



Parental sexual ornamentation and offspring performance in whitefish (*Coregonus lavaretus*)

Achancho Ekpombang Arnold

ABSTRACT

The expression of secondary sexual characters in both sexes (mutual ornamentation) is a relatively common, but rarely studied phenomenon in the animal kingdom. The primary aim of this study was to examine whether there is an association between mutual ornamentation and offspring performance in whitefish, *Coregonus lavaretus*. We conducted an experiment using newly hatched whitefish larvae, originated from crossing 10 females and 10 males in all possible combinations ((wild fish from River Oulujoki stock). The parental fish were examined for breeding tubercle ornamentation—height of lateral breeding tubercles (BT height), roughness class of lateral breeding tubercles (BT class), number of lateral breeding tubercles (BT side) and the number of front head breeding tubercles (BT front), and for parasites. Offspring quality was measured as predation avoidance behaviour (freezing and swimming close to the side of the tank, in the presence of predator 0-yr brown trout) and survival (in the absence of predator). The “good genes hypothesis” predicts that in the families with elaborate parental breeding tubercles the larvae are of better quality. The “compatible genes hypothesis” predicts that the variation between families in offspring quality is not consistent in relation to parental ornamentation but there is a male-female interaction in offspring quality. In accordance with the compatible genes hypothesis, we found a significant male-female interaction in predation avoidance behavior of larvae, although the male effect and female effect as such were insignificant. On the other hand, the survival of larvae was affected by the male effect (sire), only, as the maternal effect and male-female interactions were not significant. This finding is in accordance with the good genes hypothesis. However, there was a negative correlation both between the number of breeding tubercles on the front head and offspring survival, as well as between lateral breeding tubercle roughness class and offspring survival in males. Thus, breeding tubercle ornamentation of male whitefish seems not to indicate offspring survival in laboratory conditions. On the opposite, ornamentation of male whitefish may signal parasite load since there was a significant negative correlation between the roughness of lateral breeding tubercles and abundance of *Diplostomum spathaceum* eye flukes in the eye lens, as well as with the total abundance of Diplostomatidae flukes in the eye. Thus lateral skin roughness could be used by females to discriminate between male parasite status. In general, in males all the breeding tubercle ornamentation measures were independent, not correlated to each other. No association between the ornament measures of males and predation avoidance behavior of offspring was observed. Opposite to males, in females there was a positive association between BT class, BT side and BT height.

INTRODUCTION

Sexual selection

Sexual selection refers to the differences in reproduction that is brought about by variation among individuals in terms of traits that affect success in competition over mates and fertilization. In many species, females choose their mates based on the expression of male secondary sexual characteristics (Anderson 1994). Secondary sexual characters are often expressed in both sexes, but this mutual ornamentation and information content of female ornaments has been less studied when compared with elaborate sexual ornamentation in males (Admundsen, 2000; Kraaijeveld-smith & Komdeur, 2007). Female ornamentation has traditionally been considered as a genetically correlated response to selection for male ornaments (Lande, 1980), but there is growing empirical evidence for the presence of direct sexual selection by male and female reproductive competition (Admundsen, 2000; Kraaijeveld *et al.*, 2007; Clutton-brock, 2009; Watson & Simmons, 2010). As such, female ornaments have been examined for their effect on male selection as well as their relationship to female and offspring quality (Weiss, Kennedy & Bernhard, 2009). Although, some recent studies have revealed that female ornaments can predict offspring quality (Weiss *et al.*, 2009; Kekäläinen *et al.*, 2010), these are rare examples and further research is needed for a better understanding of the signalling function of female ornamentation.

Theory of Sexual selection

The sexual selection theory predicts that females choose males based on direct benefits (resources and parental care) or indirect benefits (good genes) (Hamilton & Zuk, 1982; Anderson, 1994). In the latter nonrecourse-based mating, males provide only genes to their mates (Neff & Pitcher, 2005), and as a result, females should select male genes that will maximize the survivorship or mating success of the offspring. The existence of female mating preference is seldom questioned when species in which males differ in their relative ability to provide resources or care for the offspring is considered. Furthermore, when males provide neither resources nor parental care, the maintenance of female mating preference is less unclear and has been a matter of debate. Two major classes of models could be used to explain mate preference in this case. One being the Fisher (1930) model which proposes that mate choice can be based on traits that are attractive to members of the opposite sex, but do not reveal anything apart from that (Lande, 1981; Kirkpatrick, 1982; Pomainkowski *et al.*, 1991; Pomainkowski & Iwasa, 1998). The other being the “goodness hypothesis” which predicts that mate choice enhances the survival chances of the offspring by selecting beneficial genes (reviewed in Kirkpatrick & Ryan, 1981; Anderson, 1994; Wedekind, 1994a; Westneat & Birkhead, 1998). The above models often assume that coevolution with parasites is a driving force of sexual selection, because constantly changing pressure by parasites may prevent sexual selection from depletion variation in heritable fitness (Hamilton & Zuk, 1982; Esthel & Mamilton, 1984). However, these sexual ornaments are costly to produce or maintain (Maynard Smith & Harper, 2003) and elaborate ornaments are believed to be handicap that reduces survival probability of the bearer (Zahavi, 1975).

Background information about whitefish

Alpine whitefish (*Coregonus* sp.) are typically caught during the breeding season, and their gametes are being stripped into large beakers for artificial breeding, and the eggs

and larvae are reared for sometime before they are released into the wild. This procedure does not take into consideration any potential benefit of mate choice, despite the fact that most if not all of the fish studied do not randomly mate, but have complex mating systems (Taborsky, 1994).

Breeding ornamentation

Whitefish (*Coregonus* spp.) are group-spawning freshwater fish species, where both sexes develop a distinct breeding ornamentation (i.e. breeding tubercles) prior to spawning period (Wiley & Collette, 1970, Wedekind *et al.*, 2001, 2008). These are little horn-like and colorless structures which, in the case of whitefish are distributed laterally over the skin. They grow before the breeding season and fall shortly after the breeding season.

Breeding tubercles are epidermal structures whose function in white fish has been suggested to be the maintenance of contact between individuals during spawning. They are also used by some species in the defense of nests and territories and perhaps in the stimulation of females in breeding (Wiley & Collette, 1970). Previous studies by Wedekind, 1992 and Rudolfson *et al.*, 2008 showed that the size and / or number of the male breeding tubercles in different species of fish are linked to individual condition, parasite resistance (Kortet *et al.*, 2004b) and male dominance (Kortet *et al.*, 2004a). Even though it is not known if these tubercles are costly to produce (Wedekind *et al.*, 2008; Kortet *et al.*, 2003). However, it has been shown that honest ornamentation does not need to be costly (Berglund *et al.*, 1997). Also, honest female ornamentation may require that energetic cost of ornamentation are low or absent. This is the case because in females, investment in ornamentation may directly limit investment in offspring (Fitzpatrick *et al.*, 1995; Berglund *et al.*, 1997).

