

# Unveiling the Secrets of Life

## — IOZ's Explorations and Contributions

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**Abstract:** With a history of 96 years, the Institute of Zoology (IOZ) is one of the oldest scientific research institutions of the Chinese Academy of Sciences (CAS). Since its establishment, IOZ has consistently served the national strategy and pursued the forefront of science. Since 1978 when China implemented a reform and opening up policy, especially over the past two decades, IOZ has achieved a series of world-leading science and technology achievements, deepening our understanding of mechanisms of cell programming and reprogramming, reproductive and developmental regulation, stem cell and regenerative medicine, integrated management of pest insects and rodents, endangered mechanisms and conservation of species, and animal taxonomy and evolution. In the future, IOZ will strive to seize the science highland by optimizing the layout of its disciplinary development, attracting and fostering talents, and constructing world-class research facilities and platforms, aiming to be a leading institution for the continuous innovation and development of life science both in China and the world.

**Keywords:** stem cell, regenerative medicine, cell programming and reprogramming, reproductive and developmental regulation, pest insect and rodent management, endangered species and conservation, animal taxonomy and evolution

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## Introduction

Institute of Zoology (IOZ), Chinese Academy of Sciences (CAS), is a government-funded research institution in zoological sciences. Its history can be traced back to its predecessor, Fan Memorial Institute of Biology founded in 1928. IOZ started very early to serve the country's needs with its science research outputs. Typical cases include the successful control of China's chronic agricultural pest-locust, and the control and management of other pest insects and rodents, playing a key role in the country's agricultural development. It also established nature reserves in China, facilitating the conservation of giant pandas, crested ibises and other endangered wildlife. More recent contributions of IOZ include outputs in the fields of reproduction and contraception, and fish nuclear transfer, among others.

The research efforts of IOZ

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have been directed to address both basic science and applied research to meet the national and public demands in the fields of agriculture, biodiversity, ecology, human health and reproductive biology. Currently, researchers are especially focusing on mechanisms of cell programming and reprogramming, stem cell and regenerative medicine, integrated management of pest insects and rodents, endangered mechanisms and conservation of species, animal taxonomy and evolution. This article is to place more attention to the latest research activities and review the progress and achievements of these fields in brief.

## Mechanisms of Cell Programming and Reprogramming

A mammalian individual has over 200 different cell types, all of which arise from a single initial cell—the fertilized egg—through continuous division and differentiation. At what stage does the first choice of cell fate occur during the division and development of the fertilized egg? How does this choice happen? This is an important foundational question in developmental biology. IOZ researchers have, for the first time, pushed the differentiation choice of cell fate in mice to the 2-cell embryo stage and discovered that a key molecule involved is an endogenous retroviral-related long non-coding RNA. This finding lays a significant foundation for exploring the regulation of pluripotency in early embryos and the mechanisms behind the first differentiation of cell fates, providing new theoretical references for establishing higher pluripotent or even totipotent stem

