The effect of personality measurement conditions on spontaneous swimming behavior in the pale chub *Zacco platypus* (Cyprinidae)

Shi-Jian Fu Corresp. [1]

[1] Laboratory of Evolutionary Physiology and Behavior, Chongqing Key Laboratory of Animal Biology, Chongqing Normal University, Chongqing, China

Corresponding Author: Shi-Jian Fu
Email address: shijianfu9@cqnu.edu.cn

The aim of the present study was to test whether spontaneous movement traits used in fish personality measurement are correlated or vary among different contexts in a common Chinese cyprinid fish, the pale chub (*Zacco platypus*, Cyprinidae). The median swimming speed, percent time spent moving, median turning rate and distance to the center of an arena were measured under boldness context (with a shelter available) and then exploration context (with a novel object nearby) and under the control context (i.e., with no shelter and novel object). The median swimming speed, percent time spent moving, and median turning rate all showed a positive correlation between the control and the other two contexts, which suggests that the personality of pale chubs is quite conserved and that future studies might use spontaneous swimming variables measured during exploration or boldness measurements to avoid the need to carry out a separate activity test. Further analysis comparing the distance and time latency to explore the novel object between the exploration condition (with the novel object present) and control condition (with an imaginary object at the same position) showed that only the amount of time it took for the fish to first reach the object for exploration significantly differed between the two contexts and hence might be a potential variable indicating exploration in the pale chub in the future.

The effect of personality measurement conditions on spontaneous swimming behavior in the pale chub *Zacco platypus* (Cyprinidae)

Shi-Jian Fu*

Laboratory of Evolutionary Physiology and Behavior, Chongqing Key Laboratory of Animal Biology, Chongqing Normal University, Chongqing, 401331, China

*Corresponding author

E-mail: shijianfu9@cqnu.edu.cn; Tel. and Fax: 862365910701

Running title: spontaneous activity and personality

ABSTRACT

The aim of the present study was to test whether spontaneous movement traits used in fish personality measurement are correlated or vary among different contexts in a common Chinese cyprinid fish, the pale chub (*Zacco platypus*, Cyprinidae). The median swimming speed, percent time spent moving, median turning rate and distance to the center of an arena were measured under boldness context (with a shelter available) and the exploration context (with a novel object nearby) and under the control context (i.e., with no shelter and novel object). The median swimming speed, percent time spent moving, and median turning rate all showed a positive correlation between the control and the other two contexts, which suggests that the personality of pale chubs is quite conserved and that future studies might use spontaneous swimming variables measured during exploration or boldness measurements to avoid the need to carry out a separate activity test. Further analysis comparing the distance and time latency to explore the novel object between the exploration condition (with the novel object present) and control condition (with an imaginary object at the same position) showed that only the amount of time it took for the fish to first reach the object for exploration significantly differed between the two contexts and hence might be a potential variable indicating exploration in the pale chub in the future.

Keywords personality measurement context, temperament, behavioral syndrome, activity, boldness, exploration

INTRODUCTION

The personality of an animal is a consistent difference among individuals in behaviors such as boldness, exploration and activity (Bell et al., 2009; Réale et al., 2010; Mazué & Godin, 2015; Jolles et al., 2019). Personality has been assumed to have large fitness consequences and a wide range of ecological and evolutionary implications (Smith and Blumstein, 2008; Réale et al., 2010; Sih et al., 2012); hence, it has attracted much attention recently (Jolles et al., 2017; Tang and Fu, 2019). The tendency to leave a refuge and search an open environment is traditionally referred to as boldness (Jolles et al., 2017), and studies have used latency to leaving shelter and percentage of time (or activity) outside shelter as indicators of this characteristic (Brown & Irving, 2014; Tang, Wu & Fu, 2018). The novel object test has been widely used in personality research to assess exploratory behavior (i.e., curiosity towards novelty) and neophobia (i.e., fear of novelty) (Galhardo, Vitorino & Oliveira, 2012). Exploration is usually evaluated in terms of the distance or time latency associated with the inspection of a novel object (Liu & Fu, 2017). For activity measurements, fish biologists usually use the characteristics of spontaneous activities (movements without external stimulus) in an arena, such as the median swimming speed and percent time moving during a given period (Brown & Irving, 2014; Liu & Fu, 2017).It has been long recognized that many animals exhibit so-called 'behavior syndrome' as the bold individuals are typically more exploratory and active than other individuals (Sih, Bell & Johnson, 2004; Sih et al., 2012; Martins & Bhat, 2014), i.e., personalities are highly correlated, at least with regard to boldness, exploration and activity. A study involving principal component analysis also suggested that most variables can be reduced to the so-called 'activity component', which is distinct from the 'sociability component' (Tang, 2019). If the variables are correlated across different personality contexts, researchers may be able to acquire data for variables related to different personality traits from one measurement. For example, if correlated, one could measure the activity variables mentioned above from a spontaneous movement trajectory of fish under boldness or exploration contexts. Thus, the present study aimed to determine whether traits of swimming behavior are correlated within individuals across different personality contexts. The context condition was as follows: boldness context with a shelter available, exploration context with a novel object nearby and activity context with no novel object or shelter.

To achieve our goals, I selected the pale chub (*Zacco platypus*), a common small cyprinid fish species distributed across east Asia, as an experimental model. This omnivorous fish species usually occupies fast-flow streams and prefers to shoal in open water but lie under or behind plants and stones against water flow to forage or avoid predators. We recorded videos of the experimental fish individuals under different personality contexts with a webcam. The variables we selected for comparison are median swimming speed, percent time spent moving, median turning rate of body centroid (which usually serves as an indicator of exploration tendency but is also closely correlated with activity), and the distance to the center of the arena (which has frequently been used as an indicator of boldness) (Couzin et al., 2011; Ioannou, Singh & Couzin, 2015; Sumpter et al., 2018).

The results found that most variables between exploration context and control context are highly correlated and not significantly different, which is unexpected. This might suggest that the novel object used in the present study was not a sufficient stimulus for exploration in pale chub. Thus, I conducted an additional analysis comparing the distance to the novel object and the latency to explore the novel object in the exploration context between those to a virtual object in the activity context. The aim of this additional analysis is to test whether the novel object is a sufficient stimulus, and the correlation between variables of activity and exploration is an artifact of the measurement protocols, i.e., the model. More active fish (individuals who spend more time moving and with a faster speed) might by chance spend less time reaching a novel object independent of their exploration tendency. Because no investigation has been conducted to test this issue before, it might provide a useful result for a study of exploration measurement in the future.