The mating system of whitefish is not well known. It was suspected to be similar to that in roach (*Rutilus rutilus*), which is another group-spawning fish of similar size to whitefish and which develops breeding tubercles as well. Roach have a lek-like mating system with different male reproductive strategies, where females differ in their spawning preferences (Wedekind, 1996). In white fish (*Coregonus lavaretus*), males do not provide paternal care to the offspring and females produce large number of eggs, which are fertilized externally, making the species an ideal model for the test of genetic effects of sexual selection.

Breeding tubercle as an indicator of fish quality

Furthermore, (Wedekind *et al.*, 2001) showed a strong maternal and paternal effect in egg mortality of whitefish and reported a negative association between the size of the male breeding tubercles and embryonic mortality (i.e. mortality caused by bacterial infection could be predicted by male ornamentation). Due to the inconsistent results obtained with respect to the embryonic mortality of whitefish, recent studies by (Wedekind *et al.*, 2008) suggested that only some aspects of genetic quality are revealed by breeding tubercles.

Numerous studies have demonstrated the association between secondary sexual traits and survival of the offspring (Møller & Alatalo, 1999; (Wedekind *et al.*, 2001, 2008) but fitness related offspring performance is much less studied. Also, past studies revealed that there is no parental effect of male ornamental coloration on offspring swimming ability (Evans *et al.*, 2004). Nevertheless, Nicoletto, 1991, showed a positive correlation between the intensity of male carotenoid ornamentation and swimming performance in adult guppy (*Poecilia reticulata*).

Relationship between immune system and bacterial infection in whitefish

Previous studies by Wedekind *et al.*, (2004), showed that there is no evidence for MHC-linked gamete fusion and also found a strong connection between the MHC class II genotype and embryo susceptibility to *Pseudomonas fluorescens* in one of the seven subships used. They concluded that the MHC class II genotype can considerably influence embryo survival in whitefish, but gamete fusion seemed to be random with respect to the major histocompatibility complex. Furthermore, Siebenthal *et al.*, (2009) studied the tolerance of whitefish embryos to *Pseudomonas fluorescens* and found that a first non-lethal exposure had immunosuppressive effects: pre-exposed embryos were more susceptible to future challenges with the same pathogen. At intermediate and high levels of pathogen intensity, maternal effects turned out to be crucial for the embryos' tolerance to infection. Paternal (i.e. genetic) effects played a significant role at the strongest level of infection. The results suggest that whitefish embryos are largely protected by maternally transmitted substances, but build up their own innate immunocompetence several days before hatching.

Relationship between fish larvae and predators

Fish larvae are highly vulnerable to a broad array of vertebrate and invertebrate predators because their small size and incomplete development limit their abilities to detect and respond effectively to predators (Bailey and Houde 1989, Fuiman and Magurran 1994). Starvation also may be a serious risk because of the high mass-specific metabolic rates and low energy reserves of fish larvae (Fuiman 2002) and their patchily distributed food resources (Houde 1989). Poor foraging skills of inexperienced juveniles have been suggested to result in starvation in birds (Sullivan, 1989; Ward & Kennedy, 1996).

Recent studies by Huuskonen *et al.*, (2009), showed that offspring first feeding success had a strong paternal effect and it was also strongly correlated with the size of the male breeding tubercles, indicating that ornamentation can function as an honest indicator of their genetic quality. The behavior of newly hatched fish larvae is an important factor determining subsequent growth and of individuals (Fuiman *et al.*, 2003). Previous studies by Ylönen *et al.*, 2007 revealed that both perch and ruffe responded to predator fish odors. They were abt passive and erected the spiny dorsal fins. Fin erection showed clearly the black warning ornamentation in the fin, hence erected fin may function both in mechanical defense and also as warning ornament for an approaching predator.

Objective and Hypotheses

The objective of this study was to examine if the parental breeding tubercles indicate quality of offspring in whitefish (*Coregonus lavaretus*). In order to achieve this objective, we conducted an experiment using newly hatched whitefish larvae and brown trout in an attempt to determine if offspring from parents whose breeding tubercles are big are better in their quality.

The following hypotheses were put forward:

1. In the families where the paternal breeding tubercles are elaborate, the larvae are better in their quality (predator avoidance).
2. In the families where the paternal breeding tubercles are elaborate, the survival rate of the larvae is higher (when exposed to bacterial infection).

MATERIAL AND METHODS

Breeding design

Parental whitefish were captured from River Oulujoki while they were descending to their breeding grounds in September 2009. At the University of Oulu, the fish were striped for milt and eggs and examined microscopically for parasites in the laboratory. In addition, the fish were examined by hand for lateral breeding tubercle ornamentation (skin roughness, classified from 0-6, i.e. smooth skin to very rough skin, respectively; variable BT class) and the number of breeding tubercle on the front head was counted (variable BT front). Furthermore, a plastered cast was made on the lateral sides of fish to be used for breeding tubercle height measurements (variable BT height). In addition, the number of breeding tubercles on the side of fish was counted along a line descending from the front edge of the dorsal fin (variable BT side).

Table 1. Parental whitefish, breeding tubercle ornamentation and parasites

Sex	N	Mass (g)	Length (cm)	BT_ height	BT_ class	BT_ front	BT_ side	Dipl_ lens	Dipl_ vb	Dipl_ tot	Icht
Males	1	455	37.9	0.39	4.00	82	255	18	3	21	0
	2	396	38.6	0.32	4.00	250	293	3	1	4	1
	3	500	39.0	0.44	5.00	190	317	4	1	5	2
	4	503	39.5	0.37	4.00	60	309	5	3	8	0
	5	556	40.5	0.41	5	194	276	2	3	5	0
	6	497	39.2	0.36	4	178	244	4	0	4	12
	7	472	39.2	0.35	4	98	294	8	3	11	3
	8	472	39.9	0.39	6	122	238	1	1	2	1
	9	527	37.3	0.32	4	124	321	7	0	7	1
	10	519	39.9	0.32	6	344	271	2	1	3	8
Females	1	660	41.3	0.40	3	8	302	3	5	8	281
	2	491	40.4	0.21	3	42	172	2	1	3	0
	3	515	42.1	0.12	3	7	57	5	1	6	0
	4	522	41.2	0.16	3	0	193	2	3	5	1
	5	479	36.9	0.21	2	24	230	2	0	2	1
	6	533	41.3	0.08	0	9	0	6	0	6	4
	7	427	36.9	0	0	0	12	1	1	2	0
	8	452	40.6	0.09	1	6	20	5	1	6	0
	9	473	40.0	0.25	3	3	191	10	1	11	0
	10	434	37.0	0.25	4	52	167	1	1	2	2

Ten males and ten females (Table 1) with varying breeding tubercle ornamentation were chosen to breed in all possible combinations. Fertilized eggs were maintained at the University of Joensuu until they hatched. Shortly after hatching the larvae were transported to Konnevesi Research Station, University of Jyväskylä, in February 2010.

