4 00387

UNIVERSIDAD NACIONAL AUTÓNOMA DE MÉXICO

POSGRADO EN CIENCIAS
BIOLÓGICAS

INSTITUTO DE ECOLOGÍA

LA ELECCIÓN DE PAREJA EN LOS GOODEIDOS: DIFERENCIAS DENTRO Y ENTRE ESPECIES

T E S I S

QUE PARA OBTENER EL GRADO ACADÉMICO DE

DOCTOR EN CIENCIAS

P R E S E N T A

ALEJANDRO CLEMENTE MOYAHO MARTÍNEZ

DIRECTOR DE TESIS: DR. CONSTANTINO MACÍAS GARCÍA

MÉXICO, D.F.   SEPTIEMBRE 2002

POSGRADO EN CIENCIAS BIOLÓGICAS
INSTITUTO DE ECOLOGIA

VNIVERSIDAD NACIONAL
AVTONOMA DE
MEXICO

ING. LEOPOLDO SILVA GUTIERREZ
Director General de
Administración Escolar, UNAM
P r e s e n t e.

Por la presente me permito informar a usted que en reunión ordinaria del Comité Académico del Posgrado en Ciencias Biológicas, celebrada el 8 de abril de 2002, se acordó poner a su consideración el siguiente jurado para el examen de grado de Doctor en Ecología del alumno: **ALEJANDRO CLEMENTE MOYAHO MARTINEZ** con No. de Cta. 96800485 Exp. 30961100 con la tesis titulada: "La elección de pareja en los Goodeidos: Diferencias dentro y entre especies", bajo la dirección del: **DR. CONSTANTINO MACIAS GARCIA.**

Presidente: Juan José Morrone Lupi
Secretario: Constantino Macías García
Vocal: Guillermina Alcaráz Zubeldía
Vocal: Alejandro Córdoba Aguilar
Vocal: Carlos Cordero Macedo
Suplente: José Luis Osorno Cepeda
Suplente: Oscar Flores Villela

Sin otro particular quedo de usted.

A t e n t a m e n t e.
"POR MI RAZA HABLARA EL ESPÍRITU"
Cd. Universitaria, D. F., 26 de agosto de 2002

DRA. TILA MARIA PEREZ ORTIZ
Coordinadora

AGRADECIMIENTOS

RESUMEN

Los goodeidos son peces vivíparos de agua dulce, nativos de la Meseta Central de México, cuyos machos requieren de la cooperación de la hembra para aparearse. Como consecuencia, los machos compiten continuamente por tener acceso a las hembras, lo que parece haber promovido la evolución de caracteres sexuales dimórficos. Puesto que el hábitat de los goodeidos varía de ríos a lagos, existe la posibilidad que la variación de caracteres masculinos conspicuos sea el resultado de adaptaciones a condiciones locales, propias de los lugares que habitan.

Aquí evalué la función de la selección natural en la presencia de caracteres sexualmente seleccionados en los goodeidos. Para evaluar esta fuerza selectiva, sobrepuse los estados de caracteres sexuales dimórficos en un arreglo sistemático de la familia Goodeidae. Luego conté el número de aquellos eventos que evolucionaron independientemente y probé si estaban asociados con aguas corrientes o con lagos. Además analicé, por una parte, la relación entre concurrencia (i.e., simpatría de las especies) y la edad nodal (i.e., parentesco), y por otra, el contraste de hábitat o cuenca y el grado de concurrencia. Predije que el grado de concurrencia crecería con la edad nodal, y que los contrastes en hábitat o cuenca aumentarían con la concurrencia.

Otro tema de investigación fue las causas de la variación de caracteres sexuales secundarios entre poblaciones coespecíficas. Estudié 4 poblaciones de Xenotoca variata cuyos machos presentan una amplia variación en la cantidad de escamas iridiscentes que lucen como espejos. El número de los espejos por población tiende a correlacionar, de manera positiva, con la transparencia del agua, y de forma negativa con la cantidad de culebras acuáticas Thamnophis melanogaster presentes en cada localidad. Comparé 4 poblaciones de peces para poner a prueba la hipótesis de que la variación en la fuerza de la elección de pareja de apareamiento por las hembras, contribuía a la variación geográfica en el número de espejos. Presenté a hembras pares de machos cuyos miembros diferían en la cantidad de espejos. Hice los experimentos con agua clara

3

y agua turbia para analizar sus efectos en la elección de pareja de apareamiento. Pares de peces macho que diferían en el número de espejos fueron presentados a culebras en agua clara y transparente.

Obtuve parte de los datos en el campo y parte en el laboratorio a través de experimentos controlados. También recabé datos de la información publicada.

No encontré asociaciones significativas entre los caracteres sexuales dimórficos y los tipos de hábitat, aunque todos los caracteres, excepto uno, aparecieron en peces que viven en aguas corrientes. También encontré una tendencia de las especies más dimórficas, en su tamaño relativo de aletas dorsales, a provenir de géneros con el mayor número de especies. La concurrencia y la edad nodal no estuvieron relacionadas en ninguno de los dos clados principales de la familia Goodeidae. Tampoco el contraste de hábitat y la concurrencia se relacionaron significativamente. En el caso del contraste de cuenca, hubo una correlación negativa con el grado de concurrencia en uno de los clados: aquél cuyas especies habitan mayoritariamente la cuenca del río Lerma.

Las hembras de San Francisco del Rincón y las de Rancho Viejo, que fueron las localidades con los machos con más espejos, pasaron más tiempo con los machos con espejos que las hembras provenientes de los lagos de Zacapu y Cuitzeo. Las preferencias solo se expresaron en agua clara. Las culebras también prefirieron aproximarse a los machos con más espejos, en agua clara y turbia; sin embargo, la magnitud de la preferencia fue mayor en agua clara.

Los resultados sugieren que la participación de la adaptación ecológica en la variedad de especies de goodeidos es limitada. Por el contrario, la selección sexual parece haber tenido una función determinante. Aun así, no puedo desechar la participación probable de otros factores (e.g., deriva génica o mutación).

El estudio de caso de las poblaciones de Xenotoca variata mostró que la elección de pareja por las hembras y el riesgo de depredación explican la variación geográfica en el número de espejos. La expresión de éstos, no obstante, está influenciada por la transparencia del agua.

4

En conclusión, la selección sexual parece tener una función
determinante en la variedad de especies de la familia Goodeidae.

ABSTRACT

Goodeids are viviparous freshwater fish native to Central Mexico. The males lack an intromittent organ, requiring the cooperation of the females to copulate, so they are likely to live under circumstances of constant competition over the females. Female choice of mate seems to have driven evolution of sexually dimorphic traits in males. There is the possibility, on the other hand, that the geographical variation of conspicuous male traits in goodeids is the result of adaptation to local conditions, given the heterogeneity of habitats occupied by them.

I assessed the role of natural selection in sexually selected characters of goodeids. To investigate this selective force I mapped sexually dimorphic characters on a systematic arrangement of the Goodeidae family. I counted the evolutionary independent events, and tested whether they were associated with either of two contrasting types of habitat: running waters and lakes. In addition, I analysed the relationship between co-occurrence (i.e., sympatry of species) and age of node (i.e., relatedness), between habitat contrast and co-occurrence, and between drainage contrast and co-occurrence. I predicted that sympatric speciation would be the predominant geographical mode of speciation, and that habitat and drainage contrasts would increase with co-occurrence, because phenotypic differences would be necessary for sympatric species to coexist.

There were no significant associations between sexually dimorphic traits and any type of habitat, although all but one trait appeared in running water fish. There was also a slight trend for the most dimorphic species in dorsal fin size to form the largest genera, suggesting that sexual selection may have promoted speciation. The association between co-occurrence and node age suggested that the main geographical mode of speciation was sympatric in the two main clades of the Goodeidae family. Habitat contrast and co-occurrence were not significantly

associated. In the case of drainage contrast, there was a negative correlation with co-occurrence for one of the clades, that whose species mostly inhabit the Lerma basin. This correlation suggests that phenotypic differences occurred as a consequence of geographical variation in drainage conditions.

Another issue considered here was to assess the causes of the variation of secondary sexual traits within a species. I studied populations of Xenotoca variata in which males show a broad range of geographical variation in 'speckles'. The number of speckles may be associated with water turbidity and with the presence of predatory aquatic snakes, Thamnophis melanogaster. I compared 4 populations of fish to test whether variations in female choice of mate may have contributed to the geographical variation in speckles. Pairs of males which differed in the number of speckles were presented to females. The trials were conducted in clear and in turbid water to determine their effect on mate choice. Pairs of fish with more and with fewer speckles were also faced with snakes in clear and in turbid water.

Females from San Francisco del Rincón and Rancho Viejo, the localities in which males had more speckles, stayed more time next to speckled fish than did females from Lake Zacapu and Lake Cuitzeo. The preferences were expressed only in clear water. The snakes approached males with more speckles than males with fewer speckles, in both clear and turbid water. The magnitude of the preference, however, was greater in clear water.

The results suggest that the role of adaptation to local conditions in the diversity of goodeid species is limited. Sexual selection might have played a determining role in generating reproductive isolation. Yet, I cannot rule out the role of other factors (e.g., genetic drift and mutation).

The case study of populations of X. variata shows that female choice of mate and predation risk both contribute to the geographical variation in number of speckles. In addition, the

7

number of speckles could be affected by water turbidity.

In conclusion, the variation of sexually dimorphic traits of goodeids cannot be explained by adaptation to conditions of lakes and running waters. It is likely that sexual selection had contributed to the richness of goodeid species. The geographical variation in number of speckles seems to be the result of the interaction of sexual selection by female choice of mate with natural selection caused by predation, and limits set by water turbidity.

TABLA DE CONTENIDOS

CAPÍTULO 1

INTRODUCCIÓN

1. El método comparativo

¿Por qué las especies estrechamente relacionadas a menudo difieren en su conducta y morfología? Podríamos esperar que estas diferencias se originaran, por lo menos en parte, por el hábitat en el que evolucionaron. Ésta es una expectativa razonable puesto que la comunicación animal, siendo la base para las interacciones sociales, se basa en exhibiciones cuya transmisión depende en muchos sentidos del ambiente físico (Endler 1992). La expectativa de concordancia entre el hábitat y el fenotipo asume que los patrones geográficos actuales han permanecido inalterados, lo cual es improbable. Así que de algún modo tenemos que rastrear su historia evolutiva para poder apoyar aquellas relaciones probables entre la conducta y la morfología, por una parte, y las variables ecológicas por la otra (Krebs & Davies 1993).

Hay consenso general entre biólogos evolucionistas en que los organismos responden a presiones selectivas, y que la variación observada en muchos rasgos corresponde a las adaptaciones a ambientes particulares. Una herramienta poderosa para evaluar la correspondencia entre el fenotipo y las variables ecológicas es el método comparativo. Resulta ser útil para probar predicciones derivadas de la teoría de la selección sexual, en particular aquella que propone que la especiación ocurrirá en una proporción mayor en los linajes donde la selección sexual es intensa (Arnqvist, et al. 2000; West-Eberhard 1983) (i.e., apareamiento concordante intenso). Sin embargo, hay todavía pocos estudios que sean concluyentes (Price 1998). Por lo tanto, vale la pena llevar a cabo estudios para clarificar la influencia de la selección sexual en la taza de especiation.

El presente trabajo buscó asociaciones significantes entre rasgos dimórficos sexuales y atribuciones de los habitas ocupados por peces vivíparos (vea sección siguiente). Tracé los estados de caracteres en una filogenia basada en allozimas de la familia Goodeidae cuyas especies exhiben dimorfismo sexual intenso en conducta y morfología, e

identifiqué la historia evolutiva de los eventos independientes. El
diseño facilita obtener evidencia de las asociaciones posibles entre la
conducta y la ecología. El estudio también identificó modos geográficos
de especiación (i.e., simpatría vs. allopatría) analizando la co-
ocurrencia de especies respecto a la edad de los nodos (i.e., donde la
separación de especies ocurre). Además, evalué si la coexistencia de
especies de goodeidos requiere de la evolución de diferencias
fenotípicas. Con este fin analicé la relación entre el contraste del
hábitat (i.e., diferencia promedio de valores estimados para los tipos
de habitas trazados en pares de especies que comparten a un antepasado
común) y la co-ocurrencia de especies.

2. La variación geográfica de la conducta
Los estudios comparativos basados en filogenias reconstruidas nos
ofrecen una perspectiva amplia para identificar la adaptación de la
conducta, pero no nos dice *cómo las poblaciones o las especies divergen*.
Una revisión de la literatura revela que no es fácil identificar los
procesos que originan diversidad biológica (Butlin & Tregenza 1998). Un
primer procedimiento puede ser, buscar correlatos que nos digan qué
variables predicen divergencia de señales entre poblaciones o especies.
Éste es un procedimiento conveniente porque se piensa que los caracteres
que contribuyen al aislamiento reproductivo son importantes para la
generación y mantenimiento de la diversidad biológica (Butlin & Ritchie
1994). Aparté de los mecanismos de selección como tal, hay otros
procesos que operan dentro de las poblaciones y eso puede conducir
incidentalmente a cambios que prevengan cruzamiento (Butlin & Ritchie
1994).

Se ha postulado que la adaptación al ambiente, la selección sexual
y la deriva genética tengan una función en los procesos que ocurren
dentro de las poblaciones (Price 1998). La selección Sexual parece ser
el factor más influyente (Butlin & Ritchie 1994), aunque se ha mostrado
que un efecto combinado de la evolución adaptativa y la selección sexual
también puede ser de importancia. Por ejemplo, la turbiedad del agua y
el riesgo de predación afectan la coloración conspicua y la intensidad

de la conducta de cortejo en gupis (para una revisión, ver Endler 1995).
De manera similar, la eutroficación creciente del Lago Victoria está
impidiendo a las hembras de peces cíclidos escoger a parejas de
apareamiento (Seehausen et al. 1997). En ambos casos parece haber un
compromiso entre la adaptación al ambiente y la selección sexual lo cual
produce diferencias entre poblaciones. Pero ningún estudio presenta
evidencia directa de tal compromiso.

La investigación presente comparó cuatro poblaciones de X.
variata, una especie de goodeido. Los machos muestran en sus flancos
escamas coloridas e iridiscentes (de ahora en adelante llamadas espejos)
cuyo número varía entre y dentro de las poblaciones (Figura 1). La
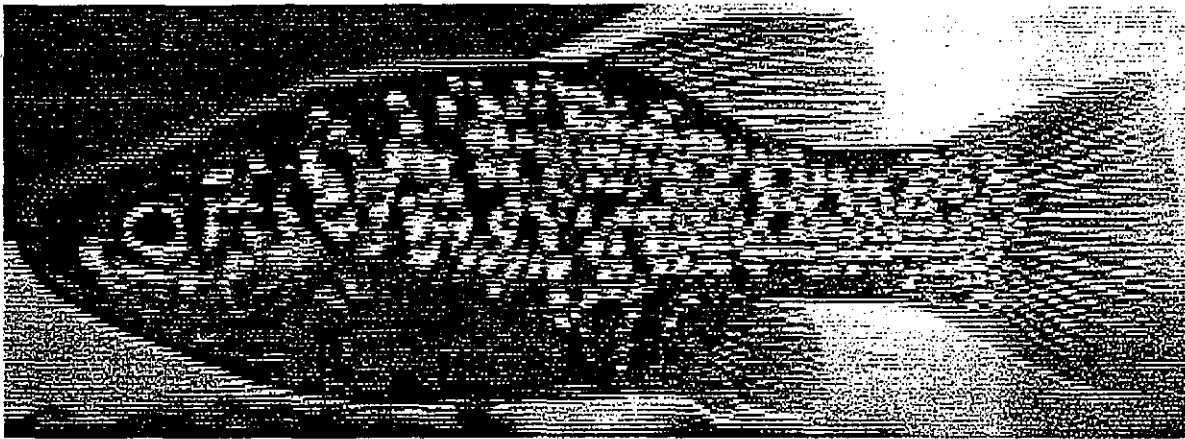
Figura 1. Pez macho de Xenotoca. variata

variación puede estar asociada con la turbiedad del agua: los machos de
aguas claras parecen tener más espejos que los machos de aguas turbias
(obs. pers.). El estudio puso a prueba si las hembras de cuatro
poblaciones preferirían a los machos con más espejos que a los machos
con menos espejos, y si la preferencia sería más intensa en agua clara
que en agua turbia. La información derivada de este estudio permitirá
entender la manera cómo los procesos evolutivos en la diversificación de
los goodeidos están relacionados con el ambiente físico. Además, el
estudio puso a prueba si la culebra jarretera (Thamnophis melanogaster)
se acercaría más a peces macho con más espejos que a peces macho con
menos espejos. Esta culebra habita muchas de las localidades donde los
goodeidos viven (Conant 1963) y hay evidencia circunstancial de que su

dieta incluye goodeidos (Macías Garcia & Drummond 1995; Manjarrez & Drummond 1996). De hecho, Macías Garcia et al., (1998) mostraron que estas culebras se alimentaban más de machos que de hembras de Gyrardinichthys multiradiatus, una especie de goodeido en la cual los machos tienen aletas medias más grandes que las hembras. Censos preliminares sugirieron que T. melanogaster abunda en las localidades donde los machos de X. variata tienen menos espejos en comparación con otras poblaciones (obs. pers.). Así que no debe sorprender que también el riesgo de depredación por esta culebra contribuya a las diferencias entre poblaciones en este carácter.

Los Goodeidos son particularmente convenientes para el estudio de la variación geográfica de fenotipos porque muestran una gran variedad de formas, colores y conducta. Habitan cada tipo de cuerpo de agua dulce, aun así, ocupan una área geográfica relativamente pequeña en el centro de México (Miller & Smith 1986). Además, estos peces presentan características reproductivas únicas (vea sección siguiente). A pesar de esta riqueza de diversidad biológica, hay pocos estudios acerca del origen y mantenimiento de tal diversidad. Por lo tanto vale la pena analizar qué factores la explican.

3. La historia natural de los goodeidos

Los goodeidos pertenecen a una familia de peces vivíparos nativos de la Meseta Central (Miller & Smith 1986; Uyeno et al. 1983). Se agrupan en 17 géneros y alrededor de 40 especies de las cuales, una proporción está extinta y otros en peligro de extinción (Grudzien et al. 1992; Miller & Smith 1986). El desarrollo embrionario es intraovarial, y extensiones especializadas del epitelio del intestino embrionario, conocidas como trofotenia, sirven para que la hembra transfiera nutrientes a su progenie (Hubbs & Turner 1939). En contraste con sus parientes cercanos, poecílidos, los machos no tienen un gonopodio verdadero. En cambio, poseen una modificación de los radios delanteros de la aleta anal (Fitzsimons 1972). La modificación consiste en una reducción de los radios, y es probable que esta estructura sea para eyectar espermatóforos en el poro vaginal (Fitzsimons 1972; Nelson 1975), aunque

14

no hay ningún estudio que confirme tal suposición. No obstante, esta característica impone restricciones de apareamiento a los machos obligándoles a que lograren la cooperación de las hembras para la cópula. Las implicaciones evolutivas de este conflicto sexual todavía tienen que ser estudiadas.

Los procesos evolutivos que promueven la diversificación de goodeidos son desconocidos. La hipótesis actual es que se extendieron por una radiación adaptativa (Miller & Smith 1986), pero no hay ninguna información empírica que confirme esta sospecha. Hay de hecho una controversia acerca de la viabilidad del concepto (Brooks & McLennan 1992) puesto que los pocos estudios que han evaluado el grado de radiación adaptativa para un clado, usando algún criterio empírico, han encontrado que este postulado es poco satisfactorio (pero ver Price 1998). No obstante, la teoría construida al rededor de la radiación adaptativa como tal proporciona información útil en la cual se pueden basar estudios.

Los goodeidos habitan cada tipo de cuerpo de agua dulce, de los estanques de arroyos a los grandes lagos. Esta ubicuidad podría haber sido favorecida por la compartamentalización que la actividad volcánica y tectónica frecuente produjo, y que ha formado la Meseta Central (Barbour 1973). De hecho, la distribución presente de los goodeidos (Figura 2) refleja las características pasadas de las cuencas de la Meseta Central (Barbour 1973). Esta asociación notable no ha sido explotada por los geólogos, y sólo ocasionalmente por los biólogos. El cambio a un nuevo hábitat provoca cambios ecológicos que pueden producir la evolución de señales de comunicación usadas en el reconocimiento de la pareja de apareamiento. Así, la variación en hábitat lleva a la variación en presiones de la selección sexual afectando la facilidad con la que se perciben diferentes caracteres (Endler 1992).

Aparte de la descripción de algunos casos de especiación por vicariansa, no hay nada que nos diga cómo la ocupación de nuevos ambientes influyó en la radiación que los goodeidos experimentaron. Una manera de estudiar tal influencia es buscar correlaciones entre
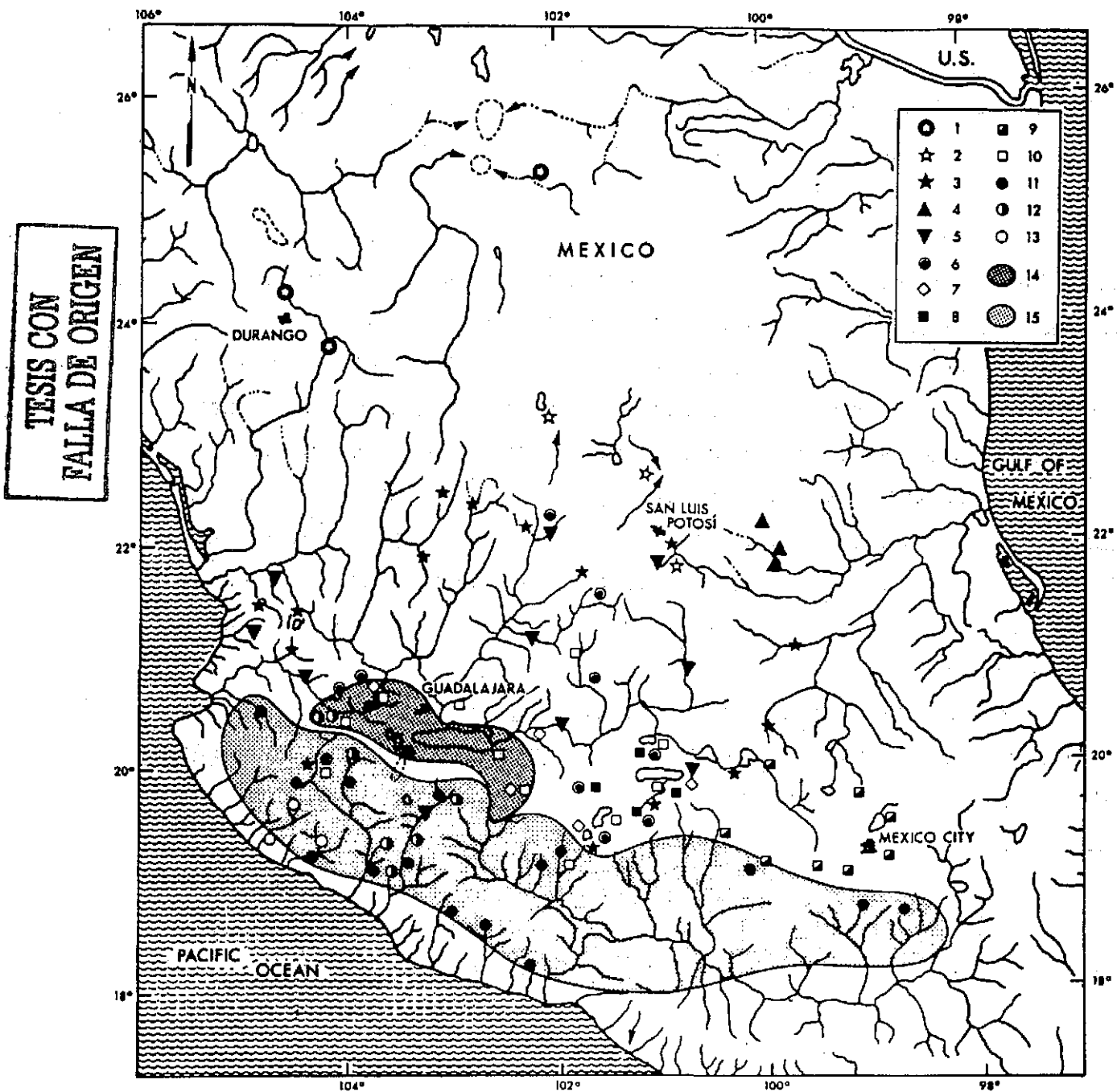
Figura 2. Distribución geográfica de los goodeidos. 1= Characodon, 2= Xenoophorus, 3= Goodea, 4= Ataeniobius, 5= Xenotoca, 6= Allotoca, 7= Skiffia, 8= Hubbsina, 9= Girardinichthys, 10= Zoogoneticus and Neoophorus, 11= Ilyodon, 12= Allodontichthys, 13= Xenotaenia, 14= área de gran concentración (11/17) de géneros, 15= área de concentración de Ilyodon. Los géneros en 14 son: Allodontichthys, Alloophorus, Allotoca, Ameca, Chapalichthys, Goodea, Ilyodon, Neoophorus, Skiffia, Xenotoca and Zoogoneticus. Tomado de Uyeno et al. 1983.

16

morfologías o conductas y variables abióticas. Pero para que estas correlaciones sean útiles, es necesario rastrear la historia evolutiva de los caracteres en estudio, pues la adaptación es a menudo una inferencia histórica acerca de la transición de condiciones ancestrales a condiciones derivadas. La técnica más accesible es el método comparativo, un procedimiento para poner a prueba hipótesis acerca del valor adaptativo de un carácter empleando comparaciones entre las especies. Se sustenta en filogenias reconstruidas para trazar eventos evolutivos independientes, que luego, son la base para hacer comparaciones insesgadas (Harvey & Pagel 1991).

La filogenia más fiable de los goodeids es una clasificación sistemática basada en datos de allozimas (Grudzien et al. 1992), aunque clasificaciones basadas en los rasgos tróficos de adultos se habían desarrollado con anterioridad. Estas clasificaciones fueron reemplazadas luego por clasificaciones basadas en atributos de la anatomía ovárica y de la trofotenia. (Hubbs & Turner 1939). Estas clasificaciones coinciden con algunas relaciones generales entre las especies de goodeidos, pero difieren en algunos aspectos específicos.

En resumen, en este trabajo estudié varios aspectos de la ecología del comportamiento de los goodeidos. Primero, tracé caracteres conductuales y morfológicos en una filogenia basada en allozimas para obtener caracteres evolutivos independientes, y su relación con variables físicas fue analizada. Además, hice análisis a nivel de especie para encontrar modos geográficos de especiación, y análisis a nivel de clado para identificar evolución fenotípica. Segundo, evalué en X. variata. la función de la elección femenina de la pareja de apareamiento en las diferencias entre poblaciones en el número de espejos, y analicé la influencia de la turbiedad del agua en este carácter sexualmente seleccionado. Finalmente, puse a prueba si las culebras T. melanogaster prefieren acercarse a los machos de X. variata con más espejos en lugar de aquellos con menos espejos.

CHAPTER 2

EVOLUTIONARY HISTORY OF PHENOTYPIC VARIATION OF GOODEIDS

Summary

The main purpose of this study is to investigate the influence of running waters and lakes on the variation of conspicuous secondary sex traits in goodeids. In addition, dimorphism in dorsal fin size was used to evaluate if sexual selection promoted speciation of goodeids; it was expected that the most dimorphic species formed the largest genera. I mapped the traits and traced the evolutionary independent events on an allozyme-based phylogeny of these fish, and assessed whether there is a close association with the type of habitat: lake or running waters. Genus size was not positively correlated with dimorphism in dorsal fin size. I did not find evidence that the studied traits had evolved as adaptations to local conditions of lakes or running waters. These results suggest that sexual selection did not promote richness of goodeids, whilst the role of habitat conditions in their diversity is limited.

18

1. Introduction

Many dimorphic traits are sexually selected as evidenced by the consequences on mating success of the bearer. The magnitude of these traits is the result of a balance between selection for reproductive success and selection for viability (Lande 1987; Partridge 1994). Despite the wide distribution of sexually selected traits amongst animals, their role in the generation of species diversity is still controversial (Price 1998). Natural selection can cause rapid divergence of populations by adaptation to local environments (Orr & Smith 1998), whereas sexual selection can accelerate speciation rates by producing many sexually selected traits (Arnqvist, et. al. 2000; Clutton-Brock & Vincent 1991), which may cause reproductive isolation (Butlin & Ritchie 1994). The connection between both selective forces is the idea that habitat, in some cases, creates a link between adaptation and speciation through sexual selection (Price 1998). Variation in morphological and behavioural traits within species and amongst species is ultimately expected to be associated with habitat conditions (Schluter 1996). However, there are traits such as genitalia of many insects that do not have any apparent connection with habitat conditions.

In many instances it is difficult to separate the relative influence of each selective force (Darwin 1871; Lande 1987), especially if the characters involved vary amongst species and amongst populations. Since this variation might be due to a diversity of factors (e.g., ecological, genetic, or behavioural), a first step is to separate these possibilities. The comparative phylogenetic methods are suitable tools to uncover those associations that are evolutionarily meaningful (e.g., Martins & Lamont 1988). Empirical studies may then experimentally clarify in detail the selective mechanisms suggested by the comparative analysis.

Goodeids are a suitable group for investigating the function of sexually selected traits in the generation of species diversification, for they show a diversity of morphological and behavioural characters that are sexually dimorphic (Fitzsimons 1972). They are viviparous freshwater fish endemic to the Mexican plateau (Miller & Smith 1986).

