# The unseen invaders: tracking phylogeographic dynamics and genetic diversity of cryptic *Pomacea canaliculata and P. maculata* (Golden Apple Snails) across Taiwan

Pritam Banerjee[1,2], Gobinda Dey[2,3], Jyoti Prakash Maity[4], Kathryn A. Stewart[5], Raju Kumar Sharma[6], Michael W.Y. Chan[2], Kuan Hsien Lee[7, *], Chien-Yen Chen[8, 9*]

[1]*Department of Environmental Science, Policy, and Management, University of California, University Avenue and Oxford St, Berkeley, CA 94720, USA*

[2]*Department of Biomedical Sciences, Graduate Institute of Molecular Biology, National Chung Cheng University, 168 University Road, Min-Hsiung, Chiayi County 62102, Taiwan.*

[3]*Department of Agricultural Chemistry, National Taiwan University, Taipei, 106319, Taiwan*

[4]*Department of Chemistry, School of Applied Sciences, KIIT Deemed to be University, Bhubaneswar, Odisha 751024, India.*

[5]*Institute of Environmental Sciences, Leiden University, 2333 CC Leiden, The Netherlands*

[6]*Doctoral Program in Science, Technology, Environment, and Mathematics, Department of Earth and Environmental Sciences, National Chung Cheng University, 168 University Rd., Chiayi County, Min-Hsiung, 62102, Taiwan*

[7]*Department of Emergency Medicine, Ditmanson Medical Foundation Chia-Yi Christian Hospital*

[8]*Department of Earth and Environmental Sciences, National Chung Cheng University, 168 University Road, Min-Hsiung, Chiayi County 62102, Taiwan.*

[9]*Center for Nano Bio-Detection, Center for Innovative Research on Aging Society, AIM-HI, National Chung Cheng University, Chiayi 62102, Taiwan*

**Corresponding Author**

Chien-Yen Chen[8, 9*] and Kuan Hsien Lee[7, *]

*Address for correspondence: Department of Earth and Environmental Sciences, National Chung Cheng University, 168 University Road, Min-Hsiung, Chiayi County 62102, Taiwan. and Department of Emergency Medicine, Ditmanson Medical Foundation Chia-Yi Christian Hospital Email: chien-yen.chen@oriel.oxon.org; yen@eq.ccu.edu.tw (C.Y. Chen); seriex.lee@gmail.com (K.H.Lee) Tel.: +886-5-2720411 (ext. 66220); Fax: +886-5-2720807.

## Abstract

The cryptic invasion of golden apple snails (*Pomacea* spp.) in Taiwan has caused significant ecological and economical damage, however, their management remains difficult due to inadequate taxonomic identification, complex phylogeny and limited population genetic information. We aim to understand the current distribution, putative population of origin, genetic diversity and potential path of invasion of *Pomacea* spp. across Taiwan to aid in improved mitigation approaches. The present investigation conducted a nationwide survey with 254 samples collected from 41 locations from 14 counties or cities across Taiwan. We identified and compared the genetic diversity of *P. canaliculata* and *P. maculata* based on mitochondrial COI in Taiwan, as well as other introduced and native countries (publicly available COI data) to understand the possible paths of invasion in Taiwan. Based on mitochondrial COI barcoding, sympatric but heterogeneous distributions of invasive *P. canaliculata* (widespread) and *P. maculata* (mostly restricted to Southern Taiwan) were noted. Our haplotype analysis and mismatch distribution suggest multiple introductions of *P. canaliculata* likely originated directly from Argentina, whereas *P. maculata* was probably introduced from a single, or a few, introduction event(s) from Argentina and Brazil, instead of directly from Brazil. Our population genetic data further demonstrated higher haplotype and genetic diversity for *P. canaliculata* and *P. maculata* in Taiwan

compared to other Asian countries, which also suggest multiple introductions affected Taiwan. Comparing the introduction history, current distribution and cryptic nature of *P. canaliculata* and *P. maculata* in Taiwan, we posit that *P. maculata* was likely introduced along with *P. canaliculata* (either intentionally or unintentionally), whereas, *P. canaliculata* plausibly has several diverse introduction histories. Based on our current understanding, the establishment of *Pomacea* spp., is alarming and widespread beyond geopolitical borders, requiring a concerted and expedited national and international invasive species mitigation program.

**Keywords**: agricultural pest, biological invasion, DNA barcoding, golden apple snails, mitochondrial COI, population genetics, phylogeography

## 1. INTRODUCTION

Invasive alien species are major drivers of biodiversity loss and have a massive impact on the economic status of invaded countries (Linders et al., 2019; McNeely, 2001), even considered the second most economically destructive event after natural hazards such as earthquakes or floods (Turbelin et al., 2023). Making matters worse, morphologically similar species may invade together, causing a masking effect to species distribution and establishment, further hindering effective management practices (Saltonstall et al., 2002). Without in-depth knowledge about the origins and population genetics of invasive species, management of natural landscapes and aquascapes is rendered speculative (Sakai et al., 2001).

Amongst some of the worst invasive species, *Pomacea canaliculata* native to South America, have been both intentionally and unintentionally introduced in several countries (Lowe et al., 2000; Zhao et al., 2022), recently becoming a serious threat to agricultural and economic

development worldwide (e.g., Asia, Europe, Africa, North America, and the Pacific Islands) (Ranamukhaarachchi & Wickramasinghe, 2006; Hayes et al., 2008; Buddie et al., 2021). Other species of *Pomacea* (e.g., *P. macualta*, *P. scalaris*) have also been introduced across numerous countries, albeit with less described environmental or economic impact. However, understanding the origins or sources of these invasive species has not been straightforward (Hayes et al., 2008). The systematics of *Pomacea* spp. is complex and murky due to high morphological similarity and phenotypic plasticity (Rama Rao et al., 2018), causing frequent misidentifications that lead to false biodiversity information, potentially obstructing effective invasion management (Cowieet al., 2006). Among the species of *Pomacea*, *P. canaliculata*, and *P. maculata* are destructive and morphologically cryptic in nature, thus difficult to distinguish with field observation alone (Rama Rao et al., 2018; Banerjee et al., 2022). Traditional methods using morphological observation (outer shell, internal tissue, and egg morphology) have failed to distinguish them properly because of their overlapping characteristics, thereby obscuring their actual distribution (Rama Rao et al., 2018; Banerjee et al., 2022). Molecular method implementation has recently been found to be the best option for distinguishing *P. canaliculata* and *P. maculata* (Matsukura & Wada, 2017; Banerjee et al., 2022), with population genetics analysis recently conducted to comprehend the invasive origin and distribution of *P. canaliculata* and *P. maculata* across Asia (Hayes et al., 2008; Yang et al., 2018; Dumidae et al., 2021; Yang et al., 2022; Zhao et al., 2022; Liu et al., 2023). Nevertheless, most analyses have not included samples from Taiwan, with the exception of a few sequences reported in Hayes et al. (2008).

Despite Taiwan encompassing a high percentage of the world's biodiversity compared to its landmass, and housing an extraordinary array of fauna that comprises evolutionary lineages from both Palearctic and Indomalaya biogeographic realms (He et al., 2018; Päckert et al, 2009),

as an island, Taiwan is highly sensitive to invasive species (Lee et al., 2019). Indeed, several invasive alien species have been introduced to Taiwan which has negatively affected its local biodiversity. Specifically, the introduction and establishment of presumably several *Pomacea* spp. (Golden apple snails) in Taiwan have resulted in serious ecological and economical destruction.

Historically, Taiwan was noted as the first Asian region to import *Pomacea* spp. for commercial utilization (dietary protein supplementation and aquarium trades) during 1979-1982 (Joshi & Sebastian, 2003; Yang et al, 2006; Cheng and Kao, 2006; Wu et al, 2010). Subsequently, *Pomacea* spp. were introduced into other Asian countries for economical trades. Initially in Taiwan, *Pomacea* spp. were considered a healthy diet alternative that contained high protein, minerals, and vitamins, thus motivating farmers in its cultivation and trade (Cheng and Kao, 2006; Hayes et al., 2008). However, the trade of *Pomacea* spp. was soon terminated because of the snail's poor texture and high production cost. Exacerbating *Pomacea* spp. invasion, farmers were not content with the low market value and thereafter released a large number of *Pomacea* spp. into the neighboring countryside (Cheng and Kao, 2006; Chiu et al., 2014). Almost immediately after release, *Pomacea* spp. became a serious threat to the local agricultural system, possibly as a result of its high reproduction rate, tolerance to various environmental stressors, and voracious appetite (Joshi & Sebastian, 2003). The problem with *Pomacea* was first recognized in 1982, raising national-level attention due to numerous damages to local agronomy; *Pomacea* spp. spread into various agricultural lands, as well as natural wetlands impacting cultivated plants such as rice seedlings, taro, and lotus, even destroying several cultivation fields (Cheng & Kao, 2006). Making matters worse, *Pomacea* spp. acts as a vector for various zoonotic diseases (e.g., *Angiostrongylus cantonensis*). Several cases of human eosinophilic meningitis have been reported in Taiwan through either direct (consumption of raw snail meat) or indirect (contaminated food) involvement

of *Pomacea* spp. (Tsai et al., 2001). More specifically, enormous annual economic losses were reported due to these invasive gastropods (Globally: 3.94 billion USD; in Asia: 3.71 billion since 1961; (Jiang et al., 2022)), with Asian countries as the predominantly affected region (Jiang et al., 2022). Across all reported gastropods, *Pomacea* spp. (particularly *P. canaliculata*) were noted to be the major contributor. Despite this, little attention has thus far been paid to understanding the invasive biology and population genetics of *Pomacea* spp. in Taiwan (Cheng & Kao, 2006; Banerjee et al., 2022), further contributing to their regional (Taiwan) and potentially global spread (Sakai et al., 2001)

Originally, two species of *Pomacea, P. canaliculata* and *P. scalaris,* were presumed in Taiwan based solely via morphological identification (Cheng et al., 2006; Yang et al., 2008; Wu et al., 2010). However, a more recent molecular study revealed the presence of another species, *P. macualta*, hiding with its cryptic relative *P. canaliculata* (Banerjee et al., 2022). Out of the three *Pomacea* species currently in Taiwan, *P. scalaris,* is considered less destructive due to its smaller hatching size and inferior growth performance (Wu et al., 2010), thus assumed geographically restricted mostly in the South of the country (Wu et al., 2011). Conversely, *P. canaliculata* and *P. maculata* are highly invasive and eco-economically destructive in nature, yet their actual distribution remains unknown (Banerjee et al., 2022). Here, our study aims to generate accurate identification of invasive *Pomacea* spp., including their nation-wide distribution and genetic diversity to better aid in adaptive management strategies. By doing so, we aspire to elucidate their (i) possible origin, thereby minimizing ongoing propagation events, and (ii) proper distribution and putative invasion path within Taiwan to maximize future eradication measures. As subsequent introductions of *P. canaliculata* and *P. maculata* may occur from trades between Taiwan into other Asian countries (e.g., China, Japan, Thailand, Malaysia) (Joshi & Sebastian, 2003), the current

lack of molecular data makes global invasive management efforts especially difficult. Here too our study may expand knowledge of global *Pomacea* distributions. To accomplish these aims, our survey conducted collections across 41 different sampling locations within 14 counties or cities across Taiwan to reveal the origin, distribution, and population genetic structure of *P. canaliculata* and *P. maculata*. We then compared the genetic diversity of *P. canaliculata* and *P. maculata* with other countries (invaded or native) to understand their possible paths of invasion.

## 2. METHODS

### 2.1 Snail sample collection and preservation

In Taiwan, *Pomacea* were presumably introduced through human activities and later distributed nationwide. Since this distribution may be affected by agricultural land or other (human-induced) distribution barriers (e.g., roads, mountains), sampling locations (populations) are centered around or near cities to understand the current nationwide distribution. We then further subdivided Taiwan into different regional parts coordinates (e.g., Southern, Northern, Eastern, Western and Middle) to more generally understand the current distribution of *Pomacea* for downstream management decisions. To accomplish this, distribution of *Pomacea* spp. were surveyed across 41 locations across 14 counties or cities in Taiwan, including Hualien (HU) and Taitung (TT) from the East, Yunlin (YL), Changhua (CH), Taichung (TC), and Miaoli (MIO) from the Middle of island, Hsinchu (HS), Taoyuan (TY), Taipei (TP), and Yilan (YI) from the North, and Pingtung (PT), Kaohsiung (KSH), Tainan (TN), and Chiayi (CY) from the South. This was achieved via manual hand-collection from crop fields and drainage ditches from April 2020 to January 2023. Sampling

locations and the number of samples are shown in Figure 1 and APPENDIX 1. Collected samples were transported directly to the laboratory as soon as possible (as per sampling protocols). In cases of remote sampling or locations away from the laboratory, specimens were heat-shocked using hot water (100$^{o}$ C) for 10-15 minutes and subsequently kept in an ice box in separate sampling bags to avoid contamination. Once in the laboratory, samples were washed thoroughly to remove soil and debris, and then if not previously applied, were heat-shocked using a microwave to remove inner tissue from shells. After removing the inner tissue from the shell, 20-50 mg of foot tissue was used to extract DNA to avoid parasite contamination, and the rest of the tissue was stored in 95% ethanol.

**2.2 Genomic DNA extraction and amplification**

Genomic DNA was extracted from the 20-50 mg *Pomacea* spp. foot tissues with the help of a DNeasy Blood and Tissue Kits (Qiagen, Germany), similar to Banerjee et al. (2022). The mitochondrial DNA barcode of *Pomacea* spp. was amplified using a conventional PCR (CLUBIO PCR system, 96-well, CB series; Scientific Biotech Corp., Taiwan) with species-specific primers: PcanCOI (F) (5'-TGG GGT ATG ATC AGG CC-3') and PinsCOI (F) (5'-ATC TGC TGC TGT TGA AAG-3') and HCO2198 (R) (5'-TAA ACT TCA GGG TGA CCA AAA AAT CA-3') for initial identification of *P. canaliculata* and *P. maculata* (Matsukura et al, 2008). The double distilled water and DNA from Giant African land snail (*Lissachatina fulica*) were used as the negative control followed by the previous study (Banerjee et al., 2022). Furthermore, successfully identified specimens from initial screening were amplified using universal mitochondrial COI primers: LCO1490 (5' GGT CAA CAA ATC ATA AAG ATA TTG G-3') and HCO2198 (5' TAA ACT TCA GGG TGA CAA AAA AAT CA-3') for a 658bp fragment. The final PCR reaction

contained 5 µL 5x Fast-RunTM Taq Master Mix with Dye (Protech Technology Enterprise, Taiwan), 0.5 µL of 10 pmol forward and reverse primers (Genomics, Taiwan), 1 µL DNA template from the tissue sample, for a final volume of 25 µL using ultrapure water (Thermo Fisher, USA). The PCR conditions were 5 min at 94°C, 35 cycles of 30s at 94°C (denaturation), 30s at 55°C (annealing), and 1 min at 72°C (extension), then the final extension was at 72°C for 5 min. All amplified products were sent to Genomics Co., Ltd, Taiwan for sequencing in both directions. All sequences derived from the present study were aligned using the MUSCLE algorithm (Edgar, 2004), and edited in MEGA 11 (Tamura et al, 2021), and GENEIOUS V11.1.5 (Kearse et al., 2012). A total of 254 of *Pomacea* spp. were amplified from 41 sampling spots in Taiwan.

### 2.3 Mitochondrial COI

The identification of 254 *Pomacea* spp. were accomplished via sequence similarity based on the NCBI database (https://blast.ncbi.nlm.nih.gov/Blast.cgi) with the highest level of query cover and percentage identity. To clarify the origins of our *Pomacea* spp. samples across Taiwan, previously identified and uploaded sequences of *P. canaliculata* and *P. maculata* were downloaded from NCBI which represent different geographic locations (Hayes et al., 2008; Kannan et al., 2021; Liu et al, 2019; Lv et al., 2013; Yang et al., 2018). Sequence quality was examined in Geneious v11.1.5 (Kearse et al., 2012), BioEdit 7.2 (Hall, 1999), and DnaSP 6.12.03 (Librado & Rozas, 2009) and filtering sequences published after 2007 to clearly distinguish *P. maculata* from *P. canaliculata* (Yang et al., 2018). A total of 1173 (948 *P. canaliculata* and 225 *P. maculata*) sequences were selected for final evaluation (APPENDIX 2, 3), and further screening was performed after a test run of phylogenetic and haplotyping network analysis (details in sections 2.5). Sequence length variation was adjusted and trimmed manually in Mega 11 (Tamura et al., 2021), allowing us to

prepare three datasets, (i) the sequences generated from the present study (samples only from Taiwan) represented 611bp for *P. maculata* and *P. canaliculata* as Dataset 1, and global sequences (Taiwan and other previously published sequences) datasets represent (ii) 579bp for *P. maculata* as Dataset 2, and (iii) 569bp for *P. canaliculata* as Dataset 3.

