Chapter 2
The Hidden Biodiversity Data Retained in Pre-Linnaean Works: A Case Study with Two Important XVII Century Italian Entomologists

Francesco Andrietti and Carlo Polidori

Abstract Before Linnaeus published the *Systema Naturae*, in which introduced the modern species concept, a huge amount of information on ecology, behaviour and diversity of many animals had been accumulated. This information, often extremely detailed, suffers from the lack of the assignation of the studied organisms to their modern specific names. Here, we examine in detail the works of Antonio Vallisneri (1661–1730), one of the most important figures of early experimental entomology in Italy. We analyse the ecological and ethological contributions of Vallisneri, as well as those that Diacinto Cestoni (1637–1718), another Italian naturalist, sent to Vallisneri, to the knowledge of parasitoid, predatory and gall-making wasps (Hymenoptera), by studying the *Saggio de' Dialoghi sopra la curiosa origine di molti Insetti* and the *Quaderni di Osservazioni* I-III, trying to assign current taxonomy to the observed insects based on eco-ethological and morphological descriptions. Valuable data have been found in the analysed works on taxonomically diverse ecological webs involving wasps. Information regarded a variety of hymenopteran parasitoids of other Hymenoptera, dipteran parasitoids of Hymenoptera, coleopteran parasitoids of Hymenoptera, and hymenopteran parasitoids associated with non-hymenopteran hosts. Overall, about 20 wasp genera could have been objects of Vallisneri and Cestoni observations, which include the first detailed ecological and ethological data on many of them. Detailed re-examinations of ancient studies may contribute to our knowledge on biodiversity by providing historical distribution data as well as unveiling trophic interactions that may have been modified due to biodiversity loss in the last century.

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E. Casetta et al. (eds.), *From Assessing to Conserving Biodiversity*,
History, Philosophy and Theory of the Life Sciences 24,
https://doi.org/10.1007/978-3-030-10991-2_2

Keywords Vallisneri · Cestoni · Hymenoptera · Parasitoid wasp · Gall wasp

2.1 Introduction

Naturalists provided through many centuries a large amount of data on the ecology, behaviour, distribution and morphological diversity of animals. However, before Linnaeus published the *Systema Naturae* (Linnaeus 1735), in which the modern species concept was introduced, this information, often extremely detailed, obviously lacked the assignation of the studied organisms to their modern specific names, making these old studies a great source of hidden biodiversity data. A detailed re-examination of these works could, however, bring these data to surface in a modern context. In particular, studying ancient works is important for several reasons: (1) it may provide historical distribution information on organisms before Linnaeus, a period generally not covered in the analyses of communities' variation through time; (2) it may provide additions on species composition and species interactions in habitats and environments that may have been lost in the last century through human activity; (3) it may unveil information on behavioural ecology of species that could be compared with information on behavioural ecology retrieved from modern populations of the same species. All these points are directly linked with three of the main shortfalls of biodiversity knowledge: the Wallacean shortfall (the knowledge on the geographic distribution of most species is incomplete), the Raunkiæran shortfall (lack of knowledge on species' traits and their ecological functions) and the Eltonian shortfall (lack of enough knowledge on species' interactions and their effects on individual survival and fitness) (Hortal et al. 2015).

The present study makes an attempt to bring to light old "hidden" biodiversity data by primarily analysing the works of Antonio Vallisneri (1661–1730), a naturalist strongly associated with the development of experimental entomology in Italy. Particularly concerning wasp (Hymenoptera) biology, Vallisneri is credited one of the first people who correctly interpreted insect parasitism. Secondarily, because additional important observations on wasp biology were also recovered (and published) by Vallisneri from letters that Diacinto Cestoni (1637–1718), another Italian naturalist, sent to him, we also analysed the Cestoni observations.

The motivation underlying Antonio Vallisneri's analysis of the world of insects proves to be two-fold:

1. The first, that we shall call simply *biological*, relating to its origin, but which, at the time, was considered to be *philosophical*, as it was set within the great debate about the science of life between the supporters of spontaneous generation and those which, instead, maintained that every animal necessarily came from a parent.
2. The second, a more specific motivation, which led him to examine in minute detail the lives and habits of these insects with a curiosity and zeal which made him a prototype of the modern naturalist and entomologist, desirous to directly verify the results of the facts he was describing and to throw off the weight of that erudite stance which still strongly characterized naturalistic studies.

These two points are actually only apparently separate as there are significant elements conjoining them. Although Redi, around the mid XVII century, had shown that the commonly held belief that many insects spontaneously materialized in rotting matter – an opinion given authority by Aristotle himself – was false, the case of insects which were *parasitoids* of others was still problematic, as was that of insects found inside plant galls (Parke 2014). In both cases the various situations seemed to confirm the theory of *equivocal* generation, which many hoped to refute. Only a very careful and thorough examination of these phenomena could clarify the problem satisfactorily.

These authors focused their studies mainly on parasitoid wasps and galler wasps, but also on predatory wasps and bees, thus covering many life-histories shown by Hymenoptera. This insect order includes ants, bees, wasps and sawflies and is one of the richest (115,000 species described worldwide), and represents a diverse and ecologically relevant component of biodiversity, since its members are involved in a wide range of interactions with plants (pollination, herbivory, gall formation), fungi (parasitism), and other animals, both invertebrates and vertebrates (parasitoidism, predation) (La Salle and Gauld 1993). Furthermore, many Hymenoptera interact with other components of environment, such as the soil (e.g. through nesting activities). Thus, any new information on species identities and their biology that could be retrieved from ancient studies may be important for biodiversity studies. Indeed, from one side the species is the most commonly used unit of biological diversity, and from the other side, adding data on behaviour and ecology to the species identification substantially increases the detail of biodiversity studies, since it gives information on the functioning of ecosystem and may also bring to light intraspecific diversity (Boenigk et al. 2015). At last, relying not only on species taxonomic identity but also on their behavioural ecology can be relevant for biodiversity conservation, since these data may help understanding how proximate and ultimate aspects of behaviour can be of value in preventing biodiversity loss (Berger-Tal et al. 2011).

In this chapter, we analyse the contribution of Vallisneri (and Cestoni) to the knowledge of wasps with two different life-style: parasitoid and predatory wasps (whole females lay eggs on or into a host and caused host death by brood feeding on it) and gall wasps (whose females lay eggs into plant tissues and cause tissue deformations (galls) serving as food for the brood). We will confine ourselves to illustrating some of the ecological and ethological aspects in which Vallisneri was particularly interested, and, more specifically, we will try to identify the considered insects. The latter task is not always easy, as the descriptions provided by Vallisneri are very different from those used in the current system, and only in a few cases the insects at issue can be properly identified, at least at the genus-level. Some of the eco-ethological data provided in the work of Vallisneri (and Cestoni) are particularly interesting in the light of biodiversity studies, since they are directly related with food webs (i.e. parasitoid – host relationships), and it is well known how food webs are vulnerable to biodiversity loss (Dunne et al. 2002).

As regards parasitoid and predatory wasps, we have used the *Saggio de' Dialoghi sopra la curiosa origine di molti Insetti* (Vallisneri 1696, 1700) as the main source,

whereas, for gall wasps, we have mainly used the *Quaderni di Osservazioni* (Vallisneri 2004; Vallisneri 2007; Vallisneri unpublished manuscript), of which volumes I and II have been recently transcribed.

2.2 Parasitoid and Predatory Wasps

The *Dialoghi sopra la curiosa origine di molti Insetti* take place between two personages, Pliny and Malpighi, the revered scholar who had died only 2 years previously, and who is no other than a stand in for Vallisneri himself. In the first *Dialogo* Pliny, who supports traditional beliefs, says

> May the Modern Gentlemen forgive me, they are depriving themselves of a great, compassionate Mother always ready to succour them in their most urgent needs. They should keep her close as a reference point, because they will eventually find forms of generation which they will not know how to explain honourably, without recourse to this universal benefactor, or some other phantom they have thought up. Amongst other things which may still be hidden, I have seen little flies hatching out of the eggs of caterpillars, and Large Flies, Flies and Wasps from the Chrysalises or Cocoons of Butterflies and moths, inside which I do not know how the mothers could have laid their eggs, or their worms. (Vallisneri 1696, p. 311)

Pliny has in fact hit on some of the most distinctive features of the parasitoids, in other words, the fact that they develop, feed and grow at the expense of eggs or larvae of other insects. In some cases these parasitoids are Diptera, but they are mostly Hymenoptera of the Parasitica (= Terebrantia) group. As to whether they are called flies or wasps or even bees should be disregarded, since Vallisneri often uses these terms interchangeably.

Malpighi replies

> One day, from 40 eggs of a big Butterfly [or moth], of the size and colour of the sort of millet they call "of the "Sun", hatched over a hundred tiny Little Flies […] in each egg I saw two holes, one big one […] and ten times bigger than the other […] out of which the little flies kept on coming out, the other just visible with the Microscope on top of each one […] The tiny hole was the one made by the worms, which penetrated the egg searching for food, […] the big hole was made by the Little Flies, after they had become Chrysalises inside the egg, and then flying insects […] In short, they went in by one door and came out of the other, and nothing was generated by itself inside there, but rather came out of the eggs of the same sort of Little Flies […] even if they were so small as to look like little flying atoms, I could make out their nubby little antennae, […] heads, and wasp-like bodies […] we might call them tiny little wasps. (Vallisneri 1696, pp. 311–312)

In these passages, Malpighi is referring to wasps which live on the eggs of a not specifically identified butterfly or moth ("Parpaglione"), inside which they hatch and develop, presumably belonging to the Trichogrammatidae or Platygastridae as already observed by Pampiglione et al. (2000, p. 16). We are here dealing with a case of endophagous oophagy, because the whole life cycle of the parasite takes place inside the egg of its host, from which the parasite emerges as an adult wasp. The small hole is probably not where the larvae got in, as Vallisneri mistakenly believes, but was made by the ovipositor of the mother wasp when laying the egg.

In another case the butterfly is easier to identify

> […] one day I saw odd little flies hatch out from half of the eggs of a butterfly with eyespots on its wings and, from the other half, black caterpillars. (Vallisneri 1696, p. 312)

The pattern on the wings and the colour of the caterpillar seem to be those of the *Inachis io* (peacock) butterfly. The eggs of this species are parasitized especially by *Telenomus* (Platygastridae) and *Trichogramma* (Trichogrammatidae) (Hondō et al. 1995), which are amongst the most active parasitoids of butterfly and moth eggs (Shaw et al. 2009, p. 147).