Predator avoidance experiment

A white-fish larva was moved into a Petri dish containing 10ml of water and its swimming activity was studied for two minutes. The white-fish was then moved to a trout box (a plastic 15 L container where was a young brown trout) and its activity was monitored for four minutes. From the white-fish, the total activity time, time by the side (less than 1cm from the side) and the time on the bottom of the box, as well as freezing

were record by means of stop watches. The brown trouts were placed into the trout boxes 12 hours before the start of the experiment for it to acclimatize in its new environment and water flow to the trout box was stopped before the experiment was done so that the behavior of the white fish could be well studied. For each family, 5 replicate whitefish larvae were used. The experiment carried out in the petri dish without the brown trout served as the control for general activity of the larva. Predator avoidance experiment was performed in 3-4 °C temperature.

Swimming on the side of the trout box was considered better predator avoidance than swimming on the bottom of the tank because trout preferred to stay on the bottom. Furthermore, freezing was considered a good predator avoidance as a swimming fish is more conspicuous in an environment with no hiding places as the present trout box. For the original variables, requirements for ANOVA were not met. Therefore a combination (predator avoidance behavior index, PABI) was calculated by adding the total activity time weighed by 0.2 to the time spent at the bottom weighed by 0.2. This way PABI actually decreases as predator avoidance increases.

Survival experiment

For the survival experiments, the statistical unit was a single petri dish, which contained 10 larvae in the beginning. There were 3 replicates per family (3 dishes per family = 3 values per family), a total of 300 dishes. Survival of larvae was monitored for 35 days. The temperature was increased 1°C per week, i.e. from 5 to 12°C during the experiment. Larvae were not fed during the experiment. The proportion of survived larvae at the end was calculated for each dish. 3 dishes out of 300 were lost, so we used the mean value of the other 2 replicates of those families as survival value for those dishes.

Statistical Analyses

Logarithm of the Predator Avoidance Behavior Index (PABI) was used in two-way ANOVA (SPSS: General Linear Model) for the swimming performance experiment. Arcsin transformed values were used in two-way ANOVA (SPSS: General Linear Model, Univariate Anova), for the survival data. Family-specific values were used as response variables and sex of parents as factor. In addition, separate one-way ANOVAs or non-parametric tests for males and females were performed on the survival data. Correlation analysis was employed to study the dependence between the breeding tubercle measures and mean predator avoidance behavior index, mean survival and parasitological variables.

RESULTS

Predation avoidance behavior

Results of ANOVA indicated that the male effect (sire, paternal effect) was not significant (Table 2). Thus, there was no difference between males in the predation avoidance behavior of offspring. Similarly, the female effect (mother, maternal effect) was also not significant (Table 2), indicating that the mean score of predator avoidance behaviour did not differ between females either. These findings were not in accordance with the good genes hypothesis. On the contrary, there was a significant male-female interaction. This implied that differences between males and females exist, but they depend on the combination, i.e. which female is mating with which male. This result was in accordance with compatible genes hypothesis.

Table 2. ANOVA statistics for Predator Avoidance Behavior Index (PABI)

Source	Type III Sum Of Squares	df	Mean Square	F	Sig
Corrected Model	3.36 ^a	99	0.034	1.445	0.008
Intercept	114.988	1	114.988	4895.014	0.000
Male	0.375	9	0.042	1.773	0.072
Female	0.298	9	0.033	1.409	0.182
Male * Female	2.687	81	0.033	1.412	0.017

a. R Squared = ,263 (Adjusted R Squared = ,081)

Family-specific scores of predator avoidance behavior are given in Fig. 1. For the male-female interaction for example, female10 had the best predator avoidance behaviour (lowest PABI value) when bred with male5, but poor avoidance behaviour (high PABI values) when bred with male3 or male10. In general, male number 4 showed the highest mean PABI (poorest avoidance behaviour) male number 9 had the lowest mean score (best avoidance). In the female category, female3 was the poorest while female5 was the best in their avoidance behaviour when assessed over all males (Fig. 1)

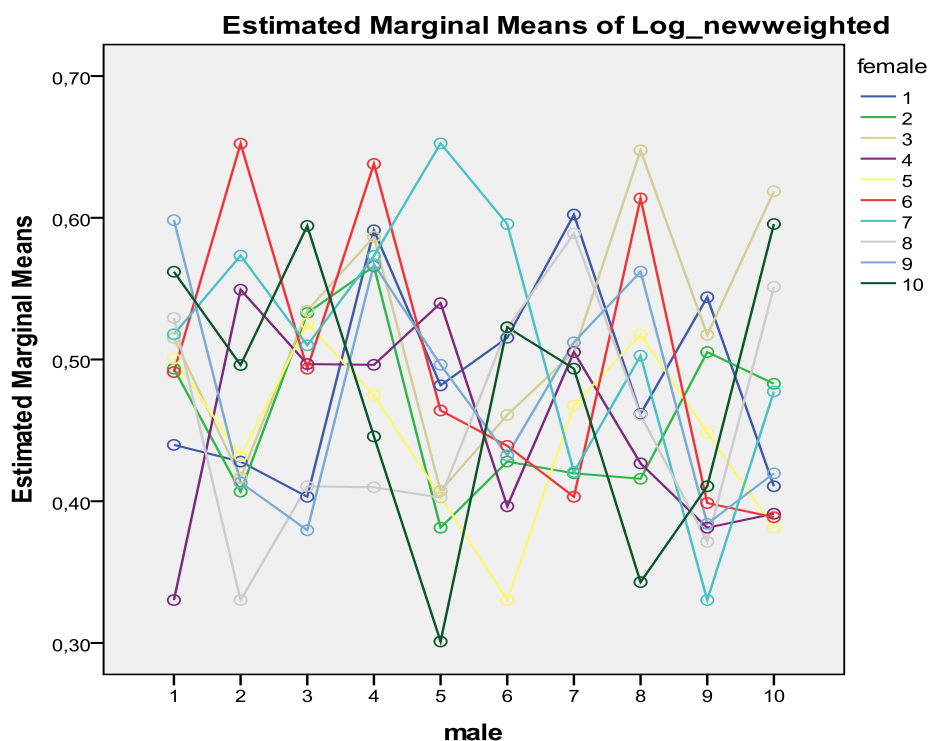


Figure 1. Mean family-specific, logarithm-transformed scores of predator avoidance behavior. Note that lower score means better avoidance behavior.

Off spring survival

Results of ANOVA on the survival of larvae indicated that the male effect (sire, paternal effect) was significant (Table 3). This means such differences between males that are consistent over all females. The finding was in accordance with good genes hypothesis. Similarly, the female effect (mother, maternal effect) was also significant (Table 3), indicating females differ in their offspring survival. This result is also in accordance with good genes hypothesis. But heterogeneity of variances may be a problem which can make $p = 0.045$ questionable. However, the male-female interaction was NOT significant as the differences between males and females did not depend on the combination, which female mated with which male. This is in contradiction with compatible genes hypothesis.