### 2.4 DNA polymorphism and phylogeographic distribution

All sequences were aligned in Mega 11 (Tamura et al., 2021) using the MUSCLE algorithm (Edgar, 2004), and DNASP 6.12.03 (Librado & Rozas, 2009) was used to calculate the number of haplotypes (H) and polymorphic sites (S) as well as haplotype diversity (Hd), nucleotide diversities ($\pi$) and average number of difference (K). Neutrality tests such as Tajima's D and Fu's Fs (Fu, 1997) were performed to understand signatures of population expansion using ARLEQUIN 3.5.2.2 (Excoffier & Lischer, 2010). Furthermore, to understand the level of genetic difference between the populations, pairwise $F_{st}$ values were calculated using ARLEQUIN 3.5.2.2 (Excoffier & Lischer, 2010), and the level of gene flow/effective migration rate was calculated using the following formula: $Nm=((1/F_{ST}) -1)/2)$ (Hudson et al., 1992). The connections between haplotypes and their phylogeographic distribution were analyzed using the median-joining network implemented in PopArt 1.7 (Leigh & Bryant, 2015). Furthermore, the mismatch distribution analyses were performed in DNASP 6.12.03 (Librado & Rozas, 2009) to understand the genetic variation for the population of *P. canaliculata* and *P. maculata*.

### 2.5 Phylogenetic analysis

The maximum-likelihood (ML) method with the GTR+G model (evolutionary model tested in jModelTest V2.1.10) was used based on 1000 bootstrap replicates (Felsenstein, 1985) to build

phylogenetic relations in Mega 11 (Tamura et al., 2021). The sequences of *P. lineata* (FJ710310, FJ710311), *P. paludosa* (EF514960, EU528590), *P. diffusa* (EU528547, FJ710317), and *P. scalaris* (collected from TC, in the present study) were added as outgroups. The inter- and intra-specific genetic distance (p-distance) of *P. canaliculata* and *P. maculata* were calculated using Mega 11.

## 3. RESULTS

### 3.1 Species distribution

Based on our collections across Taiwan, the distribution of *P. canaliculata* and *P. maculata* was observed to be sympatric, however heterogeneous. *P. canaliculata* was distributed throughout Taiwan and represented the dominant species (≈ 97%), whereas *P. maculata* represented ≈ 3% of the total number of collected individuals (Fig. 1; Map created using "ArcGIS 10.7 software; ESRI Inc., Redlands, CA, USA").

### 3.2 Phylogenic analysis

Phylogenetic analysis of all *Pomacea* spp. sequences with ML method represented with two well-separated monophyletic clades for *P. canaliculata* and *P. maculata,* with each outgroup species (*P. lineata*, *P. paludosa*, *P. diffusa*, and *P. scalaris*) representing a different clade (Fig. 2). Furthermore, there were three clades noted for *P. canaliculata* (Clade A, B, and C) and two clades for *P. maculata* (Clade D and E). However, the bootstrap value was noted to be significantly less in separating *P. canaliculata* and *P. maculata* (Fig. 2). The genetic distance (p-distance) within *P.*

*canaliculata* and *P. maculata* were 0.02 and 0.01 respectively, and genetic distance (p-distance) in between them were 0.0928 (APPENDIX 4)

### 3.3 Haplotype distribution and network analysis

#### 3.3.1 Phylogeography of *P. canaliculata* in Taiwan

A total number of 246 sequences of *P. canaliculata* from 14 different cities/counties revealed the presence of 18 haplotypes across Taiwan (Fig. 3, Table 1, 4). The most frequent haplotypes were Pc_Hap 3 (63%), Pc_Hap 2 (17.88%), and Pc_Hap 8 (6.5%) (APPENDIX 5). Pc_Hap 3 was widely distributed and found in every sampling site, whereas Pc_Hap 2 was detected in eleven sites, and Pc_Hap 8 was detected in only three sites (APPENDIX 5). The other rare but shared haplotypes were Pc_Hap 4 (2.03%), Pc_Hap 5 (2.03%), Pc_Hap 9 (1.22%), and Pc_Hap 11 (2.84%). Furthermore, eleven unique haplotypes were reported from Tainan (Total 3: Pc_Hap 1, 6, 7), Hualien (Total 2: Pc_Hap 17, 18), Chiayi (Total 2: Pc_Hap 10, 12), Yilan (Total 2: Pc_Hap 13, 14), Changhua (Pc_Hap 15) and Taitung (Pc_Hap 16) (Table 2).

Our haplotype network analysis of *P. canaliculata* showed the presence of three different networks across Taiwan (Fig. 3). The first network (Network Tw-A) was most dominant, representing 69.90% of all sequences with seven haplotypes (Pc_Hap 1, 3, 6, 11, 12,15,16) were found to correspond with Clade A in the phylogenetic tree (Fig. 2, APPENDIX 5). Among the seven haplotypes, Pc_Hap 3 was widespread and dominant; the other 6 haplotypes included five unique haplotypes from Tainan (Total 2: Pc_Hap 1, 6), Taitung (Pc_Hap 16), Chiayi (Pc_Hap 12), Changhua (Pc_Hap 15), and one shared by Chiayi, Yilan, Changhua, and Taichung (Pc_Hap 11). All of these haplotypes were only a few mutational steps away from one another, however, Pc_Hap

3 demonstrated the highest number of connections which created a star-like appearance, suggesting that Pc_Hap 3 could be a potential founder haplotype of Network Tw-A. Network Tw-B (found to correspond with Clade B) represented 30.49% of all sequences with four shared haplotypes including Pc_Hap 2 (widespread), Pc_Hap 4 (shared by Tainan, Kaohsiung, and Taichung), Pc_Hap 8 (shared by Pingtung, Yunlin, Hualien), Pc_Hap 5 (shared by Tainan, Pingtung, Chiayi, Taipei, Taichung), and five unique haplotypes from Hualien (Total 2: Pc_Hap 17, 18), Yilan (Total 2: Pc_Hap 13, 14) and Chiayi (Pc_Hap 10). All these haplotypes created a star-like appearance around Pc_Hap 2, again suggested to be a potential founder haplotype of Network Tw-B. The Network Tw-C (found to correspond with Clade C) was represented by only 1.62% of all sequences with one shared haplotype Pc_Hap 9 (Kaohsiung, Hsinchu, Changhua), and one unique haplotype Pc_Hap 7 (Tainan). The distribution Network Tw-C was limited to the South (Kaohsiung, Tainan), Middle (Changhua), and North part (Hsinchu). Compared to other cities/counties Tainan, Chiayi, Yilan, and Hualien were reported to have a higher number of shared and unique haplotypes, and interestingly the South region of Taiwan had a higher number of shared as well as unique haplotypes (Table 1,3).

### 3.3.2 Phylogeography of *P. canaliculata* worldwide

The combined mitochondrial COI datasets of *P. canaliculata* with 948 sequences across Africa, Asia, North America, Oceania, and South America were compared to one another and revealed the presence of a total of 65 haplotypes. Among all haplotypes, Wc_Hap 1 exhibited the widest geographical distribution, representing 44.67% of all global sequences (APPENDIX 6). The Wc_Hap 20 originated mostly in China (with one sequence from Japan and Taiwan) and represented the second-highest percentage of all sequences (33.75%). Despite Wc_Hap 20 prevalence, it was not widely geographically distributed. The Wc_Hap 16 represented 5.89% of

all sequences, mostly isolated in Taiwan, with a few representatives from Japan, Malaysia, China, and Kenya. Other haplotypes were less than 5% of all sequences available and represented a vast majority of unique haplotypes (APPENDIX 6). Taiwan was represented with 18 haplotypes in Dataset 1 (619bp), however in the global dataset (Dataset 3 = 569bp; 16 haplotypes from Taiwan) a slight decrease in the number of haplotypes was noted (comparison of changes recorded in APPENDIX 7). Across all countries with sequences, the total number of unique haplotypes were found to be the most prevalent in Argentina, Uruguay, Philippines, China, and Taiwan.

Our haplotype network analysis of *P. canaliculata* with a combined dataset (Dataset 3) showed three distinct networks i.e., Network Wc-A, Network Wc-B, and Network Wc-C (Fig. 4). Network Wc-A, representing 32 haplotypes, including the most widely shared dominant Wc_Hap 1, and 31 unique haplotypes from Argentina (Total 11; Wc_Hap 3-8, 10, 11, 12, 15, 17), Taiwan (Total 5; Wc_Hap 24, 26, 30, 31, 34), Philippines (Total 6; Wc_Hap 52, 53, 55, 57, 59, 61), Indonesia (Total 2; Wc_Hap 48, 49), Thailand (Total 2; Wc_Hap 62, 63), USA (Total 1; Wc_Hap 64), China (Total 1; Wc_Hap 35), Myanmar (Total 1; Wc_Hap 51), Chile (Total 1; Wc_Hap 47), Malaysia (Total 1; Wc_Hap 50). All of those unique haplotypes were connected with Wc_Hap 1. The Wc_Hap 1 was thus inferred as the founder haplotype due to its highest number of connections with other haplotypes, and its widespread global distribution. This haplotype was also found to be a widespread and dominant haplotype in Taiwan, as well as shared by other Asian countries.

Network Wc-B represented 22 haplotypes, including five shared; Wc_Hap 16 (Taiwan, China, Japan, Malaysia, and Kenya), Wc_Hap 20 (China, Taiwan, and Philippines), Wc_Hap 21 (China, Philippines), Wc_Hap 23 (Taiwan, China, Japan, Philippines), and Wc_Hap 36 (China and Japan) and 17 unique haplotypes from Taiwan (Total 5; Wc_Hap 25, 27, 29, 32, 33), China (Total 5; Wc_Hap 22, 43-46), Uruguay (Total 2; Wc_Hap 40, 41), Philippines (Total 4; Wc_Hap

54, 56, 58, 60), and Argentina (Total 1; Wc_Hap 14). All the shared haplotypes in Network Wc-B were reported from Taiwan, except Wc_Hap 21, and Wc_Hap 36, which were one to three mutational steps away from the shared one. The Wc_Hap 27 (from Taiwan) was noted to be the common connection among all the haplotypes in Network Wc-B. Lastly, Network Wc-C represented 11 haplotypes, including 2 shared; Wc_Hap 28 (Taiwan, Japan, and Singapore), Wc_Hap 9 (Taiwan and Argentina), and 9 unique haplotypes from Argentina (Total 5; Wc_Hap 2, 13, 17, 18, 65), and Uruguay (Total 4; Wc_Hap 37-39, 42). Among the haplotypes in Network Wc-C, Wc_Hap 9 was inferred to be the common ancestor, shared between Argentina and Taiwan. Most of the haplotypes from introduced regions (Asian countries and African countries) were found to be associated with Network Wc-A and B, however, few representatives were noted in Network Wc-C.

The comparison of representative sequence in Dataset 1 (representing only Taiwan sequences) and Dataset 2 (sequence from Taiwan as well as other native and non-native countries), revealed that haplotypes of Network Tw-A, Network Tw-B, and Network Tw-C, represent in Network Wc-A, Network Wc-B, and Network Wc-C, respectively without any interchange (APPENDIX 6).

### 3.3.3 Phylogeography of *P. maculata* in Taiwan

In Taiwan, eight sequences of *P. maculata* represented four different haplotypes, where Pm_Hap 19 (62.50%) was the most common and the other three haplotypes (Pm_Hap 20-22) were represented by single sequences (APPENDIX 5). Haplotype network analysis of *P. maculata* showed the presence of a single network, where Pm_Hap 1 was a few mutational steps away, however, Pm_Hap 20-22 were more closely related (Fig. 3).

### 3.3.4 Phylogeography of *P. maculata* worldwide

A total of 226 sequences of worldwide *P. maculata* revealed 46 haplotypes. The most frequently widely distributed haplotype was Wm_Hap 6 (representing 37.61% of all sequences), followed by Wm_Hap 9 (13.27%) from Japan, USA, and Argentina, where Wm_Hap 10 (9.73%) was found in USA and Argentina (APPENDIX 8). Several haplotypes were observed to have a single representation, mostly from Brazil and Argentina. In the 579bp *P. maculata* sequence database (Dataset 2), Taiwan contained four haplotypes similar to our analysis of Dataset 1 (comparison of positions in Dataset 1 and 3 recorded in APPENDIX 7).

The haplotype network analysis of *P. maculata* with a combined dataset (Dataset 2) showed seven different networks (Network Wm-A-G). Network Wm-A was widespread, consisting of 29 haplotypes which include four shared haplotypes; Wm_Hap 6 was the most widely distributed and Wm_Hap 10 was shared by Argentina and USA, Wm_Hap 9 was shared by USA, Japan, and Argentina, and Wm_Hap 1 was shared by Taiwan, Argentina, and Uruguay, including unique haplotypes from Argentina (Total 6; Wm_Hap 13-18), Uruguay (Total 3; Wm_Hap 2-4), Taiwan (Total 2; Wm_Hap 7, 8), Vietnam (Wm_Hap 5), USA (Wm_Hap 11), and 11 unique to Brazil (Fig. 5). All the other Networks were unique to Brazil. Among the shared haplotypes in Network Wm-A, all other Asian-originated haplotypes created a star-like structure around Wm_Hap 6 and Wm_Hap 1 which suggests those two haplotypes may have been founders throughout Asia. The Wm_Hap 6 and Wm_Hap 1 were reported from Taiwan.

### 3.4 Population genetic structure *P. canaliculata* and *P. maculata*

The high value of haplotype diversity (Hd), nucleotide diversity ($\pi$), and an average number of nucleotide differences (K) suggested higher genetic diversity of *P. canaliculata* in Taiwan (Table

3). Among all 14 sites, Taitung, Taichung, Pingtung, Yilan, Kaohsiung, and Chiayi showed the highest genetic diversity; Tainan, Yunlin, Miaoli, and Taoyuan showed high to moderate genetic diversity; Changhua, Hsinchu, Hualien, and Taipei were noted to have less diverse populations (Table 3). Compared to others, the populations in the South of Taiwan (PT, KSH, CY, TN) were reported to have higher haplotype diversity as well as nucleotide diversity (Table 1, 3). The significantly positive value of Fu's Fs and Tajima's D ($D < 0$) were recorded for populations Taitung, Miaoli, Yunlin, Taoyuan, Yilan, Kaohsiung, and Pingtung. Conversely, significantly negative values of Fu's Fs and Tajima's D ($D > 0$) were observed for the population in Changhua, Taipei, and Hsinchu (Table 3). Furthermore, *P. canaliculata* in Taiwan was noted to significantly differ among some provinces, where a higher $F_{st}$ value ($F_{st} > 0.2$) resulted in less gene flow ($Nm < 1$) and vice versa (Table 4). However, the negative $F_{st}$ values likely indicate more variation within the focal populations than between populations. While *P. maculata* was found to be less dominant in distribution compared to *P. canaliculata*, polymorphism in populations were higher (Table 4).

The genetic diversity of *P. canaliculata* and *P. maculata* were considerably higher in their native counties (Argentina, Brazil, and Uruguay) compared to the places in which they were introduced. Among the introduced Asian countries, Taiwan demonstrated the highest genetic diversity of *P. canaliculata* and *P. maculata*, constituting the highest number of haplotypes (16 for *P. canaliculata*, and 4 for *P. maculata*) (Table 5 and 6).

### 3.5 Mismatch distribution

The mismatch distribution results of *P. canaliculata* in Taiwan showed two bimodal well-separated peaks with some small intermediate peaks (Fig. 6a). However, in the case of *P. maculata*

three distinct peaks were observed, including two major peaks and an intermediate peak (Fig. 6b). The mismatch distribution of *P. canaliculata* and *P. maculata* from the world dataset showed multimodal distribution (Fig. 6c and d).

## 4. DISCUSSION

Based on our mitochondrial COI barcoding data, we found *P. canaliculata* to have a wide distribution of across Taiwan, whereas *P. maculata* was less abundant and restricted (mostly) to the South of Taiwan (Pingtung, Kaohsiung, Tainan, and Chiayi; with Middle - Miaoli and North – Yilan, both represented by one single individual) (Fig. 1). Thus, the distribution of *P. canaliculata* and *P. maculata* were noted to be sympatric but heterogenous across the country (Fig. 1 and Table 3), a similar distribution pattern was noted in our previous study in southern Taiwan (Banerjee et al., 2022). Our haplotype analysis further demonstrated the possible origin of *P. canaliculata* from Argentina, although *P. maculata* may have been introduced independently from Argentina and Brazil (Hayes et al., 2008; Yang et al., 2022) instead of the previously assumed hypothesis of being imported directly from Brazil (Yang et al., 2018). Alternatively, considering the cryptic nature and presence of a single network of *P. maculata*, introduction of this species during the same event with *P. canalicualta* directly from Argentina cannot be precluded. Furthermore, our haplotype network analysis agreed with the possible (re-)introduction of *P. canaliculata* and *P. maculata* from Taiwan to other Asian countries (e.g., China, Malaysia, Philippines) (Joshi & Sebastian, 2003) which may have stemmed from the trade of snails to capture new Asian markets.

