

Report by J. Poupin, expert in Marine Biology

12 August 2015

Expert report on the barnacles (*Cirripedia Crustacea*) attached to aircraft debris beached on Réunion Island

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Summary

- The crustacea attached to the flaperon are *Lepas (Anatifa) anatifera striata* de Graaf, 1952.
- This subspecies is strictly pelagic, and always lives on floating objects.
- It is cosmopolitan and disseminated in the world's oceans at tropical to temperate latitudes in waters at temperatures above 18-20°C.
- Rarely reported in the literature, this subspecies is specifically mentioned off Western Australia.
- The size of the larger specimens indicates that initial colonization may date back 15-16 months.
- The locations of the colonies of *Lepas* on the flaperon indicate that it floated with its ventral side facing upward.

Background

This expert appraisal was carried out as part of an assignment defined by a mission statement, dated 3 August 2015, by Mr. François Grangier, aviation expert designated as part of an investigation into an aircraft component that had drifted onto a beach in Réunion Island that might be a flaperon from the Boeing 777-200 on flight MH370. The chronology of events is as follows:

- 7-8 March 2014: disappearance of the Boeing 777-200 on flight MH370
- 29 July 2015: discovery of the Boeing flaperon on Réunion Island

Against this background, on Sunday, 9 August 2015, I carried out an assignment involving sampling and observation at the DGA / Toulouse, where the part was stored.

This report was written on 10-12 August 2015. It shows the results of my findings and observations, the aims of which were as follows:

- Identify the marine organisms attached to the flaperon and their origin
- Assess the immersion time of the flaperon, if possible in order to estimate the initial immersion point of the part in the ocean.

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Sampling conditions and methods (9 August 2015)

After it was found beached on Réunion Island (07/29/2015) the flaperon was taken to the premises of the DGA / Toulouse.

The observations and samples were carried out on Sunday, August 9 by Mr. Poupin with the help of DGA / Toulouse personnel, Messrs Plotka, Bordes and Hakenholz, i.e. 12 days after the part was found beached.

At first observation it appears that **the marine organisms attached to the flaperon were barnacles (Cirripedia Crustacea) of the Lepadidae family and of the *Lepas* genus**. No other organisms were found attached to the flaperon.

The colonies of barnacles dried with a fairly strong 'marine' odour. The calcareous valves of the organisms tended to crumble and easily fall off. The soft parts, feet and internal organs, were shrivelled and dried. Some individual barnacles had obviously been loosened. However, if the spread of the specimens on the part examined on Sunday August 9 is compared with its appearance at the time of its discovery on Réunion Island (see the video archives and media photos), there was no major loss of barnacles.

Examination of the colonies of barnacles on the part

A comprehensive examination of the part was performed first of all to identify the colonization points of the barnacles on the flaperon. The flaperon rested on its flat portion, the curved portion being visible only at the beginning of the examination. To observe the underside of the flaperon (flat portion), it was set on trestles to enable observation of the greater part of the flat "ventral" surface. The following observations were made:

- The barnacles attached more on the side and rear edges or on parts of the flaperon which were scratched or damaged, revealing the underlying composite material. The largest specimens were located on the left rear flare (Figure 1)
- The curved part (dorsal) had some disparate colonies of barnacles, especially on the right side. These specimens were small (Figure 2)
- The flat part of the flaperon (ventral) was colonized very little or not at all by the barnacles (Figure 3)

Cirripedia crustacea of the *Lepas* genus always live on objects floating in the sea. They are located in the water, just below the waterline of their support.

The observations made suggest that **the flaperon floated with its flat surface facing upward**, the domed part being submerged. Subsequent buoyancy tests are expected to confirm this point by checking that the parts that remain exposed to the air were much less colonized by the barnacles.

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Figure 1 - Fastening of barnacles more on the rough portions of the flaperon, from left to right and top to bottom: front left edge with the largest barnacles, small colonies on scratched areas of the dorsal side, colonies attached to the bare composite on the back edge, colony on a metal corner.



