

CROCODILE SPECIALIST GROUP NEWSLETTER

VOLUME 33 No. 4 • OCTOBER 2014 - DECEMBER 2014



CROCODILE

SPECIALIST

GROUP

NEWSLETTER

VOLUME 33 Number 4
OCTOBER 2014 - DECEMBER 2014

IUCN - Species Survival Commission

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Printed by: Uniprint NT

Charles Darwin University, NT 0909, Australia

COVER PHOTOGRAPH: "Barbara", a metal Saltwater crocodile sculpture, was designed and constructed by artists Peter Jettner and Troya Bywaters ("Get Rocked", Humpty Doo, Australia), who specialise in incorporating agricultural and mining equipment in their artwork. "Barbara" is on display at Crocodylus Park, Darwin, Australia. Photograph: Grahame Webb.

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The CSG Newsletter provides information on the conservation, status, news and current events concerning crocodilians, and on the activities of the CSG. The Newsletter is distributed to CSG members and to other interested individuals and organizations. All Newsletter recipients are asked to contribute news and other materials.

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Virginia Aquarium and Marine Science Center Foundation, Virginia Beach, Virginia, USA.

Zambia Crocodile Farmers Association, Lusaka, Zambia.

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Simone Comparini, Pantera S.R.L., S. Croce s/Arno, Italy.

Crocodile Park, Malaga, Spain.

J.K. Mercado & Sons Agriculture Enterprises Inc., Philippines.

James Hennessey, The Reptile Village Zoo, UK.

The Ebey family, New Mexico, USA.

Marco Schultz, Germany.

Editorial

Once again I sadly report the loss of two well-respected CSG members. Professor Charles Santiapillai (Sri Lanka) passed away after a brief illness in October 2014. After retiring from the University of Peradeniya, Charles taught at Rajarata and Vauniya Universities while continuing his research. He inspired a lot of young naturalists and university students and his passing is a significant loss to the scientific community. Rafael Crespo Jr. (USA), lead biologist for the University of Florida’s American Crocodile Monitoring and Assessment Program, was an enthusiastic and highly motivated crocodile worker (see page 4 for details) who had all the hallmarks of a future CSG champion.

Against this sad news, the exceptional conservation efforts and skills of some CSG members have been formally recognized, and are well deserving of congratulations from all CSG members:

- Widodo Ramono, also a member of the IUCN-SSC Asian Rhino Specialist Group received the IUCN/WCPA Fred M. Packard Award for his long-standing dedication and inspirational leadership in preventing Sumatran and Javan rhinos from becoming extinct.
- Professor R.J. Rao, was nominated to India’s National Board for Wildlife (NBWL), a Statutory Body under the Wildlife Protection Act 1972. Chaired by the Prime Minister of India, the NBWL is the apex advisory body to the Government of India on matters pertaining to wildlife conservation, particularly within Protected Areas.
- Phil Wilkinson, CSG legend, received the Lifetime Achievement Award from The Wildlife Society (South Carolina Chapter) at the group’s annual meeting in Florence, South Carolina, USA (see page 5).

The review of crocodile management in Ethiopia by Matthew Shirley, Ludwig Siege and Meseret Adamasu is an excellent update. The potential for conservation to be enhanced by the ongoing sustainable use programs is great, and what is really needed now is some industry input from CSG members skilled in these matters. Copies of the report have been widely distributed and it has also been posted on the CSG website. This is a very valuable report, and we hope it assists Ethiopia with its management.

In the Sundarbans, Bangladesh, the news is not so good. In early December an oil tanker (Southern Star 7) slammed into another vessel along the Shela River, in the world’s largest mangrove forest. The tanker sank, spilling an estimated 75,000 gallons (350,000 litres) of fuel oil into waterways, creating classic oil spill problems for the local wildlife, including crocodiles. Attempts to clean up the oil spill do not appear to have been overly successful.

The 6th IUCN World Parks Conference was held in Sydney, Australia (12-19 November 2014), with assistance from both Commonwealth and NSW Governments. The meeting focused on the need to invigorate global efforts to protect natural areas, including scaling up the protection of landscapes and oceans. The meeting takes place once a decade, and was attended by more than 5000 delegates from 160 countries, including many SSC members.

The Regional Meeting of Crocodile Specialist Group in Siem Reap, Cambodia, hosted by the Government of Cambodia in collaboration with the European Commission and Fauna & Flora International, will be held on 25-29 May 2015. This will be an important meeting for addressing regional issues openly and frankly. One highlight will be a Veterinary Workshop on 25 May. The CSG Steering Committee will meet on 24 May, and as usual all participants are welcome to attend. There are many agenda items to be advanced after the last meeting in Lake Charles (May 2014), including trade issues concerning Madagascar and Colombia. Please make every effort to attend what promises to be a remarkably instructive meeting: details are at <http://www.csg2015cambodia.org>.

The Second Symposium on Crocodiles was held in Cartagena, Colombia, in December 2014. The 27 presentations addressed conservation, ecology, genetics, management, trade, legislation, population status, ethno-zoology and environmental education. Participants from Argentina, Venezuela, Mexico, Colombia, Brazil and Spain shared shared conservation and management experiences.

I can now confirm that the 24th Working Meeting of the CSG will be held in South Africa, 23-26 May 2016, at the Nombolo Mdhluli Conference Centre, Skukuza Rest Camp, Kruger National Park. Details will be provided as they become available.

A symposium titled “Bridging Science and Practice in Crocodylian Conservation” was organized during the 51st Annual Meeting of the Association for Tropical Biology and Conservation (ATBC), on 20-24 July in Cairns, Australia. The symposium, convened by Dr. Ruchira Somaweera, provided and shared synthesis and perspectives of best-practices in

studying and conserving crocodylians. Presenters Geoff McClure, Hamish Campbell, Majintha Madawala, Matthew Brien and Ruchira Somaweera discussed topics broadly addressing: 1. What are the recent advances, challenges and future directions in studying and conserving crocodylians?; 2. How could science be integrated with conservation issues and utilised by policy makers and conservation practitioners?; and, 3. How to address the emerging human-crocodile conflict by integrating the multiple dimensions of humans, economy and crocodylians? Symposia abstracts are available at: http://www.atbc2014.org/files/Abstracts/SYMPOSIUM_abstracts_1807.pdf.

It is with great pleasure that I announce the appointment of Dr. Mark Merchant as a Joint Vice Chairman of General Research with Dr. Val Lance.

Professor Grahame Webb, *CSG Chairman*.

Obituaries

Rafel Crespo, Jr. (1983-2014)



Rafael was born to Rafael Crespo Sr. and Ileana Pelaiz in Miami, Florida. He began his career as a veterinarian technician while attending Florida Atlantic University where he graduated with a bachelor's degree in Psychobiology in 2007. After finishing his bachelor's degree, he began his career building mesocosms and surveying for crayfish in the Everglades for a professor at Florida Atlantic University. There began his affinity for the Everglades and all the wildlife within.

In 2008, Rafael began his work at the University of Florida assessing the spread and management of *Lygodium*, an invasive plant in Florida. This was followed by a 3-month trip to Zimbabwe helping researchers on a wild dog project. His passion for South Florida and the Everglades brought him back to the University of Florida where he began working with a graduate student surveying amphibians across Loxahatchee National Wildlife Refuge, as well as helping with ongoing alligator and crocodile projects.

Beginning in 2010, Rafael's passion for crocodylian

conservation took off. He was the lead biologist for the University of Florida's American crocodile (*Crocodylus acutus*) Monitoring and Assessment Program (MAP) of Everglades restoration. Rafael's main duties were collecting data on nesting, growth, and survival of American crocodiles. These data are critical in the evaluation of restoration alternatives and establishing criteria for successful restoration efforts in the Everglades. His field research brought him to various parts of Everglades National Park, Biscayne National Park, Crocodile Lake National Wildlife Refuge, and Turkey Point power plant. Rafael also participated in international projects in Belize (Morelet's crocodile, *C. moreletii*), Ecuador (Black caiman, *Melanosuchus niger*), and Cuba where he presented research on American crocodiles in Florida.

Rafael also had a passion for native landscaping. He planted native trees and shrubs in this mom's backyard to bring a little bit of the Everglades home to her. He was always willing to lend a helping hand to landscape someone's yard or give them ideas.

Rafael was a pleasure to work with and his smile always made the mosquitoes and no-see-ums more manageable and thunderstorms and lightning less scary. Everybody who met him immediately was drawn close by his charismatic personality. He enjoyed fishing and tramping around in the swamp. He was humble and well respected by everyone. Rafael was also a champion kickboxer, but you would never know by how he presented himself. He will be missed dearly by all his co-workers, by the crocodiles he helped conserve, by the Everglades he helped restore, and by everyone who was fortunate to have known him.

A conservation fund has been established to honor Rafael Crespo. Memorial contributions may be made to the Rafael Crespo Conservation Fund at the UF/IFAS Ft. Lauderdale Research and Education Center. It is the family's hope that this fund will become an endowment to provide support for the education of wildlife biologists and students studying natural resources at UF.

Visit <http://www.uff.ufl.edu/appeals/CrespoMemorial> or send a gift (memo: Crespo Memorial) to: University of Florida Foundation (IFAS), PO Box 110170, Gainesville, FL 32611-0170, USA.

Jeff Beauchamp, Brian Jeffery, Michael Cherkiss (*CSG members*) and Frank Mazzotti (*CSG Regional Vice Chair North America*), Florida, USA.

CSG Student Research Assistance Scheme

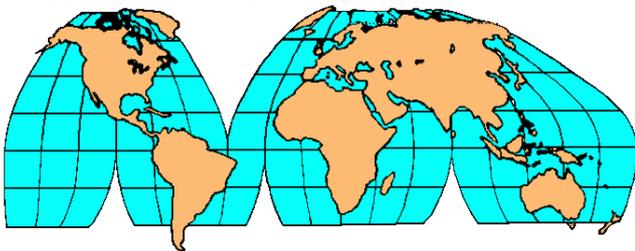
The CSG Student Research Assistance Scheme (SRAS; <http://www.iucncsg.org/pages/General-Information.html>) provided funding to 5 students in the October-December 2014 quarter.

1. Jose Avila Cervantes (Canada): Genetic and phenotypic divergence of allopatric populations of *Crocodylus acutus* in Mexico.

2. **Ciro Humboldt Paputsachis (Bolivia):** Home range and habitat use by *Caiman yacare* and *Melanosuchus niger* in the Tacana Indigenous Territory, Bolivia.
3. **Edna Paola Niño Mahecha (Colombia):** Ethno-zoological relationships and population ecology of *Crocodylus acutus* and *Caiman crocodilus fuscus* in Alto Magdalena, Colombia.
4. **Gregory Skupien (USA):** Identification of endocrine-disrupting compounds in the American alligator (*Alligator mississippiensis*) on Jekyll Island, Georgia.
5. **Asghar Mobaraki (Iran):** Sustainable management and conservation of Mugger crocodile (*Crocodylus palustris*) in Iran.

Tom Dacey, *CSG Executive Officer*, <csg@wmi.com.au>.

Regional Reports



North America

USA

CSG MEMBER RECEIVES LIFETIME ACHIEVEMENT AWARD FROM THE WILDLIFE SOCIETY. On 12 November 2014 Phil Wilkinson received the Lifetime Achievement Award from The Wildlife Society (South Carolina Chapter) at the group's annual meeting in Florence, South Carolina (SC), USA. Phil received the award "to recognize his important contributions and achievements as a wildlife biologist, researcher, conservationist, and mentor made in the art and science of wildlife management".



Phil earned a BS in Business from the University of South Carolina in 1959 and an MS in Wildlife Management from Auburn University in 1962. In 1962 he was hired by the South Carolina Department of Natural Resources (SCDNR; then SC Wildlife and Marine Resources Department) as the first resident biologist at Dirleton (later named the Samworth Wildlife Management Area).

In 1965, through an agreement with Mr. Thomas A. Yawkey and SCDNR, Phil became the first resident biologist on South Island where he was in charge of wildlife management efforts until after Mr. Yawkey's death in 1976. The property would later become the Tom Yawkey Wildlife Center Heritage Preserve (YWC), and Phil was instrumental in Mr. Yawkey's decision to leave this property to the state of South Carolina instead of the United States, which Mr. Yawkey was considering at the time. During Phil's 12 years as lead biologist at YWC, other biologists from that time recounted first-hand the gradual but tremendous transformation in both the condition and numbers of wildlife, particularly deer, turkey, and waterfowl that had never been present in such numbers or so healthy prior to that time.

Following his tenure at YWC, Phil was transferred to the SCDNR Heritage Trust Section and charged with developing and implementing biological research and surveys of non-game and endangered species in coastal South Carolina including the loggerhead sea turtle and American alligator as well as numerous species of seabirds and shorebirds. In addition to providing critical information on the population status of these species in SC, this work led to the development of multiple survey and sampling techniques that today are widely used in SC, other states, and abroad. Phil published many scientific and popular articles on SC wildlife, including the first nesting records of Forster's and Sooty Terns in SC.

Despite his diverse background, Phil's greatest contributions have been in alligator biology and management. He studied this species in SC from the late 1970s until his retirement in 1998, and has continued this work through the present day (16 years after retirement). His 1983 report, "Nesting Ecology of the American Alligator in Coastal South Carolina" is considered a seminal work in crocodylian biology, and he is internationally recognized as a crocodylian expert. As a member of the Crocodile Specialist Group (CSG), Phil has attended and presented papers at professional meetings in Venezuela, Argentina, Thailand, Mexico, Papua New Guinea, Australia, Zimbabwe, Cuba, Bolivia, and Brazil. In May 2014 Phil spoke at an international CSG conference in Lake Charles, LA, on his current alligator work at YWC which now includes collaborators from SCDNR, U.S. Fish and Wildlife Service, the Medical University of South Carolina, Hollings Marine Laboratory, NASA, and Savannah River Ecology Laboratory, among others. Phil is a legendary mentor, and as a result wildlife biologists and managers from across the USA and other countries including Argentina, Australia, Brazil, India, Japan, Myanmar, and South Africa, have journeyed to YWC to work with and learn from Phil.

In addition to his alligator work, Phil also conducted twice-

monthly seabird and shorebird surveys on Cape Romain National Wildlife Refuge (SC) for three years following retirement. He also served on the SCDNR Commissions' Waterfowl Advisory Committee for 11 years.

In 1999, Phil received the Daughters of the American Revolution (DAR) National Conservationist Award, and in 2012 he was honored by SC Governor Nikki R. Haley with the Order of the Silver Crescent, the state's highest award for an individual's commitment to the community, in recognition of his work as a pioneer in the field of wildlife biology and management in SC.

CROCODILE SYMPOSIUM. As part of the annual meeting of The Society for Integrative and Comparative Biology in West Palm Beach, Florida (3-7 January 2015), a symposium entitled "Integrative Biology of the Crocodylia" was held on 4 January. The symposium was organized and moderated by Dr. Val Lance, and was a great success. Numerous CSG members gave presentations or attended the talks, which were well received. Each speaker received several questions and lively discussions followed each presentation.