Interestingly, the invasion of *P. canaliculata* and *P. maculata* has also been noted to be sympatric and heterogenous in other nearby Asian countries, such as in China (Yang et al., 2018), Hong Kong (Yang et al., 2022), Malaysia (Rama Rao et al., 2018), and Thailand (Dumidae et al., 2021), where *P. canaliculata* is reported to be more widely distributed compared to *P. maculata*. This corroborates the dominant hypothesis of predominately *P. canaliculata* in Asia regions (Hayes et al., 2008). Interestingly, this distribution pattern is not reflected in their native range, where *P. maculata* is routinely found to be widely distributed throughout Brazil, Uruguay, and Argentina, whereas *P. canaliculata* remains restricted to North Argentina and South Uruguay (Hayes et al., 2008; Glasheen et al., 2020). The difference in the distribution in Asian regions may reflect different physiological tolerance, or species-specific biology (Hayes et al., 2008; Matsukura et al, 2016). Alternatively, it can be posited that these phylogeographical patterns and unequal distribution may be due to complex introduction histories wherein *P. canaliculata* may have been introduced as a single species for trade, with the (proportionately limited) unintentional introduction of *P. maculata* due to misidentification. Thus, the introduction of a relatively small number of *P. maculata* individuals masking with *P. canaliculata* may have resulted in a population bottleneck, exacerbating the limited southern distribution of *P. maculata*. Notably, current distribution data is based on mitochondrial DNA, and *P. canaliculata* and *P. maculata* are reported to hybridized both in their native and non-native range (Glasheen et al., 2020; Yang et al., 2020; Kannan et al., 2021). Thus, hybridization could also play an important role in their unequal phylogeographic distribution. Similar to *P. maculata*, another invasive species, *P. scalaris*, remains restricted to the southern part of Taiwan (Wu et al., 2010), underscoring that South Taiwan is likely the most heavily invaded region for multiple Golden Apple Snails species, potentially due to human impacted land use and the popularity of transportation ports.

Although the origin of *Pomacea* spp. in Asia has been widely studied by previous researchers from different countries (Hayes et al., 2008; Liu et al., 2019; Dumidae et al., 2021; Yang et al., 2022), additional data from Taiwan specifically (and for the first time) will contribute to our knowledge regarding the possible path of introduction and spread throughout Asia. Our haplotype analysis revealed the presence of all three independent Networks of *P. canaliculata* across Taiwan, whereas only two networks of *P. canaliculata* were detected from other countries of Asia (Yang et al., 2018; Yang et al., 2021). The occurrence of three largely different clades/networks across Taiwan suggesting species introduction may have occurred multiple times and possibly propagated from multiple locations (Fig. 3).

The combined mitochondrial datasets of *P. canaliculata* with 948 global sequences (originating from Africa, Asia, North America, Oceania, and South America) exhibited three different Networks, mirroring previous studies (Yang et al., 2018; Yang et al., 2021), with sequences from Taiwan represented across all networks (Fig. 4). The Wc_Hap 1, Wc_Hap 27, and Wc_Hap 9 represented the founder haplotypes of Network Wc-A, Network Wc-B, and Network Wc-C respectively. The common sharing of founder haplotypes between Taiwan and Argentina reaffirms the historical assumption of the introduction of *P. canaliculata* from Argentina to Taiwan (Joshi & Sebastian, 2003; Hayes et al., 2008). Although the absence of a shared haplotype from Argentina was noted in the case of WC_Hap 27 in Network Wc-B, which again may simply be due to a smaller number of represented sequences from Argentina in our global dataset. Furthermore, Network Wc-A and Network Wc-B were widely distributed, however the haplotypes from Network Wc-C were rare in all native and non-native countries, not just those haplotypes representative in Asia. This also does not lend support for sequence sampling bias (i.e. more

representative samples in Taiwan compared to other sampled countries), although we cannot preclude it as a possible contributing factor.

Interestingly, our haplotype analysis revealed the presence of a single Network for *P. maculata* across Taiwan, echoing patterns across other Asian countries (Yang et al., 2018; Yang et al., 2021). This is in contrast to *P. canaliculata* for which the, the haplotype distribution pattern suggest multiple introductions events (Fig. 3 and 5). Corroborating this further and despite the complex network of unique haplotypes, Taiwan shares the founder haplotype with Argentina (Wm_Hap 1). Notably, our results demonstrated the presence of Brazilian haplotypes with only a few mutational steps away from the founder haplotype (Wm_Hap 1), suggesting the introduction of *P. maculata* possibly originated via Argentina and Brazil, rather than solely from Brazil as previously hypothesized (Fig. 5). The presence of only a single network suggests less frequent introductions events, and since the network is shared with Argentina, it may be possible that previously developed populations of *P. maculata* were transported to Taiwan together with *P. canaliculata*. The presence of a single network has also been noted in all other Asian countries, where sequences within Taiwan represent the presumed founder haplotypes.

Unsurprisingly, *P. canaliculata* and *P. maculata* showed lower haplotype diversity compared to their native population counterparts (e.g., Argentina and Brazil) which can be the result of founder effects. However, compared with other Asian countries, Taiwan showed similar (e.g., China) or higher (e.g., Malaysia, Thailand, and Philippines) genetic diversity for *P. canaliculata* and *P. maculata* (Table 5 and 6). These observations insinuate that Taiwan may have faced multiple introductions during the past. By analyzing the population genetic structure and haplotype network, we speculate that (re)introduction of *P. canaliculata* and *P. maculata* from

Taiwan may have proceeded to other Asian countries along with direct introduction from Argentina and Brazil.

As per our mismatch distribution analysis, two distinct peaks with large pairwise differences created a well-separated bimodal mismatch graph for *P. canaliculata* (Fig. 6a), however mismatch distribution analysis produced an intermediate peak in between the bimodal graph for *P. maculata* (Fig. 6b). Those patterns suggest two episodes of population expansion which may coincide with multiple introductions for *P. canaliculata*, and possibly a single or continuous source for *P. maculata*. Furthermore, the multimodal mismatch distribution of *P. canaliculata* and *P. maculata* for the global dataset (Dataset 2 and 3) indicated their complex population expansion dynamics, and possibly introduction history, around several countries (Fig. 6c and d).

The genetic diversity (value of Hd, $\pi$, and K) of *P. canaliculata* and *P. maculata* in Taiwan were further noted to be significantly similar to or higher compared to other non-native countries, potentially proposing an early introduction history. Their genetic diversity also varied among the counties/cities of Taiwan which may be related to multiple sources of introductions at different times, across different land-use patterns (e.g., agriculture), and human transportation. The South and Middle portion of Taiwan are the areas where most international trade (via harbor) is situated, and these locations exhibited the highest genetic diversity for *P. canaliculata* and *P. maculata*. Here, most of the unique haplotypes were noted from Tainan (n = 3) and Chiayi (n =2). Interestingly the distribution of *P. maculata* was found to be more restricted to the South of Taiwan. Furthermore, the North of Taiwan (e.g., Taipei, Hsinchu) consists of less agricultural land which might be the reason for lower population diversity. On the Eastern side of Taiwan, the geographical locations of Hualien, Taitung, and Yilan (less agricultural land) may have

encountered different introduction events as reflected in their high number of unique haplotypes (Hualien: H= 2, Taitung: H = 1, and Yilan: H= 2). Alternatively, those locations also offer opportunities for isolation and possible environmental adaptation due to high mountain ranges from other cities and counties, although the short introduction history (40 years) may render this possibility unlikely. In general, the dispersal ability of *Pomacea* spp. is observed to be low and thus the widespread distribution across Taiwan within such a short invasion period was probably facilitated predominately from human transportation.

The neutrality test, such as the value of Fu's Fs and Tajima's D ($D < 0$), was not observed as significant for the overall population *P. canaliculata* and *P. maculata* in Taiwan, however, the higher value of Fu's Fs and Tajima's D ($D < 0$) for the population of *P. canaliculata* in Taitung, Miaoli, Yunlin, Taoyuan, Yilan, Kaohsiung, and Pingtung did demonstrate a high number of rare alleles. On the contrary, the lower value of Fu's Fs and Tajima's D ($D > 0$) for the population of *P. canaliculata* in Changhua, Taipei, and Hsinchu suggests a scarcity of rare alleles, however, these results may equally be due to sampling error. Furthermore, the significant difference in $F_{st}$ value ($F_{st} > 0.2$) and negative $F_{st}$ values (suggest more variation within the populations than in between) in Table 4 could be possible due to multiple introductions and population mixing.

## Conclusions

Our data suggested the Argentinian origin of *P. canaliculata* in Taiwan, whereas *P. maculata* might have both Argentinian and Brazilian origin. Comparing their introduction histories, current distribution, and cryptic nature of *P. canaliculata* and *P. maculata*, our data suggests that *P. maculata* may have been introduced along with *P. canaliculata* (either intentionally or

unintentionally), however, *P. canaliculata* likely has several introduction histories. The establishment of *Pomacea* spp. across all of Taiwan is alarming, and our current data suggests the urgent need for a nationwide mitigation program and proper management policy. To prevent further introductions and spread of *Pomacea* spp., eDNA-based methods have been previously demonstrated to be useful (Banerjee et al., 2022). While *Pomacea* spp. are spreading rapidly within Taiwan and across Asia (the invaded zone), the chances of further spread in non-invaded countries should be of high concern. Although physical records and morphological identification have been reported in many countries (e.g., Pakistan, and Bangladesh), proper molecular identification is needed for species-specific mitigation plans.

## ACKNOWLEDGEMENTS

P.B. has been supported by Overseas Research Scholarships (ORS) from National Chung Cheng University as well as Ministry of Education (MOE)- Industry-Academia project (Taiwan). The authors would also like to thank the Ministry of Science and Technology (Taiwan) for financial support (MOST 109-2811-M-194-502; MOST 108-2811-M-194-510).

## AUTHORS' CONTRIBUTIONS

P.B. and C.Y.C., conceived of the idea; P.B., performed experiments, P.B., K.A.S., and C.Y.C., prepared the first draft and revised the manuscript, figures and tables. G.D., R.K.S, J.P.M., and M.W.Y.C, K. H. L., gave extensive edits and revised the manuscript.

## CONFLICTS OF INTERESTS

The authors declare no conflict of interest.

## DATA AVAILABILITY STATEMENT

All data obtained in this study have been deposited to GeneBank under the accession number OQ977961 - OQ978203 and OQ977020-OQ977027.

## Figures

**Figure 1**. Geographical distribution and frequency haplotypes of *P. canaliculata* (Pc_Hap 1-18) and *P. maculata* (Pm_Hap 19-22) in Taiwan (Map created using ArcGIS 10.7 software)

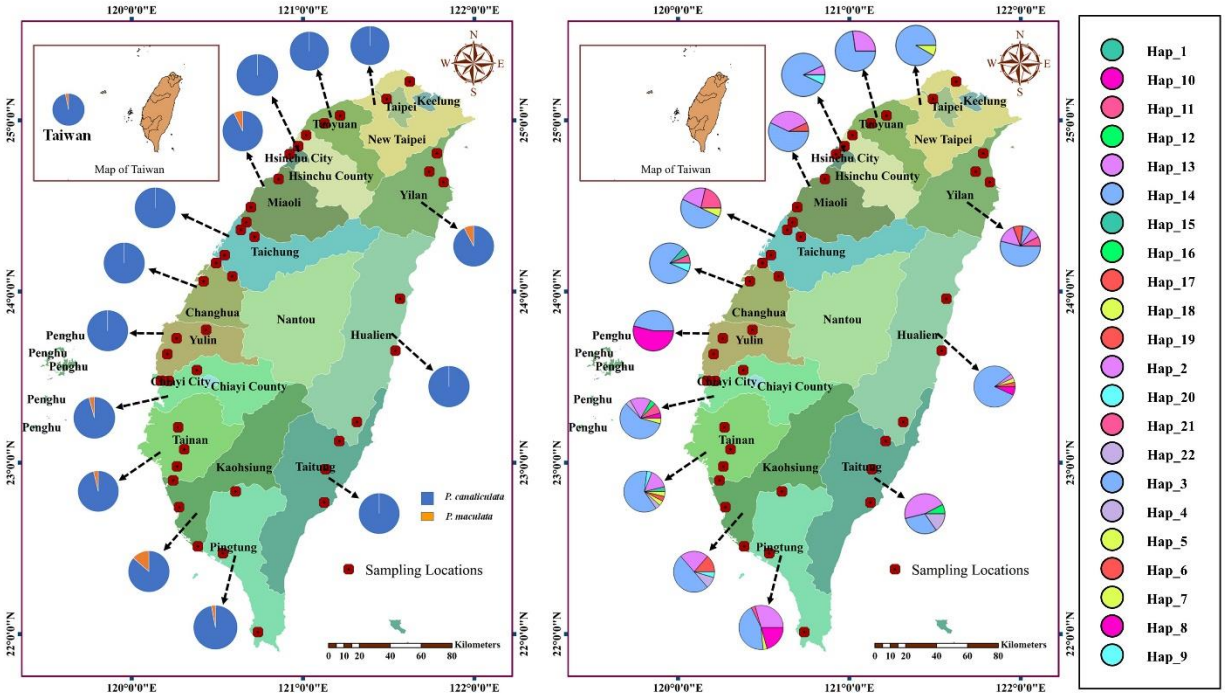


**Figure 2**. Maximum-likelihood phylogenetic tree generated from 254 sequences mitochondrial COI sequence of *P. canaliculata* and *P. maculata* collected from 41 different location (14 counties/cities) in Taiwan. The sequences of *P. lineata*, *P. paludosa*, *P. diffusa*, and *P. scalaris* were used as outgroups.

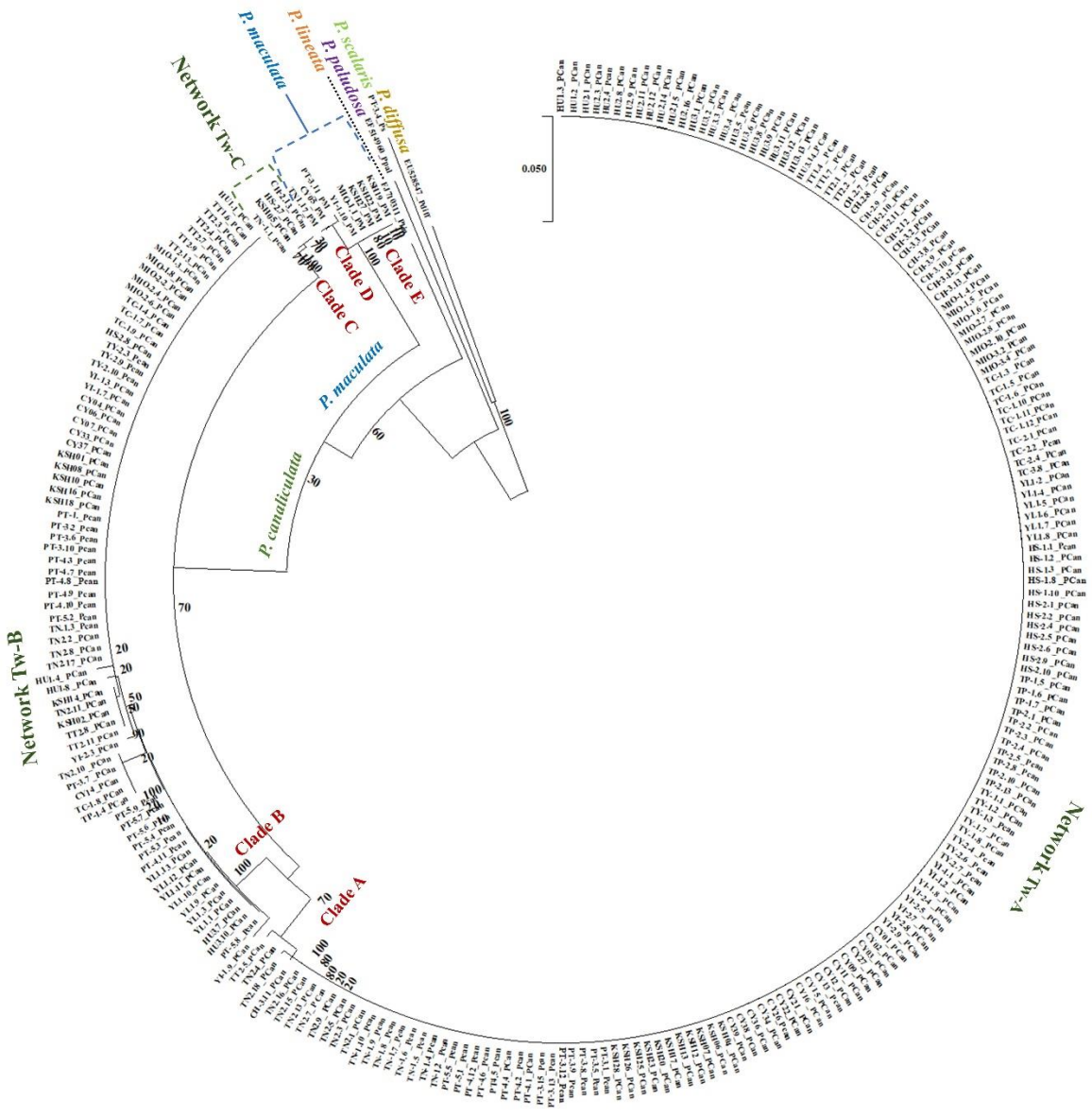


**Figure 3**. Mitochondrial COI haplotypes networks revealing frequency and relationship of *P. canaliculata* (Pc_Hap 1-18) and *P. maculata* (Pm _Hap 19-22) across Taiwan using Dataset 1.

