

# Qualitative aspects of grunts vary with pigs' mental states

# *Isabelle Veissier* based on peer reviews by *Matteo Chincarini* <sup>(D)</sup> and 1 anonymous reviewer

Avelyne S Villain, Carole Guérin, Céline Tallet (2023) The use of pigs vocalisation structure to assess the quality of human-pig relationship. bioRxiv, ver. 5, peer-reviewed and recommended by Peer Community in Animal Science.

https://doi.org/10.1101/2022.03.15.484457

Submitted: 23 March 2022, Recommended: 24 March 2023

# Cite this recommendation as:

Veissier, I. (2023) Qualitative aspects of grunts vary with pigs' mental states. *Peer Community in Animal Science*, 100190. 10.24072/pci.animsci.100190

Published: 24 March 2023

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Villain et al., (2023) investigated the structure of vocalisations in piglets in relation to human-animalrelationship. They first established a positive relationship by habituating piglets to be positively handled at weaning or later on after weaning. They then compared the reactions of piglets previously positively handled at weaning to that of non-handled piglets during tests in presence of a human (interacting or not), and also before and after the conditioning period when all piglets received positive contacts. They showed that the duration and frequency of grunts emitted in the presence of the human depends on previous contacts. More specifically, grunts are shorter and higher pitched in pigs that have been positively handled, in line with a positive human-animal relationship which is also observed through proximity of the piglets with the human. The authors concluded that the structure of pig vocalisation can reflect the quality of their relationship with humans.

The authors also showed that not only the response to humans is modified by positive contacts but also the general mood of piglets, with piglets positively handled at weaning emitting shorter grunts than non-handled piglets, whatever the context.

Another interesting finding is the temporality of behaviour of pigs habituated to positive contacts: during the first tests, they stay close to the human, probably being reassured by the proximity of the human. Then, when tests are repeated, they explore more the test room, using the human as an exploratory basis as already reported in the literature.

The hypotheses of the study are clear. The methods are reported in details so that the work is reproducible. The interpretation of results is sound. The manuscript is clearly written. This paper brings new and original knowledge in the field of animals' emotional responses and humananimal relationship: on the structure of grunts in relation to positive affects (positive emotion, positive mood) and on the temporality of the responses to human presence.

I recommend this manuscript for its originality and quality.

Isabelle Veissier

Villain, A.S., Guérin, C., Tallet, C., 2023. The use of pigs vocalisation structure to assess the quality of humanpig relationship. bioRxiv 2022.03.15.484457, ver. 5 peer-reviewed and recommended by Peer Community in Animal Science. https://doi.org/10.1101/2022.03.15.484457

# Reviews

# **Evaluation round #5**

DOI or URL of the preprint: https://doi.org/10.1101/2022.03.15.484457 Version of the preprint: 4

# Authors' reply, 24 March 2023

Dear journal, Please find attached the v5 of our manuscript, after removing Camille Noûs as author.

Download tracked changes file

# Decision by Isabelle Veissier, posted 21 March 2023, validated 21 March 2023

# Request to remove Camille Noûs as author of your article as a condition for the manuscript being recommended by PCI Animal Science

Dear authors,

The recommender handling the evaluation of your manuscript has decided to recommend your paper. You will find enclosed a letter explaining her decision. However, the managing board expressed concerns about listing Camille Noûs as author of your article. The managing board does not agree about the inclusion of a fictive author in papers submitted to PCI Animal Science. We recognize our mistake in overlooking this issue at the moment of your submission and we apologize for our late decision on this matter.

The managing board requests to have the fictive author being removed as a condition for the manuscript being recommended by PCI Animal Science. If you agree, you should upload a new version in bioRxiv without the name of Camille Noûs.

Best regards

Rafael Muñoz-Tamayo for the managing board of PCI Animal Science **Download recommender's annota-tions** 

# **Evaluation round #4**

DOI or URL of the preprint: https://www.biorxiv.org/content/10.1101/2022.03.15.484457v3 Version of the preprint: 3

# Authors' reply, 05 March 2023

Please find attached the letter to the recommender and the tracked hanges version of the manuscript.