Malpighi lengthily discusses other cases of oophagous parasitoids, particularly those emerging from the eggs of Heteroptera (bugs) and Homoptera (Vallisneri 1696, p. 312) observing that, even if there is sometimes a specific relationship between the species of host and parasitoid, some hosts may be parasitized by various parasitoid species. He then goes on to talk of cases in which the parasitoids do not emerge from the eggs of the host, but from its larva or chrysalis or cocoon

> Others from large cocoons made of the rough silk of the second Moth, inside which one day I found fourteen empty Chrysalises […] (Vallisneri 1696, p. 312)

The appearance of the cocoon and the dimension of the moth suggest the possibility that we are dealing with the genus *Saturnia* (*pavonia*?), which is known to have numerous parasites that hatch from its cocoons (Grandi 1984, Vol. II, pp. 249–251; Peigler 1994, p. 3), where they complete their development. Amongst the most common are the dipteran *Masicera* (Grandi 1984, Vol. II, pp. 249, 251, 556; Peigler 1994, p. 81) and *Agrothereutes* (= *Spilocryptus*) (Ichneumonidae) (Grandi 1984, Vol. II, p. 251; Peigler 1994, p. 23) a number of whose pupae can be found in a single moth cocoon.

Parasitoids can also be found in the hard, earthen nests of *Sceliphron* mud-dauber wasps (Sphecidae) or the cocoons of the caddis fly (Trichoptera) (Vallisneri 1696, p. 312): presumably, these are mostly parasitoids from the Ichneumonidae and Braconidae (including from the subfamily Aphidiinae), with perhaps a few dipterans. In the case of the Trichoptera, we are dealing with *Agriotypus* (Ichneumonidae: Agriotypinae).

Vallisneri seems to have noticed the difference between eggs laid on or inside the body of the host i.e. between endo- and ectoparasitoidism

> […] having seen various holes one day under the Microscope in the body of a caterpillar I had just discovered (I am not referring to their "breathing holes" [spiracles]) and, another day, various small eggs amongst their hairs, this led me to understand immediately that those too came from the Mothers […] (Vallisneri 1696, p. 312)

He also reports

> I also saw many Little Flies hatch out of the small, spherical, almost membranaceous, follicles of a little worm, which ought to turn into a certain type of wee aphid [gorgoglioncino], which dwells in the Great Mullein and the Tiny Leaves Figworts, and also out of another worm which, as it was found between the two membranes of the external part of the leaves of an elm, should have hatched out as an aphid similar in form to those found on broad beans, grass peas, and similar legumes. (Vallisneri 1696, p. 312)

This is actually an observation made by Francesco Mattacodi, which was passed on to Vallisneri, in the same words given in the above quote, in a letter sent to him, which was subsequently published by Vallisneri himself (Vallisneri 1726, pp. 61–62). Tremblay and Masutti (2005, p. 37) have apparently wrongly identified this parasitoid ("Little Flies") since, in our opinion, its host, the "wee aphid" ("gorgoglioncino") does not belong to the genus *Aphis* (Homoptera: Aphididae) as "the spherical, almost membranous follicles" remind one more of a scale insect (Homoptera: Coccidae). As concerns its parasitoid, we are spoiled for choice, even if it cannot be a genus specialized in parasitizing aphids (subfamily Aphidiinae) as was proposed (Tremblay and Masutti 2005, p. 37). It might have been a chalcidoid wasp from the subfamilies Aphelininae (Aphelinidae) or Encyrtinae (Encyrtidae). The latter are known to cause multiple infestations: thus a single egg laid by the parasitoid can produce a number of its like, as we can see from the above quote.

Moreover, Vallisneri also subsequently introduces other quotes from the same letter received from Mattacodi into his *Dialoghi,* without any reference as to their source

> From live forest Bugs, and live Beetles ["Cantaridi"] I have observed on several occasions after a single worm has come out of their nether regions, without any detriment to them. It was enclosed in a perfect egg [=cocoon], and then produced a nice little fly or, when enclosed in a little follicle, a fly elongated in shape came out of it. (Vallisneri 1696, pp. 312–313)

And, immediately afterwards,

> Many little worms, which also came out of two caterpillars raised from eggs, wrapped themselves up shortly afterwards in long cocoons, from which emerged little flies, without preventing the said caterpillars from turning into the usual Chrysalises, and afterwards the sort of whitish butterflies which are so harmful to Cabbages […] (Vallisneri 1696, pp. 312–313)

The two passages quoted above come from Mattacodi's letter (Vallisneri 1726, p. 61) and follow the same wording almost exactly. In the first case this could refer, according to Tremblay and Masutti (Tremblay and Masutti 2005, p. 37), to an infestation of Heteroptera and adult Coleoptera by tachinid Diptera; the second seems to bear witness to parasitoids emerging from the skin of the caterpillar (which, when the infestation is not excessive, may survive and finish its life cycle (e.g. Meijden et al. 2000), which will be further discussed below.

Mattacodi also observed parasitoids emerging from insect eggs, giving emphasis to the case of the "biggest moth" (*Saturnia pyri*) and believing, like Vallisneri after him, that the parasitoid entered in its larval form, not as an egg (Vallisneri 1726, p. 61). He would, moreover, observe parasitoids emerging from butterfly chrysalises, although he admitted that this had already been observed by Vallisneri (Vallisneri 1726, p. 61), who would put the fact into his *Dialoghi*. In particular Mattacodi relates that he had found a cocoon which, in addition to the remains of its maker, contained another cocoon belonging to the parasitoid, which, however, he again believed to have entered as a larva, not as an egg (Vallisneri 1726, p. 62).

The basic problem concerns the birth of the parasitoids. Indeed, Pliny goes on to ask: "And all these, and those, do they come out of an egg, or out of the worm?" ["E tutte queste, e questi, nascono dall'uovo, o dal verme?"] (Vallisneri 1696, p. 312).

There follows Malpighi's answer

> Those who can find the patience to look will always see the two holes described in all the eggs. They will never see Little Flies or Large Flies or Flies emerge from the Chrysalises if they protect them well enough, I mean the cocoons and such like, from which it can be seen that a Mother is always required. It seems far stranger that those which come out of a Caterpillar's body alive should be born of a Mother, and then make their cocoons immediately or harden into Chrysalises […] (Vallisneri 1696, p. 312)

The last sentence reported above alludes to what happens, for example, with the genus *Apanteles* (Braconidae), a parasite of lepidoptera larvae, in which they lay numerous eggs during the early stages of their host's development. After having grown to full size, the larvae emerge from their caterpillar host, making their own cocoons on or beside it.

Pliny recalls the observations of other two naturalists who had seen the same phenomenon, Aldrovandi and Redi; however, both of them had taken the cocoons of the parasitoids which had emerged from the host for eggs laid by the caterpillar (Vallisneri 1696, p. 313). To which Malpighi replies

> I doubt that both these great men, let it be said with all reverent modesty, could have been mistaken. As you know, it is not caterpillars which lay eggs, but butterflies, and even if they confused eggs with Chrysalises, I still do not believe that the caterpillars made the silk cocoons mentioned by Signor Redi, but that the worms did so as soon as they had hatched […] That this is what happens is clearly shown by the silk, which they admit to having seen around them, which certainly did not come out of the rear end of the caterpillars, and my eye showed this to me one day, as I had seen them hatch and, then and there, make the little cocoons described. (Vallisneri 1696, p. 313)

In the second *Dialogo* Pliny introduces the matter of the "vespe icneumoni". We can find mention of these in texts by Aristotle (as well as in Pliny and Aldrovandi), even if these are "very unclear, and limited" ["oscurissimi, e scarsi"] (Vallisneri 1700, p. 357), as Malpighi was then made to observe. Indeed, in chapter XX of Book 5 of Aristotle's *Historia animalium*, it is said that these lesser wasps nest in the walls and feed on spiders, even if – it is added – those of a larger size do the same thing (Vallisneri 1700, p. 357). As regards the former, Malpighi reports the following observations

> On the 20th day of June, I observed a swift Little Wasp going frequently in and out a hole made at some point by a nail in a wall in a little frequented Room […] On the 12 of July, I found it [the hole] closed up on the outside, and with the greatest of diligence daubed with a crusting of fine earth, or mud from the fields. I got the urge to open it up, and I can tell you quite frankly that the plug made of this earthen paste was a good finger in thickness. When I removed it, I saw a little cell with lots of Little Spiders inside and a fat, juicy, whitish-yellow worm, which was greedily devouring the same. When I took this away, there was another little cell further back, with a worm of the same kind, but a bit larger, shut up with other Tiny Little Spiders, and this little cell was in between two other contiguous ones, which also had their respective guests living in them, along with, as one might say, these still steaming corpses. Still further back, there were others without any flaws in them, but I

> was so clumsy when I broke [the structure] up that I made a fine mess of the whole thing, and I was unable to make […] any further observations […] I found yet another one amongst the splintered remains of a fallen in house, which I observed to be set up with a total of eleven little cells built in a most orderly manner behind a common passageway, so that almost all of them could get into it to leave [the nest] without going through the cells of the others, by gnawing through just one wall between the aforesaid passageway, and their cell. (Vallisneri 1700, p. 357)

This description could fit with the genus *Trypoxylon* (Crabronidae), which habitually builds its cells in a linear sequence inside empty plant stems or dead branches, but also in tunnels made in the wood by other insects or artificially produced holes. One would be led to think either that the walls of the room were made of wood or that the "nail" had been hammered into a supporting beam. The cells are constructed starting from the bottom of the hole, supplied with a goodly number of little spiders, an egg added and, finally, each unit is separated off from those to be subsequently built with an earth diaphragm. After the construction of the last cell, a plug of earth is applied to isolate the nest from what lies outside. The difference in size of the larvae may be due to the fact that the inner cells are made ready and provisioned first and thus the corresponding larvae would be older (and larger) than the ones nearer the outer part of the nest. Their "common passageway", on the other hand, remains unexplained.

However, the difference relating to size is interconnected with another two: the first is, again, a matter of size but, this time, relating to sex (males are smaller than females); the second is the difference between *wild* "Vespe Icneumoni" and their *domestic* counterparts. The latter, which the reader is reminded were of the type also described by Aldrovandi (Vallisneri 1700, p. 358), were observed

> […] in various places in the house, not only isolated and unfrequented ones, but continuously visited places, even under the smoky old Mantle of a Chimney, where the Kitchen fire was constantly burning. (Vallisneri 1700, p. 358)

This habit matches that of the genus *Sceliphron* (Sphecidae) as this can be confirmed from the description of the wasp which has its abdomen separated from the thorax by a "long pipe" (Vallisneri 1696, p. 358), and has a black head and thorax (which seems to indicate the species *Sceliphron spirifex*) (Pagliano and Negrisolo 2005, p. 82), from the number of cells in a nest (14 but, as would be observed further on, varying between 1 and 22) (Vallisneri 1700, pp. 358, 368), from the spiders contained in a cell (10–12) (Vallisneri 1696, p. 358), from the yellow and black of its legs, the greater length of the back legs and, finally, from its way of collecting mud from a puddle, and also due to its propensity for building its nests in warm places, like a chimney mantel, as well as on beams and under roofs or inside the walls (Vallisneri 1700, p. 359).

