Chapter 12
Azolla and Bougainville's Voyage Around the World

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12.1 Introduction

The French expedition that was led by Louis Antoine de Bougainville (1729–1811) and that sailed around the world from 1766 to 1769 is well known for its contribution to science, namely, geography, astronomy, zoology and botany, in particular the discovery of *Bougainvillea* in Brazil. Unknown to many, however, is the fact that it was during this voyage that the first *Azolla* samples for science purposes were collected and included in Commerson's herbarium. This year (2017), this botanical event commemorates 250 years. The geographic origin of these *Azolla* samples collected by Philibert Commerson (1727–1773) and Jeanne Baret (1740–1807) was and still is a controversial matter, given that the information published by Jean-Baptiste Lamarck (1744–1829) in 1783 indicates that they were collected at the Strait of Magellan (Carrapiço 2010a; Lamarck 1783). In addition, the origin of *Azolla*'s name is often a source of confusion, based on two contradictory ideas. In conclusion, much of what is known about this fern's history and biology needs to be updated and complemented with new information. It is our goal in this chapter to shed new light on the untold history and biology of this important aquatic fern.

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12.2 A New Look on the Taxonomy and Biology of *Azolla*

Azolla is a small-leafed floating or semiaquatic heterosporous fern presenting overlapping scale-like bilobed leaves covering a slender and branched rhizome that floats horizontally on the freshwater surface of tropical and temperate environments (Carrapiço 2017; Svenson 1944). The roots of this fern are simple and emerging on the ramification points of the ventral side of the stem. The genus *Azolla* belongs to the family Azollaceae (Schneller 1990; Wettstein 1903) and includes seven species divided into two sections: *Rhizosperma* (*A. pinnata* and *A. nilotica*) and *Azolla* (*A. filiculoides*, *A. caroliniana*, *A. mexicana*, *A. microphylla* and *A. rubra*) (Adams et al. 2013; Pereira et al. 2011). However, the systematics of the family Azollaceae, namely, the section *Azolla*, is an open issue still controversial and confusing. In 2004, Evrard and Van Hove carried a review of the taxonomy of the New World *Azolla* species (section *Azolla*) reintroducing the species *A. cristata*, which was introduced in 1847 by the German botanist Georg Heinrich Mettenius (1823–1866), based on the previous classification made by Georg Friedrich Kaulfuss (1786–1830) in 1824 (Evrard and Van Hove 2004; Mettenius 1847; Svenson 1944). Evrard and Van Hove based their work on three morphological characters: leaf papillae, megaspore perine surface and number of glochidia septa. They rearranged the *Azolla* cluster in two species: *A. filiculoides* (grouping *A. filiculoides*, *A. caroliniana* Willd. and *A. microphylla* Kaulf.) and *A. cristata* (including *A. caroliniana* auct. non-Willd., *A. microphylla* auct. non-Kaulf. and *A. mexicana* Presl.) (Evrard and Van Hove 2004; Pereira et al. 2011). However, one of the characters used, the "number of glochidia septa", varies among specimens of *A. caroliniana* and *A. filiculoides*, and for that reason, it is not a good taxonomic character (Pereira et al. 2011). Furthermore, our research carried out on the species of the sections *Rhizosperma* and *Azolla* obtained from the IRRI germplasm collection and based on morphological and molecular data, namely, RAPD markers, does not support this proposed classification, meaning that the results indicate the existing of a two-section ranking, the *A. pinnata* and *A. nilotica* in section *Rhizosperma* and five distinct species on section *Azolla*: *A. mexicana*, *A. microphylla*, *A. caroliniana*, *A. filiculoides* and *A. rubra* (Pereira et al. 2011).

Each bilobed leaf of *Azolla* has an immersed, thick, greenish or reddish and photosynthetic dorsal lobe and a very thin (one cell thick), immersed hyaline ventral lobe. The chlorophyllous dorsal lobe contains an ellipsoid cavity, measuring 0.15 × 0.30 mm, which opens to the exterior through a pore (Caiola and Forni 1999; Carrapiço 2002, 2010a, b; Veys et al. 1999). This extracellular compartment contains a permanent and specific endosymbiotic prokaryote community, a heterocyst-forming filamentous nitrogen fixing cyanobacterium, usually assigned to *Anabaena azollae*, as well as several genera of bacteria, such as *Arthrobacter*, *Corynebacterium* and *Agrobacterium* (Carrapiço 1991, 2017). These microsymbionts are specific of this association and live immobilized in a mucilaginous fibrillary network, which fills the peripheral area of the cavity. The mucilaginous material of the leaf cavity is delimited by two envelopes, an external and an internal one, leaving the centre empty and probably filled with gas or liquid (Carrapiço 2010a, b). The presence of the prokaryote community, which lives in permanent symbiosis with *Azolla*, plays a relevant role in the plant's survival skills when faced with new conditions (Carrapiço 2010b). This is related to the fact that this community constitutes a source of evolutionary innovation, where symbionts are the means through which a rapid renovation and adaptation of the entire symbiotic system occurs, as a response to new environmental challenges and with consequences at the metabolical, physiological, and even genetic levels (Carrapiço 2010b).