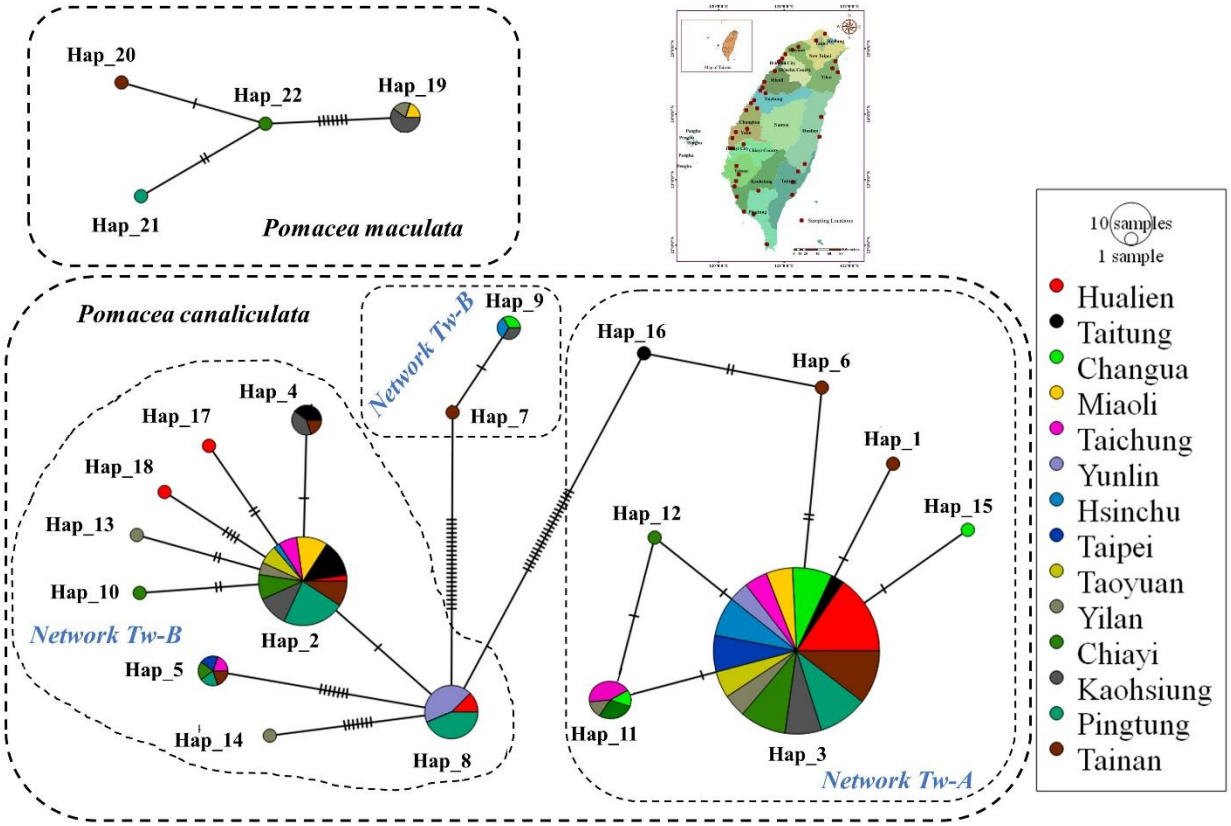


**Figure 4**. Mitochondrial COI haplotypes (Wc_Hap) revealing frequency and relationship of *P. canaliculata* from the global dataset (Dataset 3).

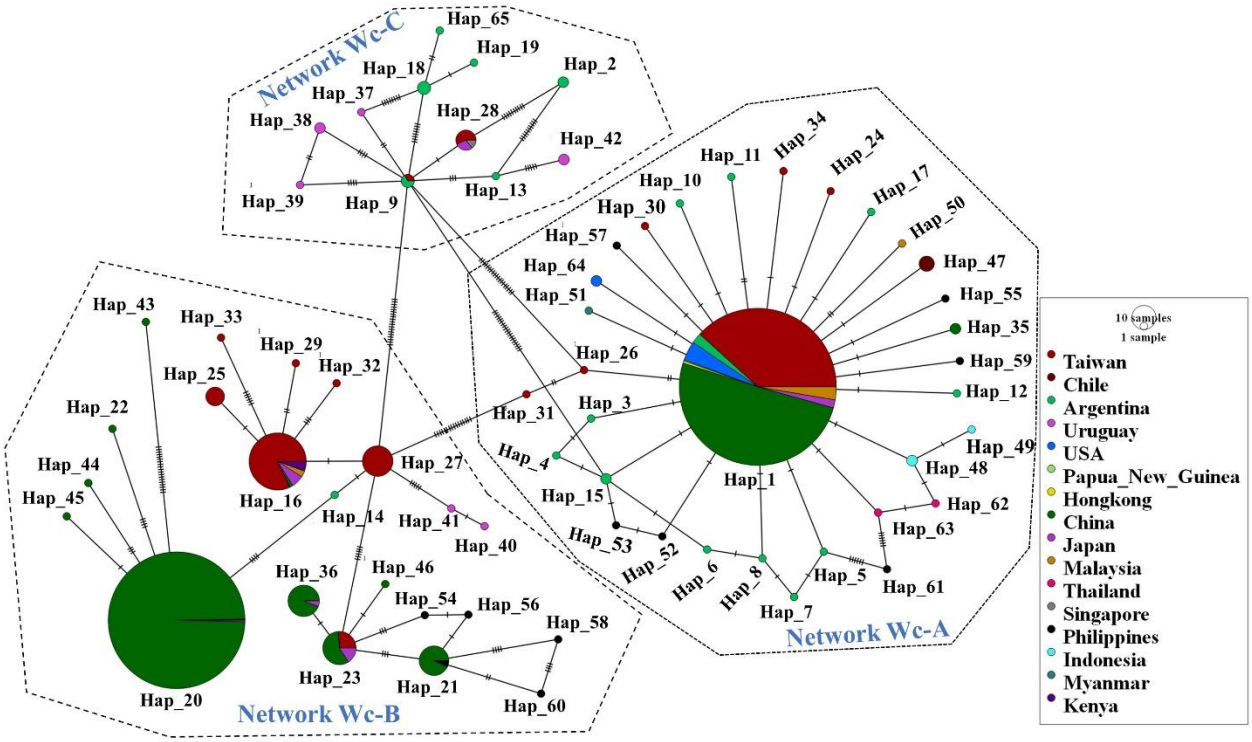


**Figure 5**. Mitochondrial COI haplotypes (Wm_Hap) revealing frequency and relationship of *P. maculata* from the global dataset (Dataset 2).

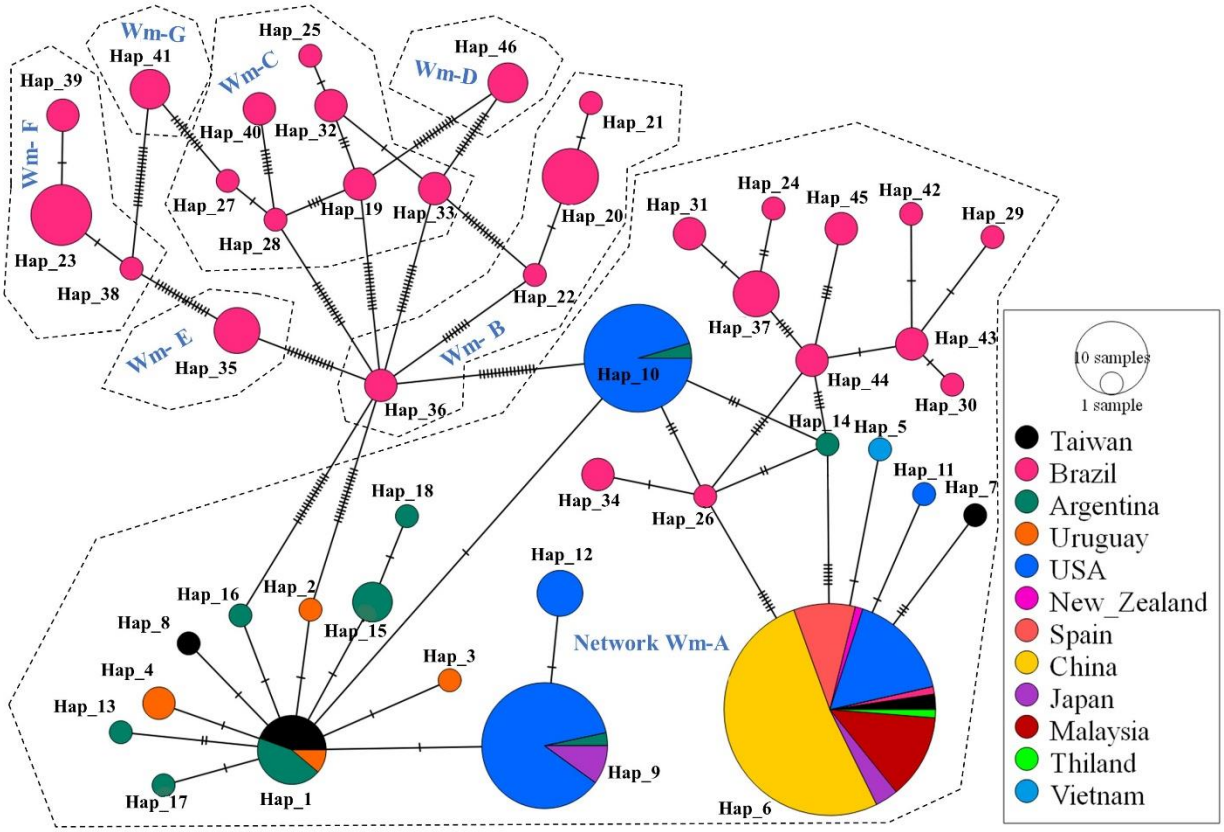
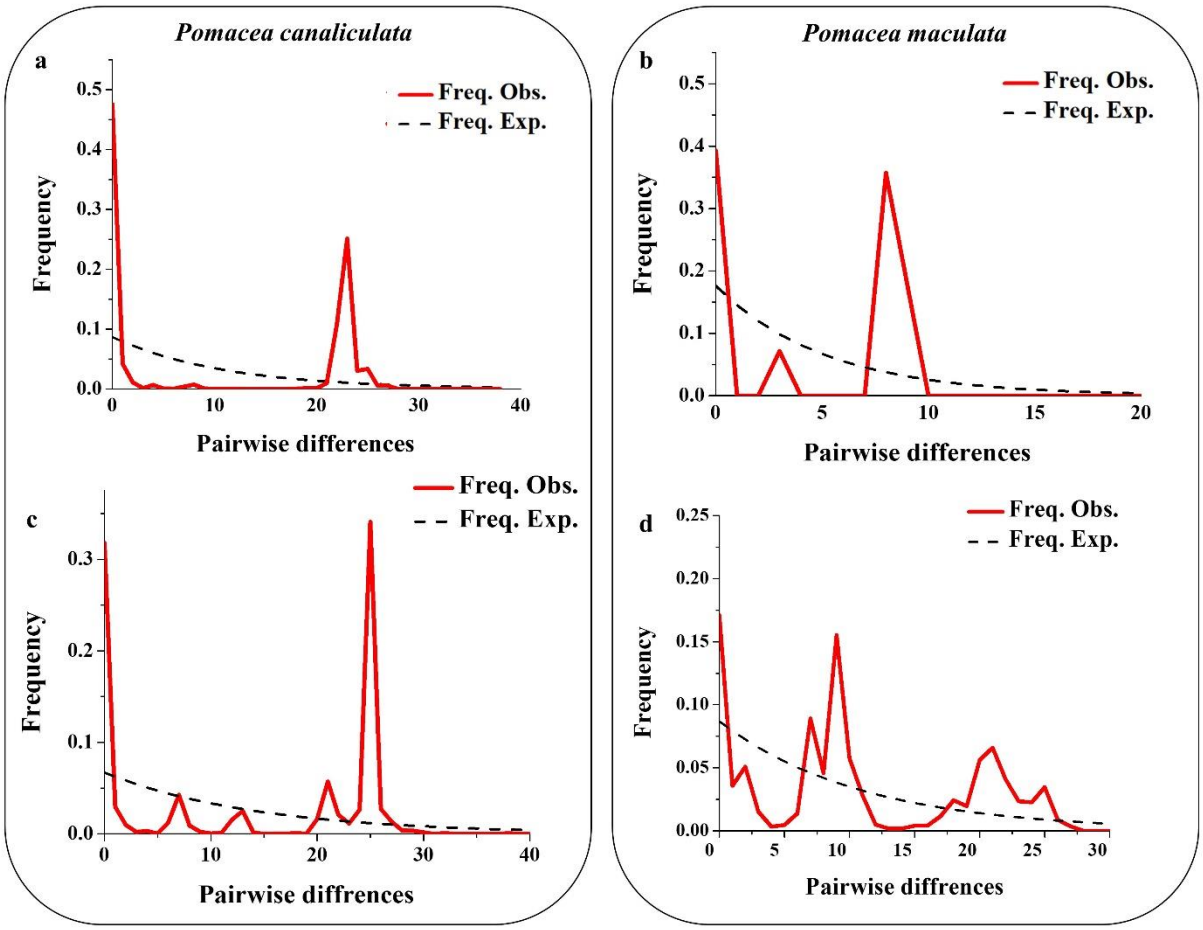


**Figure 6**. Mismatch distribution analysis of *P. canaliculata* from (a) Taiwan (Dataset 1) and (c) the global dataset (Dataset 3), and *P. maculata* (b) Taiwan (Dataset 1) and (d) the global dataset (Dataset 2)



## Tables

**Table 1.** Population genetic diversity of *P. canaliculata* and *P. maculata* across Taiwan. The significant value of Tajima's D and Fu's FS are marked in bold.

| Sampling sites (n) | Haplotypes | | | Number of segregating sites (S) | Haplotype diversity (Hd) | Nucleotide diversity (π) | Average number of differences (K) | Fu's FS | P value | Tajima's D | P value |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Total | Shared | Unique | | | | | | | | |
| ***P. canaliculata* (246)** | | | | **52** | **0.53922** | **0.01807** | **9.89940** | | | | |
| HU (29) | 5 | 3 | 2 | 31 | 0.31773 | 0.0131 | 7.99261 | **8.94284** | 0.996 | 0.04566 | 0.558 |
| TT (13) | 4 | 3 | 1 | 27 | 0.71795 | 0.02198 | 13.41026 | **9.07856** | 0.999 | **2.36073** | 1 |
| CH (15) | 4 | 3 | 1 | 26 | 0.37143 | 0.00568 | 3.46667 | 3.00518 | 0.916 | **-2.3638** | 0 |
| MIO (13) | 2 | 2 | 0 | 26 | 0.51282 | 0.02186 | 13.33333 | **15.67426** | 1 | **2.57365** | 1 |
| TC (14) | 4 | 4 | 0 | 32 | 0.7033 | 0.02012 | 12.27473 | **9.13108** | 0.997 | 0.9456 | 0.873 |
| YL (13) | 2 | 2 | 0 | 25 | 0.53846 | 0.02207 | 13.46154 | **15.75404** | 1 | **2.91314** | 1 |
| HS (14) | 3 | 3 | 0 | 36 | 0.27473 | 0.01122 | 6.84615 | **7.97682** | 0.997 | **-1.71117** | 0.024 |
| TP (12) | 2 | 2 | 0 | 28 | 0.16667 | 0.00765 | 4.66667 | **8.00904** | 0.995 | **-2.22668** | 0.001 |
| TY (11) | 2 | 2 | 0 | 26 | 0.43636 | 0.0186 | 11.34545 | **12.98712** | 1 | 1.27969 | 0.93 |
| YI (12) | 5 | 3 | 2 | 33 | 0.66667 | 0.02228 | 13.59091 | **6.17073** | 0.994 | 1.10135 | 0.911 |
| CY (23) | 6 | 4 | 2 | 33 | 0.61265 | 0.01832 | 11.17787 | **8.13701** | 0.989 | 0.95742 | 0.879 |
| KSH (19) | 4 | 4 | 0 | 37 | 0.61404 | 0.02359 | 14.39181 | **13.23563** | 1 | 1.44284 | 0.955 |
| PT (33) | 4 | 4 | 0 | 31 | 0.67614 | 0.02218 | 13.5303 | **18.39534** | 1 | **2.74772** | 1 |
| TN (25) | 7 | 4 | 3 | 42 | 0.5800 | 0.01915 | 11.68 | **7.37466** | 0.995 | 0.19116 | 0.608 |
| ***P. maculate* (8)** | | | | | | | | | | | |
| PT, KSH, TN, CY, MIO, YI | 4 | | | 10 | 0.6429 | 0.00738 | 4.5000 | | | 0.82566 | |