MATERIAL AND METHODS

Source of the fish and their care

A total of 80 juvenile pale chubs (body mass: 6.42 ± 0.14 g; mean ± S.E.) were captured by local fishers from a stream adjacent to the Wujiang River (29°24'37"N, 107°31'55"E, Wulong County, Chongqing City). All fish were reared in a 250 L recirculating system with aerated water. Twenty percent of the water was exchanged daily with fresh water. The water temperature was maintained at 25 ± 1 °C. The photoperiod was 12 h light: 12 h dark. The pale chub were hand-fed to satiation once daily (at 8:00 am) for 4 w with tubifex. The feces and uneaten food were removed with a siphon at 9:00 am. The dissolved oxygen level was kept above 90% saturation. After 4 w of acclimation, experimental fish were tagged intraperitoneally with passive integrated transponder (PIT) tags under anesthesia by neutralized tricaine methane sulfonate (MS222, 50 mg L^{-1}). Then, experimental fish recovered for 2 w. All individuals were used to measure personality.

The present study was authorized by the Animal Care and Use Committee of the Key Laboratory of Animal Biology of Chongqing (permit number Zhao-20161122-04).

Experimental setup

The experimental setup was similar to those which described in Tang & Fu (2019). Specifically, we used a rectangular tank (length × width × height: 70 × 20 × 20 cm) to measure spontaneous swimming behavior under different personality contexts, such as boldness, exploration and activity, as in previous studies (Liu & Fu, 2017; Fig. 1). The arena was surrounded with an opaque canvas to stimulus from the observers during the experiments. The arena was separated into two subareas by an opaque plastic partition that was used to separate the open area (length × width × height: 55 × 20 × 20 cm) from a shelter area (length × width × height: 15 × 20 × 20 cm), which provided a refuge with artificial plants. There was a small door (width × height: 10 × 10 cm) on the partition that allowed fish to move freely from shelter area to open area under boldness context, but it was closed under exploration and control contexts. The water depth was maintained at 10 cm during the experiment. The behaviors of the test fish were recorded using a webcam (Logitech Pro 9000; Logitech Company, Suzhou, China) placed 1.5 m directly above the aquarium and connected to a remote monitor, and the experimental tank was illuminated by fluorescent lights.

Experimental protocol

Two weeks after the tagging of PIT, the swimming behaviors of all fish under boldness, exploration and control contexts were recorded. The recordings were conducted in four identical aquariums at the same time from 8:00 am to 17:00 pm. The fish were first recorded under the boldness context, then the exploration context, and then the control context (see detail below). The aquariums were cleaned after the measurement of each fish individual.

Measurement of spontaneous swimming activities under different contexts

Spontaneous movements were recorded for all 80 individuals in the three contexts. However, the number of replicates for each test differed because some files could not be read by the software due to technical problems.

Boldness context A fish was transferred into the shelter area and acclimated for 30 min; then, the door was gently opened by the linking threat manipulated remotely, and the movement of the fish (only those in the open area) was recorded for 30 min (at 15 frames per second). Pale chub individuals showed large variation in terms of the time latency to enter the open area (ranging from nearly zero to 23 min) and the percentage of time spent in the open area (varying from 18 to 97%). Thus, the total duration of moving in the open area varied greatly among different individuals.

Exploration context After recoding under the boldness context, the fish that stayed in the shelter area were gently chased to the open area of the aquarium and acclimated for 10 min with the small door between open and hidden areas closed. Then, a novel object (a black, round, plastic ball with a diameter of approximately 2 cm, Liu & Fu., 2017) was carefully transferred to a place that faced 40 cm directly away from the small door (Fig. 1), and the movements of the fish were recorded by the webcam for 10 min.

Control context After recording the exploration context, the novel object was removed, and the fish was again acclimated for 10 min to eliminate the effect of the exploration context. Then, the movements of the fish were recorded by the webcam for 10 min.

Data calculation and analysis

The videos were imported into an automated tracking program, i.e., EthoVision XT9 (EthoVision XT 9, Nodus, Netherlands) after format conversion, which automatically calculated the x and y coordinates from pixels to cm. The trajectories were therefore smoothed using a weighted moving average with a window width of 0.5 s (Miller & Gerlai,2012). Then, the median speed while fish swimming (swimming speed above 1.75 cm s^{-1}), percent time moving, median turning rate while fish swimming and mean distance to the center of the open area were calculated for all three contexts.

The swimming speed (v, cm s^{-1}) was calculated as follows:

$$v(t) = \sqrt{(x(t) - x(t-1))^2 + (y(t) - y(t-1))^2}/d \qquad (1)$$

where $x(t)$ and $x(t-1)$ and $y(t)$ and $y(t-1)$ are the x and y coordinates, respectively, of the measured fish at time t and the time of the previous frame (t-1), and d is the length of the time interval (i.e., 0.5 s, Miller & Gerlai, 2012).

The median speed is among those when the swimming speed is above 1.75 cm s^{-1}.

The percent time moving was calculated as the percentage of time when the swimming speed was above 1.75 cm s^{-1} (Tang et al., 2017; Tang & Fu, 2019). The turning rate (ω_t, rad s^{-1}) of an individual was calculated using the absolute change in centroid (see details in Herbert-Read et al., 2013). This measure can be used to quantify exploratory as opposed to goal-directed behavior in fish, which is calculated as follows (Sumpter et al., 2018).

$$\omega_t = \frac{\cos^{-1}(\mathbf{c}_t \cdot \mathbf{c}_{t+1})}{\Delta t} \quad (2)$$

In brief, θ_t is the orientation of fish at time step *i* measured relative to the positive *x* axis of the coordinate system. Let $\mathbf{c}_t = \cos\theta_t + \sin\theta_t$ be the unit vector pointing in the direction of θ at time step *i*. Then, the change in the orientation of fish from time step *t* to time step *t* + 1 was given by $\cos^{-1}(\mathbf{c}_t \cdot \mathbf{c}_{t+1})$. Again, we calculated the median turning speed of each individual when the swimming speed was above 1.75 cm s^{-1}.

The distance from the center of the arena (cm) was calculated as

$$D_i = \sqrt{(x_i - x_0)^2 + (y_i - y_0)^2} \quad (3)$$

where x_i and y_i denote the coordinates of the fish, and x_0 and y_0 denote the coordinates of the arena center. In open fields, a large distance from the center often indicates low boldness in individual fish (Sumpter et al., 2018).