# Download author's reply Download tracked changes file

# Decision by Isabelle Veissier, posted 02 January 2023, validated 05 January 2023

### **Moderate revisions**

I thank you for following my recommendations, especially for reorganising the discussion. The discussion is much clearer now. It is still a bit tortuous and not fully convincing. You conclude both that the HAR is more positive in handled piglets and is increased with conditioning and that the short and high-pitched grunts are indicative of a positive emotion. So there should be more of these grunts at the end of conditioning, which does not seem to be the case. There must be a more elegant interpretation. If I summarise the results:

Behaviour: during the tests, the piglets positively handled stay closer to the human. The same is obtained in piglets not positively handled after they receive the conditioning, so that you can conclude that the same level of HAR is obtained after conditioning in all piglets. Also, the piglets positively handled explore more and are more mobile during the tests (both before and after the conditioning).

Grunts:

- During isolation phase: longer and lower pitch grunts

- Positively handled piglets have shorter and higher pitched grunts compared to non-positively handled ones, whatever the situation à more related to mood than to the specific context of the test

- Piglets emit shorter and higher pitched grunts when in proximity to humans, especially when the human is not familiar (e.g. the difference is more marked in non-handled piglets and – in all piglets- more marked before conditioning)

The behaviour of piglets should help you to interpret these variations in grunts:

- What difference in behaviour do you see in absence vs presence of the human? Such results are not reported. They could help you better relate grunts and behaviour

- The strong parallel I see between behaviour and grunts in your study is about exploration: Handled pigs both explore more the test pen and emit shorter and higher pitched grunts. These two seem to go together. It suggests that short and high-pitched grunts have something to do with exploration (and vice-versa). And probably the handled piglets feel more confident (thanks to a positive HAR) and explore more.

- The fact that short and high-pitched grunts are expressed preferably in proximity to human especially when it is not very familiar goes in the same direction: we can think that the human is more explored when not familiar

My feeling is that the short and high-pitched grunts are related to "positive" exploration (as opposed to "negative" exploration = looking for escape).

You may not agree with such an interpretation. I may have misunderstood your results (I got a bit lost I must admit). Also, like Reviewer 2, I am frustrated by the fact that your analyses are essentially on the quality of grunts and not their quantity. I would like you to consider the above reasoning and try to make your interpretations more convincing.

I also have minor comments on the writing. I include them on your manuscript (file attached).

All the best Download recommender's annotations

# **Evaluation round #3**

DOI or URL of the preprint: https://www.biorxiv.org/content/10.1101/2022.03.15.484457v2 Version of the preprint: V2

# Authors' reply, 16 December 2022

Dear recommender, please find attached:

- the response letter to reviewer and recommender

- the revised version of the manuscript with tracked changes.

Download author's reply Download tracked changes file

# Decision by Isabelle Veissier, posted 15 September 2022

Dear authors,

I thank you for taking the comments from reviewers into account and for providing detailed explanations on how you handle them. There are still some comments that require further attention. See attached your response where I inserted my suggestions in red text in red to further improve the manuscript. There are still few points that I would like to double check with Rev 2. I invite you to first take my suggestions into account before I liaise with Rev 2.

I thank you in advance. Download recommender's annotations

# **Evaluation round #2**

DOI or URL of the preprint: https://www.biorxiv.org/content/10.1101/2022.03.15.484457v2 Version of the preprint: V2

# Authors' reply, 22 July 2022

Please find attached a PDF with the letter to the reviewers followed by the track changes version of the manuscript.

Thank you for your patience. Download tracked changes file

# **Evaluation round #1**

DOI or URL of the preprint: https://www.biorxiv.org/content/10.1101/2022.03.15.484457v1.fu ll.pdf

# Authors' reply, 16 July 2022

Download author's reply

# Decision by Isabelle Veissier, posted 07 May 2022

# **Moderate revisions**

This manuscript is of high interest. It however deserves amendments before we can recommend it. The two reviewers made detailed and complementary comments that should help the authors.

The writing needs to be improved to help the reading. The English needs also editing by a native english speaker.

# Reviewed by Matteo Chincarini <sup>(b)</sup>, 06 April 2022

#### Dear Editor,

The manuscript "The use of pigs vocalisation structure to assess the quality of human-pig relationship" is addressing an original work on interspecific acoustic communication and explores the non-invasive emotional indicators in swine. The study design is very accurate, and the discussion of the results gives an exhaustive overview of the topic. A couple of concerns could be related to my misunderstanding. The first one regards the pen size where the experiment has been conducted and the second one is on the assumption test for PCA (please, see below). The experiment is well described and even if, working with farm animals and acoustic analysis is very challenging, the authors have worked very hard to set up elegant research. Furthermore, there is growing interest in vocal communication related to animal welfare as well as human-animal relationships. Finally, these results provide potential non-invasive indicators relevant to animal welfare. The manuscript needs also some minor revisions relative to the figures.