19

The males show a unique morphological feature: the front rays of their anal fin are shorter than the other rays (Fitzsimons 1972). The modification of the anal fin, which the males use to direct spermatophora as these are ejected (Fitzsimons 1972), is less specialised than poeciliids' gonopodium, which is a true intromittent organ (Constanz 1984; Constanz 1989). The lack of an intromittent organ avoids forced insemination, and hence copulation in goodeids is likely to occur only with female co-operation. In addition, goodeid species vary in diet, body size, and the males are colourful and their body and fins are larger than those of the females (Fitzsimons 1972; Macías García 1990; Radda 1984). These differences may potentially drive speciation in goodeids.

Goodeids inhabit every type of inland aquatic environment: from streams to the big lakes of Central Mexico. Although they occupy this diversity of habitats, each species, except Goodea atripinnis and Xenotoca variata is confined to a small geographical area or to a single basin. Some species are even restricted to a single environment: Goodea luitpoldi, Allotoca maculata, Girardinichthys viviparus and Chapalichthys encaustus are exclussively associated with lacustrine habitats (Miller & Smith 1986).

Little is known about the influence of the geographical variation of habitat conditions on the evolution of goodeids' dimorphic traits. The physical characteristics of lakes and running waters may have facilitated or constrained the evolution of sexually dimorphic traits. For example, lacustrine species should show fewer carotene-derived colours because lakes in Central Mexico are typically deep (Barbour 1973), preventing the transmission of short-wave radiation (Levine & MacNichol Jr. 1979; Lythgoe & Partridge 1991). In contrast, running waters should facilitate the presence of carotene-derived colours and some morphologies suitable for unidirectional flow of water.

The aim of this study was to look for (1) general patterns of the influence of sexual selection on species diversity and (2) associations between sexually dimorphic traits and types of habitat that could help me to understand the causes of richness of goodeid species.

20

I tested the predictions derived from the following hypotheses.

I. Hypothesis: sexual selection promotes speciation.

(A) Prediction: the largest genera would include the most

sexually dimorphic species.

To test this prediction I used dorsal fin size as an index of sexual dimorphism.

II. Hypothesis: habitat modulates the action of sexual selection.

(A) Prediction: interspecific variation in sexually selected traits

(on the assumption that they are very frequent) would be associated

with specific types of habitats.

To test this prediction I divided goodeids into those which exclusively inhabit lakes and those which may be found in more than one type of habitat (e.g., rivers, streams and springs), hereafter referred to as `running waters'. This division was intended to reduce the variation in habitat types to a few categories to strengthen the association. In addition, a broader division would have been inaccurate, as most running water species occupy more than one type of habitat.

2. Methods

In broad terms, the methods consisted in recording morphological and behavioural dimorphic traits of fish in the field and in the laboratory. Data from these observations were used to define a set of characters, whose presence or absence was counted across species. The criterion for choosing them was that they were sexually dimorphic traits present in either males or females. Although not all sexually dimorphic traits were moulded by sexual selection, it is unlikely that the traits included in this study (see below) would have evolved by forces such as ecological differentiation. Then, I looked for associations between the characters and the types of habitat using standard comparative phylogenetic methods. Dorsal fin size was used to look for an association between sexual dimorphism and species richness. I will next describe in detail each step of data adquisition.

(a) Study fish

I used an average of 12.78 ± 6.22 (± sd) sexually mature fish of both sexes of each population. The study includes as many species as possible since its objective is to characterise the interspecific variation of sexually dimorphic traits. Sexual maturity criterion in the males was the presence of a spermatopodium, which is the shortness of the front rays of the anal fin. Usually, this sexual trait appears along with other dimorphic traits, such as patterns of bright colours on the body. In contrast, females are dull and they do not show any modification of their anal fin. The age at which fish become sexually mature might vary depending on the species, being in general difficult to determine. Body size at sexual maturity depends on the species as well. In this study I assumed that the dimorphic characters used are species-specific traits and hence are present (or absent if that is the case) in every sexually mature fish of the species.

(b) Study sites

Fish were collected in spring, autumn and winter from 1996 to 1998 in the Mesa Central and Mesa del Norte, Mexico, in previously reported localities (Smith & Miller 1980). I did not take a probability sample (i.e., a probability method of sampling, such as in random sampling) of the localities visited because of the following difficulties: (1) some species are extinct and they can only be found in captivity (e.g., Skiffia francesae, Zoogoneticus tequila); (2) some are endangered species which inhabit only one locality (e.g., Chapalichthys pardalis, Ameca splendens, Characodon audax); (3) some localities have disappeared (e.g., a spring-fed pond in Tepic, Nay). This variability and in general the limited number of localities reported would have made it difficult to apply a rigorous sampling method.

The fish were collected about midday using a 4.5-m long seine that was hauled from 2 to 4 m towards the shoreline. Three samples were taken in the same or a different site, depending on the success of the previous hauling. Fish of every size were often captured, indicating that goodeids reproduce throughout the year, a suggestion which is in

22

accordance with previous observations (E. Soto, pers. commun.). The fish were placed in an ice chest filled with clean water. They were counted and their sex identified. While the fish were taken into the chest, I observed them and recorded (1) conspicuous body colour(s), and (2) conspicuous morphology (e.g., form of fins, spots, etc). I noted these observations and counts in a notebook and transcribed them later into a field book. In addition, I recorded the type of habitat as being either lake or running water.

In the laboratory, the fish were put in aquaria in groups of 3 to 10 individuals of both sexes. Artificial illumination was provided by 20-W tubes of fluorescent light with a 12 h light-dark cycle. Fish were fed on commercially prepared flakes food, twice daily from Monday to Friday and once on Saturday and Sunday.

(c) Laboratory experiments

In the laboratory I systematically observed the fish in their aquaria when feeding them, and confirmed or modified the field scores. In the laboratory also, I conducted experiments to measure median fin size dimorphism and to describe courtship behaviour.

(i) Videorecording of the fish to measure median fin size

To determine dorsal fin size I used video images from samples of variable size (Table 1). I used 2 aquaria (60 x 25 x 27 cm) each divided into 5 compartments with glass walls. Water of all compartments was continuously circulated by a system that pumped water from one extreme compartment to the other extreme. The video recording was performed in a room with aquaria illuminated with 2 tungsten lamps of 20-W.

I placed the fish in the compartments the night before the observations; 3 males were put in alternate compartments and 2 females in between. The divides of the compartments were covered with plastic board. This procedure allowed the fish to acclimate to the novel condition, and prevented them from seeing each other.

In the day of the trials (1100 to 1700 h), the boards were removed and the fish could see each other. This condition motivated males to

Table 1. Statistical summary of data on dorsal fin size dimorphism

Population	N	Sex	Mean ± SE	t	p	Dimorphism
Allotoca dugesi	11	F	0.067 ± 0.002			
(Zirahuén)	5	M	0.079 ± 0.005	2.54	0.034	1.17
Ameca splendens	2	F	0.050 ± 0.003			
(Teuchitlán)	11	M	0.081 ± 0.006	-4.80	0.009	1.64
Characodon audax	10	F	0.051 ± 0.003			
(El Toboso)	3	M	0.078 ± 0.008	-3.15	0.088	1.52
Characodon lateralis	4	F	0.046 ± 0.002			
(San Juan)	4	M	0.061 ± 0.006	2.45	0.058	1.33
Chapalichthys pardalis	4	F	0.085 ± 0.009			
(Tocumbo)	4	M	0.128 ± 0.009	-3.45	0.018	1.51
Goodea atripinnis	9	F	0.053 ± 0.003			
(Cuitzeo)	8	M	0.072 ± 0.005	4.00	0.001	1.36
G. atripinnis	4	F	0.050 ± 0.001			
(Mintzita)	4	M	0.069 ± 0.005	5.61	0.005	1.37
G. atripinnis	3	F	0.050 ± 0.008			
(San Francisco del Rincón)	2	M	0.077 ± 0.012	-1.80	0.320	1.54
G. atripinnis	10	F	0.047 ± 0.003			
(Úmecuaro)	15	M	0.077 ± 0.003	-7.13	0.000	1.66
G. atripinnis	8	F	0.060 ± 0.004			
(Yuriria)	6	M	0.078 ± 0.007	2.19	0.053	1.30
Goodea luitpoldi	8	F	0.056 ± 0.003			
(El Molino)	8	M	0.081 ± 0.004	-5.06	0.000	1.46
G. luitpoldi	4	F	0.054 ± 0.005			
(Pátzcuaro)	3	M	0.083 ± 0.001	-9.06	0.001	1.52
G. luitpoldi	14	F	0.050 ± 0.002			
(Zacapu)	10	M	0.065 ± 0.003	4.54	0.000	1.30
Girardinichthys viviparus	4	F	0.063 ± 0.006			
(Texcoco-Xochimilco)						
Allotoca sp.	9	F	0.052 ± 0.005			
(Zacapu)	9	M	0.108 ± 0.007	-6.78	0.000	2.09
Neoophorus diazi	7	F	0.057 ± 0.005			
(El Molino)	8	M	0.086 ± 0.001	-6.50	0.000	1.50
Skiffia bilineata	11	F	0.066 ± 0.003			
(Tzinzimeo)	7	M	0.111 ± 0.005	-8.67	0.000	1.67
Skiffia lermae	5	F	0.049 ± 0.005			
(El Molino)	12	M	0.085 ± 0.003	-6.00	0.001	1.71
Xenotoca eiseni	12	F	0.067 ± 0.002			
(San Leonel)	6	M	0.091 ± 0.005	-4.70	0.002	1.35
Xenotoca melanosoma	5	F	0.058 ± 0.003			
(Magdalena)	5	M	0.125 ± 0.008	9.01	0.000	2.15
Xenotoca variata	4	F	0.064 ± 0.004			
(Cuitzeo)	6	M	0.098 ± 009	3.57	0.009	1.53
X. variata	12	F	0.064 ± 0.004			
(Mintzita)	10	M	0.121 ± 0.008	-6.83	0.000	1.91
X. variata	8	F	0.062 ± 0.004			
(San Francisco del Rincón)	9	M	0.106 ± 0.005	-7.14	0.000	1.70
X. variata	5	F	0.068 ± 0.003			
(Yuriria)	2	M	0.104 ± 0.022	2.36	0.260	1.54
X. variata	3	F	0.061 ± 0.007			
(Zacapu)						
Zoogoneticus quitzeoensis	4	M	0.087 ± 0.009			
(Mintzita)						
Z. quitzeoensis	5	F	0.055 ± 0.007			
(San Francisco del Rincón)	3	M	0.078 ± 0.009	-2.05	0.130	1.74

court females, and as a result they opened and erected their fins. I videotaped each fish with a camcorder (Sony Hi 8) for about 5 min to ensure that sufficient frames had been taken in which the fish were perpendicular to the camcorder's lens and with opened fins (displayed). However, in some cases fish were discarded because they did not open their fins enough. Then the video frames were played on a monitor and the suitable images were digitised using a digitising card. Using a computer program (Image Tool) I obtained the following relative measurements (pixels, area units in which a computer screen is divided):

1. Body area, number of pixels occupied by the entire fish image;
2. Dorsal fin index, proportion of the area of the dorsal fin, number of pixels occupied, to body area.

To obtain an estimation of dimorphism, I used the ratio of the dorsal fin index of males to the dorsal fin index of females (Butler et. al. 2000).

(ii) Videorecording of courtship behaviour

I videorecorded courtship behaviour for having a permanent archive that helped me make precise descriptions. Recordings were done on 4 ± 3.91 (mean ± sd) pairs of fish (see Appendix A for a complete list). I used aquaria (60 x 25 x 27 cm) divided into two compartments with glass walls. In each compartment I placed a pair of fish (male and female) from the same population. Experimental individuals were those which had shown high motivation to court in the condition of videotaping described above, although the main criterion of selection was to have sexually mature individuals of comparable size. I sat in front of the aquaria videorecorded courtship every time it appeared. The trials were in the morning and afternoon during 2-3 days after putting the fish in the aquaria. Nevertheless, I was able to videotape the pairs only in a few instances because courtship was infrequent under these conditions in many of the species. Frequently I recorded courtship by hand-written descriptions opportunistically taken when feeding the fish, so the sample size given above was indeed larger. When videos were available I played them back on a monitor to analyse courtship displays (for a

25

detailed description of courtship behaviour see Appendix B), which I
described in terms of the following criteria (Baerends et al. 1955).

1. Orientation, the position of the male in relation to that of the
   female;

2. Displays, stereotyped movements specific to a species that a male
   shows to a female;

3. Contact movements, display movements that a male does when
   approaching a female or when is attempting to copulate with her.

(d) Comparative analyses

With the observations recorded in the field and in the laboratory,
including the experiments, I defined the following set of morphological
and behavioural characters. The criteria for choosing them were the
presence of sexually dimorphic traits in either males or females.

(i) Morphological characters

1. Carotene-derived colours (yellow-orange-red), presence of these
   colours on the outer parts of any of the median fins or on the body
   of males;

2. Black fins of males, any of the median fins completely in black;

3. Shiny speckles, presence of discrete bright and colourful scales on
   the body of males; usually they appear on the flanks;

4. Dorsal fin size dimorphism, dorsal fin area relative to body area of
   males was divided by dorsal fin area relative to body area of
   females;

5. Dark contours of males' fins;

6. Alternate brown and bright vertical bars on the flanks of females;

7. Notched dorsal fins of males.

(ii) Behavioural characters

1. Sweeping on the substrate. It consists in the male going to the
   bottom of the aquarium shaking the body so that it sweeps the sand
   with its belly whilst the female is stationary in front, a few cm
   above the substrate;

26

2. Vibration by females. This is a movement that consists in wagging the head and leaning the body in variable degrees (Macías Garcia 1990; Nelson 1975). Usually the female shows this behaviour when a male is either approaching or courting her;

3. Flagging. This is a display that a male frequently shows when he is courting a female; usually the male approaches the female whilst shaking the body at high frequency with the median fins spread (Macías Garcia 1990). The fins may be deflected either towards or away from the female, depending on the species;

4. Figure of eight or 'x'. This is a display that males of some species show in front of a female. The male moves quickly tracing a trajectory in the horizontal plane that mimics a figure of eight (Macías Garcia 1990), or x if the trajectory is short and the male moves away from the female.

   I recorded in all the species if the characters were present or absent (i.e., the state of the trait, Table 2). With few exceptions, I compared the character states I observed with reports previously published (Fitzsimons 1972; Radda 1984; Radda 1985; Scott 1987). Sometimes, when I had no recordings of a particular species from my own dataset, I relied on these reports for determining character states.

   Once defined the character states for every species, I estimated the ancestral states using parsimony criteria (Maddison et al. 1984; Maddison & Maddison 1992) and WinClada (release 0.9.99). I used a systematic arrangement based on allozyme data as an hypothesis for the Goodeidae phylogeny (Grudzien et al. 1992), in particular the optimised distance Wagner tree (Figure 3). Although the authors reported other constructed trees, I decided to use the optimised distance Wagner tree because, as the authors state, this is the most congruent with morphological and karyological studies of goodeids. Then I counted the independent evolutionary events using the method suggested by Ridley (Ridley 1983), and applied Fisher's exact test to test for significant associations between characters and type of habitat, lake or running waters.

Table 2. Data matrix of character states

Especies	Characters										
	1	2	3	4	5	6	7	8	9	10	11
Crenichthys baileyi	0**	0	0	?	0	0	0	?	?	?	?
Allodontichthys hubbsi	?	0	?	?	?	?	0	?	?	?	?
A. tamazulae	1	0	?	?	?	?	0	?	?	?	?
A. zonistius	1	0	?	?	?	?	0	?	?	?	?
Ilyiodon whitei	1	0	0	?	1	0	0	0	1	?	0
I. furcidens	1	0	0	?	1	0	0	?	?	?	?
Xenotaenia resolanae	1	?	?	?	1	?	0	?	?	?	?
Allotoca maculata	1	0	0	?	1	1	0	0	1	0	0
A. dugesi	0	0	0	1	1	1	0	1	0	1	0
Neoophorus catarinae	0	?	?	?	?	?	0	?	?	?	?
N. diazi	0	0	0	1	0	0	0	0	0	0	0
Zoogoneticus tequila	1	0	1	?	1	1	0	0	0	1	?
Z. quitzeoensis	1	0	1	1	1	1	0	0	0	1	0
Characodon audax	0	1	0	1	0	0	0	0	0	1	0
C. lateralis	1	0	0	1	0	0	0	0	1	1	0
Ataeniobius toweri	1	0	?	?	?	?	0	?	?	?	?
Goodea atripinnis	1	0	0	1	0	0	0	0	1	1	1
G. gracilis	0	0	0	?	0	0	0	0	1	1	1
G. luitpoldi	0	0	0	1	0	0	0	0	0	0	0
Xenotoca eiseni	1	0	?	1	0	0	0	0	1	1	1
X. melanosoma	0	0	0	1	0	0	0	0	0	0	0
Alloophorus robustus	1	0	1	?	0	0	0	0	0	0	0
Ameca splendens	1	0	1	1	0	0	0	0	0	0	0
Chapalichthys encaustus	?	0	1	?	0	0	0	0	1	0	0
C. pardalis	1	0	1	1	0	0	0	0	1	0	0
Xenotoca variata	1	0	1	1	0	0	0	0	1	1	1
Xenoophorus captivus	1	0	1	?	0	0	0	1	0	1	1
Skiffia bilineata	0	0	0	1	1	0	0	0	1	1	1
S. lermae	1	0	0	1	0	0	1	0	1	1	1
S. francesae	1	0	0	?	0	0	1	0	?	1	1
S. multipunctata	1	0	0	1	0	0	1	0	?	1	1
Hubbsina turneri	1	0	0	1	?	?	0	?	?	?	?
Girardinichthys multiradiatus	1	0	0	1	1	1	0	0	1	1	1
G. viviparus	0	1	0	1	0	1	0	0	0	0	0

*Characters are:
1 Carotene-derived colours;
2 Black fins;
3 Shiny speckles;
4 Median fin size dimorphism;
5 Dark contours of fins;
6 Alternate brown and bright vertical bars;
7 Notched dorsal fins;
8 Sweeping on the substrate;
9 Vibration by females;
10 Flagging;
11 Figure of eight or x

**States are:
1: presence
0: absence

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia bilineata
Skiffia lermae
Skiffia francesae
Skiffia multipunctata
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Chapalichthys pardalis
Xenotoca eiseni
Xenotoca melanosoma
Goodea atripinnis
Goodea gracilis
Goodea luitpoldi
Ataeniobius toweri
Characodon audax
Characodon lateralis
Allotoca maculata
Allotoca dugesi
Zoogoneticus tequila
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyiodon whitei
Ilyiodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae

Figure 3. Phylogenetic tree of the Goodeidae family adapted from Grudzien et al. (1992). _Allotoca maculata_, _Chapalichthys pardalis_, _Characodon audax_, _Goodea gracilis_, _Goodea luitpoldi_, _Ilyodon whitei_ and _Zoogoneticus tequila_ were not included in the original tree.

3. Results

Males of all species had dorsal fin indices larger than females (Figure 4). Populations of species clustered together. There was not a significant association between dimorphism in dorsal fin size and number of species per genus (see Figure 5; $r^2 = 0.052$; $F(1,13) = 0.71$, $p > 0.40$).

The characters with the state transitions are depicted in Appendix C. Because the original phylogeny (Grudzien et al. 1992) did not include any species necessary to compare running water fish with lake fish, I added the following species to the tree: Allotoca maculata, Chapalychthys pardalis, Characodon audax, Goodea luitpoldi, G. gracilis, Ilyodon whitei and Zoogoneticus tequila (Webb & Miller 1998). Many of them are sister species of genera well established and so monophyletic groups; it is unlikely that their inclusion could have

affected the tree structure. Although the addition of <u>Goodea</u> spp.,
caused a polytomy, whose treatment has been a controversial issue
(Ridley 1983), it was the most conservative arrangement.

Characters evolved 21 times, though none of them was significantly
associated with running waters or lakes (Table 3). Note, however, that
all the characters, except one, evolved in species inhabiting running
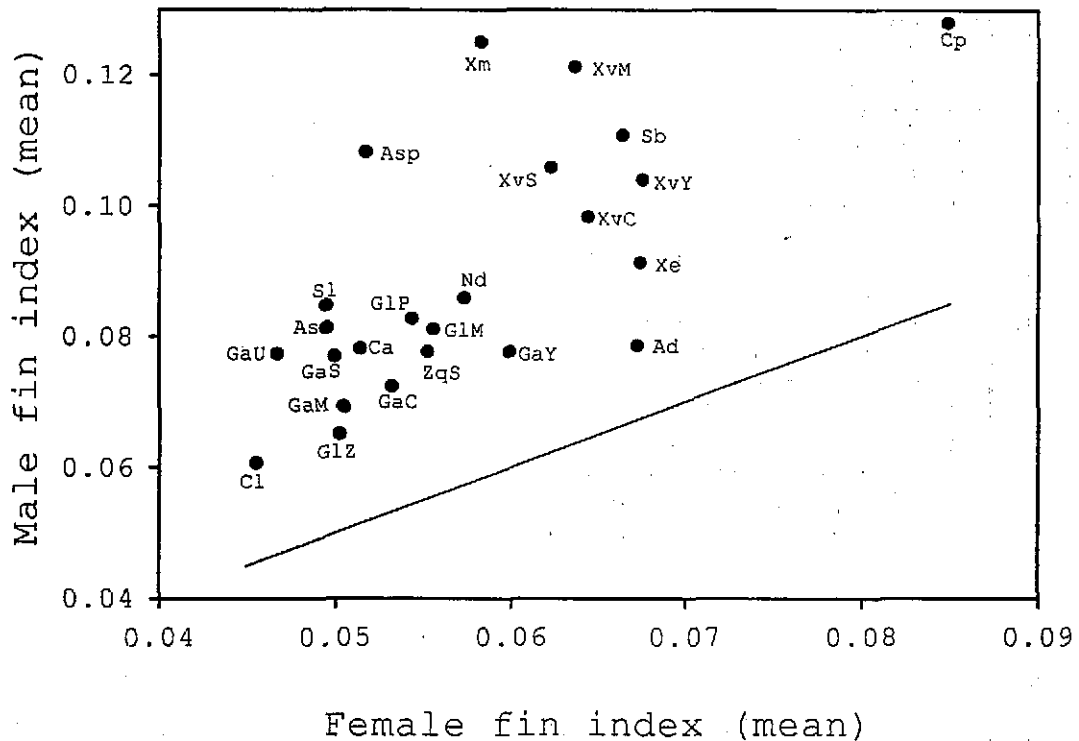
Figure 4. Dorsal fin size dimorphism in goodeid populations and
species. Each point represents the mean fin index, which is the
ratio of dorsal fin area to total area of body, of both males and
females. The solid line indicates the direction of the dimorphism:
in favour of males above the line. Asp.: *Allotoca* sp; Ad: *A. dugesi*;
As: *Ameca splendens*; Ca: *Characodon audax*; Cl: *C. lateralis*; Cp:
Chapalichthys pardalis; GaC: *Goodea atripinnis* (Cuitzeo); GaM: *G.
atripinnis* (Mintzita); GaS: *G. atripinnis* (San Francisco); GaU: *G.
atripinnis* (Úmecuaro); GaY: *G. atripinnis* (Yuriria); GlM: *Goodea
luitpoldi* (Molino); GlP: *G. luitpoldi* (Pátzcuaro); GlZ: *G. luitpoldi*
(Zacapu); Nd: *Neoophorus diazi*; Sb: *Skiffia bilineata*; Sl: *S.
lermae*; Xe. *Xenotoca eiseni*; Xm: *Xenotoca melanosoma*; XvC: *Xenotoca
Variata* (Cuitzeo); XvM: *X. variata* (Mintzita); XvS: *X. variata* (San
Francisco); XvY: *X. variata* (Yuriria); ZqS: *Zoogoneticus
quitzeoensis* (San Francisco).
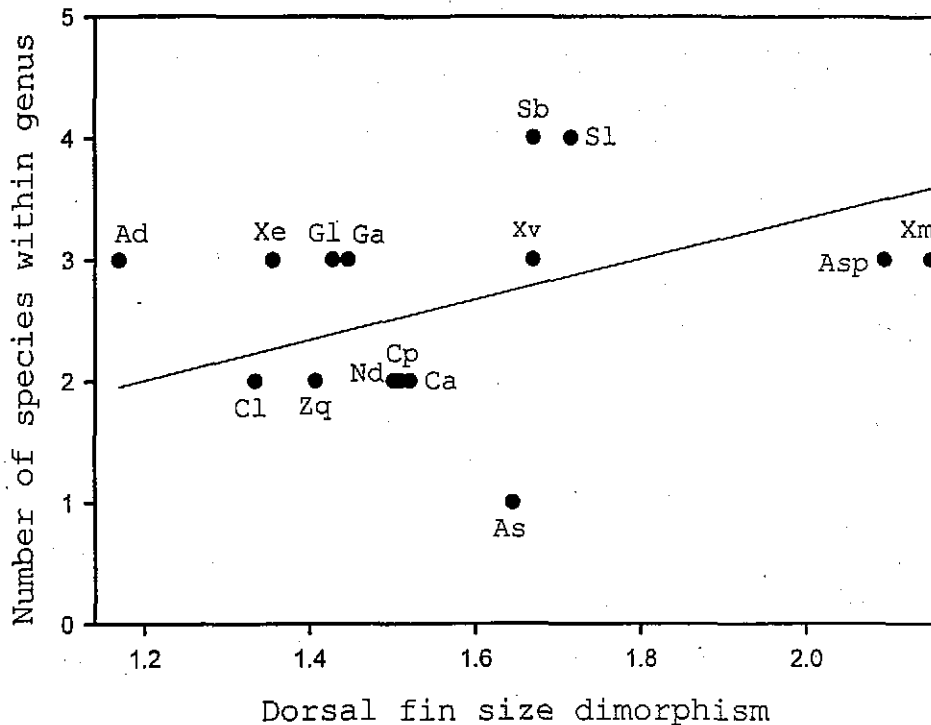
Figure 5. Relationship between dorsal fin size dimorphism and number of species within genera. Each point represents the mean ratio of dorsal fin size of males to dorsal fin size of females in species of goodeids. In the cases of more than one population within species, an average was used. See figure 4 for abbreviations.

waters.

4. Discussion

The wide presence of dorsal fin size dimorphism in goodeids indicates that this character was a suitable index for estimating the strength of sexual selection and for testing whether it promoted speciation. There was not a significant association between the magnitude of the dimorphism and the number of species within genera. This suggests that sexual selection did not play an important role in species richness of goodeids. Perhaps a comparison at a larger scale (e.g., clades) may reveal a significant relation, though the size of the two main clades of the current phylogeny for goodeids does not seem to differ significantly. Other studies (e.g., Barraclough et al. 1995) have used family-level comparisons and possibly the fact that they used

Table 3. The association of type of habitat with sexually dimorphic characters

Character	Number of independent events		p*
	Running water	Lake	
Vibration by females	3		ns
Figure of eight or x	1		ns
Carotene-derived colours	3		ns
Shiny scales ('speckles') on the flanks	2		ns
Black fins	1	1	ns
Brown-bright vertical bars (females)	2		ns
Notched fins	1		ns
Sweeping on the substrate	2		ns
Median fin size dimorphism	1		ns
Dark contours of median fins	3		ns
Flagging	1		ns

larger sample sizes helped find a significant positive relationship. If the association holds for large scale comparisons it should also hold for lower level scales. Thus, it may still be worth doing future studies by including all the goodeid species and by measuring other dimorphic traits.

I did not find definite evidence that habitat modulates the action of sexual selection in goodeids. The results of the association between sexually dimorphic traits and type of habitat indicate that although most of the traits were present in running water species, each one appeared only a few times, making it difficult to find a significant association. Five out of 11 traits evolved only once, and three only twice, so that many of them shown by extant species are shared ancestral characters (see Table 3). There is evidence that the geographical distribution of goodeids reflects former rather than present water routes (Miller & Smith 1986), and hence that the major changes in species distribution due to water routes could have occurred at the beginning of the adaptive radiation of goodeids. The fact that most characters had evolved in running water species may reflect the available opportunities to ancestral species. Therefore, the action of habitat conditions seems not to have exerted a differential influence on

the evolution of sexually selected traits between running water fish and lake fish. But genetic drift and constraints on evolutionary change might have caused mismatches between sexually dimorphic characters and environment.

The frequent appearance of dimorphic characters in running water environments suggests that natural selection could have played some role in goodeids diversity. The division of habitats into two broad categories though, does not allow me to propose what exactly the characteristics are that contribute to that differences (e.g., Leroi et al. 1994, but see Doughty 1996). Yet, there are some obvious features of lakes and running water habitats that make them look different. Lake basins arose out of (1) movements of the earth's crust, (2) events that happened in the last ice age, or (3) changes in the land surface that have happened since the end of the last ice age (Pielou 1998). This origin contrasts with that of rivers and streams which make their own valley by a self-sustaining erosive power (Pielou 1998). Similarly, flow is, although not uniform, unidirectional in running waters (Giller & Malmqvist 1998), whereas wind-induced turbulence plays a major role in mixing of the water column in lakes (Brönmark & Hansson 1998). Although, these differences between running waters and lake suggest that fish meet different demands, they do not explain why dimorphic characters evolved more frequently in running waters than in lake. One possibility is that running waters facilitated the evolution of some characters, and these in turn caused the evolution of the remainder. Candidate characters are those which evolved most, such as colour on the body. Besides turbidity of water, flow of water may be determinant for sexual displays. In fact, there is evidence that water velocity influences display behaviour in fish (Nicoletto 1996). Laboratory experiments are necessary to test whether characteristics of running water habitats such as the degree of turbidity and the flow of the water significantly affect the expression of sexually dimorphic traits.