For the exploration trials, we also calculated two variables that are commonly used to test for differences in exploratory tendency in fish, i.e., the distance to the object and latency to explore the novel object under the exploration context (Adriaenssens & Johnsson, 2011; Mazué & Godin, 2015).

The distance (cm) was calculated as $$D_i = \sqrt{(x_i - x_n)^2 + (y_i - y_n)^2} \quad (4)$$

where x_i and y_i denote the coordinates of the fish, and x_n and y_n denote the coordinates of the object. The latency to explore the novel object is defined as the amount of time it took for the fish to first swim within 7 cm (approximately 1 body length) of the object.

To assess whether the fish were responding to the novel object, we measured the same two variables as the control treatment in relation to a virtual object in the same location. The calculation assumed that the coordinates of both the novel and the virtual objects were located at the center of the object. The dimension of the novel object, i.e., a 1 cm radius, might have little effect on the distance to the coordinates and the latency to explore the object.

One-sample Kolmogorov–Smirnov test results indicated that median swimming speed, median turning rate, distance to the center and distance to the novel object were normally distributed, whereas percent time moving and latency to the novel (or virtual) object were not normally distributed. Thus, the relationships of each of the behavioral variables between the control and the two other contexts and the relationship of two variables associated with exploration between exploration and control contexts were tested with either Pearson or Spearmen correlation analysis based on whether the variables were normally distributed. If significantly correlated, the relationships were also examined using linear regressions. The effect of measurement context on median swimming speed, median turning rate and distance to the center was tested by a linear mixed model (LMM) using fish ID as a random factor. The paired t-test was used to compare the difference in median swimming speed between any two contexts. The difference in percent time moving was compared by a nonparameter Wilcoxon matched-pairs test. The distance associated with exploration under the exploration and control contexts with real or virtual novel object were compared with paired t-tests, whereas the latency to object was compared with a nonparameter Wilcoxon matched-pairs test. The program SPSS 17 was used for data analysis. *P* values < 0.05 were considered statistically significant, and all the data are presented as the mean ± S.E.

RESULTS

Effect of measurement context on spontaneous movement traits

The median swimming speed (Pearson correlation, $P < 0.001$), percent time moving (Spearman correlation, $P < 0.001$) and median turning rate (Pearson correlation, $P < 0.001$) measured under the control context were positively correlated with those measured under the other two contexts (Table 1; Fig. 2). The distance to the center measured under the control context was not correlated with that measured either under the boldness context (Pearson correlation, $P = 0.650$) or under the exploration context (Pearson correlation, $P = 0.424$).

The measurement context had a significant effect on median swimming speed (LMM, $F_{2,\ 150.29} = 70.637$, $P < 0.001$), but it showed no effect on median turning rate (LMM, $F_{2,\ 149.17} = 0.263$, $P = 0.796$) or distance to the center (LMM, $F_{2,\ 152.56} = 2.667$, $P = 0.073$) (Fig. 3). The median swimming speed of fish measured under the boldness context was significantly higher than that of those measured under both exploration (paired t-test, t_{73} = 9.723, P < 0.001) and control (paired t-test, t_{73} = 10.398, P < 0.001) contexts. The percent time spent moving measured under the boldness context was significantly higher than that measured under the exploration context (nonparameter Wilcoxon test, z = -4.873, P < 0.001), whereas the latter was significantly higher than that measured under the control context (nonparameter Wilcoxon test, z = -5.083, P < 0.001).

Difference in exploration variables between the exploration context and control context with a virtual object

The distance to the novel (or) virtual object (Pearson correlation, R = 0.435, N = 79, P < 0.001) and the latency to reach the novel (or) virtual object (Spearman correlation, R = 0.329, N = 78, P < 0.001) were positively correlated (Fig. 4).

The distance to the novel object in the exploration context showed no significant difference from that to the virtual object in the control context (paired t-test, t_{78} = -0.838, P = 0.405) (Fig. 5a). However, it took a much shorter amount of time for the fish to first reach the novel object under the exploration context than for the fish to reach the virtual object under the control context (paired t-test, t_{78} = -5.772, P < 0.001) (Fig. 5b).

DISCUSSION

The correlation of spontaneous swimming traits between the control and other two contexts

The values of activity variables, such as the median swimming speed, median turning rate and percent time moving, measured under the control context were closely correlated with those measured under the boldness and exploration contexts. This agrees with the results of a previous study that showed that variables such as median swimming speed and median turning rate were quite constant across different measurements or contexts in mosquito fish (*Gambusia affinis*) (Herbert-Read et al., 2013) and in zebrafish (*Danio Rerio*) (Toms & Echevarria, 2014). These results suggest that researchers can evaluate personality traits associated with activity using only boldness or exploration contexts and do not need to perform activity measurements separately. However, the distance to the center of the aquarium showed no relationship between the control and the other two contexts, and the values were quite similar across contexts. It might be possible that the size (relatively small) and shape (rectangle rather than round) of the aquarium greatly constrained this measure so that the values have little meaning compared to a larger arena.

The comparison of spontaneous swimming traits across contexts

All variables measured under the exploration context were almost the same as those under the control context, except for a 27% difference in percent time moving compared to those measured under the control context. Fish under the boldness context showed a 33% higher median swimming speed and 35% higher percent time moving compared to those measured under the control context. The difference in percent time moving might suggest that pale chub pursue more exploration- and boldness-related activities, such as inspecting novel environments or searching for food or potential predators. However, it might have little biological significance given the large difference among individuals. Furthermore, the difference in swimming activity between boldness and control contexts might be because some of the higher movement rates result from fish fleeing quickly back to shelter after entering the arena. Nevertheless, the distinctly different swimming patterns might suggest that variables that have been frequently used in previous studies, such as the percentage of time spent outside of a shelter, might be reliable indicators of boldness in fish species such as the pale chub (Alain et al., 2015; Mazué and Godin, 2015).

