UNIVERSIDAD DE GRANADA

DEPARTAMENTO DE BIOLOGÍA ANIMAL

LAS COMUNIDADES DE AVES Y REPTILES DEL CORREDOR VERDE DEL GUADIAMAR DESPUÉS DEL VERTIDO MINERO DE AZNALCÓLLAR

BIRDS AND REPTILES COMMUNITIES IN THE GUADIAMAR GREEN CORRIDOR AFTER THE AZNALCÓLLAR MINING SPILL

Tesis Doctoral

Rocío Márquez Ferrando

Granada 2008

Editor: Editorial de la Universidad de Granada
Autor: Rocío Marquéz Ferrando
D.L.: GR.1617-2008
ISBN: 978-84-691-4863-1

LAS COMUNIDADES DE AVES Y REPTILES DEL CORREDOR VERDE DEL GUADIAMAR TRAS DEL VERTIDO MINERO DE AZNALCÓLLAR

BIRDS AND REPTILES COMMUNITIES IN THE GUADIAMAR GREEN CORRIDOR AFTER THE AZNALCÓLLAR MINING SPILL

Memoria que lal Licenciada Rocío Marquez Ferrando presenta para aspirar al Grado de Doctor por la Universidad de Granada

Esta memoria ha sido realizada bajo la dirección de:

Dr. Juan M. Pleguezuelos Gómez
Dr. Diego Ontiveros Casado
Dr. Xavier Santos Santiró

Lda. Rocío Márquez Ferrando
Aspirante al Grado de Doctor

Granada, a 16 de mayo de 2008

Juan M. Pleguezuelos, Profesor Titular de Zoología de la Universidad de Granada, **Diego Ontiveros Casado** Profesor contratado de la Universidad de Granada y **Xavier Santos Santiró** Investigador de la Diputación Provincial de Barcelona. Como directores del presente trabajo de investigación.

INFORMAN:

Que los trabajos de investigación desarrollados en la Memoria de Tesis Doctoral: "Las comunidades de aves y reptiles del Corredor Verde del Guadiamar después del vertido minero de Aznalcóllar – Birds and reptiles communities in the Guadiamar Green Corridor after the Aznalcóllar mining spill"**, son aptos para ser presentados por el Lda. Rocío Márquez Ferrando ante el Tribunal que en su día se designe, para aspirar al Grado de Doctor por la Universidad de Granada.**

Y para que así conste, en cumplimiento de las disposiciones vigentes, firmamos el presente documento, en Granada a 16 de mayo de 2008.

JM. Pleguezuelos **D. Ontiveros** **X. Santos**

Durante el tiempo de realización de esta Tesis Doctoral la Lda. Rocío Márquez Ferrando ha disfrutado de una Beca de los Proyectos de Investigación y Seguimiento del Corredor Verde del Guadiamar (SECOVER).

La investigación presentada en esta Tesis Doctoral se ha realizado en el Departamento de Biología Animal de la Universidad de Granada.

Ella está en el horizonte.
Me acerco dos pasos.
Ella se aleja dos pasos
y el horizonte se corre
diez pasos más allá.
Por mucho que yo camine
Nunca la alcanzaré.
¿Para que sirve la utopía?
Para eso sirve:
para caminar

Eduardo Galeano "La Utopía"

Agradecimientos

En el mundo hay muchas cosas bonitas y no voy a empezar a enumerarlas, más que nada porque no acabaría nunca, pero sólo mencionaré una de ellas, que la he descubierto durante estos años.

Nunca jamás pensé hacer la tesis doctoral, lo veía algo que me superaba, pero la vida, me brindó la oportunidad de hacerla. Me tenía que presentar a una entrevista para una beca de investigación en vertebrados; pero si digo la verdad, la tesis no creía poder hacerla, y cuando Plegue me dijo que la beca era para mi, me dije a mí misma, adelante. Y ¿por qué no? formación tenía, ilusión también, miedos muchos, pero para poder superarnos hay que echarle valor a las cosas, y yo quería ver hasta donde era capaz de llegar. Y empecé poco a poco a aprender a investigar. Del campo tengo muy buenos recuerdos con todos aquellos que me han acompañado por el Corredor Verde del Guadiamar y lo mismo de la universidad de Granada, porque ha sido mi lugar de trabajo durante estos años y he conocido a mucha gente que a partir de ahora marcarán mi vida de una manera muy especial.

De ahí que halla elegido la frase de Eduardo Galeano, porque para llegar a algo tenemos que tener ganas y caminar, porque somos capaces de hacer muchas cosas y buenas, y cosas que nunca jamás te imagines, por eso hay que estar atento para que cuando la vida te abra una puerta, puedas asomarte y mirar, curiosear un poco y si te gusta, adelante, pero no digas que no eres capaz sin intentarlo antes. Y ahí empieza mi aventura y superación personal y aquí finaliza, sacando a publicar el trabajo de varios años para que quede huella de ello. Y a esto me refería cuando decía al principio, que para mi, una cosa muy bonita en esta vida es tener la oportunidad de poder demostrar lo que podemos hacer, y hacerlo, lleguemos donde lleguemos, pero superarnos es ya importante y bonito.

Se me hace esto un mundo. Ahora mismo se me está abriendo el corazón para poder expresar con todas mis ganas las GRACIAS y deciros a todos una cosa que he escuchado mil veces en boca de otros, pero que nunca sentí tanto como ahora: “sin duda esto no habría salido sin vuestra ayuda, de verdad, han sido casi cuatro años de subidas y bajadas y cada uno a su manera me ha aportado la energía suficiente para poder remontarme al final de la tesis, que ha sido, lo que más me ha costado.

… **Gracias** en primer lugar a mi familia por su apoyo constante desde que tomé la decisión, por haber confiado en mi y porque una vez más me han demostrado que son gente maravillosa. Os quiero mucho a todos.

…**Gracias** a mis directores de tesis Plegue, Diego y Xavier, tres amigos, a los que le debo mucho sobre lo que sé de reptiles, aves y de la vida, por sus consejos y por haber estado ahí siempre que los he necesitado.

… **Gracias** a Maribel, Mónica, Jesús, Marisa, Esmeralda, Juanra, Ricardo, Pizarro, Estrella y Kirk, mis compañeros del grupo de investigación de vertebrados mediterráneos que son estupendos; su compañía, sus ánimos, abrazos y ayuda ha sido muy gratificante. Sois como una gran familia para mí. Me ha encantado conoceros y trabajar con todos vosotros.

… **Gracias** a Adela, Rosa, Roca, José Antonio Hódar, Rafael Morales y Manuel Tierno Figueroa que me han me han resuelto dudas durante mi aprendizaje sobre cómo analizar los datos y muy especialmente a Adela, por ser como una hermana, compañera y amiga a la vez, por haberme regalado tantos buenos momentos, gracias chula!!!!! Te quiero mucho!!!.

… **Gracias** a la trouppe de becarios, mi amigos, mis colegas, que son la caña, qué risas y qué buenos momentos hemos pasado, y espero seguir pasando. Espero que el tiempo no nos distancie nunca!!! os quiero un montón, Muaka!!! Gracias por vuestro buen humor, apoyo constante, atención y cariño a Paqui, Jose, Kike, Pepelu, Asier, Mati, Irene, Carolina, Virginia, Valentina, Lola y Merche. Y Jose, muchas gracias por haberme echado un cable con la maquetación de la tesis, gracias gracias, gracias!!! Y Paqui, te quería agradecer especialmente el apoyo que siempre me has dado, las veces que me has llamado en los momentos más duros; eres una gran amiga y una de las cosas más buenas que me llevo de esta travesía, porque sé que siempre vas a estar ahí. Gracias también a mi amigo"J" por ese peasso de dibujo que aparece en la portada, me ha encantado que lo hiceras. Gracias también a José Antonio, ex-secretario del departamento de Biología Animal por su amababilidad cuando me hacía falta cualquier cosa. Gracias a los componentes del grupo de investigación de reptiles y anfibios de Gustavo Llorente de la Universidad de Barcelona durante mi estancia en abril del 2007, por su ayuda y atención.Gracias a Miguel Ángel Carretero por sus consejos y atención desde que lo conozco y sobre todo por su inestimable ayuda en Giao (Oporto) cuando estuve de estancia breve en Portugal y por enseñarme otras muchas cosas de la vida; me encantaron esas largas charlas de *Podarcis* spp., de historia y de la vida en general. Te aprecio mucho!!!. Gracias a Nuno Ferrand da Almeida por acogerme en el Centro de Investigación de Biodiversidad y Recursos Genéticos (CIBIO); a Xana por sus consejos para los análisis de datos de las asimetrías y por supuesto también a las niñas Caterina, Raquel y Ana por hacerme la estancia tan divertida.Y como no… **Gracias** a mis compis de piso, Luisi y Angelmari, que han sido los mejores compañeros de piso que he tenido hasta ahora, y porque han hecho que la tesis sea un poco más llevadera. Siempre que volvía a casa he encontrado buen ambiente, buena música y buena comidita Luisita!!!. Muchas gracias guapos, sois muy importantes para mí.

A todos, muchas gracias de corazón

A mis padres Antonio y
Antonia, a Noli, Mari , Ramón
y a las tres florecillas,
Carla, Paloma y Lucía.

INDICE

Resumen

El vertido de la balsa de lodos tóxicos de la mina Aznalcóllar, ocurrido en abril de 1998, fue uno de los desastres ecológicos más importantes ocurridos en España. Afectó gran parte de los ríos Agrio y Guadiamar (suroeste de la Peninsula Ibérica), alterando la estructura y el funcionamiento del ecosistema fluvial y sus márgenes.

Tras el accidente minero se puso en marcha un proyecto de limpieza y recuperación de la zona afectada; posteriormente se nombró Espacio Natural Protegido, con el nombre de *Corredor Verde del Guadiamar*; también comenzó el Programa de Investigación del Corredor Verde del Guadiamar, PICOVER, en el que intervinieron diversos grupos científicos de distintas disciplinas, para estudiar la respuesta y evolución de las distintas comunidades de organismos a este impacto ambiental.

En la presente tesis doctoral, pretendemos evaluar la evolución de las comunidades de aves y reptiles tras la restauración de la vegetación durante el periodo 2000-2006 y de este modo, valorar las medidas de restauración aplicadas. La memoria se divide en dos apartados según la comunidad de animales estudiados: los primeros dos capítulos se dedican a la comunidad de aves y los dos últimos a la de reptiles.

Para analizar la evolución espacial y temporal de la comunidad de aves nidificantes en el Corredor Verde del Guadiamar, se establecieron siete transectos a lo largo del corredor. Los censos se realizaron durante la primavera para calcular valores de riqueza en especies, abundancia y diversidad. En el último año se midieron las variables descriptoras del microhábitat y del macrohábitat de las aves, para determinar cuales son las que nos explican la mayor variación de la comunidad de aves a lo largo del corredor.

En cuanto a la comunidad de reptiles, se llevaron a cabo muestreos de a lo largo de todo el corredor; ante la escasez de individuos y refugios naturales, se plantea como hipótesis que la falta de refugios es el factor causante de la escasez de reptiles. Para testar nuestra hipótesis, realizamos un experimento en el que aportamos refugios artificiales.

Para valorar el estado de contaminación en la zona afectada, cuantificamos el nivel de metales pesados en los individuos de *P. algirus* de poblaciones con distinto grado de exposición a la contaminación.

La contaminación puede estar afectando al desarrollo de los organismos, concretamente su simetría corporal. Por lo que, medimos la asimetría presente en los poros femorales de los individuos de P. algirus de las poblaciones expuestas a contaminación.

La riqueza de especies y la diversidad de aves no mostró variación a lo largo de los seis años de seguimiento tras la restaraución (2000-2006), excepto para la abundancia. La abundancia total aumentó a lo largo de los años, pero siempre fue menor que la encontrada en el Arroyo del Alcarayón, un medio ripario, equivalente estructuralmente y próximo al río Guadiamar, pero no afectado por el vertido. Varias de las especies presentes en el la llanura de inundación incrementaron su abundancia con respecto a las especies ligadas a la vegetación de ribera. Los resultados indicaron que la comunidad de aves quedó afectada por la falta en la disponibilidad de hábitat, debido a la escasa cobertura de la vegetación presente en los primeros años tras la restauración.

La cobertura de la vegetación riparia y la heterogeneidad del paisaje que rodea al corredor, fueron las variables explicaron la mayor variación de la comunidad de aves a lo largo del corredor en el último año de estudio. La presencia de otros ecosistemas riparios cercanos al corredor podrían potencar a conectividad de la cuenca del Guadiamar y la conservación de su diversidad biológica.

Los refugios resultaron ser útiles para potenciar la recuperación de la comunidad de reptiles en zonas resturadas. *Tarentola mauritanica* parece ser la única especie que se mantuvo en los límites del corredor durante la expansión de los lodos tóxicos, refugiándose en los grandes árboles. Posteriormente, la especie que se estableció y adquirió la mayor abundancia fue *Psammodromus algirus,* por su carácter generalista, y favorecida por la estructura de la vegetación. Las especies que recolonizaron con éxito las zonas restauradas con refugios fueron *Timon lepidus*, *Malopolon monspessulanus*, *Hemorrohis hippocrepis* y *Natrix maura*.

Los individuos de las distintas poblaciones de *P. algirus* mostraron que la contaminación aún persiste en el corredor, en comparación con la población control. El As, Tl, Sn, Pb, Cd y Cu son los metales cuyo nivel de concentración se ha detectado con mayor diferencia entre los individuos de poblaciones expuestas a la contaminación y los individuos de la población control

La contaminación no influyó significativamente en el grado de asimetría fluctuante en los poros femorales de *P. algirus* . Lo que si parece es haber una tendencia a que los individuos con mayor nivel de contaminación sean más asimétricos.

A pesar del esfuerzo de los proyectos de restauración, ocho años no son suficientes para recuperar la estructura y función del ecosistema dañado. Por un lado, la contaminación persiste en formas biodisponibles para los organismos, llegando a los niveles superiores de la cadena trófica. Por otro lado, la restauración no favorecido por igual a todos las comunidades biológicas. Las aves con gran capacidad de dispersión y dependientes de la

estructura de la vegetación han colonizado rápidamente el corredor. A medida que la complejidad de la vegetación ha ido incrementando, la disponibilidad del hábitat ha aumentado y por lo tanto la abundancia total en la comunidad de aves. Sin embargo los reptiles debido a su limitada capacidad de dispersión no pudieron escapar al vertido minero, por lo que el efecto sobre estos organismos fue mayor. Además, dado su carácter ectotérmico, precisan de refugios para colonizar nuevas zonas, y la falta de éstos en los hábitats restaurados han influido en su recuperación. Sólo las especies más generalistas y abundantes han recolonizado las zonas restauradas. Por lo que los planes de gestión del corredor deberían considerar los refugios como componente esencial para facilitar la recuperación en la comunidad de reptiles.

INTRODUCCIÓN GENERAL

El crecimiento de la población y del consumo demandan cada vez más recursos naturales cuya explotación ha supuesto el aumento de beneficios económicos y el desarrollo del nivel de vida de la sociedad, pero en contraposición, ya sea mediante el desarrollo de la propia explotación o por la generación de residuos, suponen un riesgo para el medio ambiente y sus ecosistemas (Whisenant, 2002; Rodríguez & García-Cortés, 2006). La explotación de los recursos se traduce en cambios de uso del suelo, deforestación, fragmentación del paisaje, pérdida de hábitat y de especies, y contaminación (Dobson et al., 1997; Hsu et al., 2006). Paralelamente las áreas naturales afectadas son escenarios idóneos para los estudios de contaminación y de restauración ecológica (Twigg & Fox, 1991; Ireland et al., 1994; Passell, 2000; Nichols & Nichols, 2003). En la última década, la comunidad científica ha incrementado el número de estudios relacionados con la *Restauración Ecológica* y la contempla como una nueva disciplina dentro de la Ecología desde 1996 y promete ser una herramienta esperanzadora para la conservación de la biodiversidad (Allen et al., 1997; Niering, 1997; Bradshaw, 2002).

Un ecosistema se caracteriza principalmente por dos atributos, estructura y función, por lo que estos dos conceptos son utilizados para definir e ilustrar el daño que ha sufrido un ecosistema (Bradshaw, 2002). Así pues las claves del esfuerzo de la restauración residen en la recuperación de la estructura y la función de los procesos ecológicos que caracteriza a un ecosistema. Restaurar significa volver al sistema natural lo más parecido posible al estado en el cual se encontraba antes de sufrir el impacto y la *Restauración Ecológica* se define como el proceso mediante el cual se intenta devolver a los ecosistemas degradados la variabilidad estructural, funcional y la diversidad biológica presente antes del impacto, mediante prácticas sostenibles (Society for Ecological Restoration, 1996). Su reciente aceptación como ciencia ha influido en un rápido incremento de trabajos científicos sobre cómo hay que diseñar experimentos de restauración, cómo evaluar el cambio, y que consideraciones deben tenerse en cuenta en la gestión de las zonas restauradas (Michener, 1997;Cairns, 2002; Ormerod, 2003; Ruiz-Jaén & Aide, 2005).

El proceso de recuperación de un ecosistema es un proceso largo en función del tipo de restauración que se aplique, ya sea *activo,* en la que el ser humano se implica completamente en la recuperación del medio, cómo *pasivo,* si no se interviene en el proceso de restauración y dejamos que el ecosistema se recupere con el tiempo (Scott et al., 2001). Independientemente del tipo de restauración, se debe contemplar un diseño experimental dentro de una dimensión temporal y espacial que incluyan zonas restauradas con diferentes tratamientos y zonas control que nos informen de la efectividad de las medidas aplicadas. Por otro lado, es necesario llevar a cabo un seguimiento continuado durante un largo periodo de tiempo, en función de los procesos ecológicos que se deseen recuperar.

Generalmente los proyectos de restauración no evalúan efecto sobre los organismos que recolonizan la zona, siendo un pilar básico en el diseño y ejecución de estos proyectos para lograr los objetivos marcados. Block et al., (2001) sostienen que una medida directa para evaluar el éxito de la restauración es analizar la dinámica de la población, de especies paraguas que representen los requerimientos espaciales y funcionales del resto de los organismos. También plantean como alternativa el uso de medidas indirectas, indicadoras de la calidad de los ecosistemas, como el análisis de la estructura de las comunidades, el cálculo de la riqueza de especies, índices de similitud y diversidad. Todas estas consideraciones son determinantes, sin embargo lo más importante en la restauración es conocer la historia natural del ecosistema a restaurar, las interacciones entre los organismos y factores como el clima, el suelo, la presencia de depredadores, competidores y la historia evolutiva de estas interacciones (Molles, 2006). Uno se los ejemplos de restauración más exitoso que ha tenido en cuenta la historia natural en la restauración de los ecosistemas fue el de Daniel Jazen para la recuperación del bosque tropical seco, en Costa Rica (Janzen 1981a, 1981b). Janzen pudo comprobar que la falta de animales adecuados para la dispersión de semillas de una especie de árbol, el guanacaste (*Enterolobium cylocarpum*) impedía la regeneración natural del bosque. Los potenciales dispersores de sus semillas (camellos, perezosos y caballos) se habían extinguido hace aproximadamente 10. 000 años, pero aparecieron otros introducidos por los europeos como el caballo y el ganado hace 500 años. Estos animales comían y dispersaban frutos alrededor de los límites del parque y ayudaban a la regeneración del bosque natural, pero debido a la exclusión del ganado dentro del Parque la regeneración del guanacaste no evolucionaba. Así pues, Janzen mostró que incluyendo a los caballos y al ganado en el Parque como dispersores de semillas la regeneración del bosque se aceleraba. Además, Janzen implicó a la población en el proyecto para el mantenimiento de estos ecosistemas. Llamó a este tipo de restauración "*Restauración Biocultural*", en el que se demuestra que el buen manejo de los ecosistemas aporta grandes beneficios a la población.

Así pues, además de recuperar la estructura y función de los ecosistemas, la *Restauración Ecológica* tiene por objetivo sensibilizar a la sociedad sobre el mal uso de los recursos y el daño ecológico, aportando nuevas habilidades y conocimientos acorde con su mejor uso (Cairns, 1998).

Impactos de las actividades minero-metalúrgicas en los sistemas fluviales

La explotación de los yacimientos minerales causa la contaminación del medio natural. Actualmente existe una línea de investigación sobre el impacto y el riesgo ambiental de estas actividades y el posterior desarrollo de técnicas de remediación y restauración de las áreas afectadas (Rodríguez & García-Cortés, 2006). Los ecosistemas fluviales son muy vulnerables a la contaminación. El uso de sus recursos hídricos en los procesos de extracción de los minerales y la mala gestión y almacenamiento inadecuado de los residuos producidos puede provocar desastres naturales que alteran la calidad de las aguas (subterráneas, superficiales), de los suelos y por consiguiente, la salud de los organismos (González Tánago & García de Jalón, 2007). Estos ecosistemas son prioritarios para la conservación, ya que en relación directa con la población y la agricultura, y en zonas fragmentadas actúan como corredores ecológicos (Naiman et al., 1993). Sin embargo están entre los ecosistemas más alterados (Sala et al., 2000). Esto se debe a que su gestión siempre ha estado más orientada hacia un intenso aprovechamiento de sus recursos (agua, vegetación riparia, suelos de la llanura de inundación) para usos urbanos, suburbanos, agrícolas, forestales e industriales, que hacia un uso racional y conservacionista (González Tánago & García de Jalón, 2007).

Los ecosistemas fluviales son muy diversos pues albergan gran número de especies y procesos ecológicos (Gregory et al., 1991; Naiman & Décamps, 1997) cuya estructura biológica está configurada por un conjunto de comunidades de organismos pertenecientes al medio acuático, al medio hiporreico y a la vegetación riparia que utilizan el corredor fluvial de forma permanente u ocasional como refugio, área de nidificación, alimentación o migración. Dentro de estas comunidades, los macroinvertebrados son el componente de biomasa animal más importante en muchos tramos de los ríos, formando parte esencial en la alimentación de muchas especies de peces y aves (González Tánago & García de Jalón, 2007). La gestión descontrolada de estos ecosistemas traen como resultado ríos contaminados rodeados de cultivos agrícolas y zonas industriales, causando la disminución de su diversidad biológica (Hulse & Gregory, 2004); por lo que surge la necesidad de abordar la restauración de estos ecosistemas de alto valor biológico (González Tánago & García de Jalón, 2007).

Hace 10 años la balsa de contención de lodos de la mina de Aznalcóllar (provincia de Sevilla), se rompió, vertiendo los residuos mineros a los ríos de alrededor y contaminando y alterando la estructura y las funciones del ecosistema afectado (comprendida entre los 37º 30' y 37º 00' de latitud norte y los 6º 10' y 6º 20' de latitud sur.). Este acontecimiento ha sido una de las mayores catástrofes ambientales ocurridas en España. A continuación describimos detalladamente todo lo sucedido y cómo se abordó.

El vertido minero de Aznalcóllar

El yacimiento minero de Aznalcóllar-Los Frailes se localiza en la Faja Pirítica del Suroeste Ibérico, una de las provincias metalogénicas más importantes del mundo, con grandes reservas de sulfuros masivos polimetálicos, como la pirita (SFe), blenda (ZnS), galena (PbS), calcopirita ($CuFeS_2$), arsenopirita (FeAsS), entre otros (Van Geen, et al., 1999; Aguilar et al., 2003). Los filones se originaron mediante intrusiones volcánicas submarinas de carácter ácido y básico a través de materiales sedimentarios, precipitando como sulfuros metálicos en el fondo del mar creando grandes reservas de minerales, explotadas desde la época de los Romanos (Grimalt et al., 1999 Aguilar et al., 2003).

Las dos explotaciones más importantes en la cuenca del Río Guadiamar han sido "El castillo de las Guardas", que dejó de funcionar en los años 60 del pasado siglo, y Aznalcóllar-Los Frailes, explotada por la compañía, sueco-canadiense, Boliden Apirsa S.L. localizada en el municipio sevillano de Aznalcóllar (Borja et al., 2001). En esta última explotación durante el proceso de extracción de los minerales, se producían ingentes cantidades de residuos. El mineral obtenido se trituraba y se mezclaba con agua (del Río Agrio, que desemboca 5 km río abajo, en el Río Guadiamar), pasando por procesos de flotación, lixiviado y extracción de metales mediante la utilización de reactivos (Aguilar et al., 2003). Parte del agua era depurada y se vertía de nuevo al río; la otra parte se almacenaba en una balsa de decantación de estériles generados durante todo el proceso. Los residuos estaban compuestos fundamentalmente por azufre (S), hierro (Fe), cinc (Zn), plomo (Pb), arsénico (As), cobre (Cu) y antimonio (Sb) (Alaustegui et al., 1999). Desde principios de los años 80 en el Río Agrio y Guadiamar se venían detectando altas concentraciones de Fe y otros metales, procesos de acidificación de las aguas del Río Guadiamar (Olías et al., 2005). Pero, es en noviembre de 1995 cuando se anticipan serios problemas de la balsa de decantación por la mala construcción y escasa impermeabilización (Aguilar Campos, 1995).

En la madrugada del 25 de abril de 1998, se produjo una rotura de 50 m del muro de contención de la balsa, y se vertieron aproximadamente 6 millones de m^3 de residuos tóxicos y aguas ácidas (pH=2) a los ríos Agrio y Guadiamar. Se estima que los lodos vertidos estaban compuestos por 16.000 toneladas (tons) de Zn y Pb, 10.000 tons de Arsénico, 4.000 tons Cu, 100 tons de Sb, 120 tons de Co, 100 de tons Sb, 50 tons de Cd y Ag, 30 tons de Hg, 20 toneladas de Se, y otros metales, afortunadamente todos en forma insoluble (Grimalt et al., 1999). El vertido llegó a afectar 5 Km del Río Agrio y 40 Km del cauce del Río Guadiamar y unos 500m de anchura media; la contaminación llegó hasta límite del Parque Nacional de Doñana a unos 60 km de la balsa minera contaminando una superficie de aproximadamente 4.634 ha (de ellas, 2656 ha afectaron al Parque Natural y 98 ha al Parque Nacional de Doñana). El espesor de la capa de los lodos superaba los 3 m en las zonas más próximas a la mina, mientras que sólo era de unos pocos cm en el tramo más bajo, próximo a las marismas de Entremuros (Montes et al., 2003). También se apreció una variabilidad en la composición del lodo pirítico a lo largo de toda la zona afectada. (Aguilar et al., 2003). Las escasas precipitaciones y la proximidad del verano impidieron un desastre mayor, ya que las lluvias hubieran aumentado la solubilidad de los metales y el nivel de contaminación de los suelos.

Actuaciones tras el vertido minero y el Proyecto del Corredor Verde del Guadiamar

Dada la magnitud y trascendencia por la cercanía de áreas naturales de alto valor ecológico, al norte Sierra Morena y al sur el Parque Nacional y Natural de Doñana (Patrimonio de la Humanidad desde 1994), inmediatamente tras el vertido se puso en marcha un Plan de Medidas Urgentes durante siete meses (Tabla 1). La acción más inmediata fue la construcción de un dique de contención de cierre transversal en el Guadiamar, en Entremuros, antes de desembocar en el río Guadalquivir (cerca de la Vuelta de la Arena), donde el agua retenida fue depurada antes de ser vertida (Antón-Pacheco et al., 2001). Debido al desbordamiento de éste, se construyó un segundo muro. Durante la limpieza de los lodos se recogieron grandes cantidades de animales muertos y se prohibió el consumo y la comercialización de cualquier recurso de la zona afectada (Grimalt et al., 1999

Tabla 1.- Principales actuaciones del Plan de Medidas Urgentes que se llevaron a cabo en la zona afectada por el vertido minero de Aznalcóllar

Fecha	**Acciones**
25 Abril 1998 3:30 hrs.	Rotura de la balsa de contención de los lodos de la mina de Aznalcóllar
25 Abril 1998	Construcción de un dique de contención en Entremuros
26 Abril 1998	Construcción de un 2º dique de contención de las aguas en el límite del Parque Natural de Doñana
27 Abril 1998 – 27 Mayo	Retirada de los organismos que murieron por los lodos tóxicos
28 Abril	Se prohíbe pescar en los cauces afectados y en los alrededores
30 Abril	Se prohíbe el consumo de mariscos
3 Mayo 1998	Empieza la retirada de los lodos en toda la zona afectada
5 Mayo 1998	Se prohíbe el consumo de los productos agrícolas de la zona afectada
13 Mayo 1998	Empiezan a recogerse todos los productos agrícolas del área afectada para su destrucción
9-18 Junio 1998	Comienza la limpieza de las tierras contaminadas
23 Junio	Se prohíbe la comercialización de mariscos del Río Guadalquivir y de las marismas más próximas
24 Julio	Comienzan a depurarse las aguas retenidas en Entremuros
6 Noviembre	Se prohíbe la caza en las provincias de Sevilla, Huelva y Cádiz
23 Diciembre	Se vuelve a permitir la caza
13 Enero 1999	Comienza el proyecto del Corredor Verde del Guadiamar

Fuente: Grimalt et al., 1999

La labor más importante fue la retirada de la vegetación contaminada y de los lodos depositados a lo largo del cauce del río y sus alrededores, incluyendo hasta 20 cm de suelo original subyacente. Tras la limpieza, los suelos perdieron su capa fértil y estructura, más acentuado en los suelos próximos a la mina (Aguilar et al., 2003). En 1999 se comenzaron a tratar los suelos con una primera enmienda de carbonato cálcico e hidróxidos de hierro impidiendo la solubilización, absorción y dispersión de los metales pesados (Galán et al., 2002). Pero la suave topografía y la distribución diferencial de la contaminación en un patrón por mosaicos, complicó las tareas de limpieza, y fue necesaria una segunda enmienda (Querol et al., 2006; Aguilar et al., 2007). Estas labores de limpieza disminuyeron la contaminación en los primeros centímetros de suelo, pero no en los horizontes más profundos, aumentando así el riesgo de contaminación de los acuíferos (Kraus & Wiegand, 2006; Ordóñez et al., 2006).

Tras el Plan de Medidas Urgentes se establece un Plan de Acción denominado *Estrategia del Corredor Verde del Guadiamar* en el que se establecen los fundamentos teóricos, las líneas de trabajo y los procedimientos metodológicos para gestionar la zona afectada contra los efectos del vertido minero. Las cuatro líneas de trabajo establecidas fueron:

1.- Seguimiento, vigilancia, control y remediación de la contaminación.

2.- Diseño del Corredor Verde del Guadiamar.

3.- Restauración ecológica.

4.- Integración sistemas naturales humanos

Así pues, la zona afectada pasa a ser dominio de la Administración Pública y se contempla la cuenca del Guadiamar como unidad espacial de actuación y de gran importancia para el Ecosistema del Litoral de Doñana (CMA, 2001; Montes et al., 2003). Posteriormente, surge el Proyecto del Corredor Verde del Guadiamar, y se incluye a la zona dentro de la Red de Espacios Naturales de Andalucía como "Corredor Verde del Guadiamar", con el objetivo de que actúe como un sistema natural de interconexión entre Sierra Morena Occidental y el litoral de Doñana. Entre el conjunto de actuaciones que se realizan a partir del decreto de protección del Corredor Verde, surgen dos proyectos de investigación promovidos por la Consejería de Medio Ambiente de la Junta de Andalucía, en el que investigadores de diversas universidades y centros de investigación asumen la labor científico-técnica entre los años 1998-2006. El primero, "Programa de Investigación del Corredor Verde del Guadiamar" (PICOVER 1998-2002) se dividió en dos fases. La primera tuvo como objetivo principal realizar una gestión integrada de la Cuenca del Guadiamar que restableciera todas aquellas interacciones y procesos ecológicos que se perdieron tras el vertido, tanto en la llanura aluvial como en el cauce fluvial, (PICOVER, 2003) empezando por mejorar las condiciones edáficas y comunidades vegetales, imprescindibles para el funcionamiento del ecosistema (Allen et al., 2002; Davy, 2002; Whisenant, 2002). En la segunda fase, se evaluó la contaminación y los procesos de recolonización de las comunidades animales.

En el segundo proyecto, "Seguimiento del Corredor Verde del Guadiamar" (SECOVER 2004-2006) se realizó un seguimiento de los estudios realizados durante el PICOVER.

Este ha sido uno de los mayores proyectos de restauración en España, y uno de los escenarios más propicios para realizar experimentos sobre restauración de ecosistemas ribereños, con un paisaje complejo y dinámico que incluye ríos, bosques de ribera, cultivos agrícolas y zonas naturales manejadas. Con la puesta en marcha de estos proyectos podemos comprobar la aplicabilidad de la teoría ecológica y ayuda a avanzar en el conocimiento de la Restauración Ecológica.

La presente tesis doctoral se enmarca dentro de una de las cuatro líneas de investigación de la Estrategia del Corredor Verde del Guadiamar (Restauración Ecológica), en el suproyecto de "*Restauración ecológica de los ecosistemas acuáticos y terrestres: composición, estructura y dinámica de las comunidades vegetales y animales*" y dentro del SECOVER en el "*Seguimiento de los procesos de recolonización de poblaciones y comunidades*".

Nuestro objetivo de estudio fueron las comunidades de aves y reptiles

Las aves y reptiles en zonas restauradas

Como hemos señalado anteriormente, la *Restauración Ecológica* fue una de las cuatro líneas de trabajo de la *Estrategia del Corredor Verde del Guadiamar*, por lo que diversos grupos de investigación analizaron la estructura y dinámica de las comunidades vegetales y animales (micoflora, briófitos, líquenes, nematodos, macroinvertebrados acuáticos, formícidos, lepidópteros, coléopteros edáficos, odonatos, reptiles, aves, peces y mamíferos) (PICOVER, 2003).