CHAPTER 3

THE ROLE OF GEOGRAPHY IN THE SPECIATION OF GOODEID FISH

Summary

This study investigates modes of speciation and the role of geography, habitat and drainage, in the co-occurrence of the viviparous fish goodeids. Because speciation of goodeids occurred by adaptive radiation, I expected that sympatry would be the main geographical mode of speciation, and that ecological differentiation would be necessary for goodeid species to co-occur. Independent contrasts of co-occurrence, habitat types and drainage were obtained for the Goodea and Girardinichthys groups, which are the two main clades of an existing allozyme-based phylogeny of goodeids. Co-occurrence was plotted against node height and habitat and drainage contrast against co-occurrence. Sympatry was the major mode of speciation with occasional events of allopatry in both clades. Habitat contrast did not show any association with co-occurrence, but drainage contrast tended to diminish in the Goodea group and decreased significantly in the Girardinichthys group. These results suggest that goodeids diverged in sympatry whose extent does not depend on habitat or drainage differences, though they could have played a closer influence on species coexistence in the Girardinichthys group than in the Goodea group.

1. Introduction

A remarkable feature of adaptive radiations is the high degree of
sympatry amongst descendant species, although the processes involved in
such sympatry are unclear (Schluter 1996). Colonization or movement into
new habitats has been associated with ecological differentiation amongst
coexisting populations (Losos et al. 1997). The resultant ecological
adaptation seems to determine the evolution of morphology (Bouton et al.
1999; Losos et al. 1997), which in turn may drive the evolution of
mating signals (Podos 2001) and subsequent species divergence. Yet, the
variation of many morphological and behavioural traits is often
associated with geographical variation in habitat conditions (Schluter
1996).

The analysis of general patterns leading to species richness may,
in a comparative context, clarify the role of geography in speciation.
In this study geography refers to habitat, such as river, lake, etc.,
and to drainages in central Mexico, such as Lerma basin. There has been
an intense discussion on the relative importance of alternative
geographical modes of speciation (i.e., sympatry, allopatry and
parapatry). Such discussion was firstly based on the analysis of
species' ranges (Mayr 1963). Recent approaches (Chesser & Zink 1994)
have focused on counting the relative frequency of modes of speciation
using phylogenetic information, but have failed to compare observed
patterns to those predicted by specific models of geographical modes of
speciation. In addition, they have relied on arbitrary classifications
of modes of speciation. An alternative approach is to analyse the
geographical ranges of species within clades using their phylogenies
(Barraclough et al. 1998; Barraclough & Vogler 2000). Such phylogenies
trace patterns of splits leading to present day species, and so may
potentially give information about processes operating during adaptive
radiations (Barraclough et al. 1998). This approach is based on the view
that the geographical ranges of sister clades can be used as an estimate
of the geographical mode of speciation at the time of speciation.
Similarly, range changes after speciation may be explained by patterns
of geographical ranges for sister clades across all nodes in the

phylogeny (Brown & Gibson 1983; Chesser & Zink 1994; Lynch 1989). Because deeper nodes in the phylogeny represent relatively older events of speciation, changes in pattern with relative node age may reveal the extent of changes in geographical ranges over time (Barraclough & Vogler 2000).

A second issue concerning adaptive radiation is phenotypic variation in relation to the geographical ranges of species. Sympatric species richness may be the result of random shifts in the ranges of species over time, but it may be that ecological constraints to sympatry exist, so that only species with accumulated ecological differences can coexist (Losos et al. 1998). Partitioning of phenotypic variation amongst nodes in the form of contrasts (Felsenstein 1985; Harvey & Pagel 1991) may be used to distinguish between these alternatives. This approach can be used to investigate the relationship between phenotypic changes and geography (Barraclough et al. 1998; Barraclough et al. 1999).

Goodeids are freshwater fish suitable for investigating geography in relation to modes of speciation and phenotypic variation because they are indeed a case of adaptive radiation (Miller & Smith 1986). They consist of about 40 species of viviparous fish endemic to the Mexican plateau (Uyeno et al. 1983) that show interspecific divergence in morphological and behavioural traits used to exploit a variety of resource types (Fitzsimons 1972). They differ in body size and diet, and males typically are colourful with their body and fins larger than those of females (Fitzsimons 1972; Macías Garcia 1990; Radda 1984). Goodeids inhabit a variety of inland aquatic environments ranging from streams to the big lakes of Central Mexico. Even though they occupy such a diversity of habitats, most species are confined to a small geographical area or to a single basin (Miller & Smith 1986).

Although these findings on goodeids are valuable, there is little information on the evolutionary factors that contributed to their diversification. Here, I investigated the role of geography in the speciation of goodeids by testing the predictions derived from the following hypothesis:

36

I. Hypothesis: speciation of goodeids occurred by adaptive radiation.

(A) Prediction: geographical modes of speciation would be sympatric.

(B) Prediction: ecological differences would be necessary for species to co-occur.

My aim was to identify geographical modes of speciation and patterns of association between habitat differentiation and species coexistence.

2. Methods

To estimate geographical modes of speciation and patterns of association between habitat differentiation and species coexistence, I separately analysed each of the two major clades of goodeids, which here are called Goodea and Girardinichthys groups because they are the basal taxons (Figure 6). I used a systematic arrangement based on allozyme data as a hypothesis for the phylogeny of the Goodeidae (Grudzien et al. 1992), in particular the optimised distance Wagner tree. Although the authors reported other constructed trees, I decided to use the optimised distance Wagner tree because, as the authors admit, this is the most congruent one with morphological and karyological studies of goodeids.

Relative age of nodes was calculated as the inverse of the distance of each node from a perpendicular line to the bottom of the tree, and expressed as the proportion of the length of the bottom internode. I used this procedure instead of the distance of a node from the tips of the phylogeny because the branches of sister taxa differed in length. Coexistence was calculated as the number of localities where two or more member species of a clade co-occurred, weighed by the total number of species of that clade. Although there are reports on the geographical ranges of distribution of goodeids (e.g., Uyeno et al. 1983), I preferred to use co-occurrence because it offered me a finer measurement of species coexistence. Information on species localities was obtained from previous reports (Fitzsimons 1972; Grudzien et al. 1992; Radda 1984; Uyeno et al. 1983) and from my own dataset. The expected patterns of the relation between co-occurrence and node height can be summarized as follows (Barraclough et al. 1998; Barraclough &

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia francesae
Skiffia multipunctata
Skiffia bilineata
Skiffia lermae
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Goodea atripinnis
Xenotoca eiseni
Xenotoca melanosoma
Ataeniobius toweri
Characodon lateralis
Allotoca dugesi
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae

Girardinichthys group

Goodea group

Figure 6. Allozyme-based cladogram for the Goodeidae family. Redrawn from Grudzien et al. 1992. Nodes heights in the present study were calculated using the origi-nal distance Wagner tree.

Vogler 2000). If speciation is allopatric, an intercept of zero is expected because recently diverged sister species are expected to display no overlap in geographical ranges. If speciation is predominantly sympatric, an intercept greater than zero is expected because recently diverged sister species must co-occur in one or more localities. Although the intercept can, in theory, vary between a number greater than zero and one as large as localities are occupied by sister species, in practice this variation hardly overtakes one because most species inhabit a reduced number of localities. The degree of co-occurrence between anciently diverged sister clades depends on the extent of range changes after species splits (Barraclough et al. 1998; Barraclough & Vogler 2000). If speciation is sympatric, co-occurrence

will tend to decrease slightly for older nodes. If speciation is allopatric, an increase in sympatry is expected because ranges are mixed over time by random fluctuations.

I used habitat and drainage contrasts, as both give information, though at different scales, on the geographical distribution of goodeids. Contrasts were calculated as previously reported (Barraclough et al. 1998). Briefly, in the case of sister species if A and B had the same habitat type, contrast was= 0; if they had different habitat types, contrast was= 1; in the case of higher nodes if C had same habitat as A and B, contrast was= 0; if C had same habitat as A or B, but not both, contrast was= 0.5; if C had different habitat to both A and B, contrast was= 1. The rates of habitat change were scaled with respect to the relative age of nodes. I followed the same procedure for calculating drainage contrasts. Habitat and drainage data were obtained from previous reports (Miller 1986; Miller & Smith 1986). Goodeids occupy five main types of habitat: big river, stream, creek, lacustrine and spring isolates; and fourteen drainages distributed across Central Mexico (Table 4). Note that species may occupy more than one type of habitat and more than one drainage. The expected patterns of the relation between habitat contrast or drainage contrast and co-occurrence can be summarized as follows (Barraclough et al. 1998; Barraclough & Vogler 2000). A positive relation is expected if phenotypic differences are necessary for sympatric species to co-occur. A negative relation is expected if phenotypic differences arise as a consequence of geographical variation in environmental conditions.

Both types of relationship, co-occurrence against node height and habitat contrast and drainage contrast against co-occurrence, were analysed with linear regression methods (Sokal & Rohlf 1995), although the results were used for descriptive rather than inferential purposes.

3. Results

In the Goodea group, 4 out of 13 splits displayed no co-occurrence between sister taxa (Figure 7) suggesting that those four were events of vicariance with no subsequent range movements. These splits corresponded

39

Table 4. Distribution of Goodeidae in drainage

Drainage	No. of species
Santiago	2
Lerma	13
Morelia	7
Pátzcuaro	5
Zirahuén	4
San Juanico	3
Valle de México	1
Atotonilco	4
Ameca	10
Magdalena	4
Armería	4
Coahuayana	5
Balsas	8
Pánuco	4

Taken from Miller and Smith 1986
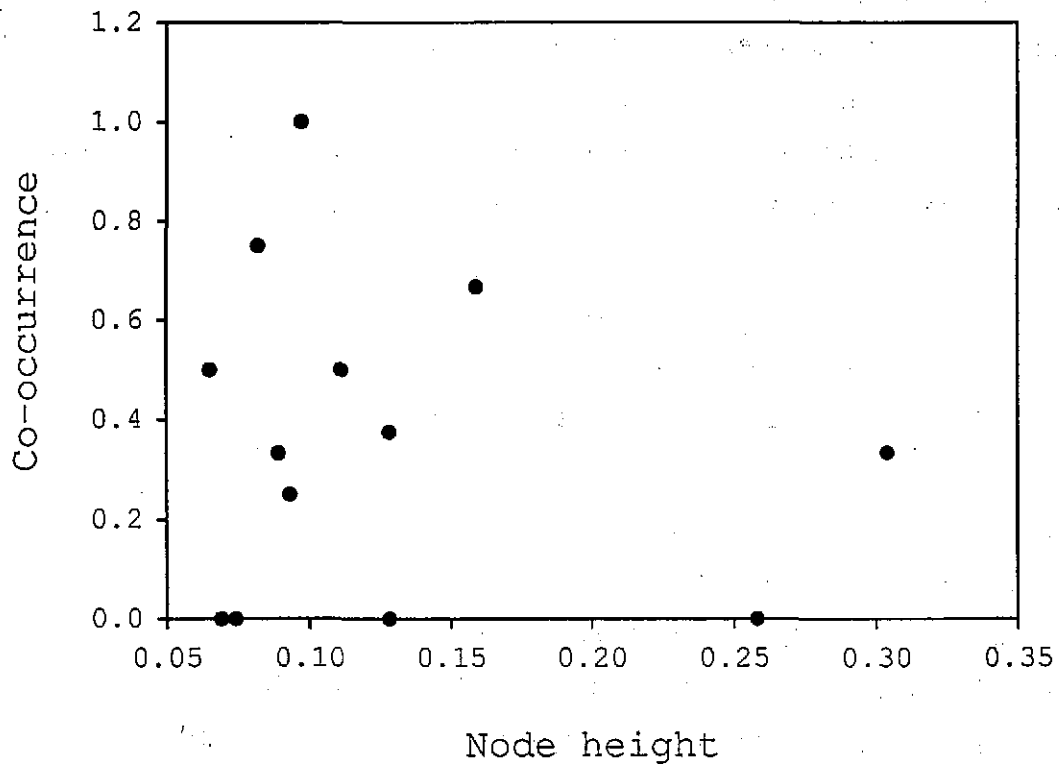
Figure 7. Relationship between the degree of co-occurrence and node height in the <u>Goodea</u> group using an optimized distance Wagner tree.

40

in chronological order to <u>Ataeniobius</u> <u>toweri</u> and its outgroup,
<u>Characodon</u> <u>lateralis</u> and its outgroup, <u>Neophorus</u> <u>catarinae</u> and <u>N</u>. <u>diazi</u>,
and <u>Allodontichthys</u> <u>zonistius</u> and <u>A</u>. spp. (see Figure 6). The other
splits showed varying degrees of co-occurrence and little change over
time. The regression slope was not significant ($r^2 = 0.24$, $F(1,11) = 0.27$,
$p > 0.6$), whereas the intercept was significantly different from zero
($t(12) = 2.37$, $p < 0.04$). In the case of the <u>Girardinichthys</u> group, 3 out
of 11 splits displayed no co-occurrence (Figure 8) suggesting that those
three, arose from vicariance events without later range movements. These
splits corresponded in chronological order to <u>Girardinichthys</u>
<u>multiradiatus</u> and <u>G</u>. <u>viviparus</u>, <u>Skiffia</u> <u>francesae</u> and <u>S</u>. <u>multipunctata</u>,
and <u>Ameca</u> <u>splendens</u> and <u>Chapalichthys</u> <u>encaustus</u> (see Figure 6). The
other splits showed varying degrees of co-occurrence and moderate
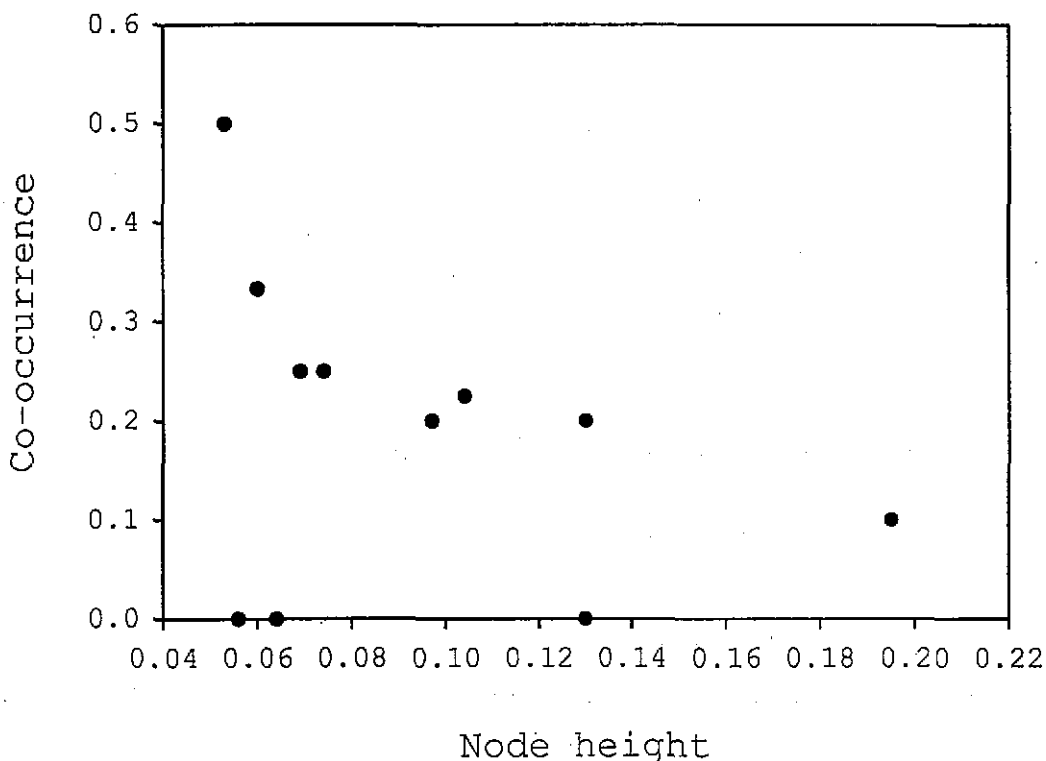
Figure 8. Relationship between the degree of co-occurrence and node
height in the <u>Girardinichthys</u> group using an optimized distance Wagner
tree.

41

subsequent range movement. The regression slope was not significant ($r^2 = 0.118$, $F(1,9) = 1.20$, $p > 0.3$), but the intercept did differ significantly from zero ($t(10) = 2.69$, $p < 0.03$).

In the <u>Goodea</u> group there was not an apparent relationship between habitat contrast and co-occurrence (Figure 9); the regression slope was
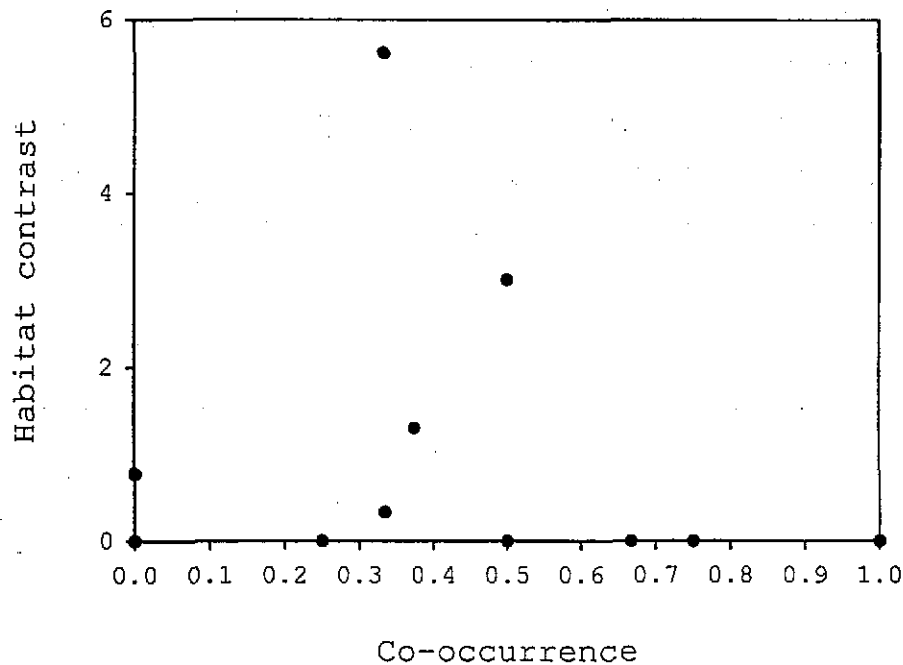
Figure 9. Relationship between habitat contrast and the degree of co-occurrence in the <u>Goodea</u> group using an optimized distance Wagner tree.

not significant ($r^2 = 0.0$, $F(1,11) = 0.0003$, $p > 0.9$). There was no relationship when drainage contrast replaced habitat contrast (Figure 10); the regression slope was not significant ($r^2 = 0.10$, $F(1,11) = 1.20$, $p > 0.2$). In the <u>Girardinichthys</u> group there was not an apparent relationship between habitat contrast and co-occurrence (Figure 11); the regression line was not significant ($r^2 = 0.159$, $F(1,9) = 1.69$, $p > 0.2$). The relation was stronger and statistically significant when drainage
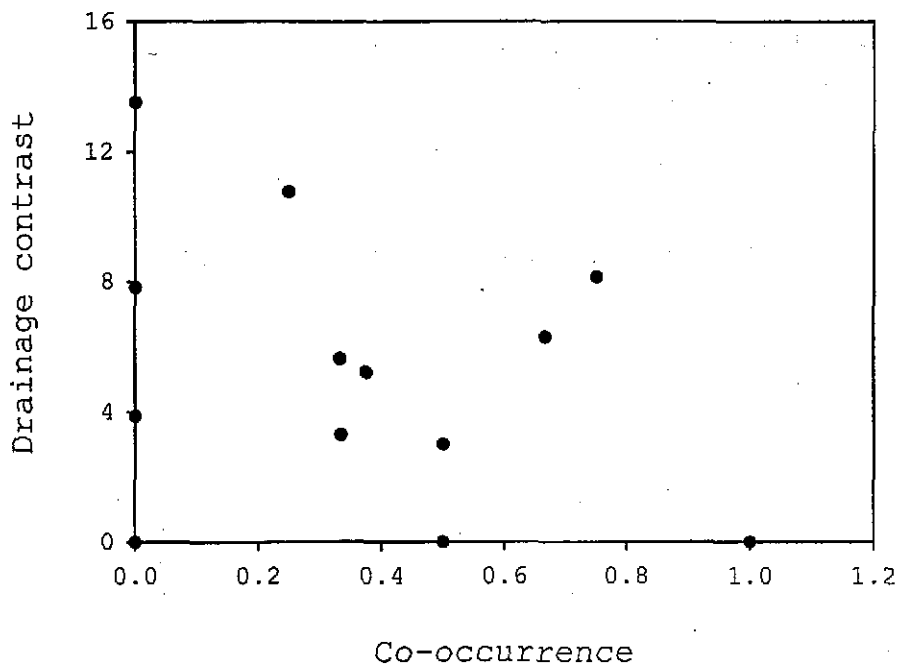
Figure 10. Relationship between drainage contrast and the degree of co-occurrence in the <u>Goodea</u> group using an optimized distance Wagner tree.
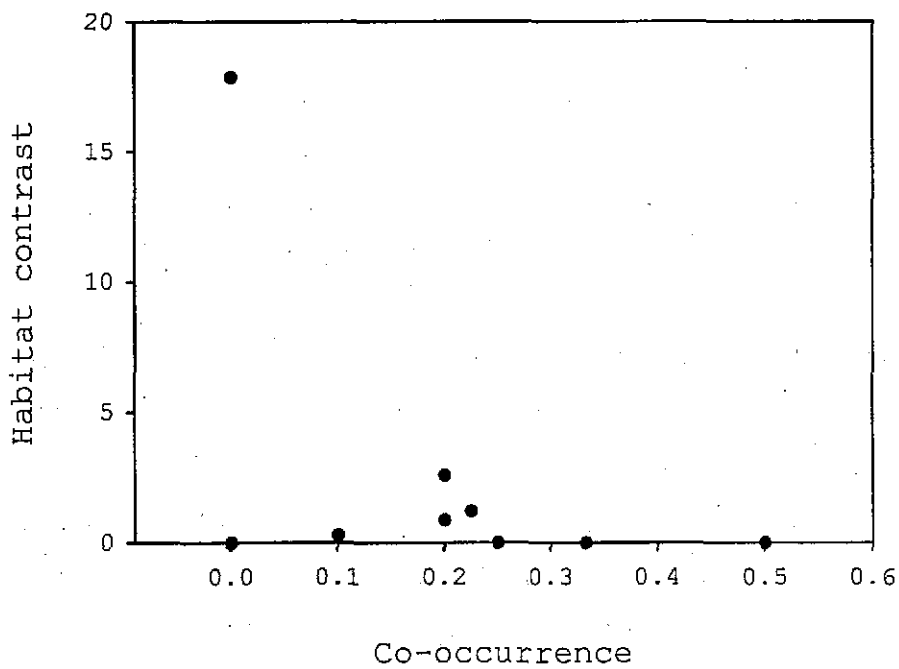
Figure 11. Relationship between habitat contrast and the degree of co-occurrence in the <u>Girardinichthys</u> group using an optimized distance Wagner tree.

contrast was used (Figure 12), suggesting that the _Girardinichthys_ group's species had less fixed habitat requirements.

4. Discussion

The pattern of the relationship between co-occurrence and node height
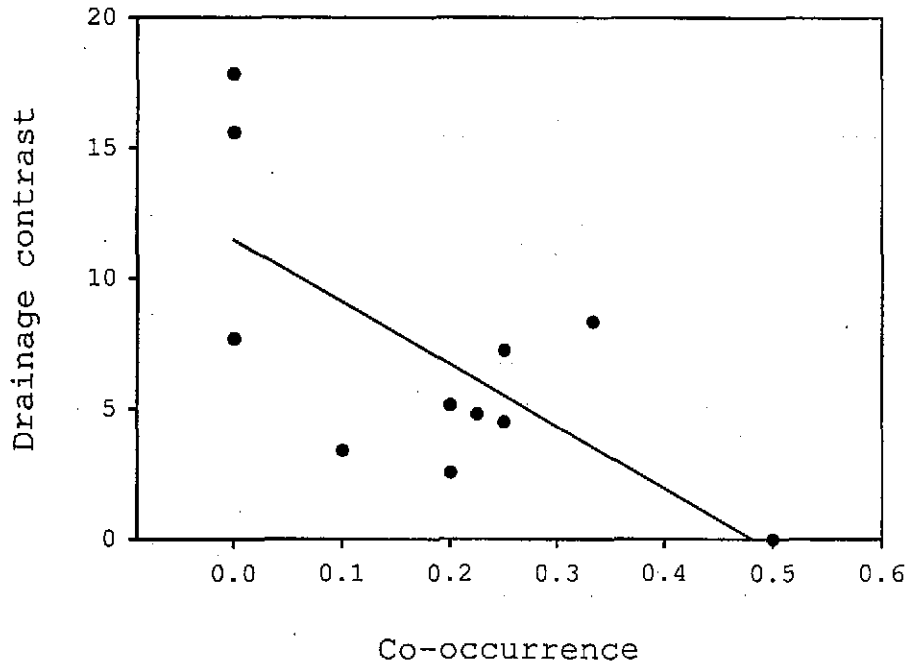
Figure 12. Relationship between drainage contrast and the degree of co-occurrence in the _Girardinichthys_ group using an optimized distance Wagner tree. $r^2 = 0.477$, $F(1,9) = 16.89$, $p < 0.018$.

was consistent with the model of sympatric speciation, as intercepts of slopes were greater than zero. It is difficult, however, to assure that speciation occurred in sympatry. Recently diverged species that tended to co-occur could have differentiated in allopatry and joined later. All I can assure is that recently diverged species exhibit a broad degree of sympatry, which accords with predictions derived from adaptive radiations of groups (Schluter 1996). Both groups presented ancient splits and recent splits with no co-occurrence, suggesting that divergence in allopatry, though infrequent, happened as well. In addition, such events indicate that species splits leading to permanent

geographical isolation occurred throughout the phylogenetic history of goodeids. This is consistent with the fact that the Mesa Central has a long history of geological instability (Barbour 1973). Vicariant species might have originated from mountain building as in the case of the Valley of Mexico (Barbour 1973), whose enclosure is believed to have separated ancestral species into G. viviparus and G. multiradiatus (Miller & Smith 1986). Hidrographic compartmentalization of drainages as result of volcanic and tectonic events could also have caused the geographical isolation of ancestral species. For example, Characodon spp. became isolated in the Mesa del Norte far from the location of the other goodeids (Smith & Miller 1986). Other species like Ataenobious toweri, Ilyodon spp., Allodontichthys spp., Xenotaenia resolanae, and Xenoophorus captivus became isolated in the Pacific and Atlantic slopes, separated from the interior of the Mesa Central (Miller & Uyeno 1980; Uyeno et al. 1983). These general patterns accord with the view that the geographical distribution of goodeids reflects former rather than present water routes (Miller & Smith, 1986).

Drainage differentiation in the Goodea group seems to have changed randomly in relation to co-occurrence, whilst it likely occurred as an incidental consequence of colonization of available drainages in the Girardinichthys group. A difference like this might be the result of distinct geographical histories. Most species of the Girardinichthys group inhabit drainages within the Mesa Central, whereas several species of the Goodea group occupy drainages outside. This differential distribution might have expanded or contracted geographical ranges distinctly, so that intrinsic differences in the rate of geographical range changes over time might have promoted differences in drainage differentiation between groups. However, there is no information, to my knowledge, that can support this hypothesis. Yet, there is suggestive evidence that many basins in the Mesa Central were interconnected (Barbour 1973) thus forcing ancestral populations to share environments. This is consistent with the fact that scattering of the points of the relationship between co-occurrence and node height was greater in the Goodea group (Figure 7) than in the Girardinichthys group (Figure 8),

45

suggesting that the determining factors of co-occurrence were more homogeneous in the Girardinichthys group.

The finding that habitat contrast did not show any association with co-occurrence suggests that the distribution into several habitat types might have resulted from stochastic processes. However, this does not explain why drainage contrast, but not habitat contrast, tended to show an association with co-occurrence. One possible explanation is that drainages are house only to a few species, whereas habitat types are shared among many species. Therefore it is easier to separate species into discrete groups according to drainage than according to habitat.

The results reported here depend on the phylogeny used, which in this case might not be an exact reflection of the true relationships amongst goodeid species, as allozymes may be less accurate than other genetic markers (Avise 1994). Yet, the division of goodeids into two main clades with the Girardinichthys group including species endemic to the Mesa Central is unlikely to change with new phylogenetic reconstructions. Moreover, the current classification accords with the view that geographical history and animal distributions are closely related (Rosen 1978). Therefore, it is expected that any phylogeny reflects general patterns of geographical distribution.

In conclusion, these results suggest that most species splits occurred in sympatry with unusual events of vicariance occurring throughout the diversification of goodeids. Drainage differentiation evolved randomly in relation to species coexistence in the Goodea group, whilst in the Girardinichthys group it was associated with differences in local opportunities between distinct drainages. Thus, geography seems to have played a closer influence on cladogenesis of the Girardinichthys group than on the Goodea group, though in none of the groups the high degree of coexistence shown by species, seems to have depended on drainages differences.

CHAPTER 4

MATING PREFERENCES AND WATER TURBIDITY ARE ASSOCIATED WITH THE GEOGRAPHICAL VARIATION OF A SEXUALLY DIMORPHIC TRAIT

Summary

Males of <u>Xenotoca</u> <u>variata</u>, a goodeid species, have 'speckles' on their flanks, the number of which varies within and amongst populations. I tested whether the differences of mate choice of females from four localities were associated with the geographical variation of speckles. I also tested if any observed preferences disappeared in turbid water, as males with many speckles tend to inhabit clear waters. Contrasting males, having more or fewer speckles were presented to females in clear and turbid water. Females from the localities with the most speckled males preferred to stay close to males with more speckles if the water was clear. These findings support the hypothesis that mate choice of females contributes to the geographical variation in speckles, and that the expression of this preference is dependent upon the transmitability of the signal.