The relationship of exploration variables between exploration and control contexts

All variables measured under the exploration context were almost the same as those under the control context. Thus, I assumed that the presence of so-called novel objects might not be a stimulus for exploration in pale chubs in the present study. It is worth noting that the size and shape of the arena and novel object are similar to those in previous studies. For example, a previous study in blue gourami (*Trichogaster trichopterus*) used a 40 × 60 × 40 cm aquarium (Bisazza, Lippolis & Vallortigara, 2001), a 90 × 30 × 45 aquarium and a 1.3 cm novel object for guppy (*Poecilia reticulata*) (De Serrana, Fong & Rodd, 2016). A 50 × 20 × 36 aquarium and 1.8 cm diameter novel object were used for zebrafish (Lucon-Xiccato & Dadda, 2014), and identical conditions were adopted in crucian carp (Liu & Fu, 2017). Thus, the fact that previous studies as well as the present study found a positive relationship between activity variables such as speed (or percent time moving) and exploration variables (distance and latency to the novel object) might have occurred because more active fish spend more time swimming at higher speed and hence show a shorter distance to the object and exploration latency in comparison to less active individuals (Liu & Fu, 2017; Tang & Fu, 2019). This is reinforced by the results indicating that the fish showed almost identical values in terms of the distance to novel objects in the same location when measured under the control context (i.e., a virtual object). However, this might be because the pale chub in the present study did not need to change their swimming behavior or position in the aquarium to explore. This might also be due to the separate stress from their conspecifics, which resulted in a pattern of swimming that was slightly affected by other test contexts, as this fish species preferred the group level in the field. Recent studies in cyprinid fish species such as qingbo (*Spinibarbus sinensis*) found that the spontaneous activity pattern varied profoundly when tested in singleton compared to those tested in dyads or in groups (Wang et al., 2019; Xu et al. 2019). However, since we measured the swimming activity under the boldness context first, it was unlikely because the aquarium is a novel environment for the fish, so adding another element of novelty may not change the situation from the fish's perspective.

It was interesting to find that the time it took for the fish to first reach the object was much shorter than that under the control context, i.e., the fish encountered the object earlier under exploration. This suggests that at least time latency can be used as an exploration indicator in fish species such as the pale chub under the situation in the present study. The reason might be that the novel object is too simple and that the pale chub only performed superficial inspection or contact with the object and then exhibited a similar spontaneous swimming pattern as those in the control context. Nevertheless, this study suggests that fish behaviorists should be cautious regarding explanations pertaining to exploration indicators previously used in other personality studies, such as the distance to an object (Wang et al., 2019; Wang, 2019). Researchers at least need to test the reliability of such measurements, for example, by comparing spontaneous movement trajectories in fish between exploration and control contexts, as in the present study. The testing of other variables under different experimental setups, such as the number of times individual guppies (*Poecilia reticulata*) traveled between containers connected by pipes (Brown & Irving, 2014), might be more appropriate for the measurement of exploration tendency.

CONCLUSIONS

In conclusion, the pale chubs showed a highly positive correlation in median swimming speed, median turning rate and percent time moving between the control and the other two contexts, which suggests that fish behaviorists can use such traits from either boldness or exploration measurement tests rather than conduct additional activity personality measurements. Further analysis revealed that the latency to an object might be a reliable indicator of exploration, whereas a traditionally used variable such as distance to a novel object might not be an appropriate indicator, at least for the pale chub in the present study.

Acknowledgements This work was supported by the National Natural Science Foundation of China (No. 31670418). We would like to thank Dr. Donald Kramer and two anonymous reviewers for insightful comments and constructive suggestions for statistical and writing that greatly improved the manuscript.

References

Adriaenssens B. Johnsson JI. 2011. Shy trout grow faster: exploring links between personality and fitness-related traits in the wild. Behavioural Ecology 24: 135-143

Alain P, Anthony S, Marie-Laure B, Yannick L, Fabrice T, Pascal F. 2015. First insight into personality traits in Northern pike (*Esox lucius*) larvae: a basis for behavioral studies of early life stages. Environmental Biology of Fish 99: 105-115Bevan PA, Gosetto I, Jenkins ER, Barnes I, Ioannou CC. 2018. Regulation between personality traits: individual social tendencies modulate whether boldness and leadership are correlated. Proc. R. Soc. B Biol. Sci. 285

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

Bisazza A , Lippolis G, Vallortigara G. 2001. Lateralization of ventral fins use during object exploration in the blue gourami (*Trichogaster trichopterus*). Physiology and Behavior, 72(4), 575-578.

Brown C, Irving E. 2014. Individual personality traits influence group exploration in a feral guppy population. Behavioural Ecology 25: 95-101

Couzin ID, Ioannou CC, Demirel G, Gross T, Torney CJ, Hartnett A, Conradt L, Levin SA, Leonard NE. 2011. Uninformed individuals promote democratic consensus in animal groups. Science 334:1578–1580

De Serrano AR, Fong C, Rodd FH. 2016. Effects of methylphenidate on responses to novelty in a teleost fish (*Poecilia reticulata*). Behavioural Brain Research, 302,53-59

Galhardo L, Vitorino A, Oliveira RF. 2012. Social familiarity modulates personality trait in a cichlid fish. Biology Letters 8:936-938

Heg D, Schuürch R, Roghenberger S. 2011. Behavioral type and growth rate in a cichlid fish. Behavioral Ecology 22: 1227-1233