Azolla is able to maintain an inoculum of the prokaryote community (cyanobacteria and bacteria) between plant generations (Carrapiço 2010a, 2017; Ran et al. 2010), and sporocarps act as the transfer vehicles during sexual reproduction (Ran et al. 2010). The majority of these studies refer to the cyanobionts, but we believe that the same process can also be applied to the bactobionts (Carrapiço 1991, 2010a, 2017). This process relies on the capacity of the cyanobiont to differentiate motile non-heterocystous hormogonial filaments, which are attracted to and enter the megasporocarp through a narrow pore (Ran et al. 2010). The mechanism by which the cyanobacteria cells become associated with the developing sporocarps involves branched epidermal trichomes, which arise on the ventral surface of the apical meristem and probably includes chemical mediators (Adams et al. 2013; Carrapiço 2017; Perkins and Peters 1993). On entering the megasporocarp, the hormogonia differentiate into akinetes in the indusium chamber above the megaspore, remaining dormant until the plant germinates (Adams et al. 2013), initiating a new generation of the life cycle of *Azolla*.

Azolla has been traditionally used as green manure for rice cultivation and animal feed not only in China and in Vietnam, in the past few centuries (Lumpkin and Plucknett 1980; Moore 1969), but also in Africa (Carrapiço 2010a) and in Central and South America (Mariano Montaño 2017, personal communication). In addition to agricultural purposes, there are other areas where the *Azolla-Anabaena* association has been applied. The use of *Azolla* as biofilter and bioremediation tool for contaminated waters has been developed in the last three decades with success (Costa et al. 1999; Costa et al. 2009; Forni et al. 2001; Sela et al. 1989). Recently, new areas of research have emerged related to this fern. The use of this symbiotic system as a model for biological, genomic, phytochemical and physiological studies (De Vries et al. 2016; Qiu and Yu 2003), namely, evolutionary developmental biology (De Vries and De Vries, in Chap. 2 of this book), and as an epistemological tool to understand the post-neo-Darwinian evolutive process (Carrapiço 2015) is currently taking place. In biotechnological research, including biological based life-support systems (BLSS), *Azolla* can be incorporated in bioregenerative space devices for cosmos exploration (Carrapiço 2010a).

The *Azolla* leaf cavity behaves as both the physiological and dynamic interface unit of this symbiotic association, where the recognition of molecular functions happens and the main metabolic and energetic flows occur (Caiola and Forni 1999; Carrapiço 2010b). Two morphologically distinct classes of epidermal trichomes (simple hairs and multicellular branched hairs), present in the leaf cavity, are involved in the transfer and uptake of metabolites from the fern to the prokaryote colony and from the endosymbionts to the host. In the vacuoles of the apical cells of the simple hairs, a mixture of several chemical compounds, namely, lipids, polysaccharides, polyphenols and alkaloids or alkaloid-like compounds, was detected (Carrapiço 2017; Carrapiço and Tavares 1989; Pereira and Carrapiço 2007). These chemical compounds may play a role in the communication between the host and the symbionts, in the selection of microorganisms that are not useful to the fern, in the control of the endosymbionts in the cavity and in the establishment and maintenance of the symbiosis (Carrapiço 2017). In this sense, this cavity can be considered to be a natural microcosm (Carrapiço 2002), a special micro-ecosystem which reveals a self-organization and an ecological defined structure, allowing the prokaryotes speciation or to form ecotypes adapted to specific environmental conditions (Carrapiço 2017).

Although traditionally considered as a lower vascular plant, *Azolla* exhibits symbiotic characteristics more evolved than the other vascular plant-cyanobacterial symbioses, such as *Cycas* and *Gunnera* (Adams et al. 2013; Carrapiço 2006). There appears to be no direct correspondence between the fern's evolutionary phylogeny and the complexity of the symbiosis (Carrapiço 2010a). This unique symbiosis is sustained throughout the fern's life cycle, where the cyanobiont and bacteria are always present and transferred from one generation to the next, either in the dorsal leaf cavities or in the megasporocarps, indicating the obligatory nature of the symbiosis (Carrapiço 2010a). It means that *Azolla* is not infected de novo, suggesting a specific phylogenetic co-evolution of the symbionts and the host, and can be considered as a natural photobioreactor with millions of years of evolution (Shi and Hall 1988). It also represents a good example of hereditary symbiosis, showing that the complexity of the relationship between the host and the symbionts can be recognized as a new level of biological organization in evolution. All this data leads to the idea that the *Azolla*-*Anabaena*-bacteria association works in a synchronized way and can be considered to be a polygenomic entity, in which the different genomes operate together in a complementary and synergistic way for the whole. It also suggests that we are in the presence of a superorganism or symbiome/holobiont in both biological and ecological terms (Carrapiço 2010a, 2015, 2017) in which new metabolic and organic capabilities are acquired and developed by the partners, which establish a new level of organization that goes beyond the individual capabilities of any individual partner. A good example of it can be found in the nitrogen metabolism shared by the host and its partners in this symbiotic system. The atmospheric nitrogen fixed by the cyanobacterium through the heterocysts is converted into ammonia and released into the leaf cavity. It has been shown that intracellular ammonia pools of symbiotically associated *Azolla* are five times bigger than those of endophyte-free *Azolla* (Braun-Howland and Nierzwicki-Bauer 1990). The activities of ammonia assimilating enzymes in the isolated trichomes of the dorsal leaf cavity were much higher than those in *Azolla* leaves, while these activities in the *Anabaena* filaments were repressed to very low levels. The host accounts for at least 90% and 80% of the total glutamine synthetase and NADH-dependent glutamate dehydrogenase activities, respectively. These results suggest that hair cells play an important role in the assimilation of the nitrogen fixed and released into the cavity by the cyanobiont. The nitrogen is transferred to the fern, after having been acquired during the development of the symbiotic process (Carrapiço 2010b; Uheda 1990). This data indicates that the synergies associated with symbiosis had and continue to have a leading role in the morphological, reproductive, physiological and metabolical complexification of the organisms in evolution (Corning 1983, 2005). In fact, the fossil records also suggest that the relationship between the fern and the cyanobiont was established in the mid-Cretaceous, which implies that the two organisms have been co-evolving for at least 80 million years (Jonathan Bujak 2017, personal communication; Bujak 2007; Bujak and Bujak 2014).