1. Introduction

The variation of sexual traits might be adaptive and arise as a result of both physical and biotic conditions. For example, efficiency of transmission of mating signals depends on environmental conditions, as well as on the properties of the implicated sensory systems (Endler 1992). The joint effect of these variables may bias the direction of the evolution of sexual traits. The process is termed sensory drive which suggests that sensory conditions drive evolution in specific directions. Sexual selection by female choice, on the other hand, enhances differences between populations and facilitates reproductive isolation (Butlin & Ritchie 1994).

There are numerous studies that demonstrate the existence of differences in sexual traits amongst different populations. For example, guppies (Poecilia retticulata) differ in male colour patterns (Endler 1983), occurrence and duration of courtship displays (Luyten & Liley 1985) and criteria of female mate choice (Endler & Houde 1995). These differences are presumably associated with the strength of predation occurrence (Endler 1978) and the degree of turbidity of water (Luyten & Liley 1985). In transparent headstreams where predation pressure is relatively low, males are more conspicuous, display for gaining access to females and in tests of female choice of mate are more successful than their turbid water-dwelling counterparts (Luyten & Liley 1991). By contrast in turbid lowland streams males depend on gonopodial thrusting for inseminating females, and in tests of female choice of mate are more successful than headwater males. Differences in female mating preferences are also associated with differences in male colour patterns (Houde 1988; Houde & Endler 1990).

Other sources of evidence are the studies on male coloration in cichlid populations of lake Victoria (Seehausen et al. 1997). In areas that are becoming turbid as a result of eutrophication, the diversity of species is being reduced in comparison to those areas that are still intact (Seehausen et al. 1997). Turbidity of water interferes with female mate choice and relaxes sexual selection, and therefore it can block the mechanisms of reproductive isolation (Seehausen et al. 1997).

48

By contrast in regions in which water is transparent, sexual selection may be stronger and lead rapidly to reproductive isolation giving rise to species flocks (Seehausen et al. 1997).

Although sexual selection drives the evolution of secondary sexual characters, the evidence reported above indicates that physical and biotic conditions may set limits to the extent to which epigamic traits evolve. In this chapter I tested whether differences in mating preferences operating under distinct conditions of water turbidity, correlated with the geographical variation in colour patterns amongst populations of Xenotoca variata (Goodeidae).

Goodeids are viviparous fish native to the Mexican plateau with unique reproductive features. There are 17 genera comprising about 40 species (Hollenberg & Wourms 1994; Smith & Miller 1980; Smith & Miller 1987). They lack an intromittent organ; the modification of their anal fin, named spermatopodium (Fitzsimons 1972), is less specialised than a gonopodium, which is an intromitent organ in poeciliids (Constanz 1984; Constanz 1989). Thus, copulation in goodeids occurs with female co-operation. Goodeids inhabit every kind of freshwater habitat, offering an opportunity to study the influence of physical and biotic variables on the evolution of secondary sexual characters.

Earlier studies suggested that X. variata individuals are sexually dimorphic in colour patterns (Fitzsimons 1972). Preliminary observations indicated that males have bright and colourful spots on their flanks (speckles), whose number seems to vary amongst populations. In addition, geographical variation in number of speckles seems to be affected by clearness of water (Fitzsimons 1972).

This study aimed, thus, to determine whether female preferences were associated with differences in the number of speckles in 4 populations of X. variata, and whether transparency of water affected female preferences measured as the visits and time spent with pairs of males.

The hypotheses of this study were the following.
I. Hypothesis: differences in female choice of mate generate population

differences in number of speckles.

(A). Prediction: females from populations with more speckles would prefer males with more speckles.

II. Hypothesis: the turbidity of water affects the ability of females to choice a mate.

(A). Prediction: females would show stronger preferences in clear water than in turbid water (on the assumption that females get a clearer view in clear than in turbid water).

2. Methods

(a) Study fish

I used 24 females and 12 males from each of four populations that represent the variation in number of speckles (see below). The fish were sexually mature individuals determined by the presence of a fully developed spermatopodium in males. Fish were collected in spring, autumn and winter from 1997 to 1998 from the following localities using a 4.5-m long seine: (1) a spring-fed pond at El Ojo de Agua, near the city of San Francisco del Rincón, State of Guanajuato. Besides the presence of X. variata, there were also other two species of goodeids: Goodea atripinnis and Zoogoneticus quitzeoensis, and one poeciliid species, Poecilopsis infans. The depth of water varied from about 40 cm to 60 cm, and the bottom was covered with Elodea sp. (2) A stream pond near Rancho Viejo, State of Guanajuato. In this locality there were also G. atripinnis and P. infans. The depth of water could vary throughout the year as a result of seasonal patterns of rain; in late spring depth varied from 20 to 100 cm. Apparently there were no perennial aquatic plants. (3) The South-Eastern shore of Lake Cuitzeo, near the village of Coro, State of Michoacán. In this site the depth of water varied from 20 cm to 30 cm; there were patches of water hyacinth, particularly on the shore. There were also other goodeids: G. atripinnis, Skiffia bilineata and Zoogoneticus quitzeoensis; carp (Ciprynus carp) and P. infans were present also. (4) The North-Eastern shore of Lake Zacapu, near Zacapu, State of Michoacán. The depth of water varied from 50 cm to 95 cm. There

was seemingly perennial vegetation in the shallow waters. Other goodeids inhabiting the Lake were: _Skiffia multipunctata_, _Goodea atripinnis_, _Allotoca_ sp., _Hubsina turneri_ and _Zoogoneticus quitzeoensis_. Populations of this study are separated by obvious physical barriers.

In the laboratory, I kept the fish in aquaria in groups of five to ten. After one week, I separated them by sex in enclosures in outdoor pools at the Instituto de Ecología.

(b) Method to measure water turbidity

Measurements of water turbidity were made 2-3 times throughout the year between 1998 and 1999. Sampling dates were chosen taking in account the following restrictions: (1) density of goodeids populations can change over seasons (Macías Garcia et al. 1998), (2) in the rainy season turbidity of water varies constantly. For these reasons and to avoid a large variation in turbidity measurements, samples were taken in spring, autumn and only once in winter (see Appendix D). In each locality I scored turbidity from 3 samples of water taken at 3 points separated by a distance that varied between 1 m and 20 m, depending on the type and extension of the waterbody. Samples were collected at approximately half the depth of the water column, and analysed _in situ_ with a water quality kit whose readings are given in Jackson units (Sawyer et al. 1994). I also measured underwater light, where it was possible, with a secchi disk to determine the depth to which photosyntetic organisms are limited (Jones & Reynolds 1996). The disc (20 cm diameter) was lowered using a line and when it ceased to be visible, the depth of the line was recorded.

(c) Data collection

After at least 1 month of isolation, I videotaped the fish to obtain permanent pictures, that could later be used to count the number of speckles in males and females. Because paired fish were matched in median fin area and body length, measurements of these variables were necessary as well. The fish were videotaped on each flank of their body

51

for approximately 5 min using a Sony Hi8 camcorder. They were individually put into a small glass tank (25 x 4.6 x 20 cm) with water. Lighting was provided by two 60-W lamps of daylight colour separated 20 cm and about 15 cm above the tank. I used this method of videotaping because it is commonly recommended by aquarists to obtain reliable photographs, but also to have a permanent archival record of each individual's phenotype. Before removing the fish, I measured its standard length (the extension in cm from the tip of the snout to the caudal peduncle where rays of the caudal fin arise) with a transparent ruler. This measure was used for subsequent calibration.

I digitised video frames from each flank of each fish, and took the following quantitative measurements using the software Image Tool:
1. Body area, total length (cm^2) of the body;
2. Median fin index, proportion of the area of median fins to body area;
3. Number of speckles on each flank. I used the average of the two flanks of each individual and divided it by body area.

Once I had counted the speckles I calculated the standard deviation of speckles for each population. To allocate male fish to a pair, I used the criterion that they did not differ in number of speckles more than half the standard deviation of their population. In this way I homogenized the relative differences in number of speckles, so that females from different populations could see the variation typical of each population. In addition, males were as similar as possible in standard length (0.19 ± 0.16 cm, mean difference).

I used six pairs of male fish from each population, and every single pair was faced with each one of four females, one from each population. The presentations occurred in clear and turbid water, so there were 192 trials in total, two for each female. Mature tap water was used as clear water and bentonite (0.3 g/l) for turbid water (Abrahams & Kattenfeld 1997). With this concentration of bentonite the turbidity of the water was of 120 JTU, which is similar to that of Lake Cuitzeo.

Each pair of males along with the four females were observed in the morning and afternoon in clear water and then in turbid water or vice versa. Such order was alternated every day. The pair of males and the four females were randomly selected every day as well as the order of presentation of the pair of males to the females. The experiments were conducted between 1000 h and 1700 h in a room with controlled temperature (22 ± 1°C) and a constant photoperiod (12 h light).

The trials were conducted in one aquarium measuring 98.5 x 24 x 23 cm. Light came from two 60-W lamps of daylight colour 30 cm above the tank. Divisions were marked in the tank resulting in three imaginary compartments 32 cm long each. They served to indicate precisely where the fish were. I placed 2 males, each one in a transparent plastic bag (26.8 x 27.9 cm., Reynolds, USA), in the extreme compartments the night before the trials. I put the males in plastic bags because previous observations had indicated that males and females could interact, and presumably the males attempted to copulate with females (C. Macías García, pers. comm.). I also placed a neutral female (from Coro, State of Michoacán) in a similar bag in the middle of the tank. This procedure allowed the males to become acclimatised to the experimental condition, as well as getting used to seeing each other. The water of each bag was continuously aerated by electric pumps using air stones. Irrespective of the experimental condition the water of the tank was clear, aged tap water.

I put the experimental female in turn in a bag similar to those used for the other fish and placed it in the central compartment. After ten minutes of acclimatisation, the female was released. Once she had crossed the divisions and entered each male's compartment, the following 15 min two observers scored the following behaviours:
1. Visits, the number and duration of occasions the female crossed the divisions and entered the males' compartments;
2. Approaches, the occasions the female was approximately at a distance of <1 body length from the male;
3. Copulation attempts, the occasions the female was close to the male

53

and he attempted to copulate with her by approaching her until he was at her side.

Then I replaced the female in her bag whilst I swapped the males to control for side preferences by the female. After that, the female was released again and the behaviour scored for 15 min.

(d) Data analysis

I evaluated frequency distributions of speckles in males from each population. I then compared the mean number of speckles using one-way ANOVA (Sokal & Rohlf 1995) to test for differences amongst population means. I added together the behavioural responses of each female to each male throughout the 30 min of observation.

I used the score difference between responses to the male with more speckles and the male with fewer speckles to estimate female preferences for speckles. To test for significant differences, I applied a factorial analysis of variance 2 x 4 x 4 with 6 replicates (i. e., two values of water turbidity, four populations of males, four populations of females). In addition, I included standard length of males as a covariate. Also, I evaluated the regression of the difference in time spent by females with each male on the difference in speckles within pairs of males. Before, an analysis of covariance (ANCOVA) was applied to detect whether slopes of females' response differed from zero.

3. Results

There were significant differences amongst male localities in the number of speckles (Figure 13; $F(3,77) = 62.3$ $p < 0.001$). The population from Coro was not included in female choice experiments because it was not found until later, and I also suspect that fish from this spring-fed pond migrate in the rainy season into Lake Cuitzeo. The histograms of the relative frequency of speckles (Figure 14) showed that the males from Rancho Viejo and San Francisco had the largest variance. Note that with the exception of Rancho Viejo, the other localities did not seem to fit into a normal distribution of speckles. In fact, Coro and Cuitzeo showed right-skewed distributions. Body size (i. e., standard length)

54
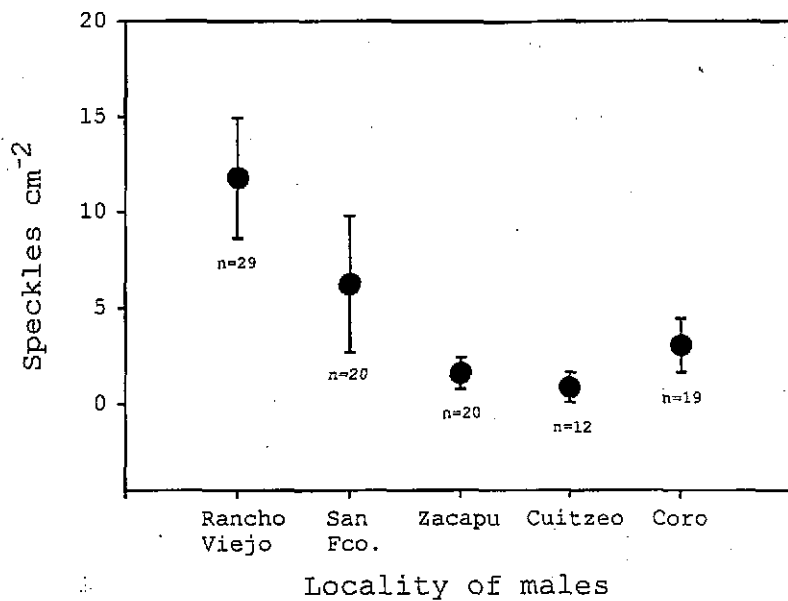
Figure 13. Variation in number of speckles (average of the two flanks) corrected for body area between populations of X. variata. Data are the means ± SEM.

did not differ amongst male localities, $(F_{(3,44)} = 2.3, p > 0.05)$. There was no difference in the number of speckles amongst female localities (Figure 15; $F_{(3,92)} = 0.91 p > 0.1$), but differences in body size (i.e., standard length) between them were detected $(F_{(3,92)} = 3.58, p < 0.05)$: Cuitzeo>Zacapu>San Francisco>Rancho Viejo.

There was not apparent relationship between number of speckles and turbidity of water (Figure 16). The locality of Rancho Viejo had an unusually high level of water turbidity that avoided any relationship between number of speckles and water turbidity.

Copulation attempts were discarded from the statistical analysis because they occurred few times. The number of visits to the males by females was marginally affected by water turbidity (Table 5), there being a positive difference in clear water (Figure 17). The number of approaches to the males was not significantly affected by either locality of males, locality of females or turbidity of water (Table 6). The duration of visits to males was affected by neither locality of males, locality of females nor turbidity of water (Table 7).

A way of assessing mate choice is evaluating the regression of the time females spent between males on the difference in speckles in pairs of males (see Milinski & Bakker 1990; Morris et al. 1996). The ANCOVA revealed that the slopes of response of the females (the four localities) to Zacapu males in clear water were significantly different
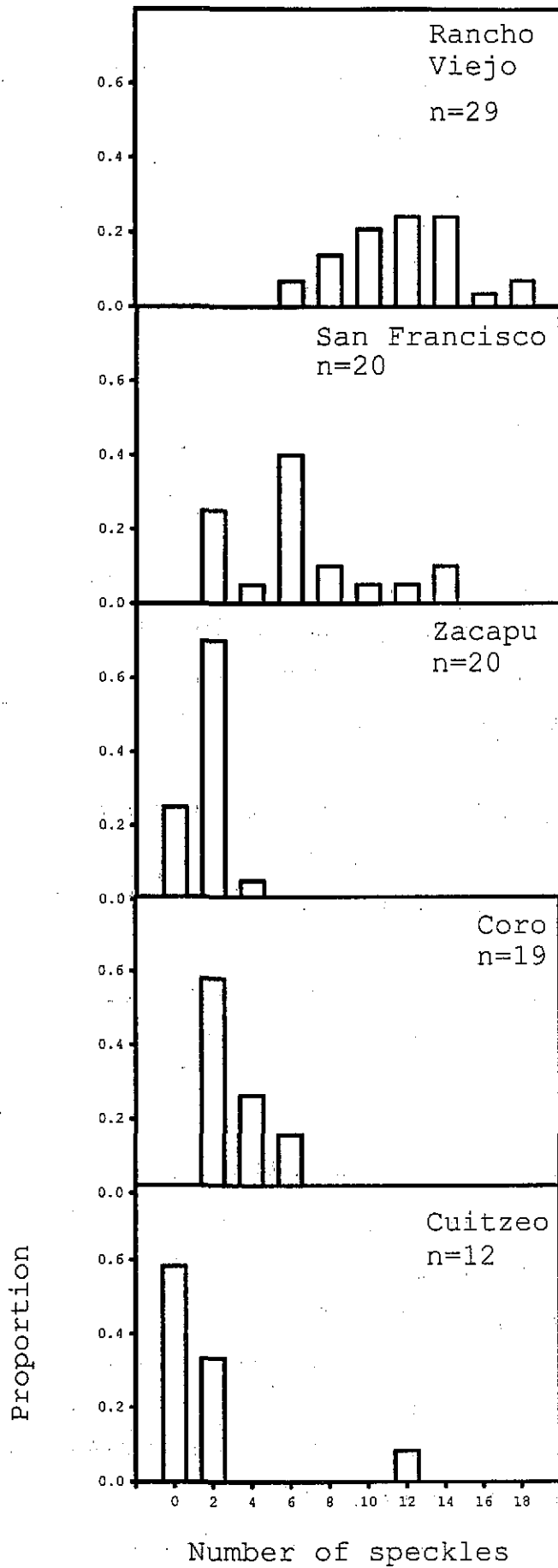
Figure 14. Population differences in frequency distributions of fish according to number of speckles. For comparison, the frequencies were expressed as relative frecuencies
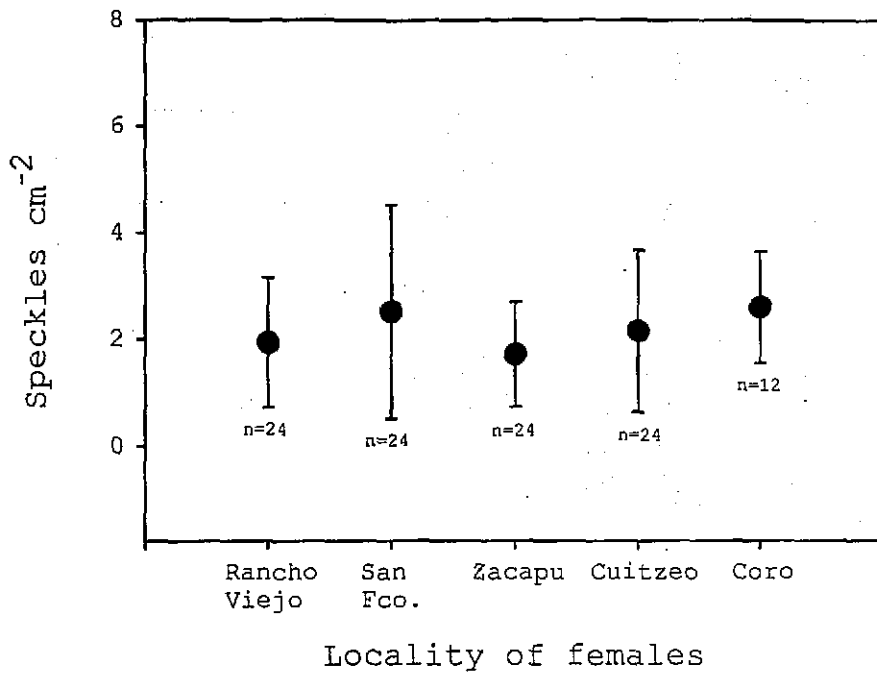
Figure 15. Number
of speckles (ave-
rage of the two
flanks) in
females. Data are
means ± SEM.

from zero $(F(1,16) = 5.82, p < 0.029)$. However, the mean response (i.e.,

the intercepts) did not differ between the four female localities
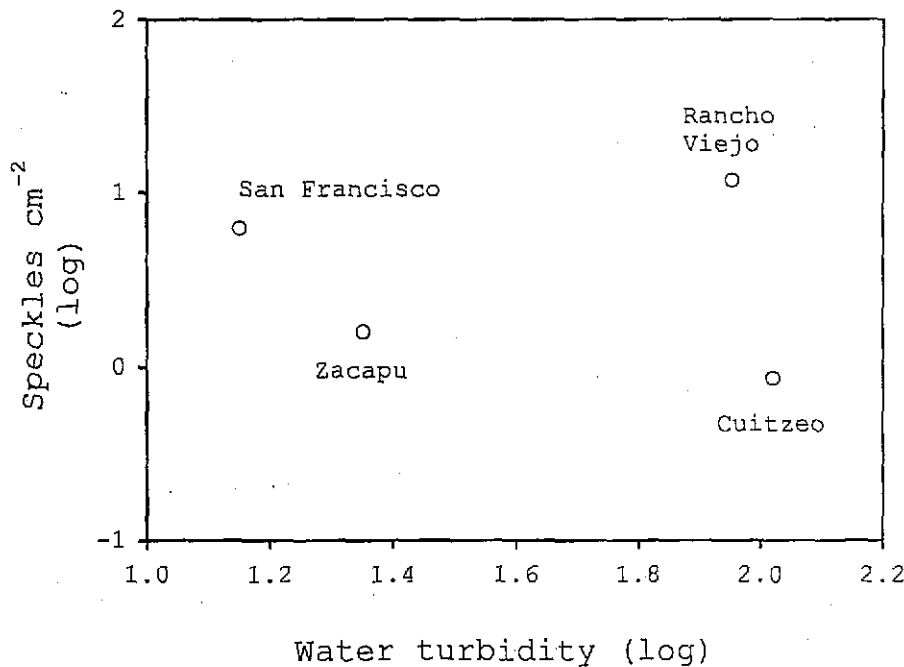
Figure 16.
Relationship
between number of
speckles in male
fish of four
localities and
turbidity of water.

Table 5. Analysis of variance on the difference in visits in pairs of males (more speckles-fewer speckles)

Factor	df	ss	ms	F	p
Male size	1	2.83	2.83	0.04	0.833
Males locality	3	228.01	76.00	1.19	0.314
Females locality	3	112.97	37.66	0.59	0.621
Turbidity of water	1	245.26	245.26	3.85	0.051
Females*water	3	162.22	54.07	0.85	0.469
Males*females	9	441.59	49.07	0.77	0.644
Males*water	3	54.02	18.01	0.28	0.838
Males*females*water	9	348.26	38.70	0.61	0.789
Error	159	10124.00	63.67		
Total	191	11716.33			

n= 24 females from each population
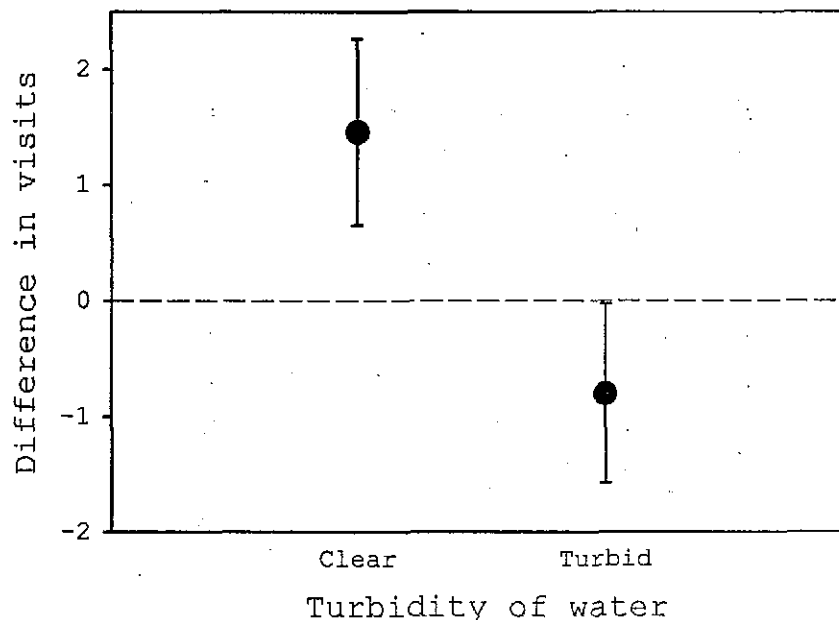
Figure 17. The number of visits in two conditions of water turbidity. Data are the mean difference (scores of males with fewer speckles subtracted from scores of males with more speckles) ± SEM, n= 96

$(F(3,16)= 0.21, p> 0.05)$, nor the slopes differed from each other $(F(3,16)= 0.78, p> 0.05)$ as expected from the corresponding ANOVA. Subsequent analyses showed significant regressions in the following cases: Rancho Viejo females against Zacapu males $(r^2= 0.663; F(1,4)= 7.87, p< 0.05$; Table 8; Figure 18) and San Francisco females against Zacapu males $(r^2= 0.846; F(1,4)= 21.97, p< 0.01$; Table 8; Figure 19). Regarding the other male localities for clear water and all of them for turbid water, the ANCOVA did not show significant differences for any of

Table 6. Analysis of variance on the difference in approaches in pairs of males (more speckles-fewer speckles)

Factor	df	ss	ms	F	p
male size	1	255.53	255.53	3.17	0.077
Males locality	3	208.37	69.46	0.86	0.462
Females locality	3	595.04	198.35	2.46	0.065
Turbidity of water	1	150.52	150.52	1.87	0.174
Females*water	3	564.69	188.23	2.34	0.076
Males*females	9	412.67	45.85	0.57	0.821
Males*water	3	54.27	18.09	0.22	0.879
Males*females*water	9	444.69	49.41	0.61	0.784
Error	159	12807.47	80.55		
Total	191	15425.92			

n= 24 females from each population

Table 7. Analysis of variance on the difference in visit duration in pairs of males (more speckles-fewer speckles)

Factor	df	ss	ms	F	p
Male size	1	56048	56048	0.25	0.621
Males locality	3	942330	314110	1.38	0.252
Females locality	3	184261	61420	0.27	0.848
Turbidity of water	1	469	469	0	0.964
Females*water	3	1071914	357305	1.57	0.200
Males*females	9	2306304	256256	1.12	0.350
Males*water	3	453639	151213	0.66	0.576
Males*females*water	9	2193353	243706	1.07	0.390
Error	159	36296454	228280		
Total	191	43487524			

n= 24 females from each population

Table 8. Summary of regression analysis of female choice of the speckled male against the difference in speckles in pairs of males from Zacapu in clear water

Female locality	Slope	S	t	p
Rancho Viejo	523.3	186.5	2.81	0.049
Cuitzeo	274.5	476.3	0.58	ns
San Francisco	508.6	108.5	4.69	0.009
Zacapu	9.4	155.3	0.06	ns
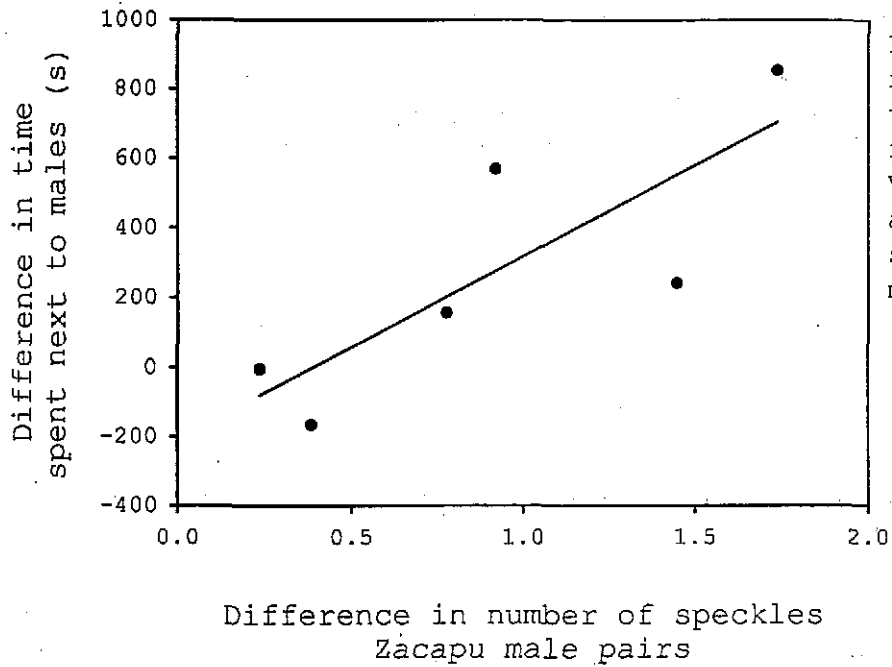
Figure 18. The relationship between the time spent by females from Rancho Viejo between males and the difference in speckles in pairs of males.

Difference in number of speckles
Zacapu male pairs
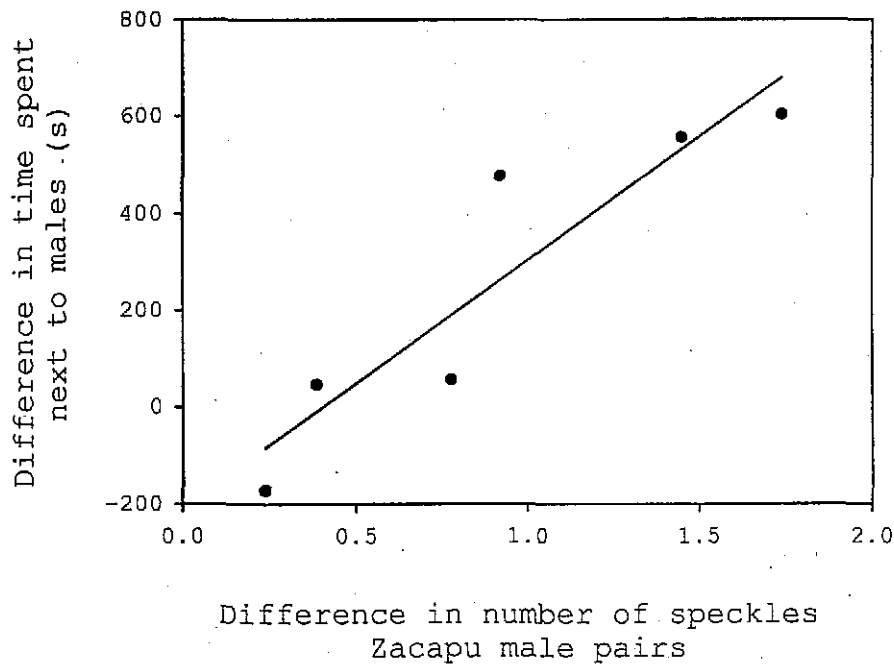
Figure 19. The relationship between the time spent by females from San Francisco between males and the difference in speckles in pairs of males.