Figure 2 – Dorsal appearance of the flaperon, domed section, from left to right and top to bottom: left side, right side, front, dorsal-posterior view (small colonies of barnacles on the right).

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Figure 3 - Ventral appearance of the flaperon, flat part, from left to right and top to bottom: left side, right side, front ventral, rear ventral.

Sample collection

Two samples were taken of 10-30 attached specimens and kept in 95° alcohol (Figure 4):

- One from the left front angle (Fig. 4 left)
- The other from a rear edge (Fig. 4 right)

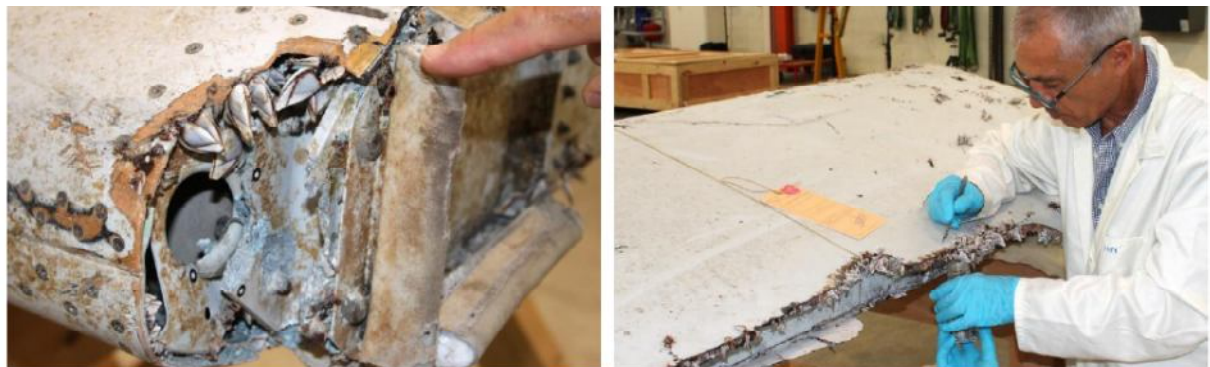


Figure 4: Location of 2 samples taken on 9 August 2015

To these two samples were added to two further lots of 10-30 specimens taken on 6 August by Mr. Hakenholz, one kept in 95° alcohol, the other in 10% formaldehyde.

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These four samples were taken by Mr. Poupin for further examination and preparation of this expert report.

Condition of the barnacles remaining on the flaperon

After being exposed to the air for several days, the colonies still attached to the flaperon were brittle with a tendency to "crumble" and fall off easily.

To facilitate the handling of the flaperon for the rest of the expert appraisal, so as to preserve the specimens not sampled, it was therefore decided, in agreement with Mr Grangier, to collect the remaining specimens, at least the largest colonies and most exposed to handling, and keep them dry in heat-sealed plastic bags.

Results of the expert appraisal by Mr. Poupin (10-12 August 2015)

Ascertainment of the species

The *Lepas* genus has 8 species listed in the World Register of Marine Species (WoRMS 2015):

1. *Lepas (Anatifa) anatifera* Linnaeus, 1758
2. *Lepas (Anatifa) anserifera* Linnaeus, 1767
3. *Lepas (Anatifa) australis* Darwin, 1851
4. *Lepas (Anatifa) beringiana* Pilsbry, 1911
5. *Lepas (Anatifa) hillii* Leach, 1818
6. *Lepas (Anatifa) pacifica* Henry, 1940
7. *Lepas (Anatifa) pectinata* Spengler, 1793
8. *Lepas (Anatifa) testudinata* Aurivillius, 1892

The species *L. anatifera* also comprises three sub-species, all mentioned in the Australian Biological Resources Study (ABRS, 2015):

1. *Lepas (Anatifa) anatifera anatifera* Linnaeus, 1758
2. *Lepas (Anatifa) anatifera dentata* Bruguière, 1789
3. *Lepas (Anatifa) anatifera striata* de Graaf, 1952

The examination was carried out under binocular microscope by observing the hard outer parts as well as the internal soft tissues that were still sufficiently well preserved. The reference works consulted were: Barnard (1924), Darwin (1842), Graaf (1952), McLaughlin (1980), and Relini (1987).