Data from the 2004 Arctic Coring Expedition (ACEX) cores, drilled in the central Arctic Ocean near the North Pole, show the presence of fossil *Azolla* in Eocene sediments (~48,5 Ma) (Brinkhuis et al. 2006). The plant's remains occur as laminations, reflecting seasonal or longer cycles, and they have been observed in more than 50 Arctic wells from northern Alaska, the Canadian Beaufort and the Chukchi Sea (Bujak 2007). The presence of repeated *Azolla* laminations in the central Arctic Ocean also indicates that the *Azolla* plants grew in situ on freshwater layers that repeatedly developed on the surface of the Arctic Ocean, rather than being transported from freshwater bodies on the surrounding land. Brinkhuis et al. (2006) and Bujak (2007) also suggested that the enormous quantities of *Azolla* inhabiting the Eocene Arctic Ocean for almost a million years may have triggered the initial shift from the Mesozoic greenhouse world towards our present icehouse state. According to their model, CO_2 absorption by the fern resulted in an abrupt reduction in this atmospheric gas with critical consequences in the climatic change and implications for the global biogeochemical cycles. This period is referred to as the *Azolla* event (Brinkhuis et al. 2006; Brouwer 2017; Bujak 2007; Carrapiço 2010a).

12.3 The Bougainville's Voyage: A Journey Through Utopia

In 1766, Louis-Antoine de Bougainville begins the political and scientific voyage around the world under the auspicious of the French King Louis XV (Taillemite 2006). This expedition involved two ships, the frigate *Boudeuse*, commanded by Bougainville and Nicolas-Pierre Duclos-Guyot (1722–1794) second in command, and the storeship *Étoile*, commanded by François Chenard de la Giraudais (1727–1776) (Carr 1968; Dunmore 2002; Williams 2013). There were 214 persons on board the *Boudeuse* and 116 on board the *Étoile* (Ridley 2010). Philibert Commerson and Jeanne Baret were travelling aboard the latter, him as royal botanist, naturalist and physician and her as his assistant or valet with knowledge in medicinal plants as it was suggested by Ridley (2010). Knapp (2011), however, considers Ridley's idea that Jeanne Baret was an "herb-woman" to be mere a speculation. Commerson was also a follower of the Genevan philosopher Jean-Jacques Rousseau (1712–1778) and disciple of the Swedish naturalist Carl Linnaeus (1707–1778), who in 1754 asked him to carry out research on marine plants, fishes and shells of the Mediterranean (Williams 2013).

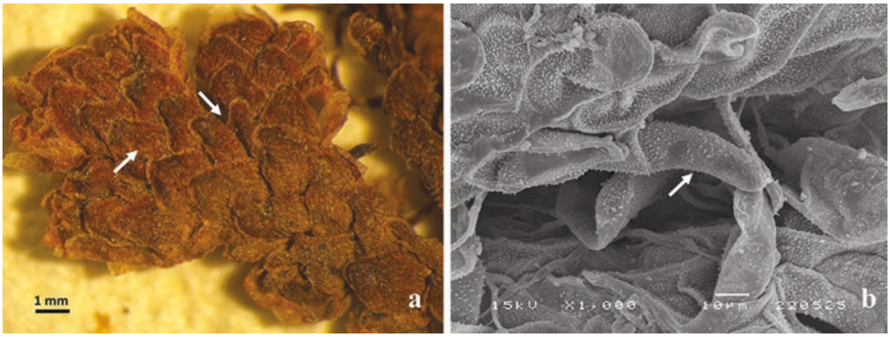

Fig. 12.1 *Azolla* samples from Commerson's herbarium, MNHN, Paris. Sporophyte of *Azolla* observed with stereo microscope and presenting the typical overlapping scale-like leaves (arrows) (**a**). Detail of unicellular papilla of dorsal leaf lobe (arrow) observed in SEM and characteristic of *A. filiculoides* (**b**)