Difference in number of speckles
Zacapu male pairs

the parameters. Because there was the possibility that the number of speckles was confounded with body size of males, I repeated the preceding analysis of regression using residuals. I regressed scores of visit duration on body size (i.e., standard length). Then, I calculated the expected score for the mean body size of males and added this number

to each female residual. I did not find apparent differences between the two analyses.

4. Discussion

The result of the number of visits in pairs of males and the time females spent next to speckled males, suggests that the dimorphism in speckles could have evolved by female choice of mate. It is unlikely that such dimorphism had arisen from other causes (e.g., ecological differences between sexes; for a review on this issue see Hedrick 1989; Slatkin 1984). Nor it seems that speckles are weapons that males use for competing over females, for they could not injure other fish.

When I took the difference in time that females stayed next to each male as an index to preferences (Milinski & Bakker 1990; Morris et al. 1996), the difference in the preferences were associated with differences amongst female localities relative to speckles. In other words, the choosiest females came from localities with the highest number of speckles. This finding supports the hypothesis that differences in female choice of mate generate population differences in number of speckles. However, it is remarkable that the analysis of variance did not reveal any significant variation of either the number of visits or their duration relative to locality of females. This does not mean that the preference for speckles does not exist, but that it cannot be estimated from these measurements. Alternatively, the magnitude of the expression of the preference may be small under the conditions of this experiment, which makes it difficult to measure.

In spite of the fact that males from Rancho Viejo and San Francisco had more speckles than those from the other localities, they were not as attractive to females as Zacapu males. There are three possible reasons to explain this discrepancy. First, females might have seen something else that correlated with the number of speckles and that only existed amongst Zacapu males. Because air stones continuously aerated the water, males were engaged in maintaining their posture and hence they had no chance to display courtship. Thus, behavioural differences do not seem to have intervened. Second, the colour of the

61

speckles could differ amongst localities; in fact, at first sight they varied from blue to golden in every locality. Even though it is difficult, with the present data, to rule out the possibility that colour had affected females' preferences, it is unlikely that females from Rancho Viejo and San Francisco prefer alien males in natural conditions. Third, the criterion to form the pairs was that males differed in speckles half the standard deviation of their specific locality. With this procedure I intended to control the differences amongst localities. Admittedly, it was difficult to have enough fish to complete the pairs, so they had a wider variation in the number of speckles than it was intended. It is possible that females from Rancho Viejo and San Francisco could not distinguish between their males because these had so many speckles that the apparent difference was imperceptible to them. Zacapu males, on the other hand, had a discrete number of speckles that contrasted well with the rest of their body. Therefore, it is possible that in this case, Rancho Viejo and San Francisco females could distinguish small differences. In addition, the error in counting speckles was greater for Rancho Viejo and San Francisco than for the other localities. Because we know little about the visual capabilities of goodeids, this interpretation should be taken cautiously, although it is the simplest one.

Geographical variation in conspicuous ornaments owing to preferences, as reported here, was also obtained in guppies (Poecilia reticulata) (Endler & Houde 1995; Houde 1988) and threespine sticklebacks, Gasterosteus aculeatus (Boughman 2001). But which factors do originate divergence in female preferences? Although variation in male traits has been studied in several species (Arnegard et al. 1999; Butlin & Ritchie 1994; Foster 1995; Hill 1994; Houde & Endler 1990; Seehausen 1997), in fish there are few works on the role of physical environments in conspicuousness of male traits (Endler 1991; Endler 1995; Luyten & Liley 1985; Luyten & Liley 1991; Seehausen et al. 1997). In aquatic environments there are constraints on the development of conspicuous ornaments caused by turbidity of water and general conditions of illumination (Endler 1992; Levine & MacNichol, Jr. 1979;

Lythgoe & Partridge 1991). Therefore, the geographical differences in female preferences may be the result of adaptations to local ecological conditions, so that male signals adjust to female perceptual sensitivity (Boughman 2001). Although in the present study the relationship between number of speckles and water turbidity was not definite, the finding that females visited the males with more speckles in clear water supports the hypothesis that visual ornaments are better seen in clear water.

Apart from the sampling of water turbidity being small, there were other factors that hid the expected relationship. For example, the unexpected high degree of turbidity of the stream-pond where Rancho Viejo fish came from, may be related to the use of the pool as a reservoir for watering cattle and for villagers to bath. In addition, there is a small dam that blocks the free flow of water, and hence contributes to the concentration of suspended materials. When I measured turbidity upstream (behind the dam, ca. 50 m) the water was clear and flowing. Therefore, it is possible that before humans settled down there, the water was clear as can be expected in a mountain stream (Pielou 1998). Consequently, the relative clearness of water might also have contributed to the variation in number of speckles amongst populations of X. variata.

The finding that females visited males with fewer speckles in turbid water may not be statistically significant because the distance of the deviation from zero (i.e., no preference) is small (Figure 17). Alternatively, it might be that in such conditions females are attracted by other kinds of traits than speckles, which likely cannot be seen in turbid waters. It is expected indeed that speckles as well as other colourful traits cannot evolve in turbid water, as the physical environment plays a relevant role in the transmission of signals (Johnstone 1997). For example, the loss of red colouration in stickleback populations has been correlated with stained waters (Reimchen 1989).

The results do not rule out the possibility that there might be other traits that females can use to assess a potential mate. Indeed,

63

there is evidence that assortative mating by body size occurs in other goodeid species (Bisazza 1997), although this trait is unlikely to be the major objective of sexual selection, and even less, that it could be the predominant mating strategy in goodeids as suggested earlier (Bisazza 1997). If this were the case, it would be useless and expensive for males to display courtship to females (Fitzsimons 1972), and to bear speckles which not only can attract potential predators, but also which may be costly to produce. The most plausible interpretation is that speckles as well as their colour, and body size account for mate choice of females. Recent studies revealed that multiple traits may co-evolve with their respective preferences (Brooks 1999), so that this alternative may be explored in goodeids.

In conclusion, these findings support the hypothesis that geographical variation in speckles is maintained by differences in preferences of female choice. These preferences are expressed in clear water. Other selective forces such as risk of predation, need to be considered to have a complete picture of the geographical variation of this trait.

CHAPTER 5

PREDATION RISK IS ASSOCIATED WITH THE GEOGRAPHICAL VARIATION OF A
SEXUALLY SELECTED TRAIT

Summary

Male secondary sex traits may increase the risk of predation because
mating signals can be perceived by potential mates but also by
predators. Males of Xenotoca variata, a freshwater fish, show on their
flanks bright and colourful spots, 'speckles', which vary geographically
in number. In this study I tested whether aquatic snakes, Thamnophis
melanogaster, which co-occur with X. variata distinguished from a pair
of fish the one with more speckles. I faced pairs of fish which differed
in the number of speckles with the snakes in clear water and in turbid
water. The snakes preferred approaching the males with more speckles to
approaching the males with fewer speckles. The preference did not change
with the turbidity of the water, although it was stronger in clear water
than in turbid water. The snakes also headed first for the males with
more speckles. These findings indicate that predation risk by T.
melanogaster may select against speckles.

1. Introduction

Geographical variation in behavioural and morphological traits is a feature common to many species (Endler & Houde 1995; Foster 1995; Foster 1999; Hill 1994; Magurran 1998; Marchetti 1993; Price 1998; Wiley 1991). It may be the result of two main factors: (1) stochastic processes (e. g., genetic drift) (2) selection pressures (i. e., natural and sexual selection). Generally, genetic drift is an evolutionary force effective when population size is small (Maynard Smith 1998). In contrast, selection pressures are continuously acting on populations.

Secondary sexual traits are deemed to represent a balance between sexual selection, primarily by mate choice, and natural selection (e. g., by predators, Endler 1992). In addition, their development and the perception by conspecifics is constrained by particular environmental features, for local conditions determine how effective the propagation of signals is.

Despite the recognition of the importance of these factors, there are few studies that have assessed the importance of natural selection on variation in the magnitude and form of sexual signals, and their interaction with environmental conditions (Andersson, 1994).

Goodeids are a livebearing family of fish native to Central Mexico. Most of their species show sexual dimorphism in both morphological and behavioural traits (Fitzsimons 1972; Macías Garcia 1990; Miller & Smith 1986; Nelson 1975). The geographical distribution of goodeids corresponds to a great extent to that of Thamnophis melanogaster (Conant 1963), an aquatically specialized garter snake that preys underwater by long distance attacks on leechees, tadpoles and fish (Drummond 1983). Temperature may limit its foraging behaviour to hours when water is above 19 °C up to 29 °C (Manjarrez & Drummond 1996). T. melanogaster rely on visual and chemical stimuli to attack their prey (Drummond 1979; Drummond 1985), preferentially those of the largest sizes that they usually ingest (Macías Garcia & Drummond 1995). They may also prey more frequently on one sex than on the other. For example, there is a study that shows that they consume more male fish than female fish from populations of Girardinichthys multiradiatus (Macías Garcia et

66

al. 1998). These males have larger median fins than the females and it
is thought that this character handicaps males in their evasive actions
(Macías Garcia & Jimenez 1994). It is possible that other goodeid males
may have a disadvantage because of bearing dimorphic sexual traits. For
example, males of _Xenotoca variata_ show bright and colourful spots
(hereafter referred to as speckles) on their flanks that could attract
T. melanogaster snakes.

Preliminary observations did not reveal an evident relationship
between turbidity of water and number of speckles (see chapter 4). There
are some localities like a spring-fed pond called Mintzita near Morelia,
Mich., and Zacapu Lake, Mich., where water is clear, yet male fish show
few speckles on their flanks. This suggests there are other factors
apart from sexual selection and water turbidity which are involved in
the geographical variation of number of speckles. One possibility is
predation by snakes, specifically _T. melanogaster_, which may be able to
see speckles in clear water.

The hypotheses and predictions of this study were the following
I. Hypothesis: speckles attract predators.
(A). Prediction: fish with fewer speckles would be common in localities
    where snakes are abundant, and fish with more speckles would be
    common in localities where snakes are rare;
(B). Prediction: snakes would approach and attack fish with more
    speckles than fish with fewer speckles.
II. Hypothesis: environmental conditions affect the ability of snakes to
    see the speckles of fish.
(A). Prediction: the number of approaches and attacks by snakes to fish
    with more speckles would be more frequent in clear than in turbid
    water.

2. Methods
(a) Census of snakes in the field
Snakes count took place from October 1996 to October 1998 in (1) the
southern shoreline of Lake Cuitzeo, Mich., (2) the southern shoreline of
Lake Zacapu, Mich., (3) a stream pond in Rancho Viejo, Gto., (4) a

spring-fed pond in San Francisco del Rincón, Gto., (5) a ditch in San Pedro Casacuarán, Gto., (6) a spring-fed pond called 'Mintzita' in Morelia, Mich., (7) a spring-fed pond in Coro in the southeast of Lake Cuitzeo, Mich. Because the snakes (Thamnophis spp.) do not forage when the water temperature is low (Manjarrez & Drummond 1996) and is hence being difficult to catch or see them, I made the censuses in spring, autumn and once in mid-winter. In Lake Cuitzeo I located one transect of 50 m long in the shoreline. I counted the snakes along bands of about 3 m wide on each side of the line transect. I walked down the bands searching and counting snakes by turning over rocks, logs and sheets of scrap paper. In the other localities I observed the bodywater from the shoreline, in a radius of about 4 m, for approximately 5 min and counted the number of snakes seen diving. I used this method because in these localities transects were inappropriate (e.g., the stream pond in Rancho Viejo had steep banks; the spring-fed pond in San Francisco del Rincón was surrounded by rough terrain; Lake Zacapu was surrounded by rough terrain with a lot of weed).

(b) Study snakes

I used 14 snakes (6 females, 4 males and 4 which were inadvertantly released without being sexed) measuring on average 42.9 ± 4.7 cm (± SD; snout-vent length). They were collected as described above from shorelines of reservoirs at Acambay (n=5), San Pedro Tlaltizapan (n=8) and El Cerrillo (n=1), State of México. The snakes were brought to the laboratory and housed individually in opaque plastic boxes (21 X 7 X 9 cm deep) containing water dishes and paper substrate. Boxes had holes on their lateral sides for aeration.

(c) Study fish

I used 12 male fish from two populations: a spring-fed pond in San Francisco del Rincón, State of Guanajuato; and Lake Zacapu, State of Michoacán. The fish were collected using a 4.5-m long seine and brought to the laboratory and maintained in enclosures (100 x 70 x 50 cm made of Nylon) in 2 adjacent (585 x 345 x 50 cm depth) concrete outdoor pools.

68

One month before the experiment, the fish were moved into the laboratory and maintained in groups of 4-5 in 40-l gravel-lined aquaria provided with filter and aeration. Photoperiod was maintained constant (12/12h dark/light) and room temperature fluctuated between 25° C and 27 °C. Before the experiment was performed, I put the male fish individually in a small glass tank (25 x 4.6 x 20cm) with water. I videotaped the fish on each flank for approximately 5 min; I used a Sony Hi 8 camcorder and two 60-W daylight colour lamps hung 15 cm above the tank. Before removing the fish, I measured their standard length (i.e., the extension from the tip of the snout to the caudal peduncle where rays of the caudal fin arise) with a transparent ruler. I used this measure for pairing fish and for subsequent calibration. The fish were 4.2 ± 0.24 cm long (mean ± sd) and were paired (3 pairs per population) according to standard length. Fish of each pair differed on average 0.17 ± 0.12 cm (± sd) in standard length. Although fish within pairs differed in number of speckles 30.17 ± 50.35 (mean difference ± sd), it was difficult for the observer to know the direction of the difference because the speckles were not counted until the end of the experiment, so that the recording of behaviour remained unbiased.

(d) Experimental tank

The tank (56 x 36 x 19.5 cm; see Figure 20) was divided into three compartments, two of them (four sides 26 x 18 x 7 x 25 cm) were adjacent and each one contained 2.5 l of water. The third (five sides: 36 x 30 x 25 x 25 x 30 cm) contained 8.7 l of water. The fish, which were in the small compartments, could not see each other or the snake, which was in the largest compartment and could see the fish through a one-way mirror. The water of the three compartments was continously aerated with air stones using an electric pump. Two 60-W daylight colour lamps were approximately 15 cm above the compartments of the fish. A black cardboard was used to cover the snake's compartment so that it was darkened. The contrast so produced allowed the snake to see the fish. The front side of the experimental tank had a small opening in the

cardboard through which the observer could watch and record the snake's behaviour.
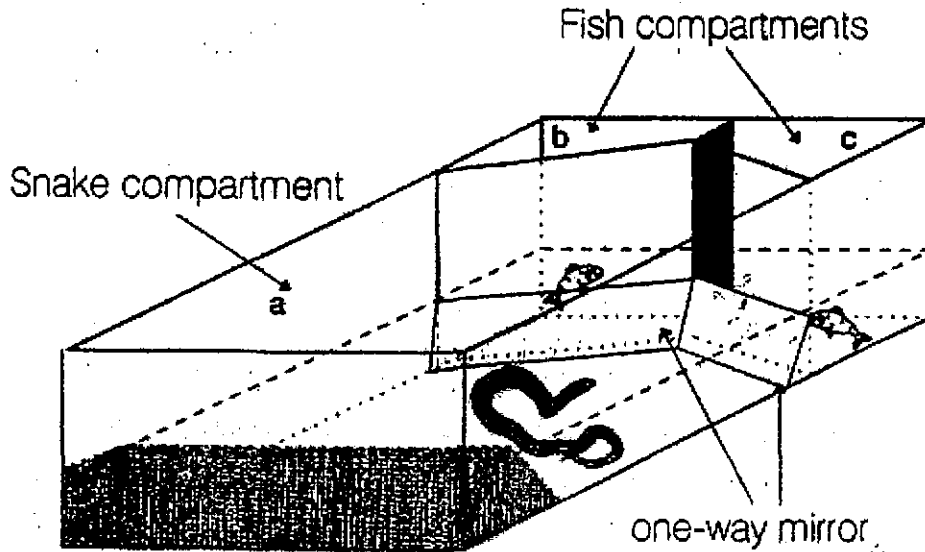
Figure 20. Tank for experiments. Taken from Macías Garcia et al. 1994

(d) Experimental design

The 6 pairs of male fish were separated into two equal groups. Four or 5 snakes were randomly assigned to each pair of the first group (i.e., the first pair of male fish was presented to 5 snakes separately; the second pair was presented to each one of 5 different snakes; and the third pair was presented to 4 different snakes). A week later the pairs of the second group of fish were presented to the same snakes. In addition, each snake was tested in clear and turbid water. So each subject was tested 4 times. In the rest of this typescript I will refer to the first and second time as the first trial and to the third and fourth time as the second trial. Mature tap water was used for clear water and bentonite (0.3g/l) for turbid water (Abrahams & Kattenfeld 1997) which was 120 JTU near to the number recorded in Lake Cuitzeo. Snakes assigned to each pair of fish were tested in a random order. The pair of fish used each day was randomly selected and the condition of the water (i.e., clear and turbid) was reversed every day.

(e) Experimental procedure

Trials took place from 0900 h to 1400 h and from 1500 h to 1800 h in an otherwise darkened room, with a room temperature of 27° ± 1°C to motivate snakes to forage (Manjarrez & Drummond 1996). To stimulate aquatic foraging, fish odour was added to the pool. This was done by keeping two to three fish in the pool the night before experimentation or between the morning and the afternoon each day.

The night before the trials, the cages with the snakes were placed upside down on a piece of acrylic put on the experimental tank. The experimental fish's compartments were filled with either clear or turbid water on the eve of the trial too. The following day the decoy fish were removed, and the corresponding experimental fish put in their compartments and allowed to settle for 10 min before observations commenced. The snakes were removed, except the first one to be tested, which was moved onto a suface, in the snake's compartment, made of two pieces of wood approximately 8 cm apart and put just above the water level. The trial began when the snake's cage was slowly taken up, leaving the snake free to get into the water, and the tank was covered with the cardboard. The observer waited for the snake's head to appear underwater, which, with the exception of one snake, occurred within the next few minutes. Then, the positioning (see below) to either fish marked the start of the observation that lasted 5 min, in which the following behaviours were recorded (Drummond 1979; Drummond 1980).
1. Orientation, rapid projection of the head that brings it to a terminal position pointing toward a fish;
2. Approach, stealthy movement in the direction of a fish;
3. Attacks, projection of the head with the jaws opened, moving rapidly forward. These are recognized by a sudden acceleration of the head in a single direction.

A maximum of 30 min was allowed for the snakes to respond to the presence of the fish. Snakes that did not respond within the 30 min were tested at the end of the series or the following day. After the 5 min of observation, the fish were exchanged to correct for any potential side preference; then, another 5 min of observation were conducted. The snake

71

was left in its compartment whilst the exchange of fish took place. All snakes were left unfed during 7 days before the trials started. At the end of each trial, they were fed thawed fish placed on their feeding dishes in their home cages.

Once I finished the experiment, I digitised video frames from each flank of each fish, and took morphometrics using a computer program (Image Tool) and measured the standard length of fish for calibration. I took the following measurements to express the number of speckles per cm^2, and to measure how similar paired fish were as to fin size.

1. Body area, total extension of the body;
2. Median fin index, proportion of the area of median fins to body area;
3. Number of speckles on each flank. I used the average of the two
   flanks of each fish in subsequent analyses.

(f) Statistical analysis

Because orientation frequently preceded approaches, the frequencies of both behaviours were pooled, hereafter called visual fixations, as suggested by Macías Garcia and Drummond (1995). Visual fixations and attacks were analysed using factorial analysis of variance (trial(2) x turbidity of water(2) x type of fish(2)). I also tested for the effect of fish population and snake population on visual fixations. The frequency of the first visual fixation to any of the fish and the frequency of the first attack by the snakes was analysed using G-tests. The relationship between snake counts and number of speckles was analysed using descriptive statistics.

3. Results

The count of snakes revealed apparent differences amongst localities (Table 9). The general pattern was that localities with snakes, though in variable numbers, had male fish with fewer speckles. Thamnophis melanogaster and T. eques were more abundant in Lake Cuitzeo than in the other localities. No snakes were seen in Rancho Viejo, Gto., and few were seen in San Pedro Casacuarán, Gto., in which male fish show many

Table 9. Data of censusing of snakes and number of speckles of male fish (*Xenotoca variata*)

Date	Locality	*Thamnophis melanogaster*	*Thamnophis eques*	Speckles (mean ± sd)
02 October 1996	Lake Cuitzeo	39[1]	5	0.86 ± 0.78
05 October 1996	Lake Zacapu		1	1.60 ± 0.82
09 November 1996	Lake Zacapu	1		
07 October 1998	Lake Zacapu	2		
17 October 1998	Lake Zacapu	2		
04 April 1998	Rancho Viejo			11.78 ± 3.15
05 February 1998	San Francisco			6.24 ± 3.54
22 March 1998	Coro	1		3.06 ± 1.40
10 October 1996	Mintzita	2		
30 May 1997	Casacuarán	1		

[1]Seven of them had fish in their stomach contents

speckles (pers. obs.). Male fish from Mintzita, Mich. show very few speckles (pers. obs.).

One snake was discarded from the analyses because she did not respond in the second trial, and only responded in clear water in the first trial. Also, I dropped, using a table of random numbers, another snake to have full cells that facilitated the statistical analysis.

The response of the snakes to the fish did not differ amongst fish pairs ($F(5,88) = 0.52$, $p=0.76$). Turbidity of water and type of fish contributed significantly to the variation of visual fixations (Table 10) which were significantly more in clear water than in turbid water (Figure 21; $F(1,67) = 5.99$, $p=0.017$), and more frequently directed to fish with more speckles than to fish with fewer speckles (Figure 22; $F(1,67) = 4.62$, $p=0.035$). There was no interaction between turbidity of

Table 10. Analysis of variance on visual fixations

Factor	df	ss	ms	F	p
Snakes	11	164.27	14.93	1.29	0.252
Trials(snakes)	12	74.64	6.22	0.54	0.884
Turbidity of water	1	69.57	69.57	5.99	0.017
Type of fish	1	53.68	53.68	4.62	0.035
Water*type of fish	1	0.07	0.07	0.01	0.940
Error	67	777.74	11.61		
Total	93	1133.33			
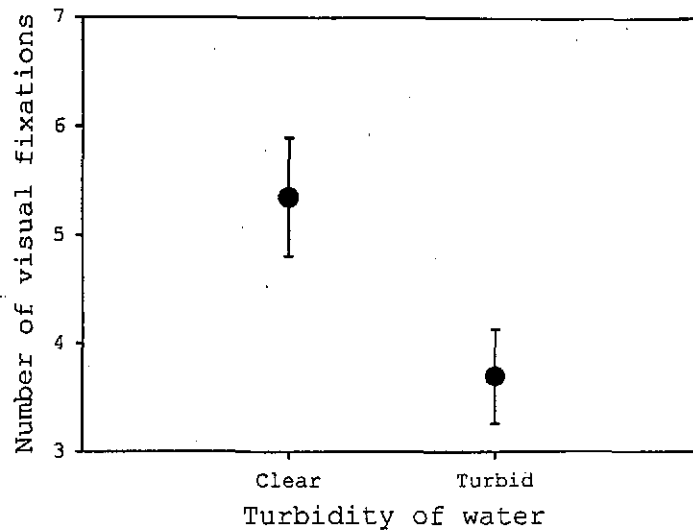
Figure 21. The number of visual fixations by snakes on fish in two conditions of water turbidity. Data are means ± SEM, n= 12
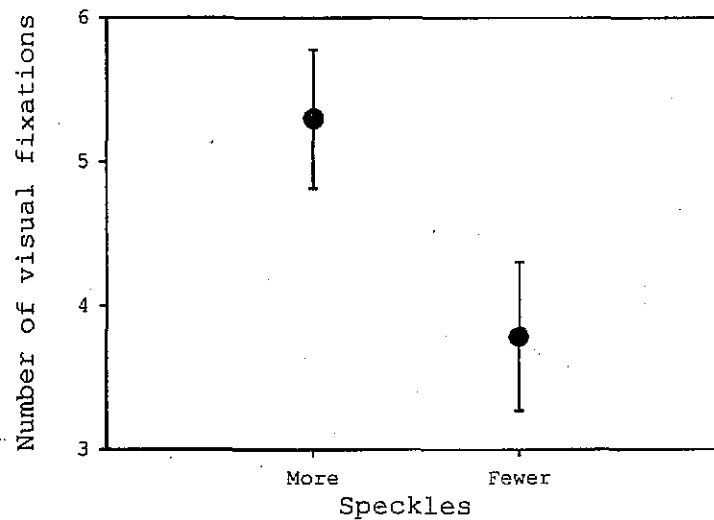
Figure 22. The number of visual fixations by snakes on two types of male fish. Data are means ± SEM, n= 12

water and type of fish (Table 10) suggesting that snakes approached speckled fish both in clear water and in turbid water. As to attacks, neither trial, turbidity of water nor type of fish contributed significantly to the variation of this behaviour (Table 11).

Table 11. Analysis of variance on attacks

Factor	df	ss	ms	F	p
Snakes	11	17.364	1.579	0.72	0.720
Trials(snakes)	12	29.361	2.447	1.11	0.368
Turbidity of water	1	0.011	0.011	0.00	0.944
Type of fish	1	0.015	0.015	0.01	0.934
Water*type of fish	1	0.866	0.866	0.39	0.533
Error	67	147.862	2.207		
Total	93	201.415			

The first visual fixation of snakes was significantly directed to fish with more speckles than to fish with fewer speckles (Table 12). The frequency of the first attack to fish with more speckles was not statistically different from the first attack to fish with fewer speckles (Table 13). There was no correlation between number of speckles and standard length of male fish that might suggest that snakes used body size rather than speckles to approach fish. The regression of number of speckles on standard length of fish with more speckles was null ($r^2 = 0.0$; $F(1,4) = 0$, $p > 0.1$). Interestingly, the number of speckles decreased with the length in fish with fewer speckles ($r^2 = 0.84$; $F(1,4) = 27.33$, $p < 0.007$).

Table 12. The frequency of the first visual fixation to male fish by snakes

| Trial | Fish with | | G-test* | p-value |
	More speckles	Fewer speckles		
1	17	6	5.4824	0.0192
2	15	9	1.516	0.2182

*The null hypothesis was that snakes would view both types of fish with equal frequency (0.5)

Table 13. The frequency of the first attack to male fish by snakes

| Trial | Fish with | | G-test* | p-value |
	More speckles	Fewer speckles		
1	6	3	1.0194	0.3127
2	10	9	1.0527	0.8185

*The null hypothesis was that snakes would attack both types of fish with equal frequency (0.5)

4. Discussion

The results showed that _T. melanogaster_ snakes distinguish from a pair the fish with more speckles, suggesting that potential predation by this snake may be associated with the geographical variation in number of speckles amongst populations of _X. variata_. This conclusion is supported

by the finding that localities with many snakes tended to be inhabited by male fish with fewer speckles. However, this tendency should be treated with caution because the size of the census sample was small, and the possibility existed that the localities were not equitable. Yet, the present results accord with other studies that show that male secondary sex traits may increase the risk of predation (Burk 1982; Macías Garcia & Jimenez 1994; Magnhagen 1991; Moodie 1972). Therefore, natural selection caused by predation may constrain the development of sexual dimorphic traits.

The direction of the visual fixations by snakes did not change with water turbidity, so it is clear that they are capable of seeing the speckles of the fish even in conditions where the transmission of the signal is attenuated, though the probability for a speckled fish to be approached in turbid water diminished significantly. Although there are previous reports on freshwater fish that indicate geographical variation in sexually selected traits caused by predation (Endler 1987; Luyten & Liley 1985; Luyten & Liley 1991), this is one of the few empirical studies to suggest a direct influence of predation on variation in a secondary sexual trait.

It is unlikely that the preference of snakes for speckled males is because of the higher exposure of males in the field than of females, or because there are more males than females (for a review of these alternatives see Andersson 1994). I believe that the snakes would capture more males because the speckles on their flanks attract them. In localities like Mintzita, State of Michoacán, male fish ($\underline{X}$. $\underline{variata}$) have few speckles despite the clearness of the water. The difference from other localities consists of the fact that there are snakes. This circumstantial evidence (Mintzita is a rocky spring where it is difficult to count or catch snakes) together with the results of the present study, suggest that predation risk selects against bright colours in male goodeid fish. This conclusion is consistent with previous studies on guppies (Endler 1980; Endler 1983).

The finding that in the second trial the snakes discovered males with more speckles as rapidly as males with fewer speckles, indicates

that snakes learned quickly to respond to both types of fish. This cannot be attributed to an identification of the fish, for these were different from the first trial. Perhaps, the snakes got more accustomed to foraging. Indeed, attacks in the first trial were twice as many as in the second trial.

In conclusion, these results indicate that predation risk may select against speckled males and hence may relax the effect of sexual selection. If so, San Francisco females would be expected to be choosier than Mitzita females. Further experiments are needed to test this prediction.

CONCLUSIONES GENERALES Y ESTUDIOS FUTUROS

1. Introducción

Este estudio partió de un análisis amplio de la historia evolutiva de rasgos dimórficos sexuales en los goodeidos, y fue complementado con una evaluación de las fuerzas selectivas subyacentes de la variación geográfica de un carácter dimórfico sexual en una sola especie. Esta aproximación incluyó varios aspectos de la ecología de los goodeidos no analizados hasta ahora. Analizó algunas características abióticas y bióticas del ambiente donde los goodeids viven y algunas fuerzas selectivas que formaron su diversidad. ¿En qué contribuye este trabajo a la comprensión de la ecología de los goodeidos?