The "wild" variety is not so easy to identify. These also have a long waist, but are very different in colour and shape from the former (Vallisneri 1696, p. 359). Here is what Malpighi says with regard to *wild* "vespe icneumoni"

> Out walking on the 15th day of March […] raising my eyes, I saw, on top of a Great Branch of a dead Oak, about eight arm's lengths high, an earthen nest, southern facing, there exposed to every gust of wind. […] I shut it up jealously in a glass jar […] it had been made the previous year, and it had been all winter in the snow and ice and wind. On the 12th day of June out hatched a Wasp with a really long pipe in its belly, but with a colour and shape rather different from the aforesaid domestic variety […] On the 14th and 15th another two […] And these were all of the same size and very similar in shape. On the 17th, Bigger Ones started hatching out, and these looked stronger and bolder […] Counting the holes, there were only fourteen, although all the Wasps together numbered seventeen, three of them having pierced the dividing wall of their cells and come out of the little windows already made by their neighbours […] I supposed, and I swear by your Aristotle that I am making a good faith guess here, that the bigger ones were female and the smaller ones […] were male […] (Vallisneri 1700, pp. 359–360)

One has the definite impression that this is a description of a eumenine wasp (Vespidae: Eumeninae), due to the fact that, unlike in the "domestic wasps", "the pipe which divides off the belly bells out like a trumpet" (Vallisneri 1700, p. 360). The observation that the males, which are smaller in size, emerge from the nest before the females is interesting, as this is typical for these Hymenoptera. The earthen nest described would seem that of a *Eumenes*, perhaps *Eumenes unguiculatus* (Eumeninae) (see, for example, Grandi 1961, Fig. 23, p. 39). Although this is correct, the assertion that they probably feed on spiders (Vallisneri 1700, p. 358), like the "domestic" ones (which is more taken for granted than supported by specific observations) is not. Indeed, the wasps in question use Lepidoptera caterpillars. On the other hand, later in the work, Malpighi would say, with regard to other (wild) wasps, which build

> […] a roundish, earthen Nest looking rather like a breast, and like a walnut in size. Once I had detached it and opened it, I found it all empty, in other words, it had just one cell, and with just one solitary worm in it […] I observed that it ate caterpillars […] It took the Little Wasp until the 20th of June of the following year to hatch. For the record, this was similar to the females of wild "icneumoni" described, but a good bit smaller […] with the very same structure, and perhaps, or even actually, there is another one of the same species, which usually builds a rough-looking nest inside walls facing East or south, which also feeds its young on little caterpillars […] and walls up ten or twelve of them, half-alive, inside each little cell as food relished by the future Little Wasps. I even found two nests in the window of a country House. They were facing East, quite close one to another, and made of hard, white clay, rather pointed towards the top, and roughly rounded off, very much smaller than the aforedescribed. When I opened one up, I saw just one white worm similar to those mentioned in a round and shiny little cell, which was greedily devouring imprisoned geometer moth larvae a lot smaller than the aforementioned caterpillar […] (Vallisneri 1700, pp. 364–365)

These passages remove all doubt as to the identity of the wasps in question (and also that of the previous one), *wild* "vespe icneumoni" which, as it is explicitly stated, have the same structure, even if they are smaller. Again we are dealing with wasps of the genus *Eumenes*: the first of this second group of smaller wasps, probably *Eumenes pomiformis*, builds mud nests with a shape reminiscent of a wineskin, is made up of a single cell (unlike *Eumenes unguiculatus*, whose nest was multicellular) and is provisioned with caterpillars of microlepidoptera.

Malpighi also speaks of "Wild (Silvestri) Bees", which he had briefly mentioned before, calling them "Forest (Silvestri) Bees" (Vallisneri 1700, p. 358), about which he says

> […] even though it works its little honeycomb out of mire and the tiniest of stones, it does still perhaps still retain the nobility of the Bees, as it feeds its little foetuses, inasmuch as I have been able to observe, solely on sweet juices. I would place these amongst the ichneumon owing to the similarity of their earth nests which, from the outside, are hardly distinguishable […] as we can also find some built by the said Wasps with almost the very same material." (Vallisneri 1700, p. 361)

But, although the structure of the nests is similar to that of the "ichneumon" wasps, the same does not hold as regards the appearance of the insect, seeing that

> The Wasps, or rather the Bees which build these [nests] have really similar features to common Bees, possessing a very different form to that of the aforementioned ichneumon […] (Vallisneri 1700, p. 362)

Both the descriptions reported above, that of the nest and that of the insect, are consistent with the bee *Megachile*, probably *Megachile parietina* (= *Chalicodoma parietina*) (Megachilidae), which builds earth nests (Grandi 1961, p. 324). Regarding these bees, Malpighi reports the following observation regarding the chrysalis of one of these, which had been separated and placed in a twist of paper so that its development could be followed

> Looking at this chrysalis on the first day of July, I found on top of the same 4 little spherical, white eggs […] and looking carefully at the twist of paper I found it had been pierced right through by a beetle, which had industriously penetrated through a little split in the first outside wrapping, and this led me to understand even better, how easily the Gentlemen who Defend spontaneous generation can be led astray […] by not observing that ingenious […] Mothers have secretly laid their eggs, precisely as happened to the aforesaid unfortunate Chrysalis, or Nymph, from which, most purposefully kept and watched over, hatched four rather hairy little worms with ringed segments and, when it was time, the Little Worms changed into Nymphs, and the Nymphs into Beetles. This did not happen to the other Nymphs of the Bees shut up in their impenetrable nest, seeing that Bees flew out of there, and not Beetles. (Vallisneri 1700, p. 362)

In all probability, we are dealing with *Trichodes* (Coleoptera: Cleridae), a parasite of bees and wasps which attacks the larvae and probably also their food stocks (Grandi 1961, p. 321; Müller et al. 1997, p. 262). Irrespective of the naturalistic interest of this observation, it is also important in a much more general sense, to counterattack the opinions of those who, when faced with the phenomenon of the apparent generation of an insect different from the one it originated from, attributed it to a case of spontaneous generation.

There later appears a consideration made by Pliny regarding a mistake made by Aldrovandi, who had allegedly confused the nests of "domestic ichneumon wasps" (*Sceliphron*) with those of the "wild bees" (*Megachile*) once described by Aristotle, not having taken into account the different colouring of the larvae and having trusted in the mistaken description of a countryman (Vallisneri 1700, pp. 362–363).

Returning to the subject of wild bees (*Megachile parietina*), Malpighi observes that larvae of two sorts come out of the same nest – one which is bigger and black

in colour, the other smaller and more colourful – considered to be, respectively, females and males (Vallisneri 1700, p. 363). Indeed, the males of *Megachile parietina* are very different in colour from the females (Grandi 1961 p. 324; Müller et al. 1997, p. 268). As we have seen, Malpighi had already revealed, in connection with wild bees and, in particular, *Megachile*, that the bees, unlike the wasps, which build earthen nests and live "of Little Spiders and Small Caterpillars and Geometers moths larvae" ["di Ragnateli, di Bruchetti, di Geometri"] (Vallisneri 1700, p. 361), feed their own larvae, at least as far as he has been able to ascertain, on a vegetarian diet, consisting of "sweet juices" ["dolci sughi"]. This is an important point, to which Malpighi subsequently returns, with an interesting observation comparing the structure of the tongue in *Megachile* and that of the honey bee

> […] a long tongue, composed of, as one might say, five little shiny, sharp tongues, almost as if they had teeth, because of some short hairs, which made them look rough and coarse. The one in the middle was twice the length of the others […] very sharp-looking, and also a bit hairy […] The other four were made differently from the aforesaid […] This new discovery as to the tongues led me to the increasing suspicion that that they feed on juices, honey, dew and other like things, because they are very similar to those of ordinary Bees, and they seem much more suited to taking away and manoeuvring all sorts of liquors with their roughnesses, whether these consist of branchy, yielding particles or sweet, sticky ones. And, indeed, I have often seen them on flowers […] (Vallisneri 1700, p. 364)

As regards the vegetarian diet, Pliny asks for confirmation

> Are you sure that they feed their little ones only on juices, and not sometimes on tender Little Mosquitoes, maggots, tiny flies, small spiders or other such things? (Vallisneri 1700, p. 364)

Malpighi, in view of the lack of other observations to add to those already given, which do not exclude the possibility of a mixed diet, tries to support his hypothesis by putting forward some further information, presumably again regarding *Megachile parietina*, reported to him in one of Cestoni's letters

> […] in Livorno there are a huge number of earth Nests attached to the stones on the facades of the Houses […] which look like so many pieces of earth thrown haphazardly by a human hand, inside the little cells of which he [Cestoni] often found a little piece of brown honey intended to nourish the worms, which further supports my suspicion, if we suppose that these were built by Bees of the aforesaid race. (Vallisneri 1700, p. 364)

The persistence on this subject is certainly justified by the importance of the question brought up by Vallisneri. The difference between the diets the larvae are fed on is indeed fundamental when making a distinction, whether ecological or systematic, which separates the large taxon made up of Hymenoptera Aculeata (bees, stinging wasps, ants) into two halves.

Another important note is contained in a passage part of which has already been reported above

> […] and it walls up ten or twelve of them, half alive, inside each cell as food relished by the future Little Wasps. […] I found two nests […] much smaller than the aforesaid. On opening one, I saw just one white worm, curled up, similar to those mentioned, in a shiny rounded little cell, which was greedily devouring imprisoned geometer moth larvae […] and I saw that there were two still alive, brought there incredibly skilfully without killing

> them […] so that they would continuously provide fresh and tender food for the little one, and so they would not rot or dry up before they [the offspring] reached the required size. And this admirable providence I have also seen practised in some of the Nests of the wild ichneumon Wasps [the *Eumenes* before considered], and perhaps, also, in domestic ones, and in all those which live in the very little holes in the walls [*Trypoxylon*] […] in which almost all the Small Spiders, which they were going to leave as food for their little ones, were alive. So I saw that it was not always true, as Aristotle said, in the Book 5 Chap. 20 quoted, that the ichneumon wasps, *Phalangia perimunt, occisaque ferunt in parietinas, aut aliquid tale foramine pervium* [they kill spiders, and bring the killed ones in ruins or anything other similar place provided with holes] […] (Vallisneri 1700, pp. 364–365)

This observation, which is of primary importance, holds with reference to *Eumenes*, *Sceliphron*, *Trypoxylon*, in other words to all the observed predatory wasps, and consists in the fact that they do not kill their prey (as Aristotle maintained) "so that they would continuously provide fresh and tender food for the little one, and so they would not rot".

The wasps Aristotle is alluding to are, with all probability, Pompilidae, spider-hunting wasps which Vallisneri does not seem to have observed personally. However, he will report Bellonio's description of how the spider is stung and carried away by the wasp (Vallisneri 1700, p. 361).