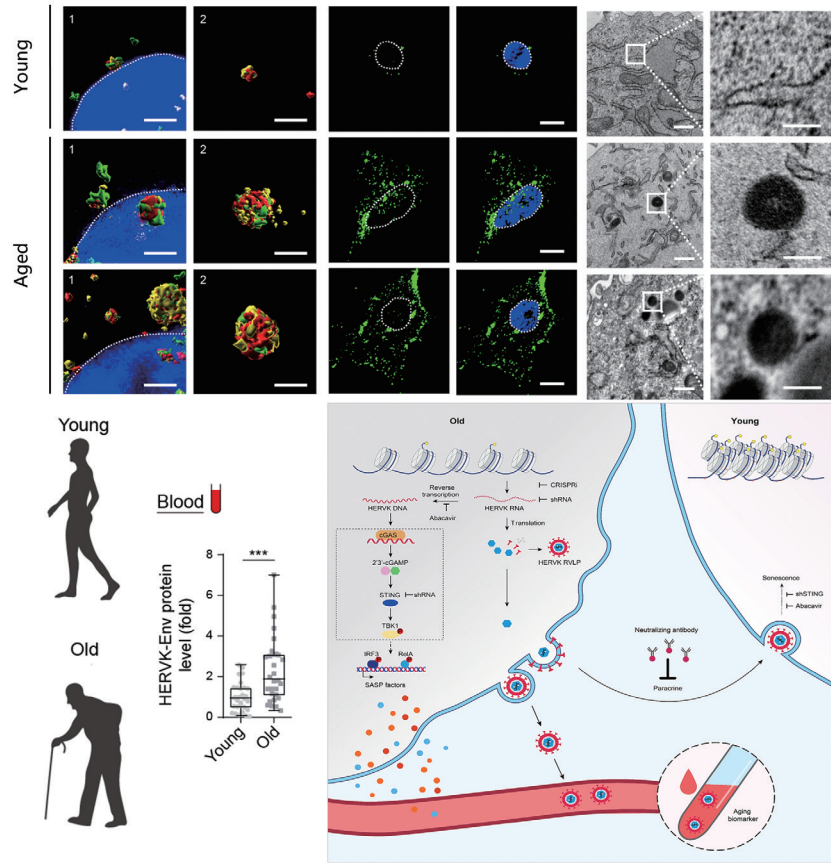


Figure 1. De-repression of the endogenous retrovirus contributes to programmed aging, adapted from Liu *et al.* (2023).

cells (Wang *et al.*, 2018). IOZ researchers further examined the developmental processes of subsequent cell type fates, revealing the epigenetic regulatory mechanisms that maintain the stemness of various adult stem cells, including neural stem cells and hematopoietic stem cells (Zhang *et al.*, 2017; Li *et al.*, 2023). IOZ also studied cellular and individual changes during aging, systematically characterizing the phenotypes, pathologies, and molecular characteristics of spinal cord aging in primates. IOZ researchers revealed a novel type of microglia that can promote motor neuron aging, which can serve as a biomarker for measuring the aging degree of the human spinal cord, thus providing

a new paradigm for researching human neurodegeneration and evaluating related drugs (Sun *et al.*, 2023). Additionally, IOZ found that individual aging is accompanied by the reactivation of “dark matter” endogenous retroviruses (ERVs) within the genome (Figure 1), elucidating how ancient virus reactivation regulates organ aging and lifespan programming (Liu *et al.*, 2023). In addition, IOZ researchers comprehensively revealed the protective effects of metformin on aging across dozens of tissues in aging male primates, significantly enhancing their cognitive function, thus providing a new milestone for aging mechanism research and intervention studies (Yang *et al.*, 2024).

## Reproductive and Developmental Regulation

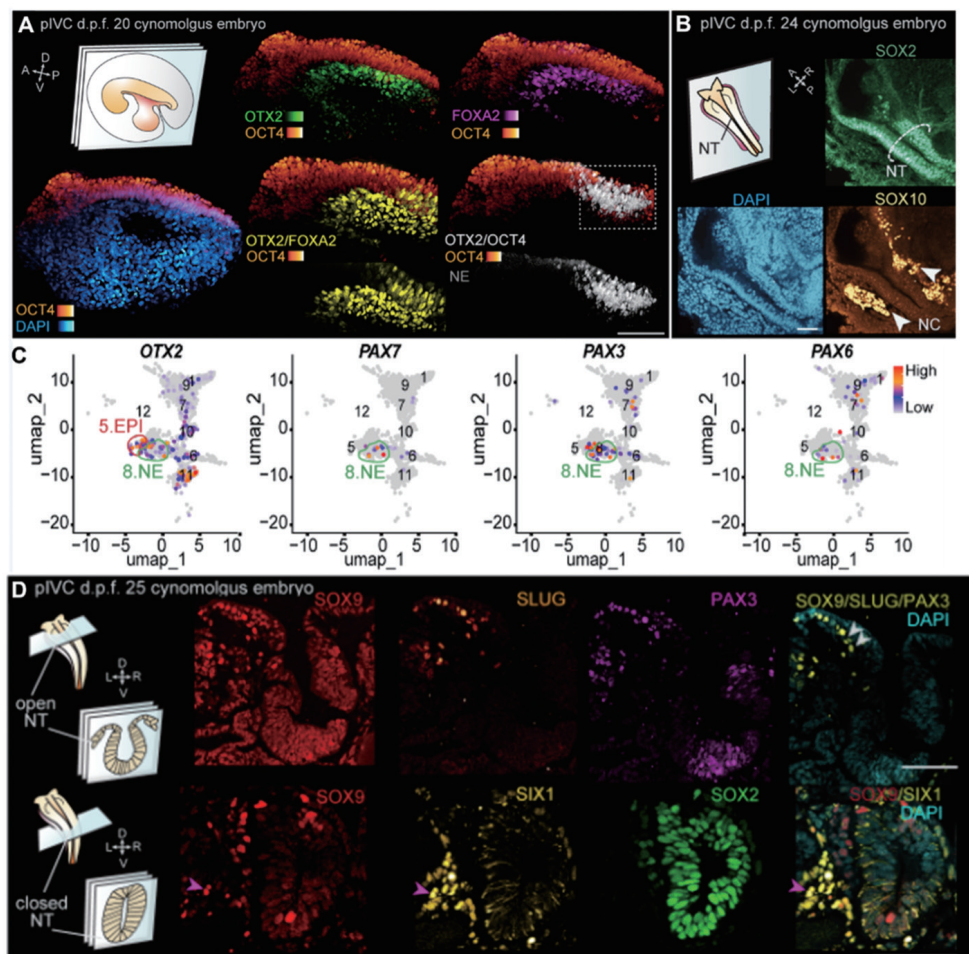
In the field of reproductive biology, significant progress has been made in understanding how sperm RNA plays a crucial role in the transgenerational inheritance of acquired traits (Chen *et al.*, 2016). By clarifying the regulatory mechanisms of sperm RNA, IOZ has unlocked a new area of study focused on how sperm RNA and its modifications influence early embryonic development (Wang *et al.*, 2018). This discovery not only advances the understanding of the molecular mechanisms involved in reproduction but also