The first speaker was Chris Brochu, who discussed neosuchian phylogeny and the origin of the Crocodylia, followed by Bhart-Anjan Bhullar discussing the origin and development of craniofacial divergence between crocodiles and birds. Mike Pritz then spoke on the evolution and development of the crocodylian forebrain, followed by Travis Glenn discussing tetrapod genomes and slow evolution of crocodylian genomes, including his recent elegant publication with collaborators in the journal "Science". After a break the morning session

continued with Colleen Farmer discussing her research on evolution of unidirectional pulmonary airflow, and Sarah Keenan spoke on her PhD work (completed in May 2014) on microbial flora of the American alligator within different segments of the gastrointestinal tract.

After the lunch break, the afternoon session began with Miryam Venegas-Anaya discussing her team's work on the American crocodile and marine-coastal habitats, after which Chris Tracy reviewed his research on digestive morphology and gut physiology in alligators and crocodiles. Following this Jeff Lang reviewed his findings on the behavioral ecology of the Gharial, and the final presentation was by Thomas Rainwater, discussing long-term recaptures in American alligator and evidence of determinate growth.

The following afternoon, a complementary session with five additional crocodylian research talks was held; details on these talks and posters are available at the SICB's web site at www.sicb.org as are all abstracts from the presentations.

We were pleased numerous CSG members could attend and participate. It was gratifying to see a former SRAS recipient (Mr. Adam Parlin) in the audience; he received a CSG SRAS grant while an undergraduate and has now initiated his graduate school work. We thank the SICB organizing committee for assistance with the crocodylian symposium, and in particular thank Dr. Val Lance for his efforts in coordinating the excellent session.

Dr. Ruth Elsey, *Louisiana Department of Wildlife and Fisheries, Rockefeller Wildlife Refuge, Grand Chenier, Louisiana 70643, USA, <relsey@wlf.la.gov>*.



Figure 1. Some of the Symposium participants. Back row, left to right: Mike Pritz, Adam Pritchard, Travis Glenn, Chris Brochu, Miryam Venegas-Anaya, Sarah Keenan, Colleen Farmer, Val Lance, Jeff Lang. Front row, left to right: Ruth Elsey, Chris Tracy, Bhart-Anjan Bhullar, Thomas Rainwater. Photograph: Kent Vliet.

Latin America and the Caribbean

Colombia

FIRST INTERNATIONAL COURSE ON CROCODYLIAN MANAGEMENT, COLOMBIA. The 1st International Course on Crocodilian Management (1er Curso Internacional de Manejo Especializado de Crocodylianos) took place in Girardot and Melgar (municipalities of Cundinamarca Department) and Prado (Tolima Department), Colombia, on 16-21 November 2014.

The course was organized by Grupo de Investigación en Biodiversidad y Dinámica de Ecosistemas Tropicales (GIBDET), supported by Programa de Doctorado en Planificación y Manejo Ambiental de Cuencas Hidrográficas (PMACH) of the University of Tolima, with the participation of: Unidad de Manejo Ambiental (UMA) Reptilario Cipactli of Centro Universitario de la Costa de la Universidad de Guadalajara; Texas Tech University; Asociación Civil Conservación, Manejo y Aprovechamiento Sustentable de la Flora y Fauna Silvestre (COMAFFAS AC); Centro de Investigaciones Científicas y Transferencia Técnica a la Producción (CICYTTP-CONICET); Fundación para la conservación de caimanes; cocodrilos de Colombia (FunCroco); and, Estación de Biología Tropical “Roberto Franco” of Universidad Nacional and Fundación Palmarito. Strategic alliances were also made with regional companies such as Colsubsidio and Comfenalco, obtaining the best places for the carrying out the theoretical-practical course on crocodilians under both *in situ* and *ex situ* conditions.

The course had 9 instructors who work with crocodilians at national and international levels: Dr. Helios Hernández Hurtado, Dr. Pablo S. Hernández Hurtado, Jerónimo Domínguez Laso (México), Dr. Carlos Piña (Argentina), Álvaro Velasco (Venezuela), Dr. Sergio Balaguera-Reina, Dr. Cristina Mora-Rivera, Sergio Medrano, Willington Martínez Barreto (Colombia). The participation of Robín Andrés Poches, M.V. Carlos Morales, Leonardo Fabio Ordoñez and Paulo Jose Murillo, and the technical support of employees of Piscilago Zoo and David Ossa and M.V. Juliana Gaviria, was greatly appreciated. Finally, logistic support was coordinated by Lorena Carvajal Bonilla and Sigifredo Clavijo.

The event was attended by participants from different Latin-American countries [Brazil (3), Bolivia (1), Chile (2), México (3), Perú (1)] and different parts of Colombia [Barranquilla (2), Bogotá (1), Cali (1), Ibagué (3), Medellín (1), Montería (1), Pereira (1), Santa Marta (2), Tolú (1), Villavicencio (1)].

With the goal of providing theoretical and practical knowledge about crocodilians, the methodology involved participatory lectures and working groups. The themes were: origin, evolution and classification of crocodilians, anatomy and physiology, biology, ecology and crocodile importance (social-cultural, economic and environmental), crocodilian aquaculture and breeding, legislation, protection and conservation, human-crocodile conflict, capture,

manipulation and management, tools (telemetry and drones), experiences and work with crocodilians, data collection (*ex situ* and *in situ*) and data analysis. Lectures were held at the Peñalisa Hotel (Girardot) and Tomogó Hotel (Prado).

Ex situ activities were undertaken at Piscilago Zoo (Melgar). The first management demonstration was with an adult Black caiman (*Melanosuchus niger*), to demonstrate different capture techniques, clinical examination and routine morphometrics. Afterwards, participants had their own experience with four babillas (*Caiman crocodilus fuscus*). Participants also made clinical examination on six adult Orinoco crocodiles (*Crocodylus intermedius*) in order to observe their health conditions and update medical records.



In situ practicals on capture and babilla management was undertaken with animals in an artificial lake in the Forest Hotel del Athan (Girardot) taking the respective individual morphometric measures, and collection of DNA samples to study genetic characterization and isolation of molecular markers for species that leads Minambiente and the Institute of Genetics at the Universidad Nacional.

Finally, spotlight surveys were carried out on the caiman population at the Prado River Hydroelectric Dam, where the numbers, sizes and distribution of animals were recorded, and some caiman captured to provide data for subsequent analysis by participants in workshops.



With the completion of this course, the instructors of the course concluded that these initiatives are highly important for the region since generate a feedback process through the exchange of knowledge about crocodylians among all participants allowing a highly productive experience. In the case of Colombia the course allowed the formation of human resources (theoretical and practical) contributing to the Colombian process of changing production system from captive breeding to ranching programs.

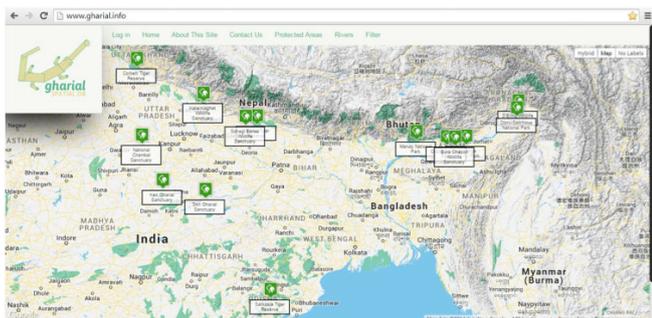


Cristina Mora-Rivera, *Coordinator of project, Grupo de Investigación en Biodiversidad y Dinámica de Ecosistemas Tropicales (GIBDET), Universidad del Tolima. Ibagué, Tolima, Colombia, <ncmorar@ut.edu.co>*.

South Asia and Iran

India

GHARIAL SPATIAL DATABASE. A spatial database (<http://gharial.info>) has been created for the Gharial (*Gavialis gangeticus*). In essence, this is a GIS-based project that displays detailed information on the current and past distribution of the species.



The project originated from the work done by John Thorbjarnarson, Frank Mazzotti and colleagues on *Crocodylus acutus*. The aim was to map precise locality data for Gharials across their range in India, based on available survey data. Using GIS technology, independent layers of information on threats and sightings have been produced. The website

that results allows registered users to add future survey information to the database to ensure it remains current.

Establishing this database is important as it provides a potent mechanism for future conservation actions for the species. Although there is a good understanding of the distribution, threats and locations of the most significant populations right now, the database provides a very clear and visual overview of these factors. This allows for better coordination in planning of surveys and projects from different organisations, a continuous mapping of the species and the threats into the future, a visual aid to secure grants from international donors, and is a great tool for awareness for the species regardless of the language of the user.

An important use for the project is for Government, Forest Department personnel, and other authorities within the range states. The database provides authorities with a very clear picture that they can use for establishing priority areas for enforcement, surveys, and in assessing permit applications.

Despite much being already known about Gharial distribution and the threats, little momentum has really been generated in conservation efforts across the species' range. Projects for the species remain independent and isolated. Part of the reason for this is that the information on the species remains largely with the experts in India and Nepal. By collating survey data and allowing international researchers access to this material, a better mechanism for setting conservation and research priorities can be achieved. This was one of the key recommendations of the Gharial Species Recovery Plan that resulted from a workshop in 2009.

The use of the database is hopefully quite straight-forward. The menu options allow for selection of either individual rivers, or protected areas to view. The 'Filter' option allows the user to select individual threat categories, states, rivers, or protected areas (or all sightings), as well as the years you wish to narrow a search down by.

This project was initiated by the Gharial Conservation Alliance, and was fully funded by San Diego Zoo (special thanks to Kim Lovich for her support).

Shakthi Sritharan (shakthi.sritharan@gmail.com) and Colin Stevenson.

NOTE ON A SERIOUSLY INJURED MALE MUGGER (*CROCODYLUS PALUSTRIS*) SURVIVING IN CAPTIVITY. The Sayaji Baug Zoo (SBZ) is one of the oldest zoos in India. SBZ was established in 1879 by Shrimant Maharaja Sayajirao Gaikwad III, the ruler of Vadodara State, with the private collection of wild animals handed over to Vadodara Municipal Corporation after India's independence. Presently the zoo functions under the rules and regulations of the Central Zoo Authority (Environment and Forest Ministry, Government of India, New Delhi). Situated on the banks of the Vishwamitri River at Vadodara, Gujarat State, SBZ has a natural population of Muggers around it (Vyas 2010).

Today, SBZ is considered to be a medium-sized zoo, housing about 874 vertebrates (79 species), including 7 species of native reptiles. Two pairs of adult Muggers are held in a semi-natural enclosure (26 x 48 m), open to the sky, with large trees and a few bamboo clumps (*Dendrocalamus strictus*) providing cover, and a water pool.

In western India, March and April are the hottest months of the year, and also mark the beginning of the breeding season for Muggers (Vyas 2012). On 25 April 2010, soon after nightfall, the two adult male Muggers were recorded to be fighting. After a lengthy fight, the animals settled down. The smaller male (2.58 m TL) had inflicted serious injuries on the larger male (3.15 m TL), the most obvious of which was to the upper jaw, which was barely attached by pieces of skin (Fig. 1). The large male lost its dominance over the water pool, and was forced into a small puddle of water in the corner of the enclosure.



Figure 1. Head of male Muger showing injured upper jaw, barely attached with some remnant pieces of skin.

The following day, as zoo staff were catching the injured male for transfer into another enclosure for medical treatment, the anterior portion of the upper jaw detached (Fig. 2) and was lost in the large water pool. Close examination of the injured male revealed a number of other injuries, but most of them were superficial, including scars on the belly, lower jaw and the dorsal region. All injuries were cleaned and disinfected periodically with Betadine (Providone-iodine solution) and the wounds covered with turmeric (*Curcuma longa*) powder. Special care was taken while feeding the animal, and to ensure no infestation of flies, parasites or maggots. Within 8-10 weeks, the animal's injuries had totally healed, and it had recovered health. The exposed bones of the upper jaw were completely covered with "new" soft tissue and skin after some 5-6 weeks.



Figure 2. Top: Zoo staff in action, catching injured male Muger. Bottom: Upper top jaw detached.

The injury and subsequent healing resulted in the loss of the nostrils and closure of the nasal passages (Fig. 3). As a consequence, the animal can only breathe through its mouth, and following periods of being underwater, must bring its head completely out of the water to breathe. It is now maintained alone in an off display enclosure at the zoo. Crocodylians are known to survive horrific injuries (eg amputation of limbs), but it is unclear whether a wild crocodile with a serious injury like this would have survived without medical intervention.



Figure 3. View of healed upper jaw.

Acknowledgements

I am very thankful to Dr. C.B. Patel (Zoo Veterinary) and other SBZ staff for help, treatments and special care provided

to the injured animal.

Literature Cited

Vyas, R. (2010). The Muggers (*Crocodylus palustris*) of Vishwamitri River: Past and Present. Report. Herpetology & Environmental Research Project (HERP): Vadodara, India.

Vyas, R. (2012). Current status of Marsh Crocodiles *Crocodylus palustris* (Reptilia: Crocodylidae) in Vishwamitri River, Vadodara City, Gujarat, India. *Journal of Threatened Taxa* 4(14): 3333-3341.

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CROC-ING IN CORBETT. Ongoing surveys in Corbett National Park (CNP) areas of the Corbett Tiger Reserve between 2012 and 2014 indicate a staggering 48.5% increase in adult Gharial (*Gavialis gangeticus*) numbers in the National Park areas of the Reserve since 2008, with present figures pegged at 52 adults (including 13 adult males) for the National Park. Surveys during 2008 recorded 35 adults in CNP, and 42 adults (including 10 adult males) for the entire Reserve (CNP and Sonanadi Wildlife Sanctuary).

However, a decline in the non-breeding juvenile and sub-adult population was also documented in CNP during this 6-year period and may be attributed to a number of natural factors, which include primarily the turnover of sub-adults to adults and low recruitment rates to the non-breeding cohorts (possibly due to predation), thereby calling for the development of conservation initiatives to maintain this viable Gharial population within the Reserve.

The largest Gharial population in the National Park is located at Dhikala, which is the confluence of the Ramganga River with Kalagarh Reservoir. The adult population here grew by 77.8% during this period along with population growth in adult gharial numbers also recorded at Boksar and the Ramganga River in Corbett NP.

Studies on the Gharial population in CNP use a combination of foot surveys, boat surveys along the shoreline, timelapse trail cameras and stationary counts from vantage points. Gharial also hatched in large numbers in Boksar (CNP) in 2013 and 2014, with approximately 600 hatchlings recorded during post-hatching surveys over these two seasons, while one-year-old Gharial recorded at Dhikala indicate that nesting does occur in the area.

The project is being undertaken by Subir Mario Chowfin in partnership with Dr. Alison Leslie, CSG Regional Vice Chair for Southern and East Africa, as the Scientific Advisor. Project collaborators are the Uttarakhand Forest Department and Corbett Tiger Reserve. The Gadoli and Manda Khal Wildlife Conservation Trust is the supporting NGO with The

University of Stellenbosch being a collaborating institution.

The project has also been expanded to include studies on nesting ecology and habitat utilization of the species in the National Park. Together with surveys of Gharial, studies have also been initiated to estimate the Mugger (*Crocodylus palustris*) population and document the turtle diversity in Corbett National Park as a part of the project.