Malpighi continues as follows

> When they are grown, the Worms make a white cocoon […] When I looked on the sixth day of June, I found two long Little Wasps had hatched, that is, one from each nest, which had emerged from the back part of the aforesaid, which was already open, by which it was attached and fixed tight to the Wall, having in the meanwhile avoided the bother of gnawing away at the front wall of the same. These are half smaller than the aforementioned ichneumons, but almost exactly the same shape. […] They have a big back and a broad chest, from which spring six legs, divided up into seven sections, the last of which are really long, and they have a long spine at the end of the third, as have the second legs too. The thorax is connected to the abdomen by a long, hard, black tube, shaped like a trumpet. The abdomen […] from the end of which is always unsheathed, and ready to strike, a very long but, as I think, innocent tricuspid sting, in the shape of a straight tail. It is almost as long as the whole of the abdomen, and it does not flatten and unsheathe it, as other Wasps do with their harmful, stinging needle. Indeed, it is adorned and hung on one side and the other with two very black and hairy threads, which are usually crooked and contorted like old, intertwined grapevines […] (Vallisneri 1696, p. 365)

Pliny replies

> These may well be the Little Wasps *ex Minuti &c.* mentioned (even if he was then talking more of Wasps), although this was believed in passing, by my Aristotle in the place you mentioned which, as he says, *nomine carent* [have no name] even though *nidos e luto parvos, aut ad sepulcra, aut ad parietinas configunt, atque in iis vermiculos pariunt* &c. [they build small nests from the mud and fix them either to sepulchres or ruins, and inside them they give birth small worms] as industriously as the larger and, if it does not seem overly bold to give a name to an insect which was not given to it by Aristotle, I would call them, at least so as to distinguish them from the other ones, *tailed domestic Ichneumons.* (Vallisneri 1700, p. 365)

The two "Little Wasps" that Vallisneri saw coming out of the back of the nest were ichneumonid parasitoids (in the currently accepted sense of this term) which had fed on the larvae of the host and its prey.

Following a discussion as to whether or not wasps which build earth nests could have wings covered with hard elytra, which seems to have been something maintained by Aristotle, as reported by Pliny, Malpighi is made to reply

> Pliny, to be quite frank with you, I really do not think that Aristotle then understood them to be Wasps, even if he was dealing with this issue, and men with a great amount of good sense, and most worthy besides, firmly believed this. And indeed, out of all those Wasps and Forest Bees that I have observed making all or a part of their mud nests, of which, besides those described, I still have many others to describe, I have never found any which have shell wings over their membranous wings […] I have often found strangers living in the aforesaid earthen nests, and false guests which have either managed to get inside, or have been laid inside by their wise and industrious Mothers, so that they can feed on this juiciest of sweet worms that there is inside[…] I found one of the domestic variety stuck tight under the arch of a public Portico, which […] contained a beautiful, live Beetle […] all of it a lovely cinnabar colour, and patterned, with wings dashingly edged with a bright purple […] (Vallisneri 1700, pp. 365–366)

Urged by Pliny to say whether he has seen other insects "dall'ali superiori di crosta ["with upper wings made out of shell"] in the wasps mud nests, Malpighi replies

> Just once, I saw a really odd one, but I imagined that it was (as it indeed turned out to be), an itinerant inhabitant of cells that were not its own. This one had a round Orange, smooth and shiny Head, and was shaped like the bare skull of a dog with a long muzzle […] This was definitely not a Wasp, as you might hear, but rather some sort of beetle of a kind of its own, quite dashing, and strangely shaped. (Vallisneri 1700, p. 366)

In the last two passages quoted, we have cases of beetles which are using or parasitizing the nests. The latter is probably a beetle belonging to the Ripiphoridae family (Coleoptera), whose genera are all parasites of Hymenoptera Aculeata. To be more precise, it could be *Macrosiagon*, a parasite of some species of predatory wasps, in particular *Sceliphron*, as could be those which build mud nests here considered (Batelka and Hohen 2007).

Malpighi's answer to Pliny's question as to how the wasps' parasites manage to get into their nests and as to whether there are others, apart from the beetles he has already described, is as follows

> […] it is probable that they sneaked in unseen when the Mother Wasp had not yet closed off the top of the cells. I have not only seen the aforesaid Insects, but also a certain sort of Fly, which lays Maggots, or worms that are infamous eaters of live meat […] and the Mother Wasps being absent, they get into the cells of the latter before they are closed and after having laid their unnoticeable little eggs on the tender little worms, go off. The Maggots, shortly after they have hatched, bore or drill holes into the worm's skin, and sucking out all its white blood […] and delicate little entrails grow greedily on the ruins, and on the carnage of others. Nor are they content to devour just one, but smelling the nearby prey they pierce the dividing walls with this sort of awl they have, which is really hard and black, attached to their mouths like a spout […] and they move from one to another, until they are swollen, and sated with those wretched little worms, they reach their destined size […] from one nest of wild ichneumon Wasps I found hatched […] four Flies, and just one Wasp. Looking at the nest I saw, in addition to the big hole that the Wasp had emerged from, a little hole also made freshly as could be seen from the detritus above a cell of its home. Opening this, and following the narrow path of the little one, and the unused tiny aperture, I found four cast offs, or empty shells of the Chrysalises of the aforesaid Flies, and two Chrysalises which were still full, along with some excrement and the remains of the devoured Worm

> […] in another wild nest closed in a box […] I found it pierced in three places, that is, with two big holes and one little one. The usual Little Wasps had come out of the big ones and six Flies of the sort described above out of the little one. Again following the trail left by the little hole, I found in the offended cell the shells of the Chrysalises of the six flies which had come out looking the same, with the same features and in the same number as those mentioned. There was also a lateral hole, which led into another cell, and in that one another hole, which led into yet another cell, both of them empty, and bereft of their legitimate owner, just leaving a dry heap of droppings, closed off with the usual webbing in one corner. This led me to suspect that, in this case, the first worm devoured was the one in the first cell, since there were no droppings of any sort there and that, once they had finished, they had got in to the other cells to eat up the others, which they had found fully grown and ready to spin their cocoons, and to change into Nymphs (since they had prepared the cell, and neatly collected the faeces), then they went back into the first one, and there they changed into the usual chrysalises. And if you are itching to know what these bold and ingeniously insolent Flies look like, I can tell you that they are very similar to those which fly and buzz about our houses every day […] although they are a bit more bristly, and a little smaller, rather more ash-grey in colour, more marbled, and they are edged with black and have a silvery head […] from other earth nests, particularly those made by domestic ichneumon, sometimes twenty-five or thirty Little Flies of the same kind as the Small Worm-eating carnivores, which also came out of eggs laid by their Mothers inside the little cell in question before the Wasp which had built the cell had closed it off, so that they might eat up the worm which Owned that cell. (Vallisneri 1700, p. 367)

This last quote is a masterly discourse on behavioural ecology regarding Diptera which parasitize *Eumenes* and *Sceliphron*. In the case of the second wasp ("domestic ichneumon") the parasite is very probably *Pachyophthalmus signatus* (= *Amobia signata*) (Diptera: Sarcophagidae), as pointed out by Tremblay and Masutti (2005, p. 37), which first feeds off the egg and then the prey stored up by the wasp (Grandi 1961, pp. 160–161).

Malpighi also reports some observations he made about bees which reutilize the abandoned nests of *Sceliphron*

> […] a certain nest of *dashing Bee*, *and tiny* and ingenious inhabitant of holes in the wall, and also of old, empty nests of domestic ichneumon Wasps […] dashing little Bees, the little ones mentioned […] which live not only in holes in the wall, but are also innocent guests in the nests of domestic ichneumon wasps found Empty. (Vallisneri 1700, p. 369)

These are probably bees of the *Osmia* genus (Megachilidae), which may make their nests in the abandoned nests of other Hymenoptera, in particular *Sceliphron* (Cane et al. 2007).

Instead, another of Malpighi's observations can easily be connected with the genus *Trypoxylon* considered above

> […] a sort of Wasp which, finding a Bramble stem hacked off, immediately dug into the yielding, spongy tissue within and, inside that long, dug-out gallery, it laid its eggs separately, neatly spaced out, and together with the eggs, Little Spiders it had caught, and then closed off the space, making a hard earth plug between one egg and another, so that each of the little worms born should have its own little cell and its own store of food. And these old, empty Brambles are […] hidden nests which are very well suited to various Insects which are believed by certain *idle and credulous Putrefactionists* to arise of themselves […] (Vallisneri 1700, p. 369)

In a situation such as the one under consideration, in other words, insects which, in the following season, had apparently sprung from a dead branch, only an analysis as thorough as that provided by Vallisneri could effectively discredit the theory of a spontaneous generation from rotting vegetable matter. This was a further example of the need for a naturalistic analysis which was sufficiently detailed to provide help for the broader "philosophical" questions implicit in the theory of generation.

If we move on from *La Galleria di Minerva* to consider the *Quaderni di osservazioni*, which in part have recently been transcribed, we find reported an important observation, made by Mattacodi, regarding another parasite of the "domestic Ichneumon". It is presumably the wasp *Acroricnus seductor* (Ichneumonidae) found in a nest of *Sceliphron* (Polidori et al. 2011)

> In one smaller little house [cell] there was a little white worm only half the size of the others. However, it was divided up into twelve rings, with a strip of transparent humour not only in its body, but also in its back […] Its little cocoon was all covered with white spittle." (Vallisneri 2004, p. 66: 27 September 1694)

The colour of the "worm", the dorsal line and the appearance of the cocoon indicate that it was an *Acroricnus* larva. In addition to this, Mattacodi reports the presence, in the nest, of the larva of a parasitic (or inquiline) beetle

> This was of a reddish colour and all hairy, with a little flattened, but very hard, head, and with six feet in the three foremost rings, using which it dragged behind it the whole bulk of the rest of its body, which was considerably bigger and longer than the forepart. (Vallisneri 2004, p. 66–67: 27 September 1694)

Judging by the description, this could have been a Ripiphoridae larva.

2.3 Diacinto Cestoni's Letter

Even if the work of this naturalist was well appreciated during his life time (Generali 2004, p. 107), after his death he was relegated to a secondary role or quite ignored, up to recent times. The reason is probably that he never published anything and preferred to confine his observations to letters that he sent mainly to Vallisneri, but also to other important researchers of his time. Only in the last years the importance of his observations has been acknowledged (see, for example, Tremblay and Masutti 2005, p. 37; Generali 2004).