opens up promising avenues for the prevention and treatment of acquired metabolic diseases (Chen *et al.*, 2016). The implications of sperm RNA's regulatory role extend beyond basic science, offering potential applications in improving reproductive health and addressing metabolic conditions passed down through generations.

In the field of embryonic development, groundbreaking advancements have been achieved by overcoming the previous limitations of long-term *in vitro* culture of primate embryos (Ma *et al.*, 2019). For the first time, human beings have successfully cultured early primate embryos for up to 25 days (Figure 2), a significant milestone in developmental biol-

gy (Ma *et al.*, 2019; Zhai *et al.*, 2022; Yan *et al.*, 2023). This breakthrough has profound implications for the study of early embryogenesis and the development of new reproductive technologies. Additionally, the creation of *in vitro* embryo models and artificial womb technologies represents a transformative leap forward in reproductive medicine (Zhai *et al.*, 2023; Yu *et al.*, 2024). These innovations provide new tools for investigating early embryonic development, offering fresh perspectives for improving assisted reproductive techniques and potentially addressing infertility challenges. Through these advances, the future of reproductive health and developmental research holds great promise.

Figure 2. Neurulation of the cynomolgus monkey embryo achieved from 3D blastocyst culture, adapted from Zhai *et al.* (2023).





## Stem Cell and Regenerative Medicine

The iPSC (induced pluripotent stem cell) technology can reprogram differentiated somatic cells back to pluripotent stem cells, thus has broad clinical application prospects. However, in the early years after the advent of the technology, whether iPSCs have the same developmental potential as that of embryonic stem cells (ESCs) was a primary question and obstacle hindering the basic research of iPSCs and their usage in regenerative medicine. By technological innovations, IOZ researchers generated, for the first time, iPSC-derived live and healthy mice and their progenies, demonstrating that iPSCs, like ESCs, can develop into healthy individuals independently (Zhao *et al.*, 2009). The work was regarded as a “breakthrough” in the iPSC research field, and has significantly promoted the development of iPSC technology.