From Table 2, the male effect (sire, paternal effect) was significant meaning differences between males exist, $p = 0.001$ (such differences that are consistent over all females). This finding was in accordance with good genes hypothesis. Similarly, the female effect (mother, maternal effect) was significant. This implied that differences between females exist $p = 0.045$ (such differences that are consistent over all females). differs from all other females except for 3.

Table 3. ANOVA statistics for survival of whitefish larvae.

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	28.199 ^a	99	.285	1.278	.074
Intercept	191.941	1	191.941	860.990	.000
Male	6.714	9	.746	3.346	.001
Female	3.946	9	.438	1.967	.045
Male x Female	17.539	81	.217	.971	.551

a. R Squared =0,387 (Adjusted R Squared =, 084)

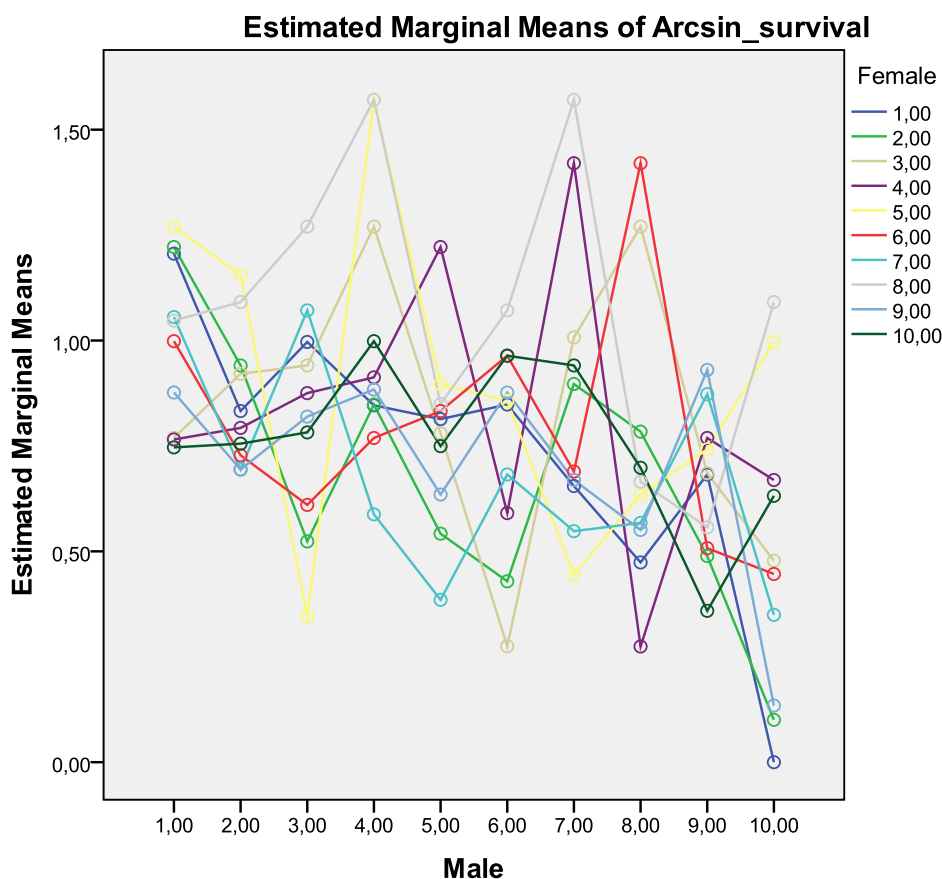


Figure 2. Mean family-specific, arcsin-transformed survivals of whitefish larvae

Parental breeding tubercle measures

Correlations were analysed for the ten males and ten females on the following measures: BT height (mean height of breeding tubercles on the side), BT front (number of breeding tubercles on front head), BT class (breeding tubercles ornamentation on lateral side classified by hand (0-6)) and BT side (number of breeding tubercle on the side) and the number of *Diplostomum* in the lense, *Diplostomum* in vitreum body, total number of *Diplostomum*, number of *Ichthyocotylurus variegates* in body cavity, total number of parasite individuls and number of parasite species of the parental fish, and survival (arcsin) and predation avoidance behavior index of offspring.

Table 4. Parental breeding tubercle and the following parasitological parameters: BT height (mean height of breeding tubercles on the side), BT front (number of breeding tubercles on front head), BT class (breeding tubercles ornamentation on lateral side classified by hand (0-6)) and BT side (number of breeding tubercle on the side) and the number of *Diplostomum* in the lense, *Diplostomum* in vitreum body, total number of *Diplostomum*, number of *Ichthyocotylurus variegates* in body cavity , total number of parasite individuls and number of parasite species of the parental fish, as well as survival and predator avoidance behaviour score.

	Males			Females	
	Mean	Std. Deviation		Mean	Std. Deviation
BT_height (mm)	0.37	0.04	10	0.18	0.11
BT_front	164.2	86.17	10	15.1	8.27
BT_class	4.6	0.84	10	2.2	1.39
BT_side	281.8	29.82	10	134.4	104.59
Survival	0.63	0.11	10	0.63	0.07
Diplo_lens	5.4	4.95	10	37	2.83
Diplo_vb	1.6	1.26	10	14	1.51
Diplo_tot	7	5.58	10	51	2.96
Ichthyocot	2.8	4.02	10	28.9	88.59
Parasite_n	12.6	5.5	10	39.5	90.68
Parasite_sp	4.3	0.95	10	4	1.41
Swimming	0.48	0.03	10	0.48	0.03

In males, all the BT measures were independent, not correlated to each other. BT front was negatively correlated with offspring survival. In the same way, BT class was negatively correlated with offspring survival (Table 5). Negative association between the number of breeding tubercles on the front head of males and survival of offspring, as well as the negative association between the lateral skin roughness and survival of offspring can be seen in Fig. 3.

Table 5. Parental males. Results of Pearson correlation analyses between different breeding tubercle measures and their relationship to offspring survival (Surv), offspring predator avoidance index (Swim), and the following parasitological parameters; number of *Diplostomum spathaceum* in eye lens (Diplo lens), Number of diplostomatid parasites in vitreum body of eye (Diplo vb), total number of diplostomatids in eyes (Diplo tot), logarithm-transformed numbers of *Ichthyocotylurus variegatus* parasites in body cavity (Log Icht), total number of parasite individual (Paras num) and total number of parasite species (Para taxa n) in fish and Index of predation avoidance behavior (PABI). Values given in upper row, middle row and lower row are correlation coefficient, significance and n studied, respectively. For BT class, non-parametric Spearmann correlation analysis was used. Statistically significant correlations are given in bold.