The letter we are concerned here is generally recognised as being of great importance, together with its great value as regards our present interests. This has been particularly emphasized by Tremblay and Masutti (2005, p. 37) who, quite rightly, point out that the descriptions and explanations of behavioural ecology provided by Cestoni are equally as good, if not sometimes even better, than those given by Vallisneri himself. This makes Cestoni's work relevant in the debated importance of building a framework unifying detailed behavioural data and conservation sciences (Berger-Tal et al. 2011). The letter, dated 1692, was sent to Vallisneri in 1698, with

the declared intent of confirming "various findings of the aforesaid Gentleman [Vallisneri] as regards the curious Origin of many Insects, described in his First and Second Dialogue" (Vallisneri 1726a, p. 89) ["vari ritrovamenti del suddetto Signore intorno la curiosa Origine di molti Insetti, descritti nel suo Primo, e Secondo Dialogo"], and subsequently published by Vallisneri himself (since the letter was sent before the publication of Vallisneri's Second Dialogue, one must assume that Cestoni had already read it, in some manuscript or preliminary version). We feel it would be appropriate to examine it in detail, as most of it is devoted to parasitoid wasps (and their hosts). Basically, this is a specialized scientific monograph, as it is exclusively dedicated to "little creatures" found on cabbage leaves, which were examined with a thoroughness and precision which could serve as a model for a modern research study.

The first observation of Cestoni relevant to the present paper regards the "fleas" that can be found on cabbages, but also on many other plants and flowers, which Aldrovandi had also mentioned, but only in passing (Vallisneri 1726a, pp. 91–92)

> These horrid little animals are idle, stupid, and very slow to react, and on any plant they live on, they all look the same, or very similar. They have a bold, round little body, very like that of Spiders, six legs, two antennae, or very long horns, two black eyes, a long, thin, sharp rostrum, with which they very often pierce the leaves, to graze on the moist, delicate and tender substances of the plants […] In short, they take on the colour or of the juices they swallow, having, amongst other things, an extremely thin skin, and being very fragile creatures […] never having personally seen any one of them up to now about the Act of Mating […] However this much I have observed that, when they are fully grown, all of them give birth, and produce their little ones alive […] when they have become as big as they will ever get, these also start to reproduce, and to produce their young alive in the same shape as the others […] Amongst the little creatures described, there are many which become winged, so I could not help but wonder whether these were of another race, notwithstanding the fact that before they grow wings, not much difference can be seen between them […] (Vallisneri 1726a, pp. 92–93)

The observation regarding the parthenogenetic viviparous females – which, therefore, do not have to be fertilized to produce their offspring – is of particular importance. This significant biological discovery (subsequently and unfairly attributed to Charles Bonnet), together with the shape of the animals, their universal distribution, the presence of winged generations which alternate with unwinged ones, allows us to identify the "fleas", without a shadow of a doubt, as aphids (Grandi 1984, Vol. I, pp. 827–830). It is thus rather peculiar that, having such precise information at their disposal, Tremblay and Masutti (2005, p. 37) came to the conclusion that the insect was *Aleyrodes proletella*, another homopteran belonging to a different family (Aleyrodidae instead of Aphididae), perhaps deceived by the name "cabbage white fly" as it is commonly called. But the described species is actually only one of the various insects found on cabbage leaves, as Cestoni clearly indicates (Vallisneri 1726a, pp. 89–105). On the other hand, Cestoni does not say he has ever seen the eggs of these *fleas* (Vallisneri 1726a, p. 95), which, are in fact easy to spot in the case of *Aleyrodes*, as they often form a distinctive pattern on the underside of the leaf, alerting the viewer to the presence of the insect in question (Grandi 1984, Vol. I, Fig. 666 p. 824).

Strange things happen to these "fleas"

> A few days later, when the aforesaid Insects have given birth, both the winged ones and those without wings, they can mostly be seen to be quite unmoving, and attached with all six feet to the same leaves, and with their rostrum forever stuck in the same, as if, just the same, they were continuing to suck; but once I had had a good look at them, I realised that they were doing anything but suck. They had actually died like that, even if their bodies remained well-preserved, big, fat, round and swollen, as if they were alive, the only difference being that they were starting to turn yellow. I started observing a number of these little creatures; whereupon I found a few of them whose heads and thoraxes had in fact dried up, and the lower part of the abdomen too. However, aside from this, when I squeezed them, I saw and felt that there was still a tiny portion of fresh matter inside. I found yet others that were not only shrivelled and all dried up, but mostly they were completely empty, so that all that remained of them was very simply the outer skin, the husk, or shell, or whatever one prefers to call it, in which a tiny hole could be seen. This observation immediately made me wonder whether there were other animals which went around devouring the insides of these Fleas; whereupon, in order to gain more insight into the matter, I took a large number of these newly-dead animals and, having separated the winged ones from the unwinged sort, I put them separately into two glass jars which I immediately covered very carefully, then, not many days later, looking into these jars again, I saw (to my great amazement), that a lot of Little black Flies had come out of these Fleas. They were very lively and slim, and they walked and flew about inside those jars incredibly quickly. And, at the same time, I observed that the Fleas had been left with just their skins, completely emptied out inside, just as I had seen happening in the other cases I described above. Being unable to imagine the reason, how such a bizarre metamorphosis could come about, and ever more desirous of discovering its cause […] and after a lot of assiduous research, I was lucky enough to discover how and whereby and why the mentioned transformation should necessarily take place. (Vallisneri 1726a, pp. 94–96)

In the end, the parasite responsible for the "transformation" observed was found and its interesting behaviour described. This was one of the first descriptions made in Europe of a true case of oviposition by a parasitoid (van Lenteren and Godfray 2005, p. 14), something Vallisneri supposed had happened, but never seen for himself.

> […] near these Fleas I saw certain Little Flies buzzing about which, after having walked and flown quite a lot around the Fleas, gradually moved closer to the largest ones, as if identifying those fit for their purpose […] I armed my eye with a really good Lens, following one of these […] I observed, that that particular Little Fly had got so close to one of the Fleas that it was almost touching it with its head. Close as it was in this way, I saw that, once it had firmly planted its feet, it raised its wings, as if it wanted to fly and, at the same time, holding its wings raised like that, it plunged its lower abdomen under its chest. As its abdomen was a little longer than the rest of its body, it thus stuck out further than its head; folded over, as its body was in that position, it bent itself so far down, and made so much effort, that it put the very end of its body under the belly of the Flea and, keeping it like that for a very short time, it went away, and I saw that it was going about the very same thing around the other ones. To clarify for myself, and see what this Little Fly might be doing by putting its abdomen under the body of that other Insect, I decided then and there to turn it over, and […] I found that that Little Fly had very kindly laid an egg under the belly of the other insect, and was doing the same thing with the others. Having this piece of good news, it was not hard for me to find out why Little Flies flew out of those Fleas which looked dead, in view of the fact that lots of little maggots hatch out of these eggs and, as soon as they hatch, the pierce the bellies of the Fleas underneath which they have been laid, and entering their bodies, they use them both as food and as a room: and when they are well enough nourished

> and grown, they make a chrysalis inside the same Flea, then in less than a Month the Little Flies coming flying out, one from each Flea. (Vallisneri 1726a, pp. 96–97)

The parasitoid in question is certainly a genus of the subfamily Aphidiinae, perhaps of the species *Aphidius*, which is characterized by this particular way of laying eggs on the underside of the aphid, bending its abdomen forward, which is made to pass under the thorax and the legs of the wasp (Grandi 1984, Vol. II, Fig. 978 p. 1004) in the way described by Cestoni. When the egg hatches, the larva of the parasite enters the host and eats it up from the inside, turning it into an empty shell, or *mummy*, as it is called in entomological jargon. In this case too, the mistake made by Tremblay e Masutti (2005, p. 37) is hard to understand. These authors hold that the parasitoid was an Aphelininae (Aphelinidae), an identification which might be appropriate, as we have said, in the case of the parasite of the "gorgoglioncino" reported by Mattacodi and discussed above.

But the aphids living on cabbage leaves studied by Cestoni have other two persecutors even if, this time, they are not parasitoid wasps. The first of these is probably a ladybird (Vallisneri 1726a, p. 97), a carnivorous beetle which, both as a larva (as observed by Cestoni) and as an adult insect, is a hearty consumer of aphids. The second (Vallisneri 1726a, p. 97) is the larva of a Syrphidae (Diptera) (hoverfly), as its green colour striped with white indicates (Rotheray 1993) and the fact that the adult of this fly

> […] which, when it flies, you very often see it hold itself in the air, exactly in the same way that larks do when they hang in the air singing. Nor should you, most Honourable Sir, think that they do this by chance; it is done on purpose, in order to observe, and see where the Insects are, and when it sees some, it lands on that Plant, and lays one or two eggs, then it goes off to fly somewhere else, since it never lays more than two eggs on the same leaf, at least as far as my observations go. (Vallisneri 1726a, p. 98)

Returning now to parasitoid wasps, Cestoni talks of those which parasitize the caterpillars of "lovely big, white cabbage butterflies (= *Pieris brassicae*) (Vallisneri 1726a, p. 98). Cestoni describes some aspects of the biology of these butterflies, such as the fact that they lay their

> […] eggs under the leaves of the aforesaid Cabbages, and arranging them with a wonderful neatness, they place about fifty, or sometimes seventy in a patch, one next to the other, tidily in one place, about the size of a nail on the hand. I said under the leaf, because very rarely do they lay them on the upper side, but habitually place them on that part of the leaf which faces the ground, so that they remain covered and are not hurt by the rays of the Sun. These eggs look yellow on the outside, and in the space of two or three days a lot of little worms hatch out of them, which immediately start eating the leaf upon which they were laid […] (Vallisneri 1726a, pp. 98–99)

When they have finished eating, the caterpillars turn into chrysalises which, after ten days, will produce new butterflies. What is more interesting is the fact that, in some cases, instead of a butterfly, "a number of Little Flies" (Vallisneri 1726a, p. 99) comes out. This was a fact that had already been reported, even if more summarily, by Vallisneri in the *Primo Dialogo*, as we have seen above, but here the discussion becomes more precise and detailed

> The bizarre birth of the aforesaid Little Flies, noticed also by yourself, Honourable Sir, encouraged me to make a move to determine their origin, as I had then fully recognised the existence [of said phenomenon] […] To fully understand the metamorphosis mentioned, or the development of so many minute Insects, one needs to know that, during the time that the above-mentioned caterpillars live and feed on the leaves of Cabbages, certain Little Black flies fly around them. They are bigger than those of wine (whose origin You discovered, Sir, in your much praised Dialoghi), very slow when they move forward, but very quick-flying, the females of which land on the most unfortunate caterpillars, and lay a number of tiny eggs on them, almost invisible to the naked eye, out of which in less than two days hatch these really tiny maggots, and then these almost invisible worms, just hatched, like the Small Worms of Mange, poke themselves in under the skin, and they get so far in that, little by little, they eat them up inside. However, these caterpillars keep on eating and growing, just the same. Nonetheless, one can clearly tell which ones are affected by these little maggots, because they start to turn yellow […] so they no longer think about making a chrysalis, but all of a sudden, when the other healthy and lucky caterpillars set about making their own Chrysalises, the infected ones burst down one side, and all these little maggots, which have fed on the caterpillar's substance, come out of the crevice in each one. These unrestrained little maggots, which have come out of the bodies of the aforesaid caterpillars (which of course then die and dry up) get silk out of their mouths, with which they bundle themselves up, and tangle themselves so much in it that they end up looking like a heap of little cocoons covered with a yellowish silk. Then, at the end of about twelve days, lots of little flies jump out. And this is not a misfortune which happens just to Cabbage caterpillars, but likewise to various other kinds of caterpillars and worms, as I and You yourself, Sir, have observed on a number of occasions, as illustrated in your above-praised Dialogue. (Vallisneri 1726a, pp. 99–100)

Here we are probably dealing with *Cotesia* (= *Apanteles*) *glomerata* (Braconidae: Microgastrinae), which attacks Lepidoptera Pieridae, laying 50 eggs or more (Grandi 1984, Vol. II, pp. 995–996). Cestoni thus clarifies once and for all the aforesaid mistakes made by Redi and Aldrovandi, who had taken the cocoons, made by the larvae of the parasitoid, for "eggs" laid by the caterpillar which could generate animals different from themselves (Vallisneri 1696, p. 313). Vallisneri had arrived at the same conclusion (Vallisneri 1696, p. 312), seemingly without ever having witnessed the act of oviposition, as Cestoni had done (Tremblay and Masutti 2005, p. 36).