Los reptiles y las aves son organismos cuya presencia puede aportarnos información sobre la restauración de un ecosistema. En general, las aves son muy sensibles a cambios en el hábitat, ocupan una posición alta e intermedia en la cadena trófica, son fáciles de detectar, abundantes y existe gran información sobre su biología (Wiens, 1989; Cantebury et al., 2000; Cardoso Da Silva & Vickery, 2002; Burger et al., 2004; Gardali et al., 2006). Dentro de los vertebrados, las aves tienen una gran capacidad de dispersión y normalmente sólo precisan de la presencia de vegetación que les proporcionen alimento, perchas para descansar, y zonas donde hacer sus nidos (McClanahan & Wolfe, 1993). Sin embargo, los reptiles también son buenos indicadores de la salud de los ecosistemas, ya que interaccionan con otras especies debido a su papel como consumidores secundarios, (Salvador, 1998; Jones, 2002); se encuentran repartidos por muchos hábitats y climas, llegan a alcanzar altos niveles de diversidad y biomasa en determinadas zonas, y su rarefacción tiene una notable incidencia en el declive de otras especies o grupos faunísticos, como aves y mamíferos (Márquez & Lizana, 2002). Además, son útiles como bioindicadores locales de polución, ya que acumulan contaminantes, tienen limitada capacidad de dispersión, alta e intermedia posición en la cadena trófica, carácter ubiquista en la dimensión espacial, y carácter generalista en recursos tróficos (Sparling, 2000; Campbell & Campbell, 2002). Los reptiles son animales ectotermos, por lo que precisan de refugios (troncos, grandes piedras y fisuras) para poder aislarse de temperaturas extremas, termorregular, colocar sus huevos, refugiarse de sus depredadores, dispersarse y colonizar nuevas zonas (Huey, 1989; Webb & Shine, 2000; Litt et al., 2001). Por lo tanto, uno de los componentes del hábitat más importante para los reptiles son los refugios. Pero muchos proyectos de restauración del hábitat para vertebrados endotermos, las aves y micromamíferos, no garantizan el reestablecimiento para vertebrados ectotermos como los reptiles, por lo que habría que considerar estos componentes del hábitat en los planes de restauración de zonas degradas (Wike et al., 2000; Buffington et al., 2000). Pero si bien, un elevado número de estudios han analizado cómo responden las aves a procesos de contaminación o restauración ecológica, pocos han sido los que han considerado a los reptiles en los procesos de restauración y en estudios de biomonitorización de contaminantes (Loumbourdis, 1997; Bowers et al., 2000; Cardoso Da Silva, 2002; Jones, 2002; Burger et al., 2006).

OBJETIVOS Y ESTRUCTURA DE LA TESIS

En la cuenca del Río Guadiamar la rápida actuación de los equipos de limpieza y restauración a lo largo de la zona afectada por el vertido minero, favoreció la pronta recuperación de muchas comunidades de organismos ligados al ecosistema ripario. En la presente tesis nuestro objetivo principal fue evaluar la evolución de las comunidades de aves y reptiles después de la limpieza y restauración de la zona afectada por el vertido. Mediante cinco capítulos hemos querido cubrir este objetivo. A continuación exponemos resumidamente el objetivo de cada uno:

- *Capítulo 1*: la hipótesis de estudio es que no ha habido un gran efecto del vertido en la comunidad de aves debido a su gran capacidad de dispersión y a la rápida actuación de las tareas de limpieza y restauración de la vegetación. Para testar esta hipótesis analizamos el gradiente espacial y temporal de los parámetros de riqueza de especies, abundancia y diversidad a lo largo de la zona de influencia del cauce del río, y los comparamos con la comunidad de aves de un arroyo próximo al corredor, no afectado por el vertido.

- *Capítulo 2*: la hipótesis de estudio de este capítulo es que la comunidad de aves a lo largo del corredor está determinada por las variables ambientales que caracterizan el ecosistema, tanto las variables del macrohábitat como las variables del microhábitat. Para testar esta hipótesis, se describirá la comunidad de aves en el último año de estudio (2006) a lo largo del corredor, determinando aquellas variables ambientales que están influyendo significativamente en la distribución espacial de la comunidad.
- *Capítulo 3*: nuestra hipótesis se basa en que la falta de refugios naturales en el las zonas restauradas debido a la retirada de los lodos, está impidiendo una rápida recuperación de la comunidad de reptiles. Para testar esta hipótesis, evaluaremos en que medida el aporte de refugios artificiales ha facilitado la recolonización de los reptiles en cuanto a riqueza de especies y abundancia en zonas restauradas del corredor.
- *Capítulo 4*: nuestra hipótesis de estudio es que la contaminación por metales pesados a pesar del tratamiento de los suelos afectados, aún persiste y está circulando por la cadena trófica. Para ello utilizamos como bioindicador a la especie de reptil más abundante, la lagartija colilarga (*P. algirus*), y determinaremos la concentración de metales en individuos de poblaciones expuestas a contaminación en la zona afectada y zonas no afectadas por el vertido. De esta manera podemos demostrar si *P. algirus* puede ser un bioindicador a largo plazo en los programas de biomonitorización de la contaminación en todo el área afectada.

- *Capítulo 5*: la hipótesis de partida es que las poblaciones de *P. algirus* expuestas a altos niveles de contaminación presentan un mayor grado de asimetría en los poros femorales durante el desarrollo ontogénico de los individuos. Para testar nuestra hipótesis de estudio, determinaremos el grado de asimetría fluctuante en los poros femorales de los individuos de la especie en poblaciones expuestas a altos niveles de contaminación en metales pesados con respecto a otras poblaciones no expuestas.

La información resultante de estas investigaciones será canalizada a los técnicos responsables de la gestión del Corredor Verde del Guadiamar para que realicen una mejor gestión y conservación de las comunidades de aves y reptiles en el ecosistema dañado por el vertido, favoreciendo así la futura conexión entre Sierra Morena Occidental y el litoral de Doñana.

Bibliografía

Aguilar Campos M. Rapport submitted to the Ministry of Environment of the Andalusian Autonomous Goverment, 1995.

Aguilar, J., C. Dorronsoro, R. Beller, E. Fernández, J. Fernández, I. García, A. Iriarte, F. Martín. I. Ortiz, M. Simón. 2003. Contaminación de suelos tras el vertido tóxico de Aznalcóllar. Universidad de Granada.

Aguilar, J., C. Dorronsoro, E. Fernández, J. Fernández, I. García, F. Martín, M. Sierra, M. Simón. 2007. Remediation of As-Contaminated Soils in the Guadiamar River basin (SW, Spain) Water Air and Soil Pollution 180: 109–118.

Antón-Pacheco C., J.C. Arranz., D. Barettino, G. Carrero, M. Giménez, J.A. Gómez, B. Martínez Pledel, E. de Miguel, J. Moreno, G. Ortíz, J.G. Rejas, A. Silgado, E.M. Vázquez. 2001. Actuaciones para el reconocimiento y retirada de los lodos depositados sobre el terreno, y su restauración edáfica y morfológica. Boletín Geológico Minero, 112 (Vol. Especial): 93-121.

Alastuey A., A. García-Sánchez, F. López, X. Querol (1999) Evolution of pyrite mud weathering and mobility of heavy metals in the Guadiamar valley after the Aznalcóllar spill, south-west Spain. Sci Total Environ 242:41-55.

Allen, E. B., W.W. Covington, D.A. Falk. 1997. Developing the conceptual basis for restoration ecology. Restoration Ecology 5, 275–276.

Allen, M. F., D. A. Jasper, and J. C. Zak. 2002. Micro-organism, in Perrow, M. R., Davy, A. J.(Eds). Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 257-278.

Block, W.M., A.B. Franklin, J.P. Ward, Jr. J.L. Ganey, G.C. White. 2001. Design implementation of monitoring studies to evaluate the success of ecological restoration on wildlife. Restoration Ecology 9, 293-303.

Borja, F., J.A. López Geta, M. Martín Machuca, R. Mantecón, C. Mediavilla, P. del Olmo, R. Palancar, R. Vives. 2001. Marco geográfico, geológico e hidrológico regional de la cuenca del Guadiamar. Boletín Geológico y Minero, 112 (Vol.Especial), 13-34.

Bowers, C.F., H.G. Hanlin, D.C. Guynn Jr, J.P. Mclendon, J.R. Davis. 2000. Herpetofaunal and vegetation of a thermally-impacted stream at the beginning of restoration. Ecological Engineering 15:101-114.

Bradshaw, A.D. 2002. Introduction and philosophy. In: Handbook of Ecological restoration. Vol.1: Principles of Restoration. Eds. M. R. Perrow and A. J. Davy, 3-9.

Buffington, J. M., J.C. Kilgo, R.A. Sargent, K.V. Miller, and B.R. Chapman. 2000. Effects of restoration techniques on breeding birds in a thermally-impacted bottomland hardwood forest. Ecological Engineering 15:115-120.

Burger, J.,R. Bowman, G.E. Woolfenden, M. Gochfeld. 2004. Metal and metalloid concentrations in the eggs of threatened Florida scrub-jays in suburban habitat f rom south-central Florida. Science of the Total Environment 328:185-193.

Burger J.,S. Murria, K. F.Gaines, J.M. Novak, T. Punshon, C. Dixon, M. Gochfeld. 2006. Element levels in snakes in South Carolina: differences between a control site and an exposed site on Savannah River site. Environmental Monitoring and Assessment Assess 112:35-52.

Cairns J.Jr. 1998. Eco-Societal restoration: rehabilitating human society's life support system in Rana BC (Ed). Damaged Ecosystems and Restoration. Sardar Patel University, India. 1-23p

Cairns J Jr. 2002. Rationale for Restoration, in Perrow, M. R., Davy, A. J.(Eds). Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 10-23.

Campbell K. R., T.S. Campbell. 2002. A logical starting for developing priorities for lizard and snake ecotoxicology: a review of available data. Environmental Toxicology and Chemistry 21:894-8.

Cantebury, G.E., T.H. Martin, D.R. Petit, L.J. Petit, D.F. Bradfords. 2000. Bird community and habitat as ecological indicators of forest condition in regional monitoring. Conservation Biology 14, 544-558.

Cardoso Da Silva, J.M., Vickery P. D. 2002. Birds, in Perrow, M. R., Davy, A. J.(Eds). Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 376-388.

CMA, 2001. El Corredor Verde del Guadiamar. Abril 1998-Abril 2001. Consejería del Medio Ambiente. Junta de Andalucía.

Davy, A. J. 2002. Establishment and manipulation of plant populations and communities in terrestrial ecosystems, in Perrow, M. R., Davy, A. J.(Eds). Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 223-241.

Dobson, A.P., A.D. Bradshaw, A. J. M. Baker. 1997. Hopes for the Future: Restoration Ecology and Conservation Biology. Science 277:515 – 522.

Galán, E., I. González, J. C. Fernández-Caliani. 2002. Residual load of soils impacted by the Aznalcóllar (Spain) mining spill after clean-up operations. The Science of the Total Environment 286:167-179.

Gardali, T., A.L. Holmes, S. Small, N. Nur, G.R. Geupel, G.H. Golet. 2006. Abundance patterns of landbirds in restored and remant riparian forest on Sacramento River, California, U.S.A. Restoration Ecology 14:391-403.

González Tanago, M., García de Jalón, D. 2007. Restauración de Río-Guía Metodológica para la elaboración de proyectos. Edita Secretaría General Técnica del Ministerio de Medio Ambiente.

Gregory, S. V., F.J. Swanson, W.A. McKee, K.W.Cummins. 1991. An ecosystems perspective of riparian zone. Bioscience, 41:540-551.

Grimalt, J.O., M. Ferrer, E. Macpherson. 1999 The mine tailing accident in Aznalcóllar. Science of the Total Environmental 242:3-11.

Hulse, D., S. Gregory. 2004. Integrating resilience into floodplain restoration. Urban Ecosystems 7:295–314.

Hsu, M.J., K. Selvaraj, G. Agoramoorthy. 2006. Taiwan's industrial heavy metal pollution threatens terrestrial biota. Environmental Pollution 143:327-334.

Ireland, T.T, G.L. Wolters, S.D. Schemnitz. 1994. Recolonization of wildlife on a coal strip-mine in northewestern New-Mexico. Southwestern Naturalist 39:53-57.

Janzen, D.H. 1981a. Guanacaste tree seed-swallowing by Costa Range horse. Ecology 62:587-592.

Janzen, D.H. 1981b. Enterolobium cyclocarpum seed passage rate and survival in horses, Costa Rican Pleistocene seed dispersal agents. Ecology 62:593-601.

Jones, C. G. 2002. Reptiles and amphibians. In: Handbook of Ecological restoration. Vol.1: Principles of Restoration (Perrow and Davy). Eds. M. R. Perrow and A. J. Davy. 355-375.

Kraus, U., J. Wiegand. 2006. Long-term effects of the Aznalcóllar mine spill—heavy metal content and mobility in soils and sediments of the Guadiamar river valley (SW Spain) Uta Science of the Total Environment 367:855-871.

Lake, P.S., N. Bond, P. Reich. 2007. Linking ecological theory with stream restoration. Freshwater Biology 52:597-615.

Litt, A.R., L. Provencher, G.W. Tanner, R. Franz. 2001. Herpetofaunal Responses to Restoration Treatments of Longleaf Pine Sandhills in Florida. Restoration Ecology 9, 462-474.

Loumbourdis, N.S. 1997. Heavy metal contamination in lizard, *Agama stellio stellio,* compared in urban, high altitude and agricultural, low altitude areas of north Greece. Environmental Contamination and Toxicology 58:945-952.

Márquez, R., Lizana, M. 2002. Conservación de los anfibios y reptiles de España. En: Atlas y Libro Rojo de los Anfibios y Reptiles de España (Pleguezuelos, J.M., R. Márquez y M. Lizana, eds). Dirección general de Conservación de la Naturaleza. Asociación Herpetológica Española (2ª impresión), Madrid, 418– 453.

McClanahan, T.R., R.W. Wolfe. 1993. Accelerating forest succession in a fragmented landscape: the role of birds and perches. Conservation Biology 7:279-288.

Michener, W.K. 1997. Quantitatively evaluating restoration experiments: research design, statistical analysis, & data management considerations. Restoration. Ecology, 5:324-338.

Molles, M.C. Jr. 2006. Ecología: Conceptos y aplicaciones. 3º ed. McGraw-Hill ,

Montes, C., J. M. Arenas, F. Borja. 2003. Iniciativa cientifico-técnica del PICOVER. En: Ciencia y Restauración del Río Guadiamar. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla.

Naiman, R.J., H. Décamps, M. Pollock. 1993. The role of riparian corridors in maintaining regional biodiversity. Ecological Applications 3:209-212

Naiman, R. J., N. Décamps. 1997. The ecology of interfaces: riparian zones. Annual Review of Ecology and Systematics 28:621-658.

Nichols,O.G, F.M. Nichols. 2003. Long-term trends in faunal recolonization after bauxite mining in the Jarrah forest of southwestern Australia. Restoration Ecology 1:261-272.

Niering, W.A. 1997. Editorial: Human-dominated ecosystems and the role of restoration ecology. Restoration Ecology 5:273-274.

Olías M., Cerón J.C., Moral F. 2005. Impacto del vertido minero de Aznalcóllar en la calidad del agua del Río Guadiamar. Geogaceta 37: 119-122.

Ormerod, S.J. 2003. Restoration in applied ecology: editor's introduction. Journal of Applied Ecology, 40:44–50.

Ordóñez, R., J.V. Giráldez, K. Vanderlinden, R. Carbonell, P. González. 2007. Temporal and spatial monitoring of the pH and heavy metals in a soil polluted by mine spill post cleaning effects. Water Air and Soil Pollution 178: 229–243.

Palmer M.A., R.F. Ambrose, N.L. Poff 1997. Ecological theory and community restoration ecology. Restoration Ecology 5:291-300.

Passell, H.D. 2000. Recovery of bird species in minimally restored Indonesian tin strip mines. Restoration Ecology 8:112-118.

PICOVER. 2003. Ciencia y Restauración del Río Guadiamar. Junta de Andalucía. (ed.), Consejería de Medio Ambiente, Sevilla. 578pp.

Querol X, Alastuey A, Moreno N, Alvarez-Ayuso E, García-Sánchez A, Cama J, Ayora C, Simón M (2006) Immobilization of heavy metals in polluted soils by the addition of zeolitic material synthesized from coal fly ash. Chemosphere 62:171-180.

Rodríguez, R., García-Cortés, A. 2006. Los Residuos Mineros Metalúrgicos en el Medio Ambiente. Instituto Geológico y Minero de España, Madrid.

Ruiz-Jaen, M.C., Aide T.M., 2005. Restoration Success: How is it being measured? Restoration Ecology, 13:569-577.

Sala, O.E., F.S., Chapin, J. J. Armesto et al., 2000. Biodiversity – global biodiversity scenarios for the year 2100. Science, 287: 1770-1774.

Salvador, A. (Coordinador), 1997. Reptiles. En: Fauna Ibérica, vol. 10. Ramos, M. A. et. al.(Eds). Museo Nacional de Ciencias Naturales. CSIC. Madrid. 705 pp.

Sanpera, C., M. Morera, X. Ruíz, L. Jover 2000. Variability of mercury and selenium levels in clutches of Audouin's gulls (*Larus audouinii*) breeding at the Chafarinas Islands, southwest Mediterranean. Environ Contam Toxicol 39:119-123.

Scott, T.A., W. Wehtje, M. Wehtje. 2001. The need for strategic planning in passive restoration of wildlife populations. Restoration Ecology 9:262-271.

Society for Ecological Restoration. 1996. Ecological Restoration: Definition. http://www.ser.org.

Sparling, D.W., C. Bishop, G. Linder. 2000. Ecotoxicology of amphibians and reptiles. 875 pp.

Twigg, L. E., B.J. Fox . 1991. Recolonization of regenerating open forest by terrestrial lizards following sand mining. Australian Journal of Ecology 16:137-148.

Urbanska, K. M., N. R.Webb, P.J. Edwards 1997. Why restoration?. Pages 3-7 in K.M. Urbanska, N.R., Webb, P.J. Edwards, editors. Restoration Ecology & sustanaible development. University Press, Cambridge, United Kingdom.

Van Straalen N.M., R.O. Butovsky, A.D. Pokarzhevskii, A.S. Zaitsev, C.S. Verhoef. 2001. Metal concentrations in soil and in invertebrates in the vicinity of a metallurgical factory near Tula (Russia). Pedobiologia 45:451-466.

Webb, J.K., Shine, R. 2000. Paving the way for habitat restoration: can artificial rocks restore degraded habitats of endangered reptiles?. Biological Conservation 92, 93-99.

Westman, W.E., 1991. Ecological restoration projects: measuring their performance. The Environamental Proffessional, 13:207-215.

Wiens, J.A., 1989. The ecology of bird communities. Cambridge University Press. Cambridge.

Wike L., Douglas M., Hanlin H., Paddock L. 2000. Small mammal populations in restored stream corridor. Ecological Engineering 15, 121-129.

Whisenant, S.G. Terrestrial systems, in Perrow, M.R., Davy, A.J.(Eds). 2002. Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 83-105.

White, P.S., Walker J.L. 1997. Approximating nature's variation: selecting and using reference information in restoration ecology. Restoration Ecology 5:338-349.

CAPÍTULO

Evolution of the breeding-bird community after the mine accident and restoration of the Guadiamar River Basin (SW Spain)

Rocío Márquez-Ferrando, Diego Ontiveros, Juan M. Pleguezuelos, Xavier Santos and Juan R. Fernández-Cardenete

(*Submitted to Journal of Environmental Management*)

Abstract

The recovery of breeding-bird community along the Guadiamar River has been studied for six years after the environmental disaster caused by the mine accident of Aznalcóllar and the subsequent restoration of the area. Values of α- and β-diversity were high just one year after vegetation restoration (three years after the mine accident) and thereafter, species richness and Shannon-Weaver diversity index remained stable during the study period, although bird abundance showed a slight trend to increase. Moreover, the Morisita-Horn similarity index showed a high degree of overlap over time in bird-species composition (close to 80% of bird-community similarity in the last few years). When compared with a control river near the Guadiamar River but not affected by the mine spill, bird communities were similar in species richness and α-diversity throughout the study period, although abundance values were higher for the river unaffected by the spill. Because of the high values for descriptors of the breeding-bird community (species richness, diversity, abundance) immediately after the disaster, we conclude that the restoration program of the Guadiamar Green Corridor has been successful for the breeding bird community due to rapid implementation (providing habitat availability), and high response capacity of the birds.

Keywords: Aznalcóllar mine spill; restoration; riparian corridor; breeding birds.

1.- Introduction

River systems are key in maintaining biodiversity in ecosystems by providing a high diversity of habitats to numerous aquatic and terrestrial biota (Naiman and Décamps, 1997; Miller et al., 2004). However, negative impact by human activities in riparian habitats, such as livestock grazing, timber harvesting, hunting, agriculture, recreation activities, mining, and wastewater discharge, diminish riverine biodiversity (Dobkin et al., 1998; Buffington et al., 2000). Although riparian recovery actions are relatively frequent in degraded habitats, little information is available on whether native faunal communities can be reassembled in conjunction with the restoration of vegetation composition (Ohmart, 1994).

On April 1998, the Agrio and Guadiamar rivers and floodplains were polluted by the Aznalcóllar acid tailings from a mine spill consisting in 6×10^6 m^3 of a sludge rich in heavy metals (zinc, lead, copper, antimony, cobalt, thallium, bismuth, cadmium, silver, mercury, manganese and arsenic). The tailings spread along 5 Km of the Agrio River and more than 40 Km downstream in the Guadiamar River, covering agricultural land, polluting marshes in the Doñana National Park, and affecting riparian vegetation and plant succession. Both qualitatively and quantitatively, this was the largest environmental pollution accident recorded in Spain (Pain et al., 1998; Gallard et al., 1999; Grimalt et al., 1999; Van Geen et al., 1999).

Immediately after the disaster, a procedure of compulsory purchase of the affected area started and a restoration program for the polluted area was established by the Regional Administration. The program involved mechanical removal of the tailings and rehabilitation of the natural vegetation in the 1999-2001 period (Aguilar et al., 2003; PICOVER, 2003). The area was protected and designated as the Guadiamar Green Corridor, and conceived to connect two large natural areas, Sierra Morena in the north and Doñana National Park in the south (Fig. 1). The great importance of the corridors to connect natural areas (Hobbs et al., 1990), and the great number of bird species that depend on these areas for nesting, migration and dispersal, promoted interest in analyzing the impact of the mine accident on the ecosystem and the breeding-bird community response to the post-restoration process.

Restoration of degraded ecosystems is based on the assumption that rebuilding the habitat will result in the rehabilitation of natural communities (Palmer et al., 1997; Block et al., 2001; Litt et al., 2001). Previous studies in the Guadiamar Green Corridor indicated that some organisms strongly affected by the spill are becoming re-established (Solá et al., 2004; Cárdenas and Hidalgo, 2006; Luque et al., 2007). However, despite the carefully planned Guadiamar restoration program, the management actions have been harmful for some taxa, such as reptiles (Márquez-Ferrando et al., in press), and the response of the breeding-bird community has not yet been evaluated.

The aim of this work was to assess the evolution of the breeding-bird community in the Guadiamar Green Corridor, studying the possible variation of avian species richness, abundance and diversity from 2001 to 2006, and compare the data with those of a nearby unaffected river.

2.- Materials and methods

2.1.- Study area

The Guadiamar River, a major tributary of the Guadalquivir River, is located in the southwestern Iberian Peninsula (Fig. 1). The climate is Mediterranean with Atlantic influence, characterized by mild winters (mean temperature of the coldest month, January, 7.1° C), hot summers (mean temperature of the hottest month, July, 26.9º C), and mean annual rainfall of 560 mm, concentrated mainly in winter and with a dry season in summer (data on 30-year standard meteorological averages, from the Sanlúcar la Mayor weather station, representative of the study area).

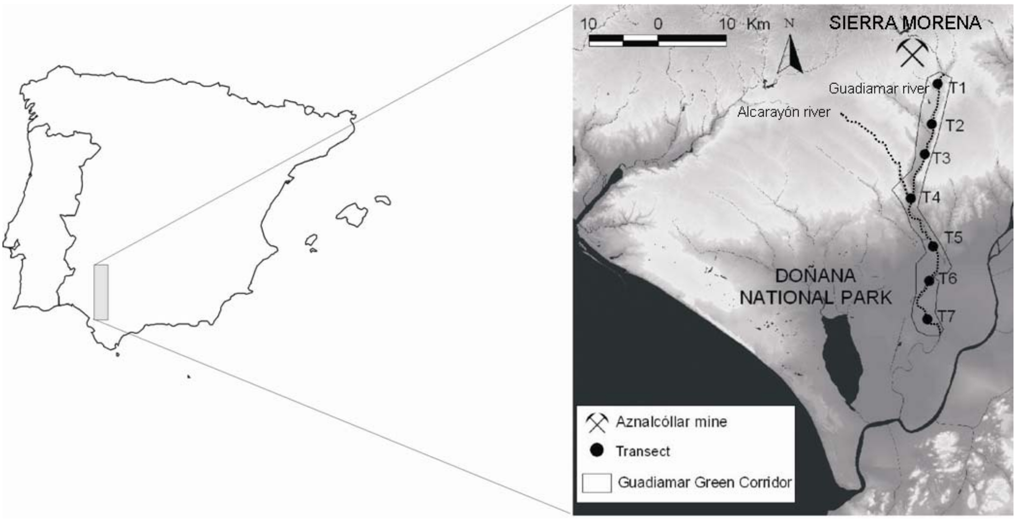


Figure 1. Map for the location of the study area in the Iberian Peninsula, and detailed map for the study area (southwestern Spain). The small circles represent transect location selected to the bird census along the Guadiamar Green Corridor.

In the Guadiamar Green Corridor, measuring 62 km long and averaging c. 500 m wide, two landscape units are clearly differentiated: the riverside (a belt of about 10 m width), and the floodplain, re-vegetated in the area reached by the spill (Guadiamar Green Corridor) after the disaster and cleanup actions. The predominant vegetation along the riverside, in the upper and middle part of the corridor, was composed of alders (*Alnus glutinosa*), willows (*Salix* sp.), poplars (*Populus alba*), and eucalyptus (*Eucaliptus globulus*) as trees, and shrubs such oleanders (*Nerium oleander*), blackberries (*Rubus ulmifolius*), ivy (*Hedera helix*), and tamarisk (*Tamarix* *africana*). Floodplain includes extensive grass cover for livestock grazing, an agroecosystem (*dehesa* in Spanish*)* composed of evergreen oak (*Quercus rotundifolia*) and cork oak (*Q. suber*), together with diverse shrub and tree species that were planted in the restoration program, such as pines (*Pinus pinaster*), olive tree (*Olea europaea*), rosemary (*Rosmarinus officinalis*), arbutus (*Arbutus unedo*), mastic plant (*Pistacia lentiscus*), and rockroses (*Cistus* sp.). In the lower part of the corridor marshlands predominate, with glassworts (*Salicornia* sp.), bulrush (*Thypha dominguensis*), and reeds (*Phragmite* sp.) along the riverside, all of them adapted to the high water saturation of the soil (Aguilar et al., 2003).

For evaluating bird response in disturbed ecosystems, previous data on bird community of the study site is ideal, but, if such data are unavailable, it is necessary to examine the natural fluctuations in the bird community of unaffected sites (Patten and Rotenberry, 1998). The Alcarayón River, untouched by the spill and 10 km western of the Guadiamar River (Fig. 1), was surveyed in order to compare bird-community disparities between the rivers. To compare affected and unaffected areas, two reference sites are recommended (Ruiz-Jaén and Aide, 2005), but in the study area this was not possible, since there is only one river close to the Guadiamar with similar characteristics. The breeding-bird community between the rivers was compared only for the sector T4 (Fig. 1) of the Guadiamar River, which was the most similar to the Alcarayón River in vegetation composition, both dominated by shrubs (height < 4m) such as oleanders, tamarisk, ivies and blackberries, and trees such as willows, eucalyptus, poplars and alders.

2.2.- Avian surveys

We established seven equidistant transects (8 km apart) along the Guadiamar Green Corridor (Fig.1). From transect one to transect four, the corridor is characterized by riparian habitat, but from transect five to seven the dominant habitat is a marshland. Birds were surveyed using line transects 2000 m in length (Bibby et al., 2000), parallel to the river and close to the riparian vegetation. Censuses were performed for five years (2001-2006 period, except year 2003), by an observer on foot, between 06.00-09.30 h., on days of good visibility, walking at a speed of 1.5 km/hour. Rainy or excessively windy days were avoided because birds are less active during these conditions and are difficult to detect. Each transect was visited twice each breeding season (within the period 15 April-15 June), and records of birds were pooled for a mean value of the two censuses. The Alcarayón River was surveyed by the same method, with a 2000 m transect in length during the breeding season, between the period 2004-2006. The distance from the birds to the observer was measured with a laser range-finder device (Bushnell Trophy^{R}; range 5-732 m; accuracy ± 1 m.) when reflective objects were available, but by eye when necessary. For a more robust estimate of bird abundance, observations were truncated for each species at a distance at which bird-detection frequency declined sharply for each species (Bibby et al., 2000).

To test possible temporal changes in bird abundance within the corridor, we split the breeding-bird species into two groups: those exclusive of riparian vegetation (riverside species), and species that used mainly the restored sites outside the riverside (floodplain species, Table 1). Swifts and swallows appeared frequently, but, being very numerous and thus uncountable as well as high overhead, they were considered only to calculate species richness. In this way, species barely detected in the censuses were also disregarded for the abundance calculation (Table 1). Species not nesting in the corridor, but in its immediate vicinity, and using the corridor for foraging or roosting, were considered.

2.3.- Data analysis

The α- and β-diversity, being measures traditionally used in the analysis of diversity in space and time (Magurran, 2004), were assessed in the Guadiamar and Alcarayón rivers during the study period. The α-diversity, the diversity of a spatially defined unit, was measured in three ways: species richness (number of species recorded per year), abundance (number of individuals/10 has.), and Shannon-Weaver diversity index (calculated from the algorithm $H' = -\sum piLnpi$; where pi is the proportion of individuals per species), performed by the EcoSim v5.53 (Gotelli and Entsminger, 2005). The β-diversity measures the resemblance of communities in species composition and abundance between two or more spatial or temporal units. In this sense, Sørensen's and Morisita-Horn's similarity indexes were computed for the 2001-2006 period in the corridor. The Sørensen's index is a qualitative measure that determines the proportion of presence/absence of the species two years have in common; it is calculated by the algorithm $Is = 2c/a+b$, where a and b are the number of the species present in the two years, and c is the number of species that the two years have in common. Morisita-Horn's index is a quantitative measure of similarity not influenced by species richness or sample size, defined as $CMH = 2\sum(a_i b_i)/(da+db)(NaNb)$, where Na is the total number of individuals at year a, Nb is the total number of individuals at year b, a_i is the number of individuals for the i species in year a, b_i is the number of individuals for i species in year b, and da (and db) is calculated as $da = \sum ai^2 Na^2$. The Sørensen and Morisita-Horn similarity indexes were calculated using

EstimateS v7.5 (Colwell, 2005). Values of these indexes closer to 1.0 indicate high similarity between communities.

3.- Results

From 2001 to 2006, 93 bird species were recorded in the breeding season in the Guadiamar Green Corridor. Within the sub-group of 63 species considered for density analyses (see Material and Methods; Table 1), only five species exhibited a significant inter-annual increase (*Bubulcus ibis, Falco tinnunculus*, *Cyanopica cyanus, Lullula arborea*, *Serinus serinus,* and *Hippolais polyglotta*), while two increased nearly to significance (*Alectoris rufa* and *Streptopelia turtur*).

The results did not indicate a trend in species richness or diversity within the Guadiamar Green Corridor during the study period after restoration, either in each transect individually (r_s <0.80, $p > 0.10$, considering species richness, abundance and Shannon-Weaver diversity index), or when the seven transects were pooled ($r_s = 0.10$, $p = 0.87$; $r_s = 0.10$, $p = 0.87$; richness and diversity, respectively). Species richness increased by 11% and diversity by 6% from 2001 to 2006, although these values really increased only between 2001 and 2002, and were practically stable from year 2002 to year 2006 (Fig. 2). However, bird abundance increased significantly and gradually up to 35% in the 2001-2006 period ($r_s = 0.90$, $p = 0.04$, Fig. 2).

Table 1. Breeding bird species and mean abundance (N indv/10 has) in the seven transects along the Guadiamar Green Corridor during the 2001-2006 period. The Spearman correlation coefficient between abundance and year of census is reported only for 63 species. 1: riverside species; 2: floodplain species (see Material and Methods); species with significant correlation were highlight in bold type (next page).