Haploid cells contain only one set of chromosomes and could show a clear mutational phenotype due to a lack of the second gene copy, thus are ideal for recessive trait studies, genetic screening and disease gene identification. However, mammalian haploid cells are only restricted to germline cells and occasionally found in tumors with massive chromosome loss; therefore, their applications are constrained. IOZ researchers generated haploid embryonic stem cells of mice and rats with either androgenetic or parthenogenetic origin, and demonstrated that the androgenetic haploid ESCs can produce viable and fertile progenies after intracytoplasmic injection into mature oocytes (Li *et al.*, 2012), and the mouse parthenogenetic haploid ESCs can produce fertile progenies by substitution of the maternal genome (Wan *et al.*, 2013), indicating the potential application of haploid ESCs in assisted reproduction. In addition, the researchers generated world-first mouse-rat allodiploid ESCs by fusing haploid ESCs of the two

species, and they could serve as a valuable tool in studying XCI and genes with functional differences between species (Li *et al.*, 2016). They also revealed the factors necessary for crossing same-sex reproduction barriers in mammals, and, through MII oocyte injection or sperm coinjection with hypomethylated haploid ESCs carrying specific imprinted region deletions, obtained, for the first time, fertile bimaternal mice of normal growth, and full-term bipaternal mice, respectively (Li *et al.*, 2018). The findings provide significant value for understanding the evolution, regulation and function of genome imprinting, and for developing new approaches for animal reproduction.

IOZ also made some significant progress in genetic engineering. The researchers employed a novel technique enabling programmable chromosome fusion and successfully produced laboratory mice with novel and sustainable karyotypes (Figure 3), genetic changes that may take a million years to occur

Figure 3. Xiaoxiao (Tiny), one of the viable mice created from induced pluripotent stem cells, adapted from Zhao *et al.* (2009).



in nature. The results provide critical insight into how chromosomal rearrangements may influence evolution, and suggest the feasibility of chromosome-level engineering in mammals (Wang *et al.*, 2022). In another study, the IOZ team utilized a systematic engineering approach to develop a novel retrotransposon gene writer, which for the first time achieved all RNA-mediated, efficient, and targeted hit-and-run gene integration in mammalian cells (Chen *et al.*, 2024). This achievement enables all-RNA-mediated targeted gene integration in human cells.

Based on the clinical-grade human ESCs and the proprietary cell differentiation technologies developed by the National Stem Cell Resource Center, IOZ has been conducting more than 10 stem cell clinical study projects that have been registered with the relevant Chinese authorities. The clinical study projects include differentiated functional cell therapy for Parkinson’s disease, age-related macular degeneration, meniscus injury, etc. Five of the projects obtained IND approval from the National Medical Products Administration of China, and four of them have entered Phase II clinical trials.

## Integrated Management of Pest Insects and Rodents

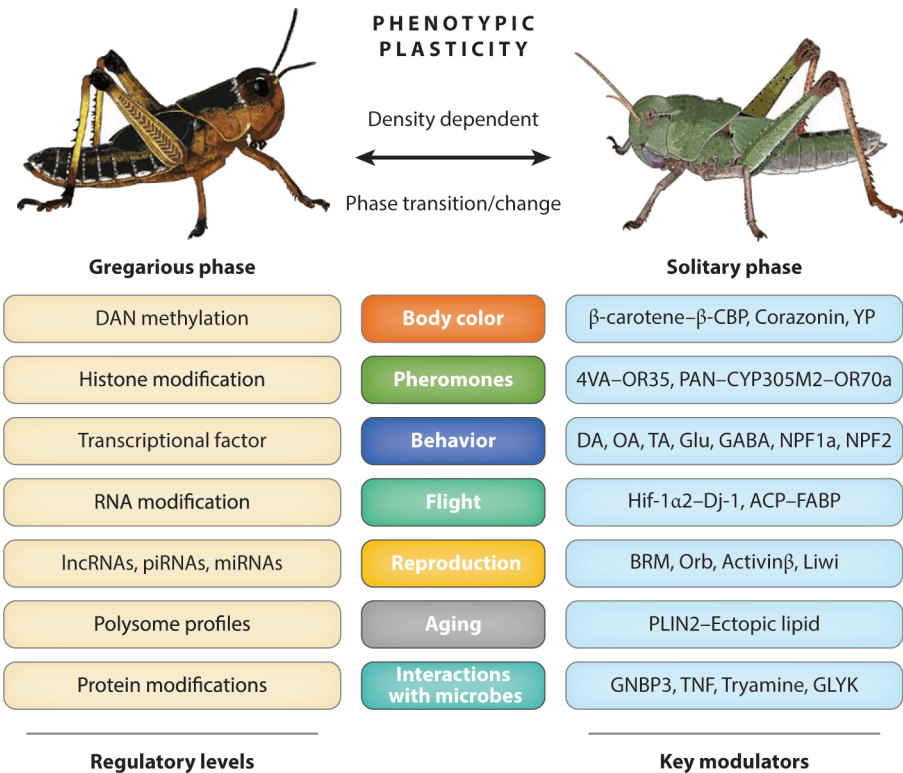
Pest outbreaks pose a substantial threat to agriculture and human health. To meet the social-economic demands of national priorities, IOZ is dedicated to advancing green control science for pest insects and rodents, focusing on critical issues related to understanding the fundamental mechanisms behind pest outbreaks,

