the fact that the reactivity to humans is planned to be included in the total merit index for Goettingen minipigs, a

huge number of piglets is tested regularly and valuable estimates for genetic parameters can be used as a guide for human responsiveness in other breeds.

For future research, genome-wide scans for detecting QTL that have effects on the expression of reactivity to humans in Goettingen minipigs can be implemented (Reiner et al., in press). With this knowledge powerful breeding programmes on the basis of genomic selection might be established.

## 5. Conclusion

Heritability estimates based on data from the current testing scheme for reactivity to humans in Goettingen minipigs enable an inclusion in the breeding programme. However, the development of a new testing scheme is proposed to overcome different problems in the analyses and interpretation.

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