Species	**Mean abundance**	**rs**	***p***	**Species**	**Mean abundance**	**rs**	***p***
Acrocephalus arundinaceus[1]	0.35	-0.78	0.12	*Hirundo daurica*	-	-	-
Acrocephalus scirpaceus[1]	3.09	0.30	0.62	*Hirundo rustica*	-	-	-
Actitis hypoleucos	-	-	-	*Ixobrychus minutus*	-	-	-
Alcedo atthis[1]	0.14	0.35	0.56	*Jynx torquilla*	-	-	-
Alectoris rufa[2]	0.26	0.87	0.05	*Lanius meridionalis*[2]	0.02	-0.22	0.72
Anas platyrhynchos[1]	1.50	-0.10	0.87	*Lanius senator*[2]	0.98	0.70	0.19
Apus apus	-	-	-	*Larus michahellis*	-	-	-
Apus pallidus	-	-	-	*Locustella luscinioides*	-	-	-
Ardea cinerea	0.22	0.56	0.32	***Lulula arborea***[2]	0.37	0.89	0.04
Ardea purpurea[1]	0.22	0.70	0.19	*Luscinia megarhynchos*[1]	0.84	0.21	0.74
Ardeola ralloides	-	-	-	*Melanocorypha calandra*	-	-	-
Bubulcus ibis[2]	0.80	1	<0.01	*Merops apiaster*[2]	3.77	0.60	0.28
Burhinus oedicnemus	-	-	-	*Milvus migrans*[2]	0.14	0.30	0.62
Buteo buteo	0.05	0	1	*Milvus milvus*	-	-	-
Calandrella brachydactyla[2]	0.81	0.30	0.62	*Motacilla flava*[1]	0.48	-0.72	0.17
Calandrella rufescens[2]	1.44	-0.10	0.87	*Nycticorax nycticorax*[1]	0.62	-0.70	0.19
Caprimulgus ruficollis	0.04	0.71	0.18	*Oriolus oriolus*[1]	0.02	0	1
Carduelis cannabina	-	-	-	*Otus scops*	-	-	-
Carduelis carduelis[2]	10.71	0.10	0.87	*Parus caeruleus*[1]	0.25	0.11	0.86
Carduelis chloris	1.62	0.70	0.19	*Parus major*[1]	0.79	0.36	0.55
Cettia cetti[1]	1.27	0.70	0.19	*Passer domesticus*	6.38	-0.10	0.87
Charadrius alexandrinus	-	-	-	*Passer hispaniolensis*	1.12	0.21	0.74
Charadrius dubius[1]	2.02	-0.82	0.09	*Passer montanus*	0.46	-0.41	0.49
Ciconia ciconia	0.16	0.30	0.62	*Phylloscopus bonelli*[1]	0.79	-0.26	0.67
Circus aeruginosus	0.01	0.67	0.22	*Phylloscopus ibericus*[1]	1.14	0.67	0.22
Circus pygargus	-	-	-	*Picus viridis*[2]	0.01	-0.35	0.56
Cisticola juncidis	5.61	-0.60	0.28	*Platalea leucorodia*[1]	0.29	0.15	0.80
Columba livia[2]	0.01	0.35	0.56	*Porphyrio porphyrio*[1]	0.55	0.21	0.74
Corvus corax	-	-	-	*Pterocles alchata*	-	-	-
Coturnix coturnix[2]	0.19	-0.21	0.74	*Riparia riparia*	-	-	-
Cuculus canorus	0.04	0	1	*Saxicola torquata*[2]	0.07	0	1
Cyanopica cyanus[2]	0.75	0.97	<0.01	***Serinus serinus***	10.35	0.90	0.04
Delichon urbica	-	-	-	*Streptopelia decaocto*[2]	0.74	0.10	0.87
Dendrocopos major	-	-	-	*Streptopelia turtur*[1]	0.19	0.87	0.05
Egretta alba[*]	-	-	-	*Sturnus unicolor*[2]	0.30	-0.10	0.87
Egretta garzetta[1]	0.13	0.60	0.28	*Sylvia atricapilla*[1]	0.18	-0.11	0.86
Elanus caeruleus	0	-	-	*Sylvia cantillans*[*]	-	-	-
Emberiza calandra[2]	1.14	0.36	0.55	*Sylvia communis*[1*]	0.05	0.29	0.64
Erithacus rubecula	0	-	-	*Sylvia conspicillata*[2]	0.2	-0.78	0.12
Falco tinnunculus[2]	0.07	0.90	0.04	*Sylvia melanocephala*[2]	0.14	-0.58	0.31
Galerida cristata[2]	5.38	0	1	*Sylvia undata*[2]	0.05	0.71	0.18
Gallinula chloropus[1]	0.65	-0.6	0.28	*Tachybaptus ruficollis*	-	-	-
Glareola pratincola	0	-	-	*Tringa totanus*[1]	0.04	0.35	0.56
Hieraaetus pennatus	0.05	-0.32	0.60	*Turdus merula*[1]	3.35	0.50	0.39
Himantopus himantopus[1]	0.10	0	1	*Turdus viscivorus*	0	-	-
Hippolais pallida[1]	0.68	0.30	0.62	*Upupa epops*[2]	0.68	-0.30	0.62
Hippolais polyglotta[1]	2.05	1	<0.01				

* Migrating species that not breed in the region

In terms of the habitat distribution within corridor, 23 species used mainly the restored sites in floodplain whereas 27 species were recorded exclusively along the riverside (Table 1). For the only variable that showed an increase over time (abundance), the increase was significant for floodplain but not for riverside species (r_s = 0.90, p = 0.04; r_s = 0.70, p = 0.19; respectively).

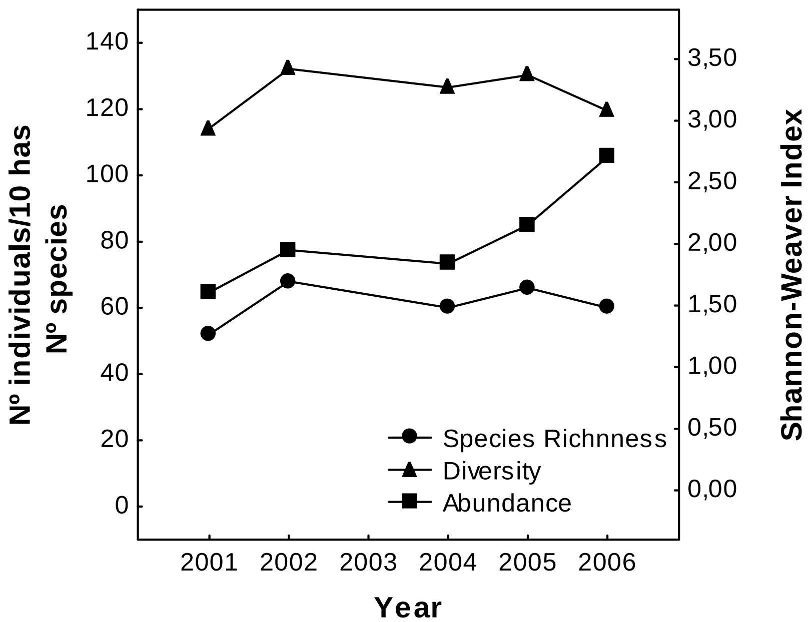


Figure 2. Species richness, abundance, and Shannon-Weaver diversity index for the breeding bird community during the 2001-2006 period in the Guadiamar Green Corridor (southwestern Spain).

Sørensen's index varied little over the study period (six years), and we found a high degree of bird-community overlap, with similitude values close to 0.70 in all the comparisons between years (Table 2). However, the

Morisita-Horn's index detected a pattern in the between-years overlap of the bird communities, decreasing with time (from 75% to 48% of overlap; Table 2).

Table 2. Sørensen Similarity Index and Morisita-Horn Index (in italics) for the breeding bird communities of the Guadiamar Green Corridor in a comparison between pairs of years.

Years	2001	2002	2004	2005	2006
2001	X	0.75	0.75	0.68	0.68
2002	*0.75*	X	0.75	0.75	0.70
2004	*0.57*	*0.83*	X	0.78	0.81
2005	*0.53*	*0.74*	*0.85*	X	0.70
2006	*0.48*	*0.73*	*0.79*	*0.74*	X

We found no differences between the two rivers in species richness and diversity (Table 3). However, the Alcarayón River had a higher value of abundance in relation to the compared stretch (T4) of the Guadiamar River (Table 3; Fig. 3).

Table 3. Comparisons for mean species richness, abundance and Shannon-Weaver diversity index between the breeding-bird communities of the Guadiamar Green Corridor (transect 4) and the Alcarayón River (reference and unaffected river), during el 2004-2006 period. Means are followed by standard error.

	Guadiamar River	Alcarayón River	Mann-Whitney test	*p*
Richness	24.66 ±3.05	23.33±1.53	0.43	0.66
Abundance	88.84±29.43	146.69±20.61	-1.96	0.05
Diversity	2.59±0.17	2.21±0.26	1.53	0.13

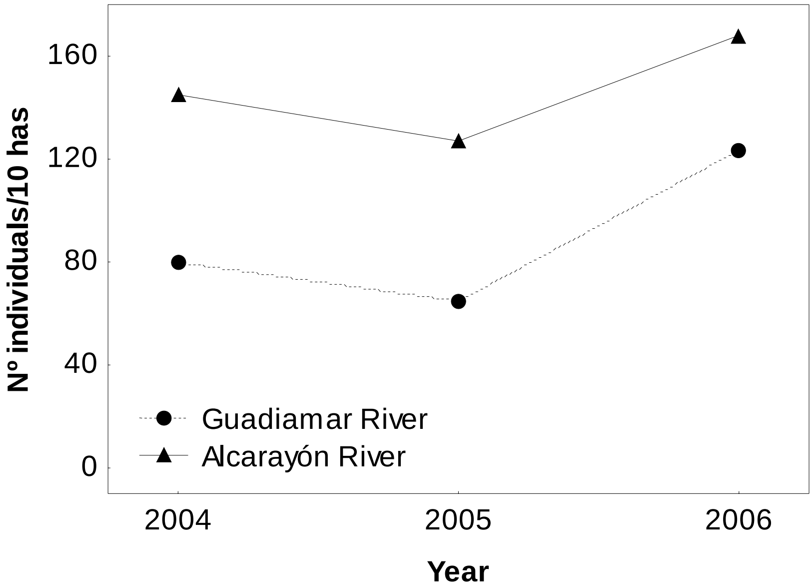


Figure 3. Abundance of the breeding bird communities in the Guadiamar Green Corridor and Alcarayón River (2004-2006 period).

4.- Discussion

The response of bioindicators such as birds should be taken into account by environmental policy makers in order to comprehend overall trends in human-caused environmental change (Read et al., 2005). Many studies have demonstrated that birds are good indicators for evaluating the success of large-scale ecosystem restoration and for stream-ecosystem bioassessment, because birds are well known and easy to detect, and are sensitive to changes in habitat structure and composition (Wiens, 1989; Cantebury et al., 2000; Cardoso da Silva and Vickery, 2002; Suárez-Seoane et al., 2002; Gardali et al., 2006).

After the toxic spill, the restoration of the Guadiamar River and floodplains was intended to augment vegetation complexity, favoring animal-community recovery. Many case studies support the idea that habitat restoration provides animal-community reestablishment by management practices that create habitat mosaics that benefit the highest number of species (Galán, 1996; Litt et al., 2001; Nichols and Nichols, 2003). In the Guadiamar basin all taxa with life cycles depending on the river, and terrestrial taxa with limited dispersion capacity such as arthropods and reptiles, were the more affected by the toxic spill (PICOVER, 2003). For these animals, species richness, abundance or diversity, have increased over time in the restored Guadiamar Green Corridor (Solá et al., 2004; Cárdenas and Hidalgo, 2006; Luque et al., 2007; Márquez-Ferrando et al., in press). However, birds have a high dispersion capacity and are more resilient than are many other animals when faced with human-induced disturbance (Wiens, 1989).

Studying restored riparian habitat in North Carolina (USA), Buffington et al. (2000) found that re-vegetation techniques employed to restore the zone had little effect on the avian communities two years post-treatment. However, Passell (2000) found that the richness, abundance, and diversity of the bird communities in Indonesian tip strip mines increased significantly over three years in restored zones. Species richness and diversity increased by 11% and 6% respectively, in the Guadiamar Green Corridor, whereas in a similar period (seven years), Nichols and Nichols (2003) found an increase of 433% and 110% respectively, in a recolonization process of a rehabilitated areas. Therefore, circumstances of restored areas can be very different according their size and biogeographic situation; consequently, it is very important to consider the degree of the previous impact and the extent of natural areas surrounding the altered zone, as they will probably act as source of colonizers (Anderson 1993; Passell 2000; Cardoso da Silva and Vickery, 2002; Brotons et al., 2005; Brawn, 2006).

Ours results have shown that in the Guadiamar Green Corridor one year after the vegetation restoration (2001), values in species richness and diversity index were rather high for the bird breeding community, and did not indicate significant variation over time. We suspect that because of the absence of the bird surveys the first two years after the disturbance (because of restricted access to the contaminated area), low values for species richness, abundance and diversity were not detected. However, bird abundance increased over time, mainly due to the group of species that used the floodplains of the corridor. These are species that nest in mosaic-habitats types, such as open grass, shrubby vegetation, and *dehesas* (Martí and Del Moral, 2003), and forage on different trophic resources (Table 1), thus, their increase in abundance did not appears to be related with changes in specific trophic resources within the corridor.

The similarity index of bird community (Sørensen's index) was homogeneous over time, and only when we considered species number plus bird abundance (Morisita-Horn index) did changes appear between years. The high similarity value in the bird community in the second half of the study period, could indicate that a stable community arrived in a few years. Other systems need 16-45 years after the disturbances to reach such similarity values in bird community (Stuart-Smith et al., 2006).

No previous data is available for the breeding-bird community in the Guadiamar River, but curiously, the comparisons of data between affected and unaffected rivers did not reveals differences in species richness or diversity index. Notwithstanding, bird abundance was higher in the unaffected river, but this difference was presumably due to the higher vegetation density in Alcarayón River (unpub. data of the authors). Similar results were reported by Stuart-Smith et al. (2006), who found that the residual tree density and tree species composition affected bird community abundance but not bird species richness and diversity. Gardali et al. (2006) suggested that, in restored areas, some large-scale phenomenon such as climate might be responsible of bird-population trends rather than the revegetation process. Thus, the similar evolution of abundance between Guadiamar and Alcarayón rivers could be explained in two ways, the beneficial effects of restoration in the Guadiamar River, and the environmental factors affecting both zones.

Conclusions and management implications

We did not detect a major change in the bird community after the restoration of the Guadiamar Green Corridor. Two main circumstances could explain this fact:

i) The accident (which occurred in 1998) was followed by a rapid soil cleanup and vegetation restoration (1999-2001), which favored a simultaneous rapid increase in birds by augmenting habitat availability (providing foraging, perching and nesting sites).

ii) The area affected by the accident was long, narrow, and in close contact to a rather well-conserved agro-ecosystem which could have acted as refuge for the breeding bird community during the cleaning-up of the toxic tailings.

We also suggest that the restoration of the bird community accomplished objectives of the environmental managers. In fact, the comparison with a reference site at the end of the study period showed similar values in species richness and diversity for affected and unaffected zones. Therefore, the recovery plan designed for the Guadiamar Green Corridor appears to be an adequate tool for similar scenarios of affected ecosystems in the Mediterranean area.

Acknowledgments

We thank Esmeralda Alaminos, Jesús Caro, Asier Herrero and Gregorio Moreno-Rueda, for helping during field work, and Adela González-Megías and Manolo Pizarro, for technical support. This study has been funded by the PICOVER and SECOVER programs, for the 2000-2002 and 2004-2006 periods. Xavier Santos was supported by a Beatriu de Pinós postdoctoral grant (BP-B1 10211) from the Generalitat de Catalunya.

References

Aguilar, J., C. Dorronsoro, R. Beller, E. Fernández, J. Fernández, I. García, A. Iriarte, F. Martín, I. Ortiz, M. Simón. 2003. Contaminación de suelos tras el vertido tóxico de Aznalcóllar. Universidad de Granada, Granada.

Anderson, A.N., 1993. Ants as indicators of restoration success at a uranium mine in tropical Australia. Restoration Ecology 9:156-167.

Bibby, J.C., N.D. Burgess, D.A Hill, S.Mustoe. 2000. Bird census techniques. Academic Press, London.

Block, W.M., A.B. Franklin, Jr. J.P. Ward, J.L. Ganey, G.C. White. 2001. Design and implementation of monitoring studies to evaluate the success of ecological restoration on wildlife. Restoration Ecology 9:293-303.

Brawn, J.D., 2006. Effects of restoring oak savannas on bird communities and populations. Conservation Biology 20:460-469.

Brotons, L., P. Pons, S. Herrando. 2005. Colonization of dynamic Mediterranean landscape: where do birds come from after fire? Journal of Biogeography 32:789-798.

Buffington, J.M., J.C. Kilgo, R.A. Sargent, K.V. Miller, B.R. Chapman. 2000. Effects of restoration techniques on breeding birds in a thermally-impacted bottomland hardwood forest. Ecological Engineering 15:115-120.

Cantebury, G.E., T.H. Martin, D.R. Petit, L.J. Petit, D.F. Bradfords. 2000. Bird community and habitat as ecological indicators of forest condition in regional monitoring. Conservation Biology 14:544-558.

Cárdenas, A.M., J.M. Hidalgo. 2006. Ecological impact assessment of the Aznalcóllar mine toxic spill on edaphic coleopteran communities in the Guadiamar River basin (Southern Iberian Peninsula). Biological Conservation 15:361-383.

Cardoso Da Silva, J. M., P. D. Vickery. 2002. Birds, in Perrow, M. R., Davy, A. J.(Eds). Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 376-388.

Colwell, R. K. 2005. EstimateS: statistical estimation of species richness and shared species from samples. Version 7.5. Persistent URL http://purl.oclc.org/estimates (accessed April 2007).

Dobkin, D.S., A.C. Rich, Pyle, W.H., 1998. Habitat and avifaunal recovery from livestock grazing in a riparian meadow system of the northwestern great basin. Conservation Biology 12, 209-221.

Galán, P., 1996. Colonization of spoil benches o fan opencast lignite mine in Northwest Spain by amphibians and reptiles. Biological Conservation 79, 187-195.

Gallart, F., Benito, G., Martín-Vide, J.P., Benito, A., Prió, J. M., Regües, D., 1999. Fluvial geomorphology and hydrology in the dispersal and fate of pyrite mud particles released by the Aznalcóllar mine tailing spill. The Science of Total Environment 242, 13-26.

Gardali, T., Holmes, A.L., Small, S., Nur, N., Geupel, G.R., Golet, G.H., 2006. Abundance patterns of landbirds in restored and remant riparian forest on Sacramento River, California, U.S.A. Restoration Ecology 14, 391-403.

Gotelli, N.J., Entsminger, D.F. 2005. EcoSim: null model software for ecology. Version 7.72. Acquired intelligence Inc. & Kesey-Bear. http://homepages.together.net/~gentmin/ecosim.htm> (accessed in April 2007).

Grimalt, J.O., Ferrer, M., Macpherson, E., 1999. The mine tailing accident in Aznalcóllar. The Science of Total Environment 242, 3-11.

Hartung, S.C., Brawn, J.D., 2005. Effects of savanna restoration on the foraging ecology of insectivorous birds. Condor 107, 879-888.

Hobbs, R.J., Hussey, B.M.J., Saunders, D.A., 1990. Nature conservation: the role of corridors. Journal of Environmental Management 31, 93-94.

Litt, A.R., Provencher, L., Tanner, G.W., Franz, R., 2001. Herpetofaunal Responses to Restoration Treatments of Longleaf Pine Sandhills in Florida. Restoration Ecology 9, 462-474.

Luque, G.M., Reyes-López, J., Fernández-Haeger, J. 2007. Recovery of ground ant (Hymenoptera: Formicidae) communities six years after a major environmental disaster. Environmental entomology 36, 337-347.

Magurran, A.E. 2004. Measuring Biological Diversity. Oxford: Blackwell Publishing.

Márquez-Ferrando, R., Pleguezuelos, J.M., Santos, X., Ontiveros, D., Fernández-Cardenete, J.R., in press., 2007. Recovering the reptile community after the mine-tailing accident of Aznalcóllar (SW Spain). Restoration Ecology.

Martí, R., Del Moral, J.C., 2003. Atlas de las aves reproductoras de España. Dirección General de Conservación de la Naturaleza-Sociedad Española de Ornitología, Madrid.

Meharg, A.A., Osborn, D., Pain, D.J., Sánchez, A., Naveso, M.A., 1999. Contamination of Doñana food-chains after the Aznalcóllar mine disaster. Environmnetal Pollution 105, 387-390.

Miller, J.R., Dixon, M.D., Turner, M.G., 2004. Response of avian communities in large-river floodplains to environmental variation at multiple scales. Ecological Applications 14, 1394-1410.

Naiman, R.J., Décamps, N., 1997. The ecology of interfaces: riparian zones. Annual

Review of Ecology and Systematics 28, 621-658.

Nichols, O.G, Nichols, F.M., 2003. Long-term trends in faunal recolonization after bauxite mining in the Jarrah forest of southwestern Australia. Restoration Ecology 11, 261-272.

Ohmart, R.D., 1994. The effects of human-induced changes on the avifauna of western riparian habitats, in Jehl, J.R., Johnson, N.K., (Eds), A century of avifaunal change in wester North America. Studies in avian biology nº15. Cooper Ornithological Society, Lawrence, Kansas, pp. 273-285.

Pain, D.J., Sánchez, A., Meharg, A.A., 1998. The Doñana disaster: contamination of a world heritage estuarine marsh ecosystem with acidified pyrite mine waste. The Science of Total Environment 222, 45-54.

Palmer, M.A., Ambrose, R.F., Poff., N.L., 1997. Ecological theory and community restoration ecology. Restoration Ecology 5, 291-300.

Passell, H.D., 2000. Recovery of bird species in minimally restored Indonesian tin strip mines. Restoration Ecology 8, 112-118.

Patten, M.A., Rotenberry, J.T., 1998. Post-disturbance changes in a desert breeding bird community. Journal of Field Ornithology 69, 614-625.

PICOVER, 2003. Ciencia y Restauración del Río Guadiamar. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla.

Read, J.L., Kovac, K.J. and Fatchen, T.J. 2005. "Biohyets": a method for displaying the extent and severity of environmental impacts. Journal of Environmental Management 77, 157-164.

Ruíz-Jaen, M.C., Mitchell, A., 2005. Restoration success: how is being measured? Restoration Ecology 13, 569-577.

Solà, C., Burgos, M., Plazuelo, A., Toja, J., Plans M., Prat, N., 2004. Heavy metal bioaccumulation and macroinvertebrate community changes in a Mediterranean stream affected by acid mine drainage and an accidental spill (Guadiamar River, SW Spain). Science of The Total Environment 333, 109-126.

Stuart-Smith, A.K., Hayes, J.P., Schieck, J., 2006. The influence of wildfire, logging and residual tree density on bird communities in the northern Rocky Mountains. Forest Ecology and Management 231, 1-17.

Suárez-Seoane, S., Osborne, P.E., Baudry, J., 2002. Responses of birds of different biogeographic origins and habitat requirements to agricultural land abandonment in northern Spain. Biological Conservation 105, 333-344.

Wiens, J.A., 1989. The ecology of bird communities. Cambridge University Press. Cambridge

.

CAPÍTULO 2

The breeding bird community of the Guadiamar Green Corridor (SW Spain) eight years after a mine tailing accident: the importance of surrounding areas

Rocío Márquez-Ferrando, Diego Ontiveros,
Juan M. Pleguezuelos and Xavier Santos

Abstract

The rivers systems are ecosystems critically strongly affected by human activities affecting with respect to their natural succession and biodiversity, and birds are contemplated as very good ecological indicators for long-term monitoring programs in rivers. In this paper, we present data of factors affecting spacial variation of breeding bird community of the Guadiamar Green Corridor at 2006, eight years after the mine-tailing spill of Aznalcóllar (SW Iberian Peninsula). We measured some landscape variables to describe the corridor and determine which explain the composition and spatial distribution of the breeding bird community. Fifty two species presently breed in the corridor and any gradient in abundance or diversity was detected along the 62 km affected by the spill; species richness was lower in marshlands than in riparian stretches. Species more abundant in the corridor were those related with open and riparian habitats, as well as generalist ones. The multivariate models showed the relationship between environmental variables and bird species distribution. Principal Component Analysis explained 76 % of the variance in bird abundance, and variables selected were those related with vegetation complexity and the amount of surrounding minor streams. A more demanding but realistic model (Stepwise Redundancy Analysis) explained 53.5 % of breeding bird abundance and distribution within the corridor. Variables selected were those related to riparian habitat (within corridor), as well as cover of olive tree orchards and amount of streams (both in surrounding areas). Conservation policies in the Guadiamar Green Corridor, must preserve riparian habitats, but also estimate the contribution of surrounding habitats in the maintenance of animal diversity within this managed green corridor.

Keywords: Aznalcóllar mine spill; restoration; riparian corridor; breeding birds.

1.- Introduction

River ecosystems has been studied in many areas of the world, and these studies have manifested that river systems support high diversity related to the characteristic of the river environment itself, but also to the surrounding lands (Gregory et al., 1991; Knopf and Samson, 1994; Machtans et al., 1996; Naiman and Décamps, 1997; Woinarski et al., 2000).The riparian zones are also of ecological significance because often includes the few habitats remaining for diverse taxa within agro-ecosystem landscapes, hence, the conservation of these systems being vital to maintaining the diversity in such landscapes (Keller et al., 1993; Deschenes et al., 2003; Henningsen and Best, 2005; Smiley et al., 2007). Although rivers are of the ecosystems most connected to human activities in populated areas (Dobkin et al., 1998; Buffington et al., 2000; Mason et al., 2006), the river ecosystems exhibit some resilient capacity, rather elevated diversity, functioning as natural corridors in fragmented landscapes (Naiman et al., 1993). It is frequent for these ecosystems to be contemplated in restoration connectivity projects (Lake et al., 2007).

The Guadiamar Green Corridor, in Southwestern Spain, is a very good example of this reality, and has been recently the subject of a major restoration program since year 2000. It is a protected and restored area, that comprises the Guadiamar River and its alluvial plain, a major tributary of the Guadalquivir River (the most important river of southern Iberian Peninsula).

In April 1998, the Guadiamar basin was polluted by the Aznalcóllar mine spill with sludge and acid tailings riches in several heavy metals that extended 62 km along and aprox. one km across the alluvial plain (Pain et al., 1998; Gallard et al., 1999). After cleaning labors, a vegetation restoration project began in the affected area trying to recover, even to ameliorate, the state previous to disaster (Aguilar et al., 2003; PICOVER, 2003). The protection and management of the Guadiamar Green Corridor progressively have gained interest at regional and national scales because it could act in the near future connecting two huge natural and protected areas, Sierra Morena in the north and Doñana Natural Park in the south (Grimalt et al., 1999).

The first to carry out a good management of this area is to determine the organism communities that use the corridor for foraging, nesting, refuge, migration and dispersion. The study of the bird community dwelling in the river systems is among the tasks that better contribute to our understanding about these ecosystems (Sorace et al., 1999; Cantebury et al., 2000). The impacts of the spoil benches of the Aznalcóllar mine on bird community of the Guadiamar Green Corridor has been previously evaluated by assessing shift in richness, abundance and ecological diversity over time (unp. data), but the spatial distribution of the bird community in relation with the actual landscape of corridor (six years after restoration) have not been yet studied. This analysis should enhance the understanding and the determination of those zones which support the highest species richness, abundances and diversity in birds, something crucial to improve the effectiveness and predictability of ecosystem restoration.

Ours aims in the present study is to analyze the situation of the breeding bird community of the Guadiamar Green Corridor eight years after the mine tail accident, and six years after the habitat restoration, in three ways: i) describe richness species, abundance and diversity of breeding bird community along the 62 km of the corridor; ii) to explain the variability in bird community composition in relation to environmental variables, and iii) provide recommendations to improve the corridor management in the future.

2.- Materials and methods

2.1. Study area

The study was carried out in the, Guadiamar Green Corridor , in the southwestern Iberian Peninsula (Fig. 1). The corridor has 62 km in length and averaged 500 m wide. Within the corridor we distinguish two very different habitats , the narrow riverside, a belt of about 10 m width (without high variation along the river), and the floodplain, re-vegetated after the mine disaster. Riverside consists of eucalyptus (*Eucaliptus globulus*) that were planted few years ago, willows (*Salix* sp.), poplars (*Populus alba*), and shrubs such as oleanders (*Nerium oleander*), blackberries (*Rubus ulmifolius*), ivy (*Hedera helix*), and tamarisk (*Tamarix africana*). The vegetation planted within the corridor was of natural habitat in the region and Mediterranean shrub species such as *Thymus vulgaris* (Thyme), *Lavandula stoechas* (Lavender), *Cistus* sp. (Rockrose), *Rosmarinus officinalis* (Rosemary), *Arbutus unedo* (Arbutus), and trees, such as *Olea europea* (Wild-olive tree), *Q. suber* (Cork oak), *Q. rotundifolia* (Holm oak) were planted.

The corridor is surrounded by a mixed woodland-farmland landscape, such as extensive agroecosystems for livestock grazing, farming and pine forest. In the southern part, marshlands predominates the landscape (PICOVER, 2003). The climate of the area is Mediterranean, characterized by temperate winters (mean temperature of the coldest month, January, 7.1º C), hot summers (mean temperature of the hottest month, July, 26.9º C), and mean annual rainfall of 560 mm, concentrated mainly in winter and with a dry season in summer (data on 30-year standard meteorological averages, from the Sanlúcar la Mayor weather station, representative of the study area).

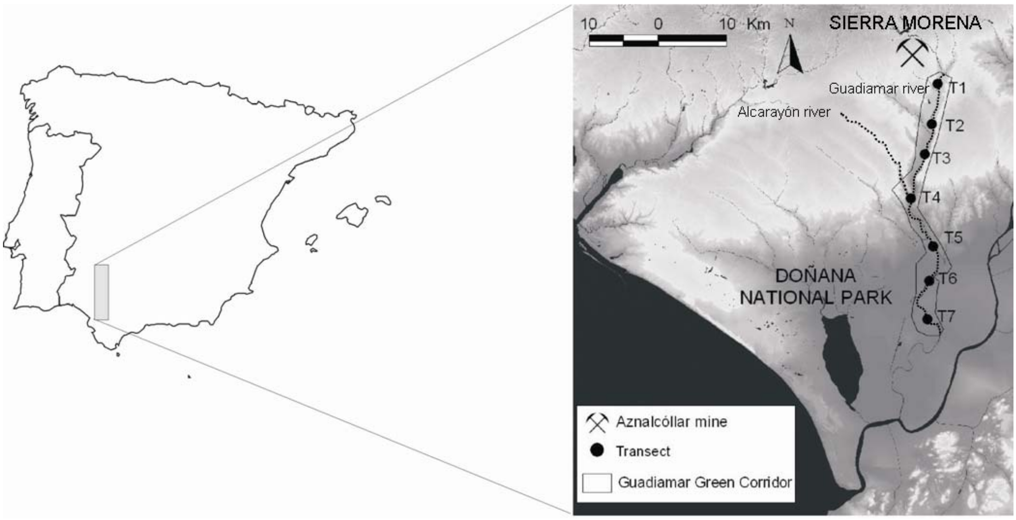


Figure 1. Map for the location of the study area in the Iberian Peninsula, and detailed map for the study area (southwestern Spain). The small circles represent transect location selected to the bird census along the Guadiamar Green Corridor.

2.2.- Samplings data

Bird sampling were carry out twice and late in spring, between May and June of 2006 (to capture peak singing of long-distant migrants), and records of birds were pooled for a mean value of the two censuses. Seven line-transects (2000 m in length) were conducted along the corridor parallel to the river and close to the riparian vegetation, and each transect was separated 8 km from the other. Censuses were completed early in the morning, between 06.00-09.30 h , by an observer on foot (at speed of 1.5 km/hour), in days of good conditions to reduce problems with detectability (Bibby et al., 2000). We recorded the number of all bird species seen and heard, the distance (in metres) from the observer to the bird, and the angle of the detection with respect the line transect (θ) to calculate bird abundance (Bibby et al., 2000). We use a laser range-finder device (Bushnell Trophy[R]; range 5-732 m; accuracy ± 1 m) to measure the distant from the birds to the observer when possible (existence of suitable reflecting surface).

The multiple birds guilds that characterized a bird community can be useful for asses status and trends in ecological conditions of the ecosystems (Canterbury et al., 2000). So, birds were lumped in guilds according to use of microhabitat for foraging: species that occur outside riparian strips as belonging to *open, shrubs*, and *trees habitats*; bird species that occur within river itself as belonging to *riparian* and *aquatic habitats*; and *generalist* for those species that do not occur in specific habitats (Table 1). The rare birds species and those very numerous overhead and uncountable (swifts and swallows) were summarized in category others, excluded of abundance computation and considered only for the measurement of species richness.

Several environmental variables were measured to describe the spatial distribution of breeding birds community (Table 2). Macrohabitat variables were measured from a Geographic Information System data of the Guadiamar Green Corridor database for year 2004, with ArcView 3.2 software, in a radius of 2000 m from central point of each transect. Microhabitats variables were measured twice every 500 m along the transects, once next to riparian vegetation and once 100 m apart, in the floodplain, that is, ten measurement points per transect. In each measurement point, we marked four radiuses, towards cardinal directions, five meters length, measuring in each meter and obtaining 20 data by measurement point. We averaged all microhabitat data for each transect to provide a single estimate of them.

Table 1.- Abundance of breeding birds species in the Guadiamar Green Corridor (SW Spain). Species are grouped in guilds.

	Code	T1	T2	T3	T4	T5	T6	T7
Open habitats								
Alectoris rufa	AlecRuf	1.25	0	0	1.67	0	0	0
Callandrella rufescens	CalRuf	0	0	0	0	3.33	3.33	5
Caprimulgus ruficollis	CapRuf	0	1.25	0	0	0	0	0
Carduelis carduelis	CarCar	5	27.5	1.25	1.67	18.33	18.75	12.5
Ciconia ciconia	CicCic	0.25	0	0	0.67	0	0.25	0
Circus aeruginosus	CirAer	0	0	0	0	0	0.33	0
Cisticola juncidis	CisJun	5	0	0.83	1.11	15.56	10.83	2.5
Falco tinnunculus	FalTin	0	0	0.18	0.48	0	0	0
Galerita cristata	GalCris	5.83	0.83	0.83	5.56	4.44	0.83	8.33
Lullula arborea	LullArb	1.25	0	0	1.67	0	2.5	1.25
Merops apiaster	MerApi	23.13	10	19.38	0	12.5	0	0
Milaria calandra	MilCal	0	0	2.5	0	0	0	2.5
Serinus serinus	SerSer	2.5	45	12.5	31.67	16.67	21.25	18.75
Upupa epops	UpuEpo	0.83	1.67	0	0	0	0	0
Species richness (abundance in total)		9 (46.04)	5 (86.85)	6 (37.47)	7 (43.83)	6 (71.66)	8 (59.07)	6 (50.83)
Shrub habitats								
Sylvia undata	SylUnd	0	1.67	0	0	0	0	0
Total of open habitat species richness		0 (0)	1 (1.67)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Forestal habitats								
Cyanopica cyanus	CyaCya	15.83	0	0	1.11	0	0	0
Dendrocopos major	DenMaj	0	0	0	0.67	0	0	0
Parus major	ParMaj	0	1.25	0	1.67	8.33	0	0
Species richness (abundance in total)		1 (15.83)	1 (1.25)	0 (0)	3 (3.45)	1 (8.33)	0 (0)	0 (0)
Riparian vegetation habitats								
Acrocephalus scirpaceus	AcrScir	6.67	7.5	15.83	8.89	10	4.17	0.83
Alcedo atthis	AlcAtt	0	1.25	0	0	0	0	0
Ardea purpurea	ArdPur	0.42	0	0.42	0.56	0	0	0
Cettia cetti	CetCet	2.5	0.63	1.88	6.67	1.67	0	0
Hippolais pallida	HipPall	0	10	0.83	2.22	0	0	0
Hippolais polyglotta	HipPol	0	0.83	2.5	0	16.67	0	0
Luscinia megarhynchos	LusMeg	1.87	0.63	4.37	3.13	1.88	0	0
Nycticorax nycticorax	NycNyc	0	0	0	0	1.67	0	0
Phylloscopus ibericus	PhyIber	0	0	0	0	0	0	6.25
Turdus merula	TurMe	2.5	22.5	2.5	26.67	0	0	0
Species richness (abundance in total)		5 (13.97)	7 (43.34)	7 (28.34)	6 (48.14)	5 (31.88)	1 (4.17)	2 (7.08)
Aquatic habitats								
Anas platyrhynchos	AnaPla	2.5	1	1	0	0	1	0
Ardea cinerea	ArdCin	0	0	0	0	0.67	0.5	0.5
Egretta garzetta	EgrGar	0	0	0	0	0	0	0.75

Gallinula chloropus	GalChlo	1.25	0	0	0	0.83	1.25	0
Platalea leucorodia	PlaLeu	0	0	0	0	0	3.5	0.25
Species richness (abundance in total)		2 (3.75)	1 (1)	1 (1)	0 (1)	2 (1.5)	3 (6.25)	3 (1.25)

(Continue)	**Code**	**T1**	**T2**	**T3**	**T4**	**T5**	**T6**	**T7**
Generalist								
Bubulcus ibis	BubIbi	3.25	0	0	7.33	6.33	0.25	0.75
Carduelis chloris	CarChlo	0.63	4.38	0.63	6.67	0	0	0
Hieraaetus pennatus	HiePen	0	0	0	0.17	0.33	0	0
Lanius senator	LanSen	7.5	0.83	0.83	3.33	1.11	0	0
Milvus milvus	MilMil	0.13	0	0	1.33	0.17	0	0
Passer domesticus	PasDom	1.25	0	15	4.17	15	0	0
Passer hipaniolensis	PasHisp	4.69	8.75	15	4.17	0	0	0
Passer montanus	PasMon	0	1.25	0	0	0	0	0
Streptopelia decaocto	StrepDec	1.25	0	0	1.67	0	0	0
Streptopelia turtur	StrepTur	2.5	0	0	0	0	0	0
Species richness (abundance in total)		8 (21.2)	4 (15.21)	4 (31.46)	8 (28.84)	5 (22.84)	1 (0.25)	1 (0.75)
Others								
Apus pallidus	-							
Apus apus	-							
Circus pygargus	-							
Delichon urbica	-							
Elanus caeruleus	-							
Glareola pratincola	-							
Hirundo daurica	-							
Hirundo rustica	-							
Riparia riparia	-							

2.3.- Data analysis

We estimated species richness (total number of species), abundance (number of individuals per 10 has) and Shannon index (H') for each units of the analysis (transects). To measure the abundance for each species, the distance from the birds to the observer was truncated at which bird-detection frequency declined sharply (Bibby et al., 2000). The H' was calculated using EcoSim v5.53 (Goetelli & Entsminger, 2005).