2. La selección sexual y la diferenciación adaptativa

Los resultados de los análisis comparativos sugieren que las aguas corriente favorecen la evolución de señales de apareamiento. Empero la especiación por adaptación a ambientes locales (i.e., aguas corrientes y lagos) parece no haber sido frecuente. Quizás la tendencia a que los caracteres dimórficos sexuales aparezcan en peces que habitan aguas corrientes simplemente sea una consecuencia de la heterogeneidad de estos ambientes. Varían de arroyos pequeños pasando por manantiales a ríos. Si el fenotipo masculino promedio es determinado por un equilibrio entre la selección natural y sexual, entonces habría tantos fenotipos como hay diferencias en la magnitud de la selección natural aun cuando la selección sexual permaneciera constante o viceversa (Lande 1987). Por consiguiente, un estudio que incluya la variación de aguas corrientes todavía puede revelar alguna asociación entre caracteres dimórficos y ambientes específicos. Necesariamente deben ser incluidas mediciones de la velocidad y turbiedad del agua. Además, variables como la transmisión de luz a través de la columna de agua y la densidad de depredadores potenciales pueden dar información útil sobre las restricciones y posibilidades de exhibir caracteres dimórficos sexuales.

La idea tradicional sobre la riqueza de especies de los goodeidos es que experimentaron una radiación adaptativa (Miller & Smith 1986)

promovida por nichos libres. Sin embargo, es difícil que los nichos ecológicos sean suficientes para explicar la presencia de especies distintas (Coyne & Orr 1999), por lo que otros factores deben ser involucrados. El estudio presentado aquí sugiere que la selección sexual, medida como la magnitud del dimorfismo del tamaño de las aletas dorsales, podría estar positivamente asociada con el número de especies por género. Ya que no todas las especies fueron incluidas, un estudio futuro debe incorporar todos ellas para confirmar la asociación. Sin embargo, queda por probar si existe atracción de la hembra por el tamaño de las aletas dorsales o si prefieren otro rasgo (e.g., tamaño del cuerpo, el cortejo) que esté correlacionado con el tamaño de las aletas dorsales.

3. La función de la geografía en la coexistencia de las especies
Aunque tradicionalmente ha sido considerado que los goodeidos experimentaron una radiación adaptativa (Miller & Smith 1986), no había estudios que probaran cualquier predicción derivada de esta hipótesis. El hallazgo de que la simpatría fue el modo geográfico predominante de especiación apoya la hipótesis de la radiación adaptativa. El hecho de que la dispersión después de la separación de especies fue ligera, también sugiere que la divergencia de las especies debe de haber involucrado mecanismos de aislamiento reproductivo que ocurren con las especies ancestrales coexistiendo. La investigación de estos mecanismos resta por hacerse. Un requisito de la simpatría es que las especies deberían diferir ecológicamente (Schluter 1996). Al analizar la relación entre diferencias fenotípicas y la geografía (i.e., hábitat y cuenca), no encontré ninguna evidencia de que diferencias de hábitat o cuenca fueran necesarias para la coexistencia de especies de goodeidos. Pero los análisis basados en otros métodos serían necesarios para descartar explicaciones alternativas.

El escenario de la ocupación de los goodeidos de México Central se desconoce. Tres posibles mecanismos por los que grupos de especies se originaron (Mayr 1988), también pueden explicar la radiación de las especies de los goodeidos. (1) Colonización repetida; los lagos en

79

México Central son localidades que pudieron favorecer la colonización múltiple. De hecho, los aterinidos, que habita la Meseta Central, pasaron por este proceso (Echelle & Echelle 1984). (2) la especiación allopátrica en porciones diferentes de lagos; aunque no hay ninguna evidencia que apoye esta hipótesis, no puedo dejar fuera la posibilidad de que los goodeidos hayan experimentado este tipo de especiación en localidades como el Lago de Zacapu. (3) La acumulación de especies a través de una fusión de cuerpos de agua dulce previamente existentes; ésta es una explicación creíble dada la historia geológica de México Central. Estudios extensos son necesarios para separar estas posibilidades.

3. La selección natural y sexual moldean los caracteres sexualmente dimórficos

Muy probablemente el número de espejos de peces macho de X. variata difiere entre las poblaciones porque las hembras varían en la magnitud de su preferencia por los espejos. A pesar de la importancia de este hallazgo, es necesario llevar a cabo experimentos en los que puedan cambiarse el número y color de los espejos. Los resultados de tales estudios ayudarán a evaluar (a) la preferencia por los machos espejosos de Zacapu, (b) la fuerza de la preferencia y (c) las capacidades sensoriales de las hembras. Sin embargo, una explicación completa de la función de los espejos en peces machos debe incluir una demostración de una relación significativa estadísticamente entre el número de espejos y el éxito reproductivo. Además de esta demostración, es deseable identificar el mecanismo(s) de selección sexual. Independientemente de la explicación de la preferencia de la hembra por los espejos del macho, creo que el sesgo sensorial es un mecanismo, aunque no evolutivo, que debe tomarse en cuenta para justificar la presencia de la preferencia.

Otra investigación tiene que ver con evaluar los costos de los espejos. Como el estudio presente ha mostrado, los peces macho que exhiben espejos probablemente serán vistos por culebras más frecuentemente, incluso en agua turbia. Analizar el efecto de una gama amplia de valores de turbiedad del agua en la probabilidad de que los

80

espejos se vean, es algo que queda por hacerse. Pero el riesgo de predación es sólo una desventaja, y otros costos también deben evaluarse. Por ejemplo, los espejos pueden ser enérgicamente costosos para los machos porque éstos pueden gastar tiempo cuidándose de los depredadores. De manera similar, un resultado indirecto de tener espejos puede ser demandas nutritivas más altas, o experimentar una mortalidad juvenil alta. Estas hipótesis pueden ser probadas con observaciones en el campo y con experimentos en el laboratorio.

Aunque he presentado evidencia de que el riesgo de depredación puede estar seleccionando contra los espejos, debería determinarse el contenido estomacal de las culebras. Estas observaciones permitirán determinar si las culebras atrapan peces espejosos en el campo. Además, se espera encontrar una correlación negativa entre la conducta de cortejo y la densidad de depredadores con base en los resultados de la elección de pareja de apareamiento.

En conclusión la variación en el número de espejos entre las poblaciones de X. variata es el resultado de tres factores: (1) la elección de pareja por la hembra; (2) el riesgo de predación; Y (3) la turbiedad del agua. Estos hallazgos son un ejemplo de cómo la selección natural y sexual pueden explicar la magnitud de caracteres conspicuos. Vale la pena investigar la contribución relativa de cada factor en detalle. Una aproximación que permitiría ventajas es modelar la función de cada factor (e.g.,., vea Maynard Smith 1978). Las predicciones de tales escenarios podrían contrastarse con datos de las observaciones en el campo y experimentos de laboratorio.

Para resumir, el objetivo principal de este trabajo fue colectar información sobre la especiación de los goodeidos. Restringí la investigación para estudiar la especiación por selección natural y sexual; los mecanismos que involucran deriva quedan por ser analizados. Los hallazgos sugieren que la selección sexual promueve especiación, mientras que la divergencia por adaptación a las condiciones generales de hábitat es poco frecuente. Aunque la simpatría fue el modo geográfico predominante de especiación, no encontré evidencia de que diferencias de hábitat o cuenca tuvieran una función en la coexistencia de los

goodeidos. Por otra parte, la variación en la elección de pareja de la hembra contribuye a la variación en el número de espejos en peces macho de X. variata. Es notorio que la atracción de la hembra por los espejos dependa de la claridad del agua. Además de este factor, es muy probable que el riesgo de depredación por culebras, Thamnophis melanogaster, ejerza una selección contra los espejos. Para continuar llevando a cabo estudios en goodeidos, es necesario proteger sus habitas. Según un estudio reciente hecho por nuestra Universidad, aproximadamente 40% de los ríos están contaminados severamente. Debemos acompañar la investigación con estrategias para proteger el hábitat de los goodeidos.

LITERATURA CITADA

Abrahams, M. & Kattenfeld, M. 1997. The role of turbidity as a
constraint on predator-prey interactions in aquatic environments.
Behavioral Ecology and Sociobiology 40, 169-174.

Andersson, M. B. 1994. Sexual Selection. Princeton: Princeton University
Press.

Arnegard, M. E., Markert, J. A., Danley, P. D., Stauffer, J. R., Jr.,
Ambali, A. J. & Kocher, T. D. 1999. Population structure and colour
variation of the cichlid fish Labeotropheus fuelleborni in a recently
formed archipelago of rocky habitat patches in southern Lake Malawi.
Proceedings of the Royal Society of London, Series B 266, 119-130.

Arnqvist, G., Edvardsson, M., Friberg, U. & Nilsson, T. 2000. Sexual
conflict promotes speciation in insects. Proceedings of the National
Academy of Sciences 97, 10460-10464.

Avise, J. C. 1994. Molecular Markers, Natural History and Evolution. New
York: Chapman & Hall.

Baerends, G. P., Brouwer, R. & Waterbolk, H. TJ. 1955. Ethological
studies on Lebistes reticulatus (Peters) I. An analysis of the male
courtship pattern. Behaviour 5, 249-334.

Barbour, C. D. 1973. A biogeographical history of Chirostoma (Pisces:
Atherinidae): a species flock from the Mexican plateau. Copeia 533-556.

Barraclough, T. G., Harvey, P. H. & Nee, S. 1995. Sexual selection and
taxonomic diversity in passerine birds. Proceedings of the Royal Society
of London, Series B 259, 211-215.

Barraclough, T. G., Hogan, J. E. & Vogler, A. P. 1999. Testing whether
ecological factors promote cladogenesis in a group of tiger beetles
(Coleoptera: Cicindelidae). Proceedings of the Royal Society of London,
Series B 266, 1061-1067.

Barraclough, T. G. & Vogler, A. P. 2000. Detecting the geographical
pattern of speciation from species-level phylogenies. American
Naturalist 155, 419-434.

Barraclough, T. G., Vogler, A. P. & Harvey, P. H. 1998. Revealing the factors that promote speciation. Philosophical Transactions of the Royal Society of London, Series B 353, 241-249.

Bisazza, A. 1997. Sexual selection constrained by internal fertilization in the livebearing fish Xenotoca eiseni. Animal Behaviour 54, 1347-1355.

Boughman, J. W. 2001. Divergent sexual selection enhances reproductive isolation in sticklebacks. Nature 411, 944-948.

Bouton, N., Witte, F., van Alphen, J. J. M., Schenk, A. & Seehausen, O. 1999. Local adaptations in populations of rock-dwelling haplochromines (Pisces: Cichlidae) from southern Lake Victoria. Proceedings of the Royal Society of London, Series B 266, 355-360.

Brooks, D. R. & McLennan, D. A. 1992. Historical ecology as a research program. In: Systematics, Historical Ecology, and North American Freshwater Fishes (Ed. R. L. Mayden), pp. 76-113. Stanford: Stanford University Press.

Brooks, R. C. V. 1999. Multiple sexual ornaments coevolve with multiple mating preferences. American Naturalist 154, 37-45.

Brown, J. H. & Gibson, A. C. 1983. Biogeography. St. Louis: Mosby.

Brönmark, C. & Hansson, L. A. 1998. The Biology of Lakes and Ponds. Oxford: Oxford University Press.

Burk, T. 1982. Evolutionary significance of predation on sexually signalling males. Florida Entomologist 65, 90-104.

Butler, M., Schoener, T. W. & Losos, J. B. 2000. The relationship between sexual size dimorphism and habitat use in greater Antillean anolis lizards. Evolution 54, 259-272.

Butlin, R. K. & Ritchie, M. G. 1994. Behaviour and speciation. In: Behaviour and Evolution (Ed. P. J. B. Slater & T. R. Halliday), pp. 43-79. Cambridge: Cambridge University Press.

Butlin, R. K. & Tregenza, T. 1998. Levels of genetic polymorphism: marker loci versus quantitative traits. Philosophical Transactions of the Royal Society of London, Series B 353, 187-198.

Chesser, R. T. & Zink, R. M. 1994. Modes of speciation in birds: a test of Lynch's method. Evolution 48, 490-497.

Clutton-Brock, T. H. & Vincent, A. C. J. 1991. Sexual selection and the potential reproductive rates of males and females. Nature 351, 58-60.

Conant, R. 1963. Semiaquatic snakes of the genus Thamnophis from the isolated drainage System of the Río Nazas and adjacent areas in Mexico. Copeia 473-499.

Constanz, G. D. 1984. Sperm competition in poeciliid fishes. In: Sperm Competition and the Evolution of Animal Mating Systems (Ed. R. L. Smith), pp. 465-485. Orlando: Academic Press.

Constanz, G. D. 1989. Reproductive biology of poeciliid fishes. In: Ecology and Evolution of Livebearing Fishes (Poeciliidae) (Ed. G. K. Meffe & F. F. Snelson, Jr.), pp. 33-50. Englewood Cliffs: Prentice Hall.

Coyne, J. A. & Orr, H. A. 1999. The evolutionary genetics of speciation. In: Evolution of Biological Diversity (Ed. A. E. Magurran & R. M. May), pp. 1-36. Oxford: Oxford University Press.

Darwin, C. 1871. The Descent of Man, and Selection in Relation to Sex. Princeton: Princeton University Press.

Doughty, P. 1996. Statistical analysis of natural experiments in evolutionary biology: comments on recent criticisms of the use of comparative methods to study adaptation. American Naturalist 148, 943-956.

Drummond, H. 1979. Stimulus control of amphibious predation in the Northern water snake (Nerodia s. sipedon). Zeitschrift für Tierpsychologie 50, 18-44.

Drummond, H. 1980. Aquatic Foraging in Some New World Natricine Snakes: Generalists and Specialists and Their Behavioral Evolution. Doctor of Phylosophy Thesis. The University of Tennessee, Knoxville.

Drummond, H. 1983. Aquatic foraging in garter snakes: a comparison of specialists and generalists. Behaviour 86, 1-30.

Drummond, H. 1985. The role of vision in the predatory behaviour of natricine snakes. Animal Behaviour 33, 206-215.

Echelle, A. A. & Echelle, A. F. 1984. Evolutionary genetics of a "species flock": atherinid fishes on the Mesa Central of Mexico. In: Evolution of Fish Species Flocks (Ed. A. A. Echelle & I. Kornfield), pp. 93-110. Orono: Orono Press.

Endler, J. A. 1978. A predator's view of animal color patterns. Evolutionary Biology 11, 319-364.

Endler, J. A. 1980. Natural selection on color patterns in Poecilia reticulata. Evolution 34, 76-91.

Endler, J. A. 1983. Natural and sexual selection on color patterns in poeciliid fishes. Environmental Biology of Fishes 9, 173-190.

Endler, J. A. 1987. Predation, light intensity and courtship behaviour in Poecilia reticulata (Pisces: Poeciliidae). Animal Behaviour 35, 1376-1385.

Endler, J. A. 1991. Variation in the appearance of guppy color patterns to guppies and their predators under different visual conditions. Vision Research 31, 587-608.

Endler, J. A. 1992. Signals, signal conditions, and the direction of evolution. American Naturalist 139, s125-s153.

Endler, J. A. 1995. Multiple-trait coevolution and environment gradients in guppies. Trends in Ecology and Evolution 10, 22-29.

Endler, J. A. & Houde, A. E. 1995. Geographic variation in female preferences for male traits in Poecilia reticulata. Evolution 49, 456-468.

Felsenstein, J. 1985. Phylogenies and the comparative method. American Naturalist 125, 1-15.

Fitzsimons, J. M. 1972. A revision fo two genera of goodeid fishes
(Cyprinodontiformes, Osteichthyes) from the Mexican plateau. Copeia 728-
756.

Foster, S. A. 1995. Understanding the evolution of behavior in
threespine stickleback: the value of geographic variation. Behaviour
132, 1107-1129.

Foster, S. A. 1999. The geography of behaviour: an evolutionary
perspective. Trends in Ecology and Evolution 14, 190-195.

Giller, P. S. & Malmqvist, B. 1998. The Biology of Streams and Rivers.
Oxford: Oxford University Press.

Grudzien, T. A., White, M. M. & Turner, B. J. 1992. Biochemical
systematics of the viviparous fish Goodeidae family. Journal of Fish
Biology 40, 801-814.

Harvey, P. H. & Pagel, M. D. 1991. The Comparative Method in
Evolutionary Biology. Oxford: Oxford University Press.

Hedrick, A. V. T. E. J. 1989. The evolution of sexual dimorphism in
animals: hypotheses and tests. Trends in Ecology and Evolution 4, 136-
138.

Hill, G. E. 1994. Geographic variation in male ornamentation and female
mate preference in the house finch: a comparative test of models of
sexual selection. Behavioral Ecology 5, 64-73.

Hollenberg, F. & Wourms, J. P. 1994. Ultrastructure and protein uptake
of the embryonic trophotaeniae of four species of goodeid fishes
(Teleostei: Atheriniformes). Journal of Morphology 219, 105-129.

Houde, A. E. 1988. Genetic difference in female choice between two guppy
populations. Animal Behaviour 36, 510-516.

Houde, A. E. & Endler, J. A. 1990. Correlated evolution of female mating
preferences and male color patterns in the guppy Poecilia reticulata.
Science 248, 1405-1408.

Hubbs, C. L. & Turner, C. L. 1939. Studies of the fishes of the order Cyprinodontes. XVI. A Revision of the Goodeidae. Miscellaneous Publications Museum of Zoology, University of Michigan 42, 80.

Johnstone, R. A. 1997. The evolution of animal signals. In: Behavioural Ecology: an Evolutionary Approach (Ed. J. R. Krebs & N. B. Davies), pp. 155-178. Oxford: Blackwell Science.

Jones, J. C. & Reynolds, J. D. 1996. Environmental variables. In: Ecological Census Techniques: A Handbook (Ed. W. J. Sutherland), pp. 281-316. Cambridge: Cambridge University Press.

Krebs, J. R. & Davies, N. B. 1993. An Introduction to Behavioural Ecology. Third edn. Oxford: Blackwell Scientific Publications.

Lande, R. 1987. Genetic correlations between the sexes in the evolution of sexual dimorphism and mating preferences. In: Sexual Selection: Testing the Alternatives (Ed. J. W. A. M. B. Bradbury), pp. 83-94. Chichester: Wiley.

Leroi, A. M., Rose, M. R. & Lauder, G. V. 1994. What does the comparative method reveal about adaptation? American Naturalist 143, 381-402.

Levine, J. S. & MacNichol, E. F., Jr. 1979. Visual pigments in teleost fishes: effects of habitat, microhabitat, and behavior on visual system evolution. Sensory Processes 3, 95-131.

Losos, J. B., Jackman, T. R., Larson, A., de Queiroz, K. & Rodríguez-Schettino, L. 1998. Contingency and determinism in replicated adaptive radiations of island lizards. Science 279, 2115-2118.

Losos, J. B., Warheit, K. I. & Schoener, T. W. 1997. Adaptive differentiation following experimental island colonization in Anolis lizards. Nature 387, 70-73.

Luyten, P. H. & Liley, N. R. 1985. Geographic variation in the sexual behaviour of the guppy, Poecilia reticulata (Peters). Behaviour 95, 164-179.

Luyten, P. H. & Liley, N. R. 1991. Sexual selection and competitive mating success of males guppies (Poecilia reticulata) from four Trinidad populations. Behavioral Ecology and Sociobiology 28, 329-336.

Lynch, J. D. 1989. The Gauge of Speciation: On the Frequency of Modes of Speciation. In: Speciation and Its Consequences (Ed. D. Otte & J. A. Endler), pp. 527-553. Sunderland: Sinauer.

Lythgoe, J. N. & Partridge, J. C. 1991. The modelling of optimal visual pigments of dichromatic teleosts in green coastal waters. Vision Research 31, 361-371.

Macías Garcia, C. & Drummond, H. 1995. Components of visual prey recognition by the Mexican aquatic garter snake *Thamnophis melanogaster*. Ethology 101, 101-111.

Macías Garcia, C. & Jimenez, G. 1994. Correlational evidence of a sexually-selected handicap. Behavioral Ecology and Sociobiology 35, 253-259.

Macías Garcia, C., Saborío, E. & Berea, C. 1998. Does male-biased predation lead to male scarcity in viviparous fish? Journal of Fish Biology 53, s104-s107.

Macías Garcia, C. 1990. Sexual Behaviour and Trade-Offs in the Viviparous Fish *Girardinichthys Multiradiatus*. Doctor of Philosophy Thesis. School of Biological Sciences, University of East Anglia, Norwich, England.

Maddison, W. P., Donoghue, M. J. & Maddison, D. R. 1984. Outgroup analysis and parsimony. Systematics Zoology. 33, 83-103.

Maddison, W. P. & Maddison, D. R. 1992. MacClade Version 3: Analysis of Phylogeny and Character Evolution. Sunderland: Sinauer Associates.

Magnhagen, C. 1991. Predation risk as a cost of reproduction. Trends in Ecology and Evolution 6, 183-186.

Magurran, A. E. 1998. Population differentiation without speciation. Philosophical Transactions of the Royal Society of London, Series B 353, 275-286.

Manjarrez, J. & Drummond, H. 1996. Temperature-limited activity in the garter snake Thamnophis melanogaster (Colubridae). Ethology 102, 146-156.

Marchetti, K. 1993. Dark habitats and bright birds illustrate the role of the environment in species divergence. Nature 362, 149-151.

Martins, E. P. & Lamont, J. 1988. Estimating ancestral states of a communicative display: a comparative study of Cyclura rock iguanas. Animal Behaviour 55, 1685-1706.

Maynard Smith, J. 1978. In defence of models. Animal Behaviour 26, 632-633.

Maynard Smith, J. 1998. Evolutionary Genetics. Second edn. Oxford: Oxford University Press.

Mayr, E. 1963. Animal Species and Evolution. Cambridge: Harvard Unversity Press.

Mayr, E. 1988. Toward a New Philosophy of Biology. Cambridge: Harvard University Press.

Milinski, M. & Bakker, T. C. M. 1990. Female sticklebacks use male coloration in mate choice and hence avoid parasitized males. Nature 344, 330-333.

Miller, R. R. 1986. Composition and derivation of the freshwater fish fauna of México. Anales de la Escuela Nacional de Ciencias Biológicas 30, 121-153.

Miller, R. R. & Smith, M. L. 1986. Origin and geography of the fishes of Central Mexico. In: The Zoogeography of North American Freshwater Fishes (Ed. C. H. Hocutt & E. O. Wiley), pp. 487-517. New York: Wiley-Interscience Publication.

Miller, R. R. & Uyeno, T. 1980. Allodontichthys hubbsi, a new species of goodeid fish from southwestern Mexico. Occasional Papers of the Museum of Zoology the University of Michigan 1-13.

Moodie, G. E. E. 1972. Predation, natural selection and adaptation in an unusual threespine stickleback. Heredity 28, 155-167.

Morris, M. R., Wagner, W. E., Jr. & Ryan, M. J. 1996. A negative correlation between trait and mate preference in Xiphophorus pygmaeus. Animal Behaviour 52, 1193-1203.

Nelson, G. G. 1975. Anatomy of the male urogenital organs of Goodea atripinnis and Characodon lateralis (Atheriniformes: Cyprinodontoidei), and G. atripinnis courtship. Copeia 475-482.

Nicoletto, P. F. 1996. The influence of water velocity on the display behavior of male guppies, Poecilia reticulata. Behavioral Ecology 7, 272-278.

Orr, M. R. & Smith, T. B. 1998. Ecology and speciation. Trends in Ecology and Evolution 13, 502-506.

Partridge, L. 1994. Genetic and nongenetic approaches to questions about sexual selection. In: Quantitative Genetic Studies of Behavioral Evolution (Ed. C. R. B. Boake), pp. 126-141. Chicago: The University of Chicago Press.

Pielou, E. C. 1998. Fresh Water. Chicago: The University of Chicago Press.

Podos, J. 2001. Correlated evolution of morphology and vocal signal structure in Darwin's finches. Nature 409, 185-188.

Price, T. 1998. Sexual selection and natural selection in bird speciation. Philosophical Transactions of the Royal Society of London, Series B 353, 251-260.

Radda, A. C. 1984. Synopsis der goodeiden Mexikos. Killifische Aus Aller Welt 8, 1-48.

Radda, A. C. 1985. Los Goodeidae de Méjico -una familia de peces interesantes, pero muy poco conocida por los acuariófilos. Primera parte. TI international: Acuariología en palabra e imagen 5-9.

Reimchen, T. E. 1989. Loss of nuptial color in the threespine sticklebacks (_Gasterosteus aculeatus_). Evolution 43, 450-460.

Ridley, M. 1983. The Explaination of Organic Diversity: The Comparative Method and Adaptations for Mating. Oxford: Oxford University Press.

Rosen, D. E. 1978. Vicariant patterns and historical explanation in biogeography. Systematics Zoology 27, 159-188.

Sawyer, C. N., McCarty, P. L. & Parkin, G. F. 1994. Chemistry for Environmental Engineering. Fourth edn. New York: Mcgraw-Hill.

Schluter, D. 1996. Ecological causes of adaptive radiation. The American Naturalist 148, s40-s64.

Scott, P. W. 1987. A Fishkeeper's Guide to Livebearing Fishes. London: Tetra Press.

Seehausen, O. 1997. Distribution of and reproductive isolation among color morphs of a rock-dwelling Lake Victoria cichlid (_Haplochromis nyererei_). Ecology of Freshwater Fish 6, 59-66.

Seehausen, O., van Alphen, J. M. & Witte, F. 1997. Cichlid fish diversity threatened by eutrophication that curbs sexual selection. Science 277, 1808-1811.

Slatkin, M. 1984. Ecological causes of sexual dimorphism. Evolution 38, 622-630.

Smith, M. L. & Miller, R. R. 1980. _Allotoca maculata_, a new species of goodeid fish from western México, with comments on _Allotoca dugesi_. Copeia 408-417.

Smith, M. L. & Miller, R. R. 1986. Mexican goodeid fishes of the genus _Characodon_, with description of a new species. American Museum Novitates 1-14.

Smith, M. L. & Miller, R. R. 1987. _Allotoca goslinei_, a new species of goodeid fish from Jalisco, Mexico. Copeia 610-616.

Sokal, R. R. & Rohlf, F. J. 1995. Biometry. Third edn. New York: Freman.

Trivers, R. L. 1972. Parental investment and sexual selection. In: Sexual Selection and the Descent of Man (Ed. B. Campbell), pp. 136-179. Chicago: Aldine.

Uyeno, T., Miller, R. R. & Fitzsimons, J. M. 1983. Karyology of the cyprinodontoid fishes of the Mexican Goodeidae family. Copeia 497-510.

Webb, S. A. & Miller, R. R. 1998. Zoogoneticus tequila, a new goodeid fish (Cyprinodontiformes) from the Ameca Drainage of Mexico, and a rediagnosis of the genus. Occasional Papers of the Museum of Zoology the University of Michigan 1-23.

West-Eberhard, M. J. 1983. Sexual selection, social competition, and speciation. Quarterly Review of Biology 58, 155-183.

Wiley, R. H. 1991. Associations of song properties with habitats for territorial oscine birds of eastern North America. American Naturalist 138, 973-993.

APÉNDICES

APÉNDICE A

BASE DE DATOS DE LAS OBSERVACIONES DE CORTEJO

Tabla A-1. Lista de datos de observaciones de cortejo

Fecha	Especies	Número de pares
24 de junio 1997	Allotoca dugesi	4
07 de febrero 1997	A. sp.	2
24 de septiembre 1997	Ameca splendens	1
24 de junio 1997	Characodon audax	1
24 de septiembre 1997	C. lateralis	1
07 de marzo 1997	Goodea atripinnis	2
13 de marzo 1997	G. atripinnis	2
18 de marzo 1997	G. atripinnis	1
02 de abril 1997	G. atripinnis	2
08 de abril 1997	G. atripinnis	1
16 de abril 1997	G. atripinnis	2
05 de mayo 1997	G. atripinnis	3
13 de enero 1998	G. gracilis	1
24 abril 1997	G. luitpoldi	2
29 abril 1997	G. luitpoldi	1
29 abril 1997	G. luitpoldi	2
02 de mayo 1997	G. luitpoldi	1
12 de mayo 1997	G. luitpoldi	3
04 de mayo 1998	Ilyidon whitei	1
14 de agosto 1996	I. whitei	1
24 de septiembre 1997	I. whitei	1
18 de febrero 1997	Neoophorus diazi	2
10 de marzo 1997	N. diazi	2
02 de abril 1997	Skiffi bilineata	2
24 de septiembre 1997	Xenotoca eiseni	1
13 de noviembre 1997	X. melanosoma	2
10 de marzo 1997	Xenotoca variata	2
01 de mayo 1997	X. variata	3
29 de mayo 1997	X. variata	3
31 de enero 1997	X. variata	2
7 de febrero 1997	X. variata	1
08 de abril 1997	Zoogoneticus quitzeoensis	2
16 de abril 1997	Z. quitzeoensis	1
10 de abril 1997	Z. quitzeoensis	1
14 de abril 1997	Z. quitzeoensis	1

APÉNDICE B

CONDUCTA DE CORTEJO DE LOS GOODEIDOS

Lo siguiente es una descripción de la conducta del cortejo de algunas especies de los goodeidos, particularmente de aquéllas de las que poco es conocido sobre sus señales de apareamiento. Para otras especies, existen descripciones anteriores (Fitzsimons 1972; Macías García 1990; Nelson 1975).