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Science



Recent Publications

Sommerlad, R., Karbe, D., Van der Straeten, K., Rauhaus, A. and Ziegler, T. (2014). Krokodile. Pp. 84-135 in *Sachkunde Gefährliche Reptilien*. VDA/DGHT Sachkunde GbR: Mannheim. [Crocodile. Pp. 84-135 in *Competency Dangerous Reptiles*]. In German.

Allen, V., Molnar, J., Parker, W., Pollard, A., Nolan, G. and Hutchinson, J.R. (2014). Comparative architectural properties of limb muscles in Crocodylidae and Alligatoridae and their relevance to divergent use of asymmetrical gaits in extant Crocodylia. *J. Anat.* 225(6): 569-582.

Abstract: Crocodiles and their kin (Crocodylidae) use asymmetrical (bounding and galloping) gaits when moving rapidly. Despite being morphologically and ecologically similar, it seems alligators and their kin (Alligatoridae) do not. To investigate a possible anatomical basis for this apparent major difference in locomotor capabilities, we measured relative masses and internal architecture (fascicle lengths and physiological cross-sectional areas) of muscles of the pectoral and pelvic limbs of 40 individuals from 6 representative species of Crocodylidae and Alligatoridae. We found that, relative to body mass, Crocodylidae have significantly longer muscle fascicles (increased working range), particularly in the pectoral limb, and generally smaller muscle physiological cross-sectional areas (decreased force-exerting capability) than Alligatoridae. We therefore hypothesise that the ability of some crocodylians to use asymmetrical gaits may be limited more by the ability to make large, rapid limb motions

(especially in the pectoral limb) than the ability to exert large limb forces. Furthermore, analysis of scaling patterns in muscle properties shows that limb anatomy in the two clades becomes more divergent during ontogeny. Limb muscle masses, fascicle lengths and physiological cross-sectional areas scale with significantly larger coefficients in Crocodylidae than Alligatoridae. This combination of factors suggests that inter-clade disparity in maximal limb power is highest in adult animals. Therefore, despite their apparent morphological similarities, both mean values and scaling patterns suggest that considerable diversity exists in the locomotor apparatus of extant Crocodylia.

Phosri, S., Mahakunakorn, P., Lueangsakulthai, J., Jangpromma, N., Swatsitang, P., Daduang, S., Dhiravisit, A. and Thammasirirak, S. (2014). An investigation of antioxidant and anti-inflammatory activities from blood components of crocodile (*Crocodylus siamensis*). Protein J. 33(5): 484-492.

Abstract: Antioxidant and anti-inflammatory activities were found from *Crocodylus siamensis* blood. The 2,2'-azino-bis(3-ethylbenzthiazoline-6-sulfonic acid) (ABTS) radical scavenging, nitric oxide scavenging, hydroxyl radical scavenging and linoleic peroxidation assays were used to investigate the antioxidant activities of the crocodile blood. Results show that crocodile blood components had antioxidant activity, especially hemoglobin (40.58% nitric oxide radical inhibition), crude leukocyte extract (78% linoleic peroxidation inhibition) and plasma (57.27% hydroxyl radical inhibition). Additionally, the anti-inflammatory activity of the crocodile blood was studied using murine macrophage (RAW 264.7) as a model. The results show that hemoglobin, crude leukocyte extract and plasma were not toxic to RAW 264.7 cells. Also they showed anti-inflammatory activity by reduced nitric oxide (NO) and interleukin 6 (IL-6) productions from lipopolysaccharide (LPS)-stimulated cells. The NO inhibition percentages of hemoglobin, crude leukocyte extract and plasma were 31.9, 48.24 and 44.27%, respectively. However, only crude leukocyte extract could inhibit IL-6 production. So, the results of this research directly indicate that hemoglobin, crude leukocyte extract and plasma of *C. siamensis* blood provide both antioxidant and anti-inflammatory activities, which could be used as a supplementary agent in pharmaceutical products.

Trillanes, C.E., Pérez-Jiménez, J.C., Rosiles-Martínez, R. and González-Jáuregui, M. (2014). Metals in the caudal scutes of Morelet's crocodile (*Crocodylus moreletii*) from the southern Gulf of Mexico. Bull. Environ. Contam. Toxicol. 93(4): 423-428.

Abstract: Caudal scutes were collected from 92 Morelet's crocodiles (*Crocodylus moreletii*) between May and August 2012, in three Wildlife Management and Use Units (UMAs, from its name in Spanish) and three wild sites in Campeche, Mexico. The UMAs are intensive, with an ex situ approach to manage crocodiles in captivity. The concentrations of arsenic, mercury, lead, nickel, cadmium and chromium were analyzed

in each sample. As and Pb were detected in all samples, Hg in 86 and Ni in 74. The metal concentrations estimated were higher than most of the concentrations reported for skin, tail tip and caudal scutes in other crocodylians around the world. The concentration of As, Pb and Ni was significantly greater in the free-ranging groups than in the captive groups in UMAs ($p < 0.05$). Negative linear relationship was estimated between the snout-vent length and the concentration of Pb (in 5 groups) and Ni (in 3 groups). In this region *C. moreletii* is exposed to metals contamination and more studies are necessary to establish if represents a risk to their populations.

Bashyal, A., Gross, B.A., Venegas-Anaya, M., Lowrance, F. and Densmore, III, L.D. (2014). Assessment of microsatellites in estimating inter- and intraspecific variation among Neotropical *Crocodylus* species. Genet. Mol. Res. 13(3): 5492-5502.

Abstract: We tested microsatellites that were developed for the saltwater crocodile (*Crocodylus porosus*) for cross-species amplification and to provide an estimate of inter- and intraspecific variation among four species of Neotropical crocodiles (*C. rhombifer*, *C. intermedius*, *C. acutus* and *C. moreletii*). Our results indicated that with the exception of 2 loci in *C. intermedius*, all 10 microsatellite loci were successfully amplified in the 4 species, producing a set of variably sized alleles that ranged in number between 2 and 14 alleles per locus. Similarly, private alleles (ie unique alleles) also were reported in all 4 species for at least 3 loci. The mean observed and expected heterozygosities (averaged across species for all 10 loci combined) ranged from 0.39 to 0.77 and from 0.44 to 0.78, respectively. In addition to this, we evaluated these microsatellites in 2 populations of *C. acutus* and *C. moreletii* to assess their utility in estimating intraspecific levels of polymorphisms. These microsatellites also showed considerable allelic variation in population level analysis. The set of 10 microsatellite loci in our study had the potential to be used as a tool in population and conservation genetic studies of Neotropical crocodiles.

Milián-García, Y., Ramos-Targarona, R., Pérez-Fleitas, E., Sosa-Rodríguez, G., Guerra-Manchena, L., Alonso-Tabet, M., Espinosa-López, G. and Russello, M.A. (2014). Genetic evidence of hybridization between the critically endangered Cuban crocodile and the American crocodile: implications for population history and *in situ/ex situ* conservation. Heredity (Edinb.). (doi: 10.1038/hdy.2014.96).

Abstract: Inter-specific hybridization may be especially detrimental when one species is extremely rare and the other is abundant owing to the potential for genetic swamping. The Cuban crocodile (*Crocodylus rhombifer*) is a critically endangered island endemic largely restricted to Zapata Swamp, where it is sympatric with the widespread American crocodile (*C. acutus*). An on-island, *C. rhombifer* captive breeding program is underway with the goals of maintaining taxonomic integrity and providing a source of individuals for reintroduction, but its conservation value is limited by lack

of genetic information. Here we collected mtDNA haplotypic and nuclear genotypic data from wild and captive *C. rhombifer* and *C. acutus* in Cuba to: (1) investigate the degree of inter-specific hybridization in natural (*in situ*) and captive (*ex situ*) populations; (2) quantify the extent, distribution and *in situ* representation of genetic variation *ex situ*; and (3) reconstruct founder relatedness to inform management. We found high levels of hybridization in the wild (49.1%) and captivity (16.1%), and additional evidence for a cryptic lineage of *C. acutus* in the Antilles. We detected marginally higher observed heterozygosity and allelic diversity *ex situ* relative to the wild population, with captive *C. rhombifer* exhibiting over twice the frequency of private alleles. Although mean relatedness was high in captivity, we identified 37 genetically important individuals that possessed individual mean kinship (MK) values lower than the population MK. Overall, these results will guide long-term conservation management of Cuban crocodiles for maintaining the genetic integrity and viability of this species of high global conservation value.

Buenfil-Rojas, A.M., Alvarez-Legorreta, T. and Cedeño-Vázquez, J.R. (2014). Metals and metallothioneins in Morelet's crocodile (*Crocodylus moreletii*) from a transboundary river between Mexico and Belize. Arch. Environ. Contam. Toxicol.

Abstract: The aim of this study was to determine concentrations of heavy metals (cadmium [Cd] and mercury [Hg]) and metallothioneins (MTs) in blood plasma and caudal scutes of Morelet's crocodile (*Crocodylus moreletii*) from Rio Hondo, a river and natural border between Mexico and Belize. Three transects of the river (approximately 20 km each) were surveyed in September 2012 and April 2013, and samples were collected from 24 crocodiles from these areas. In blood plasma, Cd (7.6 ± 9.6 ng/ml) was detected in 69% of samples (n= 9); Hg (12.2 ± 9.2 ng/ml) was detected in 46% of samples (n= 6); and MTs ($10,900 \pm 9400$ ng/ml) were detected in 92% of samples (n= 12). In caudal scutes samples, Cd (31.7 ± 39.4 ng/g) was detected in 84% of samples (n= 12) and Hg (374.1 ± 429.4 ng/g) in 83% of samples (n= 20). No MTs were detected in caudal scutes. Hg concentrations in scutes from the Rio Hondo were 2- to 5-fold greater than those previously reported in scutes from other localities in northern Belize. In blood plasma, a significant positive relationship between Hg and body size was observed. Mean concentrations of Cd and MTs in size classes suggest that MTs may be related to Cd exposure. This is the first report of MT presence in crocodile blood.

Eme, J. and Crossley, II., D.A. (2014). Chronic hypercapnic incubation increases relative organ growth and reduces blood pressure of embryonic American alligator (*Alligator mississippiensis*). Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology 182: 53-57.

Abstract: Reptilian nests can experience natural hypoxic and hypercapnic conditions. We incubated alligator eggs at a female-only producing temperature (30°C) in three conditions: 21% O₂/0.04% CO₂, 21% O₂/3.5% CO₂ and 21% O₂/7% CO₂. Alligator embryos chronically incubated in high

CO₂ were markedly hypotensive (blood pressure reduced by 46%) and had relatively (mass-specific) enlarged hearts (dry mass increased by 20%), lungs (dry mass increased by 17%), and kidneys (dry mass increased by 14%). This study is the first to chronically incubate reptilian eggs in hypercapnia and suggests that high CO₂ alters the cardiovascular phenotype of alligator embryos (low blood pressure, relatively enlarged hearts), as well as the relative size of the organs primarily responsible for acid base balance, lungs and kidneys. The lungs and kidneys are largely non-functional during embryonic development, and the embryonic phenotype of increased relative mass may be a predictive-adaptation to metabolic or respiratory acidosis, such as during exercise or high respiratory CO₂. This study demonstrates that phenotypic plasticity of alligator embryos incubated in high CO₂ may result in either preferential organ growth, or maintenance of organ growth with reduced somatic growth.

Lott, M.J., Hose, G.C., Isberg, S.R. and Power, M.L. (2014). Genetics and infection dynamics of *Paratrichosoma* sp. in farmed saltwater crocodiles (*Crocodylus porosus*). Parasitol. Res.

Abstract: *Paratrichosoma*-associated helminthiasis has been identified in saltwater crocodiles under intensive farming conditions. The development of sustainable integrated management practices is dependent on a detailed understanding of *Paratrichosoma* population genetics and infection dynamics. This study investigated the genetic relationships of *Paratrichosoma* sp in a population of commercially farmed saltwater crocodiles, *Crocodylus porosus*, in northern Australia. 18S ribosomal RNA gene sequence data were obtained from *Paratrichosoma* sp. eggs present in the epidermis of infected animals. A high level of genetic diversity was distributed within the *Paratrichosoma* sp. population (241 variable positions in the 1094 bp alignment), indicating an accelerated rate of nucleotide base-pair substitutions in this genus of nematodes. Several possible environmental correlates of the incidence and intensity of helminthiasis, including season, rainfall, and mean monthly temperature, were investigated by visual inspection of crocodile skins. Stepwise logistic regression revealed a significant negative linear relationship (P= 0.011, R²=32.69%) between mean monthly rainfall and the incidence of monthly *Paratrichosoma*-associated helminthiasis. Variation in the severity of *Paratrichosoma*-associated helminthiasis could not be explained by any of the independent environmental variables included within an ordinal regression analysis. The large genetic diversity in these nematodes indicates a high probability of anthelmintic resistant alleles occurring in the population. We discuss how the spread of these alleles may be mitigated by adopting targeted treatment protocols.

Chong, A.Y., Kojima, K.K., Jurka, J., Ray, D.A., Smit, A.F.A., Isberg, S.R. and Gongora, J. (2014). Evolution and gene capture in ancient endogenous retroviruses - insights from the crocodylian genomes. Retrovirology 11: 71.

Venegas-Anaya, M., Escobedo-Galván, A.H., Balaguera-Reina, S.A., Lowrance, F., Sanjur, O.I. and Densmore III, L.D. (2014). Population ecology of American crocodile (*Crocodylus acutus*) in Coiba National Park, Panama. *Journal of Herpetology*.

Abstract: We conducted nocturnal surveys in the insular and coastal areas of Coiba National Park (CNP) and its mainland buffer zone in Panama (Chiriquí conservation site) from 2009-2012 to determine the conservation status of *Crocodylus acutus*. In 99 nights, we surveyed 147.2 km and captured 185 animals during nocturnal transects inspection with headlamps. Overall, sex ratio was 1.00:1.01 female/male with significant differences by size/age class and year. Females were slightly larger in total length than males (115.1 ± 56.9 cm - females, 105.4 ± 71.8 cm - males). The encounter rate was calculated based on number of animals captured per km of surveyed transect. The *C. acutus* encounter rate per year was 1.8 ind/km (60 ind/33.5 km/12 places visited) in 2009, 1.0 ind/km (90 ind/87.4 km/18 places visited) in 2010, and 1.3 ind/km (35 ind/26.3 km/8 places visited) in 2012. Based on our spatial analysis, the animals showed a dispersed pattern in most sites on CNP. Captured *C. acutus* were found in 581.1 km² total area within 78% natural habitat, including mangroves and beaches, and 22% disturbed habitat on both the mainland and the islands. In addition, the spatial analysis showed reduction in natural land cover; crocodile habitat showed limited conversion to agricultural land use; and we found correlation between crocodile population size and protected areas. The differences between mainland and island populations regarding ecology suggest that a long-term monitoring program for American crocodiles is necessary to distinguish between natural fluctuations and anthropogenic changes on population dynamics and conservation status.

Whiting, E.T. and Hastings, A.K. (2014). First fossil alligator from the Late Eocene of Nebraska and the Late Paleogene record of alligators in the Great Plains. *Journal of Herpetology*.