Cestoni also observed a female parasitoid wasp smaller than the one previously described laying eggs on the same larvae or those of other Lepidoptera. The size of the parasitoid and the identity of the host indicate that they were, in all probability, *Pteromalus puparum* (Pteromalidae: Pteromalinae), which, as the name tells us, emerges from the chrysalis of *Pieris brassicae*, and also of many other butterflies. Vallisneri had given some cases which, on the surface, appeared to be similar, since they should actually have been connected with "eggs […] laid upon the Chrysalis" ["uova […] depositate sopra della Crisalide"] (Vallisneri 1726a, p. 101), as Cestoni observes after carefully reading the already quoted passage of the *Primo Dialogo* in which Malpighi asserts that

> Many Flies and Large Flies come out of cocoons, or Pupae made by worms […] others out of the their Chrysalises […] They will never see Little Flies or Large Flies or Flies emerge from the Chrysalises, if they protect them well enough, I mean the cocoons and such like […] (Vallisneri 1696, p. 312)

Instead, the parasitism observed by Cestoni is different and more sophisticated in nature

> Other Little black Flies, smaller than the ones I have described [*Apanteles glomeratus*], less than half the size, land just the same on top of the caterpillars, and offload their eggs on them, and these are so small that they cannot be seen by the eye, unless one has a superb Lens. After some time, longer than it takes for the aforesaid little flies, likewise, little maggots hatch out of these eggs, and they also get into the caterpillar but, as they are quite a bit later in feeding, the caterpillar has time to grow and make its Chrysalis (and this indeed happens) and, during this time, those nasty little maggots continue to feed in the same way on the substance of the caterpillar inside its chrysalis, without emitting faeces. When they have finished feeding, they do not emerge from the caterpillar in order to make a Chrysalis, but they do this inside the caterpillar itself, and they stay there over a Month, and then the Little Flies come out. There are really so, so many that one cannot even imagine that they could have been inside that Chrysalis, wherein the Little Flies make a tiny hole, or perhaps the first Little Fly does so, because it wants to get outside. Indeed, after that, all of them pour out of that same hole. (Vallisneri 1726a, pp. 100–101)

The fact that the egg of the parasite is not laid directly on the pupa or chrysalis, as Vallisneri seems to believe in his *Dialoghi* (something which actually occurs with other parasitoids), is an example of *delayed* parasitism, which makes more difficult to identify the parasite living in the larva of the host until it turns into the chrysalis which will, eventually, be eaten up. Thus the parasite does not emerge from the larva upon which it has been observed laying its eggs, but from the chrysalis into which the larva has turned, a situation which could have been exploited to support spontaneous generation without Cestoni's shrewd analysis.

The last observation regards the "Little white Butterflies" commonly found on the leaves of cabbages (the homopteran *Aleyrodes proletella* (=*brassicae*), already quoted), for which Cestoni gives a painstaking biological description noting, for example, how they mate and the characteristic distribution pattern of the eggs, mentioned above, and observing that they feed "on that juice, which bathes the outer skin of the leaves" (Vallisneri 1726a, pp. 103–104) ["di quel sugo, che viene a irrorare l'esterna buccia delle foglie"] apparently without harming the plant. Let us follow Cestoni's description:

> These Little Butterflies are generated in precisely the same way as happens with most animals, in other words, by means of the male and female […] which, carrying on with each other, mate, and when the females are pregnant […] they lay their eggs, which they generally arrange in a semicircle in batches of ten, twelve, fourteen, and sometimes sixteen […] from each of these emerges a little white animal with six legs and a bit of fuzz on its back […] I have decided to call them *Little Sheep* from now on […] and these start walking […] when they have got to where they have to stay […] they arrange themselves a little apart from one another, so that, as they grow, they won't touch each other; thus, seen under the Microscope, they look like a lot of little white sheep standing still in a little green field. After that, remaining still and attached, they grow […] It may seem to You, Honourable Sir, to be an error of judgement, my having given the name of little sheep to the aforementioned creatures, but thinking through what happens to these poor Insects […] it will not seem so unreasonable […] just as sheep are subject to being devoured by Wolves, these […] also have their *Wolves*, which hunt them down. The latter are a breed of Little black Flies, which […] live on nothing else but the aforesaid dear little sheep … A copious quantity of the aforementioned Little black Flies constantly buzz around the said little sheep, and some of

> them linger around the most tender ones, and slowly but surely, they suck out all their substance, so that in the end they just leave them with their outer skin. Others land on the largest ones, that is, those which have grown to their fullest size or nearly so, and sit on them for a long time; whereupon, having paid particular attention to this, and to what they were doing there, I saw that the Little wolf Flies, after having pierced the back of the little white sheep they were sitting on, proceeded nonchalantly to lay an egg in that hole, from which shortly afterwards I observed that a nasty little maggot had hatched, which started to eat up its poor little sheep […] It is an easy thing to recognise when the dear little sheep have been unfortunate, and the Little Wolf Flies have laid eggs on them, seeing that they begin to turn from their usual white, to a livid hue, and to go past the time when they would normally emerge as winged insects, which usually takes no longer than twenty days when they have not been ruined by the Wolves […] when these little maggots have finished feeding, they set about making their Chrysalises, and for their ends, they use the skin of the little white sheep themselves, which they have eaten up, inside which they can be clearly seen to be enveloped and turned into chrysalises. And there they remain for about twenty days or more, before they come out of that, than the aforesaid little butterflies. Then, at the end of that time, tearing that skin, healthy Little winged Flies come out and fly away, to start doing the same thing over again with the other little white sheep, thus continuing to reproduce at the cost of the innards and the flesh of the unfortunates, while the lovely little butterflies feed and multiply under the leaves of the Cabbages without doing any harm to them whatsoever. (Vallisneri 1726a, pp. 102–105)

Cestoni also provides a drawing of this parasite, and its host. We are dealing with *Encarsia* (Aphelinidae: Coccophaginae) which uses predominantly the more fully-grown stages of its host (the "largest ones" [le "più grosse"]) for the purposes of oviposition (Liu and Stansly 1996).

In Table 2.1 is given a summary of the species considered in the previous analysis.

2.4 Gall Wasps and Other Gall Insects

The discussion regarding the origin of galls will not be reported in the present work (see, e.g., Vallisneri 2004, p. 134 and note 653). In Table 2.2, we will confine ourselves to giving a synoptic list of what we were able to find out as regards the identity of the galls and the insects connected with them. The latter can be divided in species which form the galls on the plants, i.e. Hymenoptera (Cynipidae and Symphyta: Tenthredinidae); Diptera (Cecidomyiidae); Hemiptera (Pemphigidae), and species which are parasitoid of the gall-forming species (Pteromalidae, Torymidae, Eurytomidae, Ichneumonidae), and inquilines (Cynipidae and Coleoptera Curculionidae). Due the difficulty to identify species according to the original descriptions (see Discussion), the determination of inquilines and parasitoids was often made very tentatively.

Vallisneri explicitly points out the large number of species that can be found in just one gall

> And I did not just see beetles ["cantarelle"] in the said little swellings [galls on willows produced by *Pontania*], but the maggots of various flies, particularly carnivorous ones, all

Table 2.1 Generic or specific identifications

Genus/species	Family	Observed by	Reference and pages*	Host
Hymenopetera: Parasitica				
Agriotypus	Ichneumonidae	Vallisneri	1, p. 312	Trichoptera
Aphidius	Braconidae	Cestoni	2, pp. 96–97	Aphids (Homoptera)
Apanteles glomeratus	Braconidae	Cestoni	2, pp. 99–100	Lepidoptera larvae (Pieridae)
Trichogramma/*Teleonomus*	Trichogrammatidae/Platygastridae	Vallisneri	1, p. 312	Lepidoptera larvae
Encarsia	Aphelinidae	Cestoni	2, pp. 102–105	*Aleyrodes proletella*
Pteromalus puparum	Pteromalidae	Cestoni	2, pp. 100–101	Lepidoptera larvae (*Pieris*?)
Acroricnus	Ichneumonidae	Mattacodi	3, p. 66	*Sceliphron*
Hymenopetera: Aculeata				
Trypoxylon	Crabronidae	Vallisneri	1, pp. 357, 369	spiders
Sceliphron (*spirifex*?)	Sphecidae	Vallisneri	1, p. 359	spiders
Eumenes sp.	Vespidae	Vallisneri	1, pp. 359–360	
Eumenes pomiformis	Vespidae	Vallisneri	1, p. 364	Lepidoptera larvae
Megachile (*parietina*?)	Megachilidae	Vallisneri/Cestoni	1, p. 362	plants
Osmia	Megachilidae	Vallisneri	1, p. 369	plants
Diptera				
Pachyophthalmus signatus (=*Amobia signata*)	Sarcophagidae	Vallisneri	1, p. 367	*Sceliphron*
Coleoptera				
Trichodes	Cleridae	Vallisneri	1, p. 362	*Megachile*
Macrosiagon	Ripiphoridae	Vallisneri	1, p. 366	*Sceliphron*

*1: Vallisneri, 1696; 2: Vallisneri, 1726a; 3: Vallisneri, 2004

Table 2.2 Gall–inducing insects, parasitoids and inquilines (the families, when not otherwise indicated, are into Hymenoptera)