elucidating disease transmission pathways, and developing precise control methods.

An integrated approach was proposed and has been instrumental in achieving significant control over locust infestations in China, reducing their impact to a minimal extent by the 1970s. Over the past decade, IOZ systematically elucidated the transcriptional regulation, metabolic synthesis, and epigenetic modifications in the phase transition between gregarious and solitary locusts (Figure 4; Guo *et al.*, 2024). The true aggregation pheromone 4-vinylanisole (4VA) (Guo *et al.*, 2020), the aposematic pheromone phenylacetonitrile (PAN), and the sex pheromone dibutyl phthalate (DBP) have been identified. Simultaneously, IOZ unveiled the joint function of body color and chemical signaling in locust group defense, elucidated the key

regulatory factors for the aging plasticity (Guo *et al.*, 2023) and the intergenerational transmission of gregarious characteristics (Zhu *et al.*, 2024), and achieved the genetic modification of host-generalist fungus *Metarhizium robertsii* and host-specialist fungus, *Metarhizium acridum*. These key findings have been considered as a significant breakthrough in international entomology over the past 50 years. The chemical communications and molecular mechanisms underlying the invasion success were investigated in two major invasive species in China, the red turpentine beetle (*Dendroctonus valens*) and pinewood nematode (*Bursaphelenchus xylophilus*). IOZ, for the first time, proposed and validated the hypotheses of “symbiotic invasion” and “re-invasion”, found the maintenance mechanism of multispecies symbiotic invasion mediated by

Figure 4. Overview of locust phenotypic plasticity and its regulatory network





semiochemicals, and established the theory of “symbiotic invasion” (Ning *et al.*, 2023).

Efficient transmission of plant viruses by insect vectors is one of the important determinants for viral disease outbreaks, which is a major threat to world agriculture. IOZ identified key cofactors in insect vectors for viral infection and transmission (Lu *et al.*, 2022) and revealed the mechanisms of the immune homeostasis between virus and vector (Wang *et al.*, 2022). For viral host plants, lncRNAs were found to play a role as molecular switches in balancing plant defense and growth, and RNA helicases have broad-spectrum functions in plant resistance against viruses by altering the dynamics of membraneless organelles (Liu *et al.*, 2022). IOZ researchers constructed a graph-based pangenome and innovatively leveraged pan-genomics to mine resistance genes in crops. They found that Ecdysone-induced protein 93 is a crucial regulator of gonadotro-

phic cycles in adult female *Aedes aegypti* mosquitoes, influencing their reproductive processes. Additionally, leucine aminopeptidase 1 in male mosquitoes plays a significant role in controlling egg deposition and hatchability in females, highlighting the intricate reproductive dynamics within this species. Furthermore, gut symbiont-derived sphingosine has been shown to modulate vector competence in *Aedes* mosquitoes, potentially impacting disease transmission (Sun *et al.*, 2024). To combat flavivirus transmission from animal hosts to mosquitoes, the IOZ researchers are exploring the use of a mosquito-delivered vaccine, which could provide a novel strategy for controlling the spread of mosquito-borne diseases (Wen, *et al.*, 2022). They also revealed that replication is the key barrier during the dual-host adaptation of mosquito-borne flaviviruses.