Abstract: Fragmentary fossils have been tentatively attributed to *Alligator* from the Eocene of Wyoming, North Dakota, Saskatchewan, and Utah. The earliest definitive temporal and geographic range of *Alligator* has been limited to the late Eocene and early Oligocene Chadron and Brule Formations of the White River Badlands in South Dakota, represented by fossils of *Alligator prenasalis*. New fossils from the Chadron Formation in northwestern Nebraska expand the definitive geographic range of *Alligator* during the late Eocene. Results from a cladistic analysis of 28 alligatoroid species using 181 morphological characters support the identification of the new specimens as representing the most basally divergent taxon within *Alligator*, suggesting that they likely represent *A. prenasalis*; however, a lack of diagnostic features in the Nebraska specimens prevents a definitive assignment to this species. Limited occurrence data may indicate that *Alligator* experienced a reduction in biogeographic range following global cooling around the Eocene-Oligocene transition, although this may be an artifact of low sample size for Oligocene alligators in the Great Plains.

Mašová, Š., Baruš, V., Seifertová, M., Malala, J. and Jirků, M. (2014). Redescription and molecular characterisation of *Dujardinascaris madagascariensis* and a note on *D. dujardini* (Nematoda: Heterocheilidae), parasites of *Crocodylus niloticus*, with a key to *Dujardinascaris* spp. in crocodylians. *Zootaxa* 3893(2): 261-276.

Abstract: An examination of one specimen of Nile crocodile, *Crocodylus niloticus* (Laurenti, 1768), from Lake Turkana (Kenya), revealed the presence of two ascaridoid nematodes belonging to the genus *Dujardinascaris* Baylis, 1947. *Dujardinascaris madagascariensis* Chabaud & Caballero, 1966 was studied by scanning electron microscopy, redescribed, and differentiated from *D. dujardini* (Travassos, 1920). *Dujardinascaris madagascariensis* is the second of the genus to be sequenced. An internal fragment of the small ribosomal subunit and nuclear ribosomal DNA internal transcribed spacer 2 region were amplified-the slowly evolving 18S gene region was used for phylogenetic analysis. Molecular data confirmed affinity of *D. madagascariensis* to the family Heterocheilidae and revealed its closest relationship with *D. waltoni*. A key to the species of *Dujardinascaris* parasitizing crocodiles is provided.

Trotteyn, M.J. and Ezcurra, M.D. (2014). Osteology of *Pseudochampsia ischigualastensis* gen. et comb. nov. (Archosauriformes: Proterochampsidae) from the Early Late Triassic Ischigualasto Formation of Northwestern Argentina. *PLoS One*. 9(11):e111388.

Abstract: Proterochampsids are crocodile-like, probably semi-aquatic, quadrupedal archosauriforms characterized by an elongated and dorsoventrally low skull. The group is endemic from the Middle-Late Triassic of South America. The most recently erected proterochampsid species is “*Chanaresuchus ischigualastensis*”, based on a single, fairly complete skeleton from the early Late Triassic Ischigualasto Formation of northwestern Argentina. We describe here in detail the non-braincase cranial and postcranial anatomy of this species and revisit its taxonomy and phylogenetic relationships. The phylogenetic analysis recovered ‘*Chanaresuchus ischigualastensis*’ as part of a trichotomy together with *Gualosuchus reigi* and *Chanaresuchus bonapartei*. Accordingly, “*Chanaresuchus ischigualastensis*” can be potentially more closely related to *Gualosuchus reigi*, or even *Rhadinosuchus gracilis*, than to *Chanaresuchus bonapartei*. In addition, after discussing previously claimed synapomorphies of *Chanaresuchus*, we could not find unambiguous support for the monophyly of the genus. As a result, we propose here the erection of the new genus *Pseudochampsia* for ‘*Chanaresuchus ischigualastensis*’, which results in the new combination *Pseudochampsia ischigualastensis*. The information provided here about the anatomy and taxonomy of *Pseudochampsia ischigualastensis* will be useful for future quantitative analyses focused on the biogeography and macroevolutionary history of proterochampsids.

Zhang, R., Zhang, S.Z., Li, E., Wang, C., Wang, C.L. and Wu, X.B. (2014). Full-length cDNA cloning and structural characterization of preproinsulin in *Alligator sinensis*. *Genet. Mol. Res.* 13(4): 8845-8855.

Abstract: Insulin is an important endocrine hormone that plays a critical physiological role in regulating metabolism and glucostasis in vertebrates. In this study, the complete cDNA of *Alligator sinensis* preproinsulin gene was cloned for the first time by reverse transcription-polymerase chain reaction and rapid amplification of cDNA ends methods; the amino acid sequence encoded and protein structure were analyzed. The full-length of preproinsulin cDNA sequence consists of 528 base pairs (bp), comprising a 34-bp 5'-untranslated region, a 170-bp 3'-untranslated region and an open reading frame that is 324 bp in length. The open reading frame encodes a 107-amino acid preproinsulin with a molecular weight of approximately 12,153.8 Da, theoretical isoelectric point of 5.68, aliphatic index of 92.06, and grand average of hydropathicity of -0.157, from which a signal peptide, a B-chain, a C-peptide, and an A-chain are derived. Online analysis suggested that the deduced preproinsulin amino acid sequence contains a transmembrane region, and that it has a signal peptide whose cleavage site occurs between alanine 24 and alanine 25. Comparative analysis of preproinsulin amino acid sequences indicated that the A-chain and B-chain sequences of preproinsulins are highly conserved between reptiles and birds, and that the preproinsulin amino acid sequence of *Alligator sinensis* shares 89% similarity to that of *Chelonia mydas*, but low similarity of 48-63% to those of mammals and fishes. The phylogenetic tree constructed using the neighbor-joining method revealed that preproinsulin of *Alligator sinensis* had high homology with reptiles and birds, such as *Chelonia mydas*, *Gallus gallus*, and *Columba livia*.

Smith, A.M., Ismail, H., Henton, M.M. and Keddy, K.H. (2014). Similarities between *Salmonella enteritidis* isolated from humans and captive wild animals in South Africa. *J. Infect. Dev. Ctries.* 8(12): 1615-1619.

Abstract: *Salmonella* is well recognized as an aetiological agent of gastrointestinal and diarrhoeal disease. *Salmonella enterica* serotype *enteritidis* (*Salmonella enteritidis*) is one of the commonest serotypes associated with foodborne illness. In South Africa, we compared *S. enteritidis* strains isolated from humans with gastroenteritis and strains isolated from captive wild animals, between June 2011 and July 2012. Bacteria were phenotypically characterized using standard microbiological techniques. Genotypic relatedness of isolates was investigated by pulsed-field gel electrophoresis (PFGE) analysis. A diversity of 27 PFGE patterns amongst 196 human non-invasive isolates was shown; two PFGE patterns predominated and accounted for 74% of all human isolates. Human isolates showed a 12% prevalence rate for nalidixic acid resistance. Animal isolates from 5 different sources were investigated. With the exception of an isolate from a ground hornbill, all animal isolates (jaguar, crocodile, lion and poultry) showed PFGE pattern matches to a human isolate. Animal isolates showed susceptibility to all antimicrobial

agents tested, with the exception of nalidixic acid resistance in isolates from the lion and poultry source. Our data showed similarities between *S. enteritidis* strains isolated from humans and captive wild animals, suggesting a probable common source for strains from humans and animals.

Jaratlerdsiri, W., Deakin, J., Godinez, R.M., Shan, X., Peterson, D.G., Marthey, S., Lyons, E., McCarthy, F.M., Isberg, S.R., Higgins, D.P., Chong, A.Y., John, J.S., Glenn, T.C., Ray, D.A. and Gongora, J. (2014). Comparative genome analyses reveal distinct structure in the saltwater crocodile MHC. *PLoS One.* 9(12): e114631.

Abstract: The major histocompatibility complex (MHC) is a dynamic genome region with an essential role in the adaptive immunity of vertebrates, especially antigen presentation. The MHC is generally divided into subregions (classes I, II and III) containing genes of similar function across species, but with different gene number and organisation. Crocodylia (crocodylians) are widely distributed and represent an evolutionary distinct group among higher vertebrates, but the genomic organisation of MHC within this lineage has been largely unexplored. Here, we studied the MHC region of the saltwater crocodile (*Crocodylus porosus*) and compared it with that of other taxa. We characterised genomic clusters encompassing MHC class I and class II genes in the saltwater crocodile based on sequencing of bacterial artificial chromosomes. Six gene clusters spanning around 452 kb were identified to contain nine MHC class I genes, six MHC class II genes, three TAP genes, and a TRIM gene. These MHC class I and class II genes were in separate scaffold regions and were greater in length (2-6 times longer) than their counterparts in well-studied fowl B loci, suggesting that the compaction of avian MHC occurred after the crocodylian-avian split. Comparative analyses between the saltwater crocodile MHC and that from the alligator and gharial showed large syntenic areas (>80% identity) with similar gene order. Comparisons with other vertebrates showed that the saltwater crocodile had MHC class I genes located along with TAP, consistent with birds studied. Linkage between MHC class I and TRIM39 observed in the saltwater crocodile resembled MHC in eutherians compared, but absent in avian MHC, suggesting that the saltwater crocodile MHC appears to have gene organisation intermediate between these two lineages. These observations suggest that the structure of the saltwater crocodile MHC, and other crocodylians, can help determine the MHC that was present in the ancestors of archosaurs.

Green, R.E., Braun, E.L., Armstrong, J., Earl, D., Nguyen, N., Hickey, G., Vandeweghe, M.W., St. John, J.A., Capella-Gutiérrez, S., Castoe, T.A., Kern, C., Fujita, M.K., Opazo, J.C., Jurka, J., Kojima, K.K., Caballero, J., Hubley, R.M., Smit, A.F., Platt, R.N., Lavoie, C.A., Ramakodi, M.P., Finger, J.W. Jr., Suh, A., Isberg, S.R., Miles, L., Chong, A.Y., Jaratlerdsiri, W., Gongora, J., Moran, C., Iriarte, A., McCormack, J., Burgess, S.C., Edwards, S.V., Lyons, E., Williams, C., Breen, M., Howard, J.T., Gresham, C.R., Peterson, D.G., Schmitz, J., Pollock, D.D., Haussler, D.,

Triplett, E.W., Zhang, G., Irie, N., Jarvis, E.D., Brochu, C.A., Schmidt, C.J., McCarthy, F.M., Faircloth, B.C., Hoffmann, F.G., Glenn, T.C., Gabaldón, T., Paten, B. and Ray, D.A. (2014). Three crocodylian genomes reveal ancestral patterns of evolution among archosaurs. *Science* 346(6215): 1254449.

Abstract: To provide context for the diversification of archosaurs - the group that includes crocodylians, dinosaurs, and birds - we generated draft genomes of three crocodylians: *Alligator mississippiensis* (American alligator), *Crocodylus porosus* (saltwater crocodile), and *Gavialis gangeticus* (Indian Gharial). We observed an exceptionally slow rate of genome evolution within crocodylians at all levels, including nucleotide substitutions, indels, transposable element content and movement, gene family evolution, and chromosomal synteny. When placed within the context of related taxa including birds and turtles, this suggests that the common ancestor of all of these taxa also exhibited slow genome evolution and that the comparatively rapid evolution is derived in birds. The data also provided the opportunity to analyze heterozygosity in crocodylians, which indicates a likely reduction in population size for all three taxa through the Pleistocene. Finally, these data combined with newly published bird genomes allowed us to reconstruct the partial genome of the common ancestor of archosaurs, thereby providing a tool to investigate the genetic starting material of crocodylians, birds, and dinosaurs.

Hedley, J. and Kubiak, M. (2014). Neurologic diseases of birds and reptiles. *J. Exotic Pet Med.* (<http://dx.doi.org/10.1053/j.jepm.2014.12.004>).

Abstract: Captive reptiles and birds are often presented to veterinary hospitals exhibiting neurologic disease signs. Neurological disease conditions can be a diagnostic challenge for the clinician due to limitations of laboratory testing, non-specific clinical signs, and the wide range of possible underlying disease causes of the identified problems. Both common and seldom reported neurologic diseases associated with infectious organisms, trauma, neoplasia, metabolic disorders, toxins, and malnutrition are reviewed.

Hayman, R.B., Harvey, R.G., Mazzotti, F.J., Israel, G.D. and Woodward, A.R. (2014). Who complains about alligators? Cognitive and situational factors influence behavior toward wildlife. *Human Dimensions of Wildlife: An International Journal* 19(6): 481-497.

Abstract: Understanding perceptual and situational factors underlying nuisance complaints can help managers maintain carnivore populations while mitigating conflicts with people. Our study uses data from a mail survey (N= 467 complainants about nuisance alligators, and N= 669 random Florida residents) and a three-step binary logistic regression analysis to examine how general attitudes, specific beliefs, and situational factors influence the behavior of reporting nuisance alligators. Residence adjacent to fresh water, the presence of outdoor pets, higher risk belief scores, higher

nuisance belief scores, higher education, and older age were all related to complaining about an alligator, whereas general attitude toward alligators was not. Results are consistent with the “specificity principle” for attitude-behavior correspondence and emphasize the importance of situational factors as behavioral determinants. Targeted harvest areas can help to manage complaints in marginal habitats where risk from alligators is persistent. Information about protective behaviors and benefits of alligators can motivate residents to avoid dangerous encounters.

Siroski, P.A., Russ, N.B., Ortega, H.H. and Formentini, E.A. (2014). In vitro evaluation of synergistic activity between ciprofloxacin and broad snouted caiman serum against *Escherichia coli*. *Research in Veterinary Science* (doi: [doi:10.1016/j.rvsc.2014.11.007](https://doi.org/10.1016/j.rvsc.2014.11.007)).

Abstract: The *in vitro* synergistic activity between ciprofloxacin and serum of broad snouted caiman on *Escherichia coli* was studied. The estimated MIC value of ciprofloxacin was 0.0188 µg/ml, and two assays of kill curve during 5 hours were performed: the first one in a standard culture medium and the second one in the presence of caiman serum. Different concentrations of ciprofloxacin were tested. Ciprofloxacin showed higher values of bacterial elimination rate in the presence of caiman serum in all concentrations tested. The combined activity of sub-inhibitory concentrations of ciprofloxacin and the humoral immune factors present in caiman serum determined an increase in the bacterial elimination observed in this assay. We suggest that the antibacterial activity of complement and natural antibodies present in caiman serum, which can bind to both Gram-negative and Gram-positive bacteria and acting through the classical complement pathway, can inhibit bacterial growth of *E. coli* by lysis.

Tineo, D.E., Bona, P., Pérez, L.M., Vergani, G.D., González, G., Poiré, D.G., Gasparini, Z.N. and Legarreta, P. (2015). Palaeoenvironmental implications of the giant crocodylian *Mourasuchus* (Alligatoridae, Caimaninae) in the Yecua Formation (late Miocene) of Bolivia. *Alcheringa* 39 (doi: [10.1080/03115518.2015.967162](https://doi.org/10.1080/03115518.2015.967162)).