Characodon audax

1. Orientación. La conducta inicial del macho es orientarse hacia la hembra y seguirla cuando nada.

2. Exhibición. Después de orientarse el macho normalmente alcanza a la hembra. En esta fase la hembra se mueve lentamente cambiando su orientación de lado a lado e inclinándose ligeramente agitando la cola. El macho se posiciona bajo la hembra, e imita los mismos movimientos que ella hace. De vez en cuando, y probablemente como resultado de perder sincronización, el macho prosigue en cualquier dirección derecha o izquierda y regresa trazando un círculo delante de la hembra, hasta recuperar la posición bajo ella. Durante esta exhibición el macho vibra a una frecuencia alta y amplitud baja. Todos las exhibiciones ocurren entre la profundidad media y la superficie. He observado las mismas exhibiciones en las pozas del Instituto, donde hay una población de esta especie.

3. Movimientos de contacto. Cuando el macho está bajo la hembra, los dos entran en contacto con la aleta dorsal del macho tocando la parte ventral de la hembra. Ningún otro movimiento de contacto apareció.

Ilyodon whitei

1. Orientación. Dentro de los primeros minutos de presentación el macho puede orientarse hacia la hembra y seguirla en una posición determinada por el vigor de los movimientos de la hembra: si la hembra está nadando rápidamente, entonces el macho puede estar varios centímetros detrás de

ella; si la hembra nada despacio, el macho se queda a su lado.

2. Exhibición. Si el macho está al lado de la hembra, él puede realizar varias exhibiciones en las que el cuerpo es flexionado a una frecuencia alta y amplitud baja en el plano horizontal. Esta posición puede mantenerse durante 2 a 5 segundos; normalmente el macho arquea el cuerpo, la cola y la cabeza apuntando hacia la hembra. Con el tiempo el macho aumenta la frecuencia de flexionado y se acerca a la hembra. Durante esta exhibición las aletas dorsal y anal del macho se extienden totalmente y cuando él se acerca a la hembra, sus aletas medias se curvan hacia ella. También es posible que el macho después de orientarse hacia la hembra realice un círculo alrededor de ella, después de lo cual, él puede reanudar la persecución.

3. Movimientos de contacto. Normalmente cuando el macho está al lado de la hembra, él intenta hacer contacto con su espermatopodio cuyo éxito depende de la postura de la hembra que puede permanecer casi inmóvil o puede nadar evitando el contacto. Sin embargo, el contacto real es difícil de descubrir.

Allotoca Sp.

1. Orientación. Después de algunos minutos de presentación, el macho se puede orientar hacia la hembra y, dependiendo de su actividad, empezar a seguirla o exhibirse. Si la hembra está nadando lentamente lejos del macho, él la sigue; si la hembra está estacionaria a nivel medio del agua, el macho en el fondo empieza a exhibirse.

2. Exhibición. Si la hembra permanece estacionaria, el macho estando en el fondo empieza a realizar cualquiera de dos exhibiciones: moviendo; normalmente despacio, hacia adelante y hacia atrás; o agitándose en el plano horizontal. En ambos casos, el macho parece tocar la arena con su barriga. Al agitarse, el macho dobla ligeramente la parte posterior del cuerpo. La exhibición termina muy a menudo porque la hembra cambia de actividad. También es posible que el macho, después de seguirla, realice un movimiento frontal lentamente en figura de ocho. También he visto esta exhibición en las pozas del Instituto donde se mantiene una

población de esta especie.

3. Movimientos de contacto. Bajo estas condiciones de observación no descubrí ningún contacto real entre el macho y la hembra.

Skiffia lermae

1. Orientación. Ésta es una de las pocas especies de goodeidos en la que el cortejo ocurre frecuentemente; también es más elaborado que en las otras especies, excepto por sus especies congenéricas y Girardinichthys multiradiatus. Muy a menudo el macho se localiza a unos centímetros de la hembra.

2. Exhibición. Incluye varios movimientos. El primero consiste en interceptar a la hembra y realizar una serie de movimientos lentos de zig-zag en el plano horizontal delante de ella a aproximadamente 10cm, orientado en la misma dirección que ella. Durante este movimiento, el macho puede agitarse ligeramente. En la segunda exhibición el macho realiza una serie de círculos, en el plano horizontal, delante de la hembra que por un segundo permanece inmóvil. La tercera exhibición consiste en que el macho realiza un movimiento en forma de espiral junto a la hembra. En la cuarta exhibición el macho realiza una serie de figuras de 'figuras de ocho', en el plano horizontal delante de la hembra. La exhibición empieza por moverse hacia la hembra, volviéndose hacia fuera, yendo de nuevo hacia la hembra, y volviéndose hacia fuera, y así sucesivamente. La extensión de las trayectorias es corta, cerca de la hembra.

3. Movimientos de contacto. De vez en cuando el macho se acerca a la hembra desde cualquier lado, y mientras vibra, él intenta tocarla.

Characodon lateralis

1. Orientación. Es frecuente que macho y hembra estén en el fondo, y que el macho se oriente hacia la hembra y se le acerque.

2. Exhibiciones. Cuando la hembra es alcanzada por el macho desde cualquier lado, empieza a agitarse de lado a lado a una frecuencia lenta y una amplitud alta. Mientras la hembra se agita no se mueve a otra

parte del acuario. El macho se mueve lateralmente intentando tocar a la hembra con la aleta anal; debido a la inercia del movimiento, la cola tiende a moverse hacia fuera. El macho puede realizar la misma exhibición, pero acercándose a la hembra hacia atrás.

3. Movimientos de contacto. Cada vez que el macho ejecuta la exhibición lateral intenta hacer contacto con el área genital de la hembra. Muy a menudo la hembra reacciona marchándose.

Skiffia multipunctata

1. Orientación. Los machos de esta especie cortejan frecuentemente, y siempre están siguiendo a las hembras que normalmente nadan a profundidad media donde las exhibiciones del cortejo ocurren.

2. Exhibiciones. El macho intercepta a la hembra y empieza a agitarse a una frecuencia alta y amplitud baja; se estaciona a aproximadamente 2 cm de la hembra y a un ángulo de aproximadamente 45° respecto a la hembra, los dos están en la misma dirección. Cuando el macho parece aumentar la frecuencia de agitación lentamente se inclina se erecta a aproximadamente 90°, dependiendo del tiempo que la hembra permanece inmóvil, y con la aleta caudal apuntando hacia afuera. Eventualmente la hembra se retira y el macho la intercepta de nuevo y repite la misma exhibición, pero mostrando el otro flanco. Aunque las aletas medias del macho se abren y erectan, la estructura física particular de la aleta dorsal, los últimos 3 radios anteriores están separados, hace que su parte exterior se mueva aparentemente independiente y más despacio que el resto de la aleta. Además, el macho puede realizar una serie de movimientos en el sentido de las agujas del reloj frente a la hembra a aproximadamente 45°; a veces él se mueve al lado opuesto y repite la exhibición. También es posible que el macho interrumpa las exhibiciones laterales y se mueva varias veces alrededor de la hembra, en el sentido de las agujas del reloj.

3. Movimientos de contacto. De vez en cuando la hembra permanece inmóvil durante varios segundos permitiendo que el macho intente acercársele y hacer contacto con su poro genital. En el campo he observado que muchos

de estos intentos son interrumpidos por la presencia de un macho
intruso.

Zoogoneticus quitzeoensis

1. Orientación. Muy a menudo el macho sigue la hembra que nada lejos de
él en el fondo.

2. Exhibiciones. Cuando la hembra no se esta moviendo, el macho se le
acerca por el frente hasta estar a un lado, y en dirección opuesta.
Entonces el macho empieza a mover al revés mientras vibra a una
frecuencia moderada y amplitud baja. Unos segundos después él se da la
vuelta para quedar cerca de la hembra y en la misma dirección; él
intenta hacer contacto con la hembra que a menudo nada lejos. También es
posible que el macho estando orientando hacia la hembra y en la misma
dirección, se le acerque por un lado mientras vibra; él dobla ambos
extremos hacia la hembra. De vez en cuando el macho realiza movimientos
como los de un látigo frente a la hembra, pero en la misma dirección que
ella.

3. Movimientos de contacto. Cuando el macho logra acercarse a la hembra,
él vibra e intenta hacer contacto; ella se agita y se va a otro lugar.

Xenotoca eiseni

1. Orientación. Como otras especies de la familia, esta también muestra
frecuentemente conducta de cortejo. Los machos siempre están siguiendo
hembras con quienes intentan copular. El cortejo ocurre entre
profundidad media y el fondo.

2. exhibiciones. Por lo menos pueden describirse 7 exhibiciones
diferentes: a) El macho realiza una serie de círculos en forma de
espiral; empieza delante de la hembra (45°) en el plano horizontal
moviendo la cola de lado a lado ('aleteando'), luego pasa moviéndose en
el sentido de las agujas del reloj atrás de la hembra, quedando un poco
arriba y aleteando. Luego pasa al frente de la hembra aletea y regresa
detrás de ella, pero pasando sobre ella. Después de aletear persigue se
va hacia el frente moviéndose en sentido contrario al movimiento de las

agujas del reloj; repite el golpeteo de la cola y se mueve en el sentido de las agujas del reloj, sube pasando sobre la hembra, pero aleteando de nuevo antes de ir a la superficie. Mientras el macho está realizando estas exhibiciones, la hembra está vibrando a una frecuencia y amplitud moderadas; ella está ligeramente inclinada (15°). Esta es una conducta que ella muestra durante cada exhibición del macho. b) Mientras la hembra permanece estacionaria, el macho, delante de la hembra y orientado hacia ella (40°) empieza a golpear la cola, se mueve, en el sentido de las agujas del reloj trazando un semicírculo, al frente de la hembra. Luego empieza a realizar una serie de movimientos como 'X', aleteando cada vez que él se vuelve del lado opuesto. c) El macho gira alrededor de la hembra en el sentido de las agujas del reloj, y aletea detrás y delante de ella. d) El macho realiza un movimiento que se parece a un ocho y que otros goodeidos presentan; él cruza de izquierda a derecha justo delante de la hembra, se vuelve en el sentido de las agujas del reloj y se mueve hacia la hembra, y luego alejándose de ella. e) Otra exhibición es cuando el macho, delante de la hembra, en el plano horizontal, realiza una sucesión de movimientos en forma de ola; cada vez que muestra un flanco a la hembra, golpea la cola. f) Otra variante de los movimientos de X arriba mencionados es cuando el macho, delante de la hembra, en el plano horizontal, realiza movimientos de zig-zag cerca de ella; cada vez que el macho muestra un flanco agita la cola que casi golpea la hembra. g) Finalmente, el macho estando estacionario aproximadamente 7 cm delante (45 °)de la hembra, ligeramente sobre el plano horizontal, agita su cola. Cuando la hembra para de vibrar y nada lejos, el macho la sigue violentamente.

3. Movimientos de contacto. He visto sólo movimientos de contacto de machos que no están cortejando a la hembra. Muy a menudo interrumpen a un macho cortejando e intentan acercarse por un lado a la hembra. A veces el intruso se las arregla para hacer contacto con la hembra, pero muy a menudo el macho que cortejaba lo detiene.

<u>Zoogoneticus</u> <u>quitzeoensis</u>

1. Orientación. La conducta de cortejo es rara a menos que el macho se motive después de estar aislado o que la hembra esté receptiva. Bajo cualquiera de estas circunstancias, el macho se orienta hacia la hembra y la sigue.

2. Exhibiciones. Cuando la hembra está inmóvil, el macho se le acerca por el frente. Él empieza a agitarse a una frecuencia alta y amplitud baja en el plano horizontal, mostrando un flanco a la hembra. Cuando el macho parece aumentar la frecuencia de vibración, empieza a rotar en sentido contrario al movimiento de las agujas del reloj hasta quedar acostado en su flanco. Eventualmente la hembra nada lejos y el macho regresa a su postura anterior.

3. Movimientos de contacto. A veces el macho se acerca a la hembra por atrás e intenta hacer contacto con ella por un lado; podría ser que cuando el macho está intentando hacer contacto con la hembra, se agite a una frecuencia alta y una amplitud muy baja, pero esta observación debe confirmarse.

APÉNDICE C

DENDROGRAMAS DE TRANSICIONES EVOLUTIVAS INDEPENDIENTES

103

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia bilineata
Skiffia lermae
Skiffia francesae
Skiffia multipunctata
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Chapalichthys pardalis
Xenotoca eiseni
Xenotoca melanosoma
Goodea atripinnis
Goodea gracilis
Goodea luitpoldi
Ataeniobius toweri
Characodon audax
Characodon lateralis
Allotoca maculata
Allotoca dugesi
Zoogoneticus tequila
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyiodon whitei
Ilyiodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae

Figura C-1. Árbol filogenético de la familia Goodeidae (Grudzien et al. 1992). Allotoca maculata, Chapalichthys pardalis, Characodon audax, Goodea luitpoldi, Goodea gracilis, Ilyodon whitei and Zoogoneticus tequila no estaban incluídas en el árbol original.
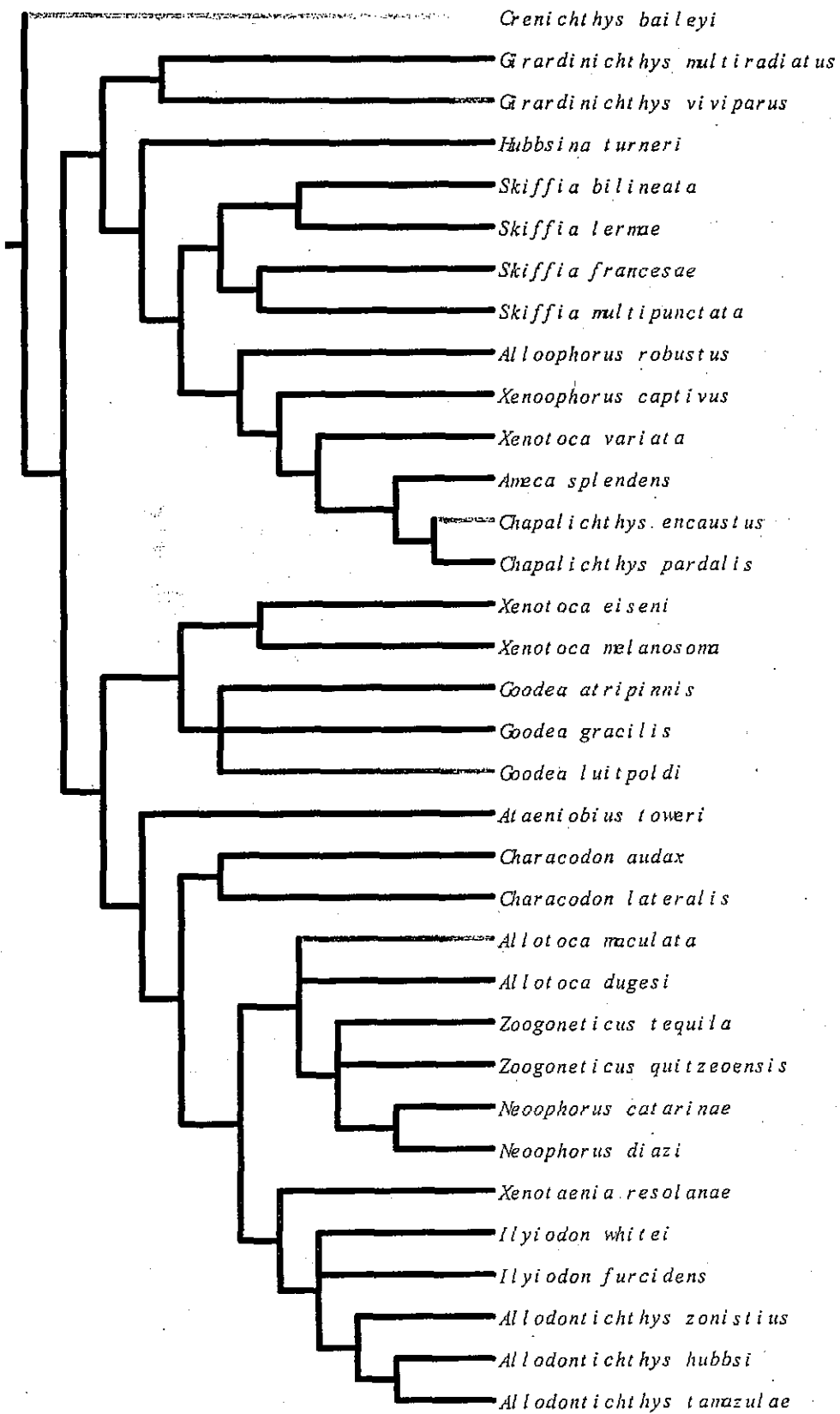
Figura C-2. Tipo de hábitat en los goodeidos. Transiciónes en el árbol
(líneas sólidas: aguas corrientes; líneas grises: lagos; líneas pálidas:
ambigüedad) fueron obtenidas usando Winclada.
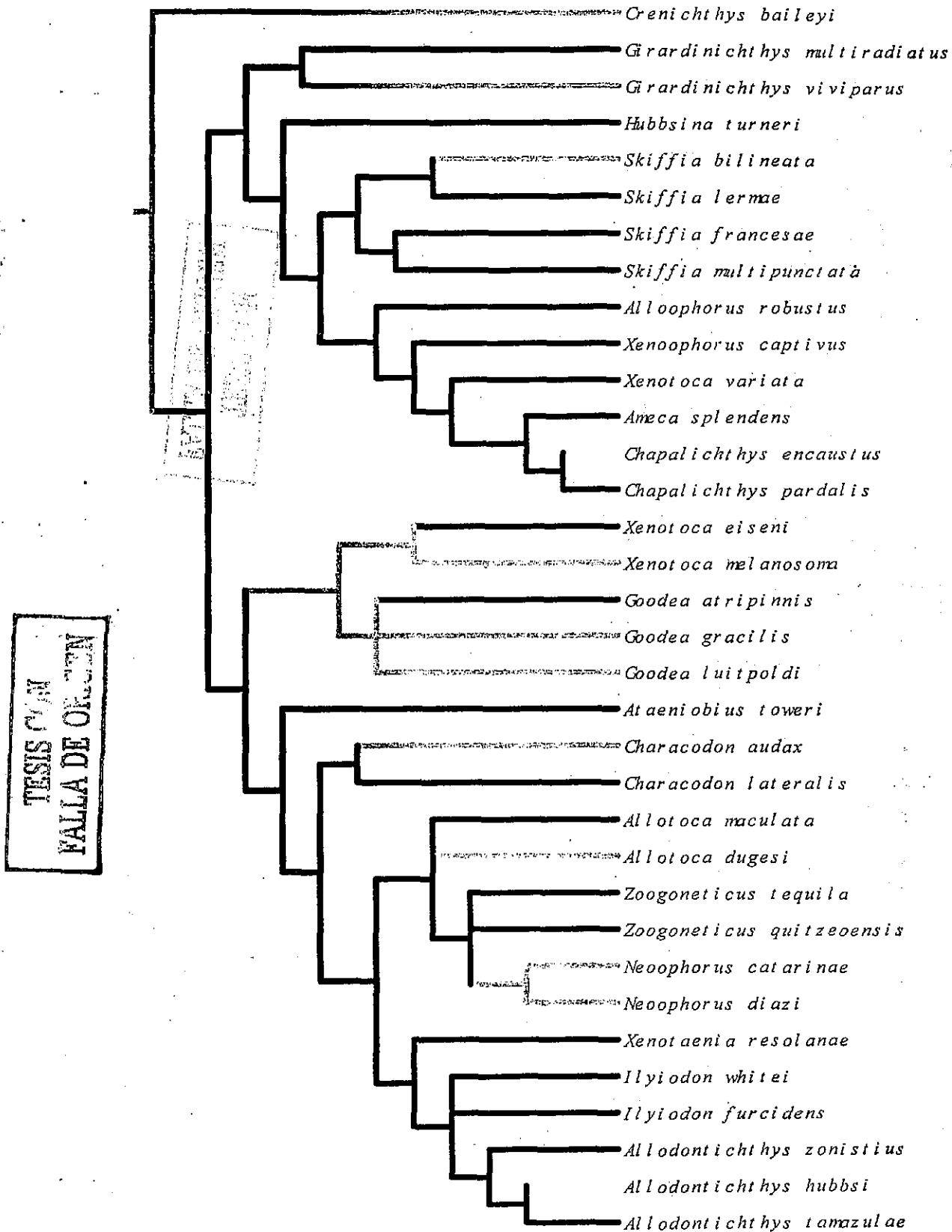
Figura C-3. Colores derivados de caroteno en los machos. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.
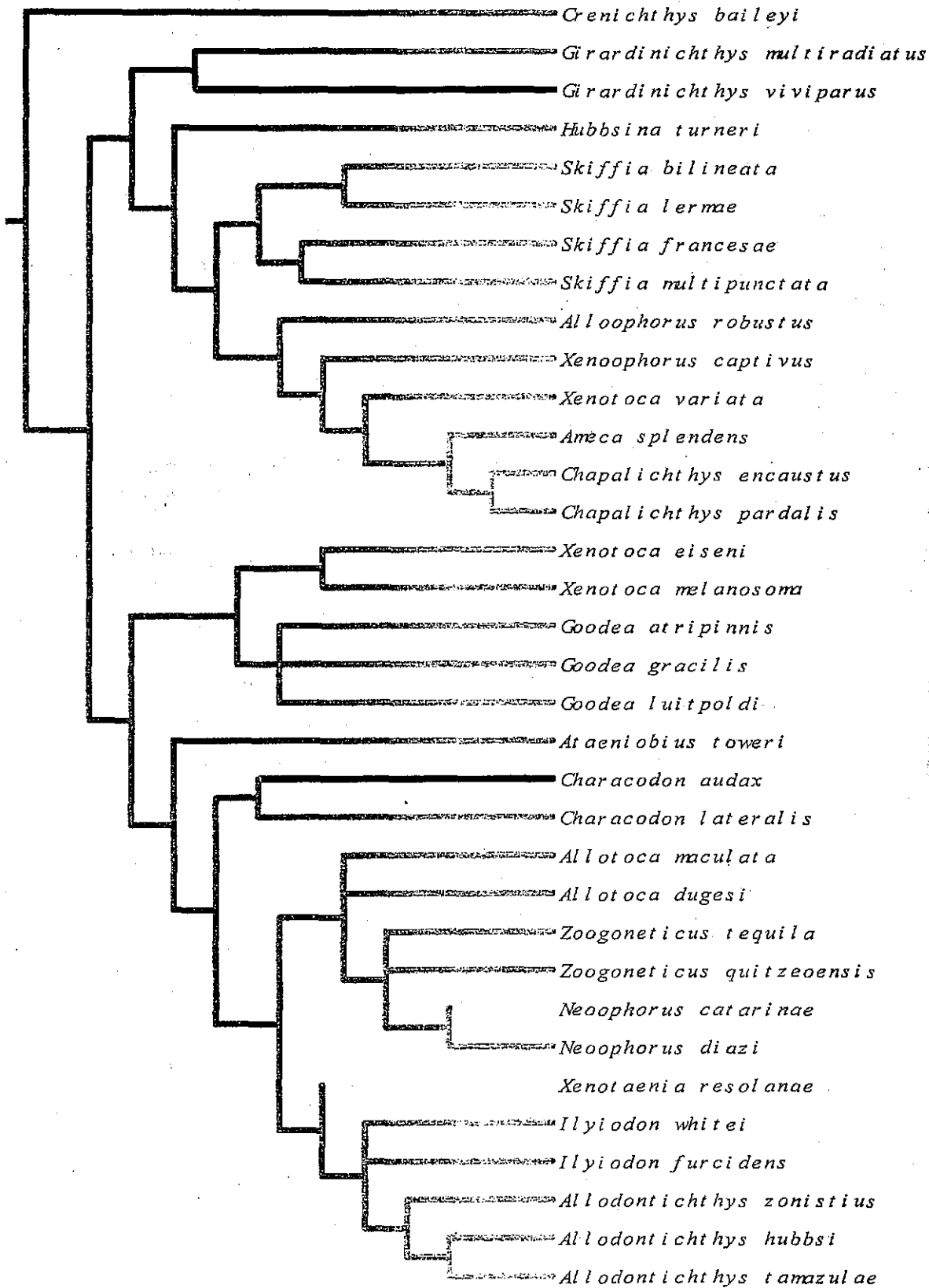
Figura C-4. Aletas negras en los machos. Las transiciones en el árbol
(líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas:
ambigüedad) fueron obtenidas usando Winclada.
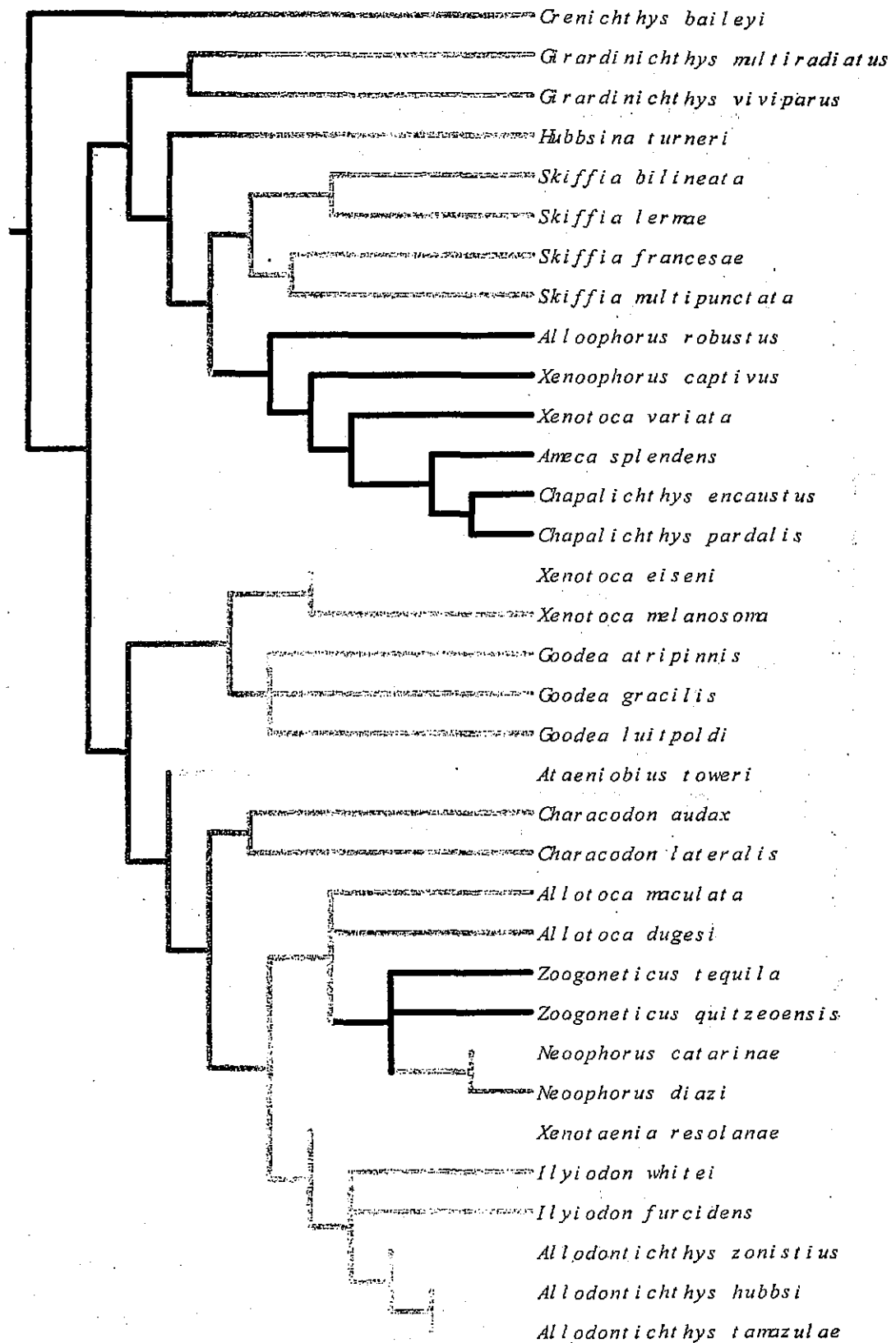
Figura C-5. Espejos en los flancos de los machos. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.