We used multivariate methods to explain variability in bird community composition in relation to environmental variables measured across the corridor. The analyses were performed by CANOCO 4.5 (Leps & Smilauer, 2003), to detect bird assemblages with similar habitat associations and habitat types supporting similar bird assemblages. Both indirect and direct gradient analyses were performed because our aim is to extract patterns from the variation explained by the species (indirect gradient analysis) and the variation explained by each environmental variable with respect to bird species data (direct gradient analysis). The indirect gradient method, analyze all variance of biological data in relation with variance of environmental data; opposite, the direct gradient analysis operate only with the portion of biological data that are related with environmental variation. Thus, Detrended Correspondence Analyses (DCA) for indirect gradient analysis and Detrended Canonical Correspondence Analysis (DCCA) for direct gradient analysis was used to determine the adequate approach (unimodal or linear) that adjusted better with our data set (Leps & Smilauer, 2003).

Firstly, we used the full range of environmental variables (18 variables; Table 2) to quantify the effect upon the bird community across the corridor, but as analysis highlighted those variables that exhibiting high inflation values (detecting multicollinearity), for the final analyses we reduce the number of variables.

For indirect gradient analysis, we only considered the canocical axes which explained more than average variability explained by individual axis (Leps & Smilauer, 2003), and for direct gradient analysis we generated a model using a forward stepwise procedure to select the environmental variables and their importance for explain species abundance. The significant level of the analyses was assessed by a Monte-Carlo permutation test, with

499 randomizations. Significant environmental variables were grouped into two sets; macrohabitat and microhabitat variables, to detect their significant effect separately.

The variables measured in percentage were transformed by $y' =$ arcoSen (Sqrt y/ 100), and the rest of environmental variables by $y' = \log(y+1)$. The abundance of the species was transformed by CANOCO software by a log-transformation (Leps & Smilauer, 2003).

Table 2.- Environmental variables considered to characterise landscape in the Guadiamar Green Corridor (SW Spain).

Variables	Description
Macrohabitat variables	
DISTMIN	Distance from transect to Aznalcóllar mine (m)
MINREACH	Average width that mine sludge reached at each transect point when accident (m)
DISTSTRE	distance from transect to nearest stream (m)
LENGSTRE	Average length of the streams within a 2 km radius circle in each transect (m)
OLIVCOV	Percentage of olive tree orchard within a 2 km radius circle from the middle of each transect
OAKCOV	Percentage of dehesa (oak trees) within a 2 km radius circle from the middle of each transect
SCRUBFOR	Percentage of mixed scrubs and forest within a 2 km radius circle from the middle of each transect
CULTGRAIN	Percentage of cultivated grain field within a 2 km radius circle from the middle of each transect
PASTCOV	Percentage of pasture within a 2 km radius circle from the middle of each transect
FRUITCOV	Percentage of tree orchards within a 2 km radius circle from the middle of each transect
IRRIGCROP	Percentage of irrigated crop within circle of 2 km of radius
WETLAND	Percentage of wetland within circle of 2 km of radius
RIPHAB	Percentage of riparian habitat within circle of 2 km of radius
Microhabitat variables	
BARGRO	Percentage of bare ground on each transect
HERB	Percentage of herbaceous cover on each transect
SHRUB<50	Percentage of low shrub (< 50 cm) cover on each transect
SHRUB>50	Percentage of high shrub (> 50 cm) cover on each transect
TREE*	Percentage of tree cover of each transect

3.- Results

3.1.- Bird communities along the corridor

A total of 52 bird species were recorded along the Guadiamar Green Corridor in the 2006 breeding season, corresponding to 24 families, being *Sylvidae* and *Ardeidae* the best represented in species richness (Table 1). Species richness trends to be higher close the mine responsible for the tailing spill eight years before (r_s = -0.78, P=0.04), although we not found variation in abundance and ecological diversity along the corridor (r_s = -0.57, P = 0.18; r_s = -0.50, P = 0.25; respectively; Table 3).

Table 3.- Mean values (± SE) in species richness, abundance and diversity of the seven transects surveyed along the Guadiamar Green Corridor during 2006. Bird species diversity was computed using $-\sum p_i \ln p_i$.

Site	T1	T2	T3	T4	T5	T6	T7
Richness	27	21	21	28	19	17	16
Abundance	99.78 ± 14.92	148.70 ±12.63	98.27 ±2.76	124.9 ±9.14	136.6 ±15.59	71.24 ±4.26	62.66 ±13.99
Diversity	2.54 ±0.17	2.23 ±0.17	2.13 ±0.14	2.75 ±0.19	2.51 ±0.13	2.16 ±0.01	2.09 ±0.17

Most species recorded along the corridor were associated with open and riparian vegetation habitats, being *Serinus serinus* and *Carduelis carduelis* the more abundant. Riparian and generalist guilds showed a tendency to decrease toward the south, both in species richness and abundance (Table 1), but we not found any significant decrease or increase along the Guadiamar corridor in richness and abundance of all the two parameters in all guilds (r_s <0.19, p > 0.10). The guilds with lower species number in the corridor were those harboring species associated with shrubs, trees and water.

3.2.-Indirect gradient analysis: relationship between species composition and environmental variables

The mean value of the environmental variables and final variables included in the multivariate analyses appeared in Table 4. The DCA indicated a value of beta diversity for bird community below 3.0; hence, a linear ordination model such a Principal Components Analysis (PCA) was the method appropriate to summarize bird community variation (Braak & Smilauer 2002). The first three axes explained 76 % of the variability in species data (table 5) and three groups of variables implied in the model could be differentiated (table 6). The first axis included variables that were related with a vegetation change gradient and amount of nearby streams, greater presence of riparian vegetation, trees, shrubs; average length of streams appeared in the negative end of the axis, and absence of this vegetation types in the positive. The second ordination axis was correlated mainly with neighboring vegetation outside to the corridor, concretely with the presence of olive tree orchards; and the axis 3 was correlated with variables referring to irrigated land crops in the southernmost transects

Table 4.- Mean values (± SE) of environmental variables measured at macro- and microhabitat level, to describe the spatial context of the transects for bird census in the Guadiamar Green Corridor (SW Spain). * Variables excluded in the multivariate analysis due to multicollinearity

Variables	Value (mean ± SE)
Macrohabitat variables	
DISTMIN*	27310.57 ± 15019.77
MINREACH*	507.71 ± 315.62
DISTSTRE	583.14 ± 729.63
LENGSTRE	1996.71 ± 1683.41
OLIVCOV	12.98 ± 18.79
OAKCOV	9.77 ± 12.62
SCRUBFOR	12.82 ± 14.88
CULTGRAIN*	15.90 ± 25.81

PASTCOV	6.98 ± 6.59
FRUITCOV*	5.72 ± 10.36
IRRIGCROP	14.82 ± 27.57
WETCOV	17.87 ± 35.81
RIPHAB	2.57 ± 2.53
Micr habitat variables	
BARGRO*	7.29 ± 12.36
HERB	73.14 ± 24.48
SHRUB<50*	2.29 ± 3.25
SHRUB>50	5.86 ± 5.96
TREE*	11.43 ± 11.51

Table 5. Canonical axis values obtained in indirect and direct analysis between environmental variables and bird species abundance (performed with CANOCO) in the Guadiamar Green Corridor (SW Spain). Axis values greater than the average variability explained per axis, are noted in bold type.

Axes	1	2	3	4
Indirect analysis				
Eigenvalues	**0.384**	**0.197**	**0.179**	0.109
Cumulative percentage variance	38.4	58.1	**76.0**	86.9
Direct analiyysis				
Eigenvalues	**0.321**	**0.234**	0.149	0.121
Cumulative percentage variance	32.1	**55.5**	70.5	82.6

Table 6.- Intraset correlation of environmental variables with the three axes of PCA for bird species data in the Guadiamar Green Corridor (SW Spain).

Variables	**Axis 1**	**Axis 2**	**Axis 3**
LENGSTRE	-0.89	0.09	0.35
DISTSTRE	-0.81	0.29	0.35
OLIVCOV	-0.56	0.73	0.01
OAKCOV	-0.72	-0.24	0.21
PASTCOV	0.17	0.29	-0.04
CROPSCOV	0.08	-0.38	0.83
RIPHAB	-0.87	0.29	-0.30
TREESHRUB	-0.79	0.32	0.18
WETLAND	0.81	-0.11	-0.36

HERB	0.97	-0.08	0.08
SHRUB>50	-0.96	0.11	0.15
Eigenvalues	*0.384*	*0.197*	*0.179*
Cumulative percentage of variance explained	*38.40*	*58.10*	*76.00*

3.3.- Direct gradient analysis: detecting significant effect of each environmental variable to bird species data

In the DCCA, the length of the greater gradient axis indicated a value of beta diversity for bird species data below 3.0, thus, a linear ordination model was appropriate such a Redundancy Detrended Analysis (RDA) to relate species abundance and environmental variables. The forward selection procedure indicates ranking and importance of the environmental variables explaining bird species abundance. Eigenvalues for considered axes (1 and 2) explained 55.5 % of the bird abundance variability (Table 5). This analysis included five macrohabitat variables and one microhabitat variable, being percentage of herbaceous layer and riparian vegetation cover, the most important variables in the model (Table 7). Notwithstanding, the herbaceous layer cover was a variable acting opposite to most of other variables, and appearing in the negative extreme of biplots (Fig. 2).

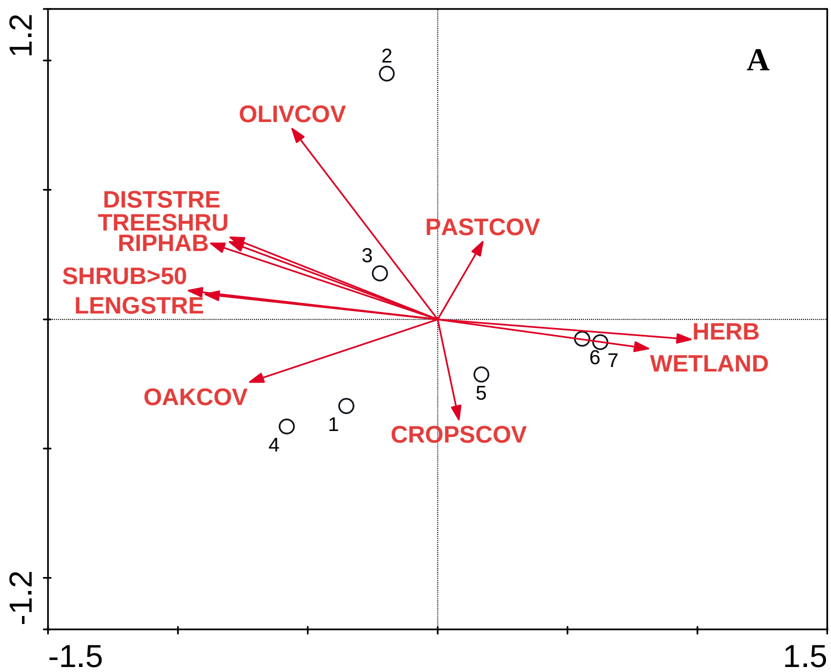
Table 7. Set of environmental variables selected by Stepwise Redundancy Analysis explaining bird abundance in the Guadiamar Green Corridor (SW Spain), according with the spatial scale of the variables considered. The *f*-values of restricted Monte Carlo test, and their signification (*P*) are shown.

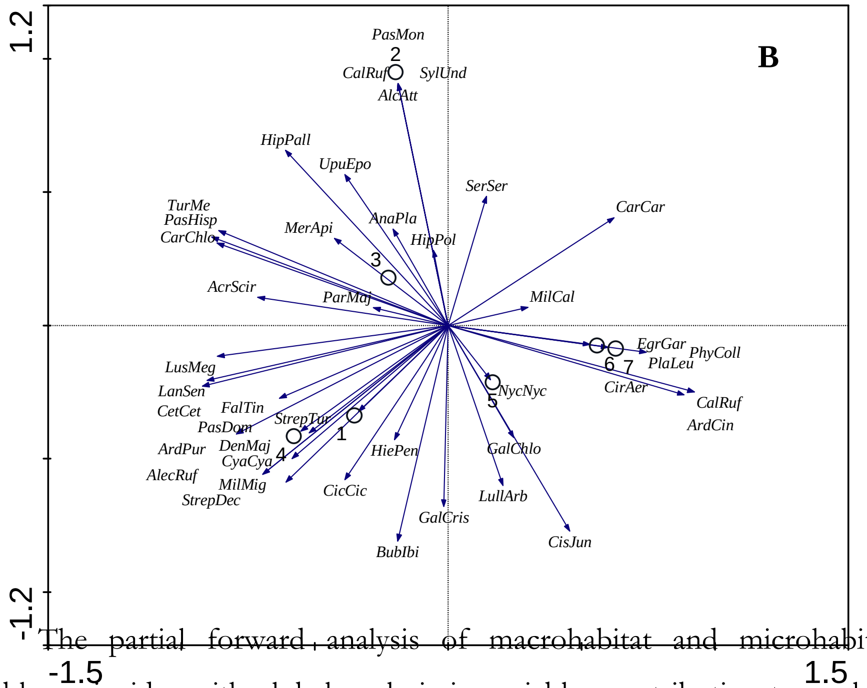
	f	*P*	*Explained variance (%)*
Macrohabitat variables	2.26	0.002	
LENGSTRE	2.0	0.002	
OLIVCOV	1.6	0.046	55.5

Microhabitat variables	2.26	0.002	
HERB	0.51	0.002	63.4
All variables			
HERB	2.26	0.002	
RIPHAB	1.32	0.002	
OLIVCOV	1.72	0.06	55.5

The biplots between transects and environmental variables, and transects and bird species, indicate the habitat differences between the first stretch of the corridor (transects 1-4) and the second (transects 5-7), which are shown in the negative and positive extremes of graphic, respectively (Fig. 2A). In this way, species which thrive in open habitats are present in the positive extreme and species with are related with shrub and/or trees in the negative extreme (Fig. 2B).

Figure 2.- Location of transect scores in the multivariable space defined by the Redundancy Detrended Analysis (RDA) in relation with environmental variables (A) and bird species (B) in the Guadiamar Green Corridor. Biplot B is performed with the subset of 50 % of the species best represented by the environmental variables.





The partial forward analysis of macrohabitat and microhabitat variables coincides with global analysis in variables contributing to explain variation in bird abundance, but revealed a new important variable in the first case, the average length of the surrounding streams.

4.- Discussion

Birds are good ecological indicators to asses status and trends over broad geographic regions (Canterbury et al., 2000). Eight years after the mine tailing accident and six years after the restoration project of the Guadiamar floodplain the breeding bird community responded very fast to the habitat restoration (unpub. data of the authors) and today, the corridor supports a diverse breeding bird community. However, will be necessary more years to detect a good structured community. Today there is a poor community in forest and shrubs species, where are abundant many generalist, open habitat and riparian guilds, in which we failed to detect any gradient related with distance to the Aznalcóllar mine. The species richness was higher closer to the mine and was lowest in the last zone. This can be due to the diversity in the land use that surround the corridor in the upper and middle stretch. These results are consistent with others findings as such as has been observed in others animal groups like aquatics macroinvertebrates, ants and beetles (Solà et al., 2004; Cárdenas & Hidalgo, 2006; Luque et al., 2007). Presently, the Guadiamar Green Corridor contrasts sharply with the surrounding area, a mosaic resulting from diverse human land uses along the second half of the past century (PICOVER, 2003). The multivariate analysis separated out two zones different in relation with and by environmental variables and consequently few differences in breeding bird community were observed among them. The heterogeneity of the first stretch (T1-T4), with a mosaic of natural vegetation and crops, allows a greater richness in generalist species and species related with riparian habitats; while the second (T5-T7), consisting in a homogeneous marshland landscape with grassy opening lands, harbor less bird species richness related with this open and aquatics habitats. But the multivariate analysis showed that the most of the structure of the breeding bird community can be explained by the existence of three different factors, the riparian habitat, and the presence in surrounding areas of olive groves and streams. Ours results indicated that the tasks of the managers of the corridor must preserve these heterogeneous landscapes as may enhance the connectivity of the matrix area in the Guadiamar Basin, estimating the contribution of surrounding orchards and streams in the maintenance of a rather diverse breeding bird. In previous studies in the Guadiamar Green Corridor the action proposal for enhancing the corridor´s functionality and the Guadiamar basin connectivity have highlighted the importance of the conservation of wooded land as olive grove and the small riparian formations linked to tributaries of the Guadiamar in the surroundings (Arenas, 2003).

The homogenization and agriculture intensification have negative effects in biodiversity and the heterogeneity within agricultural landscape are associated with higher in bird species richness (Benton et al., 2003; Devictor and Jiguet, 2007). In general, the important of remnant riparian habitat as buffer strip has been showed in several studies contributing to increase landscape diversity in impacted riparian zones. Hanowiski et al., (2007) and Smiley et al., (2007) have highlighted the woody vegetation and the buffer widths in the conservation of the riparian habitats functions. Matchtans et al,.(1996) showed that the buffer strips are important consideration to facilitate juveniles fluxes through the landscape in fragmentized landscape. Others studies have showed that in riparian ecosystems and agricultural landscape the configuration of the fragments and the habitat quality have important effects on bird community dynamic, and as a consequence, in bird species richness and abundance (Saab, 1999; Dauber et al., 2003; Miller et al., 2004; Mitchell et al., 2006). In the same way have found that floodplain land use, proximity of others waterbodies, anthropogenic structures, and the structural characteristic of the bank channel vegetation, are also determinants in the abundance or richness in bird communities (Strong and Bock,1990). Notwithstanding, others environmental variables have been important in bird community as channel characteristic itself as well as fluviogeomorphological features (as river width), water quality, and aquatic invertebrate abundance (Rushton et al., 1994; Iwata et al., 2003;Mason et al., 2006; Vaughan et al., 2007).

The conservation oriented to streams as corridors in the landscape of Guadiamar could provide a better conservation results in the connectivity in the Guadiamar Basin. Ours results have showed that birds community structure is sensitive to the vegetation structure along the Guadiamar Green Corridor and of the surroundings, and also to isolation to others refuges and feeding areas.

So, the fast actions performed in the Guadiamar Green Corridor, implying mechanical removal of the tailings and rehabilitation of the natural vegetation, provided a good example of ecosystem restoration after an ecological disaster, however we recommends that the management policies must consider the conservation of the heterogeneity of the surrounding habitats with wider corridors to preserve the biodiversity in the Guadiamar Green Corridor.

Acknowledgments

We appreciate the assistant Adela González-Megías for helping during analysis data. This study has been funded by the PICOVER and SECOVER programs, for 2004-2006 periods.

References

Aguilar, J., C. Dorronsoro, R. Beller, E. Fernández, J. Fernández, I. García, A. Iriarte, F. Martín, I. Ortiz, and M. Simón. 2003. Contaminación de suelos tras el vertido tóxico de Aznalcóllar. Universidad de Granada, Granada.

Bibby, J.C., N.D. Burgess, D.A. Hill, S. Mustoe. 2000. Bird census techniques. Academic Press, London.

Buffington, J.M., J.C. Kilgo, R.A. Sargent, K.V. Miller, B.R. Chapman. 2000. Effects of restoration techniques on breeding birds in a thermally-impacted bottomland hardwood forest. Ecological Engineering 15:115-120.

Cantebury, G.E., T.H. Martin, D.R. Petit, L.J. Petit, D.F. Bradfords. 2000. Bird community and habitat as ecological indicators of forest condition in regional monitoring. Conservation Biology 14:544-558.

Dauber J., Hirsh M., Simmering D., Waldhardt R., Otte A., Wolters V. 2003. landscape structure as a indicator of biodiversity: matrix effects on species richness. Agric. Ecosyst. Environ. 98:321-329.

Deschenes, M., L. Belanger, J. Giroux. 2003. Use of farmland riparian strips by declining and crop damaging birds. Agric. Ecosyst. Environ. 95:567-577.

Devictor V., Jiguet F. 2007. Community richness and stability in agricultural landscapes: the importance of the surrounding habitats. Agriculture, Ecosystems and Environment 120:179-184.

Dobkin, D.S., A.C. Rich, W.H Pyle. 1998. Habitat and avifaunal recovery from livestock grazing in a riparian meadow system of the northwestern great basin. Conservation Biology 12:209-221.

Gallart, F., G. Benito, J.P. Martín-Vide. 1999. Fluvial geomorphology and hydrology in the dispersal and fate of pyrite mud particles released by the Aznalcóllar mine tailing spill. The Science of Total Environment 242:13-26.

Gotelli, N.J., Entsminger, D.F. 2005. EcoSim: null model software for ecology. Version 7.72. Acquired intelligence Inc. & Kesey-Bear. http://homepages.together.net/~gentmin/ecosim.htm> (accessed in April 2007).

Gregory S.V., F.J. Swanson, W.A. McKee, K.W. Cummins. 1991. An ecosystems perspective of riparian zone. Bioscience, 41:540-551.

Grimalt, J.O., Ferrer, M., Macpherson, E., 1999. The mine tailing accident in Aznalcóllar. The Science of Total Environment 242:3-11.

Henningsen, J.C., Best, L. B., 2005. Grassland bird use of riparian filter strips in southeast Iowa. J. Wildl. Manage. 69:198-210.

Iwata T., Nakano S., Murakami M. 2003. Stream meanders increase insectivorous bid abundance in riparian deciduous forest. Ecography 26:325-337.

Keller, C.M.E., Robbins, C. S., Hafield, J. S., 1993. Avian communities in riparian forest of different widths in Mayrland and Delaware. Wetlands 13:137-144.

Knopf FL, and Samson FB. 1994. Scales perspective on avian diversity in western riparian ecosystems. Conservation Biology 8:669-679.

Lake P.S., Bond N., Reich P. 2007. Linking ecological theory with stream restoration. Freshwater Biology 52:597-615.

Leps J., Smilauer P., 2003. Multivariate analysis of ecological data using CANOCO. Cambridge University Press. UK. 269 pp.

Mason, C, Hofman TA., Macdonald SM. 2006. The winter bird community of river corridors in eastern England in relation to habitat variables. Ornis Fenica 83:73-85.

Machtans CS, Villard MA and Hannon SJ. 1996. Use of riparian buffer strips as a movement corridors by forest birds. Conservation Biology, 10: 1366-1379.

Mitchell M.S., Rutzomoser S.H., Wigley T.B., Loehle C., Gerwin J.A., Keyser P.D., Lancia R.A., Perry R.W., Reynolds C.J., Thill R. E., Weih R., White D., Wood P.B. 2006. Relationship between avian richness and landscape structure at multiple scales using multiple landscape. 221:155-169.

Miller, J.R., Dixon, M.D., Turner, M.G., 2004. Response of avian communities in large-river floodplains to environmental variation at multiple scales. Ecological Applications 14:1394-1410.

Naiman, R.J., Décamps, N., 1997. The ecology of interfaces: riparian zones. Annual Review of Ecology and Systematics 28, 621-658.

Naiman, R.J., Décamps, H and Pollock, M. 1993. The role of riparian corridors in maintaining regional biodiversity. Ecological Applications 3:209-212.

Pain, D.J., A. Sánchez, A.A. Meharg, 1998. The Doñana disaster: contamination of a world heritage estuarine marsh ecosystem with acidified pyrite mine waste. The Science of Total Environment 222: 45-54.

PICOVER, 2003. Ciencia y Restauración del Río Guadiamar. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla.

Rushton S.P., D. Hill, S.P. Carter. 1994. The abundance of river corridor birds in relation to their habitats- a modelling approach. Journal of applied ecology 31, 313-328.

Saab V., 1999. Importance of spatial scale to habitat use by breeding birds in riparian forest: a hierarchial analysis. Ecological Applications 9, 135-151.

Smiley Jr. P. C., Maul J.D., C. M. Cooper. Avian community structure among restored riparian habitats in northwestern Mississipi. Agriculture, Ecosystems and Environment 122: 149-156.

Solà C, M. Burgos, A. Plazuelo, J. Toja, M. Plans, N. Prat. 2004. Heavy metal bioaccumulation and macroinvertebrate community changes in a Mediterranean stream affected by acid mine drainage and an accidental spill (Guadiamar River, SW Spain). Science of the Total Environment, 333:109-126.

Sorace A., P. Colombari, E. Cordiner. 1999. Bird communities and extended biotic index (EBI) in some tributaries of the Tiber river. Aquatic conservation-marine and freshwater ecosystems 9:279-290.

Strong T. R., Bock C. E. 1990. Bird species distribution patterns in riparian habitats in southeastern Arizona. The Condor 92:866-885.

Ter Braak, C.J.F., Smilauer, P. 2002. CANOCO Reference Manual and CANODRAW for Windows User's Guide version 4.5. Microcomputer Power, Ithaca, NY:

Vaughan I.P., D.G. Noble, S.J. Ormerod 2007. Combinig surveys of rivers habitats and river bids to appraise riverine hydromorphology. Freswater biology 52:2270-2284.

Woinarski J.C.Z., Brock C., Armstrong M., Hempel C., Cheal D., Brennan. 2000. Bird distribution in riparian vegetation in the extensive natural landscape of Australia´s tropical savanna: a broad-scale survey and analysis of a distributional data base.

CAPÍTULO

Recovering the reptile community after the mine-tailing accident of Aznalcóllar (SW Spain)

Rocío Márquez-Ferrando, Juan M. Pleguezuelos, Xavier Santos,
Diego Ontiveros and J. R. Fernández-Cardenete

(*Restoration Ecology*)

Abstract

Ecosystem restoration requires that habitat requirements of all species be considered. Among animal communities in Mediterranean ecosystems, reptiles, as ectothermic vertebrates, need refuges for avoidance of extreme environmental temperatures, concealment from predators, and oviposition sites. In 1998 a massive amount of tailings broke out of the holding pond of the Aznalcóllar mine (SW Spain) and polluted the Guadiamar river valley. After the accident, a soil- and vegetation-restoration program began, and the Guadiamar Green Corridor was created to connect two huge natural areas, Doñana National Park and the Sierra Morena. Within this corridor the reptile community remained dramatically impoverished, probably because of elimination of all natural refuges during the soil-restoration program. To test this hypothesis, we set an array of artificial refuges (logs) in a large experimental plot. During the five years of the experiment (2002-2006) the area managed with artificial refuges exhibited a better and faster recovery of the reptile community in species richness and individual abundance than did the control area with no artificial refuges. Moreover, reptile colonization of the Guadiamar Green Corridor was transverse rather than lineal -- that is, it did not act as a corridor for reptiles, at least in the first stages of colonisation. This suggests that landscape-restoration programs should not neglect refuge availability, a limiting resource for reptile species.

Keywords: recovering animal communities, reptiles, artificial refuges, mine spill, Spain.

1.- Introduction

Restoration of degraded ecosystems is based on the assumption that rebuilding habitat will rehabilitate natural communities (Litt et al. 2001). Because of ecosystem complexity, however, even the most carefully designed habitat rebuilding programs rarely take the requirements of all natural communities into account. Programs for vegetation restoration usually do not meet requisites for faunal recovery, while those focused on mammal and bird communities do not satisfy requirements for ectothermic vertebrates (Buffington et al. 2000; Wike et al. 2000; Martín & López 2002; Nichols & Nichols 2003) and monitoring success of restoration programs with respect to different communities of organisms is essential (Palmer et al. 1997; White & Walker 1997; Block et al. 2001; Scott et al. 2001).

Reptiles play an important role in ecosystems, serving as prey, predators, and seed dispersers (Schoener & Spiller 1996; Jones 2002). They require natural cover that offers protection against visually oriented predators or microclimatic conditions favoring thermoregulation and reproduction (i.e. nesting sites) that differs from that required by birds and mammals (Huey et al. 1989). Scarcity of shelter negatively affects reptiles at individual, population and community levels (Goode et al. 2005) and how habitat management will affect reptile communities can be tested in degraded ecosystem restoration programs.

North of Doñana National Park, in southwestern Spain, a mine-tailing spill polluted a large area, giving rise to an interesting case study involving soil and vegetation restoration and recovery of the terrestrial reptile community in the contaminated area. On 25 April 1998, the wall of the holding pond for tailings of the Aznalcóllar mine collapsed and more than 6 million m^3 of toxic solids and acidic waters with high concentrations of arsenic and heavy metals (Zn, Pb, Cu, Sb, Tl, Cd) were released into the Guadiamar floodplain. Waste waters spread to a width of 1000 m in some areas along 62 km of the river (4286 ha in area) and included the northern zone of Doñana Natural and National Parks (a Biosphere Reserve; Pain et al. 1998; Gallart et al. 1999; Montes 2002).

From qualitative and quantitative standpoints this, the largest environmental pollution accident recorded in Spain (Grimalt et al. 1999), precipitated a process of compulsory purchase of all polluted areas and a program for ecological restoration of the affected area. The main points of this program were, i) mechanical mud withdrawal, ii) removal of original soil to a depth of 20 cm, iii) application of amendment materials (liming), iv) addition of a new layer of fertile soil, and v) rehabilitation of the natural vegetation (Simón et al. 1999; Antón–Pacheco et al. 2001; Aguilar et al. 2003). The site was fenced, traffic restricted, and the entire area protected under the aegis of the Guadiamar Green Corridor (PICOVER 2003).

Soon after habitat-restoration began, some species assemblages in natural communities of fluvial and terrestrial ecosystems (plants, ants, beetles, butterflies, fishes, reptiles, birds, mammals) began to be monitored and most increased in terms of species richness, abundance and/or ecological diversity (PICOVER 2003; Cárdenas & Hidalgo 2006). In 2000 when we studied the reptile community along the entire corridor both species richness and abundance results were quite poor (unpubl.data). Restoration programs for a natural community must identify habitat components most important to its total recovery (Caughley & Gunn 1995) but this was not achieved for the reptile community in the Guadiamar Green Corridor. Removing original soil to a depth of 20 cm eliminated all refuges available for reptiles; most reptiles known to have inhabited the study area were associated with shelters (Salvador 1998). The value of restored sites for reptiles should be improved by adding key habitats not likely to develop on restored sites under natural processes (Kanowski et al. 2006).

A corridor is a linear habitat connecting two or more large areas with ecological and landscaping value (Beier & Noss 1998). The Guadiamar Green Corridor emerged to connect Doñana National Park in the south and natural landscapes of Sierra Morena in the North. Riparian corridors are potential dispersal areas, mainly in agricultural landscapes, that diminish the effects of habitat fragmentation for vertebrates (Kjoss & Litvaitis 2001; Maisonneuve & Rioux 2001; Montori et al. 2001; Driscoll 2004). While corridors account for many of the ecological requirements of passage species (i.e. some birds and medium-sized mammals), this is not the case for most potential corridor dwellers (Burbrink et al. 1998). In this sense, many studies demonstrate the functionality of corridors for butterflies, small mammals and birds, but very few studies are oriented to organisms with limited home-ranges, such as reptiles (Beier & Noss 1998; Burbrink et al. 1998; Haddad et al. 2003; Huddens & Haddad 2003).

In 2002 we began an experiment focused on recovery of the reptile community within the Green Corridor that consisted of treatment with artificial refuges in a large area. Our objective was to assess effectiveness of using artificial refuges for restoring the reptile community in an area intensively managed to restore the landscape, but deprived of shelters, and to determine the colonization process of the corridor by the community of reptiles.

2.- Materials and methods

2.1. Study area

The Guadiamar River, a tributary of the Guadalquivir River, flows north to south in southwestern Spain (Fig. 1). The floodplain of this river has a long history of human alteration, and natural riparian vegetation contrasts with cultivation (mainly orchards; Jiménez et al. 2003). Within the upper and middle part of the floodplain there are also *dehesas*, an agro-ecosystem composed of scattered oaks, *Quercus rotundifolia* (Holm oak) and *Q. suber* (Cork oak), and there are marshlands in the lower corridor. During 1998-2000 the landscape within the corridor was managed to recover floral characteristics and complexity of natural habitat in the region and Mediterranean shrub species such as *Thymus vulgaris* (Thyme), *Lavandula stoechas* (Lavender), *Cistus* sp. (Rockrose), *Rosmarinus officinalis* (Rosemary), *Arbutus unedo* (Arbutus), and trees, such as *Olea europea* (Wild-olive tree), *Q. suber* (Cork oak), *Q. rotundifolia* (Holm oak) were planted.

The study area is characterized by mild winters (mean temperature of the coldest month, January, 7.1º C), hot summers (mean temperature of the hottest month, July, 26.9º C), and moderate rainfall (560 mm; data from 30 year standard meteorological averages from the Sanlúcar la Mayor weather station, representative of the study area).