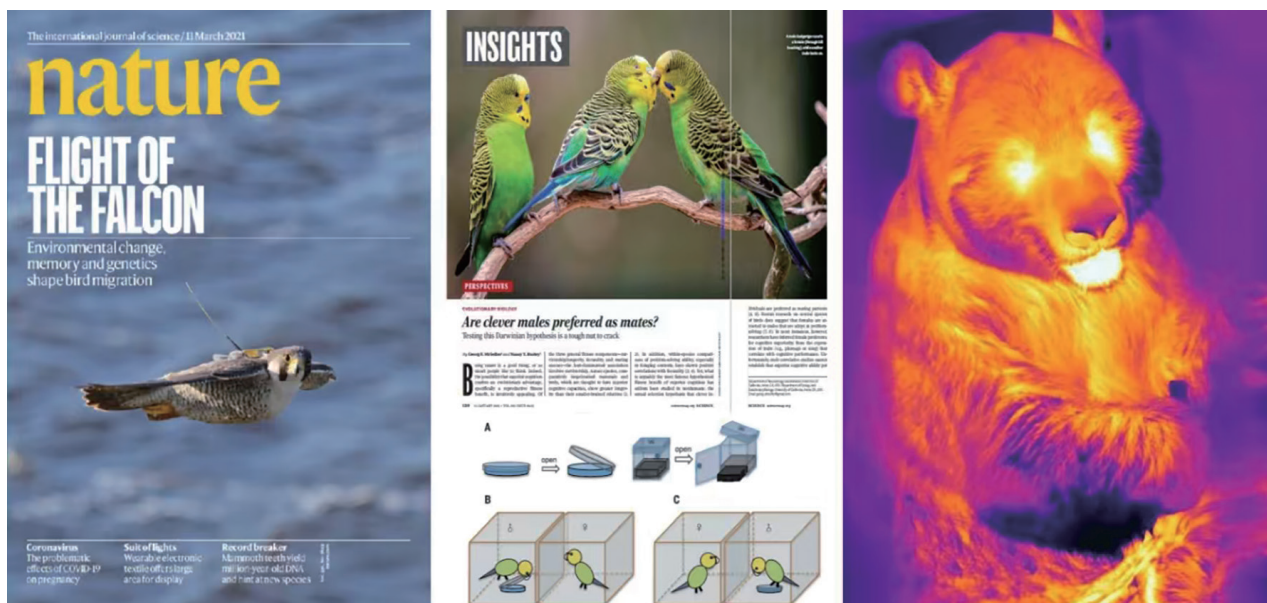
IOZ has developed new techniques for controlling insects and rodents. Researchers found EP-1

(Quinestrol and Levonorgestrel) can significantly inhibit the reproduction of many wild rodent species under both laboratory and field conditions. They have successively developed 12 kinds of baculoviral insecticides including 3 technical materials and 9 formulations since 2002. These products have been distributed domestically and internationally to combat pests such as the cotton bollworm, beet armyworm, tobacco cutworm, fall armyworm, and diamondback moth.

## Endangered Mechanisms and Conservation of Species

In the field of threatened animal conservation, a series of remarkable historic achievements have been made. IOZ scientists performed the pioneering studies of wild giant panda ecology in the 1960s, rediscovered the world's only seven crested ibis in

**Figure 5. Ecological and genetic mechanisms of adaptation in wild animals, adapted from Gu *et al.* (2021), Nie *et al.* (2015), and Striedter & Burley (2019).**



the 1980s, published a book on conservation biology in the early 1990s and established China's conservation biology discipline.

In recent years, IOZ continues to make remarkable research breakthroughs in conservation biology of wild animals (Figure 5). In the aspects of endangerment mechanisms and conservation strategies of threatened animals, based on the population genomics, metagenomics and meta-analysis analyses, IOZ researchers elucidated the genomic diversity, genetic structure, endangered history and adaptive evolution mechanisms of the giant panda, red panda, takin, saker falcon, and crocodile lizard, to propose precise conservation strategies for species and genetic diversity conservation (Hu *et al.*, 2020; Hu *et al.*, 2022; Xie *et al.*, 2022; Guan *et al.*, 2024). IOZ teams found that nature reserves serve as a refugia for amphibians and reptiles worldwide in the face of climate change (Mi *et al.*, 2023), and established a life strategy index to predict the threatened status of species based on the species' ecological potential, evolutionary potential, and colonization potential (Chen *et al.*, 2024). The achievements in the adaptive evolution and endangered mechanisms of giant pandas have won the second prize of the National Natural Science Award.