Herbert-Read JE, Krause S, Morrell LJ, Schaerf TM, Krause J, Ward AJW. 2013. The role of individuality in collective group movement. Proceeding Royal Society B 280: 20122564
Ioannou CC, Singh M, Couzin ID. 2015. Potential leaders trade off goal-oriented and socially oriented behavior in mobile animal groups. American Naturist 186: 284–293
Jolles JW, Boogert NJ, Sridhar VH, Couzin ID, Manica A. 2017. Consistent individual differences drive collective behavior and group functioning of schooling fish. Current Biology 27: 2862-2868
Jolles JW, Briggs H, Araya Y, Boogert NJ. 2019. Personality, plasticity and predictability in sticklebacks: bold fish are less plastic and more predictable than shy fish. Animal Behaviour 154:193-202
Liu S., Fu SJ. 2017. Effects of food availability on metabolism, behaviour, growth and their relationships in a triploid carp. Journal of Experimental Biology. 220: 4711-4719
Lloren JI, Davidson SM, Twardek WM, Elvidge CK. 2019. Baseline activity and shoal type determine antipredator behaviors in bluegill from a southern ontario lake. Behavioral Ecology and Sociobiology, 73(5):57
Lucon-Xiccato T, Dadda M. 2014. Assessing memory in zebrafish using the one-trial test. Behavioural Processes, 106, 1-4
Martins EP, Bhat A. 2014. Population-level personalities in zebrafish: aggression-boldness across but not within populations. Behaviroural Ecology 25: 368–373
Mazué GPF, Godin JGJ. 2015. Boldness-exploration behavioural syndrome: interfamily variability and repeatability of personality traits in the young of the convict cichlid (*Amatitlania siquia*). Behaviroural Ecology 26: 900-908
Miller N, Gerlai R. 2012. Automated tracking of zebrafish shoals and the analysis of shoaling behavior. In: Kalueff AV, Stewart AM, eds. Zebrafish protocols for neurobehavioral research. New York, Humana Press, pp 217-230
Réale D, Garant D, Humphries MM, Bergeron P, Careau V, Montiglio PO. 2010. Personality and the emergence of the pace-of-life syndrome concept at the population level. Proceeding Royal Society B 365: 4051-4063
Sih A, Cote J, Evans M, Fogarty S, Pruitt J. 2012. Ecological implications of behavioural syndromes. Ecology Letter 15: 278-289
Sih A, Bell A, Johnson JC. 2004. Behavioral syndromes: an ecological and evolutionary overview. Trends in Ecology and Evolution 19(7): 372-378
Smith BR, Blumstein DT. 2008. Fitness consequences of personality: a meta-analysis. Behavioural Ecology 19: 448–455
Sumpter DJT, Szorkovszky A, Kotrschal A, Kolm N, Herbert-Read JE. 2018. Using activity and sociability to characterize collective motion. Philosophical Transactions of Royal Society B 373: 20170015
Tang J. 2019. The personality of pale chub (*Zacco Platypus*). Master Degree thesis. Chongqing Normal University Chongqing. China
Tang ZH, Fu SJ. 2019. Qingbo (*Spinibarbus sinensis*) personalities and their effect on shoaling behavior. Acta Ethology 22:135-144
Tang ZH, Wu H, Huang Q, Kuang L, Fu SJ. 2017. The shoaling behavior of two cyprinid species in conspecific and heterospecific groups. Peer J 5:e3397
Tang ZH, Wu QY, Fu SJ. 2018. Inspection behaviour and inter-individual cooperation in juvenile qingbo: the effects of prior predator exposure and food deprivation. Journal of Ethology 36:181-190
Toms CN, Echevarria DJ. 2014. Back to basics: searching for a comprehensive framework for exploring individual differences in zebrafish (*Danio Rerio*) behavior. Zebrafish, 11; 325-340
Wang Y, Fu SJ, Fu C. 2019. Behavioral adjustments to prior predation experience and food deprivation of a common cyprinid fish species vary between singletons and a group. PeerJ 7: e7236
Wang L. 2019. The effect of food abundance on standard metabolic rate, personality and shoal behavior of pale chub (*Zacco Platypus*). Master Degree thesis. Chongqing Normal University Chongqing. China
Wang L, Tang JY, Qin YL, Zeng LQ, Peng JL, Fu SJ. 2019. Effect of starvation on energy metabolism,

fish behavior, and schooling behavior of *Spinibarbus sinensis*. Acta Ecologica Sinica 39(3):1095-1104

Wang Y, Fu SJ, Fu C. 2019. Behavioral adjustments to prior predation experience and food deprivation of a common cyprinid fish species vary between singletons and a group. PeerJ 7: e7236

Xu JJ, Fu SJ, Fu C. 2019. Physiological and behavioral stress responses to predators are altered by prior predator experience in juvenile qingbo (*Spinibarbus sinensis*). Biology Open 8, bio041012.

Table 1(on next page)

The statisticalresults of correlation between the control and other two contexts

Pearson or Spearman (percent time moving) correlations between measurements conducted under the control context and those conducted under either boldness or exploration contexts.

Table 1 Pearson or Spearman (percent time moving) correlations between measurements conducted under the control context and those conducted under either boldness or exploration contexts.

		Context	
		Boldness	Exploration
Median swimming speed			
	N	74	78
	R	0.644	0.656
	P-value	<0.001	<0.001
Percent time moving			
	N	72	67
	R	0.450	0.619
	P-value	<0.001	<0.001
Median turning rate			
	N	74	78
	R	0.660	0.664
	P-value	<0.001	<0.001
Distance to center			
	N	74	78
	R	-0.054	0.092
	P-value	0.650	0.424

Figure 1

Design of the experimental aquarium used for the measurement of activity, exploration and boldness in the study.

A) hidden area (length × width × height: 15 × 35 × 35 cm); B) open area (length × width × height: 55 × 35 × 35 cm); C) removable opaque PVC divider; and D) small door (10 × 10 cm)