The terminology used for the morphology (e.g. capitulum) is taken from McLaughlin (1980) (Figure 5)

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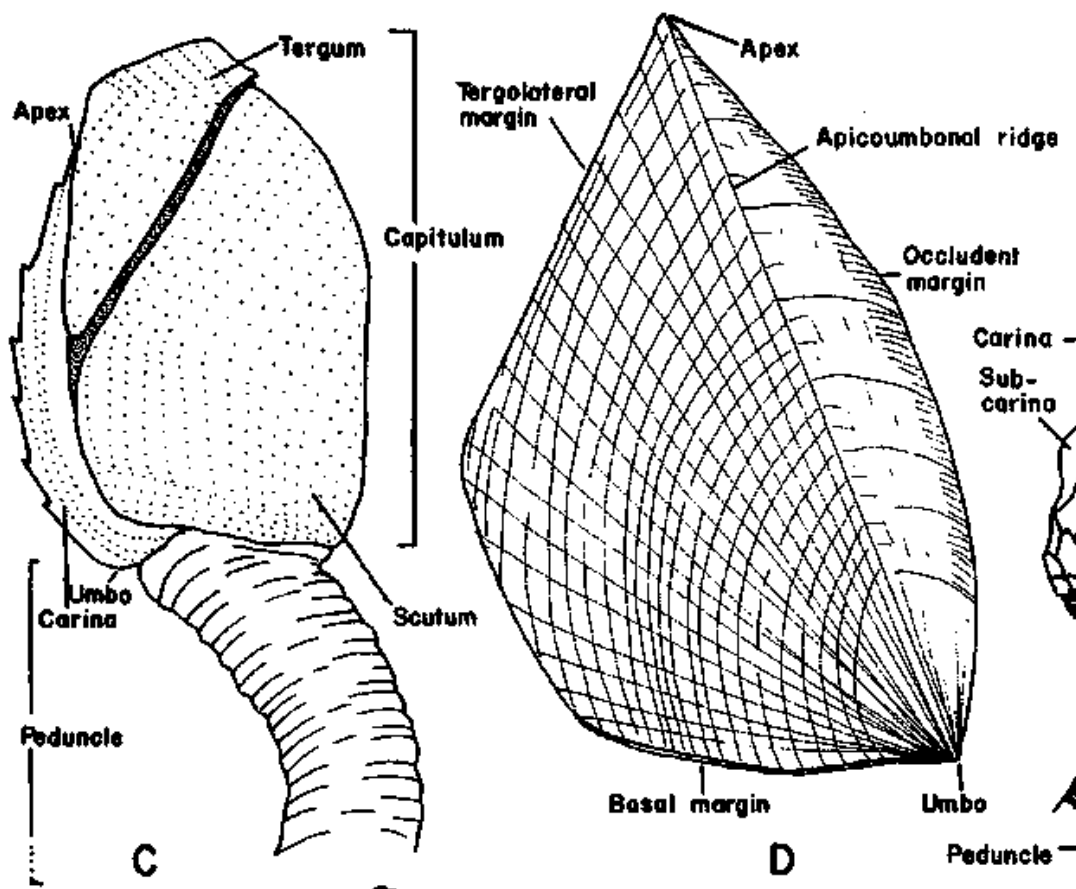


Figure 5 - External morphology of the plates of a Lepadidae according to McLaughlin (1980, fig. 17)

Graaf (1952) mentions that the external morphological characteristics, such as defined by Darwin (1852) to distinguish *L. anatifera* and *L. anserifera*, are highly variable and that the two species can therefore be confused. In his opinion, the only valid morphological criterion for recognizing these two species is the number of filamentary appendages on the soft parts (p. 1-2 *The valves of the capitulum of L. anatifera and L. anserifera are subject to a rather extreme variability - After these considerations it must be clear that in many cases only the number of the filamentary appendages can render a safe identification possible*).