108
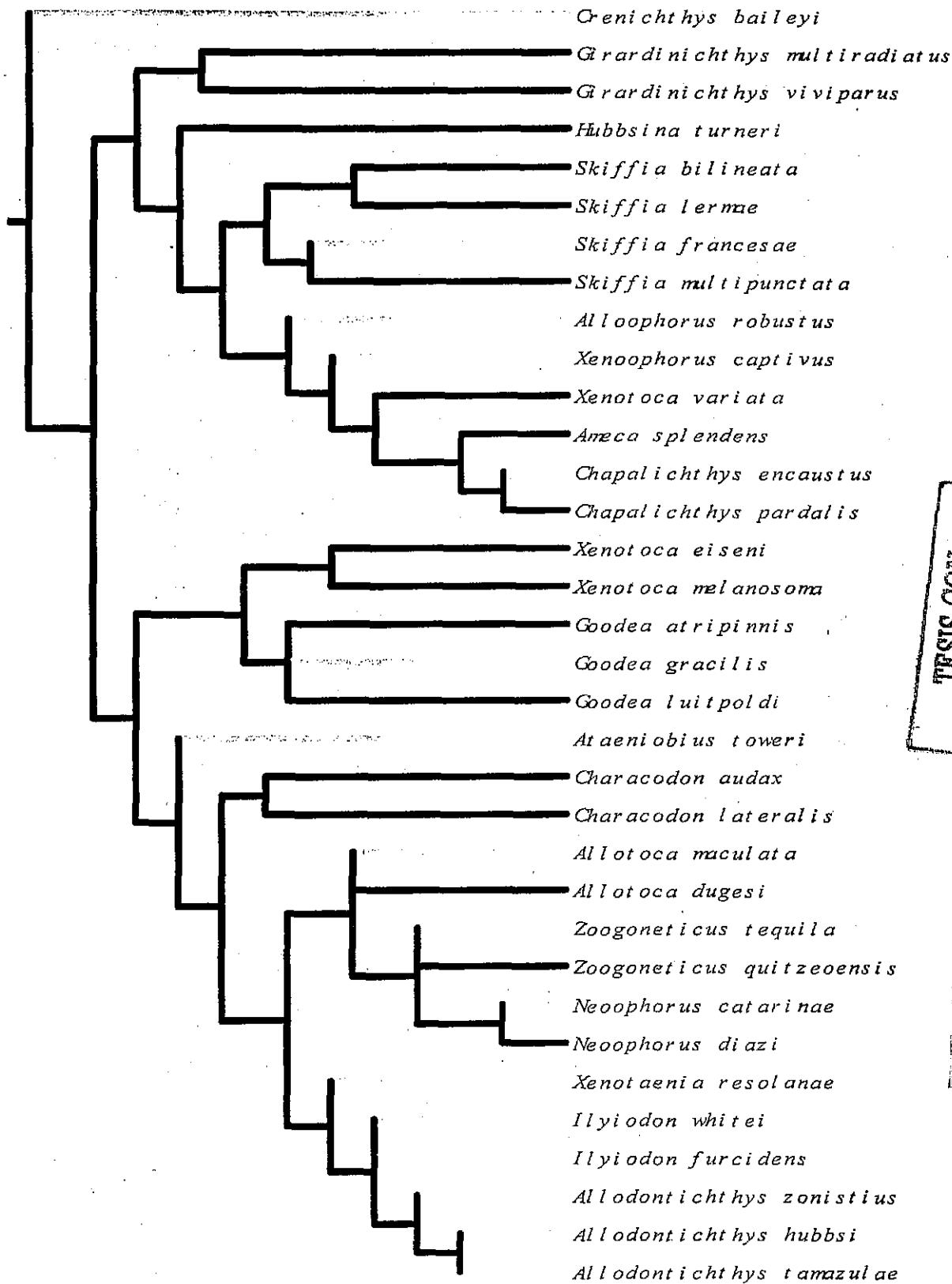
Figura C-6. Dimorfismo del tamaño de la aleta dorsal. Las transiciones en el árbol (líneas sólidas: presencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia bilineata
Skiffia lermae
Skiffia francesae
Skiffia multipunctata
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Chapalichthys pardalis
Xenotoca eiseni
Xenotoca melanosoma
Goodea atripinnis
Goodea gracilis
Goodea luitpoldi
Ataeniobius toweri
Characodon audax
Characodon lateralis
Allotoca maculata
Allotoca dugesi
Zoogoneticus tequila
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyiodon whitei
Ilyiodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae

Figura C-7. Contornos oscuros de las aletas medias en los machos. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia bilineata
Skiffia lermae
Skiffia francesae
Skiffia multipunctata
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Chapalichthys pardalis
Xenotoca eiseni
Xenotoca melanosoma
Goodea atripinnis
Goodea gracilis
Goodea luitpoldi
Ataeniobius toweri
Characodon audax
Characodon lateralis
Allotoca maculata
Allotoca dugesi
Zoogoneticus tequila
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyiodon whitei
Ilyiodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae

Figura C-8. Barras verticales brillantes y cafés en las hembras. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia bilineata
Skiffia lermae
Skiffia francesae
Skiffia multipunctata
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Chapalichthys pardalis
Xenotoca eiseni
Xenotoca melanosoma
Goodea atripinnis
Goodea gracilis
Goodea luitpoldi
Ataeniobius toweri
Characodon audax
Characodon lateralis
Allotoca maculata
Allotoca dugesi
Zoogoneticus tequila
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyiodon whitei
Ilyiodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae
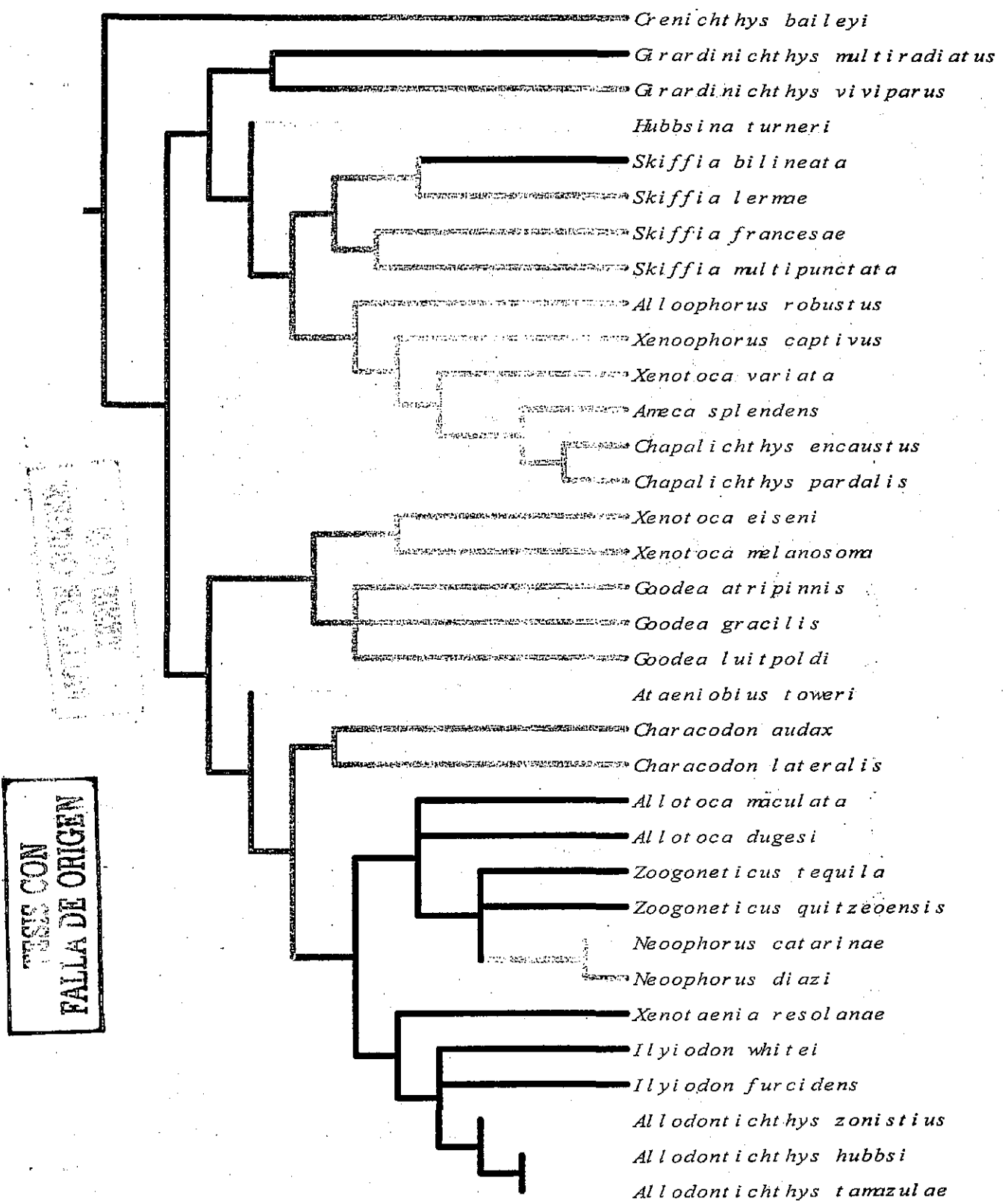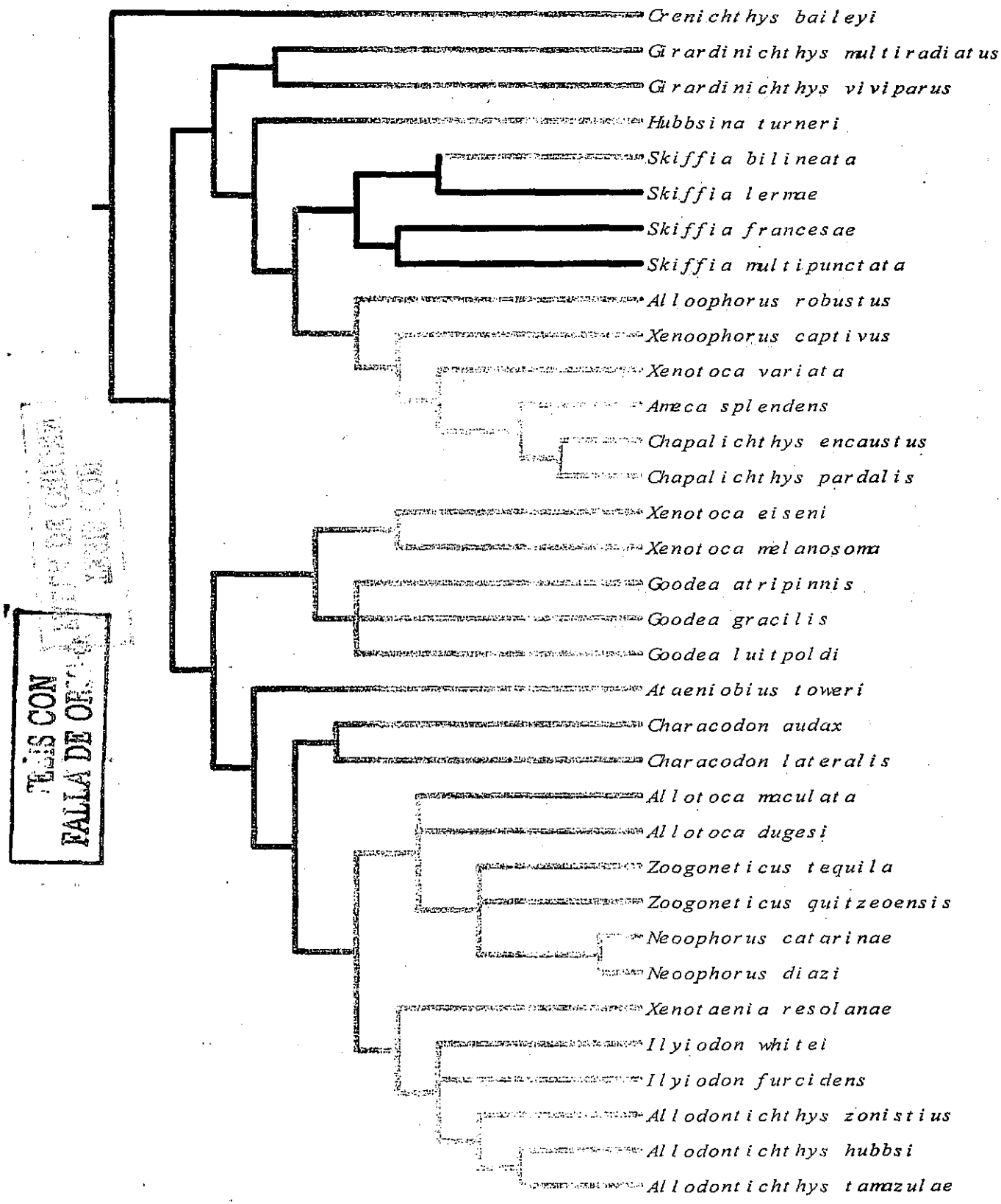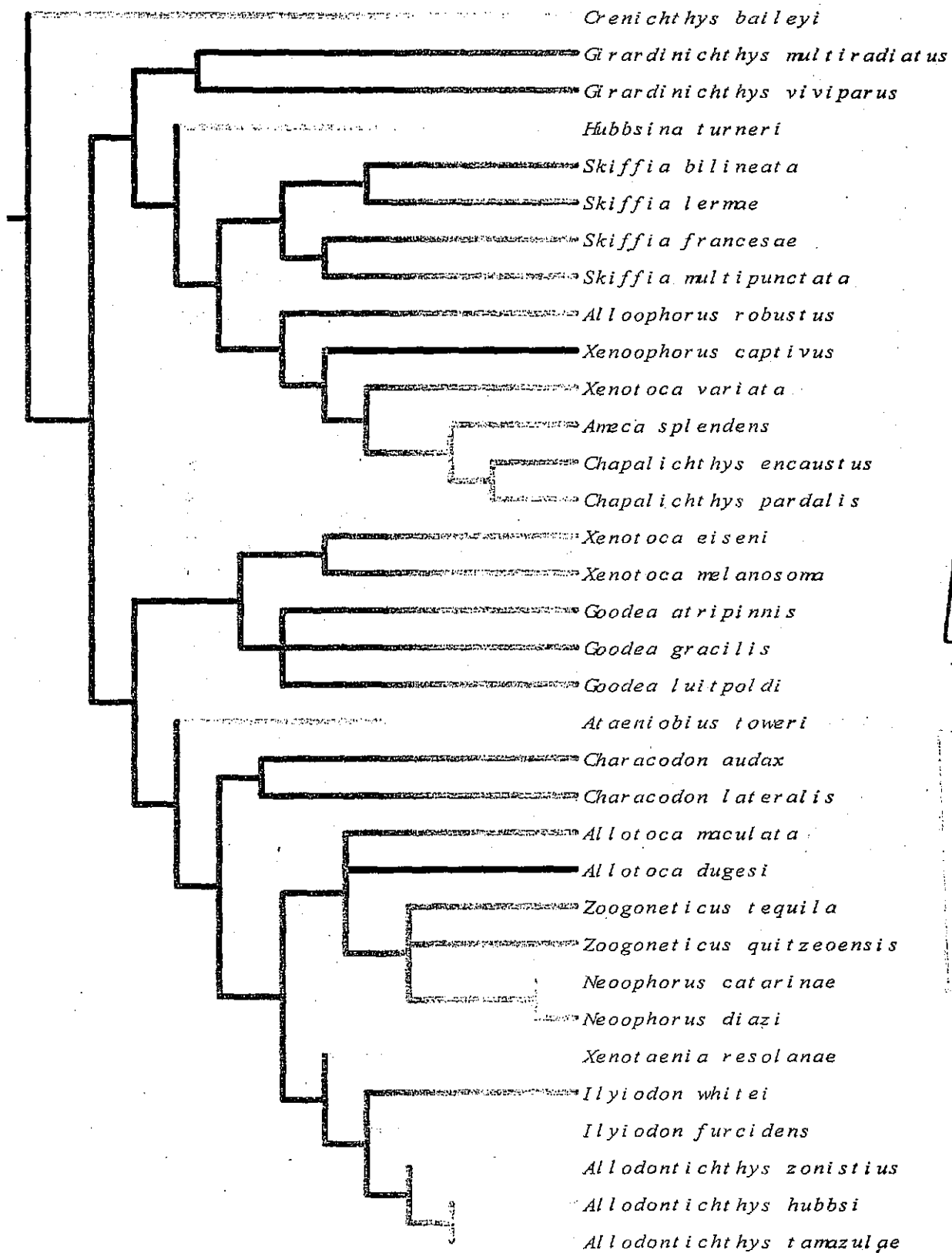
Figura C-9. Aletas hendidas en los machos. Las transiciones en el árbol
(líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas:
ambigüedad) fueron obtenidas usando Winclada.

Crenichthys baileyi

Girardinichthys multiradiatus

Girardinichthys viviparus

Hubbsina turneri

Skiffia bilineata

Skiffia lermae

Skiffia francesae

Skiffia multipunctata

Alloophorus robustus

Xenoophorus captivus

Xenotoca variata

Ameca splendens

Chapalichthys encaustus

Chapalichthys pardalis

Xenotoca eiseni

Xenotoca melanosoma

Goodea atripinnis

Goodea gracilis

Goodea luitpoldi

Ataeniobius toweri

Characodon audax

Characodon lateralis

Allotoca maculata

Allotoca dugesi

Zoogoneticus tequila

Zoogoneticus quitzeoensis

Neoophorus catarinae

Neoophorus diazi

Xenotaenia resolanae

Ilyiodon whitei

Ilyiodon furcidens

Allodontichthys zonistius

Allodontichthys hubbsi

Allodontichthys tamazulae

Figura C-10. Sobarse el abdomen en el sustrato, conducta de los machos. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.
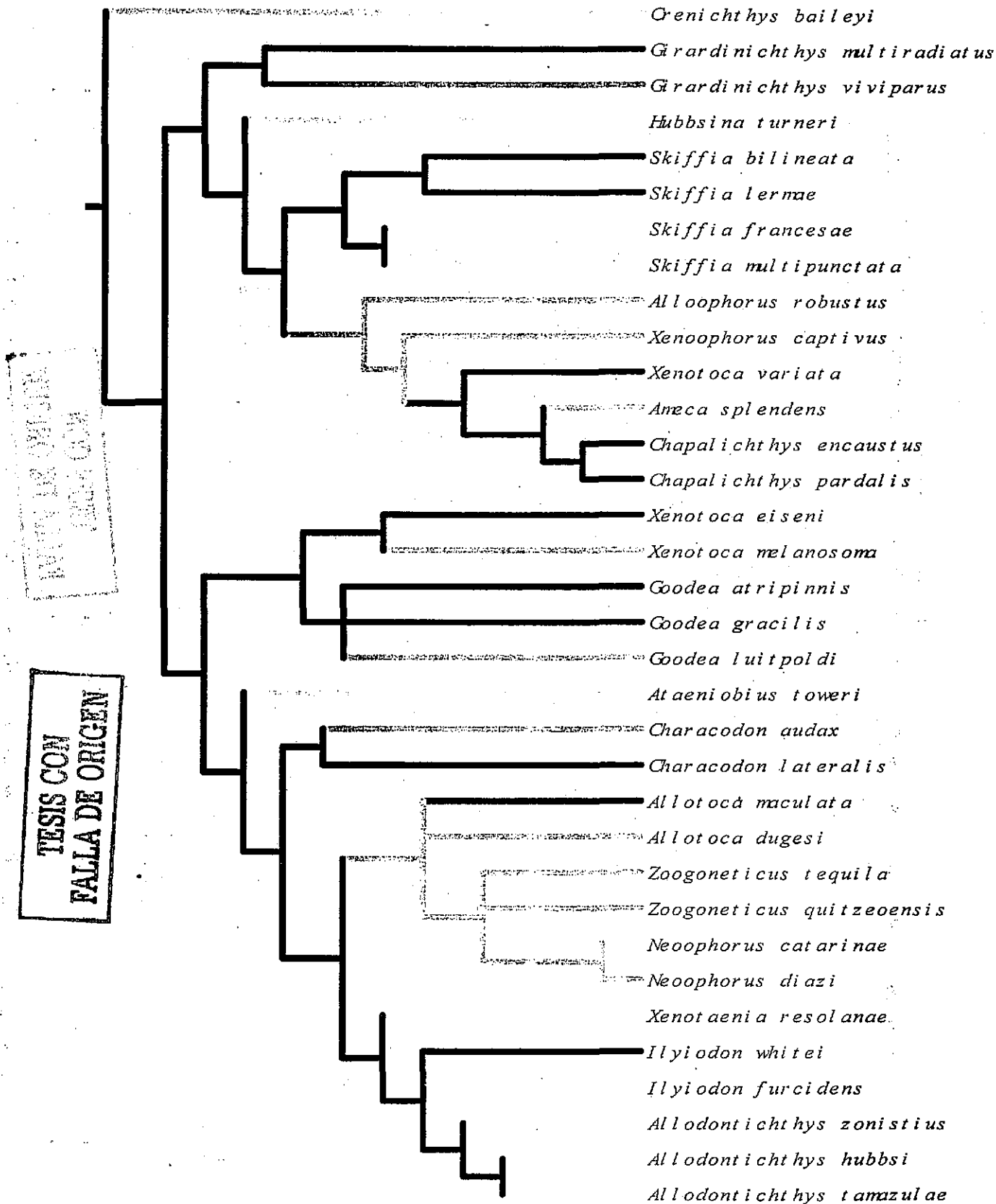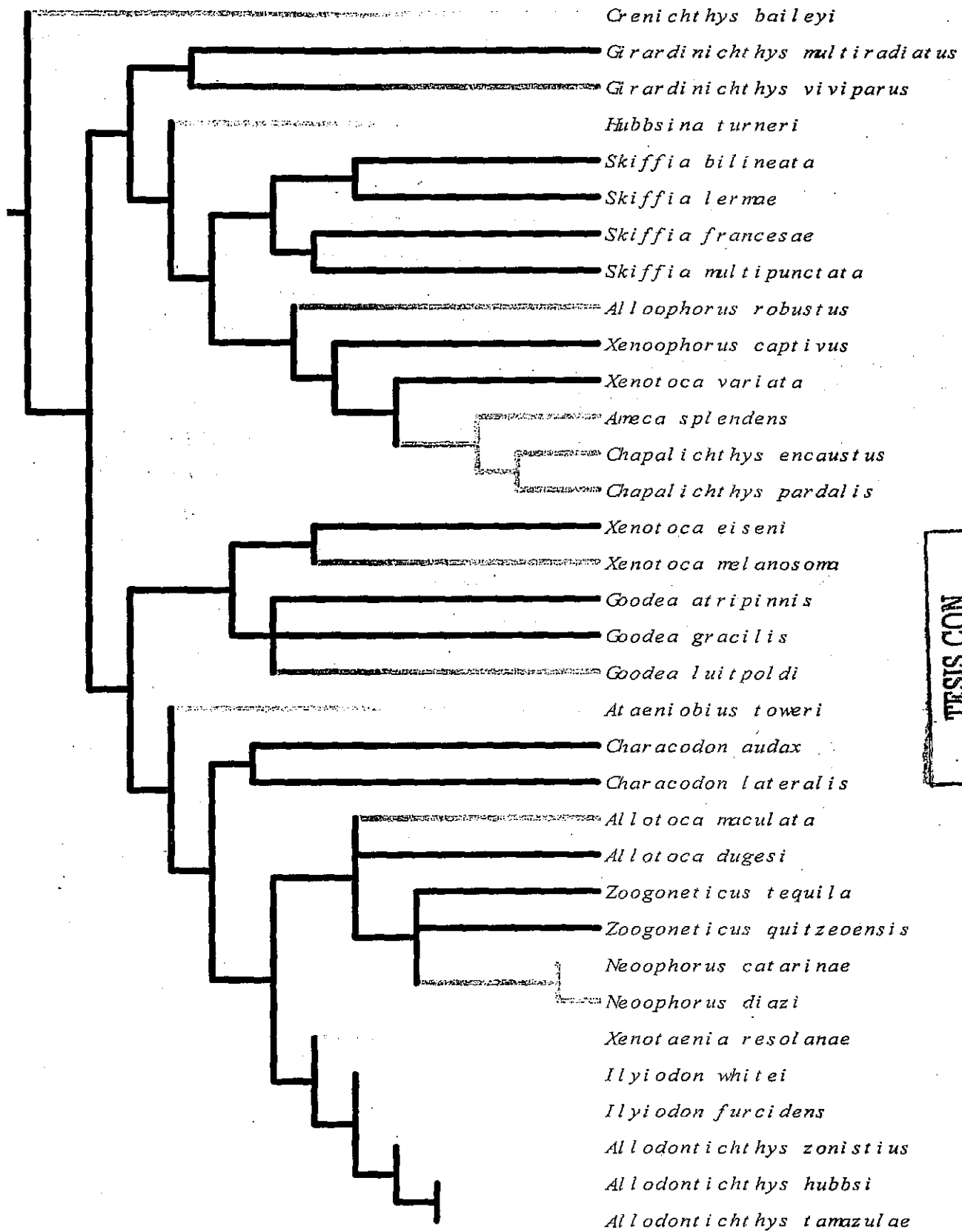
Figura C-11. Conducta de vibración de las hembras. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.

Crenichthys baileyi
Girardinichthys multiradiatus
Girardinichthys viviparus
Hubbsina turneri
Skiffia bilineata
Skiffia lermae
Skiffia francesae
Skiffia multipunctata
Alloophorus robustus
Xenoophorus captivus
Xenotoca variata
Ameca splendens
Chapalichthys encaustus
Chapalichthys pardalis
Xenotoca eiseni
Xenotoca melanosoma
Goodea atripinnis
Goodea gracilis
Goodea luitpoldi
Ataeniobius toweri
Characodon audax
Characodon lateralis
Allotoca maculata
Allotoca dugesi
Zoogoneticus tequila
Zoogoneticus quitzeoensis
Neoophorus catarinae
Neoophorus diazi
Xenotaenia resolanae
Ilyiodon whitei
Ilyiodon furcidens
Allodontichthys zonistius
Allodontichthys hubbsi
Allodontichthys tamazulae

Figura C-12. Conducta de bandereo de los machos. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.
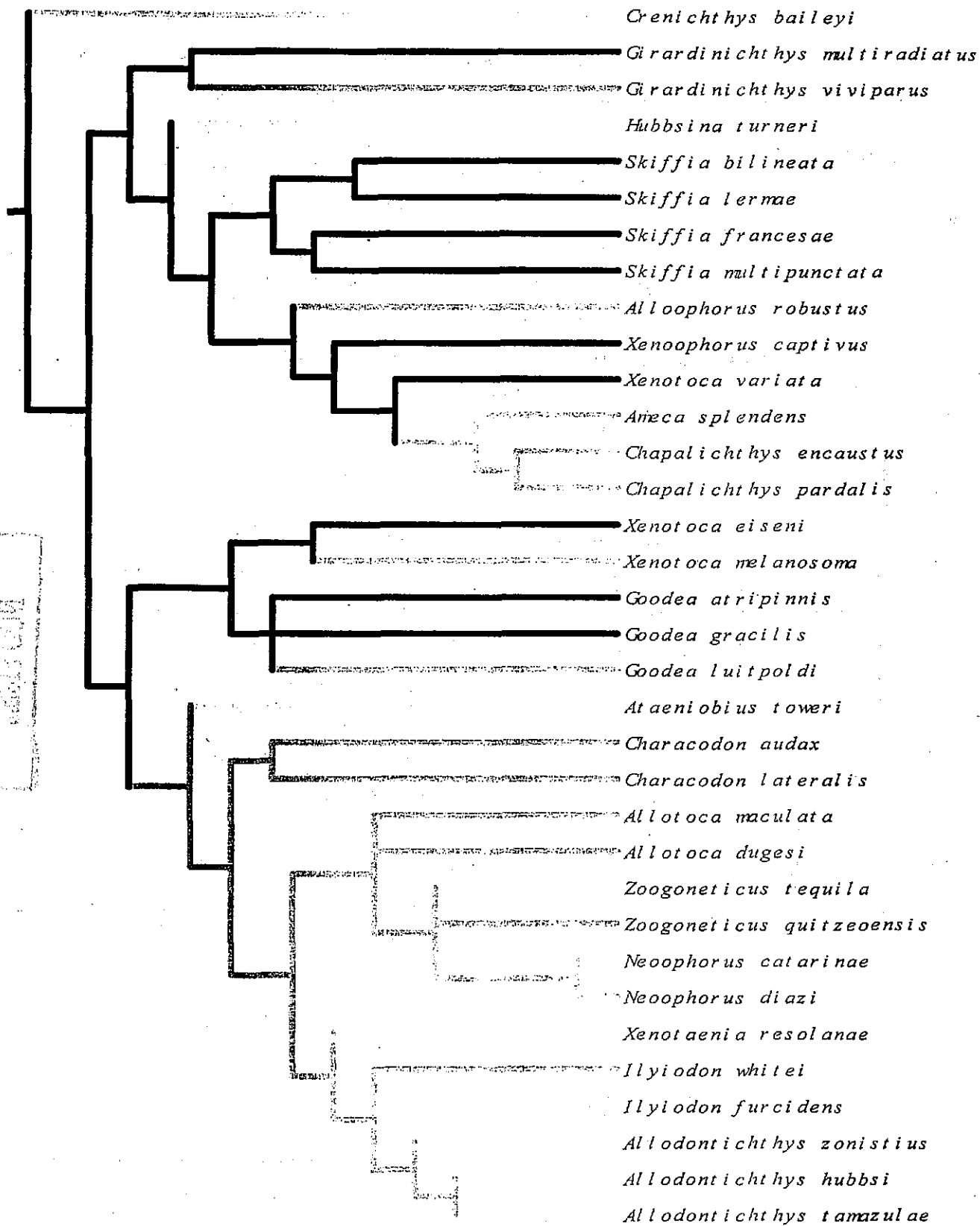
Figura C-13. Figura de ocho o de x, conducta de los machos. Las transiciones en el árbol (líneas sólidas: presencia; líneas grises: ausencia; líneas pálidas: ambigüedad) fueron obtenidas usando Winclada.

BASE DE DATOS DE LA TURBIEDAD DEL AGUA

Tabla D-1. Datos de turbiedad del agua usando agua clara como estándar de comparación

Fecha	Localidad	Turbiedad (TJU)
22 de marzo 1998	Cuitzeo	150
13 de diciembre 1998[1]	Cuitzeo	60
24 de abril 1999	Cuitzeo	300
04 de abril 1998	Rancho Viejo	120
01 de mayo 1999	Rancho Viejo	60
07 de octubre 1999[2]	Rancho Viejo	12.5
05 de febrero 1998	San Francisco	5
03 de junio 1999	San Francisco	35
21 de noviembre 1997	Yuriria	23.33
17 de octubre 1998	Zacapu	22.5
07 de mayo 1999	Zacapu	10

[1]Las muestras fueron tomadas de una poza adyacente al lago.
[2]Río arriba de la poza donde fueron capturados los peces.

Las siguientes mediciones de profundidad fueron tomadas usando un disco de seki (20 cm, diámetro).

Tabla D-2. Datos de la profundidad a la cual los organismos fotosintéticos están limitados

Fecha	Localidad	Cm
07 de octubre 1999	Cuitzeo	8.5
05 de octubre 1999	San Francisco	86
06 de octubre 1999	Zacapu	94.5