The first ship to sail from France (Nantes) was the *Boudeuse* on 15 November 1766. However, but due to a storm in the Bay of Biscay, the ship had to return to France, to Brest, where she was repaired and returned to the sea on 6 December. From there, she sailed to South of America en route to the Falklands (Malvinas in Spanish or Malouines in French) (Bougainville 1771; Monnier et al. 1993). The *Étoile* was not ready to sail at that time and only left France (Rochefort) on 1 February 1767 (Monnier et al. 1993). The first mission of Bougainville was to deliver the Falklands to Spain, which happened on 1 April 1767 (Monnier et al. 1993; Taillemite 2006). After that, the *Boudeuse* sailed to Rio de Janeiro to meet the *Étoile*, which has arrived to this city on 13 June. A month later, on 15 July, both ships sailed to the Rio de la Plata, having arrived on 31 July to the city of Montevideo (Monnier et al. 1993; Taillemite 2006). They stayed there until November, visiting Buenos Aires too. During this period, several field trips were carried out, in which many biological samples were collected by Commerson and Baret (Williams 2013), including the first *Azolla* samples for scientific purposes. Some of this material prepared for herbarium was sporulated, which enabled us to identify its species using scanning electron microscopy (SEM) of the megaspore perine surface and also the leaf papillae. As it was expected, the *Azolla* species was *A. filiculoides* (Figs. 12.1 and 12.2).

We must mention that these field trips in the region of La Plata were really challenging for Commerson, who was hindered by a bad leg (Williams 2013), meaning that the success of the botanizing work was in great part due to Baret. In fact, François Vivez (1744–1828), surgeon of the *Étoile*, referring to Jeanne Baret's work in his journal, reinforces this idea saying that "she worked like a black. During our period of call at the River Plate, she went to collect plants in the plain, in the mountains two or three leagues away carrying a musket, a game-bag, food supply and paper for the plants" (Taillemite 2006; Williams 2013).

On 5 December 1767, the two ships began crossing the Strait of Magellan, which gives access to the Pacific Ocean. The crossing took almost 2 months, more precisely

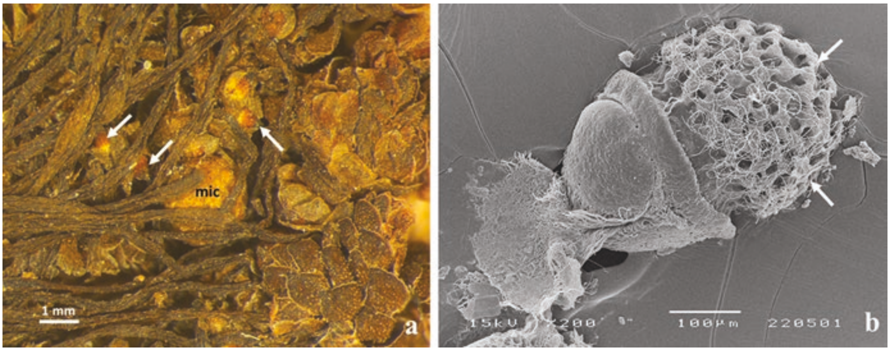

Fig. 12.2 *Azolla* samples from Commerson's herbarium, MNHN, Paris. Some of this material was sporulated. Megasporocarps (arrows) and microsporocarp (mic) observed with stereo microscope (**a**). Megasporocarp with a visible megaspore perine surface ornamentation (arrows) observed in SEM and characteristic of *A. filiculoides* (**b**)

52 days (Taillemite 2006) with a stormy and cold weather in unpleasant environmental conditions (Williams 2013). It is interesting to mention that the Portuguese navigator and explorer Fernão de Magalhães (1480–1521), at the service of the Spanish Kingdom, took 27 days to cross the Strait in 1520 but with much better weather conditions (Taillemite 2006). During the crossing, Baret and Commerson did several trips inland to collect biological specimens (Dunmore 2002; Olivier 1909; Williams 2013). François Vivez referring to Jeanne Baret in this period said "she spent entire days in the forest with snow, rain and ice, to seek plants, or along the seashore for shells. It remains for me to say in her praise that she generally surprised everyone by the work she did" (Dunmore 2002; Taillemite 2006).

After sailing through the Strait of Magellan, the two ships entered into the Pacific Ocean en route to the unknown Tahiti, where they arrived on 2 April 1768 (Taillemite 2006). This island was named "Nouvelle Cythère" by the Bougainville's French expedition, without knowing that in June of the previous year the British naval officer and explorer Samuel Wallis (1728–1795), commander of the *Dolphin*, declared Tahiti to be an English Crown territory on behalf of King George III (Dunmore 2002; Ridley 2010; Williams 2013). In fact, Wallis only returned to England in May 1768 which explains why Bougainville had no knowledge of this discovery (Dunmore 2002). The name "Nouvelle Cythère" was "inspired from the Greek island where Aphrodite, the goddess of love and beauty, was said to have been born" (Dunmore 2002). The island was also claimed as French territory (Williams 2013) and is currently part of the French Polynesia (French Overseas Country) following its annexation in 1880, when it was proclaimed a colony of France.