Plant and location	Vallisneri's reference	Number of species	Gall (G), Larva (L), Adult (A)	Modern reference	Notes
Quaderno I[a]					
Oak (underleaf)	[99r]: p. 120[b]		(G) Cynipidae: *Neuroterus tricolor* (?)[e]	1 (p. 59 and Fig. X.22 p. 59)	Inducer (reported by Mattacodi)
	[101r]: p. 123		(G) Cynipidae: *Neuroterus quercusbaccarum* (?)	1 (p. 59 and Fig. X.20 p. 59; Pl. 1.7b)	Inducer
	[107v]: pp. 132–133		(G) Cynipidae: *Neuroterus tricolor* (?)	1 (p. 59 and Fig. X.22 p. 59)	Inducer
Oak[d] (leaf buds?) marble gall?	[124r–126v]: pp. 155–158	2	(G+L) Cynipidae: *Andricus kollari*?	1 (p. 62, Fig. 160 p. 35)	Inducer
			(G+L) Cynipidae: *Synergus umbraculus*?		Inquiline
			+ aphids and Lepidoptera larvae		
Oak (inflorescence?)	[173v]: pp. 210–211	1	(G + A) Cynipidae: *Andricus* (*fecundator*? *solitarius*?)		Inducer
Rose[d] (bedeguar = cynorrhodon)	[113r–115r]: pp. 141–144	2	(G+L) Curculionidae (Coleoptera): *Curculio* (=*Balanius*) *villosus*? or *Anthonomus rubi*?	2, 6	Probable transient inquiline
			(G+L) Cynipidae: *Diplolepis rosae*		Inducer
			(G+L) *Periclistus brandtii*?		Inquiline

(continued)

Table 2.2 (continued)

Plant and location	Vallisneri's reference	Number of species	Gall (G), Larva (L), Adult (A)	Modern reference	Notes
Quaderno II[c]					
Oak[d] (leaf shoots?) marble gall ?	[72v–77r]: pp. 60–64	>=9	(A) Pteromalidae, Torymidae: *Torymus* sp.?, *Torymus nitens*?, *Megastigmus*?,	1 (p. 87); 3	Parasitoids of *Andricus kollari*
			(A) Eurytomidae: *Eurytoma brunniventris*?		Parasitoid of *Synergus* (inquiline)
Rose[d] (bedeguar = cynorrhodon)	[78r–81v]: pp. 64–66	5	(A) Ichneumonidae?: *Orthopelma mediator*?	2 (pp. 220, 223); 1 (p. 85)	Parasitoids of *Diplolepis*
			(A) Torymidae?: *Torymus bedeguaris*?, *Glyphomerus stigma*?		
			(A) Eurytomidae?: *Eurytoma rosae*?		Parasitoid
			(A) Curculionidae (Coleoptera): *Curculio (=Balanius) villosus*? *Anthonomus rubi*?		Inquiline
Oak			(A) Microlepidoptera	1 (p. 39)	
Quaderno III[i]					
Oak (leaf shoots?) oak apple ?	[113r]		(G + L) Cynipidae: *Biorhiza pallida*?	1 (p. 62 and Fig. X.34 p. 62)	Inducer
Oak (underleaf, close to the central vein) oyster gall	[15r]	1	(G + L) Cynipidae: *Andricus anthracina*?	1 (p. 58 and Fig. X.14 p. 58)	Inducer
Idea nuova d'una Division generale degl'Insetti[f]					
Beech (leaves)	p. 49		(G) Cecidomyiidae (Diptera): *Mikiola fagi*	4	Inducer
Poplar	p. 51		(G) Pemphigidae (Hemiptera): *Pemphigus spirothecae*?	1 (p. 53)	Inducer

La galleria di Minerva[g]					
Willow (leaves)	pp. 316–317	1 + 7	(G + L + A) Tenthredinidae: *Pontania* (*Euura*?) + 7 connected (inquilines, parasitoids)	5; 1 (p. 73)	Vallisneri notes that the fully grown larvae of *Pontania* leave the gall and climb down to earth, where they chrysalize (pupate)
			Curculionidae (Coleoptera)		
(Mattacodi) Nuova giunta di osservazioni, e di esperienze…[h]					
Willow (leaves)	pp. 65–66		(G + L + A) Tenthredinidae	1 (p. 73)	
			Microlepidoptera: *Pontania* (*Euura*?)		
Elm poplar	p. 66		(G + L) (parasites and inducer)		
Rose	p. 66		(G) (“cappelluta”) + (A) Cynipidae: *Diplolepis*	1 (p. 69)	Inducer
Rose	p. 66		(G) (“non cappelluta”) + (A) Cynipidae: *Diplolepis*	1 (p. 69)	Inducer

[a]Vallisneri (2004); [b]Numbers between [ ] refer to the original manuscript, the other to the pages of the printed transcript; [c]Vallisneri (2007); [d]Galls which were first opened (to observe the larvae), then re-closed to await the emergence of the adults; [e](?) means that the both genus and species are uncertain; ? after the species name means that only species is uncertain; [f]Vallisneri (1713); [g]Vallisneri (1696); [h]Vallisneri (1726); [i]Vallisneri (unpublished manuscript); 1 = Redfern and Askew (1992); 2 = László and Tothmeresz (2011); 3 = INTERNET reference at entry “oak marble gall wasp *Andricus kollari*”; 4 = INTERNET reference at the entry “ *Mikiola fagi*”; 5 = Nyman et al. (2000); 6 = INTERNET reference at entry “*Diplolepis rosae bedeguar*”

> of them bastards, foreigners and uninvited guests. I also observed the same thing in the scaly ends of willow shoots throwing out narrow leaves. I found inside, apart from the central worm or fly, which is black with long antennae and a really long sting, all sorts of other little flies, and I counted seven different kinds one day […] And not only did the said Little Flies which there ought to have been come out, but all these others, of Father unknown, and wild. (Vallisneri 1696, pp. 316–317)

The finding of Vallisneri regarding the large number of insects associated to a single gall is in fact true. For example, Grandi (1984, Vol. II, note 1 p. 1013) reports that Fahringer (1924) had observed 101 species of insects associated with the galls of *Andricus kollari*, which is probably fewer than the number found in more recent studies.

However, it was Mattacodi who identified, more clearly than Vallisneri ever did, the presence of parasitoids in the gall, which live at the expense of the inducer insect

> So in the blisters formed on elms, and in any sort of goitres found on poplars, the worms of flies can often be found, which kill the little flies that are born there by sucking at them, and they live in this womb they have appropriated for a long time, and yet those worms did not cause that goitre […] (Vallisneri 1726, p. 66)

The galls were therefore not caused "by a variety of Insects" but were induced

> […] solely by the aforesaid little caterpillar, and the little worms then got inside it in order to eat up the caterpillar, and gain any other nourishment they could find. (Vallisneri 1726, pp. 65)

Mattacodi therefore holds that it were the larvae of the parasitoid which somehow entered the host gall, as happened when eggs (and chrysalises) were parasitized, an opinion with which Vallisneri himself agreed as we have seen.

Another interesting observation regards the life cycle of *Pontania* (Tenthredinidae) (Table 2.2), whose larvae, once they are fully grown, come out from the gall (confirmed by Mattacodi (Vallisneri 1726, pp. 65)) and go down to earth to pupate (see, for example, Kopelke 1998)

> Having carried out some necessary duties, I found that the true caterpillar [the inducer of the gall, not one of the many others connected] of the Willow in question, when it is fully grown, emerges from the Swelling [the gall], and comes down to earth, where it buries itself, and hides itself up, there making its little cocoon […] to protect itself against the winter cold […] (Vallisneri 1696, p. 316)

Mattacodi also reports that the inquilines of a gall occupy positions further away from the centre than the larvae of the inducer

> […] up until now, I have always observed the same fly in the middle of the gall, and it nearly always comes out in the Autumn [to pupate, see above], whereas the other inhabitants place themselves between the middle of it and the perimeter, having got into the fully developed gall, where they even later complete their metamorphosis, and they cannot leave the gall before the next Summer. (Vallisneri 1726, p. 66)

In confirmation of the above Vallisneri would write, again as regards oak galls (Quaderno I: [124r-126v])

> I have observed many galls, which I collected until September last year, and I found, on the 8th January 1695, many completely empty, many without their original worm, but with the

> original hole where the worm emerged full of flies and ants and other little insects, with the mouth of the hole closed off with earth. Other galls, although they did not contain the original central worm in it, nonetheless had lots of tiny little worms around the centre, and in one I counted as many as eight. (Vallisneri 2004, p. 155)

One is dealing with galls of *Andricus kollari* (Cynipidae) (Table 2.2), in which the cell of the larva of the inducer insect lies at the heart, whereas the smaller ones, very presumably of the inquiline *Synergus umbraculus* (Cynipidae) (Table 2.2), establish themselves in the peripheral area around it (Redfern and Askew 1992, p. 35, Fig. 160; Pénzes et al. 2012, p. 4).

Another interesting observation, again from Mattacodi, relating to the specificity of the relationship between the gall and the galler insect (Table 2.2), is: two rose galls, both of them spongy, but one of them hairless ("non cappelluta"), the other hairy ("cappelluta"), are actually caused by two different "kinds of little flies" (Vallisneri 1726, p. 67) (presumably two different species of *Diplolepis* (Cynipidae) (Redfern and Askew 1992, p. 69)).

However, there are still a few cases, reported both by Mattacodi and Vallisneri (Vallisneri 1726, p. 66; Vallineri 1696, p. 316), in which a willow gall is used only as a temporary shelter by an insect which lives part of the time outside it and, in Mattacodi's opinion, this can be explained as reuse of a gall which the larva has left in order to migrate into the earth, as we have said.

2.5 Discussion

This work is not the first attempt to produce a systematic determination of the parasitoid wasps identified by Antonio Vallisneri and his cohorts. To our knowledge, it has been preceded by at least two other studies here cited: one by Pampiglione et al. (2000), the other by Tremblay and Masutti (2005). However, these works are not of great help when trying to identify the insects described: the former, because it confines itself to describing the work Vallisneri did on these organisms very generically, with few identifications of the wasps he actually studied; the latter, as it proposes implausible identities, which are sometimes decidedly erroneous, as pointed out above. Moreover, as regards the galler insects described by Vallisneri, we do not know of any other modern analysis. And this should not be surprising, considering that these descriptions are mostly taken from unpublished works, from the *Quaderni di Osservazioni* to be precise (only the first and second of which have recently been transcribed), while just a few are from published works (Table 2.2). Instead, the descriptions of parasitoid wasps are mostly taken from *La Galleria di Minerva* and from a published letter written by Cestoni.

Valuable data have been found in the works we analysed on various aspects of parasitoid-host relationships in taxonomically diverse ecological webs. Vallisneri and Cestoni provided information that could be associated with a variety of hymenopteran parasitoids of other Hymenoptera (e.g. Ichneumonidae attacking Sphecidae, Torymidae attacking Cynipidae), dipteran parasitoids (e.g. Tachinidae,

Sarcophagidae) of Hymenoptera, coleopteran parasitoids (e.g. Ripiphoridae, Cleridae) of Hymenoptera, and hymenopteran parasitoids (e.g. Chalcididae, Pteromalidae) associated with non-hymenopteran hosts such as Homoptera, Lepidoptera and spiders. Overall, about 20 wasp genera could have been objects of Vallisneri and Cestoni observations. Noteworthy, Vallisneri and Cestoni provided about 300 years ago the first detailed ecological and ethological data on many wasp species from a wide range of life-histories.