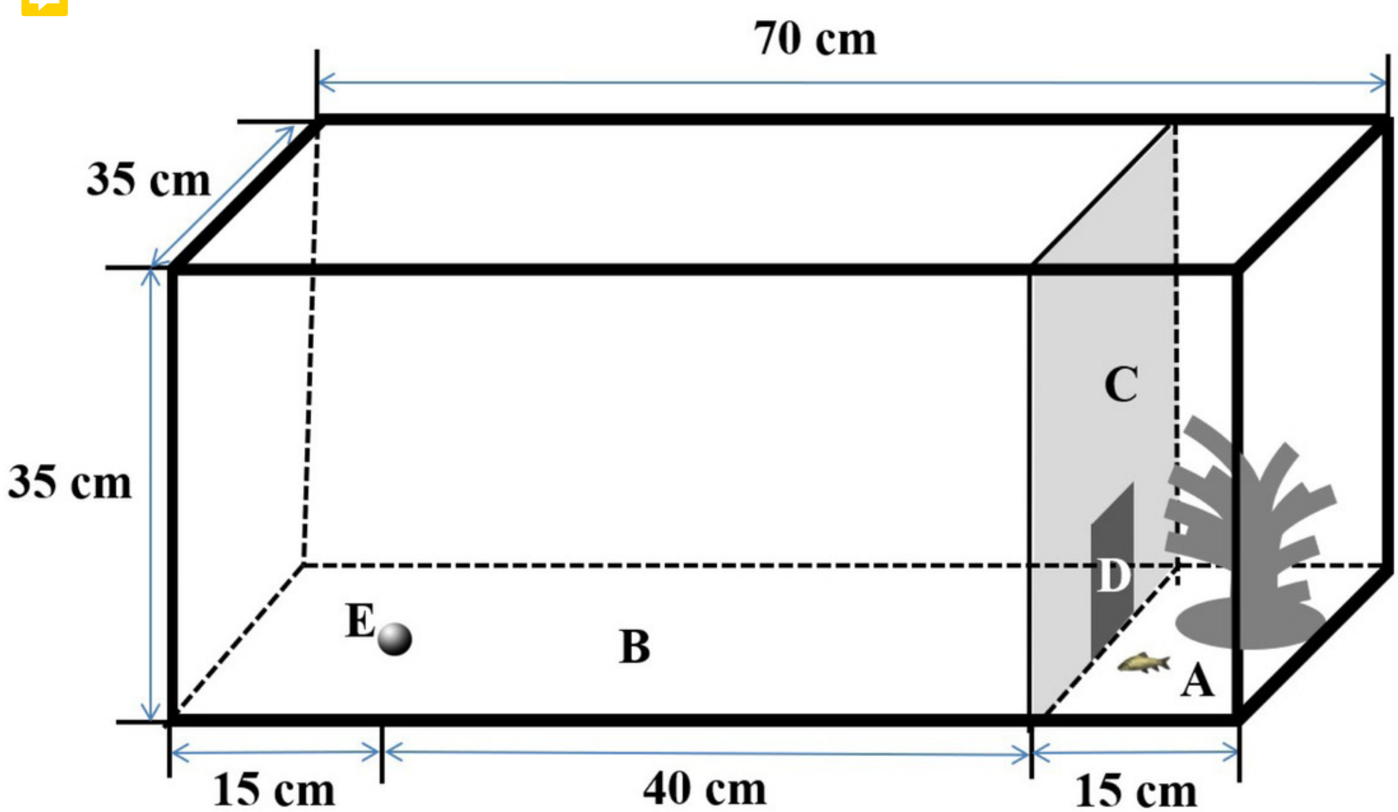


Figure 2

Correlations between the control context and two other contexts, boldness (red dots) and exploration (blue dots) for three measures of activity

[median swimming speed (A), percent time moving (B) and median turning rate (C)] in pale chubs. Dotted lines represent the relationships between two different contexts using linear regressions.

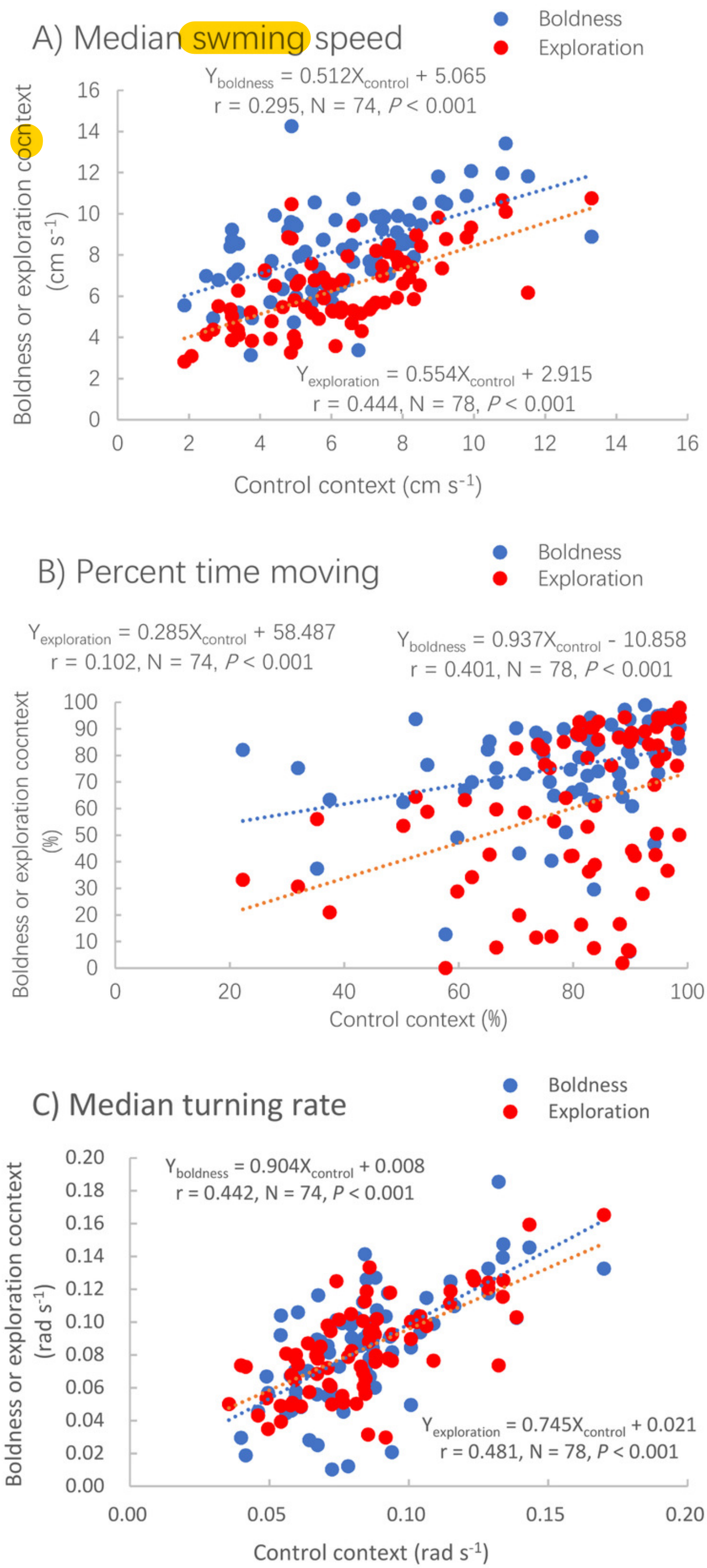

A) Median swming speed
Boldness
Exploration
Yboldness = 0.512Xcontrol + 5.065
r = 0.295, N = 74, P < 0.001
Yexploration = 0.554Xcontrol + 2.915
r = 0.444, N = 78, P < 0.001
Boldness or exploration cocntext (cm s-1)
Control context (cm s-1)
B) Percent time moving
Boldness
Exploration
Yexploration = 0.285Xcontrol + 58.487
r = 0.102, N = 74, P < 0.001
Yboldness = 0.937Xcontrol - 10.858
r = 0.401, N = 78, P < 0.001
Boldness or exploration cocntext (%)
Control context (%)
C) Median turning rate
Boldness
Exploration
Yboldness = 0.904Xcontrol + 0.008
r = 0.442, N = 74, P < 0.001
Yexploration = 0.745Xcontrol + 0.021
r = 0.481, N = 78, P < 0.001
Boldness or exploration cocntext (rad s-1)
Control context (rad s-1)

Figure 3

Mean ± S.E. of four measures of spontaneous swimming activity

[median swimming speed (A), percent time moving (B), median turning rate (C) and distance to center of the aquarium (D)] in pale chubs in the boldness (N = 74), exploration (N = 78) and control (N = 79), contexts. Bars with different letters are significantly different ($P < 0.05$).