	BT height	BT front	BT class	BT side	Diplo lens	Dipl Vb	Diplo tot	Log Icht	Para num	Para taxa n	Surv	PABI
BT height	1	-0.3	0.32	-.093	0.06	0.34	0.13	-0.29	-0.04	0.28	0.39	0.33
		0.39	0.37	0.27	0.88	0.34	0.73	0.41	0.91	0.43	0.27	0.36
	10	10	10	10	10	10	10	10	10	10	10	10
BT front		1	0.47	-0.08	-0.53	0.43	-0.56	0.47	-0.29	0.07	-0.73	-0.43
			0.17	0.82	0.118	0.22	0.09	0.17	0.42	0.87	0.017	0.21
		10	10	10	10	10	10	10	10	10	10	10
BT class			1	-0.33	-0.79	-0.06	-0.67	0.14	-0.44	0.39	-0.63	.000
				0.351	0.007	0.87	0.036	0.69	0.2	0.27	0.049	1.000
			10	10	10	10	10	10	10	10	10	10
BT side				1	-0.02	0.04	-0.01	-0.39	-0.34	-0.1	0.19	-0.06
					0.95	0.92	0.98	0.26	0.33	0.78	0.74	0.87
				10	10	10	10	10	10	10	10	10

In females, breeding tubercle measures were inter-dependent so that BT class and BT side correlated statistically significantly with BT height (Table 6). None of the breeding tubercle measures correlated with survival or predator avoidance of offspring, but BT height correlated significantly positively with the total numbers of parasite individuals and number of parasite species found in the fish. In addition, the number of parasite species found was significantly positively correlated also to BT class and BT side. Furthermore, and there was a marginally significant, positive correlation with BT side and the total number of parasite individuals (Table 6).

Table 6. Parental females. Results of Pearson correlation analyses between different breeding tubercle measures and their relationship to offspring survival (Surv), offspring predator avoidance index (Swim), and the following parasitological parameters; number of *Diplostomum spathaceum* in eye lens (Diplo lens), Number of diplostomatid parasites in vitreum body of eye (Diplo vb), total number of diplostomatids in eyes (Diplo tot), number of *Ichthyocotylurus variegatus* parasites in body cavity (Icht), logarithm-transformed total number of parasite individual (Log para n) and total number of parasite species (Para taxa n) in fish and Index of predation avoidance behavior (PABI). Values given in upper row, middle row and lower row are correlation coefficient, significance and n studied, respectively. For BT class and number of *Ichthyocotylurus variegatus* parasites in body cavity (Icht), non-parametric Spearman correlation analysis was used. Statistically significant correlations are given in bold. Of the breeding tubercle measures, values of BT front were logarithm-transformed, as well as the total number parasite individual, to achieve normal distributions.

	BT height	Log BT front	BT class	BT side	Diplo lens	Dipl Vb	Diplo tot	Icht	Log Para n.	Para taxa n	Surv	PABI
BT height	1	0.43	.783	.908	0.03	0.6	0.34	0.36	0.78	0.92	-0.21	-0.32
		0.21	0.007	<.001	0.93	0.07	0.34	0.31	0.008	<.001	0.56	0.37
	10	10	10	10	10	10	10	10	10	10	10	10
Log BT front		1	0.34	.257	-0.14	-0.27	-0.27	0.39	0.03	0.09	0.03	-0.23
		10	0.34	.473	0.7	0.45	0.45	0.27	0.95	0.79	0.94	0.52
BT class			1	0.55	-0.17	0.49	0.07	0.13	0.17	0.65	-0.07	-0.12
				0.103	0.64	0.14	0.86	0.73	0.64	0.04	0.86	0.73
			10	10	10	10	10	10	10	10	10	10
BT side				1	-0.15	0.58	0.15	0.29	0.61	0.89	-0.24	-0.49
					0.68	0.08	0.66	0.4	0.06	0.001	0.49	0.15
				10	10	10	10	10	10	10	10	10

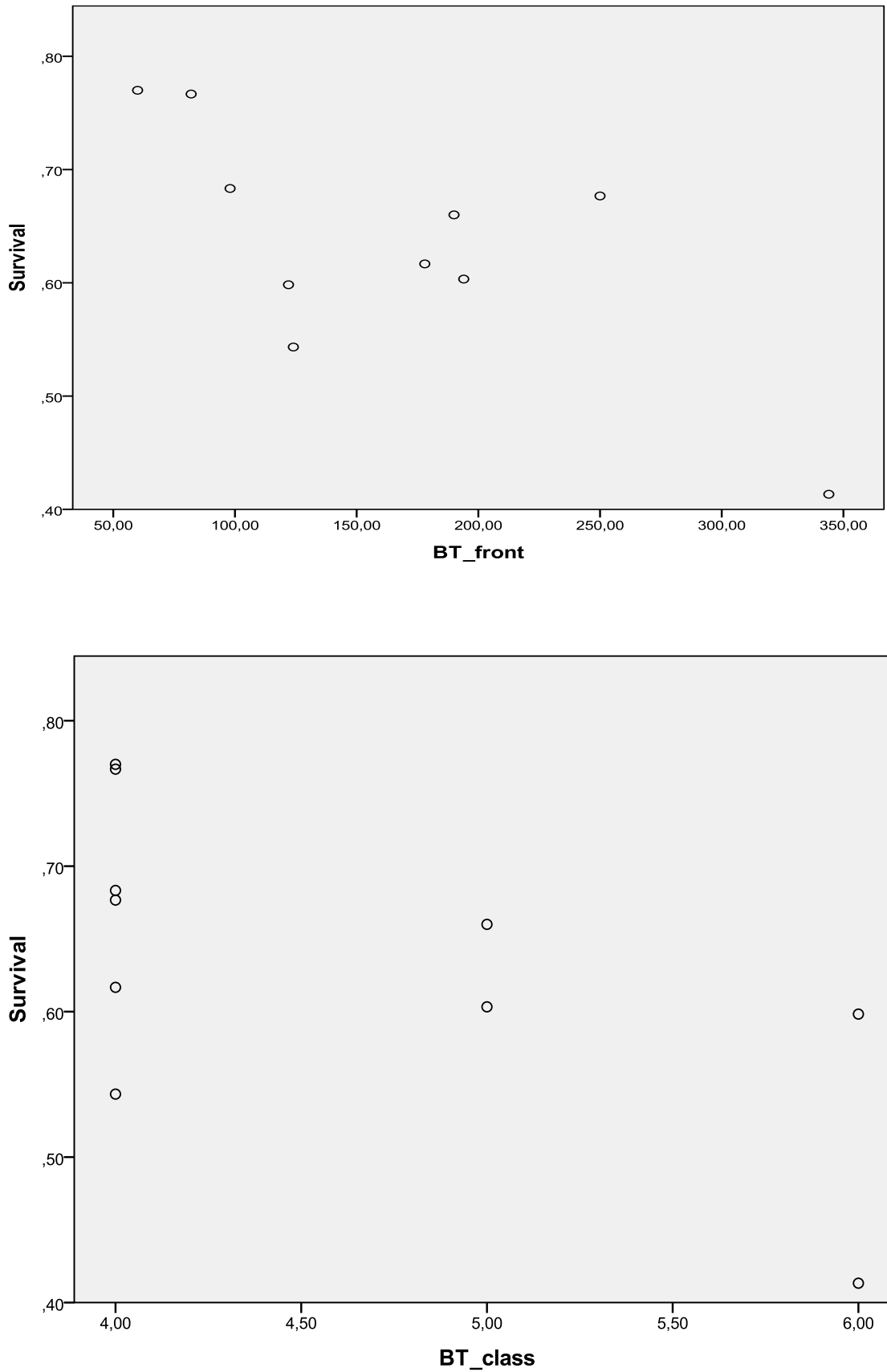


Fig. 3. The relationship between off spring survival to number of front head breeding tubercles (BT_front, upper picture) and to lateral skin roughness (BT_class, lower picture) in males.