Many of the officers of the Bougainville's expedition were Rousseau's followers, including Bougainville himself as well as Commerson. For this reason, they had high expectations in relation to the inhabitants of the New Cythera island and the evolution of their society. The theory of the "noble savage", according to which man in its natural state was born essentially good, without guilty, was widely accepted in the eighteenth century and served as a key philosophical reference for these men. In earlier primitive societies, mankind would have been happier and less exploitative than in modern societies where corruption and oppression established the pattern for development, exploitation and poverty (Dunmore 2002; Williams 2013). According to personal journals of several members of the expedition, Tahiti seemed to correspond to a utopian and romantic world, where happiness and love were the main rules in society (Dunmore 2002; Ridley 2010; Williams 2013). The island's society, however, was not as romantic and peaceful as they thought (Williams 2013).

It was in these conditions that an incident happened with Jeanne Baret (while disguised as a man) when she was collecting samples onshore in Tahiti. It was Bougainville who described it in his journal. When Commerson and Baret were doing herborization work, a number of native men surround her yelling "Ayenne" (girl in local language) and attempted to carry her off, revealing that the Commerson's assistant was a woman (Williams 2013). Jeanne Baret travelled much of the trip disguised as a man until she was discovered on April 1768 (Dunmore 2002; Ridley 2010; Schiebinger 2003; Williams 2013). Although she has denied that Commerson knew her real identity, we know that they had lived together after Commerson's wife, Antoinette Vivante Beau, passed away on 19 April 1762, following the birth of a son 3 days earlier (Ridley 2010). In fact, Jeanne Baret was hired as housekeeper in 1764 when she was 5 months pregnant. When she was declaring her pregnancy (as required by law) on 22 August 1764, in Digoin outside Lyon, she refused to tell the name of the child's father (Ridley 2010; Schiebinger 2003, 2004). In September, they both moved to Paris, where in January 1765, a baby boy, called Jean-Pierre Baret, was born. The baby only lived a few months (Monnier et al. 1993; Ridley 2010; Schiebinger 2003, 2004).

After leaving the paradisiacal Tahiti, the French expedition visited several Pacific islands before arriving to Port Louis in the Isle de France (Mauritius) on November 1768. This island had become a French territory in 1715 and would remain so until 1810, when Great Britain took control of the island (Dunmore 2002). In 1768, the intendant was Pierre Poivre (1719–1786), a naturalist and friend of Commerson, who invited him to stay on the island to carry out botanical work (Dunmore 2002; Williams 2013). Having accepted the invitation, Commerson and Baret would conduct research, over the following 4 years, on the natural resources of Isle de France, Bourbon (Réunion) and Madagascar. They focused in particular on plants with pharmacological interest, as well as on spice plants whose trade was controlled by the Dutch. For this natural work, they were helped by two young illustrators provided by Pierre Poivre: Paul Philippe Sanguin de Jossigny (1750–1827) and Poivre's godson, Pierre Sonnerat (1748–1814) (Williams 2013).

In the meantime, Commerson's health had been deteriorating. In a letter dated 19 October 1772 and sent from the Isle of France to Paris, to his friend the astronomer Jérôme Lalande (1732–1807), he refers to these health problems in a prophetic way saying that "after an attack of rheumatic gout, which kept me in bed for nearly three months, I thought I was convalescent, when, in addition, dysentery attacked me; up to the present it was been incurable, and it was brought me to the very edge of the grave. My strength is almost utterly exhausted, and I am already more than half worn out. If the country air and diet of rice and fish do not cure me of this attack, you may well, as you once said (prophetically, no doubt), begin to work on the history of my martyrology". (Olivier 1909). He tried to obtain some relief from these health problems by moving to the East coast of the Isle of France, but on 13 March 1773 at 11:45 p.m., Commerson died at Le Flacq, at a friend's house, only a few weeks after his last botanical excursion and before the news that he had been elected member of the Académie des Sciences could reach him on 21 March 1773 (Dunmore 2002; Olivier 1909; Williams 2013). He was buried the next day in the Flacq cemetery, although his grave was never identified (Dunmore 2002).