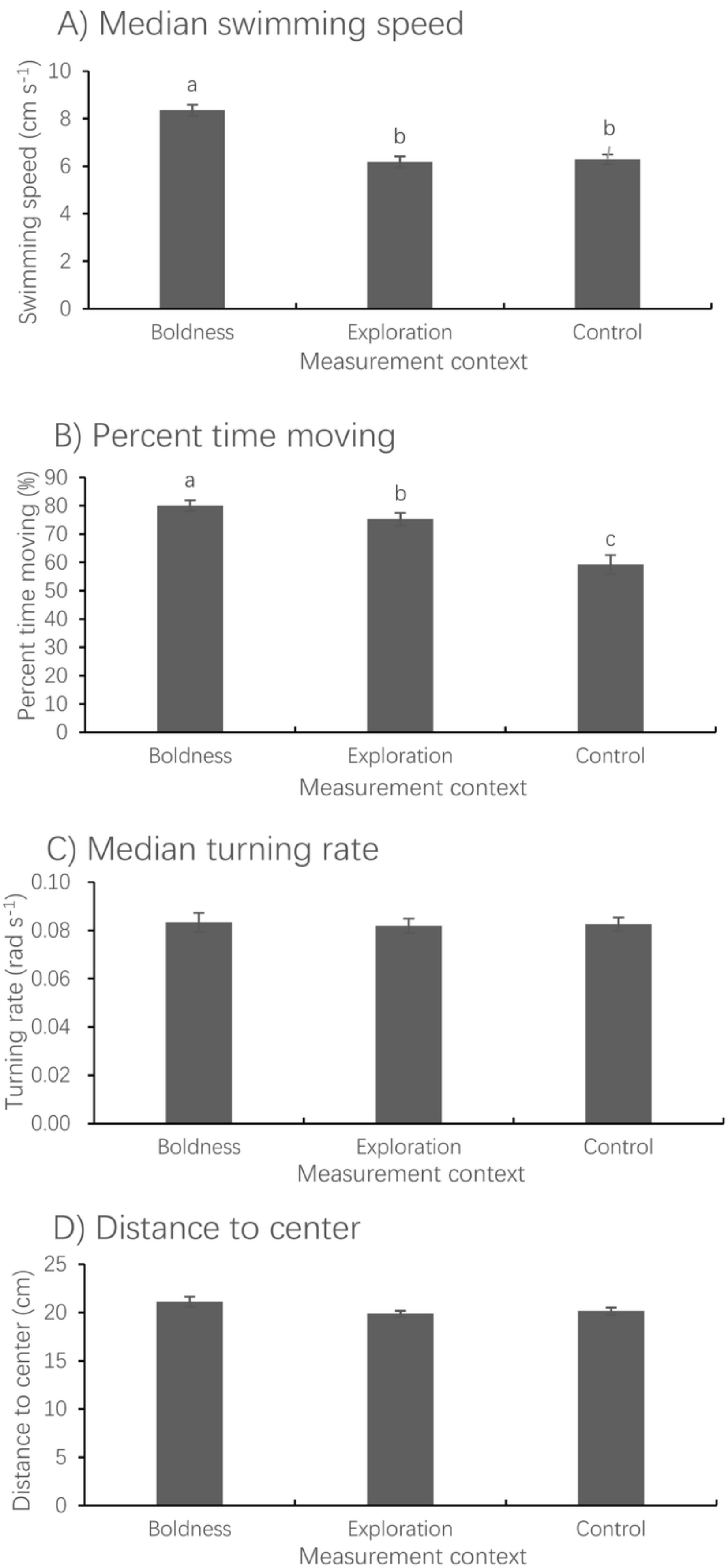

A) Median swimming speed
Swimming speed (cm s-1)
0
2
4
6
8
10
a
b
b
Boldness
Exploration
Control
Measurement context
B) Percent time moving
Percent time moving (%)
0
10
20
30
40
50
60
70
80
90
a
b
c
Boldness
Exploration
Control
Measurement context
C) Median turning rate
Turning rate (rad s-1)
0.00
0.02
0.04
0.06
0.08
0.10
Boldness
Exploration
Control
Measurement context
D) Distance to center
Distance to center (cm)
0
5
10
15
20
25
Boldness
Exploration
Control
Measurement context

Figure 4

Correlations between the exploration and control contexts

Correlations between the exploration and control contexts for measures of distance to the novel object (A) and latency to the object (B). Dotted lines represent the relationships between two different contexts using linear regressions.