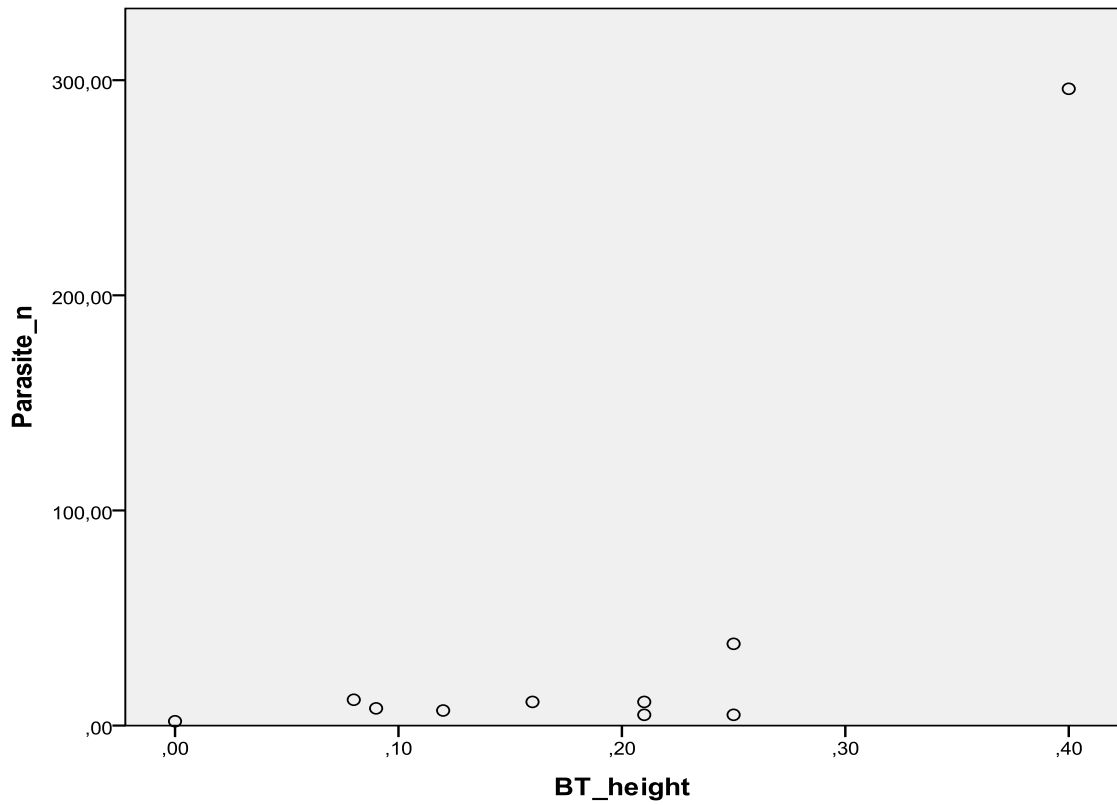


Fig. 4: The relationship between parasite number and breeding tubercle height in male whirefish.

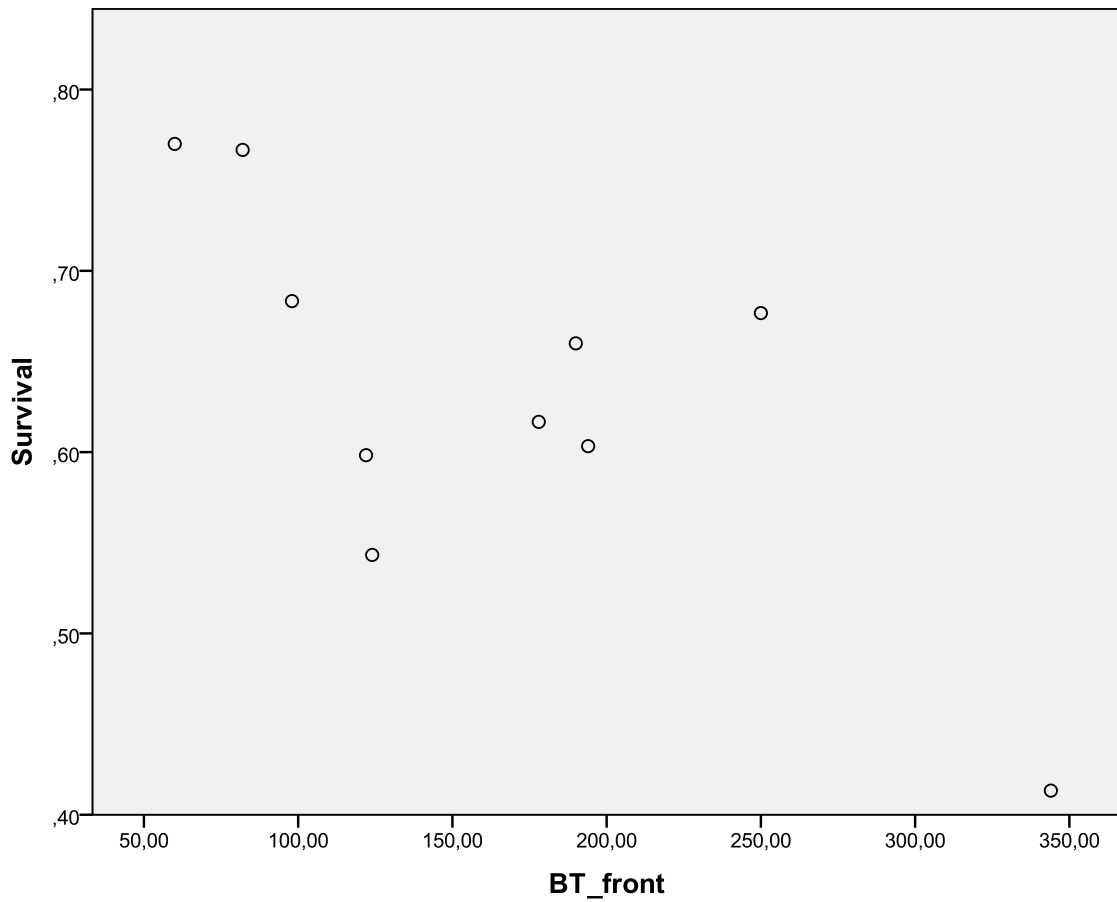


Fig.5: The relationship between off spring survial and front head breeding tubercle

DISCUSSION

The good gene hypothesis predicts that males advertise their quality with different sexual ornaments and that females are able to recognise the genetic quality of males by evaluating these characteristics. The survival of larvae was affected by the male effect only, as the maternal effect and male-female interactions were not significant. Findings are in accordance with the good genes hypothesis.