**Table 2.** Genetic differentiation coefficient ($F_{st}$) below diagonal and gene flow ($N_m$) above diagonal based on mitochondrial COI sequences of *P. canaliculata* from 14 counties or cities of Taiwan. Abbreviations: HU: Hualien, and TT: Taitung from the East, YL: Yunlin, CH: Changhua, TC: Taichung, and MIO: Miaoli from the Middle, HS: Hsinchu, TY: Taoyuan, TP: Taipei, and YI: Yilan from the North, and PT: Pingtung, KSH: Kaohsiung, TN: Tainan, and CY: Chiayi from the

South. The higher $F_{st}$ value ($F_{st}$ >0.2) resulted in less gene flow (Nm <1) between the counties or cities (marked in bold).

| | **TN** | **PT** | **KSH** | **CY** | **YI** | **TY** | **TP** | **HS** | **YL** | **TC** | **MIO** | **CH** | **TT** | **HU** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **TN** | | 4.040 | NA | NA | NA | NA | 13.27 | 73.904 | 4.788 | NA | NA | 4.904 | 2.13 | NA |
| **PT** | 0.110 | | 261.28 | 3.906 | 13.778 | 6.224 | 1.148 | **1.368** | NA | 5.85 | NA | **0.858** | NA | **1.752** |
| **KSH** | -0.007 | 0.002 | | NA | NA | NA | 2.584 | 3.83 | NA | NA | NA | **1.692** | 12.49 | 5.622 |
| **CY** | -0.038 | 0.113 | 0.000 | | NA | NA | 13.724 | 35.758 | 4.478 | NA | NA | 4.464 | 2.026 | NA |
| **YI** | -0.046 | 0.035 | -0.051 | -0.050 | | NA | 4.816 | 8.668 | 36.4 | NA | NA | 2.332 | 4.618 | 42.198 |
| **TY** | -0.065 | 0.074 | -0.039 | -0.067 | -0.083 | | 9.828 | 60.032 | 7.904 | NA | NA | 3.286 | 2.754 | NA |
| **TP** | 0.036 | **0.304** | 0.162 | 0.035 | 0.094 | 0.048 | | NA | **0.998** | 9.844 | 2.51 | NA | **0.63** | NA |
| **HS** | 0.007 | **0.268** | 0.115 | 0.014 | 0.055 | 0.008 | -0.053 | | **1.272** | 21.712 | 3.744 | NA | **0.782** | NA |
| **YL** | 0.095 | -0.049 | -0.015 | 0.100 | 0.014 | 0.060 | **0.334** | **0.282** | | 7.608 | NA | **0.694** | NA | **1.71** |
| **TC** | -0.050 | 0.079 | -0.026 | -0.058 | -0.070 | -0.080 | 0.048 | 0.023 | 0.062 | | NA | 3.434 | 2.714 | NA |
| **MIO** | -0.022 | -0.006 | -0.064 | -0.021 | -0.072 | -0.061 | 0.166 | 0.118 | -0.022 | -0.046 | | **1.488** | 12.258 | 7.12 |
| **CH** | 0.093 | **0.368** | **0.228** | 0.101 | 0.177 | 0.132 | -0.038 | -0.041 | **0.419** | 0.127 | **0.251** | | **0.464** | 13.7 |
| **TT** | 0.190 | -0.030 | 0.038 | 0.198 | 0.098 | 0.154 | **0.443** | **0.390** | -0.032 | 0.156 | 0.039 | **0.519** | | **0.974** |
| **HU** | -0.011 | **0.222** | 0.082 | -0.014 | 0.012 | -0.031 | -0.017 | -0.029 | **0.226** | -0.011 | 0.066 | 0.035 | **0.339** | |

**Table 3.** Haplotype distribution (Pc_Hap) of *Pomacea canaliculata* in 14 counties/cities across Taiwan.

| **Sample sites (Hn)** | **Sample ID** | **Shared Haplotypes** | **Unique Haplotypes** |
|---|---|---|---|
| **East (7)** | HU and TT | **Total 4**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**, **Hap 4**, **Hap 8** | **Total 3**: Network Tw-A: **Hap 16**; Network Tw-B: **Hap 17**, **Hap 18** |
| Hualien (5) | HU | **Total 3**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**, **Hap 8** | **Total 2**: Network Tw-B; **Hap 17**, **Hap 18** |
| Taitung (4) | TT | **Total 3**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**, **Hap 4** | Network Tw-A: **Hap 16** |
| **Middle (7)** | CH, MIO, TC, YL | **Total 6**: Network Tw-A: **Hap 3**, **Hap 11**; Network Tw-B: **Hap 2**, **Hap 5**, **Hap 8**; Network Tw-C: **Hap 9** | Network Tw-A: **Hap 15** |
| Changhua (4) | CH | **Total 3**: Network Tw-A: **Hap 3**, **Hap 11**; Network Tw-C: **Hap 9** | Network Tw-A: **Hap 15** |
| Miaoli (2) | MIO | **Total 2**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2** | - |
| Taichung (4) | TC | **Total 4**: Network Tw-A: **Hap 3**, **Hap 11**; Network Tw-B: **Hap 2**, **Hap 5**, | - |
| Yunlin (2) | YL | **Total 2**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 8** | - |
| **North (7)** | HS, TP, TY, YI | **Total 3**: Network Tw-A: **Hap 3**; **Hap 11**; Network Tw-B: **Hap 2**, **Hap 5**; Network Tw-C: **Hap 9** | **Total 2**: Network Tw-B: **Hap 14**; Network Tw-A: **Hap 15** |
| Hsinchu (3) | HS | **Total 3**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**; Network Tw-C: **Hap 9** | - |
| Taipei (2) | TP | **Total 2**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 5** | - |
| Taoyuan (2) | TY | **Total 2**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2** | |
| Yilan (5) | YI | **Total 3**: Network Tw-A: **Hap 3**, **Hap 11**; Network Tw-B: **Hap 2** | **Total 2**: Network Tw-B: **Hap 14**; Network Tw-A: **Hap 15** |
| **South (9)** | CY, KSH, PT, TN | **Total 4**: Network Tw-A: **Hap 3**, **Hap 11**; Network Tw-B: Hap 2, Hap 4, Hap 5, Hap 8; Network Tw-C: Hap 9 | **Total 5**: Network Tw-A: **Hap 1**, **Hap 6**, **Hap 12**; Network Tw-B: **Hap 10**; Network Tw-C: **Hap 7** |
| Chiayi (6) | CY | **Total 4**: Network Tw-A: **Hap 3**, **Hap 11**; Network Tw-B: **Hap 2**, **Hap 5** | **Total 2**: Network Tw-A: **Hap 12**; Network Tw-B: **Hap 10** |
| Kaohsiung (4) | KSH | **Total 4**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**, **Hap 4**; Network Tw-C: **Hap 9** | - |

| | | | |
|---|---|---|---|
| Pingtung (4) | PT | **Total 4**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**, **Hap 5**, **Hap 8** | - |
| Tainan (7) | TN | **Total 4**: Network Tw-A: **Hap 3**; Network Tw-B: **Hap 2**, **Hap 4**, **Hap 5** | Total 3: Network Tw-A: **Hap 1**, **Hap 6**; Network Tw-C: **Hap 7** |

**Table 4.** Haplotype distribution (Pm_Hap) of *Pomacea maculata* in 14 counties/cities across Taiwan.

| **Sample sites (Hn)** | **Sample ID** | **Shared Haplotypes** | **Unique Haplotypes** |
|---|---|---|---|
| **Middle (1)** | MIO | **Hap 19** | - |
| Miaoli (1) | MIO | **Hap 19** | - |
| **North (1)** | YI | **Hap 19** | - |
| Yilan (1) | YI | **Hap 19** | - |
| **South (4)** | CY, KSH, PT, TN | **Hap 19** | Total 3: **Hap 20**, **Hap 21**, **Hap 22** |
| Chiayi (1) | CY | - | **Hap 22** |
| Kaohsiung (3) | KSH | **Hap 19** | |
| Pingtung (1) | PT | - | **Hap 21** |
| Tainan (1) | TN | - | **Hap 20** |

**Table 5.** Population genetic diversity of *Pomacea canaliculata* from the global mitochondrial COI dataset (Dataset 3)

| **Country*** | **n** | **Hn** | **S** | **Hd** | **π** | **K** |
|---|---|---|---|---|---|---|
| Argentina | 32 | 19 | 49 | 0.91532 | 0.02282 | 12.59879 |
| Uruguay | 8 | 6 | 35 | 0.92857 | 0.02737 | 15.10714 |
| USA | 19 | 2 | 1 | 0.19883 | 0.00036 | 0.19883 |
| Chile | 4 | 1 | 0 | 0 | 0 | 0 |
| China | 582 | 12 | 57 | 0.56362 | 0.02231 | 12.31556 |
| Taiwan | 249 | 16 | 51 | 0.53922 | 0.01788 | 9.86789 |
| Indonesia | 3 | 2 | 1 | 0.66667 | 0.00121 | 0.66667 |
| Japan | 18 | 6 | 41 | 0.79739 | 0.02767 | 15.27451 |
| Malaysia | 14 | 3 | 22 | 0.38462 | 0.01047 | 5.78022 |
| Philippines | 12 | 12 | 39 | 1 | 0.02907 | 16.04545 |
| Kenya | 3 | 1 | 0 | 0 | 0 | 0 |
| Thailand | 2 | 2 | 1 | 1 | 0.00181 | 1 |
| China-Hongkong** | 2+ 139 | 5 | - | 0.441 | 0.01771 | 8.91 |

**Note:** n = Number of sequences; Hn = Number of Haplotype, S = Number of segregating sites; Hd = Haplotype diversity; π = Nucleotide diversity; K = Average number of differences

*Countries represented with 1 sequence were not shown here.

* 139 samples added from Yang et al., 2022, as their sequence are not available publicly yet.

**Table 6.** Population genetic diversity of *Pomacea maculata* from the global mitochondrial COI dataset (Dataset 2)

| Country* | n | Hn | S | Hd | π | K |
|---|---|---|---|---|---|---|
| Argentina | 16 | 11 | 36 | 0.925 | 0.01101 | 6.375 |
| Uruguay | 5 | 4 | 3 | 0.90000 | 0.00242 | 1.4 |
| Brazil | 61 | 29 | 69 | 0.96448 | 0.02907 | 16.83279 |
| USA | 66 | 5 | 11 | 0.70536 | 0.0063 | 3.64942 |
| Spain | 8 | 1 | 0 | 0 | 0 | 0 |
| Taiwan | 8 | 4 | 11 | 0.75 | 0.00851 | 4.92857 |
| Malaysia | 11 | 1 | 0 | 0 | 0 | 0 |
| Japan | 6 | 2 | 9 | 0.6 | 0.00933 | 5.4 |
| China | 44 | 1 | 0 | 0 | 0 | 0 |
| China- Hong Kong* | 10 | 2 | - | 0.356 | 0.00353 | 1.778 |

**Note:** n = Number of sequences; Hn = Number of Haplotype, S = Number of segregating sites; Hd = Haplotype diversity; π = Nucleotide diversity; K = Average number of differences
*Countries represented with 1 sequence were not shown here.
* 10 samples added from Yang et al., 2022, as their sequence are not available publicly yet.

## Reference


ArcGIS 10.7 software; ESRI Inc., Redlands, CA, USA. Retrieved from https://www.esri.com/en-us/arcgis/products/arcgis-desktop/overview

Banerjee, P., Stewart, K. A., Dey, G., Sharma, R. K., Maity, J. P., Chan, M. W., . . . Chen, C.-Y. (2022). When conventional methods fall short: identification of invasive cryptic Golden Apple Snails (Pomacea canaliculata; P. maculata) using environmental DNA. *Hydrobiologia, 849*(19), 4241-4257.

Bouckaert, R., Heled, J., Kühnert, D., Vaughan, T., Wu, C.-H., Xie, D., . . . Drummond, A. J. (2014). BEAST 2: a software platform for Bayesian evolutionary analysis. *PLoS computational biology, 10*(4), e1003537.

Buddie, A. G., Rwomushana, I., Offord, L. C., Kibet, S., Makale, F., Djeddour, D., . . . Chacha, D. (2021). First report of the invasive snail Pomacea canaliculata in Kenya. *CABI Agriculture and Bioscience, 2*(1), 1-10.

Cheng, E. Y., & Kao, C. (2006). Control of golden apple snail, Pomacea canaliculata (Lamarck), in Taiwan. *Global advances in ecology and management of golden apple snails*, 155-167.

Chiu, Y.-W., Wu, J.-P., Hsieh, T.-C., Liang, S.-H., Chen, C.-M., & Huang, D.-J. (2014). Alterations of biochemical indicators in hepatopancreas of the golden apple snail, Pomacea canaliculata, from paddy fields in Taiwan. *Journal of environmental biology, 35*(4), 667.

Cowie, R., Hayes, K., & Thiengo, S. (2006). What Are Apple Snails? Confused Taxonomy and Some Preliminary Resolution. Dalam Global Advances in Ecology and Management of Golden Apple Snails. RC Joshi and LS Sebastian (Eds), 3-23. *PhilRice. Philippines*.

Dumidae, A., Janthu, P., Subkrasae, C., Polseela, R., Mangkit, B., Thanwisai, A., & Vitta, A. (2021). Population genetics analysis of a Pomacea snail (Gastropoda: Ampullariidae) in Thailand and its low infection by Angiostrongylus cantonensis. *Zoological Studies, 60*.

Edgar, R. C. (2004). MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic acids research, 32*(5), 1792-1797.

Excoffier, L., & Lischer, H. E. (2010). Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. *Molecular ecology resources, 10*(3), 564-567.

Felsenstein, J. (1985). Confidence limits on phylogenies: an approach using the bootstrap. *evolution, 39*(4), 783-791.

Fu, Y.-X. (1997). Statistical tests of neutrality of mutations against population growth, hitchhiking and background selection. *Genetics, 147*(2), 915-925.

Glasheen, P. M., Burks, R. L., Campos, S. R., & Hayes, K. A. (2020). First evidence of introgressive hybridization of apple snails (Pomacea spp.) in their native range. *Journal of Molluscan Studies, 86*(2), 96-103.

Hall, T. A. (1999). *BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT.* Paper presented at the Nucleic acids symposium series.

Hayes, K. A., Joshi, R., Thiengo, S. C., & Cowie, R. (2008). Out of South America: multiple origins of non-native apple snails in Asia. *Diversity and distributions, 14*(4), 701-712.

He, J., Gao, Z., Su, Y., Lin, S., & Jiang, H. (2018). Geographical and temporal origins of terrestrial vertebrates endemic to Taiwan. *Journal of Biogeography, 45*(11), 2458-2470.

Jarić, I., Heger, T., Monzon, F. C., Jeschke, J. M., Kowarik, I., McConkey, K. R., . . . Essl, F. (2019). Crypticity in biological invasions. *Trends in ecology & evolution, 34*(4), 291-302.

Jiang, X., Zheng, P., Soto, I., Haubrock, P. J., Chen, J., & Ji, L. (2022). Global economic costs and knowledge gaps of invasive gastropods. *Ecological Indicators, 145*, 109614.

Joshi, R. C., & Sebastian, L. S. (2003). Global advances in ecology and management of golden apple snails.

Kannan, A., Ratnayeke, S., & Yow, Y.-Y. (2021). Molecular evidence of hybridisation in two invasive species of Pomacea (Gastropoda: Ampullariidae) in Peninsular Malaysia. *RAFFLES BULLETIN OF ZOOLOGY, 69*, 570-585.