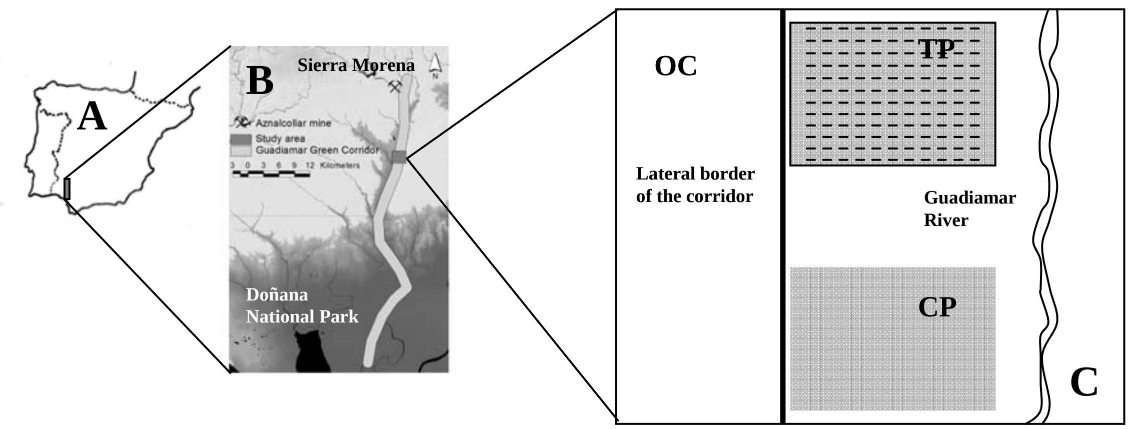


Figure 1.- Maps of A) the Iberian Peninsula showing location of the Guadiamar Green Corridor, B) the Green Corridor showing location of experimental plots, and C) the shape of the experimental plots.

2.2. Sampling sites

Since 2000 we have monitored reptile species richness and abundance. After two years of very poor results (see Results, below) we started a reptile-habitat recovery program in 2002 that involved setting up a large array of artificial refuges. The study site within the corridor was selected because, i) it was surrounded by almost natural habitat (*dehesa*) with well-conserved bush cover, to provide an immediate source area for reptile recolonization, ii) it was located in a wide section of the corridor (at least 500 m in width, to test for possible lateral colonization), and iii) it was the site where the vegetation-restoration program had begun earlier. We selected two large (24 ha) and close together (400 m apart) plots within the corridor and provided one, the treatment plot (TP), with 120 artificial refuges; the control plot (CP) was not provided with artificial refuges (Fig. 1). Plots shared the same external habitat (*dehesa*) as source areas for reptile recolonization. In addition, samples were taken from outside the corridor (OC; within a belt 500 m from the corridor and unaffected by the spill) for recording some traits of the reptile community in the surrounding source area. In the TP, refuges were distributed in a squared mesh array, approximately 40 m cell size (depending on site vegetation). The array contained 12 lines of refuges directed parallel to the main axis of the corridor, with 10 refuges in each line. To avoid random effects of temperature on refuges used by reptiles, all refuges were placed in sun-exposed conditions. The study area and surroundings are in a Plio-Pleistocene alluvial sedimentary basin (Borja et al. 2001) in which rocks as natural refuges are almost absent and most natural refuges available to reptiles are logs. We used timber for artificial refuges because it was the most abundant refuge in the region and other materials (metal, stones) become too hot when exposed to the summer sun in Mediterranean ecosystems and could not be integrated into the natural habitat (Webb & Shine 2000). Wood offers good insolation from extreme summer temperatures, maintains moisture, integrates into the landscape, degrades with time, is not expensive, and does not need to be removed after the experiment (Grant et al. 1992). Each artificial refuge consisted of two logs (approximately 1.2 m long and 0.2 m in diameter) placed side by side.

2.3. Reptile census

Reptile censuses began two years after the mine accident and immediately after the vegetation-restoration program. Censuses were time-constrained and relative densities of reptiles were analyzed in terms of encounter rates (number of species and number of individuals per hour per person; Crump & Scott 1994) and ontogenetic stage of individuals (newborn, juvenile, adult). Surveys were consistently performed by two observers to facilitate the moving of refuges in some cases, particularly in TP. Because of low diversity in the reptile community and rather clear structure of the vegetation species identification was easy and we assumed no bias among observers in reptile sampling. Plots within the corridor and zone OC were surveyed a minimum three times/year (spring and autumn) starting two hours after sunrise for about four/five hours per sampling (approximately 60 sampling hours in each of the three areas) for the period 2002-2006. Every individual record, either below or outside a refuge, was geo-referenced with a 12 channel GPS device offering ±4 m error. Records within the TP were plotted on a digital map generated by the Regional Administration (1:10,000) with ARCVIEW 3.2 software, facilitating calculation of the distance of individuals from the plot's external borders.

Differences in reptiles among the three sites were analyzed in terms of species richness and number of individuals per hour. Non-parametric analyses were performed because some data violated assumptions of normality; sampling plots were considered the treatment and data for one observer hour as cases.

The TP was designed almost square shaped and large enough (500 m x 480 m) to detect possible gradients in species richness and abundance. At beginning stages in the colonization process, i) the highest number of records (either for species or individuals) in the northern border of the plot (Fig. 1) should suggest that reptile colonization occurred linearly through the corridor, ii) the highest number of records in the external border of the plot, that adjacent to the side of the corridor (Fig. 1), should suggest that reptile colonization of the corridor was transverse instead of linear, and iii) the lack of any gradient suggests that colonization occurred from all directions. There were no barriers for terrestrial reptiles in habitats closest to TP, either outside or inside the corridor, and connectivity between habitats of both lineal and transverse colonization directions were apparently equivalent. The period four years after the refuge treatment (2005) was chosen to analyze direction of the colonization process because it was then that the reptile community within the corridor began to be significant, both in species richness and abundance (see Results). To test each prediction we correlated number of species and individuals along both possible spatial gradients within the plot with distance to the border of the plot. Both gradients were divided into sectors 25 m long and Spearman Rank Correlation (r) was used.

2.4. Habitat structure

Habitat structure has been shown to be a key trait to explain species composition in reptile communities (Pianka 1966; Enge & Marion 1986). To test for eventual differences in habitat between TP and CP some measurements were conducted in both plots. In each experimental plot we designed a square mesh, 50 m on a side, and measured habitat structure at each mesh intersection. We marked a sub-plot of 5 m radius by placing four 5 m tapes in the four directions of the compass (N, S, E, W). Height data were taken using 3 m rods graduated into three sections (0-0.5 m, 0.5-2.0 m, > 2.0 m) and placed vertically at every meter. We analyzed two variables, ground cover (bare ground, herbaceous, shrubs and trees) and vertical complexity (number of contacts with vegetation for each section of the rod). Data were collected during June 2006 at the peak of the annual growth period. Ground cover and vertical complexity of the vegetation were compared by the Mann-Whitney U test.

3.- Results

Historical data reported 13 species of reptiles inhabiting areas surrounding the Guadiamar Green Corridor (Pleguezuelos et al. 2002; J.P. González de la Vega 2006, AHE, Spain, personal communication; Table 1). Only six of these species have to date been detected within the TP (three lizards and three snakes) and only five species in the equal-sized, almost adjacent CP (Table 1). Data for the last three years of field searching (2004-2006) indicated species richness was greater in OC than CP (T = 3.12, p = 0.001, *d.f.* = 91), but revealed no difference between OC and TP (T = 0.57, p = 0.56, *d.f.* = 92). Abundance was higher in OC than in CP (T = 2.44, p = 0.01, *d.f.* = 91), but lower between OC and TP (T = -3.48, p << 0.001, *d.f.* = 92). These data suggest reptile recolonization of the corridor is far from complete in terms of species richness in the CP seven years after the vegetation-restoration program, and four years after the reptile-specific treatment program in the TP.

Within the corridor, we found no differences between TP and CP for herb (T = - 1.67, p = 0.09, *d.f.* = 23), shrub (T = 0.29, p = 0.77, *d.f.* = 23) and tree (T= -0.09, p = 0.93, *d.f.* = 23) density or for vertical complexity of vegetation (T= -0.23, p = 0.82, *d.f.* = 23). The plots differed only marginally in the percentage of bare ground, higher in TP (T = 2.14, p = 0.03, *d.f.* = 23) (raw habitat structure data are available upon request from corresponding author). Therefore, habitat complexity was rather similar between plots.

The average number of species and number of individuals per unit sampling effort were higher in TP than in CP (T= 2.78, p = 0.005, *d.f.* = 94; T = 5.48; $p < 0.001$, *d.f.* = 94, respectively) during the last three years of field searching (2004 -2006). This difference was maintained when comparing number of individuals per unit sampling effort independently for the two species with significant sample sizes in each plot, *Psammodromus algirus* (large psammodromus; T = 5.35, $p < 0.001$, *d.f.* = 94), and *Hemorrhois hippocrepis* (horseshoe whip snake; T = 2.44, p = 0.01; *d.f.* = 94). During the seven years of the study (2000-2006), species richness and abundance increased in both plots (TP r = 0.88, $p< 0.05$, CP r = 0.96, $p<0.05$; TP r = 0.90, $p < 0.05$, CP r = 0.91, $p < 0.05$, respectively), but abundance increased more abruptly in TP than in CP (Fig. 2). High scores for correlations suggested that colonization of the reptile community was driven by time.

In the TP, newborn individuals for five of the six species (all except *Natrix maura* [Viperine snake]) were found, and no newborn specimens were found in the CP.

There were negative and significant relationships between distance to the lateral border of the corridor and the average number of species and number of individuals per unit sampling effort (r = -0.72, p <0.01; r = -0.84, p < 0.01 respectively; Fig. 3). There was no gradient in species richness or number of individuals (r = 0.03, p = 0.92; r = -0.16, p = 0. 49 respectively) along the main axis of the corridor.

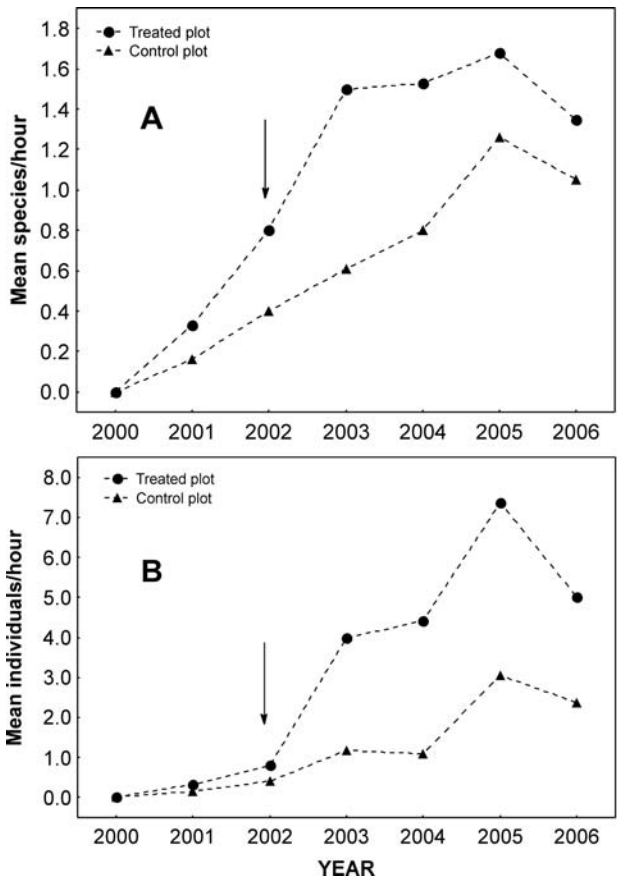


Figure 2.- Evolution of the reptile community of the Guadiamar Green Corridor (southwestern Spain) in the experimental plots (Treated [TP] and Control [CP]) during 2000-2006: A) species richness (mean number of species per unit sampling effort), and B) abundance (mean number of individuals per unit sampling effort). Arrows represent the year in which treatment with refuges began.

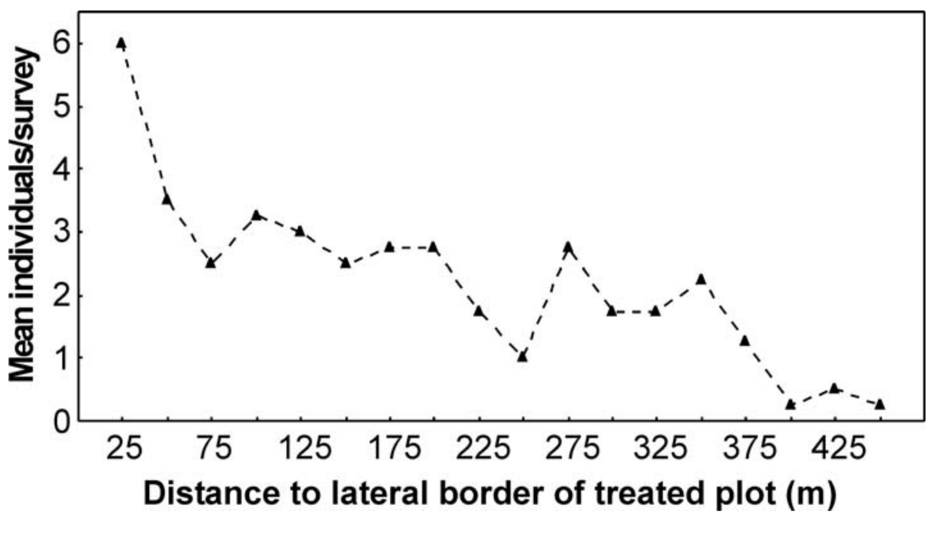


Figure 3. - Mean number of reptiles per unit sampling effort in a gradient defined by the distance from the external border of the Guadiamar Green Corridor (southwestern Spain) to the inner part of the corridor (Guadiamar River area). Data correspond to the experimental plot three years after the treatment with refuges (year 2005). For more details, see the Methods section.

Reptile species	2000		2001		2002		2003		2004		2005		2006		2004-2006		
	TP	CP	TP	CP	TP	CP	TP	CP	TP	CP	TP	CP	TP	CP	TP	CP	OC
Blanus cinereus																	
Tarentola mauritanica			0.33	0.1	0.20		0.25			0.20	0.18	0.05			0.06	0.08	1.00
Acanthodactylus erythrurus																	0.11
Timon lepidus									0.13		0.04	0.10	0.29	0.11	0.15	0.07	0.09
Podarcis hispanica																	0.28
Psammodromus algirus					0.60	0.40	3.50		3.87	0.90	6.64	2.79	4.53	2.22	5.01	1.97	1.58
Coronella girondica																	
Hemorrhois hippocrepis									0.13		0.18	0.05	0.12		0.14	0.01	
Macroprotodon brevis																	
Malpolon monspessulanus							0.25		0.20		0.32	0.05	0.06	0.05	0.19	0.03	0.20
Natrix maura									0.06						0.02		
Natrix natrix																	
Rhinechis scalaris																	0.02

4.- Discussion

For restoration of damaged habitats to be successful the basic requirements of target communities to be recovered must be considered (Palmer et al. 1997; Jones 2002). In the Guadiamar Green Corridor, plans for restoration of natural vegetation following the mine accident implied large-scale landscape management but failed to meet some basic requirements for re-establishment of the reptile fauna. Our data demonstrate that one unfulfilled need for better and faster recovery of the reptile community was the presence of shelters. The addition of shelter sites to degraded or strongly-managed habitats helped improve the reptile community through the advantage that refuges offer to adults, to their main prey, or to their clutches (Zappalorti & Reinert 1994; Castilla & Swallow 1995; Webb & Shine 2000) and probably contributed to making enemy-free space (Cole et al. 2005). After treatment with artificial refuges, the reptile community increased from one species (*Tarentola mauritanica*, Moorish gecko) during 2000-2001 to six species in 2006, and abundance increased from fewer than one to more than five individuals per unit sampling effort. These increments could simply be the consequence of natural colonization of the restored area over the seven years of the study period. Significant differences in species richness and overall abundance between treated and untreated plots, however, suggest that recovery of the reptile community in TP was due to the addition of artificial refuges. Similarity in vegetation structure between the two experimental plots eliminates the possibility that differences in the reptile community were an artifact of habitat complexity in such plots. The only habitat structure trait for which we found a marginal difference was the importance of bare ground, which was higher in the TP. This possible difference might reinforce our hypothesis as the main reptile colonizer in the study area, *P. algirus*, selected high canopy cover of the vegetation, thus rejecting bare ground (Carretero et al. 2002). In general, providing habitats with artificial refuges as part of landscape management proved very effective for other reptile communities, particularly in studies made in Australasia. Nichols & Nichols (2003), for example, reported that establishment of ground shelters in the form of logs and rocks, and varying vegetation density to include requirements of diverse reptile species, would promote faunal return. Webb & Shine (2000) added artificial resources (shelter sites) in degraded habitats in the successful recovery of the endangered *Holplocephalus hungaroides* (Broad-headed snake) and its main prey *Oedura lesveurii* (Velvet gecko). Souter et al. (2004) added burrows to enhance a population of the endangered *Tiliqua adelaidensis* (Pygmy blue-tongue lizard).

Artificial refuges can improve reptile communities by providing shelter to adults and juveniles (Reading 1997; Bowers et al. 2000; Webb & Shine 2000), and by providing egg-laying sites (Castilla & Swallow 1995). In reptiles, eggs may be the most vulnerable stage (Shine 1988), and availability of appropriate areas in which to lay eggs is therefore fundamental for success in reptile community recovery in a degraded area (Shine & Fitzgerald 1989). During censuses, the difference in the occurrence of newborns between plots confirmed that reptile colonization was a success in the TP, as most colonizing species were breeding.

Some reptile species that, according to the literature, apparently occupied the corridor before the disaster were not found within the limits of the corridor after seven years of monitoring. These species can be grouped as fossorial species such as *Blanus cinereus* (Amphisbaenian) that require deep and well-structured soils, not yet available in the corridor; rupicolous species such as *Podarcis hispanica* (Iberian wall lizard) for which vertical and hard surfaces are not yet present; and six species having very patchy distributions in the region (Table 1). Appearance of these species within corridor limits seems to require a period of more than seven years after the beginning of habitat restoration. For these we do not recommend translocation programs, as we suspect the managed area still does not meet their habitat requirements. Moreover, there are few examples of successful translocation programs involving reptiles (Dodd & Seigel 1991; Kanowski et al. 2006). In a similar study in north-western Spain, more than 10 years were required for the herpetofaunal community to attain pre-disturbance characteristics (Galán 1996) and, in general, reptiles are slow to colonize newly planted areas (Kanowski et al., 2006). The species currently in the corridor are among the most abundant (*T. mauritanica, N. maura*), generalist *(Timon lepidus, H. hippocrepis, Malpolon monspessulanus*), and small sized (*P. algirus*) in the region's reptile community (see review in Salvador 1998). Biological traits of these species matched well with the paradigm of ecological successions, which states that the first steps of any colonization process are performed by opportunistic, small-sized and r-strategist species (Odum & Barrett 2005). The restored area within the corridor was more effective for such species as lizards (*P. algirus* in our case study) that have fast-growing populations (Huddens & Haddad 2003). In other studies, lizards have also been found to be pioneer colonizers in restored habitats. For instance, *Podarcis bocagei* (Bocage's wall lizard) was the first colonizer in a restored mine in north-western Spain (Galán 1996).

With respect to the use of the Guadiamar Green Corridor by the community of reptiles, the two core areas for the corridor are the Sierra Morena in the north and Doñana National Park in the south. We should expect colonization by the reptiles of the newly-generated habitats of the corridor to be in the northern-southern axis, in either direction. Our results, however, suggest that colonization by reptiles in the corridor was transverse rather than linear, that is, the Green Corridor did not act as a corridor for the reptile community, at least not during the first years following landscape restoration. Our results also suggest that, in a colonization process involving terrestrial reptiles, proximity to a source area (the immediate habitat to TP, outside the corridor) appears to be a more critical factor than size of the source area (the huge surface of the northern protected areas, Sierra Morena; see also Burbrink et al. 1998). Studying factors affecting the use of restored sites by reptiles in rainforest landscapes of Australia, Kanowski et al. (2006) also found that restoration sites where more reptile species appeared were those more spatially linked to natural source areas. This is an expected result for organisms like lizards and snakes, those with very small home ranges and limited dispersal capacity, particularly when the intervening habitat between source and target areas is unsuitable (Stow et al. 2001; Kanowski et al. 2006).

Implications for Practice

- After eight years of the mine accident, seven years of vegetation restoration, and four years of reptile-specific habitat restoration (addition of artificial shelters), the colonizing community of reptiles in the Guadiamar Green Corridor did not achieve pre-disturbance richness; only opportunistic species were able to recolonise the experimental plot.
- Restoration of highly-modified habitat for terrestrial reptiles in Mediterranean ecosystems may require deliberate creation of particular components of the habitat (such as refuges) that would be slow or unlikely to develop on re-vegetated sites under natural processes.

- Extensive use of inexpensive artificial refuges (such as logs) over large areas (the Guadiamar Green Corridor, 60 km long) should favor faster and more natural recovery of the reptile community.
- Species not recovered in the restored area here considered were those most specialized in microhabitat (burrowing, rupicolous, litter-dwelling). For these species we do not recommend translocation programs, as we suspect the managed area still does not meet their habitat requirements.

Acknowledgments

Esmeralda Alaminos, Jesús Caro, Asier Herrero and Gregorio Moreno-Rueda, collaborated in field sampling. G. Moreno-Rueda thoroughly revised the manuscript and Stephen D. Busack edited and improved the manuscript. This study was funded by the PICOVER (2000-2002) and SECOVER (2004-2006) programs of the Guadiamar Corridor Agency, Consejería de Medio Ambiente, Junta de Andalucía.

References

Aguilar, J., C. Dorronsoro, R. Beller, E. Fernández, J. Fernández, I. García, A. Iriarte, F. Martín, I. Ortiz, M. Simón. 2003. Contaminación de suelos tras el vertido tóxico de Aznalcóllar. Universidad de Granada, Granada.

Antón-Pacheco, C., J.C. Arranz, D. Barettino, G. Carrero, M. Jiménez, J.A. Gómez, J.C. Gumiel, E. López-Pamo, J.A. Martín Rubí, B. Martínez Pledel, E. de Miguel, J. Moreno, G. Ortiz, J.G. Rejas, A. Silgado, E. Vázquez. 2001. Actuaciones para el reconocimiento y retirada de los lodos depositados sobre el terreno, y su restauración edáfica y morfológica. Boletín Geológico y Minero 112:93–121.

Beier, P., R. F. Noss.1998. Do habitat corridors really provide connectivity?. Conservation Biology 12:1241-1252.

Block, W. M., A. B. Franklin, J. P. Ward, Jr. J.L. Ganey, G. C. White. 2001. Design implementation of monitoring studies to evaluate the success of ecological restoration on wildlife. Restoration Ecology 9:293-303.

Borja, F., J. A. López Geta, M. Martín Machuca, R. Mantecón, C. Mediavilla, P. del Olmo, R. Palancar, R. Vives. 2001. Marco geográfico, geológico e hidrológico regional de la cuenca del Guadiamar. Boletín Geológico y Minero 112:3-34.

Bowers, C. F., H. G. Hanlin, D. C. Guynn Jr, J. P. Mclendon, J. R. Davis. 2000. Herpetofaunal and vegetation of a thermally-impacted stream at the beginning of restoration. Ecological Engineering 15:101-114.

Buffington, J. M., J. C. Kilgo, R. A. Sargent, K. V. Miller, and B. R. Chapman. 2000. Effects of restoration techniques on breeding birds in a thermally-impacted bottomland hardwood forest. Ecological Engineering 15:115-120.

Burbrink, F. T., C. A. Philips, E. J. Heske. 1998. A riparian zone in southern Illinois as a potential dispersal corridor for reptiles and amphibians. Biological Conservation 86:107-115.

Cárdenas, A. M., J. M. Hidalgo. 2006. Ecological impact assessment of the Aznalcóllar mine toxic spill on edaphic coleopteran communities in the Guadiamar River basin (Southern Iberian Peninsula). Biological Conservation 15:361-383.

Carretero, M. A., X. Santos, A. Montori, G. Llorente. 2002. *Psammodromus algirus* (Fitzinger, 1826). Pages 260-262 in J. M. Pleguezuelos, R.

Márquez and M. Lizana, editors. Atlas y Libro Rojo de los anfibios y reptiles de España. DGCN-AHE, Madrid.

Castilla, A. M., J. G. Swallow. 1995. Artificial egg-laying sites for lizards: a conservation strategy. Biological Conservation 72:387-391.

Caughley, G., A. Gunn. 1995. Conservation biology in theory and practice. Blackwell Science, Massachusetts.

Cole, N. C., C. G. Jones, S. Harris. 2005. The need for enemy-free space: the impact of an invasive gecko on island endemics. Biological Conservation 125:467- 474.

Crump, M. L., N. J. Scott Jr. 1994. Visual encounter surveys. Pages 84-92 in W. R. Heyer, M.A. Donnelly, R.W. McDiarmid, L.C. Hayek and M.S. Foster, editors. Measuring and monitoring biological diversity: standard methods for amphibians. Smithsonian Institution Press, Washington.

Dodd, C. K. Jr, R. A. Seigel. 1991. Relocations, repatriation and trans-location of amphibians and reptiles: are they conservation strategies that work?. Herpetologica 47:336-350.

Driscoll, D. A. 2004. Extinction and outbreaks accompany fragmentation of a reptile community. Ecological Applications 14:220-240.

Enge, K. M., and W. R. Marion. 1986. Effects of clearcutting and site preparation on herpetofauna of a north Florida flatwoods. Forest Ecology and Management 14:177-192.

Galán, P. 1996. Colonization of spoil benches of an opencast lignite mine in northwest Spain by amphibians and reptiles. Biological Conservation 79:187-195.

Gallart, F., G. Benito, J. P. Martín-Vide, A. Benito, J. M. Prió, and D. Regües. 1999. Fluvial geomorphology and hydrology in the dispersal and fate of pyrite mud particles released by the Aznalcóllar mine tailing spill. The Science of Total Environment 242:13-26.

Goode, M. J., W. C. Horrace, M. J. Sredl, and J. M. Howland. 2005. Habitat destruction by collectors associated with decreased abundance of rock-dwelling lizards. Biological Conservation 125:47-54.

Grant, B. W., A. D. Tucker, J. E. Lovich, A. M. Mills, P. M. Dixon, J. W. Gibbons. 1992. The use of coverboards in estimating patterns of reptile and amphibian biodiversity. Pages 379-403 in D. R. McCullough and R. H. Barrett, editors. Wildlife 2001: Populations. Elsevier Science Publ., Inc. London, England.

Grimalt, J. O., M. Ferrer, E. Macpherson. 1999. The mine tailing accident in Aznalcóllar. The Science of Total Environment 242:3-11.

Haddad, N. M., D. R. Browne, A. Cunningham, B. J. Danielson, D. J. Levey, S. Sargent, T. Spira. 2003. Corridor use by diverse taxa. Ecology 84:609-615.

Huddens, B. R., N. M. Haddad. 2003. Predicting which species will benefice from corridors in fragmented landscapes from population growth models. The American Naturalist 161:808-820.

Huey, R.B., C.R. Peterson, S.J. Arnold, W.P. Porter. 1989. Hot rocks and not-so-hot rocks: retreat-site selection by garter snakes and its thermal consequences. Ecology 70:931-944.

Jiménez, Y., A. Paniza, J. Gómez. 2003. Análisis de los paisajes del Guadiamar y de su evolución reciente como fundamento del diagnóstico territorial de su cuenca. Pages 482-492. Ciencia y restauración del río Guadiamar. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla.

Jones, C. G. 2002. Reptiles and amphibians. Pages 355-375 in M. R. Perrow and A. J. Davy, editors. Handbook of ecological restoration. Vol.1: Principles of restoration. Cambridge University Press, Cambridge.

Kanowski, J. J., T. M. Reis, C. P. Catterall, S. D. Piper. 2006. Factors affecting the use of reforested sites by reptiles in cleared rainforest landscapes in tropical and subtropical Australia. 2006. Restoration Ecology 14:67-76.

Kjoss, V. A., J. A. Litvaitis. 2001. Community structure of snakes in a human-dominated landscape. Biological Conservation 98:285-292.

Litt, A. R., L. Provencher, G. W. Tanner, and R. Franz. 2001. Herpetofaunal responses to restoration treatments of longleaf pine sandhills in Florida. Restoration Ecology 9:462-474.

Maisonneuve, C., and S. Rioux. 2001. Importance of riparian habitat for small mammal and herpetofaunal communities in agricultural landscapes of southern Québec. Agriculture, Ecosystems and Environment 83:165-175.

Martín, J., and P. López. 2002. The effect of mediterranean dehesa management on lizard distribution and conservation. Biological Conservation 108:213-219.

Montes, C. 2002. Lecciones aprendidas en tres años de restauración de ecosistemas en el Corredor Verde del Guadiamar. Ecosistemas 11:1-9.

Montori, A., G. A. Llorente, M. A. Carretero, and X. Santos. 2001. La gestión forestal con la herpetofauna. Pages 251-289 in J. Camprodoms and E. Plana, editors. Conservación de la biodiversidad y gestión forestal. Su aplicación en la fauna vertebrada. Universitatd de Barcelona, Barcelona.

Nichols, O. G, and F. M. Nichols. 2003. Long-term trends in faunal recolonization after bauxite mining in the Jarrah forest of southwestern Australia. Restoration Ecology 1:261-272.

Odum, E. P., and G. W. Barrett. 2005. Fundamentals of Ecology. Thomson Brooks, California.

Pain, D. J., A. Sánchez, and A. A. Meharg. 1998. The Doñana disaster: Contamination of a world heritage estuarine marsh ecosystem with acidified pyrite mine waste. The Science of Total Environment 222:45-54.

Palmer, M. A., R. F. Ambrose, and N. L. Poff. 1997. Ecological theory and community restoration ecology. Restoration Ecology 5:291-300.

Pianka, E.R. 1966. Convexity, desert lizards, and spatial heterogeneity. Ecology 47:1055-1059.

PICOVER. 2003. Ciencia y Restauración del Río Guadiamar. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla.

Pleguezuelos, J. M., R. Márquez, and M. Lizana. 2002. Atlas y Libro Rojo de los anfibios y reptiles de España. DGCN-AHE, Madrid.

Reading, C. J. 1997. A proposed standard method for surveying reptiles on dry lowland heath. Journal of Applied Ecology 34:1057-1069.

Salvador, A. (Coord.).1998. Reptiles. Vol. 10. in M. A. Ramos, editor. Reptiles. MNCN, CSIC. Madrid.

Schoener, T.W., and D. A. Spiller. 1996. Devastation of prey diversity by experimental introduced predators in the field. Nature 381:691-694.

Scott, T. A., W. Wehtje, and M. Wehtje. 2001. The need for strategic planning in passive restoration of wildlife populations. Restoration Ecology 9:262-271.

Shine, R. 1988. Parental care in reptiles. Pages 275-330 in C. Gans and R. B. Huey, editors. Biology of the Reptilia, Vol. 16, Ecology B, Alan R. Liss Inc., New York.

Shine, R., and M. Fitzgerald. 1989. Conservation and reproduction of an endangered species: the broad-headed snake, *Hoplocephalus bungaroides* (Elapidae). Australian Zoologist 25:65-67.

Simón, M., I. Ortiz, I. Garcıa, E. Fernández, J. Fernández, C. Dorronsoro, and J. Aguilar. 1999. Pollution of soils by the toxic spill of a pyrite mine (Aznalcóllar, Spain). The Science of the Total Environment 242:105-115.

Souter, N.J., C.M. Bull, and M.N. Hutchinson. 2004. Adding burrows to enhance a population of the endangered pygmy blue tongue lizard, *Tiliqua adelaidensis*. Biological Conservation 111:403-408.

Stow, A.J., P. Sunnucks, D.A. Briscoe, and M.G. Gardner. 2001. The impact of habitat fragmentation on dispersal of Cunningham's skink (*Egernia cunninghami*). Evidence from allelic and genotypic analyses of microsatellites. Molecular Ecology 10:867-878.

Webb, J. K., and R. Shine. 2000. Paving the way for habitat restoration: can artificial rocks restore degraded habitats of endangered reptiles?. Biological Conservation 92:93-99.

White, P. S., and J. L. Walker. 1997. Approximating nature's variation: selecting and using reference information in restoration ecology. Restoration Ecology 5:338-349.

Wike, L., M. Douglas, H. Hanlin, and L. Paddock. 2000. Small mammal populations in restored stream corridor. Ecological Engineering 15:121-129.

Zappalorti, R. T., and H. K. Reinert. 1994. Artificial refugia as a habitat-improvement strategy for snake conservation. Pages 369-375 in J. B. Murphy, K. Alder and J. T. Collins, editors. Captive management and conservation of amphibians and reptiles. SSAR, London.

CAPÍTULO 4

Bioaccumulation of heavy metals in the lizard *Psammodromus algirus* after a tailing-dam collapse in Aznalcóllar (SW Spain)

Rocío Márquez-Ferrando, Xavier Santos, Juan M. Pleguezuelos and Diego Ontiveros

(*Submitted to Archives of Environmental Contamination and Toxicology*)

Abstract

Quantification of heavy metal concentrations in biota is a common technique that helps environmental managers measuring level of pollutants circulating in ecosystems. Despite interest in heavy metals as indicators of localized pollution, few studies have assessed these pollutants in reptiles. In 1998, the tailing pond of a pyrite mine near Aznalcóllar (SW of Spain), containing 6 million m^3 of mud with high heavy metal concentrations, collapsed releasing toxic sludge into the Guadiamar Basin. Here we analyze heavy metal concentrations in the most common reptile of the area, the large psammodromus, *Psammodromus algirus*, a rather small sized lizard. We quantified levels of several elements (Hg, Sb, Cd, Cr, Tl, Sn, Ba, Cu, Pb, Sr, Mn, Rb, As and Zn) in lizard tail clips collected in and around the affected area during the springs of 2005 and 2006. Samples were collected from two contaminated localities, one directly affected by the spill, and another adjacent to the tailing pond, but not covered by toxic mud. We also collected samples from a non-polluted control site in the same basin. We found higher concentrations of As, Tl, Sn, Pb, Cd and Cu in lizards from the affected area than in lizards from the control site, indicating the continued presence of heavy metal pollutants in the terrestrial food chain eight years after the mine accident. We did not uncover sexual or annual differences in heavy metal concentrations, although concentrations increased with lizard size. We discuss how heavy metals moved across the food chain to lizards despite intensive restoration efforts after the accident, and suggest reptiles to be included in biomonitoring programs of heavy metals pollution in terrestrial habitats.

Keywords: *Psammodromus algirus*; heavy metals; Aznalcóllar mine; Guadiamar River; bioaccumulation.