Abstract: Outcrops of the Yecua Formation (late Miocene) are exposed for approximately 230 m along the La Angostura section of the Piraf River (50 km southwest of Santa Cruz de la Sierra). These reveal massive (argillic palaeosols) and laminated (quiet-water lacustrine and marsh settings) mudstones interbedded with thin sandstones containing microfossils, molluscs and vertebrate remains. Significantly, the succession hosts a giant crocodylian, *Mourasuchus* (Alligatoridae, Caimaninae), which is represented by both skull and postcranial fragments found in association with freshwater turtles and fishes. *Mourasuchus* was distributed widely from the middle Miocene of Colombia to upper Miocene of Venezuela, Brazil and Argentina, suggesting connections between major fluvial systems and an active mechanism for dispersal of South American freshwater

vertebrates during the Miocene.

Martin, J.E., Smith, T., De Lapparent de Broin, F., Escuillie, F. and Delfino, M. (2014). Late Palaeocene eusuchian remains from Mont de Berru, France, and the origin of the alligatoroid *Diplocynodon*. *Zoological Journal of the Linnean Society* 172(4): 867-891.

Abstract: Crocodylian remains from the late Palaeocene continental locality of Mont de Berru (Marne, France) offer the opportunity to reassess the taxonomic identity of the oldest diplocynodontid from Europe. Owing to significant morphological differences from previously described species of *Diplocynodon*, which include the presence of a splenial symphysis, a new species, *Diplocynodon remensis* sp. nov., is erected here. Its inclusion in a phylogenetic framework for Eusuchia leads to its positioning as a derived member of diplocynodontids. Diplocynodontidae are viewed as a basal alligatoroid taxon, and, because morphological affinities with the Late Cretaceous-early Eocene North American genus *Borealosuchus* were mentioned in earlier studies, a comparison amongst *D. remensis* sp. nov., *Leidyosuchus* and *Borealosuchus* spp. is presented. Although *D. remensis* sp. nov. is the geologically oldest diplocynodontid, according to our results, it is not the phylogenetically most primitive. Other morphological discrepancies are highlighted, indicating that the topology recovered here is only tentative. From a biogeographical point of view, the appearance of *Diplocynodon* in Europe prior to the Palaeocene/Eocene boundary indicates that it did not disperse with North American taxa that reached Europe around the time of the Palaeocene-Eocene thermal maximum (PETM). Therefore, a pre-PETM dispersal from North America at the same times as other vertebrates with clear North American affinities also occurring in the Palaeocene of Europe cannot be excluded. The description of *D. remensis* sp. nov. adds substantial new, albeit conflicting, information, highlighting the need for a better phylogenetic framework with a revision of other critical taxa (*Menatalligator*, *Borealosuchus*) from the Palaeocene of Europe and North America.

Fukuda, Y. and Saalfeld, K. (2014). Abundance of saltwater crocodile hatchlings is related to rainfall in the preceding wet season in northern Australia. *Herpetologica* 70(4): 439-448.

Abstract: The reproduction of Saltwater Crocodiles (*Crocodylus porosus*) is seasonal, coinciding with the wet season (October-May) in the Northern Territory, Australia. Understanding environmental factors influencing the population dynamics of *C. porosus* can have important implications for the management of this species. We examined the relationship between hatchling density and monthly rainfall in the preceding wet season using crocodile population data from standardized spotlight surveys in 10 tidal rivers from 1976 to 2012 and historical weather data. Hatchling densities in four rivers were negatively correlated with monthly rainfall around the peak of the wet season (January or February). Hatchling densities in four rivers were

positively correlated with rainfall after the peak months. Hatchling densities in 8 rivers were positively correlated with rainfall after the wet season peak (April or May). Our results support previous observations that: (1) the length of the wet season determines the extent of reproduction; and (2) flooding caused by heavy rains in the middle of the wet season leads to higher rates of egg mortality. These relationships could be used to estimate reproductive success from year to year, and to guide management decisions such as adjusting the harvest intensity of eggs and hatchlings.

Wang, H., Zhang, S., Zhou, N., Wang, C. and Wu, X. (2014). Distribution of endocrine cells in the digestive tract of *Alligator sinensis* during the active and hibernating period. *Tissue Cell*. 46(5): 343-51.

Abstract: The digestive tract is the largest endocrine organ in the body; the distribution pattern of endocrine cells varies with different pathological and physiological states. The aim of the present study was to investigate the distributed density of 5-hydroxytryptamine (5-HT), gastrin (GAS), somatostatin (SS) and vasoactive intestinal peptide (VIP) immunoreactive (IR) cells in the digestive tract of *Alligator sinensis* during the active and hibernating period by immunohistochemical (IHC) method. The results indicated that 5-HT-IR cells were distributed throughout the entire digestive tract, which were most predominant in duodenum and jejunum. The density increased significantly in stomach and duodenum during hibernation. GAS-IR cells were limited in small stomach and small intestine. The density decreased significantly in small stomach during hibernation, while increased in duodenum. What's more, most of the endocrine cells in duodenum were generally spindle shaped with long cytoplasmic processes ending in the lumen during hibernation. SS-IR cells were limited in stomach and small stomach. The density increased in stomach while decreased in small stomach during hibernation, meanwhile, fewer IR cells occurred in small intestine. VIP-IR cells occurred in stomach and small stomach. The density decreased in small stomach, while increased in stomach during hibernation. These results indicated that the endocrine cells in different parts of digestive tract varied differently during hibernation, their changes were adaptive response to the hibernation.

Fei, R., Tang, M., Wu, Y., Zhou, Y. and Lu, C. (2014). Hematology of three age groups of captive Chinese alligator during deep and late hibernation. *Comparative Clinical Pathology*.

Abstract: As the numbers and density of the farmed Chinese alligator (*Alligator sinensis*) increase, diseases are more common. Death often occurs during the recovery period from hibernation and this phenomenon is particularly prominent in juveniles. A practical way is therefore needed to monitor the health of the alligator. Research on hematology can provide an early warning and a rapid diagnosis of a disease and will help decrease mortality during recovery from hibernation. The aim of this study was to determine

the hematologic values of the Chinese alligator at different stages of hibernation. We collected 30 blood samples from each of three age groups (adults, sub-adults, juveniles) during deep and late hibernation. We obtained counts of red blood cells, thrombocytes, and total and different white blood cells using manual methods. The thrombocyte numbers in the three groups decreased significantly during late compared with deep hibernation. The white blood cell counts for the sub-adults and juveniles increased significantly during late hibernation, whereas this did not occur in the adults. The immunity of the sub-adults and juveniles clearly increased during late hibernation. The results suggest that, for breeding programs, environmental stress may be the cause of the higher mortality in the juveniles during recovery from hibernation

Goldstein, E.J.C. and Abrahamian, F.M. (2015). Animal bites and zoonoses: from A to Z: alligators to zebras. Pp. 659-679 in *Zoonoses - Infections Affecting Humans and Animals*, ed. by A. Sing. Springer-Science + Business Media: Dordrecht.

Abstract: Worldwide, animal bite-related injuries to humans are a common daily occurrence. Injuries can range from minor puncture wounds to extensive crush injuries and even amputations and death. Increasing population, continued spread of habitation, and the popularity of owning various types of both traditional and non-traditional pets has made it easier for humans to have contact with various types of animals. In general, the oral flora of the biting animal will be concordant with the bacteria isolated from the wound. These oral flora organisms often originate from the normal oral residents and environmental flora as well as the skin and intestinal bacteria of the animals food sources and prey.

Herrera, J., Solari, A. and Lucifora, L.O. (2014). Unanticipated effect of climate change on an aquatic top predator of the Atlantic rainforest. *Aquatic Conservation Marine and Freshwater Ecosystems* (doi: 10.1002/aqc.2536).

Abstract: From a biodiversity conservation viewpoint it is crucial to estimate the sensitivity of species and populations to climate change, especially of key species such as top predators. Here, the El Niño-Southern Oscillation phenomenon (ENSO) was used as a natural experiment to estimate the sensitivity of a population of the rainforest-dwelling broad-snouted caiman, *Caiman latirostris*, to extreme floods. Caiman abundance declined by 35% annually between 1996 and 1998, and then increased, without reaching 1996 levels, indicating a marked effect of the 1997 El Niño. Accordingly, the Southern Oscillation Index was positively correlated with caiman abundance, indicating lower caiman abundance with warm ENSO events. The relationship between the relative annual mean abundance of caimans and the maximum daily annual discharge of the Iguazú River was analysed. This relationship was parabolic, with caiman abundance increasing at discharges from 500 to 1500 m³ s⁻¹ and then decreasing at discharges from 1500 to 2500 m³ s⁻¹, indicating an adverse effect of both extreme low and high river discharge. No effect of illegal hunting was measurable. This

study evaluated whether the negative effect of extremely high water levels on caiman abundance could be due to a decrease in the availability of the habitat more commonly used by small (<60 cm total length, TL) and medium (60-120 cm TL) caimans. Small and medium-sized caimans used herbaceous/shrub habitats more frequently than large caimans (>120 cm TL), ie the type of habitat flooded during extreme floods. An increase in extreme floods, as forecast for the Atlantic rainforest owing to climate change, may seriously affect the population of rainforest caimans through the reduction of adequate habitat for juveniles. This counter-intuitive result, in which an excess of water reduces the abundance of an aquatic top predator, should be considered in conservation plans of rainforest-dwelling crocodylians.

Hanson, J.O., Salisbury, S.W., Campbell, H.A., Dwyer, R.G., Jardine, T.D. and Franklin, C.E. (2014). Feeding across the food web: The interaction between diet, movement and body size in estuarine crocodiles (*Crocodylus porosus*). *Austral Ecology* (doi: 10.1111/aec.12212).

Abstract: The estuarine crocodile (*Crocodylus porosus*) is an apex predator across freshwater, estuarine and coastal environments. The impact of a changing *C.porosus* population upon the ecosystem is unknown, but due to large ontogenetic changes in body mass (>1000-fold) their impact may be wide reaching and substantial. Here we investigated the relationship between diet, movement and body size in a population of *C.porosus* inhabiting a tidal river in northern Australia. Subcutaneous acoustic transmitters and fixed underwater receivers were used to determine the activity space and movement patterns of 42 individuals (202-451 cm TL). There was no size-related spatial partitioning among different sized crocodiles. Large individuals (snout-vent length (SVL): 160 cm<SVL<188.5 cm) did, however, exhibit a much larger activity space than other size classes. Diet and individual specialization was assessed using the composition of stable carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotopes in tissues with different turnover rates. There was a quadratic relationship between body size and $\delta^{15}N$, suggesting that medium-sized individuals (110 cm<SVL<160 cm) incorporated a greater proportion of high trophic prey into their diets than small (SVL<110 cm) or large individuals (SVL>160 cm). Tissue $\delta^{13}C$ composition on the other hand was positively correlated with body size, indicating that different size classes were trophically linked to primary producers in different habitats. Individual-level analyses showed that small crocodiles were generalist feeders while medium and large size classes specialized on particular prey items within the food webs they fed. The findings further our understanding of ontogenetic variation in *C.porosus* diet, and suggest that change in *C.porosus* population size or demographics may be influential at various levels across the local food web.

De Buffrenil, V., Clarac, F., Fau, M., Martin, S., Martin, B., Pelle, E. and Laurin, M. (2014). Differentiation and growth of bone ornamentation in vertebrates: A comparative histological study among the Crocodylomorpha. *J. Morphol.* (doi: 10.1002/jmor.20351).

Abstract: Bone ornamentation, that is, hollow (pits and grooves) or protruding (ridges) repetitive reliefs on the surface of dermal bones, is a frequent, though poorly studied and understood, feature in vertebrates. One of the most typical examples of this characteristic is given by the Crurotarsi, a taxon formed by the crocodylians and their closest allies, which generally display deep ornamentation on skull roof and osteoderms. However, the ontogenetic process responsible for the differentiation and development of this character remains controversial. This study was conducted to settle the question on histological and microanatomical evidence in several crurotarsan taxa. Observational and experimental data in extant and extinct crocodyliforms show that bone ornamentation is initially created, and later maintained during somatic growth (that is indefinite in crocodylians), by a complex process of bone remodeling comprising local resorption of superficial bone cortices, followed by partial reconstruction. The superficial reliefs of crocodylian dermal bones are thus permanently modified through pit enlargement, drift, stretching, shrinking, or complete filling. Ridges are also remodeled in corresponding ways. These processes allow accommodation of unitary ornamental motifs to the overall dimensions of the bones during growth. A parsimony optimization based on the results of this study, but integrating also published data on bone histology in non-crocodyliform crurotarsans and some non-crurotarsan taxa, suggests that the peculiar mechanism described above for creating and maintaining bone ornamentation is a general feature of the Crurotarsi and is quite distinct from that attributed by previous authors to other vertebrates.

Bloor, P., Ibanez, C. and Vilorio-Lagares, T.A. (2014). Mitochondrial DNA analysis reveals hidden genetic diversity in captive populations of the threatened American crocodile (*Crocodylus acutus*) in Colombia. *Ecology and Evolution* (doi: 10.1002/ece3.1307).

Abstract: Identification of units within species worthy of separate management consideration is an important area within conservation. Mitochondrial DNA (mtDNA) surveys can potentially contribute to this by identifying phylogenetic and population structure below the species level. The American crocodile (*Crocodylus acutus*) is broadly distributed throughout the Neotropics. Its numbers have been reduced severely with the species threatened throughout much of its distribution. In Colombia, the release of individuals from commercial captive populations has emerged as a possible conservation strategy that could contribute to species recovery. However, no studies have addressed levels of genetic differentiation or diversity within *C. acutus* in Colombia, thus complicating conservation and management decisions. Here, sequence variation was studied in mtDNA cytochrome b and cytochrome oxidase I gene sequences in three Colombian captive populations of *C. acutus*. Two distinct lineages were identified: *C. acutus*-I, corresponding to haplotypes from Colombia and closely related Central American haplotypes; and *C. acutus*-II, corresponding to all remaining haplotypes from Colombia. Comparison with findings from other studies indicates the presence of a single

“northern” lineage (corresponding to *C. acutus*-I) distributed from North America (southern Florida), through Central America and into northern South America. The absence of *C. acutus*-II haplotypes from North and Central America indicates that the *C. acutus*-II lineage probably represents a separate South American lineage. There appears to be sufficient divergence between lineages to suggest that they could represent two distinct evolutionary units. We suggest that this differentiation needs to be recognized for conservation purposes because it clearly contributes to the overall genetic diversity of the species. All Colombian captive populations included in this study contained a mixture of representatives of both lineages. As such, we recommend against the use of captive-bred individuals for conservation strategies until further genetic information is available.

Cherkiss, M.S., Mazzotti, F.J., Hord, L. and Aldecoa, M. (2014). Remarkable movements of an American crocodile (*Crocodylus acutus*) in Florida. *Southeastern Naturalist* 13(4): N52-N56.

Abstract: Here we present the remarkable movements of an individual *Crocodylus acutus* (American Crocodile) over a 14-year period. The crocodile was originally marked in Homestead, FL, as a young-of-the-year in 1999, and was later recaptured multiple times more than 388 km away along the southwest coast of Florida. After several relocations and numerous sightings, this individual who has become known as Yellow Number 1 was found back within the same canal system in which it was first captured.