12.4 The Winds of Liberty and the Emotions of the Natural World

As we have mentioned previously, Philibert Commerson and Jeanne Baret travelled in the same cabin on the *Étoile*. It was a very risky and delicate decision, but they were in a lovely relation not only with science (Knapp 2011; Ridley 2010; Schiebinger 2003, 2004). The sanitary conditions in both ships were very precarious, namely, inadequate for women (Carr 1968), which reveals a very brave and courageous attitude on the part of Jeanne Baret who decided to travel on the *Étoile* with Commerson and with an all-male crew. When the voyage took place, it was forbidden by royal naval ordinance for officers and sailors to have women on board, which led her to disguise herself as a man (Schiebinger 2003, 2004). The importance of her work to herbalize and to discover new botanical species has not been emphasized by many authors, but it was thanks to her work that many of the biological specimens have seen the daylight and reached France on May 1774 after the Commerson's death at Flacq in the Mauritius Island (Isle de France) on 13 March 1773 at the age of 45 (Monnier et al. 1993). Despite Philibert Commerson being a very good and determined botanist with a large expertise, it is well known that he was not a very organized person and was often ill or physically diminished during the voyage, namely, due to an injured leg made by a coach accident in Niort prior the journey and later gout crisis, which jeopardized part of his goals as naturalist (Bour 2015; Dunmore 2002; Williams 2013). However, Jeanne Baret, 13 years younger than Commerson, provided him with support and assistance during field trips, namely, by collecting and preparing biological specimens. As Bougainville referred in his 1771 historical book *Voyage autour du monde par la frégate du roi la Boudeuse et la flûte l'Étoile; en 1766,1767,1768&1769*, Jeanne Baret was "an expert botanist", and also Commerson "praised her for the countless plants she collected, the herbaria she constructed, the many collections of insects and shells she curated" (Schiebinger 2003). As a tribute to Baret's work, Commerson named one of the collected plants as *Baretia bonnafidia*, a flowering plant from Madagascar known for its "doubtful sexual characteristics" (Schiebinger 2004). However, Commerson never published it, and posteriorly this plant was renamed and included in the genus *Turraea* belonging to the family Meliaceae (Knapp 2011; Ridley 2010; Schiebinger 2004). It was only in 2012 that a definitive tribute to Jeanne Baret was achieved in recognition to the importance of her botanical work in science through the paper of Tepe et al. (2012), which named a new species of *Solanum* (*S. baretiae*) in her honour.

A considerable part of the biological material first collected in Brazil and La Plata failed to arrive to France (Olivier 1909; Williams 2013), leading Commerson to vow never to separate himself from his collections (Olivier 1909). After Commerson's death, several bureaucratic problems emerged between the Isle de France administration and the Commerson's biological material executors, which complicated the transfer of the collections to the Jardin du Roi in Paris (renamed Jardin des Plantes in 1793, after the 1789 revolution) (Williams 2013). Furthermore, many of the collections and manuscripts were not organized. Since it was decided to expedite the transfer of the material to France, it was not possible for the executors to catalogue it appropriately (Williams 2013). In November 1773, the boxes with the biological collections and his manuscripts were sent to the Jardin du Roi in Paris through the ship the *Victoire* which arrived at Lorient on May 1774 (Taillemite 2006). The number of boxes sent is still a matter for discussion. According to Christinat (1996), it seems there were 32 boxes, although the number indicated by the inventory letter sent by the Intendant Jacques Maillart-Dumesle (1730–1782) from Port Louis was 34. It was Paul de Jossigny, who has worked with Commerson as illustrator in the Isle de France, that travelled and escorted this material to Paris and delivered it to Georges-Louis Leclerc de Buffon (1707–1788) and Bernard de Jussieu (1699–1777) at the Cabinet d'Histoire Naturelle du Roi, including the drawings of the specimens collected by Commerson (Bour 2015; Williams 2013).

Using the new excellent database of the Muséum National d'Histoire Naturelle (MNHN) in Paris, we have found 3986 specimens of vascular plants collected by Philibert Commerson and, although not referred, also by Jeanne Baret in several locations during the Bougainville's voyage and also during his stay at Mauritius, Madagascar and Réunion (Le Bras et al. 2017). These plants sent to Paris were in most cases new for science. This material and other biological samples collected by Commerson and Baret were almost "cannibalized" by the scholarly world, namely, by the established French scientific community, though often without proper attributions (Williams 2013). I believe that a good example of it can be found in the well-known *Bougainvillea*, a flowering plant from Brazil and named by Commerson in honour of Bougainville in 1767. This plant, although with the name of *Buginvillaea*, and other 36 with their attributions were described by Antoine Laurent de Jussieu in 1789 in the *Genera Plantarum* (Jussieu 1789; Williams 2013) few days before the storming of the Bastille and the beginning of the French Revolution. In 1799, the German botanist Carl Ludwig Willdenow (1765–1812) published in the *Species Plantarum* the description of the species *Buginvillaea spectabilis* Willd. (currently *Bougainvillea spectabilis* Willd.) (Willdenow 1799) without any reference to Commerson. The same seems to have happened to the *Azolla* specimens collected by Philibert Commerson and Jeanne Baret, which were studied and published on by several scientists without reference to the original herborization work and research. Nevertheless, it is important to mention that Commerson never published his observations, as he has died before having the possibility to deepen his research. According to Georges Cuvier (1769–1832), "it is astounding that one man should have been able to do so much in so short of time and in a tropical climate" (Olivier 1909; Williams 2013). The collaborative partnership between Commerson and Baret was key in completing this task successfully (Knapp 2011).