1. Introduction

Heavy metals contamination associated with mining activities has caused environmental problems in several countries (Hsu *et al,* 2006), hence, environmental managers are particularly interested in developing methods to detect heavy metals loads in biota (Lambert *et al,* 1996; Loumbourdis, 1997; Meharg *et al,* 1999; Burger *et al,* 2006). The events occurred in the Guadiamar Basin, in the southwestern Iberian Peninsula (Spain), provides resource managers with a model case-study for biomonitoring heavy metals in the ecosystems. On April 1998, the wall of a large pond containing sulfide ore deposits collapsed, spilling more than 6 million m^3 of acidic water and toxic sludge directly into the Agrio and Guadiamar rivers (Gallart *et al,* 1999; Grimalt *et al,* 1999; Dorronsoro *et al,* 2002). The main toxic metals spilled were Pb, Zn, As, Cu and Cd (Alastuey *et al,* 1999; Cabrera *et al,* 1999). Tailing materials reached an area 40 km long and 0.5 km wide in the Guadiamar basin, with sludge covering the ground in a layer 0.3-3.0 m thick, depending to the distance from the collapsed dam. Environmental managers first (year 1998) attempted to clean-up the pyrite slurry by mechanically removing the mud and the first cm of the underlying soil (Simón *et al,* 1999) However, characteristics of the Guadiamar Basin, including low-profile topography and a mosaic pattern of contamination, complicated soil recovery. Thus, a second clean-up activity was undertaken (year 1999), adding different amendments to the soil to immobilize pollutants (Querol *et al,* 2006; Aguilar *et al,* 2007). These actions helped decrease contaminants in upper soil layers, however, pollutant levels increased greatly in deeper horizons of the soil, enhancing the risk of groundwater contamination (Kraus and Wiegand, 2006; Ordóñez *et al,* 2006). After these clean-up operations in the spill-affected area, a reforestation program was initiated (year 2000) as part of an effort to designate the area as a natural space, the Guadiamar Green Corridor.

Between the years 1999-2006, several research teams monitored the accumulation of heavy metals in soils, running waters and organisms, including aquatic macro-invertebrates, fishes, amphibians, mammals, reptiles, birds, shrubs and trees, to examine the transfer of heavy metals through the food chain and to assess environmental managing tasks (Benito *et al,* 1999; Cabrera et al.,1999; PICOVER, 2003; Madejón *et al,* 2004; Solá *et al,* 2004; Alcorlo *et al,* 2006; Olías *et al,* 2006; Taggart *et al,* 2006). In the polluted area, only one reptile species, rather marginal in the ecosystem because its tree dwelling habits, the Moorish gecko (*Tarentola mauritanica*), has been included in analyses of heavy metal bioaccumulation (Fletcher *et al,* 2006). Indeed, ecotoxicological studies on reptiles have been scarce, a trend that has only recently begun to change (Avery et al., 1983; Hopkins *et al,* 2000; Linder and Grillitsch, 2000; Campbell and Campbell, 2002; Mann *et al,* 2006). However, terrestrial reptiles are good bioindicators of high metals concentrations, because they occupy intermediate or high levels in food chains, frequently are generalist diet, and have low vagility (Loumbordis, 1997; Campbell and Campbell, 2002; Burger *et al,* 2004).

During the years 2000-2006, we monitored recolonization of the restored Guadiamar Green Corridor by the reptile community. The first colonizer, and most abundant reptile, was the large psammodromus, *Psammodromus algirus* (Márquez-Ferrando *et al,* 2008). This small and opportunistic lizard is a generalist feeder of arthropods, exhibits fast-growing populations, has a short lifespan (mean lifespan, two years) and low vagility (Diaz and Carrascal, 1990; Carretero and Llorente, 1997; Salvador, 1998). These biological traits make this species a suitable model for monitoring localized bioaccumulation of heavy metals in contaminated Mediterranean terrestrial habitats.

The objectives of this study are to: i) assess the quantity of heavy metals accumulated by *P. algirus* in the Guadiamar Green Corridor, seven-eight years after the mine spill; ii) determine sexual, size-related and inter-annual differences in heavy metal accumulation; iii) detect simultaneous accumulation patterns of different metals; and iv) contribute to the biomonitoring program of the Guadiamar Green Corridor.

2- Materials and methods

2.1. Study site

The Guadiamar River is situated in the southwest of the Iberian Peninsula (Fig. 1). Within the Guadiamar Basin, several studies have detected heavy metal accumulation in organisms from sites directly affected by the Aznalcóllar mine spill (Solá *et al,* 2004; Alcorco *et al,* 2006) and in nearby sites that were not covered by the toxic mud, but were impacted by atmospheric pollution (Madejón *et al,* 2006). Thus, we collected *P. algirus* from three localities differently impacted by the mine spill. Two study sites are located within the Guadiamar floodplain (Las Doblas bridge [Site A] and the Agrio-Guadiamar confluence [Site B]). The third site is located just outside the floodplain (Villamanrrique Pinewood [Site C]). Site A, situated in the middle of the Green Corridor and 11 km downstream from the mine (Fig. 1), was severely affected by the spill and restored following the procedures described above. Site B was not covered by toxic mud, although it is located very close to the affected area (0.1 km). Site C, an unpolluted control site, is located 25 km downstream of the mine and 2 km outside of the Guadiamar floodplain.

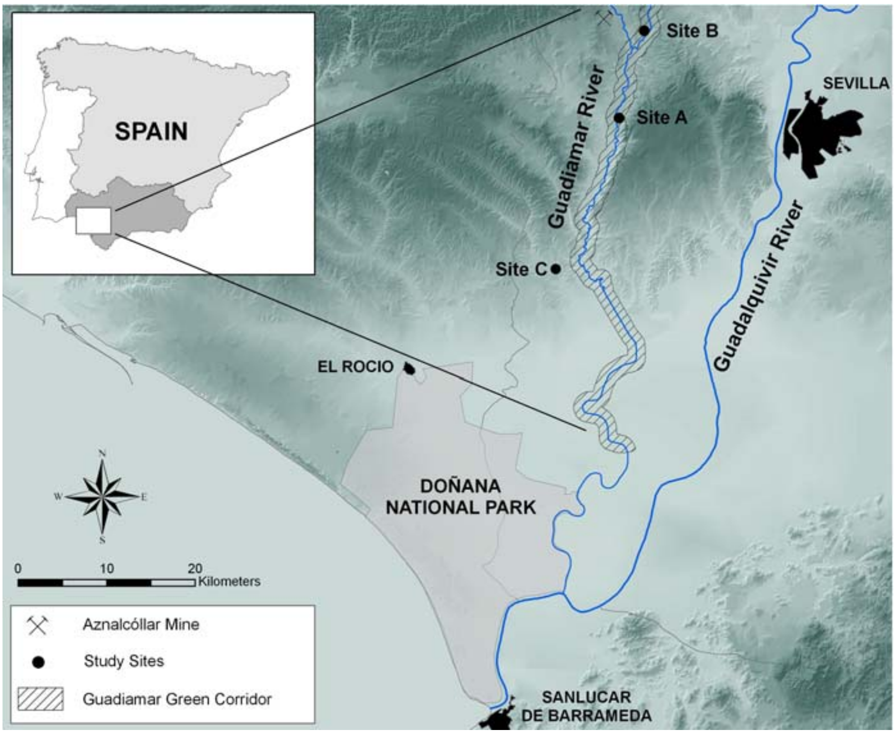


Figure 1.- Study area. Maps showing the location of the Guadiamar Green Corridor in the Iberian Peninsula and the locations of the three study sites within the Guadiamar Basin.

2. 2. Lizard handling

We collected lizards by hand during May-June of 2005 and 2006. Thirty lizards from site A, 15 from site B and 20 from site C, were captured. We measured (snout-vent length [SVL], to the nearest mm) and sexed lizards (by color pattern and morphology of the femoral pores) to check for the effect of size and sex in metal accumulation, as has been reported in other reptile species (Linder and Grillitsch, 2000). Non-lethal measurements can be used to assess pollutant levels in squamate reptiles (Hopkins *et al*, 2001; Burger *et al*, 2005; Fletcher *et al*, 2006), and the use of such methods here is advisory to reduce man-induced alterations, as the Guadiamar Green Corridor is currently a protected area. Thus, we collected a tail clip (< 30 mm) from each individual to assess levels of heavy metals. Each lizard was later released at its capture site. We assumed that the probability of recapture in the second sampling year in the polluted site was minimal due to the high population density (Márquez-Ferrando *et al*, 2008); furthermore, during the 2006 sampling period, we did not capture individuals with regenerated tail.

2. 3. Laboratory procedures

We analyzed concentrations of 15 metals (Hg, Sb, Cd, Cr, Tl, Sn, Ba, Cu, Pb, Sr, Mn, Rb, As and Zn). Of these, Pb, Zn, As, Cu and Cd were abundant in the toxic mud (Alastuey *et al*, 1999), whereas the other metals, although less abundant in the mud, have been previously analyzed in several organisms of the Guadiamar Basin, and hence contribute to a wider overview of heavy metal mobilization along the food chain (Solá *et al*, 2004; Fletcher *et al*, 2006). After collection, tail clips were cleaned with deionized water and freeze-dried. Samples were oven dried at 60°C until they attained constant weights, and digested for 8 hours with 2 ml HNO_3 and 1 ml H_2O_2 in Teflon vessels. Samples were brought to a final volume with deionized water. Metal concentrations were measured through mass spectroscopy (Perkin-Elmer ELAN-6000) by Scientific-Technical Services at the University of Barcelona. We included ten blanks in digestion and analysis procedures as controls. Results of element levels are expressed in $\mu g\ g^{-1}$ on a dry weight basis. For lizards collected in the control population we found concentrations below the detection limits for several metals. In these cases, we used one-half the detection limits as surrogate values for non-detects (Hesel, 1990).

2. 4. Statistical analyses

We used non-parametric tests when data did not fit normality after log-transformation, and compared heavy metal concentrations among the three localities with a two-way ANOVA rank-transformed dependent variables, considering sex, location and their interaction, as factors. Differences between pairs of localities were tested using Tukey post-hoc tests. We checked the relationship between body size and heavy metal levels in lizards from site A using Spearman rank correlations. This analysis was also used to investigate patterns of accumulation in pairs of heavy metals. Scores of the correlation matrix were used to create a cluster tree of similarities among metals according to Euclidean distances. Thus, metals were organized as functions of the linkage distances between them, using single linkage as aggregation algorithm. In all the tests, significant differences were assumed at $p < 0.05$.

3. Results

3. 1. Differences among localities

We failed to detect difference in lizard size among localities (mean SVL = 66.6±1.6 mm [site A], 67.0±2.1 mm [site B], 64.0±2.1 mm [site C]; ANOVA test, $F_{2,48} = 0.69$, $p = 0.51$). We found differences among localities in all metal concentrations (Table 1). Results of Two-way ANOVA showed differences between populations ($F_{2,48} = 7.36$, $p < 0.01$), between sexes ($F_{1,24} = 2.64$, $p = 0.02$) and in the interaction sex and location ($F_{2,48} = 1.93$, $p = 0.02$) for overall heavy metal concentration. Tukey post-hoc test indicated a similar pattern in 10 of the 14 elements: lizards collected at site A showed higher levels than did lizards from the other two localities. Lizards from site A showed 21-, 8- 7- 5- and 4-fold greater concentrations of As, Tl, Sn, Pb, Cd and Cu, respectively, when compared to lizards from site C. We also detected differences between lizards from site B and lizards from site C in Tl, As, Hg, Rb, Cu, Pb, and Zn. In each case, lizards from site B had greater metal concentrations than did lizards from site C.

Table 1.- Means and standard errors of metal concentrations (μg g^{-1} dry weight) found in tails of the lizard *Psammodromus algirus* collected at three locations in the Guadiamar Basin (Southwestern Spain). Comparisons were tested by two-way ANOVA with rank-transformed data. Site A, Guadiamar River floodplain, fully affected by the spill; Site B, Guadiamar River floodplain, very near to the affected area; Site C, Villamanrrique pinewood, 2 km away from the affected area.

	Site A (n = 30)	Site B (n = 15)	Site C (n = 20)	*F* (P)	
Hg	0.17 ± 0.02	0.24 ± 0.03	0.09 ± 0.01	12.73(<0.01)	B and A > C
Sb	0.38 ± 0.06	0.08 ± 0.02	0.17 ± 0.14	19.47 (<0.01)	A > C> B
Cd	0.14 ± 0.02	0.05 ± 0.02	0.03 ± 0.02	10.85 (< 0.01)	A and B > C
Cr	2.05 ± 0.16	1.43 ± 0.08	1.41 ± 0.07	1.85 (0.17)	-
Tl	0.08 ± 0.01	0.05 ± 0.02	0.00 ± 0.00	88.52 (<0.01)	A > B > C
Sn	0.20 ± 0.06	0.02 ± 0.00	0.03 ± 0.01	4.09 (0.02)	A > C
Ba	2.88 ± 0.33	1.76 ± 0.38	4.38 ± 0.50	6.45 (<0.01)	C > B
Cu	10.54 ± 1.55	4.25 ± 0.38	3.03 ± 0.17	24.29 (<0.01)	A > B and C
Pb	10.18 ± 1.94	2.60 ± 0.69	2.17 ± 1.00	10.82 (<0.01)	A > B > C
Sr	10.56 ± 0.89	6.30 ± 1.36	8.86 ± 0.88	1.35 (0.27)	-
Mn	13.71 ± 1.90	6.72 ± 0.98	8.32 ± 1.01	1.88 (0.17)	-
Rb	7.60 ± 0.81	6.96 ± 0.52	3.38 ± 0.25	29.64 (<0.01)	A and B> C
As	6.26 ± 1.05	1.20 ± 0.23	0.30 ± 0.03	62.72 (<0.01)	A > B > C
Zn	106.76 ± 4.20	86.92 ± 5.61	85.08 ± 8.45	1.91 (0.16)	A > B > C

3. 2. Sexual, inter-annual and size-related differences

There was no significant sexual difference in body-size among individuals collected at site A (mean SVL, males 68.5±7.8 mm; females 64.5±4.5 mm; ANOVA test $F_{1,11}$= 0.92, p = 0.36). Sexes did not differ in heavy metal levels except for Cr (Table 2). For this reason, we did not separate sexes in further analyses. Likewise, there was no difference in body size between individuals collected in 2005 and 2006 at site A (mean SVL; year 2005, 66.7±9.0 mm; year 2006, 66.3±1.9 mm; ANOVA test $F_{1,20} = 0.01$, $p = 0.91$). We did not find significant differences in any metal concentrations, except for Hg and Sn, between lizards collected in 2005 and 2006 at site A. Mercury level was higher in lizards collected during 2006 (Table 3), and Sn levels were lower in 2006. The relationship between metal concentration and lizard size approached statistical significance for several metals, but was only significant for Cd (Fig. 2).

Table 2.- Means and standard errors of metal concentrations (μg g^{-1} dry weight) in tail clips of male and female *Psammodromus algirus* collected in the most affected area by the Azanalcóllar mine spill (Site A, see Materials and Methods section) of the Guadiamar Basin (Southwestern Spain). Differences between sexes were tested by one-way ANOVA with rank-transformed data.

	Male (n=9)	Female (n=5)	F (P)
Hg	0.16 ± 0.02	0.20 ± 0.05	0.82 (0.38)
Sb	0.47 ± 0.13	0.34 ± 0.10	0.45 (0.51)
Cd	0.19 ± 0.05	0.06 ± 0.03	2.88 (0.12)
Cr	2.32 ± 0.35	1.37 ± 0.08	5.81 (0.03)
Tl	0.09 ± 0.02	0.09 ± 0.04	0.13 (0.73)
Sn	0.24 ± 0.13	0.08 ± 0.04	0.62 (0.45)
Ba	3.06 ± 0.58	2.13 ± 0.54	0.63 (0.44)
Cu	14.17 ± 4.17	5.92 ± 2.34	2.25 (0.16)
Pb	12.59 ± 3.12	6.18 ± 1.06	1.77 (0.21)
Sr	10.53 ± 1.77	7.99 ± 1.05	1.61 (0.23)
Mn	12.76 ± 1.99	8.13 ± 1.60	4.58 (0.05)
Rb	7.10 ± 1.04	10.89 ± 2.78	2.73 (0.13)
As	8.19 ± 2.23	5.23 ± 1.99	0.42 (0.53)
Zn	102.77 ± 47.46	95.36 ± 11.92	0.68 (0.43)

Table 3.- Means and standard errors of metal concentrations (μg g^{-1} dry weight) in tail clips of the lizard *Psammodromus. algirus* collected in 2005 and 2006 in the most affected area by the Aznalcóllar mine spill (Site A, see Materials and Methods section) of the Guadiamar Basin (Southwestern Spain). Differences between years were tested by one-way ANOVA with rank-transformed data.

	2005 (n=20)	2006 (n=10)	F (P)
Hg	0.12 ± 0.01	0.28 ± 0.03	29.95 (<0.01)

Sb	0.43 ± 0.08	0.29 ± 0.09	1.32 (0.26)
Cd	0.14 ± 0.03	0.14 ± 0.03	0.55 (0.46)
Cr	2.20 ± 0.21	1.74 ± 0.21	3.35 (0.08)
Tl	0.08 ± 0.02	0.07 ± 0.02	0.27 (0.61)
Sn	0.28 ± 0.09	0.04 ± 0.01	11.98 (<0.01)
Ba	2.82 ± 0.39	3.01 ± 0.62	0.03 (0.86)
Cu	9.74 ± 2.28	11.05 ± 3.78	0.07 (0.93)
Pb	11.21 ± 2.17	9.20 ± 1.77	0.15 (0.69)
Sr	11.82 ± 1.14	8.04 ± 1.05	3.94 (0.06)
Mn	12.09 ± 1.42	16.96 ± 4.96	0.55 (0.46)
Rb	6.55 ± 0.75	9.70 ± 1.77	3.94 (0.06)
As	6.76 ± 1.35	5.25 ± 1.67	0.37 (0.55)
Zn	109.34 ± 4.94	101.60 ± 7.91	0.93 (0.34)

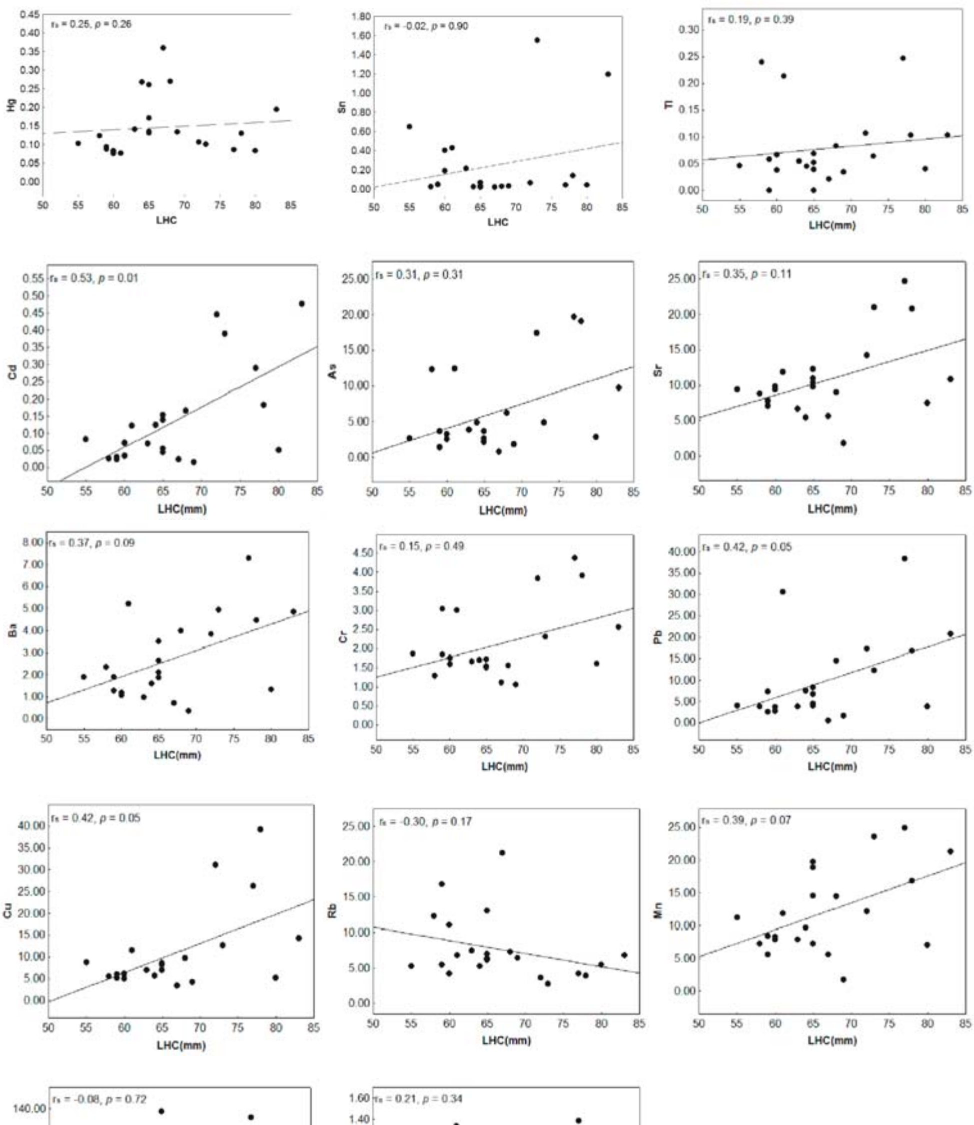


Figure 2.- Size-related differences in metal concentrations. Spearman correlations between metal concentrations (µg g^{-1} dry weight) in tail clips of *Psammodromus algirus* (n=22) and lizard size (SVL), in the most affected area by the Azanalcóllar mine spill (Site A, see Materials and Methods section) of the Guadiamar Basin (Southwestern Spain).

3. 3. Correlations among metal concentrations

At site A, metal concentrations were positively correlated in 41 out of 98 pairs of elements (Table 4), indicating that many elements shared similar accumulation trends (Fig. 3). The group of Sb, As, and Tl were all strongly associated with Spearman coefficients higher than 0.8. A second group was composed of Cd, Ba, Mn, Cu, and Pb with correlation coefficients greater 0.7. The pair formed by Hg and Rb was positively correlated with a value of 0.5 and exhibited sharp differences with respect to other elements. Finally, a group consisting of Sr, Cr, Zn and Sn exhibited very low correlations with the rest of metals.

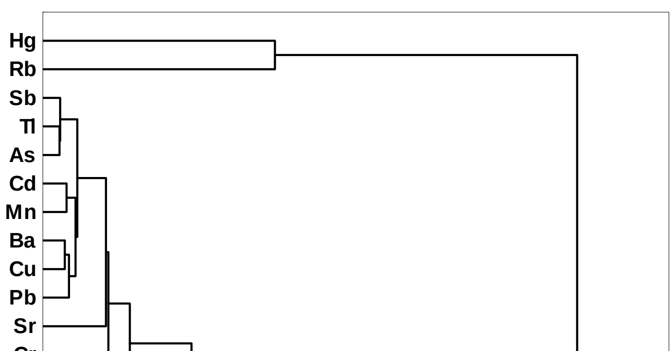


Figure 3.- Correlations among metal concentrations. Cluster tree that show the linkage (Euclidean distance) between metal concentrations by single linkage as aggregation algorithm in tail clips of the lizard *Psammodrolus algirus* in the most affected area by the Azanalcóllar mine spill (Site A, see Materials and Methods section) of the Guadiamar Basin (Southwestern Spain).

Table 4.- (next page) Spearman correlations among metal concentrations in tail clips of the lizard *Psammodromus algirus* (n=30) collected in the most affected area by the mine spill of Aznalcóllar (Site A, see Materials and Methods section) of the Guadiamar Basin (Southwestern Spain). After sequential Bonferroni correction for multiple test the p value was adjusted for 5% of the nominal level, p<0.001.

Ba	**Sn**	**Tl**	**Cr**	**Cd**	**Sb**	**Hg**
0.11 (0.55)	**-0.38** (0.04)	-0.02 (0.91)	-0.26 (0.16)	0.34 (0.06)	-0.09 (0.06)	1.00
0.73 (<0.001)	0.32 (0.09)	**0.84** (<0.001)	**0.68** (<0.001)	**0.60** (<0.001)	1.00	
0.74 (<0.001)	0.26 (0.16)	**0.59** (<0.001)	**0.59** (<0.001)	1.00		
0.62 (<0.001)	**0.45** (0.01)	**0.53** (0.003)	1.00			
0.68 (<0.001)	0.25 (0.18)	1.00				
0.26 (0.17)	1.00					
1.00						

	Zn	As	Rb	Mn	Sr	Cu	Pb
Hg	-0.04 (0.64)	0.06 (0.85)	0.34 (0.76)	0.29 (0.06)	-0.19 (0.12)	0.14 (0.31)	0.19 (0.46)
Sb	0.29 (0.12)	**0.89** (<0.001)	0.02 (0.91)	**0.59** (<0.001)	**0.39** (0.03)	**0.65** (<0.001)	**0.82** (<0.001)
Cd	0.35 (0.06)	**0.71** (<0.001)	-0.26 (0.165)	**0.84** (<0.001)	**0.50** (0.005)	**0.81** (<0.001)	**0.85** (<0.001)
Cr	**0.56** (<0.001)	**0.63** (<0.001)	-0.35 (0.06)	**0.60** (<0.001)	**0.50** (0.005)	**0.69** (<0.001)	**0.71** (<0.001)
Tl	0.27 (0.15)	**0.88** (<0.001)	0.01 (0.96)	**0.48** (0.008)	**0.39** (0.03)	**0.62** (<0.001)	**0.76** (<0.001)
Sn	0.32 (0.08)	0.25 (0.18)	-0.32 (0.08)	0.26 (0.17)	0.31 (0.09)	**0.39** (0.04)	0.34 (0.07)
Ba	**0.45** (0.01)	**0.73** (<0.001)	-0.22 (0.25)	**0.84** (<0.001)	**0.68** (<0.001)	**0.84** (<0.001)	**0.86** (<0.001)
Pb	**0.41** (0.02)	**0.82** (<0.001)	-0.11 (0.57)	**0.81** (<0.001)	**0.45** (0.01)	**0.85** (<0.001)	1.00
Cu	**0.61** (<0.001)	**0.65** (<0.001)	-0.29 (0.12)	**0.83** (<0.001)	**0.67** (<0.001)	1.00	
Sr	**0.60** (<0.001)	**0.40** (0.027)	**-0.58** (<0.001)	**0.58** (<0.001)	1.00		
Mn	**0.42** (0.02)	**0.58** (<0.001)	-0.16 (0.41)	1.00			
Rb	-0.25 (0.17)	-0.18 (0.35)	1.00				
As	0.23 (0.22)	1.00					
Zn	1.00						

4. Discussion

4. 1. Differences among localities

This study describes heavy metal contamination of terrestrial organisms from the Guadiamar Green Corridor following the collapse of the Aznalcóllar mine tailing pond. Lizards from the most impacted site exhibited higher metal concentrations eight years after the spill than did both lizards collected at a control site and lizards from a nearby locality not covered by toxic mud. Water quality in the Guadiamar River increased from 2002 onwards (Olías *et al,* 2006), and consequentially, species richness and the community structure of freshwater organisms improved (Toja *et al,* 2003). In contrast, and despite extensive cleaning actions, the soils of the Doblas (site A) still exhibited high concentrations of heavy metals four years after the spill because they persist in deeper horizons (Kraus and Wiegand, 2006). These contaminants are taken up by plants through their roots and, depending on the mobility of the chemicals, are moved to vegetative parts (in general to leaves) of the plants (Madejón *et al,* 2004). Leaves, as well as other plant parts, may be eaten by insects that are reptile prey. Several studies have showed that the principal avenue of pollutant acquisition by reptiles is through the ingestion of polluted prey (Hopkins *et al,* 2001, 2002; Fletcher *et al,* 2006; Mann *et al,* 2006). *Psammodromus algirus* is a generalist forager upon epigeous invertebrates, being Colepotera, Heteroptera and Araneae the main prey for populations of this lizard from Southwestern Spain (Pérez Quintero and Rubio García, 1997) which live in the upper levels of soils and can be exposed to several metals, such as Cd and Pb, in polluted areas (Jelaska *et al,* 2007). Furthermore, a complementary avenue for acquisition of contaminants in lizards is the accidental ingestion of sediment particles with food (Fletcher *et al,* 2006; Mann *et al,* 2006), very important in *P. algirus* due to its foraging habits (Carretero and Llorente, 1993).

Arsenic was the toxic element that showed the highest difference between individuals dwelling in contaminated and control sites. This metal was very important in the toxic mud (Alastuey *et al,* 1999) and its monitoring in the food chain is potentially critical because its neurotoxic effects upon a variety of organism (Chang 1996), as well as its negative effects on the embryonic development in the Iberian rock lizard (*Lacerta monticola cyrenni*; Marco *et al*, 2004).

We also detected high significant differences in thallium among populations. Tl persists for long periods in terrestrial ecosystems and is still detectable at high levels in the contaminated areas, indicating wide dispersal through the terrestrial food-chain (Madejón *et al,* 2004; Sánchez-Chardi, 2007), although the exact mechanism of toxicity is still unclear (Jon Peter and Viraraghavan, 2005).

Among the metals analyzed, Zn showed the highest concentrations in lizard tails, reinforcing the pattern previously observed in *T. mauritanica* (Fletcher *et al,* 2006). Concentrations of Zn may remain higher than other metals because Zn can bind to specific metallothioneins in reptiles, interfering with the organism's detoxification processes (Linder and Grillitsch, 2000; Lance *et al,* 2005). Other metals, such as Mn and Cr, were very scarce in the sludge and in the surrounding area (Cabrera *et al,* 1999; Simón *et al,* 1999), although we detected accumulations in lizards (Table 1). These elements are lithophilic, and therefore, despite occurring in low levels, became increasingly available to plants as a consequence of soil acidification following the spill (Madejón *et al,* 2006).

Although site B was not covered by the toxic sludge, lizard tails from this site showed intermediate metal concentrations (Table 1). Madejón *et al.* (2006) suggested that metals may spread from contamination sites via atmospheric transport and deposition of contaminants over surrounding areas. This process may have been exacerbated by the removing of affected soils during clean-up activities in the 1998-1999 (Querol *et al,* 2000). Consequently, deposition of aerosolized elements may explain the contamination of lizards in areas surrounding the affected site. Surprisingly, lizards from site B showed higher levels of Hg than lizards from site A. Mercury was present in low levels within the sludge (Alaustey *et al,* 1999; Cabrera *et al,* 1999), and was of great concern in aquatic ecosystems (Pain *et al,* 1998; Sanpera *et al,* 2000), but information concerning impacts and transport of this metal in terrestrial food chains is scarce.

Our results were similar to those reported in whole-body samples of *T. mauritanica* from the same study area (Fletcher *et al,* 2006). In this gecko, As, Tl, Cd and Pb levels were also higher among individuals at contaminated sites than in control geckos. However, tail samples from *P. algirus* had higher concentrations of As, Tl, Mn, Sb, Pb and Cu than did whole-body samples of *T. mauritanica*. Both studies suggest that reptile species are useful indicators of heavy metal levels in the Guadiamar Green Corridor.

4. 2. Sexual, annual and lizard size comparisons

In some reptiles, sex can influence metal accumulation due to differences in physiology, size and diet (Linder and Grillitsch, 2000; Van Straalen *et al,* 2001; Hopkins, 2002; Burger *et al,* 2004; Hopkins *et al,* 2005; Jelaska *et al,* 2007). In site A, *P. algirus* did not exhibit sexual size dimorphism, in contrast to other Iberian populations (Mellado and Martínez, 1974; Carretero and Kaliontzopoulou, 2000), however this can be because in our study there is a low number of males and females compared Furthermore, this lizard did not display sexual in diet and microhabitat use (Díaz and Carrascal, 1990; Salvador, 1998; Carretero *et al,* 2002). Accordingly, it is not surprising that we did not detect sexual differences in metal concentrations.

Likewise, we did not detect any differences between samples collected in 2005 and 2006, even though several authors have found that metal levels in water, soil and plants, declined over time in the Agrio and Guadiamar river floodplains (Madejón *et al,* 2006; Olías *et al,* 2006; Querol *et al,* 2006). It is likely that a longer term study would be needed to detect temporal changes in accumulation of metals by lizards.

Cadmium was the only metal that increased significantly with lizard size in *P. algirus*. Cadmium accumulation seems to be dependent on the duration of metal exposure (Mann *et al*, 2007). Correlations between metal concentrations and lizard size have been documented for Cd and Pb in *Podarcis muralis* and *Anolis sagrei*, and for Pb and Cu in *Zootoca vivipara* (Schidt, 1980, 1988; Burger *et al*, 2004). In general, larger individuals can accumulate higher amounts of pollutants, although considerable differences exist among species based, at least in part, on species longevity (Santos *et al*, 1999; Linder and Grillitsch, 2000). *Psammodromus algirus* is a short-lived species and all individuals we captured were likely between one and two years old. Thus, the lack of correlation in the most of heavy metals with lizard size is not surprising, since the correlation between size and longevity in adults is very weak in *P. algirus* (Carretero, com. pers.).

4. 3. Relationships among elements

In general the presence of simultaneous heavy metals within an ecosystem may favour the existence of interactions between them (Beyersman, 1991). In our study large, positive correlations among eight elements suggest that these metals were accumulated simultaneously. A similar pattern occurred in *T. mauritanica* from the Guadiamar River (Fletcher *et al*, 2006). Low correlations between Sr, Cr, Zn and Sn and the rest of the heavy metals suggests that these elements may be acquired or uptaken differently that are the first group of metals, although whole body analyses of metal concentrations might yield different results and are necessary to rinse the pattern of correlation of heavy metal accumulated by reptiles.

5. Conclusions

Over the last years several studies have focused on bioaccumulation of heavy metals by wildlife in the Guadiamar Green Corridor following the release of mining by-products in this area. Our study adds to the knowledge of this issue in terrestrial animals, and demonstrates that even eight years after a toxic spill and subsequent clean-up activities, the terrestrial food chain still demonstrates exposure to high levels of heavy metals. Lizards from contaminated sites showed significantly greater concentrations of several metals than did lizards from non-contaminated sites. Because of their biological traits (low vagility, middle and upper position in the trophic food chain, generalist in diet, rapid population turnover, short lifespan), we suggest that small reptiles such as *P. algirus* are good bioindicators of local heavy metal contamination (see also Fletcher *et al,* 2006, for the same study area). Long-term studies of trace-element accumulation in aquatic and terrestrial biota are necessary to understand how pollutants move across food chains, and to assess the continued impact of the mining spill on Guadiamar Green Corridor wildlife.