Manolo, R.I. and Alcala, A.C. (2014). Conservation of the Philippine crocodile *Crocodylus mindorensis* (Schmidt 1935): *in situ* and *ex situ* measures. *International Zoo Yearbook* (doi: 10.1111/izy.12080).

Abstract: The endemic Philippine crocodile *Crocodylus mindorensis* is one of the most Critically Endangered crocodylian species in the world. Four major captive-breeding programmes for the Philippine crocodile have greatly contributed to our knowledge about the biology and natural history of the species. *In-situ* conservation actions, such as reintroduction, conservation breeding in large semi-wild areas with no supplementary feeding and head-starting programmes, could result in increasing the numbers of extant wild and semi-wild populations. If concerted efforts are directed at *in-situ* and *ex-situ* conservation, and locating quality habitats in which to establish new conservation sites for Philippine crocodiles, the species is likely to become widespread in the next few decades. Conservation measures that address species-specific issues promoting healthy viable populations in natural habitats are presented in this paper.

Stuebing, R., Sommerlad, R. and Staniewicz, A. (2015). Conservation of the Sunda gharial *Tomistoma schlegelii* in Lake Mesangat, Indonesia. *International Zoo Yearbook* 49.

Abstract: Although featured in many international zoo collections for decades, little was known about the natural history of Sunda gharial *Tomistoma schlegelii* until recently. Zoos rarely keep large individuals and breeding success has been low. As late as 1998, even though most conservationists regarded the conservation status of the species as Endangered, the reality was that over most of the range the actual status of the Sunda gharial was Data Deficient. Beginning with surveys of the species by international and local scientists in Indonesian Sumatra and, later, in Kalimantan, Indonesian Borneo, supported by the International Union for Conservation of Nature/Species Survival Commission Crocodile Specialist Group, more details on the broad distribution of this crocodylian came to light. Ironically, rediscovery of a large and healthy population of *T. schlegelii* in East Kalimantan arose from an oil-palm company accessing the area to develop a site called Danau Mesangat. Subsequently through a cooperation agreement with the oil-palm company's conservation department and a local foundation, a group of dedicated zoos in Europe and America, and the Tomistoma Task Force set up by the Crocodile Specialist Group, provided funding for three consecutive years of field studies by a research group. These studies investigated the ecology of *T. schlegelii* and of a sympatric population of the Critically Endangered Siamese crocodile *Crocodylus siamensis*. The role of the zoos, including their role in the development of the research programme, is described. Accounts are given of the characteristics of the Mesangat habitat, some new details about the distribution and abundance of Sunda gharial and Siamese crocodiles in the habitat, and information about reproduction in *T. schlegelii*. In summary, an overview of threats and suggestions for conservation actions needed at the Mesangat site are provided.

Weigl, R. (2014). Longevity of crocodylians in captivity. *International Zoo News* 61(5): 363-373.

Abstract: Since 1978 the author has collected numerous records on animal longevities. This data has been collected during personal examination of archives at zoological collections worldwide and through correspondence with zoo staff members. Taxonomy in this list follows that used in the publications of the Crocodile Specialist Group of the IUCN. This paper is the first comprehensive listing for every species and subspecies of crocodylian for which the author has a verified and known longevity record, including initial date of entry (hatch, capture, arrival) and a known date of death and/or verification as still living. Documentation of longevities at the subspecies level has not been included in previous treatments of the Crocodylia. Anecdotal records have not been accepted.

Von Baczko, M.B., Desojo, J.B. and Pol, D. (2014). Anatomy and phylogenetic position of *Venaticosuchus rusconii* Bonaparte, 1970 (Archosauria, Pseudosuchia), from the Ischigualasto Formation (Late Triassic), La Rioja, Argentina. *Journal of Vertebrate Paleontology* 34(6): 1342-1356.

Abstract: Ornithosuchidae is a group of Late Triassic pseudosuchian archosaurs of controversial phylogenetic affinities. This clade currently comprises three taxa: *Venaticosuchus rusconii* and *Riojasuchus tenuisiceps*, both from Argentina, and *Ornithosuchus longidens*, from Scotland. *Venaticosuchus rusconii* was erected on the basis of an incomplete skull with articulated lower jaws and some elements of the postcranium that are currently lost. *Venaticosuchus rusconii* is redescribed here and included for the first time in a phylogenetic analysis to test its affinities with ornithosuchids. The bizarre morphology of *V. rusconii* has a unique combination of traits that distinguishes it from the other ornithosuchids, such as basiptyergoid processes directed ventrally, dentary with the dorsal margin of the anterior end dorsally expanded, articular without a foramen on its medial side, and the absence of a surangular foramen, corroborating the validity of this taxon. *Venaticosuchus rusconii* is recovered as an ornithosuchid more closely related to *R. tenuisiceps* than to *O. longidens*, based on the presence of a nearly pointed anterior margin of the antorbital fenestra, and a vertical bony strut of the jugal that separates the antorbital fenestra from the infratemporal fenestra.

Francis, M. (2014). Improving water stability of extrudate feed for *C. porosus* using sodium alginate. MSc Thesis, Massey University, Manawatu, New Zealand.

Abstract: The extrudate sausage-pellet feed that is used currently to feed farmed *C. porosus* disintegrates on contact with water, which leads to leaching of nutrients and results in approximately 50% of feed being wasted. The objective of this study was to ascertain if Na alginate could be used to improve feed stability in water and then examine its effects on animal digestibility. The approach first looked at *in vitro* laboratory experiments to examine optimal conditions for Na alginate use. The second phase then applied those findings on-farm in a digestibility trial to measure if Na alginate affected digestion. Acceptance of feed containing Na alginate by crocodiles was also observed. In the laboratory, a new diet was replicated from the 'MHCF 2012/2 Fine' feed formulation obtained from Mainland Holdings Crocodile Farm (MHCF) and had three sodium alginate products (Protanal XP 3639, Manuacol DM and Kimica) added separately at 1.7 and 3.3% (as-fed). Each of these was cross-linked with either CaCaO3 or CaCl2 at 1.9% w/w rate. Feed was subjected to sensory evaluation and water stability measurements. For sensory evaluation, the diets were assessed by physical observation of their texture by sight and by feel and scores were given for traits including cohesiveness, viscosity, adhesiveness, and wetness. For water stability, a sample of feed for each alginate product, alginate level, and Ca source (15-30 g) was collected and subjected to water submersion for 18-24 hours and then was oven dried. Its dry matter recovery (DMR) was calculated as a proportion of the DM remaining after water immersion to the initial DM added to the water. Reaction with CaCO₃ resulted in greater DMR (80.68% DMR; P<0.05) compared with CaCl₂ (16.08% DMR). There was a greater than 10-fold increase in DMR when Na alginate was used with CaCO₃ compared to the control (86.7% vs 6.2% DMR),

however there were no differences in DMR among all Na alginates and inclusion levels with CaCO₃. Therefore, the least expensive Na alginate product at the lower inclusion level with CaCO₃ was recommended for use on-farm and in the digestibility experiment. For the on-farm digestibility experiment, ten juvenile crocodiles (2.2-2.4 years of age, 1.2-1.9 kg BW) were chosen from farm raised stocks and fed the extrudate chicken by-product-based diets with and without 1.4% sodium alginate and 1.9% CaCO₃ added. The percentage composition of sodium alginate was reduced due to increase in component of other ingredients (except sodium alginate) to meet the capacity of the processing machine. Animals were fed 2% BW for 12 d, with faeces collected the last 5 d. Animals were then slaughtered and digesta sampled from the ileum. Acid insoluble ash was used an internal marker. There were no effects of alginate on faecal digestibility of DM (0.350 vs 0.664, SE= 0.101) and N (0.249 vs 0.556, SE= 0.15) as well as ileal digestibilities for AA, DM, OM and N between the two diets (DM= 0.360 vs 0.410, SE= 0.146; OM= 0.396 vs 0.468, SE= 0.128; N= 0.558 vs 0.650, SE= 0.122), except OM and energy faecal digestibilities were greater with Na alginate (OM= 0.392 vs 0.698, SE= 0.091; energy= 0.444 vs 0.722, SE= 0.083) These results indicate there were no deleterious effects of alginate on digestibility of nutrients in *C. porosus*. In conclusion, this study showed that Na alginate had the potential to effectively reduce feed wastage and cost by preventing the feed from disintegrating and dissolving on contact with water. Furthermore, Na alginate did not interfere with feed digestion in *C. porosus*.

Temple, B.L., Finger, Jr., J.W., Jones, C.A., Gabbard, J.D., Jelesijevic, T., Uhl, E.W., Hogan, R.J., Glenn, T.C. and Tompkins, S.M. (2014). *In ovo* and *in vitro* susceptibility of American alligators (*Alligator mississippiensis*) to avian influenza virus infection. *Journal of Wildlife Diseases* (doi: <http://dx.doi.org/10.7589/2013-12-321>).

Abstract: Avian influenza has emerged as one of the most ubiquitous viruses within our biosphere. Wild aquatic birds are believed to be the primary reservoir of all influenza viruses; however, the spillover of H5N1 highly pathogenic avian influenza (HPAI) and the recent swine-origin pandemic H1N1 viruses have sparked increased interest in identifying and understanding which and how many species can be infected. Moreover, novel influenza virus sequences were recently isolated from New World bats. Crocodylians have a slow rate of molecular evolution and are the sister group to birds; thus they are a logical reptilian group to explore susceptibility to influenza virus infection and they provide a link between birds and mammals. A primary American alligator (*Alligator mississippiensis*) cell line, and embryos, were infected with four, low pathogenic avian influenza (LPAI) strains to assess susceptibility to infection. Embryonated alligator eggs supported virus replication, as evidenced by the influenza virus M gene and infectious virus detected in allantoic fluid and by virus antigen staining in embryo tissues. Primary alligator cells were also inoculated with the LPAI viruses and showed susceptibility based upon antigen staining; however, the requirement for trypsin to support replication

in cell culture limited replication. To assess influenza virus replication in culture, primary alligator cells were inoculated with H1N1 human influenza or H5N1 HPAI viruses that replicate independent of trypsin. Both viruses replicated efficiently in culture, even at the 30°C temperature preferred by the alligator cells. This research demonstrates the ability of wild-type influenza viruses to infect and replicate within two crocodylian substrates and suggests the need for further research to assess crocodylians as a species potentially susceptible to influenza virus infection.

Kerfoot, J.R., Fern, M.P. and Elsey, R.M. (2014). Scaling the feeding mechanism of captive *Alligator mississippiensis* from hatchling to juvenile. *Biology (Basel)* 3(4): 724-738.

Abstract: Small changes in size can lead to potential performance consequences and may influence an organism's ability to utilize resources in its environment. As the American alligator (*Alligator mississippiensis*) transitions between neonate, juvenile and adult habitats (ontogenetic niche shifts), there are inevitably dynamic changes in their feeding performance. This study sought to investigate the scaling of the feeding mechanism and its performance from hatchling to juvenile size classes in *A. mississippiensis*. Feeding events were recorded during March 2011 at Rockefeller Wildlife Refuge (Grand Chenier, Louisiana). Thirty-six captive individuals were randomly sampled, ranging from 30.5 cm to 91.5 cm TL, and feeding events were recorded using a high speed camera at a rate of 300 fps. Results indicated that many linear, angular and timing kinematic variables scale allometrically with cranium length; whereas maximum gape velocity and duration of feeding bout do not scale with cranium length and remain constant between these size classes. Although it has been shown that there is an isometric relationship between cranial elements and body size in *A. mississippiensis*, this relationship is not transferred to linear and timing variables of prey-capture events. These allometric relationships echo other investigations of scaling relationships such as bite-force production and terrestrial locomotion.

Barão-Nóbrega, J.A.L., Marioni, B., Villamarín, F., Soares, A.M.V.M., Magnusson, W.E. and Da Silveira, R. (2014). Researcher disturbance has minimal impact on natural predation of caiman nests in Central Amazonia. *Journal of Herpetology* 48(3): 338-342.

Abstract: Predation is one of the major causes of crocodylian egg mortality. Many studies have suggested that opening nests, handling eggs, and capturing attending females may increase predation rates, but the influence of such activities on nest predation has not been quantified. For that reason, we studied the impacts of different research activities on nests of wild Spectacled Caimans (*Caiman crocodilus crocodilus*) from a flooded forest (várzea) in the Piagaçu-Purus Sustainable Development Reserve, Central Amazonia. During the 2010 nesting season, predation on eggs in disturbed nests was generally higher than in nests without research activities found in the same area. However, natural predation rates in

similar habitat were not significantly different, suggesting that predation rates on disturbed nests, though elevated, remain within the broad range found in undisturbed nests. We conclude that the benefits in knowledge gained from research activities will generally out-weigh the small increase in nest predation that could result from research activities.

Colello, J.M.A. (2014). Estado Actual de la Poblacion de Caimanes (*Caiman crocodilus*) y Possibilidades Socioeconomicas para su Manejo en el Refugio Nacional De Vida Silvestre Caño Negro, Costa Rica. MSc thesis, Universidad Nacional, Heredia, Costa Rica.

Schneider, M., Klein, B., Krämer, D., Knezevic, K., Tiflova, L., Vogt, S., Rauhaus, A., van der Straeten, K., Karbe, D., Sommerlad, R. and Ziegler, T. (2014). First observations on the courtship, mating, and nest visit behaviour of the Philippine crocodile (*Crocodylus mindorensis*) at the Cologne Zoo. *Journal of Zoo and Aquarium Research* 2(4): 123-129.

Abstract: The aim of this study was to gain a better understanding of the social and in particular reproductive behaviour of the Critically Endangered Philippine crocodile. *Crocodylus mindorensis* has been a focus for international conservation breeding measures for about two decades. Since little scientific data have been gathered on the biology and ecology of the species so far, its breeding remains a challenge. In order to identify behavioural patterns that trigger courtship behaviour, and to determine when sociopositive interactions increase and the animals are ready for reproduction, a pair of two adult *C. mindorensis* at Cologne Zoo was systematically observed between August 2011 and July 2012 for a total of 583 hours. Observations took place using all occurrences recording and scan sampling, focusing on pre-, post- and actual mating behaviour. We present a detailed documentation of copulations with behaviours such as growling, roaring, and bubbling. Bubbling in both sexes was observed prior to copulations and decreased with the end of the mating season, supporting the assumption that it can be referred to as courtship behaviour. Behaviours that indicate the approach of the breeding season, such as approaches to the dividing slide, bubbling and nest building, could be distinguished. Our findings should help to improve breeding efforts under captive husbandry conditions and thus contribute to the conservation breeding of this highly endangered and difficult to keep crocodylian species.

Tanaka, K. and Zelenitsky, D.K. (2014). Comparisons between experimental and morphometric water vapor conductance in the eggs of extant birds and crocodiles: implications for predicting nest type in dinosaurs. *Canadian Journal of Zoology* (doi: 10.1139/cjz-2014-0078).

Abstract: Estimates of water vapor conductance for the eggs (ie diffusive capacity of eggshell; GH_2O , $\text{mgH}_2\text{O day}^{-1} \text{ torr}^{-1}$) of extinct archosaur species have been used to infer their nest type (ie covered vs open) because experimental GH_2O values for living archosaur species appear to reflect nest type.