What happened to Jeanne Baret after Commerson's death? We know that she became the owner of a small tavern at Port Louis (Isle de France), because she was fined for selling alcohol during the mass period in December 1773 (Dunmore 2002; Ridley 2010). After this reference, her name reappears in the official records in connection with her marriage to Jean Duberna (or Dubernat), a French officer, on 17 May 1774 in Port Louis (Dunmore 2002; Knapp 2011; Ridley 2010). The couple returned to France in early to mid 1775, settling down in Saint-Aulaye in Dordogne, some 80 km north of Bordeaux (Dunmore 2002). In 1785, she was granted a pension of 200 livres a year by the Ministry of Marine, as well as the official and public recognition of being an "extraordinary woman" for assisting Commerson's work during the voyage, for sharing with him the dangers of the mission and for showing great courage in her duty (Dunmore 2002). Although not mentioned, it is suspected that Bougainville was behind this decision (Dunmore 2002). On 5 August 1807, she died at home in the small village of Saint-Aulaye at the age of 67 (Dunmore 2002), leaving everything she possessed to Commerson's heirs, namely, his son Archambault (1762–1834) (Olivier 1909). She was the first woman to do the circumnavigation of the world (Ridley 2010).

12.5 Who Discovered *Azolla*?

The first reference and crude illustration of this plant in the literature was made by the French priest and naturalist Louis Feuillée (1660–1732), based on a Peruvian specimen. Published in 1725 in the *Journal des Observations Physiques, Mathematiques et Botaniques* under the name of *Muscus squamosas aquaticus elegantissimus*, the reference also included that the plant was used for improving the production of chicken eggs (Carrapiço 2010a, b, 2017; Evrard and Van Hove 2004; Feuillée 1725). The first samples for herbarium purposes of this aquatic pteridophyte were collected in 1767 by Philibert Commerson and Jeanne Baret in Montevideo and Buenos Aires region. After Commerson's death, this material was freely studied by several naturalists, among them were Marc Antoine Louis Claret de La Tourrette (1729–1793), Jean-Baptiste Lamarck, Antoine Laurent de Jussieu (1748–1836), Carl Ludwig Willdenow, Augustin Pyramus de Candolle (1778–1841) and Antoine Laurent Apollinaire Fée (1789–1874). Although Lamarck describes in 1783, in the *Encyclopédie Méthodique*, the species *Azolla filiculoides* referring that the plant was brought from Magellan (Carrapiço 2010a; Lamarck 1783), we believe this was an error. This assumption is based on the information in Bougainville's journal, which makes reference to these places, dates and existing samples at the

Fig. 12.3 Sheets from Commerson's herbarium featuring *Azolla* samples stemming from several botanical institutions: Muséum National d'Histoire Naturelle de Paris (**a**), Conservatoire et Jardin botaniques de la Ville de Genève (**b**), Jardin botanique de Lyon (**c**), Institut de Botanique de l'Université de Montpellier (**d**)

Herbarium of the Jardin botanique de Lyon, the Herbarium of the Muséum National d'Histoire Naturelle de Paris, the Herbarium of the Institut de Botanique de l'Université de Montpellier and the Herbarium of the Conservatoire et Jardin botaniques de la Ville de Genève (Fig. 12.3). An interesting note can be found in the Antoine Laurent de Jussieu's herbarium sheet existing at the Muséum National d'Histoire Naturelle of Paris (MNHN-P-P00668168) and probably written by the hand of this botanist, indicating that "M. Lamarck l'a annoncé du detroit de Magellan, par erreur" and referring to these samples from Buenos Aires and Montevideo and were included in Commerson herbarium (Fig. 12.4). It is also important to mention that the environmental conditions of the Strait of Magellan were not adequate to the biology, development and ecology of *A. filiculoides* (Roberto Rodríguez-Rios 2010, personal communication).

The *Azolla* ecological and biological description made by Lamarck was only possible if he had the opportunity to observe the live plant. This was, however, not the case, as *Azoll*a had arrived in dried form on a herbarium sheet, and this fern did not exist in Europe at the time (Lamarck 1783; Marsh 1914). In fact, *Azolla* was only introduced in Europe in 1872 in several botanic gardens, from which it escaped into ditches and ponds (Marsh 1914). The solution to this Lamarck's enigma can probably be found in the information included by Philibert Commerson with the collected *Azolla* and its ecology, which was observed by him and by Jeanne Baret in loco (Carrapiço 2010a). The published description made by Lamarck indicates that *Azolla* "C'est une petite plante aquatique, qui paroît flotter à la surface des eaux à la manière des Lenticules, (Lemna) avec lesquelles elle semble avoir beaucoup de