Acknowledgments

We thank A. Herrero and M. Lorenzo for field assistance. We also thank M. Pizarro and J. M. Poquet for technical support. M. C. Cruces, C. Sanpera and X. Ruiz made valuable comments on an earlier version of this manuscript. Kirk Setser improved the style of the manuscript. This study was supported by the SECOVER program (Regional Environmental Agency, Sevilla, Spain) from 2004 to 2006. Xavier Santos was supported by a Beatriu de Pinós postdoctoral grant from the Generalitat de Catalunya (BP-B1 10211).

References

Aguilar J, Dorronsoro C, Fernández E, Fernández J, García I, Martín F, Sierra M, Simón M (2007) Remediation of As-Contaminated Soils in the Guadiamar River basin (SW, Spain). Water Air Soil Pollut 180:109-118.

Alastuey A, García-Sánchez A, López F, Querol X (1999) Evolution of pyrite mud weathering and mobility of heavy metals in the Guadiamar valley after the Aznalcóllar spill, south-west Spain. Sci Total Environ 242:41-55.

Alcorlo P, Otero M, Crehuet M, Baltanás A, Montes C (2006) The use of the red-swamp crayfish (*Procambarus clarkii*, Girard) as indicator of the bioavailability of heavy metals in environmental monitoring in the River Guadiamar (SW, Spain). Sci Total Environ 366:380-390.

Avery RA, White AS, Martín NH, Hopkin SP (1983) Concetrations of heavy metals in common lizards (*Lacerta vivipara*) and their food and environment. Amphibia-Reptilia 4:205-213.

Baxer C.V., Fausch K.D., SAUNDERS W.C. 2005.Tangled webs: reciprocal flows of invertebrate prey link streams and riparian zones. Freshwater Biology. 50:201-220.

Benito V, Devesa V, Munñoz O, Suñer MA, Montoro R, Baos R, Hiraldo F, Ferrer M, Fernández M, González MJ (1999) Trace elements in blood collected from birds feeding in the area around Doñana National Park affected by the toxic spill from the Aznalcóllar mine. Sci Total Environ 242:309-23.

Beyersmann D (1991) The significance of interactions in metal essentiality and toxicity. In: Merian E (ed) Metals and their compounds in the environment. Occurrence, analysis and biological relevance. Weinheim, Germany: VCH Verlagsgesellscheft.

Burger J, Campbell KR, Campbell TS (2004) Gender and spatial patterns in metal concentrations in brown anoles (*Anolis sagrei*) in southern Florida, USA. Environ Toxicol Chem 23:712-718.

Burger J, Campbell KR, Campbell TS, Shukla T, Jeitner C, Gochfeld M (2005) Use of skin and blood as non-lethal indicators of heavy metal contamination in

northern water snakes (*Nerodia sipedon*). Arch Environ Contam Toxicol 49:232-238.

Burger J, Murria S, Gaines KF, Novak JM, Punshon T, Dixon C, Gochfeld M (2006) Element levels in snakes in South Carolina: differences between a control site and an exposed site on Savannah River site. Environ Monit Assess 112:35-52.

Cabrera F, Clemente L, Díaz Barrientos E, López R, Murillo JM (1999) Heavy metal pollution of soils affected by the Guadiamar toxic flood. Sci Total Environ 242:117-129.

Campbell KR, Campbell TS (2002) A logical starting for developing priorities for lizard and snake ecotoxicology: a review of available data. Environ Toxicol Chem 21:894-898.

Carretero MA, Llorente GA (1993) Feeding of two sympatric lacertids in a sandy coastal area (Ebro Delta, Spain). In: Valakos ED, Bohme W, Pérez-Mellado V, Maragou P (eds) Lacertids of the Mediterranean region. A biological approach. Hellenic Zoological Society, Athens, p 155.

Carretero MA, Llorente GA (1997) Habitat preferences of two sympatric lacertid in the Ebro Delta (NE Spain). In: Bohme W, Bischoff W, Ziegler T (eds) Herpetologia Bonnensis. Societas Europaea Herpetologica, Bonn, p 51.

Carretero MA, Kaliontzopoulou A (2005) Size and shape variation between populations of the lizard *Psammodromus algirus*. In: 5th Worls Congress of Herpetology 19-24 June. Abstract book. Stellonbosch (South Africa).

Carretero MA, Montori A, Llorente GA, Santos X (2002) *Psammodromus algirus* (Linné, 1758) Lagartija colilarga. In: Pleguezuelos JM, Márquez R, Lizana M (eds) Atlas y Libro Rojo de los Anfibios y Reptiles de España. Segunda impresión. Dirección General de Conservación de la Naturaleza-Asociación Herpetológica Española, Madrid, p 260.

Chang LW (1996) Toxicology of metals. Lewis Publ, Boca Raton, FL, USA.

Díaz JA, Carrascal LM (1990) Prey size and food selection of *Psammodromus algirus* (Lacertidae) in central Spain. J Herpetol 2:342-347.

Dorronsoro C, Martin F, Ortiz I, García I, Simón M, Fernández (2002) Migration of trace elements from pyrite tailings in carbonate soils. J Environ Qual 31:829-35.

Fletcher DE, Hopkins WA, Saldaña T, Baionno JA, Arribas C, Standora MM, Fernández-Delgado C (2006) Geckos as indicators of mining pollution. Environ Toxicol Chem 25:2432-2445.

Gallart F, Benito G, Martín-Vide JP (1999) Fluvial geomorphology and hydrology in the dispersal and fate of pyrite mud particles released by the Aznalcóllar mine tailings spill. Sci Total Environ 242:13-26.

Grimalt, JO, Ferrer M, Macpherson E (1999) The mine tailing accident in Aznalcóllar. Sci Total Environ 242:3-11.

Hesel DR (1990) Less than obvious: Statistical treatment of data below the detection limit. Environ Sci Technol 39:419-423.

Hopkins WA. Reptile toxicology: challenges and opportunities on the last frontier in vertebrate ecotoxicology (2000). Environ Toxicol Chem 19:2391-2393.

Hopkins WA, Roe JH, Snodgrass JW, Jackson BP, Kling DE, Rowe CL, Congdon JD (2001) Nondestructive indices of trace element exposure in squamate reptiles. Environ Pollut 115:1-7.

Hopkins WA, Roe JH, Snodgrass JW, Staub BP, Jackson BP, Congdon JD (2002) Effects of chronic dietary exposure to trace elements on banded water snake (*Nerodia fasciata*). Environ Toxicol Chem 21:906-913.

Hopkins WA, P. Stauba BP, Baionnoa JA, Jacksona B P, Talentb LG (2005) Transfer of selenium from prey to predators in a simulated terrestrial food chain. Environ Pollut 134:447-456.

Hsu MJ, Selvaraj K, Agoramoorthy G (2006) Taiwan's industrial heavy metal pollution threatens terrestrial biota. Environ Pollut 143:327-334.

Jelaska LS, Blanus M, Durbes P, Jelaska SD (2007) Heavy metal concentrations in ground beetles, leaf litter, and soil of a forest ecosystem. Ecotoxicol Environ Saf 66:74-81.

Jon Peter AL, Viraraghavan T (2005) Thallium: a review of public health and environmental concerns. Environ Int 31:493-501.

Krauss U, Wiegand J (2006) Long-term effects of the Aznalcóllar mine spill-heavy metal content and mobility in soils and sediments of the Guadiamar river valley (SW Spain). Sci Total Environ 367:855-871.

Lambert MRK (1997) Environmental effects of heavy spillage from a destroyed pesticide store near Hargeisa (Somaliland) assessed during the dry season, using reptiles and amphibians as bioindicators. Arch Environ Contam Toxicol 32:80-93.

Lance VA, Cort T, Masuoka J, Lawson R, Saltman P (1995) Unusual high zinc concentrations in snake plasma, with observations on plasma zinc concentrations in lizards, turtles and alligators. J Zool 235:577-585.

Linder G, Grillitsch B. Ecotoxicology of metals. In: Sparling DW, Linder G, Bishop CA (eds) Ecotoxicology of amphibians and reptiles. SETAC, Pensacola, FL. p 325.

Loumbordis NS (1997) Heavy metals contamination in a lizard, *Agama stellio stellio*, compared in urban, high altitude and agricultural, low altitude areas of north Greece. Environ Contam Toxicol 58:945-952.

Madejón P, Marañón T, Murillo JA, Robinson B (2004) White poplar (*Populus alba*) as a biomonitor of trace elements in contaminated riparian forests. Environ Pollut 132:145-155.

Madejón P, Marañón T, Murillo JA (2006) Biomonitoring of trace elements in the leaves and fruits of wild olive and Holm oak trees. Sci Total Environ 355:187-203.

Mann RM, Serra EA, Soares AMVM (2006) Assimilation of cadmium in a European lacertid lizard: is trophic transfer important? Environ Toxicol Chem 25:3199-3203.

Mann RM, Sánchez-Hernández JC, Serra EA, Soares AMVM (2007). Bioaccumulation of Cd by a European lacertid lizard after chronic exposure to Cd-contaminated food. Chemosphere 68:1525-1534.

Marco A, López-Vicente M, Pérz-Mellado V (2004) Arsenic uptake by reptile flexible-shelled eggs from contamined nest substrates and toxic effect of embryos. Bull Environ Contam Toxicol 72:983-990.

Marco A, López-Vicente ML, Pérez-Mellado V (2005) Soils acidification negatively affects embryonic development of flexible-shelled lizard eggs. Herpetol J 15:107-111.

Márquez- Ferrando R, Pleguezuelos JM, Santos X, Ontive D, Fernández-Cardenete JR. Recovering the reptile community after the mine-tailing accident of Aznalcóllar (SW Spain). Res Ecol 2007 in press.

Meharg AA, Osborn D, Pain DJ, Sánchez A, Naveso MA (1999) Contamination of Doñana food-chains after the Aznalcóllar mine disaster. Environ Pollut 105:387-390.

Olías M, Cerón JC, Moral F, Ruiz F (2006) Water quality of the Guadiamar River after the Aznalcóllar spill (SW Spain). Chemosphere 62:213-225.

Ordóñez R, Giráldez JV, Vanderlinden K, Carbonell R, González P (2006) Temporal and spatial monitoring of the pH and heavy metals in a soil polluted by mine spill post cleaning effects. Water Air Soil Pollut 178:229-243.

Pain DJ, Sánchez A, Meharg AA (1998) The Doñana disaster: Contamination of a world heritage estuarine marsh ecosystem with acidified pyrite mine waste. Sci Total Environ 222:45-54.

Pérez-Quintero JC, Rubio-García JC (1997) Alimentación de la lagartija colilarga, Psammodromus algirus (L) (Sauria, Lacertidae), en el litoral de Huelva (SO España). Doñana-Acta Vertebrata 34:3-26.

Pérez-Quintero JC (1996) Reproductive characteristic of three Mediterranean lizards: Psammodromus algirus (L.), Psammodromus hispanicus (Fitzinger) and Acanthodactylus erythrurus (Schinz). Amphibia-Reptilia 17:197-208.

PICOVER (2003) Ciencia y Restauración del Río Guadiamar. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla.

Querol X, Alastuey A, López-Soler A, Plana F (2000) Levels and chemistry of atmospheric particulates induced by spill of heavy metal mining wastes in the Doñana area, southwest Spain. Atmos Environ 34:239-253.

Querol X, Alastuey A, Moreno N, Alvarez-Ayuso E, García-Sánchez A, Cama J, Ayora C, Simón M (2006) Immobilization of heavy metals in polluted soils by the addition of zeolitic material synthesized from coal fly ash. Chemosphere 62:171-180.

Salvador A. Reptiles (1998) In: Ramos MA, Coord. Fauna Ibérica. Vol 10. Museo Nacional de Ciencias Naturales, CSIC, Madrid.

Sanpera C, Morera M, Ruíz X, Jover L (2000) Variability of mercury and selenium levels in clutches of Audouin's gulls (*Larus audouinii*) breeding at the Chafarinas Islands, southwest Mediterranean. Environ Contam Toxicol 39:119-123.

Sánchez-Chardi A (2007) Tissue, age, and sex distribution of thallium in shrews from Doñana, a protected area in SW Spain. Sci Total Environ 383:237-240.

Santos X, Pastor D, Llorente GA, Albaigés J (1999) Organochlorine levels in viperine snake *Natrix maura* carcasses from the Ebro Delta (NE Spain): sexual and size-related differences. Chemosphere 39:2641-2650.

Schmidt J (1980) Blei- und Cadmium-Rückstände bei inner- und ausserstädtischen Lacerta populationen. Verhand Gesell Ökologie 9:297-300.

Schmidt J (1988) Indikatorbedeutung von *Lacerta agilis* für die Bewertung urbaner Raume. Mertensiella 1:195-204.

Simón M, Ortiz I, García I, Fernández E, Fernández J, Dorronsoro C (1999). Pollution of soils by the toxic spill of a pyrite mine (Aznalcóllar, Spain). Sci Total Environ 242:105-115.

Solá C, Burgos M, Plazuelo A, Toja J, Plansa M, Prat N (2004) Heavy metal bioaccumulation and macroinvertebrate community changes in a Mediterranean stream affected by acid mine drainage and an accidental spill (Guadiamar River, SW Spain). Sci Total Environ 333:109-126.

Taggart MA, Figuerola J, Green AJ, Mateo R, Deacon C, Osborn D, Meharg AA (2006) After the Aznalcóllar mine spill: Arsenic, zinc, selenium, lead and copper levels in the livers and bones of five waterfowl species. Environ Res 100:349-361.

Toja J, Alcalá E, Burgos MD, Martín G, Plazuelo A, de Schutter T (2003) Efecto del vertido tóxico en las comunidades de plancton y perifiton del río Guadiamar. In: Arenas JM, Martínez-Faraco FR, Mora A (eds) PICOVER 1998–2002. Ciencia y restauración del río Guadiamar. Junta de Andalucía: Consejería de Medio Ambiente, Sevilla, p 94.

Van Straalen NM, Butovsky RO, Pokarzhevskii AD, Zaitsev AS, Verhoef CS (2001) Metal concentrations in soil and in invertebrates in the vicinity of a metallurgical factory near Tula (Russia). Pedobiologia 45:451-466.

CAPÍTULO 5

Análisis de asimetría corporal en la lagartija colilarga (*Psammodromus algirus*) en el Corredor Verde del Guadiamar

Rocío Márquez-Ferrando, Xavier Santos, Juan M. Pleguezuelos y Diego Ontiveros

Resumen

En este estudio se analizó el grado de asimetría en los poros femorales que presentan las poblaciones de la lagartija colilarga (*Psammodromus algirus*) expuestas a niveles altos de contaminación por metales pesados, y se compararon con otras cuatro poblaciones cercanas geográficamente con diferente grado de exposición a la misma contaminación. No hubo diferencias en el grado de la asimetría fluctuante entre las poblaciones. Aparentemente esto nos muestra que no existe un efecto significativo de la contaminación por metales pesados en la asimetría en los poros femorales de *Psammodromus algirus*.

Palabras clave: *Psammodromus algirus*, asimetría fluctuante, metales pesados, vertido minero de Aznacóllar

1. Introducción

El análisis de la asimetría fluctuante en la población de una especie es una de las técnicas usadas para estimar la estabilidad en el desarrollo de los organismos. En varios casos, se ha comprobado que poblaciones expuestas a cierto estrés ambiental (temperaturas adversas, escasez de reservas aportadas por la madre, perturbación por la alta densidad poblacional, altos niveles de toxicidad, etc…) incrementan la asimetría fluctuante (Leary & Allendorf, 1989; Palmer & Strobeck, 2003; Crnobrnja-Isailovic et al., 2005).

Hay diversos métodos que se pueden utilizar para identificar los efectos cuantificando el grado en que un factor estresante del ambiente está afectando el desarrollo de los organismos (Tarlow et al., 2007). El estudio de la asimetría en estructuras morfológicas o merísticas bilaterales es una de ellas. Hay tres modalidades de asimetría: *antiasimetría*, *asimetría bilateral* y *asimetría fluctuante*. La *antiasimetría* se caracteriza por una distribución hacia la bimodalidad o hacia la platicurtosis de la diferencia entre el lado derecho e izquierdo del carácter, con media de (D-I) generalmente igual a cero. En la *asimetría direccional* se detecta mayores diferencias entre los lados pero siempre hay un lado que presenta la mayor magnitud del carácter. La *asimetría fluctuante* se caracteriza por una distribución normal de las diferencias entre (D-I), con media igual a cero (Clarke, 1993).

En abril de 1998, la balsa de contención de lodos tóxicos de la mina de Aznalcóllar provocó el vertido de aproximadamente 2 millones de litros de lodos piríticos y 4 millones de litros de aguas ácidas, con una elevada concentración de metales pesados como Zn, As, Cu, Sb, Tl, Cd, etc… (Grimalt et al., 1999). Ocho años después de la restauración de la zona afectada, la comunidad de reptiles se recupera gracias a proyectos de restauración específicos para estos organismos (Capítulo 4).

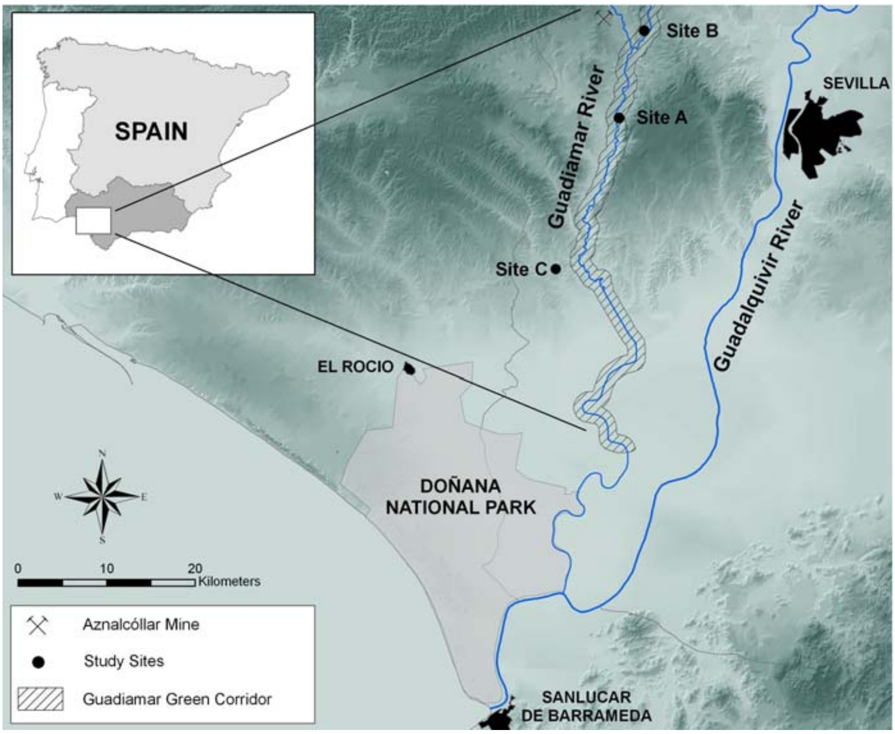


Figura 1.- Área de estudio en la que se observa la localización del Corredor Verde del Guadiamar y de las tres poblaciones de *Psammodromus algirus*.

Actualmente la lagartija colilarga (*Psammodromus algirus*) es una especie muy abundante en el Corredor Verde del Guadiamar y como reptil es un excelente bioindicador de contaminación (Capítulo 4). La lagartija colilarga (*Psammodromus algirus*) es un lacértido de mediano tamaño (LHC 60-80 mm). Se extiende por la mayor parte de áreas mediterráneas ibéricas donde es muy abundante, sobre todo en zonas de cierta cobertura arbustiva (Salvador & Pleguezuelos, 2002). Su carácter oportunista le confiere una alta plasticidad para recolonizar zonas restauradas llegando a ser uno de los primeros colonizadores después de la restauración del Corredor Verde del

Guadiamar (Capítulo 3). Su densidad ha ido aumentando progresivamente a medida que el hábitat se ha ido recuperando. Su elevada abundancia local, su posición en la cadena trófica (se alimenta de gran variedad de artrópodos; Pérez-Quintero y Rubio-García, 1997), su rápida tasa de renovación poblacional (alcanza la madurez sexual al primer año de vida; Marí et al., 1996) y su limitada capacidad de dispersión, hace de ella un buen bioindicador potencial de zonas contaminadas. En el capítulo 4, hemos analizado el nivel de concentración de metales pesados en individuos de poblaciones localizadas en zonas afectadas directamente e indirectamente por el vertido, y una población control. Los datos mostraron que los individuos que viven en zonas contaminadas acumulan altos niveles de metales pesados de As, Tl, Sn, Pb, Cd y Cu. Los efectos de la contaminación en la lagartija colilarga, podrían reflejarse a nivel de individuo y a nivel poblacional.

Los poros femorales es una carácter que puede ser medido para determinar el grado de asimetría fluctuante en lacértidos (Martín et al., 2002; Carretero, 2003); se localizan en la cara ventral de los miembros posteriores y presentan dimorfismo sexual en número y tamaño (habitualmente mayores y más numerosos en las machos, Blasco, 1975) y diferencias entre poblaciones (Carretero, 2003). Las secreciones de los poros femorales marcan el área de campeo y/o informan sobre el estatus social y la competitividad del emisor (Mason, 1992). La asimetría en los poros femorales puede afectar a la eficacia biológica de los individuos entendida ésta en términos de éxito reproductivo, ya que la simetría en los poros femorales parece estar relacionada con la calidad sexual de los machos en la lagartija serrana (*Iberolacerta monticola*); las hembras parecen elegir a los machos machos más simétricos en poros femorales, ya que producen más feromonas que manifiesta su buena calidad (Martín et al., 2002).

Nuestra hipótesis es que las poblaciones expuestas a altos niveles de contaminación sufren una mayor inestabilidad en el desarrollo de los individuos de la lagartija colilarga (*P. algirus*), la cual se verá reflejada en un mayor grado de asimetría fluctuante en los poros femorales. Por lo tanto el objetivo de este estudio se centra en comprobar: i) si existen diferencias significativas en la asimetría fluctuante entre las poblaciones de estudio en función del nivel de contaminación al que están expuestas; y ii) si existe correlación positiva entre el nivel de contaminación y la asimetría que presentan los individuos.

2- Materiales y métodos

2.1. Área de estudio y metodología

Durante las primavera de 2006-2007 se capturaron individuos de lagartija colilarga (*P. algirus)* en dos localidades diferentes del Corredor Verde del Guadiamar (Figura 1): una población localizada a 20 km de la balsa minera (N=18; Puente de las Doblas; Población 1) que quedó totalmente cubierta de lodos y en la cual se realizaron tareas de limpieza y restauración; otra próxima a la balsa minera pero que no quedó cubierta por lodos (N= 12; Confluencia Río Agrio-Guadiamar: Población 2) y una tercera población perteneciente a una zona control (Pinares de Villamanrrique: Población 3) alejada 25 km de la mina y fuera del Corredor (N= 17). Para este estudio, también se tuvieron en cuenta los datos de dos poblaciones de *P. algirus* colectadas aproximadamente 30 años antes del vertido en la provincia de Huelva, pertenecientes a la colección herpetológica de la Estación Biológica de Doñana (El Mustio: Población 4 y El Quintillo: Población 5).

Para medir el grado de asimetría fluctuante, se seleccionó el número de poros femorales como variable merística. En las poblaciones de lacértidos, el número de poros femorales sigue una distribución normal, y presenta asimetría fluctuante (Carretero, 2003). Las ventajas que tiene analizar la asimetría fluctuante mediante el número de poros femorales y otros caracteres folidóticos, es que la probabilidad de cometer errores durante la toma de datos es mínima, ya que se observa fácilmente a simple vista (Crnobrnja-Isailovic et al., 2005). A cada individuo se le midió su tamaño corporal (Longitud Hocico Cloaca (LHC) en mm), el número de poros femorales a cada lado de los miembros posteriores y se sexó según el patrón en la coloración corporal y según los poros femorales (muy visibles en machos). Para asegurarnos del mínimo error en los datos se contaron hasta tres veces el número de poros femorales y sólo por la misma persona.
De los individuos pertenecientes al año 2006 se recogieron 5 cm de cola para su posterior análisis. Después se devolvieron los ejemplares al mismo lugar de captura. Los análisis de los metales pesados mostraron que las poblaciones expuestas a niveles de contaminación fueron la población 1 y 2 (ésta última en menor grado; Capítulo 4). Por lo quelas poblaciones control son la 3, 4 y 5 (Tabla 1).

2.2. Análisis estadísticos

La covariación entre el número de poros a la derecha y el número de poros a la izquierda (D-I) fue testada para determinar si la varianza de la diferencia es interpretada como una simple medida de la inestabilidad en el desarrollo (DI) (valor positivo de la covarianza) o es una medida compleja (valor negativo de la covarianza), ya que además de estar influenciada por la inestabilidad en el desarrollo también puede estar afectada por el ambiente y por el genotipo (Palmer and Strobeck, 2003). La covarianza fue calculada mediante la siguiente fórmula:

$$(DiIi) = \sum [(Di - Dx)(Ii - Ix)] / (N - 1)$$

Donde:

Di y Ii es el número de poros a la derecha y a la izquierda de cada individuo.

Dx es el número medio de poros a la derecha.

Ix es la media de poros a la izquierda.

N es el número total de individuos

En todos los análisis se consideró como variable el índice de asimetría corregido por el efecto del tamaño sobre el carácter **(D-I)/0.5(D-I)**, ya que los individuos más grandes suelen tener mayor número de poros (Carretero, 2003). Antes de determinar las diferencias en la asimetría entre poblaciones, se determinó el tipo asimetría (direccional o fluctuante) presente en cada población mediante el test de normalidad de Kolmorogov-Smirnov y el t-test para determinar si la media de la diferencia de **(D-I)/0.5(D-I)** difería de cero. En cada población se testó la Kurtosis de la distribución de los datos de asimetría para determinar si había antiasimetría mediante el programa Statistica v7.0.

Las diferencias en el grado de asimetría fluctuante entre poblaciones se analizaron de dos formas diferentes; por un lado, testando la media del valor de la asimetría fluctuante mediante el índice **|D-I|/0.5(D+I)** usando una ANOVA, y por otro lado testando la heterogeneidad de las varianzas de cada población en la asimetría mediante el índice **(D-I)/0.5(D+I),** usando el test de Levene's.

Para determinar si había diferencia en el nivel de contaminación por metales pesados entre los individuos simétricos o asimétricos machos y hembras se usó una ANOVA de dos vías, en el que establecimos como factores el sexo y la asimetría, y como variable dependiente la concentración de cada metal pesado en las muestras de cola de cada individuo (μg/g).

3. Resultados

Debido al bajo número de machos y hembras capturados en las dos poblaciones de *P. algirus* afectadas por del vertido (1 y 2), no fue posible detectar el tipo de asimetría por sexos, por lo que sólo analizamos la asimetría en la población independientemente del factor sexo (Tabla 1).

Tabla 1.- Valores medios de machos (M) y hembras (H) en la Longitud Hocico Cloaca (LHCen mm) en el número de poros femorales a la derecha y a la izquierda (PORFD y PORFI), los valores medios de los índices de asimetría sin la corrección para el efecto del tamaño sobre el carácter (D-I)/0.5*(D+I) y con la corrección del efecto del tamaño |D-I|/0.5*(D+I)

Localidad	**Sexo**	**Nº indiv.**	**Media LHC**	**Media PorFD**	**Media PorFI**	**Media PORF**	**Media D-I**	**Media (D-I) /0.5*(D+I)**	**Media |D-I| /0.5*(D+I)**
Doblas	M	11	72	18.73	18.73	18.73	0.00	-0.00002	0.028
	H	7	70.71	17.28	17.57	17.42	-0.28	-0.018	0.034
Agrio	M	6	64.33	18.83	18.83	18.83	-0.00	0.00053	0.0
	H	6	69.66	17.33	17.17	17.25	0.167	0.01	0.05
Pinar	M	7	70.15	18.86	17.50	17.70	-0.20	-0.01	0.04
	H	10	59.70	17.50	17.70	17.65	-0.10	-0.01	0.04
El Mustio	M	28	69.54	18.57	18.64	18.61	-0.14	-0.01	0.04
	H	13	65.31	17.54	17.77	17.65	0.05	0.0028	0.04
El Quintillo	M	22	64.77	18.71	18.81	18.73	0.11	0.01	0.03
	H	18	64.67	17.83	17.66	17.75	0.11	0.01	0.02
Doblas	M y H	18	71.50	18.17	18.28	18.22	-0.11	-0.007	0.03
Agrio	M y H	12	67.00	18.08	18.00	18.04	0.08	0.004	0.05
Pinar	M y H	17	64.00	18.06	18.24	18.15	-0.17	-0.01	0.04
El Mustio	M y H	41	68.19	18.24	18.37	18.31	-0.12	-0.01	0.04
El Quintillo	M y H	40	64.73	18.30	18.27	18.29	0.025	0.03	0.00

La covariación entre el número de poros a la derecha y a la izquierda (D-I) fue positiva, por lo que la medición de la asimetría en los individuos de *P. algirus* mediante los poros femorales es una medida simple de la inestabilidad durante el desarrollo. El test de Kolmorogov-Smirnov determinó la distribución normal para las poblaciones de las Doblas, el Río Agrio y del Pinar de Villamanrrique, pero no para las poblaciones de *P. algirus* 4 y 5 previas al vertido (Figura 2).

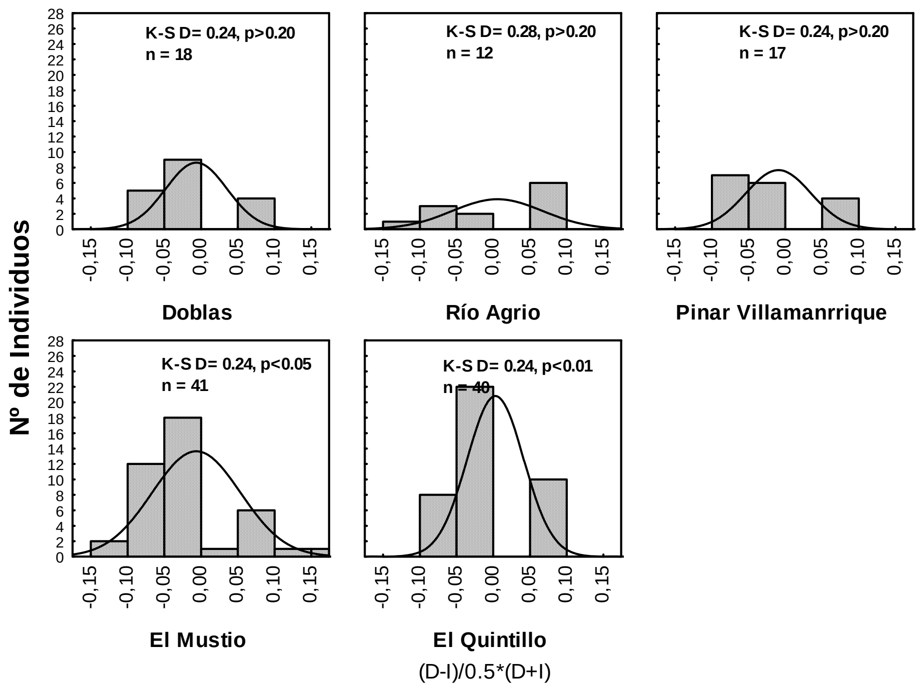


Figura 2. Test de Kolmorogov-Smirnov para la distribución de los datos de asimetría en los poros femorales de la lagartija colilarga (*Psammodromus algirus*) en las distintas poblaciones de estudio del SO de la Península Ibérica en dos poblaciones expuesta a niveles de contaminación (Doblas y Río Agrio) y otras cuatro poblaciones no expuestas a contaminación (Pinar de Villamanrrique, El Mustio y El Quintillo).

Los valores de las medias de la asimetría en las poblaciones no mostraron ser diferente de cero ($t < 0.34$, $p < 0.79$). Por lo que las poblaciones de *P. algirus* afectadas por el vertido y la población control (1, 2 y 3 respectivamente) mostraron tener asimetría fluctuante. Las poblaciones previas al vertido mostraron un patrón confuso en la asimetría. La población 4 mostró un valor alto de Kurtosis = 2.63; sin embargo, la población 5 mostró un valor más bajo = -0.84.

Para las poblaciones en las que se determinaron asimetría fluctuante, los análisis de ANOVA no detectaron diferencias significativas en la asimetría entre poblaciones (F(2, 44) = 2.22, p = 0.12) entre las poblaciones donde se determinó asimetría fluctuante. Por otro lado el test de Levene's (F(2,44) = 2.28, p = 0.12) tampoco determinó diferencias en la asimetría fluctuante entre estas poblaciones.

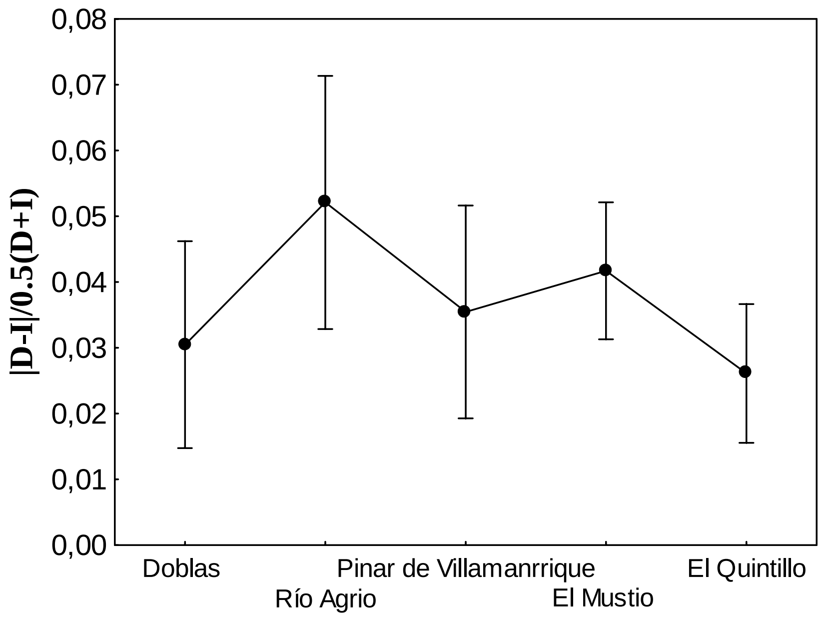


Figura 3. Medias de los valores de asimetría en los poros femorales de la lagartija colilarga (*Psammodromus algirus*) en las distintas poblaciones de estudio del SO de la Península Ibérica en dos poblaciones expuesta a niveles de contaminación (Doblas y Río Agrio) y otras cuatro poblaciones no expuestas a contaminación (Pinar de Villamanrrique, El Mustio y El Quintillo).