The methods used to derive GH_2O for fossil eggs (eggshell morphometrics) differs from that for extant eggs (experimental measurements), and it remains unknown if these two methods are comparable or if morphometric GH_2O values correspond to nest type. Although previous studies assumed that GH_2O values derived from the two methods were comparable, this assumption has not been statistically evaluated in a large sample size that includes both crocodiles and birds. Here, GH_2O values for over 100 species of living archosaurs for both morphometric and experimental methods were compiled and compared using statistical analyses. Results found that although experimental and morphometric GH_2O values are significantly correlated, there is disagreement between the methods particularly apparent in small eggs, likely due to systematic errors. These results suggest that morphometric and experimental GH_2O of living species are not necessarily comparable, although the reason for the discrepancy remains uncertain. Thus, direct comparisons between morphometric GH_2O of dinosaurs and experimental GH_2O of living species should be avoided when inferring the nest type for dinosaurs.

MacCormick, A. (2014). *Man-Eaters of the World: True Accounts of Predators Hunting Humans*. Skyhorse Publishing: New York.

Rabi, M. and Sebok, N. (2014). A revised Eurogondwana model: Late Cretaceous notosuchian crocodyliforms and other vertebrate taxa suggest the retention of episodic faunal links between Europe and Gondwana during most of the Cretaceous. *Gondwana Research* (doi: 10.1016/j.gr.2014.09.015).

Abstract: We describe new remains of the enigmatic Late Cretaceous crocodyliform *Doratodon carcharidens* coming from the Santonian Csehbánya Formation of Hungary. The material includes isolated upper and lower jaw elements and teeth that represent the earliest occurrence of this genus. Previous reports of *Doratodon* restricted the range of this taxon to the Campanian-Maastrichtian of the areas that are now Romania, Austria and Spain. Using data from both the new remains and the type series from the lower Campanian of Austria, our phylogenetic analysis finds *D. carcharidens* deeply nested within Gondwanan notosuchians as part of the predominantly South American clade Sebecosuchia. The suggested affinity of *D. carcharidens* is generally in agreement with previous cladistic assessments and confirms the presence of Gondwanan faunal elements in the Late Cretaceous western Tethyan archipelago (the region that is now southern Europe). Together with published reports on Gondwanan tetrapod immigrants from the Barremian, Albian, Cenomanian and Santonian of Europe this record is not compatible with a recently proposed paleobiogeographical model predicting a post-Hauterivian separation of Europe from Africa and subsequent biogeographical reconnection in the Campanian lasting till the Eocene. Instead, the European fossil record suggests that episodic faunal and geographical links were retained with Gondwana during most of the Cretaceous.

Young, M.T., Hua, S., Steel, L., Foffa, D., Brusatte, S.L., Thuring, S., Mateus, O., Ruiz-Omenaca, J.I., Havlik, P., Lepage, Y. and De Andrade, M.B. (2014). Revision of the Late Jurassic teleosaurid genus *Machimosaurus* (Crocodylomorpha, Thalattosuchia). *R. Soc. open sci.* 1: 140222.

Abstract: *Machimosaurus* was a large-bodied genus of teleosaurid crocodylomorph, considered to have been durophagous/chelonivorous, and which frequented coastal marine/estuarine ecosystems during the Late Jurassic. Here, we revise the genus based on previously described specimens and revise the species within this genus. We conclude that there were three European *Machimosaurus* species and another taxon in Ethiopia. This conclusion is based on numerous lines of evidence: craniomandibular, dental and postcranial morphologies; differences in estimated total body length; geological age; geographical distribution; and hypothetical lifestyle. We re-diagnose the type species *Machimosaurus hugii* and limit referred specimens to only those from Upper Kimmeridgian-Lower Tithonian of Switzerland, Portugal and Spain. We also re-diagnose *Machimosaurus mosae*, demonstrate that it is an available name and restrict the species to the uppermost Kimmeridgian-lowermost Tithonian of northeastern France. We re-diagnose and validate the species *Machimosaurus nowackianus* from Harrar, Ethiopia. Finally, we establish a new species, *Machimosaurus buffetauti*, for the Lower Kimmeridgian specimens of France and Germany (and possibly England and Poland). We hypothesize that *Machimosaurus* may have been analogous to the Pliocene-Holocene genus *Crocodylus* in having one large-bodied taxon suited to traversing marine barriers and additional, geographically limited taxa across its range.

Nifong, J.C., Layman, C.A. and Silliman, B.R. (2014). Size, sex, and individual-level behavior drive intra-population variation in cross-ecosystem foraging of a top-predator. *Journal of Animal Ecology* (doi: 10.1111/1365-2656.12306).

Abstract: Large-bodied, top-predators are often highly mobile, with the potential to provide important linkages between spatially distinct food webs. What biological factors contribute to variation in cross-ecosystem movements, however, have rarely been examined. Here, we investigated how ontogeny (body size), sex, and individual-level behavior impacts intra-population variation in cross-ecosystem foraging (ie between freshwater and marine systems), by the top-predator *Alligator mississippiensis*. Field surveys revealed *A. mississippiensis* uses marine ecosystems regularly and are abundant in estuarine tidal creeks (from 0.3-6.3 individuals/km of creek, n= 45 surveys). *Alligator mississippiensis* captured in marine/estuarine habitats were significantly larger than individuals captured in freshwater and intermediate habitats. Stomach content analysis showed that small juveniles consumed marine/estuarine prey less frequently (6.7% of individuals) than did large juveniles (57.8%), sub-adult (73%), and adult (78%) size classes. Isotopic mixing model analysis (SIAR) also suggests substantial variation

in use of marine/estuarine prey resources with differences among and within size classes between sexes and individuals (range of median estimates for marine/estuarine diet contribution = 0.05-0.76). These results demonstrate the importance of intra-population characteristics (body size, sex, and individual specialization) as key determinants of the strength of predator-driven ecosystem connectivity resulting from cross-ecosystem foraging behaviors. Understanding the factors which contribute to variation in cross-ecosystem foraging behaviors will improve our predictive understanding of the effects of top-predators on community structure and ecosystem function.

Tennant J.P. and Mannion, P.D. (2014) Revision of the Late Jurassic crocodyliform *Alligatorellus*, and evidence for allopatric speciation driving high diversity in western European atoposaurids. *PeerJ* 2: e599 (<http://dx.doi.org/10.7717/peerj.599>).

Abstract: Atoposaurid crocodyliforms represent an important faunal component of Late Jurassic to Early Cretaceous Laurasian semi-aquatic to terrestrial ecosystems, with numerous spatiotemporally contemporaneous atoposaurids known from western Europe. In particular, the Late Jurassic of France and Germany records evidence for high diversity and possible sympatric atoposaurid species belonging to *Alligatorellus*, *Alligatorium* and *Atoposaurus*. However, atoposaurid taxonomy has received little attention, and many species are in need of revision. As such, this potentially high European diversity within a narrow spatiotemporal range might be a taxonomic artefact. Here we provide a taxonomic and anatomical revision of the Late Jurassic atoposaurid *Alligatorellus*. Initially described as *A. beaumonti* from the Kimmeridgian of Cerin, eastern France, additional material from the Tithonian of Solnhofen, south-eastern Germany, was subsequently referred to this species, with the two occurrences differentiated as *A. beaumonti beaumonti* and *A. beaumonti bavaricus*, respectively. We provide a revised diagnosis for the genus *Alligatorellus*, and note a number of anatomical differences between the French and German specimens, including osteoderm morphology and the configuration and pattern of sculpting of cranial elements. Consequently, we restrict the name *Alligatorellus beaumonti* to include only the French remains, and raise the rank of the German material to a distinct species: *Alligatorellus bavaricus*. A new diagnosis is provided for both species, and we suggest that a recently referred specimen from a coeval German locality cannot be conclusively referred to *Alligatorellus*. Although it has previously been suggested that *Alligatorellus*, *Alligatorium* and *Atoposaurus* might represent a single growth series of one species, we find no conclusive evidence to support this proposal, and provide a number of morphological differences to distinguish these three taxa that appear to be independent of ontogeny. Consequently, we interpret high atoposaurid diversity in the Late Jurassic island archipelago of western Europe as a genuine biological signal, with closely related species of *Alligatorellus*, *Alligatorium* and *Atoposaurus* in both French and German basins providing evidence

for allopatric speciation, potentially driven by fluctuating highstand sea levels.

Siroski, P.A., Poletta, G.L., Parachú Marcó, M.V., Ortega, H.H. and Merchant, M.E. (2014). Presence of chitinase enzymes in crocodylians. *Acta Herpetologica* 9(2): 139-146.

Abstract: Chitin is an abundant bio-polymer present as a structural component of many organisms such as arthropods, nematodes, mollusks, insects, and fungi, among others. Chitinolytic enzymes are synthesized for organisms to defend themselves against chitin-containing pathogens. Chitotriosidase (CHT) is a chitinase enzyme and one of the main proteins secreted by activated macrophages. It plays an important role in mechanisms of immunity by hydrolyzing chitin, thus protecting against chitin-containing pathogens. In this study, CHT was detected in *Caiman latirostris* plasma, and characterized under laboratory controlled conditions of temperature, reaction time, plasma concentration, pH and salinity. The results complement other immunological studies performed in caimans and demonstrate that they possess an efficient and well-developed immune system that resists the attack of some pathogens. Based on the current knowledge of the properties and homologies of CHT, it would be highly valuable to evaluate its possible therapeutic application in the veterinary clinical setting.

Arp, D. Jr. (2014). Alligators, crocodiles and cutlasses: trade mark infringement in Guyana. *Journal of Intellectual Property Law & Practice* (doi: 10.1093/jiplp/jpu182).

Abstract: The markets of the developing world present a rich environment for infringement of trademarks and intellectual property, offering perpetrators quick and substantial profit with limited risk of interference from local law enforcement. In Guyana, such a scheme infringed on the trademark of a United Kingdom-based tool manufacturer and played on the cultural heritage buyers had with both the tool and the trademark. Fieldwork in developing nations is critical to investigate and re-investigate infringements on trademarks and intellectual property.

Chowfin, S. (2014). A second chance. *SAEVUS* December 2014: 27-33.

Campbell, H.A., Dwyer, R.G., Wilson, H., Irwin, T.R. and Franklin, C.E. (2014). Predicting the probability of large carnivore occurrence: a strategy to promote crocodile and human coexistence. *Animal Conservation* (doi: 10.1111/acv.12186).

Abstract: Preserving large carnivores that perceive humans as prey brings conservation values into direct conflict with human security. Informing when and where humans and large carnivores occupy the same space may reduce attack

frequency and promote coexistence. Here, we demonstrate a methodology to better understand the spatiotemporal relationship between a population of large carnivores and humans. The carnivore of study was the estuarine crocodile *Crocodylus porosus*, a large semi-aquatic predator responsible for 705 recorded human attacks over the last 20 years. Crocodiles were captured every August over 3 years and individuals greater than 2.5 m in length were implanted with an acoustic transmitter (n=84). The transmitter emitted a coded pulse detected when in proximity to underwater hydrophones deployed throughout the river. The telemetry data informed which previously captured crocodiles were present during subsequent trapping episodes and adult population size was estimated using a closed-population model. Over 3 years, 24 of the tagged crocodiles were detected 269 times moving through a shallow-water area where humans frequently entered the water. The tagged crocodile presence was extrapolated to the population level to provide a probability of adult crocodile presence across a range of temporal scales. The results showed that between September and December, the probability of crocodile presence within the human entry zone was 0.97 ± 0.01 during darkness but decreased to 0.07 ± 0.01 during daylight, except around periods of high tide when it increased to 0.71 ± 0.02 . Human visitors confined their activity to shallow water during daylight hours, but no consideration was given to the significant rise in crocodile presence with season and tide. The observed patterns in crocodile and human behaviour, around this shallow-water river crossing, exhibited parallels with historical incidences of crocodile attack.

Elsy, R.M., King, R., Gribbins, K., Like, D.E. and Lance, V. (2014). *Alligator mississippiensis* (American Alligator). Large presumptive female: a case of mistaken identity. *Herpetological Review* 45(1): 119-120.

Rojtinnakorn, N., Kupittayanant, S., Temsiripong, Y. and Griangsak, E. (2014). Antibacterial activity of plasma fractions from Siamese crocodile (*Crocodylus siamensis*) on Ceftazidime-resistant *Enterobacter cloacae*. Pp. 138-143 in *Proceedings of the 5th International Conference on Natural products for Health and Beauty*.

Abstract: The resistance of bacteria, is a major problem in the world including Thailand, lead to increasing the mortality and cost of medical care. Thus, the objective of this study was to investigate the activity of plasma fractions from Siamese crocodile (*Crocodylus siamensis*) against drug resistant bacteria, when use alone and in combination with β -lactams. The crocodile plasma was purified to give five fractions (P1, P2, P3, P4 and P5) using column chromatography. The MICs of P1, P2, P3, P4 and P5 against ceftazidime-resistant *Enterobacter cloacae* DMST21394 (CREnC) revealed 1024, >1024, >1024, >1024 and 1024 mg/ml, respectively, while MICs for methicillin-resistant *Staphylococcus aureus* DMST 20651 (MRSA) displayed 1024 mg/ml for all fractions. The FICs index of either P1 or P5 plus ceftazidime against

CREnC revealed both equal at 0.062, also these fractions plus cloxacillin demonstrated both equal 0.375 against MRSA. The killing curves confirmed that both P1 and P5 in combination with either ceftazidime or cloxacillin caused markedly decrease of CREnC or MRSA cells from 6 throughout 24 h. CREnC treated with the ceftazidime plus either P1 or P5 combination revealed a dramatically significant smaller cell size than the control cells ($p < 0.01$), cell distortion, cell envelope damage, increased the OM and CM permeability, β -lactamase inhibition ($p < 0.01$) and OMPG associated protein synthesis disruption. In conclusion, the combination of either P1 or P5 and ceftazidime showed strong synergistic activity against CREnC strain. These findings provide evidence that these combinations can reverse resistance strain to be susceptible to its primary antibiotic.

Saber, A.S. and Hassanin, A. (2014). Some morphological studies on the jaw joint of the Australian Saltwater Crocodile (*Crocodylus porosus*). J. Vet. Anat. 7(2): 55-74.

Abstract: The saltwater crocodile is the largest of all living reptiles. It is also known and proved that it has the largest biting forces. The magnitude of the biting force exerted by the jaw leads to thoughts about the anatomical structure and construction of the jaw joint in this reptile. Thirteen skulls of the saltwater crocodile (*C. porosus*) were used for this study. Some skulls were used for X-raying and morphology, and the others for histological slides which were prepared from the articular cartilages, capsule and collateral ligaments. The quadrate/articular joint (jaw joint) was diarthroidal, formed by two articular surfaces, which were fibrocartilage in nature at the periphery of the articular surface, hyaline in the rest of the articular surface, and ossifies on reaching the underlying bone. The thick lateral and thin medial collateral ligaments were formed of collagenous fibers. An articular disc was missing in the crocodile quadrate/articular joint. The joint was surrounded by a complex massive group of muscles responsible for the firm closure and opening of the mouth. The results were supported by 10 images and 4 tables, and were discussed with other amphibians, domestic animals and man when needed.