In the aspects of the adaptive strategies and evolutionary mechanisms of complex behaviors in wild animals, IOZ researchers integrated multiple years of satellite tracking data and population genomic data and revealed the main causes of the peregrine falcon's migratory routes and the key genes for long-distance migration. This achievement is selected as one of the top ten science advances and the top ten life

science advances in China (Gu *et al.*, 2021). IOZ researchers provided strong evidence for one of Darwin's hypotheses that cognitive performance could affect the sexual preference of the opposite sex (Chen *et al.*, 2019), observed the first case of allomaternal nursing in Old World monkeys (Xiang *et al.*, 2019), and found that the embryos of turtles can regulate their own sexual destinies through behavioral thermoregulation and the underlying molecular sensors for temperature detection (Ye *et al.*, 2019).

## Animal Taxonomy and Evolution

Since the establishment of CAS 75 years ago, a focus of IOZ has been on key scientific challenges in animal diversity and taxonomy and has conducted extensive work such as resource surveys, taxonomic identification, specimen preservation, distribution patterns, and strategic utilization of animal resources. These studies have driven a paradigm shift in taxonomic research and played a leading role in biodiversity surveys and taxonomic studies in China. The Institute has organized and published a series of major zoological works such as *Fauna Sinica* (Zheng *et al.*, 1978; Xue *et al.*, 1999; Qiao *et al.*, 2018; Wu *et al.*, 2023), *Economic Animals Fauna of China*, *Economic Insect Fauna of China*, and *Animal Atlas of China* (Figure 6). The Institute has led eight national resource survey projects and two key national R&D programs which identified more than 70,000 species, and more than 9,000 new species. In addition, the Institute formulated international standards for the collection, identification, storage and sharing of animal specimens, led the development of the "Belt

and Road" national biological specimen preservation, which has become the national center for the modernization of animal specimens (Figure 6). It has also built the largest platform for animal resource preservation in Asia. Based on these achievements, IOZ researchers have won a number of awards. For example, the projects "Blockade and Control Technology of Invasive Pest Potato Beetles" and "Ecological Management Technology of Xinjiang Cotton Aphid" have won the "Second Prize of the National Science and Technology Progress Award" (Zhang *et al.*, 2007). The "Identification and Control of Invasive Pests" has won the "Outstanding Scientific and Technological Achievement Award of Chinese Academy of Sciences".

Meanwhile, IOZ researchers have conducted research surrounding the central theme of "the Formation and Maintenance Mechanism of Animal Diversity", by integrating animal diversity information, multi-omics technology, bioinformatics and other methods. Important studies of this kind include "the Evolution of Beak Diversity in Highland Birds", "the Convergence of Alpine Phenotypes from the Sea" (Liu *et al.*, 2023), "the Convergent Evolution of Stickleback Species", "the Influence of Ecological Factors and Evolutionary History on the Formation of Avian Diversity" (Cheng *et al.*, 2021), "the Vertical Differentiation and Diversity of Mountain Birds" (Qu *et al.*, 2020), "the Diversity Distribution Pattern of Asian Mammals" and "the Umbrella Species Effect of Highland Pika Under Climate Change" (Chen *et al.*, 2023). In addition, a new mechanism of cell population diversification and adaptation, and the largest dataset of active DNA transposons have been found (Zhang *et al.*, 2024). In





Figure 6. National Animal Collection Resource Center and *Fauna Sinica*.

summary, IOZ systematically revealed the joint effects of genetic diversity, species distribution, ecology, and evolution.

## Conclusions

In the new era, IOZ has always remained true to its original aspiration in serving the national campaigns such as the “Healthy

China” and the “Beautiful China,” focusing on the major needs of China in the fields of human health, modern agriculture, and biodiversity conservation to carry out basic, prospective, and strategic research and conduct core technological research and development. Under IOZ, there are two state key laboratories, two national resource centers, one national museum, one national observation

and research station, one national and one CAS research facility. In the future, IOZ will strive to seize the science highland by optimizing its layout of disciplinary development, attracting and fostering talents, and building world-class research facilities and platforms, aiming to be a leading institution for the continuous innovation and development of life science both in China and the world.

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