Los niveles de metales pesados no se pudieron normalizar, por lo que utilizamos una ANOVA factorial por rangos. El análisis no mostró diferencias significativas entre el nivel de contaminación por metales pesados entre los individuos simétricos y asimétricos, pero la mayoría de los datos sugieren que los individuos asimétricos tienden a mostrar una mayor nivel de contaminación (Tabla 2; Figura 4).

Tabla 2.- Valores de la ANOVA factorial por rangos en el nivel de contaminación, para los distintos metales pesados analizados, entre sexos, entre los individuos asimétricos y simétricos y en la interacción Sexo*, correspondientes a todos los individuos de las tres poblaciones (Las Doblas, Río Agrio y Pinar de Villamanrrique) en las que se analizó el nivel de contaminación**.**

	Sexos		**Asimetría**		**Sexo*Asimetría**	
	F(1,27)	*p*	*F*(1,27)	*p*	*F*(1,27)	*p*
Hg	0.29	0.59	0.78	0.39	1.48	0.23
Sb	0.08	0.78	1.05	0.32	0.07	0.80
Cd	0.47	0.50	1.30	0.26	0.24	0.63
Cr	1.53	0.23	8.98	0.01	0.01	0.94
Tl	0.00	0.98	0.47	0.50	0.50	0.48
Sn	0.39	0.54	0.39	0.54	0.39	0.54
Ba	0.57	0.46	2.90	0.10	0.28	0.60
Pb	0.59	0.45	1.46	0.24	0.08	0.78
Cu	0.85	0.37	0.00	1.00	2.28	0.14
Sr	2.03	0.17	1.48	0.23	0.50	0.49
Mn	3.70	0.07	1.41	0.24	0.12	0.73
Rb	0.06	0.81	0.14	0.71	0.50	0.49
As	0.27	0.61	0.00	0.99	0.00	1.00
Zn	0.00	0.97	2.95	0.10	0.28	0.60

4. Discusión

El Corredor Verde del Guadiamar es un escenario idóneo para hacer estudios sobre el efecto que puede tener la contaminación por metales pesados sobre los organismos durante su desarrollo y su crecimiento. Los resultados en el estudio sobre la bioacumulación de metales pesados en *P.*

algirus muestran que es un excelente modelo como bioindicador de contaminación del Corredor Verde del Guadiamar. Nuestra hipótesis de partida era que poblaciones *P. algirus* expuestas a altos niveles de metales pesados podrían presentar mayor asimetría fluctuante en los poros femorales que aquellas poblaciones menos expuestas o no expuestas a contaminación. Entre las poblaciones estudiadas no hemos encontrado diferencias significativas en la asimetría fluctuante, lo cual sugiere que los individuos expuestos a contaminación no influyen en la simetría de los poros femorales. Pero debemos tener en cuenta varias consideraciones en estos resultados. Por un lado, la baja muestra de individuos en las zonas contaminadas puede ser la causa de no encontrar altos grados de asimetría fluctuante, ya que se necesita un tamaño muestral más grande para detectar este tipo de asimetrías. El test de Kolmorogov-Smirnov para testar la normalidad de la distribución de datos de la diferencia D-I puede dar un valor erróneo de la normalidad de los datos para caracteres merísticos y tamaño muestral pequeño (Palmer y Strobeck, 2003); por lo que no estamos seguros de que la distribución de los datos en asimetría en las poblaciones sigan una distribución normal; por otro lado sólo estamos considerando individuos adultos y el hecho de no considerar a los juveniles puede ser que estemos obteniendo resultados no representativos de la población. La contaminación de los ecosistemas puede aumentar la tasa de mortalidad de juveniles, ya que determinados estudios en las poblaciones de la lagartija roquera (*Iberolacerta monticola cyrenni*) demuestran que los suelos contaminados alteran la permeabilidad de la cáscara del huevo y por lo tanto afectan a la tasa de intercambio de humedad con el medio externo; de esta manera los individuos nacen muy pequeños en tamaño y peso y la capacidad de locomoción disminuye, alterando así el fitness del individuo (Marco et al., 2004, 2005). Otra cuestión a considerar es que sólo estamos midiendo un carácter en el individuo. Un estudio de asimetría que comprenda el mayor número de caracteres posibles puede responder más claramente a si la contaminación por metales pesados puede influir en la inestabilidad sobre el desarrollo, que la elección de un solo carácter no es suficiente para concluir que hay habido inestabilidad durante el desarrollo (Palmer, 1994; Tarlow et al., 2007).

Debido a la gran variabilidad encontrada en los niveles de metales entre individuos de una población, la misma cuestión la hemos intentado responder relacionando el nivel de contaminación con la asimetría o simetría en el individuo, independientemente de la población a la que pertenecían (dentro de las tres analizadas para la contaminación; 1, 2 y 3). El problema que nos encontramos es que aquí la relación causa-efecto que intentamos determinar no es directa. No sabemos en que medida la inestabilidad en el desarrollo está relacionada con el alto nivel de contaminación, ya que suponemos que el nivel de contaminación aumenta a medida que el individuo crece, lo cual es variable en el tiempo, y el efecto de la asimetría en los poros femorales es invariable. Aún así, llama la atención que parece ver una tendencia en que los individuos con mayores niveles de contaminación son asimétricos; pero no hemos detectado diferencias significativas en el nivel de contaminación entre estas dos categorías, lo cual no determina con claridad esta relación.

En un futuro esperamos determinar el efecto de la contaminación por metales pesados sobre la inestabilidad durante el desarrollo de los individuos, por lo que tendríamos que tener en cuenta todas estas consideraciones. Así pues se podrían hacer dos cosas, lo primero sería aumentar el número de individuos de las muestras de las poblaciones contaminadas, cogiendo individuos tanto juveniles como adultos. Por otro lado, también se podría completar este estudio midiendo la asimetría fluctuante en otro carácter como puede ser el número de escamas a un lado y otro de la cabeza.

Agradecimientos

Muchas gracias a Marcial y a Jesús Caro, que nos ayudaron en la captura de los individuos de la lagartija colilarga. A Miguel Angel carretero y a Xana por su ayuda en la resolución de las diversas dudas para el análisis de los datos. Y a la Universidad de Granada por su ayuda económica para realizar este estudio durante una estancia breve en el Centro de Investigaciones sobre Biodiversidad y Recursos Genéticos (CIBIO) en Vairao (Oporto) durante los meses de enero-abril (2008).

Bibliografía

Blasco, M. 1975. El dimorfismo sexual en cinco especies de la familia Lacertidae (Reptilia). Boletín Real de la Sociedad Española de Historia Natural (Biología) 73:237-242.

Bergstrom, C. A., T. E. Reimchen. 2002. Geographical variation in asymmetry in Gasterosteus aculeatus. Biological Journal of the Linnean Society 77: 9-22.

Bowers, C.F., H.G. Hanlin, D.C. Guynn Jr, J.P. Mclendon, J.R. Davis. 2000. Herpetofaunal and vegetation of a thermally-impacted stream at the begining of restoration. Ecological Engineering 15:101-114.

Campell, K.R., Campell T.S. 2002. A logical starting point for developing priorities for lizard and snake ecotoxicology: a review of avaible data. Enviorn Toxicol Chem 21:894-8.

Carbonell, R., Tellería J.L.1999. Feather traits and philochronology as indicators of stress in Iberian Blackcaps *Sylvia atricapilla.* Bird Study 46:161-169.

Carretero, M.A. 2003. Is femoralia a valid measure of fluctuating asymmetry in Lacertidae? 12th Ordinary General Meeting, Societas Europaea Herpetologica. St. Petersburg (Russia), 12-16 August 2003.

Carretero, M. A. and Llorente, G. A. 1993. Morfometría en una comunidad de lacértidos mediterráneos, y su relación con la ecología. Historia Animalium, 2:77-79.

Carretero, M. A. and Llorente, G. A. 1997. Reproduction of *Psammodromus algirus* in coastal sandy areas of NE Spain. Amphibia-Reptilia, 18: 369-382.

Clarke, G.M. 1992. Fluctuating asymmetry: A technique for measure developmental stress of genetic and environmental origin. Acta Zoologica Fennica 191:31-35.

Clarke, G.M. 1993. Fluctuating asymmetry of invertebrate populations as a biological indicator of environmental quality. Environmental Pollution 82:207-211.

Crnobrnja-Isailovic, J., I. Aleksic, D. Bejakovic. 2005. Fluctuating asymmetry in *Podarcis muralis* populations from Southern Montenegro: detection of environmental stress in insular populations. Amphibia-Reptilia 26:149-158.

Grimalt, J.O, M. Ferrer, E. Macpherson. 1999. The mine tailing accident in Guadiamar. The Science of the Total Environment 242 (1-3): 3-12.

Herrando, S. 2001. Habitat disturbance in Mediterranean landscapes: effects of fire and fragmentation on birds. Tesis Doctoral.

Hopkins, W.A. 2000. Reptile ecotoxicology: Challenges and opportunities on the last frontier in vertebrate toxicology. Environ Toxicol Chem 19:2391-2393.

Jones, C.G., 2002. Reptiles and amphibians. En: Handbook of Ecological restoration. Vol.1: Principles of Restoration (Perrow and Davy). 355-375.

Kaliontzopoulou, A., M.A. Carretero, Llorente, G.A. 2007. Multivariate and geometric morphometrics in the analysis of sexual dimorphism variation in *Podarcis* lizards. Journal of Morphology, 268:152.165.

Kaliontzopoulou, A., M. A. Carretero, G.A Llorente. 2008. Head shape allometry and proximate causes of head sexual dimorphism in *Podarcis* lizards: joining linear and geometric morphometrics. Biological Journal of the Linnaean Society, 93:111-124.

Lambert, M. R. K. 1993. Effects of the DDT ground-spraying against tsetse flies on lizards in NW Zimbawe. Envioron Pollut 82: 231-237.

Leary, R. F., F. W. Allendorf. 1989. Fluctuating asymmetry as indicator of stress: implications for conservations biology. Trends in Ecology & Evolution 4:214-217.

Lima, A., A. Kaliontzopoulou, M. A. Carretero. 2007. Asymmetry in *Podarcis bocagei* and *P. carbonelli*: a preliminary study.14th European Congress of Herpetology. Oporto (Portugal), 19-23 Septiembre 2007.

López P., Muñoz A., Martín J. 2002. Symmetry, male dominance and female mate preferences in the Iberian rock lizard, *Lacerta monticola.* Behavior, Ecology and Sociobiology 52: 342-347.

Loumbourdis, N.S. 1997. Heavy metal contamination in a lizard, *Agama stellio stellio,* compared in urban, high altitude and agricultural, low altitude, low altitude areas of the north Greece. Environ Contam Toxicol 58:945-952.

Palmer, A.R. 1994. Fluctuanting asymmetry analyses: a primer. In: Markow, T.A. (Ed.), Development Instability: Its origins and evolutionary implications. Kluwer, Dordrecht, Netherlands, pp. 335-364.

Pérez-Quintero J.C., Rubio-García J.C. 1997. Alimentación de la lagartija colilarga, Psammodromus algirus (L) (Sauria, Lacertidae), en el litoral de Huelva (SO España). Doñana-Acta Vertebrata 34:3-26.

Marco A, M. López-Vicente, V. Pérez-Mellado 2004. Arsenic uptake by reptile flexible-shelled eggs from contamined nest substrates and toxic effect of embryos. Bull Environ Contam Toxicol 72:983-990.

Marco A, M.L. López-Vicente, V. Pérez-Mellado. 2005. Soils acidification negatively affects embryonic development of flexible-shelled lizard eggs. Herpetological Jorunal 15:107-111.

Marí N., M.A. Carretero, G.A. Llorente 1996. Estructura de lãs edades em dos poblaciones costeras de Psammodromus algirus. Herpetologia IV Congreso Luso-Español VIII Congreso Español 5-8 December. Abstract book. Oporto, Portugal.

Márquez-Ferrando R., J.M Pleguezuelos, X. Santos, D. Ontiveros, J.R. Fernández- Cardenete. 2007. Recovering the reptile community after the mine-tailing accident of Aznalcóllar (SW Spain). Restoration Ecology, in press.

Márquez-Ferrando R., X. Santos, J. M. Pleguezuelos, D. Ontiveros. 2007. Bioaccumulation of heavy metals in the lizard *Psammodromus algirus* after a tailing-dam collapse in Aznalcollar (SW Spain). Submitted in Science of Total of Environment.

Mason, R.T. 1992. Reptilian pheromones. pp. 114-228. En: Gans, C. and Crews, D. (eds.), Biology of the Reptilia Vol. 18. Hormones, Brain, and Behavior. University of Chicago Press, Chicago.

Ryabov, I.N., Kryshev, I.I, 1991. Assessment of Level of Fluctuating Asymmetry and Radiation Dose of Fishes from a Marine Water-Cooling Pond of a Nuclear Power Plant. Journal of Ichthyology JITHAZ, 31:133-139.

Ruíz-Laguna, J., D. Bonilla, J. Ballesteros, J. López-Barea y S. Moreno. 2003. Aplicación de biomarcadores bioquímicos al seguimiento de los efectos biológicos del vertido de Aznalcóllar en el ratón moruno (*Mus spretus*). En Montes C., Borja F., Arenas J.M., Martínez-Faraco F.R., Mora A. (Eds.): *Ciencia y Restauración del Río Guadiamar*. PICOVER 1998-2002. Págs: 226-235. Consejería de Medio Ambiente. Junta de Andalucía. Sevilla.

Santos, X., D. Pastor, G.A. Llorente, J. Albaigés. 1999. Organochlorine levels in viperine snake *Natrix maura* carcasses from the Ebro Delta (NE Spain): sexual and size-related differences. Chemosphere 39: 2641-2650.

Salvador A., Pleguezuelos J.M. 2002. Reptiles españoles. Identificación. Historia natural y distribución. Canseco Ed.

Van Valen, L., 1962. A study of fluctuating asymmetry. Evolution 16, 125-142.

DISCUSIÓN GENERAL

El vertido minero de Aznalcóllar afectó a las comunidades de organismos ligados al al cauce fluvial y a la llanura de inundación. Pero, la rápida actuación de los equipos de limpieza y restauración a lo largo de la zona afectada favoreció la pronta recuperación de estas comunidades biológicas gracias a la disminución de la contaminación en el agua y en el suelo (PICOVER, 2003). La estructura de la vegetación de ribera y de la llanura de inundación quedó intensamente afectada por la retirada de los lodos, sólo los árboles más grandes aguantaron el empuje del vertido (Aguilar et al., 2003). Las aves dependientes de la vegetación y con gran capacidad de dispersión, pudieron refugiarse en las zonas de alrededor no impactadas. A partir de la restauración de la vegetación, nuestro estudio sobre la evolución temporal de la comunidad de aves, mostró que la riqueza y la diversidad se mantuvo constante entre el período 2000-2006. Sin embargo, la abundancia total de aves aumentó considerablemente. Este incremento en la abundancia pudo ser debido al aumento de la complejidad estructural de la vegetación, ya que, la diversidad y abundancia de aves está relacionada directamente su la estructura y densidad (Dechenes et al., 2003; Smile et al., 2004). La restauración, favoreció a especies asociadas al hábitat de la llanura de inundación. Sin embargo, el corredor todavía no alberga una disponibilidad de hábitat comparable con otros medios riparios no impactados, como el Arroyo Alcarayón próximo al Río Guadiamar. El arroyo Alcarayón mantiene una abundancia de aves aún mayor en la que predominan especies más ligadas a la vegetación riparia; mientras que en el corredor las especies que más abundan son las especies de hábitos generalistas y ligadas a medios abiertos. Otros estudios de restauración de ecosistemas alterados han mostrado que han de pasar muchos años para que la comunidad de aves sea similar a aquellas zonas no alteradas (Stuart-Smith et al., 2006). Por lo que, ocho años después del vertido, la vegetación restaurada ha aumentado pero no ha alcanzado el grado de desarrollo suficiente para albergar a una comunidad de aves similar a aquellas zonas no impactadas, ya que aún, predominan los espacios abiertos, con gran cobertura de herbáceas y poco matorral.

En nuestro segundo trabajo (Capítulo 2) los resultados de los análisis multivariantes mostraron que la vegetación riparia y la heterogeneidad paisajística del corredor fueron determinantes en la composición y abundancia de la comunidad de aves. La presencia de zonas forestales, ganaderas y agrícolas junto con otros medios riparios, puede haber influido en el alto número de especies observadas en la zona próxima a la mina. También se destacó la importancia de los olivares en los tramos alto y medio, debido a que son agroecosistemas que ofrecen refugio y recursos tróficos para muchas especies de aves (Muñoz-Cobos et al., 2003). Las especies más abundantes en estos tramos del corredor fueron especies de medios abiertos (fringílidos y aláudidos) y especies ligadas a la vegetación riparia (silvidos y túrdidos). Sin embargo, en el tramo bajo se encontraron los valores más bajos en riqueza de especies, abundando las especies ligadas al medio acuático (ardeidos) y a los espacios abiertos (fringílidos y aláudidos).

En general, la heterogeneidad paisajística en paisajes agrícolas está asociada a una alta riqueza de especies en comunidades avifauníticas (Benton et al., 2003; Devictor and Jiguet, 2007). Diversos estudios han mostrado la importancia del paisaje, con respecto al tamaño de los fragmentos, su configuración en el espacio y su calidad ecológica, en el mantenimiento de las funciones de los ecosistemas fluviales (Anderson, 1993; Passell, 2000; Brotons & Herrando, 2001; Cardoso Silva & Vickery, 2002; Uezu et al., 2005; Brawn, 2006). Otros han mostrado que la presencia y conservación de sistemas fluviales cercanos ayudan al mantenimiento de la conectividad en los ecosistemas fragmentados y a la dinámica metapoblacional de las comunidades de aves (Hanski, 1999; Machtans et al., 1996; Hanowiski et al., 2007; Smiley et al., 2007). Pero las variables descriptoras del cauce fluvial como la anchura, la calidad del agua y la abundancia de macroinvertebrados también pueden jugar un papel muy importante en la riqueza y abundancia de aves ligadas a estos ecosistemas (Rushton et al., 1994; Iwata et al., 2003;Vaughan et al., 2007).

En relación a los reptiles, la restauración de la vegetación no fue suficiente para su rápida y eficaz recuperación. El aporte de refugios artificiales, como componente esencial del hábitat para reptiles en zonas restauradas, ha quedado de manifiesto en el presente estudio (Capítulo 3). El incremento de la riqueza específica y abundancia en la comunidad de reptiles en la parcela tratada con refugios, pudo haber sido fruto de la complejidad estructural de la vegetación, ya que es un factor que puede influir en la composición y estructura de las comunidades de reptiles (Mushinsky, 1985; Galán, 1996; Lit et *al*., 2001; Montori et *al*., 2001; Nichols & Nichols, 2003). Sin embargo la recuperación de la comunidad de reptiles en la zona de estudio que se empleó como testigo (en la que no se colocaron refugios artificiales y que tenía parecida estructura de vegetación y la misma zona externa como fuente de individuos para la colonización) fue mucho más pobre y lenta.

Entre la comunidad de reptiles presentes en la región, las especies más abundantes como la salamanquesa común (*Tarentola mauritanica*), la culebra viperina (*Natrix maura*), las especies más generalistas como el lagarto ocelado (*Timon lepidus*), la culebra de herradura (*Hemorrohis Hippocrepis*), la culebra bastarda (*Malpolon monspoessulanus*), y las especies de menor tamaño como la lagartija colilarga (*Psammodromus algirus*), han sido las que hasta la finalización de nuestra monitorización han recolonizado el corredor. *Tarentola mauritanica* y *P. algirus* fueron las primeras especies registradas en la zona de estudio. Lo más probable es que *P. algirus* fuera la primera colonizadora, ya que *T. mauritanica* se observó en los troncos de árboles que quedaron después del vertido (obs. Pers.); posiblemente los árboles más grandes sirvieron de refugio cuando ocurrió el desastre. Los ofidios son estrictamente carnívoros (Salvador, 1998) y su posición como depredadores en la red trófica supone una destacada madurez del medio y una suficiente disponibilidad de alimento, por lo que han sido los últimos en recolonizar las zonas restauradas.

Psammodromus algirus fue la especie más abundante a lo largo del periodo de estudio en las dos parcelas. Esta especie muestra una clara dependencia de la estructura de la vegetación, abundando en zonas donde el matorral y los árboles alternan con suelo desnudo (Carretero & Llorente, 1993). Por lo tanto, no es una especie que *a priori* se pueda ver favorecida por la presencia de refugios, aunque el aumento de individuos en la parcela experimental a partir de la colocación de refugios fue evidente. Así pues, cabe pensar que los refugios podrían favorecer a la especie por otras razones como el aporte de recurso trófico que pueden encontrar bajo los refugios de madera (areneidos y coleópteros; observación de los autores) ya que su dieta se compone básicamente de estos dos grupos (Pérez-Quintero y Rubio-García, 1997).

El hecho de que *P. algirus* sea más abundante en el corredor que en el encinar adehesado, fuera del corredor, puede ser debido a la diferencia en la estructura de la vegetación. Los encinares adehesados son ecosistemas manejados por el hombre y no albergan la estructura y densidad de vegetación necesaria para esta especie, lo cual pueden afectar negativamente a sus poblaciones (Martín & López, 2002; Díaz, 2004). Por todo ello, cabe concluir que la restauración de la vegetación, unida a la colocación de refugios, han creado hábitats favorables para *P. algirus* (Márquez-Ferrando et al. 2008).

El resto de las especies presentes en la región según el "Atlas y Libro Rojo de Anfibios y Reptiles de España" y que no se observaron en los muestreos son especies sujetas a componentes muy específicos del hábitat, aún no presentes en el biotopo relativamente reciente que constituye el corredor.

Aunque los reptiles representan una cuarta parte de las especies de vertebrados terrestres y se hallan repartidos por muchos hábitats y climas, pocas veces se han utilizado como bioindicadores de la calidad del hábitat (Burger et al., 2004). Recientes estudios han resaltado la importancia de incluir a los reptiles como bioindicadores de la calidad de los ecosistemas en relación con los estudios ecotoxicológicos (Lambert, 1993; Loumbourdis, 1997; Hopkins, 2000; Santos et al., 2000). Los reptiles, y en especial los pequeños lacértidos, son especies muy útiles como bioindicadores de los ecosistemas debido a su abundancia, conspicuidad, facilidad de muestro y limitada capacidad de dispersión (Cambell & Campell, 2002). Nuestro estudio demostró la utilidad de *P. algirus* en la biomonitorización de la contaminación del Corredor Verde del Gaudiamar. Los análisis de concentración de metales pesados en individuos de *P. algirus* del corredor pone de manifiesto que el nivel de contaminación en este medio es aún preocupante por su alta biodisponibilidad para los organismos. La diferencia de las concentraciones de As, Tl, Sn, Pb, Cd y Cu entre la población del corredor cubierta por los lodos y la población control (1 km alejado de la zona afectada), muestran la persistencia y los altos niveles de contaminación en el corredor. Los estudios de bioacumulación en la población de *T.*

mauritanica presente en el Corredor Verde del Guadiamar sirven de apoyo a nuestros resultados, ya que se cuantificaron niveles altos de As, Tl, Cd y Pb en las poblaciones expuestas en zonas directamente contaminadas (Fletcher et al., 2006). Los datos del Pb, As y Cd son metales de gran interés en el seguimiento de la contaminación en el corredor, ya que fueron componentes importantes en los lodos vertidos (Alastuey et al., 1999; Cabrera et al., 1999) y el Tl por la alta toxicidad y desconocimiento del comportamiento en el suelo y en los organismos (Aguilar et al., 2003).

Cabe comentar que los niveles medios de concentración de metales detectados en la población de *P. algirus* localizada en el corredor pero no afectada directamente por los lodos (a 0.1 km de la zona afectada) demuestran que el vertido no ha afectado exclusivamente a las zonas cubiertas por los lodos, sino que también ha contaminado los ecosistemas más próximos. Esto puede ser debido por un lado, a las tareas de remoción de los lodos con maquinaria pesada, que provocó la dispersión de los sólidos en suspensión y su propagación hacia los alrededores (Aguilar et al., 2003; Madejón et al., 2006); y por otro lado debido a la gran conexión entre el sistema fluvial y sus alrededores mediante el intercambio de nutrientes, energía y e interacciones ecológicas entre especies de una zona y otra (Baxer et al., 2005).

El efecto que puede derivarse de la acumulación por metales pesados en los organismos puede traer consecuencias a nivel de individuo y a nivel poblacional, ya que puede afectar a la eficacia biológica (Sparling et al., 2000). El agua y los gases que los huevos absorben son determinantes para el buen desarrollo embrionario (Overall, 1994). Marcos et al. (2004; 2005) demostraron que los suelos contaminados o la acidificación de éstos afectan al desarrollo embrionario de la lagartija roquera (*Lacerta monticola cyrenni*), debido al efecto que tiene en la permeabilidad de los huevos. Los huevos eran más pequeños y como consecuencia, los individuos nacían con menor masa corporal, afectando a la locomoción. Como consecuencia del mal desarrollo de los individuos, la tasa de mortalidad de los juveniles podría incrementar y esto afectaría a la dinámica poblacional de la especie. Por lo que, la contaminación altera las condiciones naturales de los ecosistemas, provocando estrés ambiental en los organismos. En el Capítulo 5 de esta memoria nuestra hipótesis de partida era que las poblaciones de *P. algirus* expuestas a altos niveles de contaminación presentarían un mayor grado de asimetría en los poros femorales. Sin embargo, en el análisis de los datos no hemos detectado diferencias en el grado de asimetría fluctuante entre las poblaciones. Lo que si parece es haber una tendencia a que los individuos con mayor nivel de contaminación sean los asimétricos. Esta relación asimetría-concentración en metales pesados es difícil de interpretar ya que la acumulación de metales pesados aumenta posiblemente con la edad del individuo (Linder & Grillitsch, 2000), pero la asimetría en los poros femorales se fija durante el desarrollo ontogénico. Lo cual, nos hace pensar que está relación causa-efecto sólo se podría explicar si los individuos asimétricos en poros femorales ya tenían en su origen altos niveles de contaminación en metales pesados. Por lo que nuestra hipótesis no se ha cumplido, pero sería interesante aumentar el número de individuos que incluya a juveniles para determinar con más claridad la relación causa-efecto entre nivel de contaminación y asimetría en poros femorales.

Bibliografía

Aguilar, J., C. Dorronsoro, R. Beller, E. Fernández, J. Fernández, I. García, A. Iriarte, F. Martín. I. Ortiz, M. Simón. 2003.

Contaminación de suelos tras el vertido tóxico de Aznalcóllar. Universidad de Granada.

Alastuey A, García-Sánchez A, López F, Querol X (1999) Evolution of pyrite mud weathering and mobility of heavy metals in the Guadiamar valley after the Aznalcóllar spill, south-west Spain. Sci Total Environ 242:41-55.

Benton, TG, Vickery JA, Wilson JD, 2003. Farmland biodiversity: is habitat heterogeneity the key? Trends Ecol Evol 18, 182:188.

Brawn, J.D., 2006. Effects of restoring oak savannas on bird communities and populations. Conservation Biology 20:460-469.

Clergeau, P., Jokimäki J, Savard JPL. 2001. Are urban bird community influences by the bird diversity of adjacent landscape? J. Appl.Eccol 38, 1122-1134

Burger, J., R. Bowman, G. E. Woolfenden, M. Gochfeld. 2004. Metal and metalloid concentrations in the eggs of threatened Florida scrub-jays in suburban habitat f rom south-central Florida. Science of the Total Environment 328:185-193.

Campbell K. R., T. S. Campbell. 2002. A logical starting for developing priorities for lizard and snake ecotoxicology: a review of available data. Environmental Toxicology and Chemistry 21:894-8.

Cantebury, G.E., Martin, T.H., Petit D.R., Petit L.J., Bradfords, D.F., 2000. Bird community and habitat as ecological indicators of forest condition in regional monitoring. Conservation Biology 14, 544-558.

Cardoso Da Silva, J. M., Vickery P. D. 2002. Birds, in Perrow, M. R., Davy, A. J.(Eds). Handbook of Ecological restoration. Vol.1: Principles of Restoration. Cambridge University Press, Cambridge, pp. 376-388.

Devictor V., Jiguet F. 2007. Community richness and stability in agricultural landscapes: the importance of the surrounding habitats. Agriculture, Ecosystems and Environment 120, 179-184.

Iwata T., Nakano S., Murakami M. 2003. Stream meanders increase insectivorous bid abundance in riparian deciduous forest. Ecography 26, 325-337.

Litt, A. R., L. Provencher, G. W. Tanner and R. Franz. 2001. Herpetofaunal Responses to Restoration Treatments of Longleaf Pine Sandhills in Florida. Restoration Ecology 9, 462-474.

Loumbourdis, N. S. 1997. Heavy metal contamination in lizard, *Agama stellio stellio,* compared in urban, high altitude and agricultural, low altitude areas of north Greece. Environmental Contamination and Toxicology 58:945-952.

Madejón P, Marañón T, Murillo JA (2006) Biomonitoring of trace elements in the leaves and fruits of wild olive and Holm oak trees. Sci Total Environ 355:187-203.

Marco A, López-Vicente M, Pérz-Mellado V (2004) Arsenic uptake by reptile flexible-shelled eggs from contamined nest substrates and toxic effect of embryos. Bull Environ Contam Toxicol 72:983-990.

Marco A, López-Vicente ML, Pérez-Mellado V (2005) Soils acidification negatively affects embryonic development of flexible-shelled lizard eggs. Herpetol J 15:107-111.

Nichols, O. G, F. M. Nichols. 2003. Long-term trends in faunal recolonization after bauxite mining in the Jarrah forest of southwestern Australia. Restoration Ecology 1:261-272.

Passell, H. D. 2000. Recovery of bird species in minimally restored Indonesian tin strip mines. Restoration Ecology 8:112-118.

PICOVER. 2003. Ciencia y Restauración del Río Guadiamar. Junta de Andalucía. (ed.), Consejería de Medio Ambiente, Sevilla. 578pp.

Sparling, D. W., C. Bishop, G. Linder. 2000. Ecotoxicology of amphibians and reptiles. 875 pp.

Matchtans C.S., Villard M.A., Hannon S. 1996. Use of the riparian buffer strips as movement corridors by forest birds. Conservation Biology 10:1366-1379.

Stuart-Smith, A.K., Hayes, J.P., Schieck, J., 2006. The influence of wildfire, logging and residual tree density on bird communities in the northern Rocky Mountains. Forest Ecology and Management 231, 1-17.

Swift, B.L. Larson J.S., DeGraaf, R.M., 1984. Relationship of breeding bird density and diversity to habitat variables in forested wetlands. Wilson Bulletin, 96, 48-59.

Van Straalen NM, Butovsky RO, Pokarzhevskii AD, Zaitsev AS, Verhoef CS (2001) Metal concentrations in soil and in invertebrates in the vicinity of a metallurgical factory near Tula (Russia). Pedobiologia 45:451-466.

CONCLUSIONES

De acuerdo con los resultados expuestos, extraemos las siguientes conclusiones:

1.- El vertido minero afectó la disponibilidad del hábitat para las aves, debido a su dependencia de la vegetación. Pero, debido a su gran capacidad de dispersión pudieron refugiarse en los alrededores en los primeros años durante las tareas de restauración. La restauración de la vegetación favoreció la rápida recolonización de las aves y el aumento de la abundancia. Sin embargo, ocho años después del vertido, la vegetación no ha alcanzado el grado de desarrollo suficiente para albergar a una comunidad de aves similar a aquellas zonas no impactadas, ya que aún, predominan los espacios abiertos, con gran cobertura de herbáceas y poco matorral.

2.- La vegetación de ribera, los olivares y la presencia de los ecosistemas riparios que rodean al corredor, fueron las variables que determinaron la comunidad en el tramo alto-medio del corredor. Sin embargo, la cobertura de las herbáceas fue la más importante en el tramo inferior. Por tanto, además de la conservación de la vegetación riparia, hay que tener en cuenta la conservación de la heterogeneidad paisajística en futuros proyectos sobre la conectividad y conservación de la comunidad de aves en el Corredor Verde del Guadiamar.

3.- La comunidad de reptiles no se recuperó con los proyectos de restauración genéricos del hábitat. La comunidad de reptiles sin embargo sí se recuperó de manera más rápida y eficaz en una zona experimental en la que se realizó una restauración específica para este grupo. Los refugios artificiales para reptiles aportados en la parcela experimental favorecen el reestablecimiento de los comunidad en ambientes alterados de forma más rápida y eficaz. Seis de las trece especies que se han registrado en la región, han recolonizado las áreas restauradas con refugios en un plazo de seis años., mientras que menos especies, y sobre todo, con menor abundancia, ha colonizado durante el mismo tiempo otra parcela testigo, sin tratamiento con refugios. Esta información podría ser utilizada por el equipo técnico de mantenimiento para potenciar la recuperación de los reptiles a lo largo de todo el corredor Verde del Guadiamar.

4.- Ocho años después del vertido minero, los individuos de lagartija colilarga que viven en las zonas cubiertas por los lodos indicaron que la contaminación de As, Tl, Sn, Pb, Cd y Cu persiste en altos niveles en formas biodisponibles para los organismos, a pesar de los esfuerzos de limpieza y restauración realizados.

5.- La exposición a la contaminación no mostró efectos estadísticamente significativos en la asimetría de los poros femorales en las poblaciones de lagartija colilarga, aunque parece haber una tendencia a que los individuos con mayores niveles de contaminación sean más asimétricos.

6.- El vertido tóxico de Aznalcóllar afectó de diferente manera a las comunidades de organismos ligadas al ecosistema dañado, ya que todos no tienen la misma capacidad de colonizar nuevas zonas y los requerimientos en los componentes del hábitat no son los mismos para todos. La evolución de la comunidad de las aves y reptiles pasa por una buena conservación de la vegetación riparia y de la conservación de la heterogeneidad paisajística que lo rodea, destacando los ecosistemas riparios más cercanos y los olivares, ya que favorecen el mantenimiento de la conectividad en la Cuenca del Guadiamar. Sin embargo los reptiles dependerán de la disponibilidad de refugios que haya en el medio, fundamentales para colonizar las zonas restauradas y dispersarse.