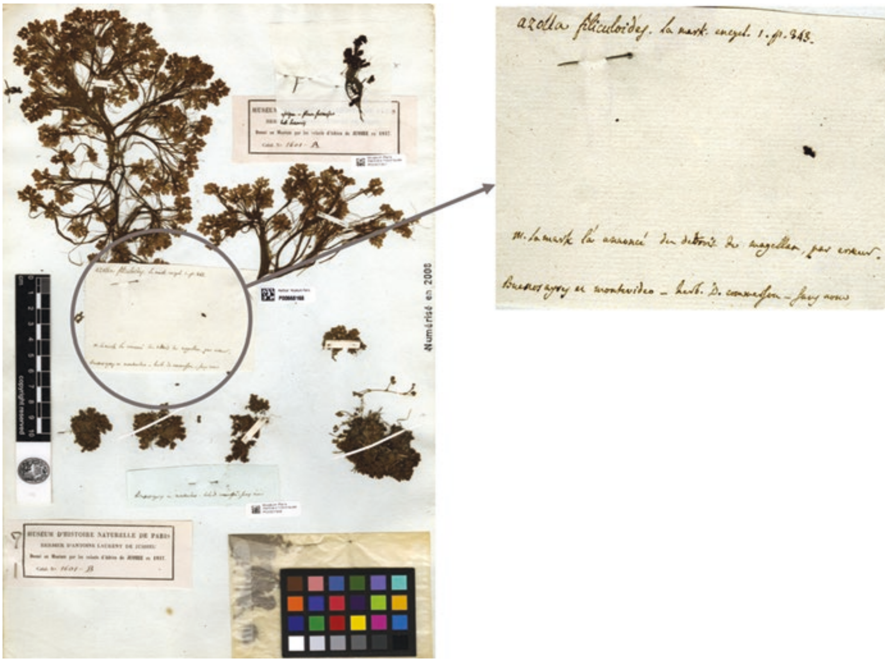

Fig. 12.4 Antoine Laurent de Jussieu's herbarium sheet with a note referring to Lamarck's error, Muséum National d'Histoire Naturelle of Paris (MNHN-P-P00668168). Probably handwritten by Jussieu, indicating that "M. Lamarck l'a annoncé du detroit de Magellan, par erreur"

rapports, et qui a néanmoins l'aspect d'une très petite fougère" and "cette plante a eté rapportée de Magellan par M. de Commerson" (Carrapiço 2010a; Lamarck 1783). This error made by Lamarck related to the *Azolla* location had consequences for the fern systematics. As an example, Carl Ludwig Willdenow studied the *Azolla* samples from the Commerson herbarium and published in 1810 in *Species Plantarum* that there was a new species of this fern, named *Azolla magellanica* Willd. (Willdenow 1810) (meaning *Azolla* from the Strait of Magellan). This species was in fact *A. filiculoides*. It was also in this publication that *Azolla caroliniana* was described for the first time based on material derived from Richard (Louis Claude Richard, 1754–1821) in Paris, and probably collected by Michaux (André Michaux, 1746–1802) in the Southeastern United States (Svenson 1944), referring to the geographic location of the plant collected.

Regarding the origin of the name *Azoll*a, we know that Lamarck described *A. filiculoides* without referring to it. Several authors have suggested that the genus is a result of two words of Greek origin: *azo*, to dry, and *ollyo*, to kill, alluding to death from drought (Gledhill 2008; Lumpkin and Plucknett 1980; Moore 1969; Stearn 1992). This interpretation seems contrived, and we believe that the name may correspond to a native word for the plant (Dyce 1988, edited and expanded by John Edgington in 2016; Carrapiço 2010a). We do not know for certain who was responsible for coining the name *Azolla*, but Commerson and Baret have probably adopted the designation used by the native population in Buenos Aires or in Montevideo having included it in the description sent to the Jardin des Plantes. Commerson's description was eventually used by Lamarck in the *Encyclopédie Méthodique* (Lamarck 1783) although without proper attributions.

12.6 Conclusion

Azolla is an aquatic fern that presents an endosymbiotic prokaryotic community (cyanobacteria and bacteria) in the chlorophyllous dorsal lobe of the leaf. This symbiotic association is sustained throughout the fern's life cycle, where the cyanobiont and bacteria are always present and transferred from one generation to the next, either in the dorsal leaf cavities or in the megasporocarps, indicating the obligatory nature of the symbiosis. Given all the referred main physiological and ecological characteristics, the *Azolla* leaf cavity can be seen as the "Rosetta Stone" of the symbiosis, allowing us to understand the dynamic and the synergistic processes of this symbiotic system (Carrapiço 2017). It represents the key to comprehending *Azolla* as a superorganism (Carrapiço 2010a) and to understanding how this remarkable and unique fern can be "domesticated" in view of an improved biobased future economy (Brouwer et al. 2014).

Azolla did not exist in Europe when the Bougainville's voyage around the world took place in the eighteenth century, from 1766 to 1769. It was in Montevideo and Buenos Aires, and not in the Strait of Magellan, that this plant was collected for the first time for science purposes and prepared for herbarium by Philibert Commerson and Jeanne Baret. Following Commerson's death in the Isle de France (Mauritius) in 1773, his herbarium and manuscripts, including all biological specimens collected by himself and Baret, were shipped to the Jardin du Roi in Paris and distributed among several scientists affiliated to a number of scientific institutions. In 1783, Lamarck describes in the *Encyclopédie Méthodique* the species *Azolla filiculoides* referring that the plant was brought from Magellan by Philibert Commerson (Carrapiço 2010a; Lamarck 1783). This reference, however, was an error related to the geographic location of the biological material collected by Commerson.

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