Kearse, M., Moir, R., Wilson, A., Stones-Havas, S., Cheung, M., Sturrock, S., . . . Duran, C. (2012). Geneious Basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics, 28*(12), 1647-1649.

Lee, K.-H., Chen, T.-H., Shang, G., Clulow, S., Yang, Y.-J., & Lin, S.-M. (2019). A check list and population trends of invasive amphibians and reptiles in Taiwan. *ZooKeys, 829*, 85.

Leigh, J. W., & Bryant, D. (2015). POPART: full-feature software for haplotype network construction. *Methods in ecology and evolution, 6*(9), 1110-1116.

Librado, P., & Rozas, J. (2009). DnaSP v5: a software for comprehensive analysis of DNA polymorphism data. *Bioinformatics, 25*(11), 1451-1452.

Linders, T. E. W., Schaffner, U., Eschen, R., Abebe, A., Choge, S. K., Nigatu, L., . . . Allan, E. (2019). Direct and indirect effects of invasive species: Biodiversity loss is a major mechanism by which an invasive tree affects ecosystem functioning. *Journal of Ecology, 107*(6), 2660-2672.

Liu, X., Zhou, Y., Ouyang, S., & Wu, X. (2019). Phylogeographic patterns and demographic history of Pomacea canaliculata and Pomacea maculata from different countries (Ampullariidae, Gastropoda, Mollusca). *Nature Conservation, 36*, 71-92.

Lv, S., Zhang, Y., Liu, H. X., Hu, L., Liu, Q., Wei, F. R., . . . Zhou, X. N. (2013). Phylogenetic evidence for multiple and secondary introductions of invasive snails: Pomacea species in the People's Republic of China. *Diversity and distributions, 19*(2), 147-156.

Matsukura, K., Izumi, Y., Yoshida, K., & Wada, T. (2016). Cold tolerance of invasive freshwater snails, Pomacea canaliculata, P. maculata, and their hybrids helps explain their different distributions. *Freshwater Biology, 61*(1), 80-87.

Matsukura, K., Okuda, M., Kubota, K., & Wada, T. (2008). Genetic divergence of the genus Pomacea (Gastropoda: Ampullariidae) distributed in Japan, and a simple molecular method to distinguish P. canaliculata and P. insularum. *Applied Entomology and Zoology, 43*(4), 535-540.

Matsukura, K., & Wada, T. (2017). Identification of Pomacea species using molecular techniques. *Biology and management of invasive apple snails*, 33-43.

McNeely, J. (2001). Invasive species: a costly catastrophe for native biodiversity. *Land Use and Water Resources Research, 1*(1732-2016-140260).

Päckert, M., Martens, J., & Severinghaus, L. L. (2009). The Taiwan Firecrest (Regulus goodfellowi) belongs to the Goldcrest assemblage (Regulus regulus sl): evidence from mitochondrial DNA and the territorial song of the Regulidae. *Journal of Ornithology, 150*, 205-220.

Pyšek, P., Jarošík, V., Hulme, P. E., Kühn, I., Wild, J., Arianoutsou, M., . . . Essl, F. (2010). Disentangling the role of environmental and human pressures on biological invasions across Europe. *Proceedings of the National Academy of Sciences, 107*(27), 12157-12162.

Rama Rao, S., Liew, T.-S., Yow, Y.-Y., & Ratnayeke, S. (2018). Cryptic diversity: Two morphologically similar species of invasive apple snail in Peninsular Malaysia. *PLoS One, 13*(5), e0196582.

Rambaut, A., & Drummond, A. (2010). FigTree v1. 4.4. Institute of Evolutionary Biology, University of Edinburgh. In.

Ranamukhaarachchi, S., & Wickramasinghe, S. (2006). Golden apple snails in the world: introduction, impact, and control measures. *Global advances in ecology and management of golden apple snails. Philippine Rice Research Institute, Nueva Ecija*, 133-152.

Richardson, D. M., & Rejmánek, M. (2011). Trees and shrubs as invasive alien species–a global review. *Diversity and distributions, 17*(5), 788-809.

Tamura, K., Stecher, G., & Kumar, S. (2021). MEGA11: molecular evolutionary genetics analysis version 11. *Molecular biology and evolution, 38*(7), 3022-3027.

Tsai, H.-C., Liu, Y.-C., Kunin, C. M., Lee, S. S.-J., Chen, Y.-S., Lin, H.-H., . . . Yen, M.-Y. (2001). Eosinophilic meningitis caused byAngiostrongylus cantonensis: report of 17 cases. *The American journal of medicine, 111*(2), 109-114.

Turbelin, A. J., Cuthbert, R. N., Essl, F., Haubrock, P. J., Ricciardi, A., & Courchamp, F. (2023). Biological invasions are as costly as natural hazards. *Perspectives in Ecology and Conservation*.

Wu, J.-Y., Meng, P.-J., Liu, M.-Y., Chiu, Y.-W., & Liu, L.-L. (2010). A high incidence of imposex in Pomacea apple snails in Taiwan: a decade after triphenyltin was banned. *Zool Stud, 49*, 85-93.

Yang, P., Chen, Y., Lee, W., & Chen, Y. (2006). Golden apple snail management and prevention in Taiwan. *Global advances in ecology and management of golden apple snails*, 169-179.

Yang, Q.-Q., Liu, S.-W., He, C., & Yu, X.-P. (2018). Distribution and the origin of invasive apple snails, Pomacea canaliculata and P. maculata (Gastropoda: Ampullariidae) in China. *Scientific Reports, 8*(1), 1-8.

Yang, Q. Q., Ip, J. C. H., Zhao, X. X., Li, J. N., Jin, Y. J., Yu, X. P., & Qiu, J. W. (2022). Molecular analyses revealed three morphologically similar species of non-native apple snails and their patterns of distribution in freshwater wetlands of Hong Kong. *Diversity and distributions, 28*(1), 97-111.

Cheng, E.Y. and Kao, C., 2006. Control of golden apple snail, Pomacea canaliculata (Lamarck), in Taiwan. Global advances in ecology and management of golden apple snails, pp.155-167.

Darriba, D., Taboada, G.L., Doallo, R. and Posada, D., 2012. jModelTest 2: more models, new heuristics and parallel computing. Nat Meth 9: 772. Go to original source.

Hayes, K.A., Joshi, R.C., Thiengo, S.C. and Cowie, R.H., 2008. Out of South America: multiple origins of non-native apple snails in Asia. Diversity and distributions, 14(4), pp.701-712.

Wu, J.Y., Wu, Y.T., Li, M.C., Chiu, Y.W., Liu, M.Y. and Liu, L.L., 2011. Reproduction and Juvenile Growth of the Invasive Apple Snails Pomacea canaliculata and P. scalaris(Gastropoda: Ampullariidae) in Taiwan. Zoological Studies, 50(1), pp.61-68.

Zhao, B., Luo, M., Zhang, J., Liu, Y., Deng, Z., & Gong, X. (2022). Genetic diversity of two globally invasive snails in Asia and Americas in relation with agricultural habitats and climate factors. Diversity, 14(12), 1069.

Dumidae, A., Janthu, P., Subkrasae, C., Polseela, R., Mangkit, B., Thanwisai, A. and Vitta, A., 2021. Population genetics analysis of a Pomacea snail (Gastropoda: Ampullariidae) in Thailand and its low infection by Angiostrongylus cantonensis. Zoological Studies, 60.

Liu, X., Zhou, Y., Ouyang, S. and Wu, X., 2023. Environmental Correlates to Genetic Diversity and Structure in Invasive Apple Snail (Pomacea canaliculata) Populations in China. Diversity, 15(10), p.1048.

Saltonstall, K., 2002. Cryptic invasion by a non-native genotype of the common reed, Phragmites australis, into North America. Proceedings of the National Academy of Sciences, 99(4), pp.2445-2449.

Sakai, A.K., Allendorf, F.W., Holt, J.S., Lodge, D.M., Molofsky, J., With, K.A., Baughman, S., Cabin, R.J., Cohen, J.E., Ellstrand, N.C. and McCauley, D.E., 2001. The population biology of invasive species. Annual review of ecology and systematics, 32(1), pp.305-332.

Lowe, S., Browne, M., Boudjelas, S. and De Poorter, M., 2000. 100 of the world's worst invasive alien species: a selection from the global invasive species database (Vol. 12). Auckland: Invasive Species Specialist Group

Glasheen, P.M., Burks, R.L., Campos, S.R. and Hayes, K.A., 2020. First evidence of introgressive hybridization of apple snails (Pomacea spp.) in their native range. Journal of Molluscan Studies, 86(2), pp.96-103.

Yang, Q.Q., He, C., Liu, G.F., Yin, C.L., Xu, Y.P., Liu, S.W., Qiu, J.W. and Yu, X.P., 2020. Introgressive hybridization between two non-native apple snails in China: widespread hybridization and homogenization in egg morphology. Pest management science, 76(12), pp.4231-4239.

Kannan, A., Ratnayeke, S. and Yow, Y.Y., 2021. Molecular evidence of hybridisation in two invasive species of Pomacea (Gastropoda: Ampullariidae) in Peninsular Malaysia. RAFFLES BULLETIN OF ZOOLOGY, 69, pp.570-585.

Hudson, R.R., Slatkin, M. and Maddison, W.P., 1992. Estimation of levels of gene flow from DNA sequence data. Genetics, 132(2), pp.583-589.

**APPENDIX 1.**

Details of sampling sites, number of samples (n), and sampling locations across Taiwan for *Pomacea* spp,

| **Sample site (n)** | **Sample ID** | **Location** | **Date of sampling** |
|---|---|---|---|
| East (42) | ES (HU+TT) | | |
| Hualien (29) | HU1 | 23°57'29.3"N 121°33'57.0"E | 11/5/2022 |
| | HU2 | 23°39'15.9"N 121°32'20.5"E | 11/5/2022 |
| | HU3 | 23°14'18.7"N 121°18'51.0"E | 12/5/2022 |
| Taitung (13) | TT1 | 23°07'34.6"N 121°12'42.5"E | 13/5/2022 |
| | TT2 | 22°57'45.7"N 121°07'52.0"E | 13/5/2022 |
| | TT3 | 22°46'05.7"N 121°07'23.7"E | 14/5/2022 |
| Middle (56) | MD (CH+MIO+TC+YL) | | |
| Changhua (15) | CH1 | 24°03'37.1"N 120°25'09.5"E | 2/9/2021 |
| | CH2 | 24°05'18.5"N 120°35'15.3"E | 2/9/2021 |
| | CH3 | 24°09'58.5"N 120°29'36.1"E | 2/9/2021 |
| Miaoli (14) | MIO1 | 24°39'23.9"N 120°51'25.6"E | 7/7/2022 |
| | MIO2 | 24°29'32.3"N 120°41'45.3"E | 7/7/2022 |
| | MIO3 | 24°24'13.6"N 120°40'06.9"E | 8/7/2022 |

| | | | |
|---|---|---|---|
| Taichung (14) | TC1 | 24°12'46.9"N 120°32'33.8"E | 9/7/2022 |
| | TC2 | 24°21'36.9"N 120°38'14.0"E | 9/7/2022 |
| | TC3 | 24°19'10.2"N 120°43'00.6"E | 10/7/2022 |
| Yunlin (13) | YL1 | 23°38'05.5"N 120°12'29.9"E | 2/8/2022 |
| | YL2 | 23°43'36.2"N 120°15'42.9"E | 2/8/2022 |
| | YL3 | 23°46'34.8"N 120°26'04.7"E | 2/8/2022 |
| North (50) | NR (HS+TP+TY+YI) | | |
| Hsinchu (14) | HS1 | 24°48'05.7"N 120°55'17.1"E | 5/8/2022 |
| | HS2 | 24°50'57.8"N 120°58'17.6"E | 5/8/2022 |
| | HS3 | 24°54'47.3"N 121°01'06.8"E | 5/8/2022 |
| Taipei (12) | TP1, | 25°07'29.0"N 121°29'15.1"E | 12/7/2022 |
| | TP2 | 25°13'34.6"N 121°37'21.8"E | 12/7/2022 |
| Taoyuan (11) | TY1 | 24°58'56.2"N 121°07'29.7"E | 12/7/2022 |
| | TY2 | 25°01'40.8"N 121°13'00.2"E | 15/7/2022 |
| | TY3 | | 15/7/2022 |
| Yilan (13) | YI1 | 24°38'20.1"N 121°49'10.9"E | 4/9/2021 |
| | YI2 | 24°42'01.3"N 121°44'10.6"E | 4/9/2021 |
| | YI3 | 24°48'27.4"N 121°46'51.5"E | 4/9/2021 |
| South (106) | SU (CH+KSH+PT+TN) | | |
| Chiayi (24) | CH1 | 23°32'26.6"N 120°22'50.2"E | 1/9/2021 |
| | CH2 | 23°28'44.5"N 120°10'10.5"E | 1/9/2021 |
| | CH3 | 23°28'41.7"N 120°12'55.8"E | 1/9/2021 |
| Kaohsiung (22) | KSH1 | 22°30'42.0"N 120°23'10.7"E | 12/4/2022 |
| | KSH2 | 22°44'28.8"N 120°16'34.7"E | 12/4/2022 |
| | KSH3 | 22°49'10.7"N 120°17'53.1"E | 12/4/2022 |
| Pingtung (34) | PT1 | 22°00'40.2"N 120°44'13.2"E | 05/01/2023 |
| | PT2 | 22°49'56.2"N 120°36'23.7"E | 05/01/2023 |
| | PT3 | 22°28'18.4"N 120°31'56.4"E | 05/01/2023 |
| Tainan (26) | TN1 | 22°58'43.7"N 120°15'50.9"E | 3/9/2021 |
| | TN2 | 23°04'41.6"N 120°18'24.8"E | 3/9/2021 |
| | TN3 | 23°12'26.1"N 120°16'15.6"E | 3/9/2021 |

**APPENDIX 2.**

List of GenBank accession numbers used to prepare *Pomacea canaliculata* global mitochondrial COI dataset (Dataset 3)

| **Continent** | **Country** | **n** | **Sequence** |
|---|---|---|---|
| Africa | Kenya | 3 | KN_MW363565; KN_MW363566; KN_MW363568 |
| Asia | China | 583 | CHI_MK862299; CHI_MK862295; CHI_MK862294; CHI_MK862293; CHI_MK189419; CHI_MK189413; |

| | | |
|---|---|---|
| CHI_MK189403; | CHI_MK189394; | CHI_MK189393; |
| CHI_MK189362; | CHI_MK189360; | CHI_MK189359; |
| CHI_MK189356; | CHI_MK189354; | CHI_MK189350; |
| CHI_MK189349; | CHI_MK189348; | CHI_MK189347; |
| CHI_MK189345; | CHI_MK189344; | CHI_MK189342; |
| CHI_MK189340; | CHI_MK189339; | CHI_MK189338; |
| CHI_MK189332; | CHI_MK189331; | CHI_MK189330; |
| CHI_MK189329; | CHI_MH293388; | CHI_MH293375; |
| CHI_MH293373; | CHI_MH293372; | CHI_MH293370; |
| CHI_MH293369; | CHI_MH293368; | CHI_MH293367; |
| CHI_MH293366; | CHI_MH293364; | CHI_MH293363; |
| CHI_MH293362; | CHI_MH293361; | CHI_MH293360; |
| CHI_MH293358; | CHI_MH293357; | CHI_MH293356; |
| CHI_MH293354; | CHI_MH293353; | CHI_MH293345; |
| CHI_MH293337; | CHI_MH293336; | CHI_MH293333; |
| CHI_MH211415; | CHI_MH211414; | CHI_MH211413; |
| CHI_MH211412; | CHI_MH211410; | CHI_MH211409; |
| CHI_MH211408; | CHI_MH211407; | CHI_MH211406; |
| CHI_MH211405; | CHI_MH211404; | CHI_MH211403; |
| CHI_MH211402; | CHI_KT884521; | CHI_KT884519; |
| CHI_KT852762; | CHI_KT852761; | CHI_KT852760; |
| CHI_KT852759; | CHI_KT852758; | CHI_KT852722; |
| CHI_KT852721; | CHI_KT852720; | CHI_KT852719; |
| CHI_KT852718; | CHI_KT852717; | CHI_KT852716; |
| CHI_KT852715; | CHI_KT852714; | CHI_KT852713; |
| CHI_KT852712; | CHI_KT852711; | CHI_KT852710; |
| CHI_KT852709; | CHI_KT852708; | CHI_KT852707; |
| CHI_KT693026; | CHI_KT693023; | CHI_KT693017; |
| CHI_KT693015; | CHI_KT693013; | CHI_KT693011; |
| CHI_KT693010; | CHI_KT693007; | CHI_KT693006; |
| CHI_KT693005; | CHI_KT693003; | CHI_KT693002; |
| CHI_KT693000; | CHI_KT692999; | CHI_KT692997; |
| CHI_KT692996; | CHI_KT692995; | CHI_KT692994; |
| CHI_KT692992; | CHI_KT692990; | CHI_KT692989; |
| CHI_KR021013; | CHI_KR021012; | CHI_KR021011; |
| CHI_KR021010; | CHI_KR021008; | CHI_KR021007; |
| CHI_KR021006; | CHI_KR021004; | CHI_KR021003; |
| CHI_KR021002; | CHI_KR021001; | CHI_KR021000; |
| CHI_KR020999; | CHI_KR020998; | CHI_KR020997; |
| CHI_KR020996; | CHI_KR020995; | CHI_KR020994; |
| CHI_KR020978; | CHI_KR020977; | CHI_KR020976; |
| CHI_KR020975; | CHI_KR020974; | CHI_KR020973; |
| CHI_KR020972; | CHI_KR020971; | CHI_KR020970; |
| CHI_KR020969; | CHI_KR020967; | CHI_KR020966; |
| CHI_KR020965; | CHI_KR020964; | CHI_KR020963; |
| CHI_KR020962; | CHI_KP310435; | CHI_KP310434; |