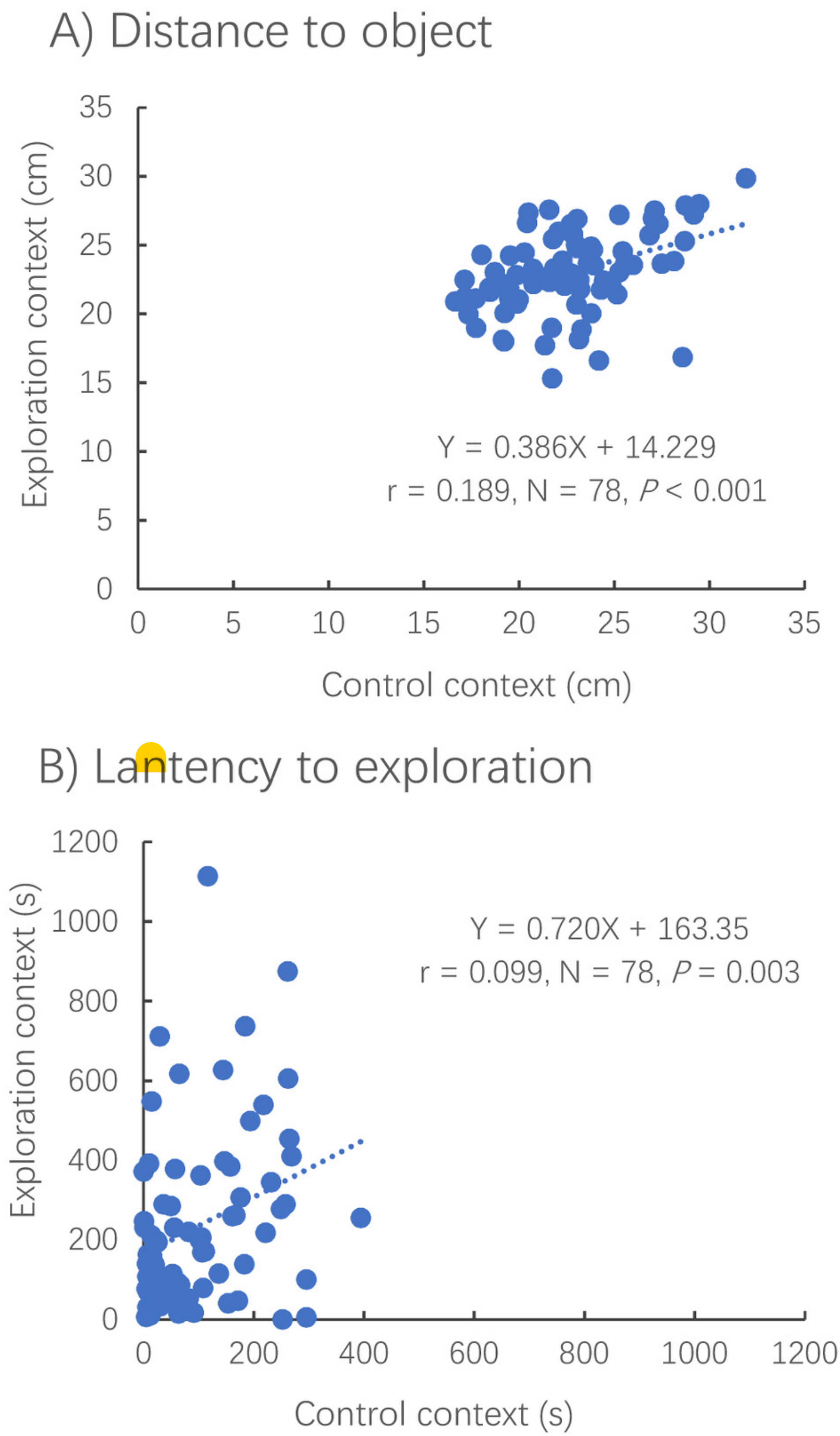
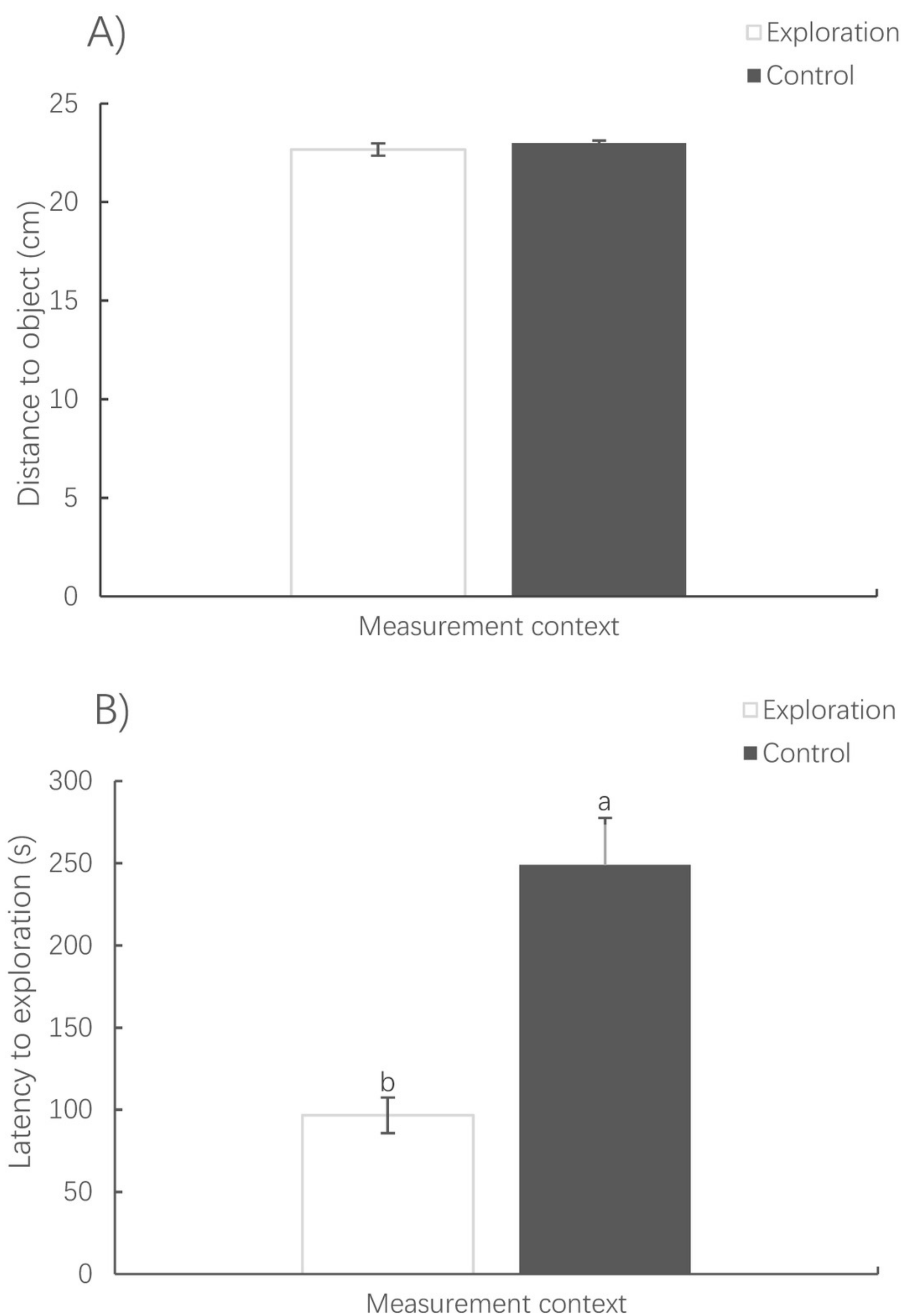

A) Distance to object
Exploration context (cm)
35
30
25
20
15
10
5
0
0
5
10
15
20
25
30
35
Control context (cm)
Y = 0.386X + 14.229
r = 0.189, N = 78, P < 0.001
B) Lantency to exploration
Exploration context (s)
1200
1000
800
600
400
200
0
0
200
400
600
800
1000
1200
Control context (s)
Y = 0.720X + 163.35
r = 0.099, N = 78, P = 0.003

Figure 5

Mean distance (A) and latency to exploration (B) under the exploration context and the virtual object under the activity context

(mean ±S.E., N = 78). Bars with different letters are significantly different ($P < 0.05$).


A)
Exploration
Control
25
20
15
10
5
0
Distance to object (cm)
Measurement context
B)
Exploration
Control
300
250
200
150
100
50
0
Latency to exploration (s)
a
b
Measurement context