Alibardi, L. and Minelli, D. (2015). Sites of cell proliferation during scute morphogenesis in turtle and alligator are different from those of lepidosaurian scales. Acta Zoologica (doi: 10.1111/azo.12114).

Abstract: Cell proliferation in forming shield scutes has been studied by immunofluorescence in embryos of turtle, alligator and snake after injection of 5-bromo-deoxy-uridine. Hinge regions of scutes in alligator and turtle carapace derive from an initial waving and invagination of the epidermis that contains 5-bromo-deoxy-uridine-labelled cells. This suggests that down growth of the epidermis into the dermis is driven by local proliferation in addition to dermal anchorage and stabilization of hinge regions. Few keratinocytes migrate into suprabasal layers 1 day after injection of 5-bromo-

deoxy-uridine and keratinocytes reach the precorneous layer in about 5 days. Proliferating keratinocytes are randomly distributed in the outer scale surface of symmetric scutes but are more numerous in the outer scale surface of asymmetric or overlapped scutes indicating epidermal expansion. Higher localization of proliferating cells along hinge regions of embryonic turtle and alligator scutes is maintained in adult scutes where most growth occurs. In snake, skin proliferation becomes prevalent on the elongating outer side of the asymmetric scale. Comparison between proliferation sites in turtle-alligator-chick scales with lepidosaurian scales indicates that placodes are present only in turtle-alligator-chick scales. Conversely, scale primordia detected only using gene markers are found in most crocodilian and lepidosaurians embryonic skin.

Submitted Publications

Mugger Crocodiles (*Crocodylus palustris*) in Iran: a preliminary genetic study

Introduction

Mugger crocodiles (*Crocodylus palustris*) are found in the southeastern corner of Iran, southern Pakistan, India and Sri Lanka and the southeast edge of Nepal, but extinction in the wild is reported for Bhutan, Bangladesh and Myanmar (Da Silva and Lenin 2010). The species is listed by the IUCN Red List as Vulnerable due to a decline in global population size of 20% or more in the last three generations and an estimated global population of less than 2500 adults that are located in fragmented habitat of declining quality (Da Silva and Lenin 2010). In Iran, small groups of Muggers are distributed in the Sistan and Baluchestan Provinces. The total population is estimated to be about 500 non-hatchlings (Mobaraki and Abtin 2013) that occupy a diversity of aquatic habitats, including artificial ponds and reservoirs (Mobaraki 2002). The population exists as several sub-populations, with the largest groups found along the Kaju River to the west and along the Sarbaz and Balukalat Rivers in the north and central parts of the region. A major portion of the Mugger's range in Iran has been designated as the Gandou Protected Area (Fig. 1), which encompasses 465,181 ha, with a focal area along the Sarbaz and Bahukalat Rivers. Gandou is the local name for the Mugger, reflecting the importance of this species in establishing the protected area (Mobaraki 2002).

Movement between habitats is a feature of Muggers in Iran, and the recent construction of dams poses a potential threat by limiting movement between isolated sub-populations. Muggers are known to move between habitats during periods of drought and depending on local water availability and the size of crocodiles, the distance moved is variable. Dead crocodiles have been found on roads a few kilometres from water bodies, where they were killed by cars (Mobaraki and Abtin 2007). Additionally, dead juveniles <1.5 m TL have also been found far from any water bodies where the origin or destination could not be determined (Mobaraki and Abtin

2007). Over the last 15 years, dams have been constructed along the main rivers, including the Pishin and Shirgovaz Dams on the Bahukalat River, and the recently completed Zirdan Dam on the Kaju River (Fig. 1). These large dams constitute obstacles for the crocodiles by preventing any in-river contact between the sub-populations.

The extent to which Mugger populations in Iran have been isolated from those in neighbouring Pakistan is unknown. Muggers in both Iran and Pakistan are mainly found in the Baluchestan areas of the both countries and there are some reports of movement of crocodiles through the border between some habitats (Mobaraki 2000). In the northeastern part of the area, close to Saravan, a sub-population of crocodiles exists in the Nahang River. This river has tributaries in both Iran and Pakistan (where it is renamed as the Dasht River) and it is likely to support a shared Mugger population between the two countries, due to connectivity during some rainy seasons. To investigate population history and the extent of isolation of Iranian crocodiles, this paper reports on a preliminary genetic survey of Muggers in Iran that will support future comparisons of the genetic structure of Mugger populations in Iran to those of neighbouring countries.

Methods

Ten samples were collected from Muggers captured from the Sarbaz and Bahou Kalat Rivers (Fig. 1) and analyzed for variation in the mtDNA control region. The greatest river distance between the sample locations was approximately 75 km. Tissue samples were taken from the vertical tail scutes of wild-caught animals, ranging from hatchling to adult. Samples were stored in a solution of 20% dimethyl sulfoxide that was saturated with NaCl. DNA extractions followed a simple salting-out protocol (FitzSimmons *et al.* 2002). PCR amplification was carried out using primers L15463 (5'-cgctggccttgaagacaga-3') and H16260 (5'-cactaaaattacagaaaagccgac-3'), which were designed by Travis Glen and reported in FitzSimmons *et al.* (2002). These primers amplified a 788 bp fragment of the control region and adjacent tRNAs. PCR reactions were carried out in 25

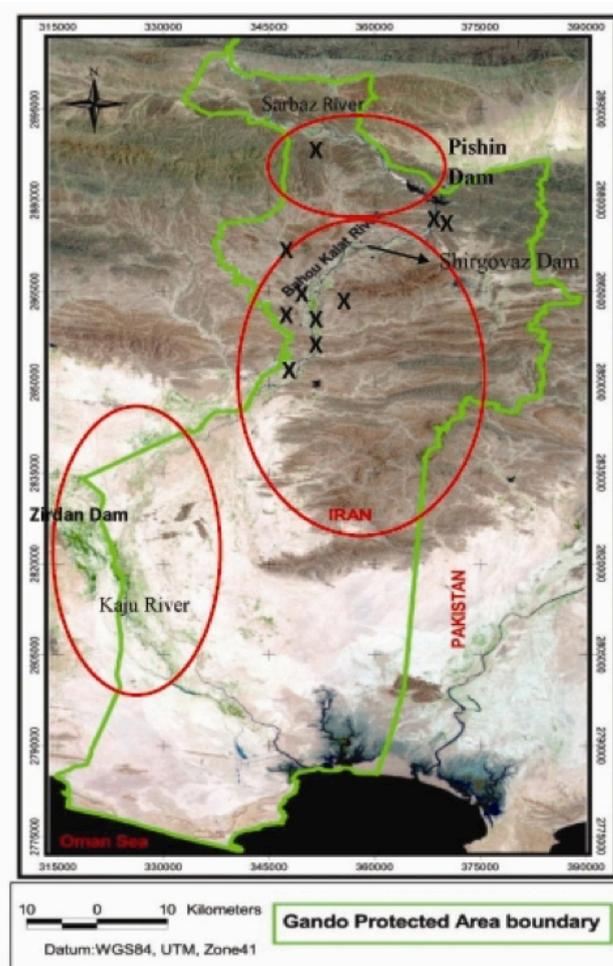


Figure 1. *Crocodylus palustris* habitat in Iran and sample locations for genetic analysis. Red circles indicate the sub-populations of crocodiles in the region, the green boundary indicates Gandou protected area, and Xs denote sample locations.

µl volumes containing 0.8 mM dNTPs, 2.5 mM MgCl₂, 0.02 mg BSA (bovine serum albumin), 1 µM each primer, 1 unit Astral Red Taq and 1 x Astral buffer. PCR temperature profiles were 94°C for 3 min, followed by 32 cycles of 94°C

Table 1. Variable sites in the mtDNA control region of Mugger crocodiles. Base pair (bp) distances are taken from the start of the Thr anticodon of tRNA-Thr as identified in sample GU144286.1 (Feng *et al.* 2010). ¹same sequence for AY138885.1-AY138888.1; ²this study.

GenBank ¹ Sample	Basepair (bp) Location											
	19	129-130	131 indel	198-209 indel	215-219 indel	558	570	650	667	685	718	746 indel
Iran ²	C	-	G	CAACAAACCAAC	ACCAC	C	T	C	C	G	C	-
JF315358.1	C	-	G	CAACAAACCAAC	ACCAC	C	T	C	C	G	C	-
JF315359.1	C	-	G	CAACAAACCAAC	ACCAC	C	T	C	C	G	C	-
GU144286.1	C	-	-	-	-	C	T	C	C	G	C	-
NC_014706.1	C	-	-	-	-	C	T	C	C	G	C	-
AF460212.1	?	?	?	?AACAAACCAAC	ACCAC	C	T	C	C	G	C	A
HM488007.1	T	TA	-	CAACAAACCAAC	ACCAC	T	C	A	T	A	C	-
AY138884.1 ¹	?	?	?	?	?	?	?	?	?	?	T	-

for 30s, 48°C for 45s and 72°C for 55s, with a final extension of 72°C for 5 min. PCR products were sent to Macrogen (Korea) for sequencing in both directions using the same primers. All sequences were aligned using Geneious 6.0.4 software and checked visually and GenBank was searched for matching sequences. Sequences were truncated to 750 bp for comparison to those available on GenBank

Results

All crocodile samples from Iran had the same 750 bp haplotype. For comparative purposes, we aligned this haplotype to several Mugger mtDNA control region sequences available on GenBank. These included: AF460212.1 (Ray and Densmore 2002), JF315358.1 and JF315359.1 (Oakes 2011), and complete mtDNA genomes of NC_014706.1 and GU144286.1 (Feng *et al.* 2010) and HM488007.1 (Meganathan *et al.* 2012). Among all sequences there were 7 substitutions, including 6 transitions and one transversion (Table 1). Five indels were observed, including a 5 bp and a 12 bp indel observed in the Iran and other samples (Table 1). Five additional samples from Genbank (AY138884.1 – AY138888.1; Ray and Densmore 2003) only provided data from the 703-740 bp region, in which all shared the same sequence, which was one bp different from the Iranian haplotype (Table 1). If the observed indels are each counted as a single mutation event, the uncorrected maximum divergence (p) among haplotypes is 1.5%. Unfortunately the original sample locations of the crocodiles represented by these different haplotypes was not provided in the previous studies and the authors we were able to contact did not know the origins of the samples.

Discussion

All samples from Iran had the same haplotype, suggesting a past connection among Muggers at the sample locations. It is likely that since the construction of the Pishin Dam in 1990, previous gene flow would have ceased between the Sarbaz River, upstream of Pishin Dam, and downstream of the dam wall, given the dry rugged country adjacent to the dam site. Muggers are found in the reservoir of Pishin Dam but nothing is known about their movements. If further studies indicate that the dam has caused the isolation of fragmented populations, it would be possible to translocate crocodiles between the two rivers.

Further sampling is needed throughout the range of *C. palustris* to reveal whether the haplotype diversity as evidenced on GenBank can be found in the wild and whether this can provide insights into population history and genetic isolation. The haplotype observed among the Iranian samples was identical along the 750 bp to two samples reported in Oakes (2011), which were obtained from Kent Vliet (USA) from animals with unknown origins. These samples shared the 5 bp and 12 bp insertion mutations that were also observed in the samples reported in Ray and Densmore (2002) and

Meganathan *et al.* (2012), one of which (HM488007.1) came from a collection in India of unknown origin. In contrast, neither insertions were observed in the two samples of Feng *et al.* (2010) that were obtained from the Anhui Normal University in China and are also of unknown origin.

Additional samples are needed to assess the extent of genetic diversity within the Mugger populations in Iran and throughout their range to determine the extent of genetic structure and degree of isolation among the local populations. These analyses are needed to support the development of conservation plans for this species. Cooperative works between Mugger range states is recommended and we would welcome other interested people joining our ongoing works.

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TWO BEHAVIOURAL APPROACHES TO EATING A TOAD. Decades ago in northeastern North America, I experimented to see if a captive and hungry young American crocodile (*Crocodylus acutus*) would eat an American toad (*Bufo americanus*), or alternatively if it would reject the toad as food. I had predicted that the toxic defensive excretions from the skin of the toad would make it taste too bad, but to my surprise my pet crocodile, in a bath tub of shallow water and with all four feet comfortably on the bottom, quickly captured its prey, held it in its mouth and chewed it a while, then rinsed it in the water employing a prolonged and deliberate and vigorous sidewise movement of its own head, then chewed it some more and then rinsed it, and repeated the process and often performed both operations simultaneously, for at least 15 minutes before eventually swallowing the unhappy and exhausted and now detoxified toad alive.

Born in Colombia and shipped airfreight from Barranquilla to New York City and then sold at a young age at a pet store, back in the 1960s when it was legal, my captive crocodilian had physically stimulated the toad to secrete its defensive toxins, and therefore when the white colored "bufotoxin" liquid excretions were leached, lost and depleted, and had all been washed and rinsed away in the water, it became safe to eat the remaining part of the toad that was not poisonous. In this case, and under the circumstances of being alone in a bath tub, my crocodile "Felix" was hungry and got its meal, but notably only through being remarkably patient and methodical about properly and appropriately preparing its food for consumption.

It appeared that there was some sort of forethought and planning involved in my South American crocodile's behavioural solution to a problem presented to it in Ontario, Canada. However, I do not know what would have happened if this feeding experiment had been tried on land without sufficient water, nor what would have happened if there had been other crocodilians present and threatening to grab the food if it was not swallowed quickly and without significant

delay. It is my understanding that when a living toad is swallowed alive, it will release its bufotoxin secretions while inside the crocodile's stomach. However, this threat only applies when and if the still alive toad has not already been detoxified by having its toxins exhausted and lost prior to being swallowed.

Oddly, in a similar experiment in Australia with hatchling Australian freshies (*C. johnstoni*) being fed introduced (feral) cane toads (*Bufo marinus*), it was found that the crocodiles with prior germane experience mostly swallowed their toads more quickly than expected, so that the bad taste of the toad spent as little time in the predator's mouth as possible. Alternatively some of the cane toads in the Australian experiment were rejected. Thus in a simplistic sense the experienced young *C. johnstoni* would either eat its toad quickly, or not at all, while in contrast my *C. acutus* ate its toads slowly, and in a bath tub of shallow water and plenty of time to prepare it. Felix never refused local toad as a snack. Further, with a sufficient body of comfortably shallow water and no competition, I noticed no ill effects of my occasional addition of toad to my captive Colombian crocodile's diet.

About the young Australian freshies being offered *B. marinus* (renamed as *Rhinella marina*) as an experiment, see Somaweera *et al.* (2011), and concerning the introduced cane toads being a dietary threat to selected Australian wildlife see the Smith and Phillips (2006) and Doody *et al.* (2009). However, at least some wild *C. johnstoni* (in Lake Argyle, Western Australia) are apparently able to ingest *R. marina* without it being fatal to the crocodile (Somaweera and Shine 2012), which reported that sometimes it only takes one cane toad to kill an Australian freshie, while at other times or in other places the result is different, because the crocodile survives alive after ingesting a toad or toads.

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