| | | |
|---|---|---|
| CHI_KP310433; | CHI_KP310432; | CHI_KP310431; |
| CHI_KP310430; | CHI_KP310429; | CHI_KP310428; |
| CHI_KP310427; | CHI_KP310426; | CHI_KP310425; |
| CHI_KP310424; | CHI_KP310423; | CHI_KP310422; |
| CHI_KP310421; | CHI_KP310420; | CHI_KP310419; |
| CHI_KP310418; | CHI_KP310417; | CHI_KP310416; |
| CHI_KP310415; | CHI_KP310414; | CHI_KP310413; |
| CHI_KP310412; | CHI_KP310411; | CHI_KP310410; |
| CHI_KP310409; | CHI_KP310408; | CHI_KP310407; |
| CHI_KP310406; | CHI_KP310405; | CHI_KP310404; |
| CHI_KP310403; | CHI_KP310402; | CHI_KP310401; |
| CHI_KP310400; | CHI_KP310399; | CHI_KP310398; |
| CHI_KP310397; | CHI_KP310396; | cCHI_KP310395; |
| CHI_KP310394; | CHI_KP310393; | CHI_KP310392; |
| CHI_KP310391; | CHI_KP310390; | CHI_KP310389; |
| CHI_KP310388; | CHI_KP310387; | CHI_KP310386; |
| CHI_KP310385; | CHI_KP310384; | CHI_KP310383; |
| CHI_KP310382; | CHI_KP310381; | CHI_KP310380; |
| CHI_KP310379; | CHI_KP310378; | CHI_KP310377; |
| CHI_KP310376; | CHI_KP310375; | CHI_KP310374; |
| CHI_KP310373; | CHI_KP310372; | CHI_KP310371; |
| CHI_KP310370; | CHI_KP310369; | CHI_KP310368; |
| CHI_KP310367; | CHI_KP310366; | CHI_FJ946820; |
| CHI_KR021009; | CHI_KP310290; | CHI_MK189424; |
| CHI_MK189410; | CHI_MK189412; | CHI_MK189407; |
| CHI_MK189404; | CHI_MK189401; | CHI_MK189395; |
| CHI_MK189391; | CHI_MK189387; | CHI_MK189385; |
| CHI_MK189383; | CHI_MK189382; | CHI_MK189381; |
| CHI_MK189380; | CHI_MK189379; | CHI_MK189378; |
| CHI_MK189377; | CHI_MK189375; | CHI_MK189374; |
| CHI_MK189373; | CHI_MK189372; | CHI_MK189370; |
| CHI_MK189369; | CHI_MK189368; | CHI_MK189365; |
| CHI_MK189364; | CHI_MK189363; | CHI_MK189361; |
| CHI_MK189358; | CHI_MK189357; | CHI_MK189355; |
| CHI_MK189352; | CHI_MK189351; | CHI_MK189343; |
| CHI_MK189341; | CHI_MK189337; | CHI_MK189336; |
| CHI_MK189335; | CHI_MK189334; | CHI_MK189333; |
| CHI_MK189328; | CHI_MK189327; | CHI_MK189326; |
| CHI_MK189325; | CHI_MK189324; | CHI_MK189323; |
| CHI_MH293450; | CHI_MH293449; | CHI_MH293447; |
| CHI_MH293446; | CHI_MH293445; | CHI_MH293444; |
| CHI_MH293443; | CHI_MH293441; | CHI_MH293440; |
| CHI_MH293438; | CHI_MH293437; | CHI_MH293436; |
| CHI_MH293435; | CHI_MH293434; | CHI_MH293433; |
| CHI_MH293432; | CHI_MH293431; | CHI_MH293429; |
| CHI_MH293428; | CHI_MH293427; | CHI_MH293426; |

| | | |
|---|---|---|
| CHI_MH293425; | CHI_MH293424; | CHI_MH293422; |
| CHI_MH293421; | CHI_MH293420; | CHI_MH293419; |
| CHI_MH293418; | CHI_MH293417; | CHI_MH293416; |
| CHI_MH293415; | CHI_MH293414; | CHI_MH293413; |
| CHI_MH293412; | CHI_MH293411; | CHI_MH293410; |
| CHI_MH293409; | CHI_MH293408; | CHI_MH293407; |
| CHI_MH293406; | CHI_MH293405; | CHI_MH293404; |
| CHI_MH293403; | CHI_MH293402; | CHI_MH293401; |
| CHI_MH293400; | CHI_MH293399; | CHI_MH293398; |
| CHI_MH293397; | CHI_MH293396; | CHI_MH293395; |
| CHI_MH293394; | CHI_MH293393; | CHI_MH293392; |
| CHI_MH293390; | CHI_MH293389; | CHI_MH293386; |
| CHI_MH293385; | CHI_MH293384; | CHI_MH293383; |
| CHI_MH293382; | CHI_MH293381; | CHI_MH293380; |
| CHI_MH293379; | CHI_MH293378; | CHI_MH293377; |
| CHI_MH293352; | CHI_MH293351; | CHI_MH293350; |
| CHI_MH293349; | CHI_MH293348; | CHI_MH293347; |
| CHI_MH293346; | CHI_MH293344; | CHI_MH293343; |
| CHI_MH293342; | CHI_MH293341; | CHI_MH293340; |
| CHI_MH293339; | CHI_MH293338; | CHI_MH293334; |
| CHI_MH293332; | CHI_MH293331; | CHI_MH293330; |
| CHI_MH293329; | CHI_MH293328; | CHI_MH293327; |
| CHI_MH293326; | CHI_MH293325; | CHI_MH293324; |
| CHI_MH293323; | CHI_MH293322; | CHI_MH211411; |
| CHI_MH211401; | CHI_MH211400; | CHI_KT884522; |
| CHI_KT884520; | CHI_KT884518; | CHI_KT852756; |
| CHI_KT852755; | CHI_KT852754; | CHI_KT852753; |
| CHI_KT852752; | CHI_KT852751; | CHI_KT852750; |
| CHI_KT852749; | CHI_KT852748; | CHI_KT852747; |
| CHI_KT852746; | CHI_KT852745; | CHI_KT852744; |
| CHI_KT852743; | CHI_KT852742; | CHI_KT852741; |
| CHI_KT852740; | CHI_KT852739; | CHI_KT852738; |
| CHI_KT852737; | CHI_KT852736; | CHI_KT852735; |
| CHI_KT852734; | CHI_KT852733; | CHI_KT852732; |
| CHI_KT852731; | CHI_KT852730; | CHI_KT852729; |
| CHI_KT852728; | CHI_KT852727; | CHI_KT693022; |
| CHI_KT693021; | CHI_KT693020; | CHI_KT693019; |
| CHI_KT693018; | CHI_KT693016; | CHI_KT693014; |
| CHI_KT693009; | CHI_KT692988; | CHI_KR020993; |
| CHI_KR020992; | CHI_KR020991; | CHI_KR020990; |
| CHI_KR020989; | CHI_KR020988; | CHI_KR020987; |
| CHI_KR020986; | CHI_KR020985; | CHI_KR020984; |
| CHI_KR020983; | CHI_KR020980; | CHI_KR020961; |
| CHI_KR020960; | CHI_KR020959; | CHI_KR020958; |
| CHI_KR020957; | CHI_KR020956; | CHI_KR020955; |
| CHI_KR020954; | CHI_KR020953; | CHI_KR020952; |

| | | | |
|---|---|---|---|
| | CHI_KR020951; | CHI_KR020950; | CHI_KR020949; |
| | CHI_KR020948; | CHI_KR020947; | CHI_KR020946; |
| | CHI_KR020945; | CHI_KR020944; | CHI_KR020943; |
| | CHI_KR020942; | CHI_KP310365; | CHI_KP310364; |
| | CHI_KP310363; | CHI_KP310362; | CHI_KP310361; |
| | CHI_KP310360; | CHI_MK189423; | CHI_KP310359; |
| | CHI_KP310358; | CHI_KP310357; | CHI_KP310356; |
| | CHI_KP310355; | CHI_KP310354; | CHI_KP310353; |
| | CHI_KP310352; | CHI_KP310351; | CHI_KP310350; |
| | CHI_KP310349; | CHI_KP310348; | CHI_KP310347; |
| | CHI_KP310346; | CHI_KP310345; | CHI_KP310344; |
| | CHI_KP310343; | CHI_KP310342; | CHI_KP310341; |
| | CHI_KP310340; | CHI_KP310339; | CHI_KP310338; |
| | CHI_KP310337; | CHI_KP310336; | CHI_KP310335; |
| | CHI_KP310334; | CHI_KP310333; | CHI_KP310332; |
| | CHI_KP310331; | CHI_KP310330; | CHI_KP310329; |
| | CHI_KP310328; | CHI_KP310327; | CHI_KP310326; |
| | CHI_KP310325; | CHI_KP310324; | CHI_KP310323; |
| | CHI_KP310322; | CHI_KP310321; | CHI_KP310320; |
| | CHI_KP310319; | CHI_KP310318; | CHI_KP310317; |
| | CHI_KP310316; | CHI_KP310315; | CHI_KP310314; |
| | CHI_KP310313; | CHI_KP310312; | CHI_KP310311; |
| | CHI_KP310310; | CHI_KP310309; | CHI_KP310308; |
| | CHI_KP310306; | CHI_KP310305; | CHI_KP310304; |
| | CHI_KP310303; | CHI_KP310302; | CHI_KP310301; |
| | CHI_KP310300; | CHI_KP310299; | CHI_KP310298; |
| | CHI_KP310297; | CHI_KP310296; | CHI_KP310295; |
| | CHI_KP310294; | CHI_KP310293; | CHI_KP310292; |
| | CHI_KP310291; | CHI_KP310289; | CHI_KP310288; |
| | CHI_KP310287; | CHI_KP310286; | CHI_KP310285; |
| | CHI_KP310284; | CHI_KP310283; | CHI_KP310282; |
| | CHI_KP310281; | CHI_KP310280; | CHI_KP310279; |
| | CHI_KP310278; | CHI_KP310277; | CHI_KP310276; |
| | CHI_KP310275; | CHI_KP310274; | CHI_KP310273; |
| | CHI_KP310272; | CHI_KP310271; | CHI_KP310270; |
| | CHI_KP310269; | CHI_KP310268; | CHI_KP310267; |
| | CHI_KP310266; | CHI_KP310265; | CHI_KP310264; |
| | CHI_FJ946824; | CHI_EU528579; | CHI_KP310307; |
| | CHI_MH293391; | CHI_MH293387; | CHI_MH293376; |
| | CHI_MH293374; | CHI_MH293371; | CHI_MH293365; |
| | CHI_MH293359; | CHI_MH293355; | CHI_KT852726; |
| | CHI_KT852725; | CHI_KP310444; | CHI_KP310443; |
| | CHI_KP310442; | CHI_FJ946823; | CHI_KT852757; |
| | CHI_KT852724; | CHI_KT852723; | CHI_KR021014; |
| | CHI_KR020981; | CHI_KR020979; | CHI_KP310445; |
| | CHI_KP310441; | CHI_KP310440; | CHI_KP310439; |

| | | | |
|---|---|---|---|
| | | | CHI_KP310436; CHI_FJ946822; CHI_KT852706; CHI_KR021005; CHI_KT693029; CHI_KT693028; CHI_KT693027; CHI_KT693025; CHI_KT693024; CHI_KR106989; CHI_KR021020; CHI_KR021019; CHI_KR021018; CHI_KR021017; CHI_KR021016; CHI_KR021015; CHI_KR020982; CHI_KP310438; CHI_KP310437; CHI_EU528578; CHI_FJ946827; CHI_FJ946826; CHI_FJ946825; CHI_FJ946821 |
| Asia | China-Hong Kong | 2 | HK_KT313034; HK_KT313035 |
| | Indonesia | 3 | IN_EU528580; IN_EU528581; IN_KY574007 |
| | Japan | 18 | JP_AB433757; JP_AB433758; JP_AB433759; JP_AB433760; JP_AB433761; JP_AB433762; JP_AB433763; JP_AB433764; JP_AB433765; JP_AB433766; JP_AB433767; JP_AB433768; JP_AB433769; JP_AB433770; JP_AB433771; JP_AB433772; JP_AB433773; JP_AB433774 |
| | Malaysia | 23 | ML_MN623427; ML_MN623428; ML_MN623429; ML_MN623430; ML_MN623431; ML_MN623433; ML_MN623434; ML_MN623435; ML_MN623436; ML_MN623437; Ml_MN623438; ML_MN623439; ML_MN623440; ML_MN758725; ML_MN758726; ML_MN758727; ML_MN758728; ML_MN758729; ML_MN758730; ML_MN758731; ML_MN758732; ML_MW591143 |
| | Myanmar | 1 | MY_EU528582 |
| | Philippines | 12 | PH_EU528483; PH_EU528484; PH_EU528485; PH_EU528486; PH_EU528487; PH_EU528488; PH_EU528490; PH_EU528491; PH_EU528492; PH_EU528493; PH_EU528494 |
| | Singapore | 1 | SN_KY081757 |
| | Taiwan | 249 | TW_EU528584; TW_EU528586; TW_EU528587 + 246 Present study |
| | Thailand | 2 | TH_EU528497; TH_EU528498 |
| North America | USA | 19 | US_EF514967; US_EF514968; US_EF514969; US_EF514970; US_EF514971; US_EF514972; US_EF514973; US_EF514974; US_EF514975; US_EF514976; US_EF514977; US_EF514978; US_EF514979; US_EF514980; US_EF514981; US_EF514982; US_EF514983; US_EF514984; US_EU523129 |
| Oceania | Papua New Guinea | 1 | PNG_EU528499 |
| South America | Argentina | 32 | ARG_AB728574; ARG_AB728575; ARG_AB728576; ARG_AB728577; ARG_AB728578; ARG_AB728585; ARG_AB728586; ARG_AB728587; ARG_AB728588; ARG_EF514961; ARG_EF514962; ARG_EF514963; ARG_EF514964; ARG_EF514965; ARG_EF514966; |

| | | | |
|---|---|---|---|
| | | | ARG_EU528482; ARG_EU528489; ARG_EU528508; ARG_EU528509; ARG_EU528510; ARG_EU528513; ARG_EU528515; ARG_EU528516; ARG_EU528520; ARG_EU528522; ARG_EU528523; ARG_EU528524; ARG_EU528525; ARG_EU528528; ARG_EU528529; ARG_FJ710314; ARG_FJ710315 |
| | Chile | 4 | Cle_KX965671; Cle_KX965672; Cle_KX965673; Cle_KX965674 |
| | Uruguay | 8 | UR_EU528593; UR_FJ710313; UR_MK992483; UR_MK992484; UR_MK992485; UR_MK992486; UR_MK992487; UR_MK992488 |

**APPENDIX 3.**

**Genetic distance (p-distance) among the taxa used in the phylogenetic tree, Pcan:*P. canaliculate*; Pma: *P. maculata*; Plin: *P. lineata*, Ppal: *P. paludosa*, Pdif: *P. diffusa*, and PC: *P. scalaris***

| | **Pcan** | **Ppal** | **Pdif** | **Plin** | **Pma** | **PC** |
|---|---|---|---|---|---|---|
| **Pcan** | | | | | | |
| **Ppal** | 0.1333 | | | | | |
| **Pdif** | 0.1353 | 0.1429 | | | | |
| **Plin** | 0.1020 | 0.1252 | 0.1411 | | | |
| **Pma** | 0.0928 | 0.1246 | 0.1523 | 0.0852 | | |
| **PC** | 0.1456 | 0.1658 | 0.1340 | 0.1517 | 0.1584 | |