Below are some specific comments to the authors for minor revisions:

### Title

The title clearly reflects the content of the article.

# Abstract

The abstract is concise and presents the main findings of the study. I've only one concern regarding the first sentence: "In domestic species, studying human-animal interactions and their consequences on the establishment of a positive Human-Animal Relationship (HAR) would have applications for both improving animal welfare." I'm not a native English speaker but here maybe it is possible to rephrase. I understand that authors are referring to animals and humans when they use the term "both" but now it seems it is referring to "interactions" and "consequences".

## Introduction

Hypotheses have been explicit very clearly and they are supported by several papers representing the state of the art in this field.

LL 46-47: please consider merging these two very short sentences.

# Materials and methods

This section is, in general, well explained and detailed.

# **Ethical note**

L 103: please, consider citing the French and European legislation (this will be relevant for the pen size, see below)

# Subject and housing conditions

L 112: please, verify the pen size according to your authorisation. According to the EU Directive 63/2010, the minimum enclosure size should be 2,0 m2 (Table 7.3). This could be not your case but it needs to be justified (it can be smaller due to experimental grounds).

#### Conditioning

L 137: you say that conditioning took place between day 42 and 62 after weaning, so it would be between 70 and 90 days of life? At L 107 it is reported that piglets involved in the study were from 28 to 62 days of life, please double check it.

L 145: I'm not sure what "Hens" means here

# Behavioural monitoring and analysis

Please, specify if the behaviour has been analysed either by the same or different persons.

#### Acoustic monitoring and analysis

L 178: Even if Praat is well-known software in this field, I think it would be better to cite it using a reference. Especially to be clear about the version that has been used (please, you can give a look here: https://www.fon.hum.uva.nl/praat/manual/FAQ\_How\_to\_cite\_Praat.html)

# **Statistical analysis**

I am not a statistician. However, when applying PCA I think it is worth reporting the value of Kaiser-Meyer Olkin (KMO) and Bartlett's test of sphericity as preliminary tests (or explain why not). Maybe, you could also consider using the Measure of Sampling Adequacy (MSA) if some variable needs to be excluded.

From Table 4, I'm understanding that you transformed some variables (like using log or sqrt). Please, consider adding this information also in the text when you write about symmetrical distribution L 206.

L 207: "pca", did you mean function "dudi.pca"?

L 254: inside the code, I think ID/time/Phase should be ID/Time/Phase

# Results

General: most of the script has been reported, could you add also the PCA analysis?

L 344: figure 3 is not present in the manuscript

L 369: figure 4, I'm not sure that is referring to the actual figure

# **Tables and figures**

Please, see above

# Discussion

The discussion is exhaustive and well supported by the literature. Still, the conclusions are not overstated. L 475: there are two "first"

# References

Fine.

# Reviewed by anonymous reviewer 1, 04 May 2022

Review of "The use of pigs vocalisation structure to assess the quality of human-pig relationship" by Vilain et al.

This is an interesting study aiming to investigate if (changes in) pig vocalisation reflects the quality of humananimal relationships. I have, however, some major concerns and a few other issues, which I have summarised below.

### Major concerns:

The study sets out to analyse pig vocalisation in different situations, comparing changes in sound structure both within and between pigs when treated differently over time. However, the data are immediately reduced to only grunts (line 178) because they were the most frequent. Is frequency important for all calls? One scream may say more than a thousand grunts, to paraphrase Ibsen. I would like to see a couple of phrases explaining this a bit more, as this is an important aspect of your data editing. You also focus on vocal quality, but what about vocal quantity? You mention vocal activity in line97, but refer to some qualitative aspects there, too.

I am missing which specific hypotheses you are testing? Or rather, in the Discussion, you dismiss one of the hypotheses, and then appear to suggest two new ones (lines 539-544).

Was the vocalisation when conspecifics were social partners (line 141) used at all? I may have missed where that is presented. And if not, why was this included? And if used, how did you separate the vocalisation of the different pigs?