Our study provides thus an example of the "hidden" biodiversity data that could be brought to light through detailed analysis of pre-Linnean works. By assigning taxonomic names (with the precision that was possible given the descriptions available, see below) we have provided information that could be incorporated in biodiversity databanks, that clearly play a fundamental role in biodiversity science (including conservation science).[1] Contribution to biodiversity data through the inspection of pre-Linnean works was already highlighted for botany, e.g. by analysing ancient herbaria and thus retrieving information on distribution and diversity of plant species >300 years ago (e.g. Pulvirenti et al. 2015), though such studies seem to be scarcer for zoology. In addition, our study revealed many aspects of the behaviour and ecology of a number of species, notably on their interactions with other species, thus providing data on ecological networks at the time of Vallisneri and Cestoni. Geographical data associated with pre-Linnean insect identification have furthermore the potential to better reconstruct a species' distribution more than 300 years ago, thus revealing changes in distribution across the last centuries.

The result of the present work is actually fragmentary, due to the difficult identification of insects based on the morphological descriptions; however, its is interesting in that several ecological and behavioural characteristics of these species could be used to help such identification. However, these aspects are rather lacking in the case of gall insects, apart from the few cases mentioned. Thus the insects have to be identified on the basis of morphological descriptions, which, although painstakingly thorough and rich in detail, often lack the features utilized in modern systematic criteria. Despite such obvious flaws in our species identification, we still believe in the importance of inspecting ancient studies and do the best to assign a taxonomy to the observed individuals. This could in most cases be the genus but not the species. However, for the problems associated with the co-occurrence of cryptic species, a valuable option may be to consider a taxonomic sufficiency approach, i.e. using higher taxa instead of the species in biodiversity analyses.[2]

At the end of the XVII century, as for most of the Eighteenth century, naturalists did not possess systematic methods to precisely identify the majority of insects, which have a variety and a complexity of forms unaccountable by using descriptions of an intuitive nature, even if they are as exhaustive as possible, as are those given by Vallisneri. This was a problem Réamur, too, would come up against in his monumental work devoted to these organisms, around 40 years later. The identifica-

[1] See Barberousse and Bary, Chap. 3, in this volume.

[2] See Chenuil et al., Chap. 4, in this volume.

tion of the insects according to their morphological features in the times of Vallisneri, like the one produced by Ulisse Aldrovandi around a century before Vallisneri or the then very recent example given by John Ray, did not go much beyond an elaboration of what Aristotle had said in his *Historia Animalium*. It should be remembered that even the distinction between the two most important groups that Vallisneri is examining in connection with the studies which concern us here – Diptera and Hymenoptera – is often hazy and unclear. Even if, in this case, the difference is very obvious – Diptera are two winged, Hymenoptera four – something which Vallisneri definitely realised; their general appearance, which is often very similar, seems to eclipse this fundamental difference. Francesco Mattacodi proves that this problem exists, in one of the letters already considered

> However, I should think that these little mosquitoes are incorrectly called so, like those from the blisters found on elms and poplars, regarding which I must beg you, when you have leisure to do so, to explain how Lyster makes his distinctions between mosquitoes, wasps and flies etc., as I fail to understand how various sorts of Insects with four wings, even if their features conform to those of flies, should not be called by that name, seeing that flies have, again according to Aristotle himself, only two wings. (Vallisneri 1726, p. 67)

Here we can see direct evidence of the lack of a "methodical" system, which would allow a few, clearly defined distinctive features to be isolated in order to avoid this sort of ambiguity. On the other hand, even Linnaeus, the inventor of the "Method", would not go far beyond subdividing insects into their principal orders. For example, in the tenth edition of *Systema Naturae* (Linnaeus 1758, pp. 553–583), Hymenoptera were divided into eight genera*: Cynipis* (Cynipidae, gall-making Hymenoptera Parasitica), *Tenthredo* (Tenthredinidae and other Hymenoptera Symphyta), *Ichneumon* (Ichneumonidae), *Sphex* (Sphecidae), *Wasp* (social and sphecid wasps), *Apis*, *Formica*, *Mutilla* (a genus of Hymenoptera Aculeata parasitoid of other Hymenoptera Aculeata), giving a total of 229 species for the whole Order.

Vallisneri's and Cestoni's "systematic" classification does not make use of this or other similar terminology, as the only valid grouping they give is actually the individual "species" ["spezie"]. At this level, the number of species described is greater by several orders of magnitude than the corresponding lists provided by Linnaeus or Ray, and their description immeasurably more elaborate and detailed. However, one might ask: what is the taxonomic benefit of such meticulous description, if it does not allow the reader to identify the 'species' in question?" We think it proved particularly useful for Vallsneri himself, as a sort of personal reminder, so he could subsequently recognise a species he had previously observed. This might be one explanation for these minutely detailed descriptions, which are even greater in number in the *Quaderni* than they are in the printed works. However, it seems less likely that they could be used to enable readers to identify the species described, due to the difficulties already indicated, in addition to the use of generic qualitative adjectives, such as "large", "little", "long", "short" etc., lacking any standard reference indication.

In fact, as early as 1713, in *Idea nuova d'una Division generale degl'Insetti*,[3] Vallisneri had set forth a traditional type of systematic classification which he had, admittedly, taken from other contemporary authors (and which, indeed, was based on that proposed by John Ray, 3 years earlier, in his *Historia Insectorum* (Ray 1710)), in which identification was based on the number of wings and "feet". This bore an explicit analogy with the botanical systematics of Tournefort (Vallisneri 1713, p. 67), who had chosen, amongst all other possible bases, to use only the features of the flowers and the seeds to draw up a classification of plants. However, this division, which was "methodical" if a bit rough and ready, seems to have been proposed only as a form of respect for the new methodical systems which were then beginning to arise; Vallisneri thought it should be supported by another method, on which he focussed his energies and interest, which would be ecological in nature. With a strange turnaround, disregarding the classification normally used, perhaps with the aim of diminishing the importance of the "methodical" system, he would use ecological features in order to initially subdivide the insects into "Classes" (Vallisneri 1713, p. 42). The other method, instead, would confine itself to determining the "specific differences" (Vallisneri 1713, p. 65).

We are now in a position to understand why Vallisneri often seems to treat flies and wasps as altogether similar insects. The mere fact that they possess one or two pairs of wings - a feature considered to be so important by the majority of zoologists who preceded, were contemporaries of or came after Vallisneri -, is instead judged by him to be just a "specific difference"; to Vallisneri, the fact that both flies and wasps shared a habitat is much more significant, justifying their inclusion in the same "Class". Thus, when Vallisneri finally needed to define the species, he would use only a description, set forth in minute detail, of all the morphological features of the insect.

However, it is definitely not the ability of Vallisneri and his circle to systematize which should attract our attention, as much as the wealth of that finely drawn detail in his ecological and behavioural observations, which show the level reached by naturalistic observation, at least in regard to insects, towards the end of the seventeenth century. This would be what ushered in a great era devoted to the study of the behaviour of insects, which would see its crowning achievements with Réamur in France.

It could be argued that Vallisneri considered himself to have been responsible for the discovery of the phenomena relating to parasitoid wasps (Vallisneri 1710, p. 15), apparently not taking into account what other authors had already written about this matter before him.

Even if he was aware of what Swammerdam had already written about insects, "diversae speciei" of which emerged from caterpillars and chrysalises (Vallisneri 1710, pp. 13–14), he believed that this author, who reported the phenomenon "ad

[3] See also (Andrietti and Generali 2004, pp. 110–115) for a brief examination of this study.

quartum mutationis ordo" ["fourth order of metamorphosis"], attributed it to "an internal principle" (Vallisneri 1710, p. 14; Swammerdam 1693, p. 131) and not an *ex ovo* origin. Van Lenteren and Godfray (2005, p. 17) opt for a different interpretation: in view of the fact that Swammerdam's "fourth order of metamorphosis" is always based on an egg, they suggest that he viewed parasitoids in the same way. In any case, in his next work, "Bybel der Natur", Swammerdam reports the observation (made by Otto Marsilius van Schriek) of a caterpillar being stung by a "fly", which he connects with the immission of eggs by the parasitoid (van Lenteren and Godfray 2005, p. 18).

Vallisneri certainly could not have known of this second work by Swammerdam (finished in 1679 but only published in 1737–1738) (van Lenteren and Godfray 2005, p. 17). However, he did not even seem to be informed (or at least he did not make any mention) of other researchers which, in the two decades preceding his *Dialoghi*, had already made similar suppositions and, in some cases, actually observed the process of parasitoids laying eggs in their host (for a list of various researchers who reached similar conclusions before Vallisneri, see van Lenteren and Godfray 2005). Before being called by the Università di Padova the sources of Vallisneri's entomological studies were mainly Italian, and his knowledge of European literature was quite limited (Generali, *Introduzione*, in Vallisneri 2004, pp. XXVIII-XXXVI), with the exception of Martin Lister, constantly quoted in Vallisneri's *Quaderni* for his edition and notes to *De Insectis* of Johannes Goedaert (1685). Already in some letters published in the 1670–1671, Lister had suggested the possibility that certain insects could lay their eggs inside living caterpillars (van Lenteren and Godfray 2005, pp. 18–19). In addition, in his note 4 at page 9 of the mentioned work, he writes: "Muscae octoginta duae [observed by Goedaert], hic memoratae, fuerunt cuiusdam muscae Ichneumonis progenies, in Erucae corpus delata; quo pacto autem illuc deferatur, fateor me adhuc non satis ossequi; sed conjecturam facio, illud ab Ichneumone Parente fieri" ["The 82 flies that emerged from the pupa are the progeny of an ichneumon fly, which had gotten into the caterpillar in a manner that is still not entirely clear to me. In all likelihood they were laid right there by the mother fly"] (translation by van Lenteren and Godfray 2005, p. 19).

In this regard, we should recall also Cestoni's observations of the plant flea mentioned above, in which he declared, rather ironically, that the *Aphidius* "had very kindly laid an egg" under its belly. If it is true that his letter is dated 25th July 1698, it appears that it was terminated from the year 1692 (Cestoni 1787, note at page 271; Tremblay and Masutti 2005, p. 37).

Acknowledgements The authors are indebted with Alessandra Cuminale for her help in bibliographical researches and with Dario Generali for his many helpful comments and suggestions. We thank Elena Casetta and Davide Vecchi for the interest shown in our contribution and the invitation to join the book project. Part of the present work has been realized making use of a PRIN 2007 funding (Università degli Studi del Piemonte Orientale, di Milano, Ferrara, Pavia e Insubria) coordinated by Maria Teresa Monti. CP was funded by a Post-Doctoral contract from the Universidad de Castilla-La Mancha.

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