Also, the compatible gene hypothesis predicts that the variation in families in terms of offspring quality is not straight forward with respect to parental breeding tubercle ornamentation. There was a significant male x female interaction in predation avoidance behavior of whitefish larvae, since offspring quality depended on the male-female combination. This result was in accordance with the compatible gene hypothesis and are similar to results by Kekäläinen *et al.*, 2010 who found a statistically significant male x female interaction in the offspring swimming performance experiments, but different in that the male and female effects were also statistically significant.

There was also a negative correlation both between the number of breeding tubercles on the front head and offspring survival and between lateral breeding tubercle roughness class and offspring survival in males. Thus, breeding tubercle ornamentation of male whitefish seems not to indicate offspring survival in laboratory conditions. Also, the correlation between BT front and survival indicated that the higher the number of breeding tubercles on the front head, the lower was survival of offspring and the higher the breeding tubercles on the side of the fish, the lower was the survival of offspring. Females could choose males if they want to increase the probability of offspring survival under such circumstances as in our experiment. Females should choose males with a low number of breeding tubercles on front head and on the side. There has to be a selective advantage of choosing males or females with elaborate breeding tubercles, otherwise the tubercles will disappear in the long run.

In females, BT height was positively correlated to total parasite number and parasite species number. However, there was no correlation between this breeding tubercle measure and survival. Generally, in females, there was no connection between offspring survival and ornamentation, whereas in males, there was a relationship between these two parameters. Males should think that females with high breeding tubercles are of low quality, because it has got lots of parasites. Also, males could think that females with high breeding tubercles are of good quality, since they are able to produce elaborate ornaments. From the evolution of breeding tubercles, in order to maintain this ornament when there is a positive correlation between breeding tubercle height and parasites, there should be a selection against these ornaments. This can be possible only if these fish trade off parasites for immune defence as stipulated by the Immunocompetence handicap hypothesis (which states that testosterone is needed to produce sexual ornaments, but on the other hand, testosterone is an immune suppressive substance. This means that an individual with high breeding tubercles will produce more testosterone, which will suppress its immune system).

In males, the evolution of breeding tubercles could not be explained due to the negative correlation between the breeding tubercle measures and parasites. This did not support the good gene hypothesis.

The predator-avoidance experiments should be repeated using the same methods described in the material and methods section, using brown trouts which are used to feed on whitefish larvae, so that the results could be compared with the situation in the wild. Another reason for repeating this experiment is probable because we did the observations by standing closer to the trout box, which had an effect on the behaviour of the trout and

the white-fish larvae. The alarm system on the stop watches used might also have influenced the behaviour of the trout and white-fish larva. This experiment could be repeated using an inbuilt video system connected to a monitor for the observations and timing to be done without actually influencing the behaviour of the fish in any way.

Our behaviour experiment did not actually measure the fitness of the offspring due to the need to repeat that same experiment as explained above. On the other hand, our survival experiment did measured offspring fitness in my opinion as the situation in the petri dishes and trout box could be compared with the situation in the wild.

CONCLUSION

In our study, we observed that in males as the number of breeding tubercles on the front head and that on the side increases, survival of offspring decreases. In this respect, we conclude that females should choose males with a decreased number of front head and side breeding tubercles so as to increase the chances of offspring survival.

From the observed negative correlation between the breeding tubercle measures and parasites, only bad genes will be favoured instead of the good genes if elaborate breeding tubercles are selected. This is probably the case as our results were not in support of the good genes hypothesis.

ACKNOWLEDGEMENTS

My gratitude goes to my supervisors Prof. Jouni Taskinen and Dr. Jukka Kekäläinen for their guidance, countless discussions and criticisms at every stage of this piece of work. I express my appreciation to Prof. Raine Kortet in the very beginning of the experiments at the Konneversi Research station.

I wish to acknowledge the contributions of Niko and Sarah who assisted greatly with the experiments.

I owe special thanks to my parents, brothers, sister, other family relatives and friends whose names have not been mentioned herein for their support throughout this work.

Finally, I thank God Almighty for his endless love and comfort throughout this period.

REFERENCES

- Admundsen, T. 2000. Why are female birds ornamented? *Trends in Ecology and Evolution* **15**:149-155.
- Andersson, M. 1994. *Sexual Selection*. Princeton, NJ: Princeton University Press.
- Bailey, K. M., and Houde E. D. 1989. Predation on eggs and larvae of marine fishes and the recruitment problem. *Advances in Marine Biology* **25**:1–83.
- Berglund, A., Rosenqvist, G., Bernet, P. (1997) Ornamentation predicts reproductive success in female pipefish. *Behavioural Ecology and Sociobiology* **40**:145-150.
- Clutton-Brock, T. 2009. Sexual selection in females. *Animal Behaviour* **77**:3-11.
- Esther, I. and Hamilton, W.D. 1984. Parent-offspring correlation in fitness under fluctuating selection. *Proc. R. Soc. Lond. B* **222**: 1-14.
- Evans, J.P, Kelley, J.L, Bisazza, A., Finazzo, E., Pilastro, A. 2004. Sire attractiveness influences offspring performance in guppies. *Proceedings of the Royal Society of London Series B, Biological Sciences* **271**: 2035–2042.
- Fisher, R.A. 1930. *The genetic theory of natural selection*. Clarendon Press, Oxford.
- Fitzpatrick, S., Berglund, A., Rosenqvist, G. (1995) Ornaments or Offspring: costs to reproductive success restrict sexual selection processes. *Biological Journal of the Linnean Society* **55**:251-260.
- Fuiman, L.A, Cowan, J.H., Jr (2003) Behavior and recruitment success in fish larvae: repeatability and covariation of survival skills. *Ecology* **84**:53-56.
- Fuiman, L. A. 2002. Special considerations of fish eggs and larvae. Pages 1–32.
- Fuiman, L.A. and R. G. Werner, editors. *Fishery science: the unique contributions of early life stages*. Blackwell Science, Oxford, UK.
- Fuiman, L. A. and A. E. Magurran. 1994. Development of predator defenses in fishes. *Reviews in Fish Biology and Fisheries* **4**:145–183.
- Hamilton, W.D. & Zuk, M. 1982. Heritable true fitness and bright birds: a role for parasites? *Science* **218**: 384–387.
- Hammer, C. 1995. Fatigue and exercise tests with fish. *Comparative Biochemistry and Physiology* **112A**: 1–20.
- Houde, E. D. 1989. Subtleties and episodes in the early life of fishes. *Journal of Fish Biology* **35** :(Supplement A) 29–38.
- Huuskonen, H., Haakana, H. and Kekäläinen, J. 2009. Offspring performance is linked to parental identity and male breeding ornamentation in whitefish. *Biological Journal of the Linnean Society* **98**: 532–539.
- Kekäläinen, J., Huuskonen, H., Tuomaala, M., Kortet, R. 2010. Both male and female sexual ornaments reflect offspring performance in a whitefish. *Evolution* **64**:3149-3157.
- Kirkpatrick, M. 1982. Sexual selection and the evolution of female choice. *Evolution* **30**: 1-12.
- Kirkpatrick, M. and Ryan, M.J. 1991. The evolution of mating preference and the paradox of the lek. *Nature* **350**: 33-38.
- Kortet, R., Vainikka, A., Rantala, M., Jokinen, I. and Taskinen, J. 2003. Sexual ornamentation, androgens and papillomatosis in roach (*Rutilus rutilus*). *Evolutionary Ecology Research* **5**: 411–419.