**APPENDIX 4.**

List of GenBank accession numbers used to prepare *Pomacea maculata* mitochondrial COI dataset (Dataset 2)

| **Continent** | **Country** | **n** | **Sequences** |
|---|---|---|---|
| Asia | China | 44 | CHI_FJ946828; CHI_KP310480; CHI_KP310481; CHI_KP310482; CHI_KP310483; CHI_KP310484; CHI_KP310485; CHI_KP310486; CHI_KP310487; CHI_KP310488; CHI_KP310489; CHI_KP310490; CHI_KP310491; CHI_KP310492; CHI_KP310493; |

| | | | |
|---|---|---|---|
| | | | CHI_KP310494; CHI_KP310495; CHI_KP310496; CHI_MK189346; CHI_MK189353; CHI_MK189388; CHI_MK189389; CHI_MK189390; CHI_MK189392; CHI_MK189396; CHI_MK189397; CHI_MK189398; CHI_MK189399; CHI_MK189400; CHI_MK189402; CHI_MK189405; CHI_MK189406; CHI_MK189408; CHI_MK189409; CHI_MK189411; CHI_MK189414; CHI_MK189415; CHI_MK189416; CHI_MK189417; CHI_MK189418; CHI_MK189420; CHI_MK189421; CHI_MK189422; CHI_MK862301 |
| | Japan | 6 | JP_AB433776; JP_AB433777; JP_AB433778; JP_AB433779; JP_AB433780; JP_AB433781 |
| | Malaysia | 12 | ML_MN623417; ML_MN623418; ML_MN623419; ML_MN623420; ML_MN623421; ML_MN623422; ML_MN623423; ML_MN623424; ML_MN623425; ML_MN623426; ML_MN623432 |
| | Taiwan | 8 | Present Study |
| | Thailand | 1 | TH_EU528496 |
| | Vietnam | 1 | VT_EU528502 |
| Europe | Spain | 8 | SP_GU133205; SP_GU133206; SP_GU236486; SP_GU236487; SP_GU236488; SP_GU236489; SP_GU236490; SP_GU236491 |
| North America | USA | 66 | US_EF514942; US_EF514943; US_EF514944; US_EF514985; US_EF515013; US_EF515014; US_EF515015; US_EF515016; US_EF515017; US_EF515018; US_EF515019; US_EF515020; US_EF515021; US_EF515022; US_EF515023; US_EF515024; US_EF515025; US_EF515026; US_EF515027; US_EF515028; US_EF515029; US_EF515030; US_EF515031; US_EF515032; US_EF515033; US_EF515034; US_EF515035; US_EF515036; US_EF515037; US_EF515038; US_EF515039; US_EF515040; US_EF515041; US_EF515042; US_EF515043; US_EF515044; US_EF515045; US_EF515046; US_EF515047; US_EF515048; US_EF515049; US_EF515050; US_EF515051; US_EF515052; US_EF515053; US_EF515054; US_EF515055; US_EF515056; US_EF515057; US_EF515058; US_JX845573; US_KM107825; US_KM107826; US_KM107827; US_KM107828; US_KM107829; US_KM107830; US_KR350494; US_KR350495; US_KR350496; US_KR350497; US_KR350498; US_KR350499; US_KX149128; US_KX810826; US_MK992475 |
| Oceania | New Zealand | 1 | NZ_HQ908057 |

| | | | |
|---|---|---|---|
| South America | Argentina | 14 | ARG_AB728579; ARG_AB728580; ARG_AB728582; ARG_AB728583; ARG_AB728584; ARG_EF514945; ARG_EU528481; ARG_EU528495; ARG_EU528514; ARG_EU528521; ARG_EU528526; ARG_EU528527; ARG_EU528530; ARG_EU528531 |
| | Brazil | 60 | BR_EF514986; BR_EF514987; BR_EF514988; BR_EF514989; BR_EF514990; BR_EF514991; BR_EF514992; BR_EF514993; BR_EF514994; BR_EF514995; BR_EF514996; BR_EF514997; BR_EF514998; BR_EF514999; BR_EF515000; BR_EF515001; BR_EF515002; BR_EF515003; BR_EF515004; BR_EF515005; BR_EF515006; BR_EF515007; BR_EF515008; BR_EF515009; BR_EF515010; BR_EF515011; BR_EF515012; BR_EU528533; BR_EU528538; BR_EU528539; BR_EU528540; BR_EU528544; BR_EU528549; BR_EU528550; BR_EU528551; BR_EU528552; BR_EU528554; BR_EU528557; BR_EU528558; BR_EU528559; BR_EU528560; BR_EU528561; BR_EU528562; BR_EU528563; BR_EU528566; BR_EU528567; BR_EU528568; BR_EU528569; BR_EU528570; BR_EU528571; BR_EU528572; BR_EU528575; BR_EU528576; BR_MK992470; BR_MK992471; BR_MK992472 ; BR_MK992479 ; BR_MK992480; BR_MK992481; BR_MK992482 |
| | Uruguay | 5 | UR_MK992473; UR_MK992474; UR_MK992476; UR_MK992477; UR_MK992478 |

**APPENDIX 5.**

Haplotype distribution of *Pomacea* spp. in 14 counties/cities across Taiwan (full names of abbreviation can be found in Table S1.)

| ***Pomacea canaliculata*** | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **HU** | **TT** | **CH** | **MIO** | **TC** | **YL** | **HS** | **TP** | **TY** | **YI** | **CY** | **KSH** | **PT** | **TN** | **Total** | **%** |
| **Hap_1** | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 1 | 0.407 |
| **Hap_2** | 1 | 6 | - | 5 | 3 | - | 1 | - | 3 | 2 | 4 | 5 | 10 | 4 | 44 | 17.886 |
| **Hap_3** | 24 | 4 | 12 | 8 | 7 | 6 | 12 | 11 | 8 | 7 | 14 | 11 | 15 | 16 | 155 | 63.008 |
| **Hap_4** | - | 2 | - | - | - | - | - | - | - | - | - | 2 | - | 1 | 5 | 2.033 |
| **Hap_5** | - | - | - | - | 1 | - | - | 1 | - | - | 1 | - | 1 | 1 | 5 | 2.033 |
| **Hap_6** | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 1 | 0.407 |
| **Hap_7** | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 1 | 0.407 |

| | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Hap_8** | 2 | - | - | - | - | 7 | - | - | - | - | - | - | 7 | - | 16 | 6.504 |
| **Hap_9** | - | - | 1 | - | - | - | 1 | - | - | - | - | 1 | - | - | 3 | 1.220 |
| **Hap_10** | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.407 |
| **Hap_11** | - | - | 1 | - | 3 | - | - | - | - | 1 | 2 | - | - | - | 7 | 2.846 |
| **Hap_12** | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.407 |
| **Hap_13** | - | - | - | - | - | - | - | - | - | 1 | - | - | - | - | 1 | 0.407 |
| **Hap_14** | - | - | - | - | - | - | - | - | - | 1 | - | - | - | - | 1 | 0.407 |
| **Hap_15** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.407 |
| **Hap_16** | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.407 |
| **Hap_17** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.407 |
| **Hap_18** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.407 |
| ***Pomacea maculata*** | | | | | | | | | | | | | | | | |
| | **HU** | **TT** | **CH** | **MIO** | **TC** | **YL** | **HS** | **TP** | **TY** | **YI** | **CY** | **KSH** | **PT** | **TN** | **Total** | **%** |
| **Hap_19** | - | - | - | 1 | - | - | - | - | - | 1 | - | 3 | - | - | 5 | 62.5 |
| **Hap_20** | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 1 | 12.5 |
| **Hap_21** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | 1 | 12.5 |
| **Hap_22** | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 12.5 |

**APPENDIX 6.**

Haplotype distribution of *Pomacea canaliculata* from the global mitochondrial COI dataset (Dataset 3)

| | **TW** | **Cle** | **AR** | **UR** | **US** | **PNG** | **HK** | **CH** | **JP** | **ML** | **TH** | **SN** | **PH** | **IN** | **MY** | **KN** | **Total** | **%** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Hap_1** | 162 | - | 9 | - | 17 | 1 | 2 | 214 | 7 | 11 | - | - | - | - | - | - | 423 | 44.48 |
| **Hap_2** | - | - | 2 | - | - | - | - | - | - | - | - | - | - | - | - | - | 2 | 0.21 |
| **Hap_3** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_4** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_5** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_6** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_7** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_8** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_9** | 1 | - | 2 | - | - | - | - | - | - | - | - | - | - | - | - | - | 3 | 0.32 |
| **Hap_10** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_11** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_12** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_13** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_14** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |

| | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Hap_15** | - | - | 2 | - | - | - | - | - | - | - | - | - | - | - | - | - | 2 | 0.21 |
| **Hap_16** | 46 | - | - | - | - | - | - | 1 | 4 | 2 | - | - | - | - | - | 3 | 56 | 5.89 |
| **Hap_17** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_18** | - | - | 3 | - | - | - | - | - | - | - | - | - | - | - | - | - | 3 | 0.32 |
| **Hap_19** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_20** | 1 | - | - | - | - | - | - | 319 | 1 | - | - | - | - | - | - | - | 321 | 33.75 |
| **Hap_21** | - | - | - | - | - | - | - | 14 | - | - | - | - | 1 | - | - | - | 15 | 1.58 |
| **Hap_22** | - | - | - | - | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_23** | 5 | - | - | - | - | - | - | 11 | 3 | - | - | - | - | - | - | - | 19 | 2.00 |
| **Hap_24** | 1 | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 2 | 0.21 |
| **Hap_25** | 6 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 6 | 0.63 |
| **Hap_26** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_27** | 16 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 16 | 1.68 |
| **Hap_28** | 4 | - | - | - | - | - | - | - | 2 | - | - | 1 | - | - | - | - | 7 | 0.74 |
| **Hap_29** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_30** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_31** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_32** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_33** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_34** | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_35** | - | - | - | - | - | - | - | 2 | - | - | - | - | - | - | - | - | 2 | 0.21 |
| **Hap_36** | - | - | - | - | - | - | - | 16 | 1 | - | - | - | - | - | - | - | 17 | 1.79 |
| **Hap_37** | - | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_38** | - | - | - | 2 | - | - | - | - | - | - | - | - | - | - | - | - | 2 | 0.21 |
| **Hap_39** | - | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_40** | - | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_41** | - | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_42** | - | - | - | 2 | - | - | - | - | - | - | - | - | - | - | - | - | 2 | 0.21 |
| **Hap_43** | - | - | - | - | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_44** | - | - | - | - | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_45** | - | - | - | - | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_46** | - | - | - | - | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_47** | - | 4 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 4 | 0.42 |
| **Hap_48** | - | - | - | - | - | - | - | - | - | - | - | - | - | 2 | - | - | 2 | 0.21 |
| **Hap_49** | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | 1 | 0.11 |
| **Hap_50** | - | - | - | - | - | - | - | - | - | 1 | - | - | - | - | - | - | 1 | 0.11 |
| **Hap_51** | - | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | 1 | 0.11 |
| **Hap_52** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_53** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_54** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_55** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_56** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_57** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |

| | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Hap_58** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_59** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_60** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_61** | - | - | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | 1 | 0.11 |
| **Hap_62** | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | - | - | 1 | 0.11 |
| **Hap_63** | - | - | - | - | - | - | - | - | - | - | 1 | - | - | - | - | - | 1 | 0.11 |
| **Hap_64** | - | - | - | - | 2 | - | - | - | - | - | - | - | - | - | - | - | 2 | 0.21 |
| **Hap_65** | - | - | 1 | - | - | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.11 |
| **Total** | 249 | 4 | 32 | 8 | 19 | 1 | 2 | 582 | 18 | 14 | 2 | 1 | 12 | 3 | 1 | 3 | 951 | |

**APPENDIX 7.**

Comparison of Taiwan haplotype positions in different datasets, for *Pomacea canaliculata* Dataset 1 (Taiwan sequence) compared with Dataset 2 (global dataset of *Pomacea canaliculata*), and for *Pomacea maculata* Dataset 1 (Taiwan sequences) compared with Dataset 3 (global dataset of *Pomacea maculata*)

| **Dataset 1** | **Networks** | **Dataset 3 (*P. canaliculata*)** | **Network** |
|---|---|---|---|
| *P. canaliculata* | | | |
| Hap 1 | Network Tw-A | Hap 24 | Network Wc-A |
| Hap 2 | Network Tw-B | Hap 16 | Network Wc-B |
| Hap 3 | Network Tw-A | Hap 1 | Network Wc-A |
| Hap 4 | Network Tw-B | Hap 25 | Network Wc-B |
| Hap 5 | Network Tw-B | Hap 23 | Network Wc-B |
| Hap 6 | Network Tw-A | Hap 26 | Network Wc-A |
| Hap 7 | Network Tw-C | Hap 9 | Network Wc-C |
| Hap 8 | Network Tw-B | Hap 27 | Network Wc-B |
| Hap 9 | Network Tw-C | Hap 28 | Network Wc-C |
| Hap 10 | Network Tw-B | Hap 16 | Network Wc-B |
| Hap 11 | Network Tw-A | Hap 1 | Network Wc-A |
| Hap 12 | Network Tw-A | Hap 1 | Network Wc-A |
| Hap 13 | Network Tw-B | Hap 29 | Network Wc-B |
| Hap 14 | Network Tw-B | Hap 20 | Network Wc-B |
| Hap 15 | Network Tw-A | Hap 30 | Network Wc-A |
| Hap 16 | Network Tw-A | Hap 31 | Network Wc-A |
| Hap 17 | Network Tw-B | Hap 32 | Network Wc-B |
| Hap 18 | Network Tw-B | Hap 33 | Network Wc-B |
| **Dataset1** | **Networks** | **Dataset 2 (*P. maculata*)** | **Networks** |
| *P. maculata* | | | |

| | | | |
|---|---|---|---|
| Hap 19 (exclude KSH 27) | Network Tw-A | Hap 1 | Network Wm-A |
| Hap 19 (only **KSH 27**) | Network Tw-A | Hap 8 | Network Wm-A |
| Hap 20 | Network Tw-A | Hap 6 | Network Wm-A |
| Hap 21 | Network Tw-A | Hap 7 | Network Wm-A |

**APPENDIX 8.**

Haplotype distribution of *Pomacea maculata* from the global mitochondrial COI dataset (Dataset 2)

| | **TW** | **BR** | **AR** | **UR** | **US** | **NZ** | **SP** | **CH** | **JP** | **ML** | **TH** | **VT** | **Total** | **%** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Hap_1** | 4 | - | 4 | 1 | - | - | - | - | - | - | - | - | 9 | 3.982 |
| **Hap_2** | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_3** | - | - | - | 1 | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_4** | - | - | - | 2 | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_5** | - | - | - | - | - | - | - | - | - | - | - | 1 | 1 | 0.442 |
| **Hap_6** | 2 | 1 | - | - | 14 | 1 | 8 | 44 | 3 | 11 | 1 | - | 85 | 37.611 |
| **Hap_7** | 1 | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_8** | 1 | - | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_9** | - | - | 1 | - | 26 | - | - | - | 3 | - | - | - | 30 | 13.274 |
| **Hap_1-** | - | - | 1 | - | 21 | - | - | - | - | - | - | - | 22 | 9.735 |
| **Hap_11** | - | - | - | - | 1 | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_12** | - | - | - | - | 4 | - | - | - | - | - | - | - | 4 | 1.770 |
| **Hap_13** | - | - | 1 | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_14** | - | - | 1 | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_15** | - | - | 3 | - | - | - | - | - | - | - | - | - | 3 | 1.327 |
| **Hap_16** | - | - | 1 | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_17** | - | - | 1 | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_18** | - | - | 1 | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_19** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_2-** | - | 6 | - | - | - | - | - | - | - | - | - | - | 6 | 2.655 |
| **Hap_21** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_22** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_23** | - | 7 | - | - | - | - | - | - | - | - | - | - | 7 | 3.097 |
| **Hap_24** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_25** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_26** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_27** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_28** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_29** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |

| | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Hap_3-** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_31** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_32** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_33** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_34** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_35** | - | 4 | - | - | - | - | - | - | - | - | - | - | 4 | 1.770 |
| **Hap_36** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_37** | - | 4 | - | - | - | - | - | - | - | - | - | - | 4 | 1.770 |
| **Hap_38** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_39** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_4-** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_41** | - | 3 | - | - | - | - | - | - | - | - | - | - | 3 | 1.327 |
| **Hap_42** | - | 1 | - | - | - | - | - | - | - | - | - | - | 1 | 0.442 |
| **Hap_43** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_44** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_45** | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 | 0.885 |
| **Hap_46** | - | 3 | - | - | - | - | - | - | - | - | - | - | 3 | 1.327 |