The analyses are complex and can be difficult to follow in places. Is a p-value threshold of 0.05 too large for 3-way interactions? Some (many?) of your 3-way interactions have a p-value of 0.03 (even 0.07, which you still keep?), and I am left questioning how relevant they are. It leads to results like "grunts produced closer to the human were shorter... but only in untamed piglets, effect being stronger before the conditioning" and "grunts had a higher frequency range ... when produced closer to the human..., but only in untamed piglets and before the conditioning". On a data set of this size, I wonder to what extent these results can be generalised. It makes the manuscript very long and very difficult to follow in places – and the main results drown. Is there enough power to make such detailed conclusions?

Line 445-449: Is this what you expected? Could your interpretation be affected by the nature of the treatment, in other words, you will describe the response of the H+ pigs as positive (either being touch/attention satiated,

or know they can always come back, and therefore disinterested, or confident enough to go exploring). How can this be disproven if you haven't set out expected outcomes from the start?

The last major concern is the text. The manuscript has a large number of typos (e.g. lines 405, 440, 463, 552 and elsewhere) and missing spaces, which leaves the impression that the uploading was rushed. I am usually quite forgiving when the English is a little bit rustic when the authors are writing in a second language. Unfortunately, there are places where it makes it difficult to understand what is meant (e.g. line 38: "a carrying human"; line 112: "on plastic duckboard and panels visually isolated pens"; line 568: "'AH' was more entitled to trigger higher positive states"), and as a reviewer, it can be jarring to have to second-guess the content. I would therefore recommend that the revised manuscript is copy-edited by third party before re-submission.

# Other issues:

The use of the word 'taming' is confusing, as domestic pigs (Sus scrofa domesticus) are all tame compared to the wild boar (Sus scrofa). I suggest to change this to 'positive handling'.

The authors refer to their study animals as piglets, but they are weaned and thus should be called pigs.

Line 19: objectify the quality?

Line 20: Is not all behaviour spatial?

Line 25: replace breeding with husbandry

Line 50: Animal welfare conveys? Consists of?

Line 69 and elsewhere: associated with, not to

Line 88: What is 'formant'?

Line 113: metal chain?

Line 145 and elsewhere: Hence

Figure 1. Suggest using 'area' for the distal and proximal areas, to not confuse it with the 16 zones. It is not clear where the distal area is (above or below the dashed line).

Line 161: Where the sessions recorded or was the annotation done live?

Line 170: Replace over with other

Table 1: "The number of times the piglet looked at other parts of the room" - as the pig will be looking somewhere at all times, this will always be within 1 of the previous variable (Nb looks toward human).

Table 2: The Table does not show "Behavioural response score for the reunion phase of the Isolation/Reunion test." but "Percentage of explained variance and variable loadings of the principal component analysis for the first three PCs."

Table 2 and elsewhere: You use the word 'parameter' when 'variable' is the correct term.

Table 4: Are these variables on vocalisation characteristics essentially showing the same, leading to a high loading for all of them on PC1, i.e. so highly correlated that they are not all needed?

Line 260: Vocal response variable

Lines 323 and 326: Why are these estimates?

Figure 4: It is not possible to know what comparisons the letters refer to, as some of them have no letter. What is different from what? Question if a three-way interaction on a subset of data is biological relevant? The blue and grey colours are indistinguishable.

Figure 5: What does H:N mean in the legend of A? Figure D is missing (referred to in Table heading). Not sure what C means - failed used in figure heading, missed used in y-axis label, but what does this show?

Line 369: Here you refer to the wrong Figure. This was carried over for the rest of the manuscript, so that wrong Figures were indicated.

Figure 6: Are these slopes based on linear regressions? is this justified?

Table 5: Does this not indicate either that vocal parameters are not very robust measures, or that you were unable to standardise your treatment?

Line 443: attraction instead of attractiveness

Line 467-468: This is using your interpretation of the positively handled pigs to draw conclusions on the limitations on the control group. The set-up did allow them to explore.

Line 514: Double reference

Line 568: More likely to? But does this not show that this test or variable is not generalisable?

Line 577: The major difference to the pigs is more likely to be in the difference in smells of the two handlers. Was there any thought given to soaps and perfume? Even body odours differ.

Lines 586-588: But some variables were not significant for one of the handlers.

Line 600: "We suggest that the use of vocalisations to assess quality of human-pig relationship could help to better monitor the parameters involved..." I don't know what this means?