- Kortet, R., Taskinen, J., Vainikka, A. and Ylönen, H. 2004b. Breeding tubercles, papillomatosis and dominance behaviour of male roach (*Rutilus rutilus*) during the spawning period. *Ethology* **110**: 591–601.
- Kortet, R., Vainikka, A., Rantala, M.J., Myntti, J., and Taskinen, J. 2004a. In vitro embryo survival and early viability of larvae in relation to male sexual ornaments and parasite resistance in roach, *Rutilus rutilus* L. *Journal of Evolutionary Biology* **17**: 1337–1344.
- Kraaijeveld, K., Kraaijeveld-Smith, F.J.L., Komdeur, J. 2007. The evolution of mutual ornamentation. *Animal Behaviour* **74**:657-677.
- Lande, R. 1980. Sexual dimorphism, sexual selection, and adaptation in polygenic characters. *Evolution* **34**:292-305.
- Lande, R. 1981. Models of speciation by sexual selection on polygenic traits. *Proc. Natl. Acad. Sci. USA* **78**: 3721-3725.
- Maynard Smith J. and Harper D. 2003. *Animal signals*. Oxford: Oxford University Press.
- Møller, A.P. and Alatalo, R.V. 1999. Good-genes effects in sexual selection. *Proc. R. Soc. Lond. B* **266**: 85–91.
- Neff, B.D. and Pitcher, T.E. 2005. Genetic quality and sexual selection: an integrated framework for good genes and compatible genes. *Molecular Ecology* **14**: 19-38.
- Nicoletto, P.F. 1991. The relationship between male ornamentation and swimming performance in the guppy, *Poecilia reticulata*. *Behavioral Ecology and Sociobiology* **28**: 365–370.
- Pomainkowski, A. and Iwasa, Y. 1998. Runaway ornament diversity caused by Fisherian Sexual selection. *Proc. Natl. Acad. Sci. USA* **95**: 5106-5111.
- Pomainkowski, A., Iwasa, Y. and Nee, S. 1991. The evolution of costly mate preferences. I. Fisher and biased mutation. *Evolution* **45**: 1422-1430.
- Rudolfsen, G., Müller, R., Urbach, D., Wedekind, C. 2008. Predicting the mating system from phenotypic correlations between life-history and sperm quality traits in the Alpine whitefish *Coregonus zugensis*. *Behavioral Ecology and Sociobiology* **62**: 561–597.
- Sheldon, B.C., Arponen, H., Laurila, A., Crochet, P.A., Merilä, J. 2003. Sire coloration influences offspring survival under predation risk in the moorfrog. *Journal of Evolutionary Biology* **16**: 1288–1295.
- Siebenthal, V.A.B, Alain J. and Wedekind, C. 2009. Tolerance of whitefish embryos to *Pseudomonas fluorescens* linked to genetic and maternal effects, and reduced by previous exposure. *Fish & Shellfish Immunology* **26**: 531–535.
- Sullivan, K.A. 1989. Predation and starvation: age-specific mortality in juvenile juncos (*Junco phaeotus*). *Journal of Animal Ecology* **58**: 275–286.
- Taborsky, M. 1994. Sneakers, satellites, and helpers: parasitic and cooperative behavior in fish reproduction. *Adv. Study Behav.* **23**: 1–100.
- Watson, N.L., Simmons, L.W. 2010. Reproductive competition promotes the evolution of female weaponry. *Proceedings of the Royal Society of London Series B, Biological Sciences* **277**:2035-2040.
- Ward, J.M, Kennedy PL. 1996. Effects of supplemental food on size and survival of juvenile Northern Goshawks. *The Auk* **113**: 200–208.
- Wedekind, C. 1992. Detailed information about parasites revealed by sexual ornamentation. *Proc. R. Soc. Lond. B* **247**: 169–174.
- Wedekind, C. 1996. Lek-like spawning behaviour and different female mate preferences in roach (*Rutilus rutilus*). *Behaviour* **133**: 681–695.

- Wedekind C., Müller R. and Spicher H. 2001. Potential genetic benefits of mate selection in whitefish. *Journal of Evolutionary Biology* **14**: 980–986.
- Wedekind, C. and Müller, R. 2004. Parental characteristics versus egg survival: towards an improved genetic management in the supportive breeding of lake whitefish. *Ann. Zool. Fennici* **41**: 105–115.
- Wedekind, C., Walker M., portmann J., Cenni B., Müller, R. and Binz T. 2004. MHC-linked susceptibility to a bacterial infection, but no MHC-linked cryptic female choice in whitefish. *Journal of Evolutionary Biology* **17**: 11–18.
- Wedekind C., Evanno, G., Urbach, D., Jacob, A. and Müller, R. 2008. ‘Good-genes’ and ‘compatible-genes’ effects in an Alpine whitefish and the information content of breeding tubercles over the course of the spawning season. *Genetica* **132**: 199–208.
- Weiss, S.L., Kennedy, E.A., Bernhard, J.A. 2009. Female- specific ornamentation predicts offspring quality in the striped Plateau lizard, *Sceloporus virgatus*. *Behavioural Ecology* **20**:1063-1071.
- Westneat, D.F. and Birkhead, T.R. 1998. Alternative hypothesis linking the immune system and mate choice for good genes. *Proc. R. Soc. Lond. B* **265**: 1065-1073.
- Welch A.M., Semlitsch, R.D., Gerhardt, H.C. 1998. Call duration as an indicator of genetic quality in male gray tree frogs. *Science* **280**:1928–1930.
- Wiley, M.L. and Collette, B.B. 1970. Breeding tubercles and contact organs in fishes: their occurrence, structure, and significance. *Bulletin of the American Museum of Natural History* **143**:145–216.
- Ylöneni, H., Kortet, R., Myntti, J. and Vainikka, A. 2007: Predator odour recognition and antipredatory response in fish: Does the prey know the predator diel rhythm? *Acta Oecologica*, **31**: 1-7.
- Zahavi A. 1975. Mate selection – a selection for handicap. *J. Theor. Biol.* **53**: 205–214.