According to the description by Graaf (1952) the **specimens attached to the flaperon belong to the subspecies *Lepas (Anatifa) anatifera striata* de Graaf, 1952**. In particular, the following characteristics mentioned by Graaf (1952) fully correspond the specimens examined (Figure 5):

Terga and scuta smaller than in the forma typica, more or less striated, especially the scuta. Scuta sometimes with a diagonal row of depressed quadrilateral marks of a brownish-green colour. Carina often more or less strongly dentated. Peduncle shorter than in the forma typica. Animals on the whole approaching Lepas anserifera in external appearance. Filamentary appendages two on each side as in the forma typical

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Figure 5 – Two specimens of *Lepas (Anatifa) anatifera striata* de Graaf, 1952 from the flaperon. The different size (capitulum length of 36 mm, on the left, 19 mm, on the right) illustrates the variations of the radial streaks in this species.

Geographical distribution

Like all of the *Lepas* genus, *L. (Anatifa) anatifera striata* is a cosmopolitan species. If *Lepas anatifera* has been frequently reported in the literature, this is not the case of the *striata* subspecies which was only described fairly late by Graaf (1952) and which has often been reported under the name *L. anatifera* in the broad sense, without specifying the subspecies.

It is therefore not possible to precisely define the geographical range of *L. (Anatifa) anatifera striata*, but it is known to be present in the temperate and tropical latitudes of the Atlantic, Indian and Pacific oceans.

It is interesting to note that one of the few formal records of this subspecies concerns Western Australia (ABRS, 2015).

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Ecology

L. (Anatifa) anatifera striata is a species that fastens in colonies onto floating objects of a very diverse nature, preferring their rough surfaces. The colony lives below the waterline near the surface. The species prefers water at a temperature above 18-20°C. The species is hermaphroditic, without being able to self-fertilize (Patel, 1959). The optimum reproduction temperature is between 18-25°C. Individuals are mature only 4-5 weeks after the first attachment and larvae can then be produced in large quantities.

The different size ranges observed on the flaperon, of which there were at least four (1-10 mm, 10-20 mm, 20-30 mm, > 30 mm), show that recolonization of the flaperon occurred from the first individuals that attached.

Size and Growth

The size of the specimens, measured as the maximum length of the capitulum, was between about 1 mm for the smallest specimens to 36 mm for the largest measured specimen.

The growth of *L. anatifera* barnacles was studied by Evans (1958) and Skerman (1958) with the data summarized by Thiel & Gutow (2005: 352, tab 17.). According to these authors, the growth of barnacles is rapid after the first colonization:

- The growth of the capitulum can reach 0.44-0.55 mm/day.
- The size of the capitulum at sexual maturity is 13-23 mm.
- Sexual maturity is reached after 30-37 days (4-5 weeks)

Growth data for the capitulum of a species of the *Lepas* genus (*L. anserifera*, very close to *L. anatifera*) by Evans (1958) are summarized in Table 1 (in grey).

Capitulum size (mm)	Days
8	8
21	40
25	107
36	476

Table 1 - Number of days required for different growths of the capitulum observed in barnacles of the *Lepas* genus (*L. anserifera*), very close to *L. anatifera striata*. The data in grey are taken from Evans (1958). The results in italics in the last row correspond to an extrapolation of the number of days required for growth of a size of 36 mm, estimating growth of the logarithmic type.

Evans' data (1958) are best adjusted for growth of the logarithmic type according to the equation:

$$\text{Capitulum size (mm)} = 6.7041 * \ln(\text{days}) - 5.333 \quad (r^2 = 0.9752)$$

By extrapolation, the colonization of the larger specimens (36 mm) on the flaperon dates back 476 days (Figure 6), i.e. 15-16 months.

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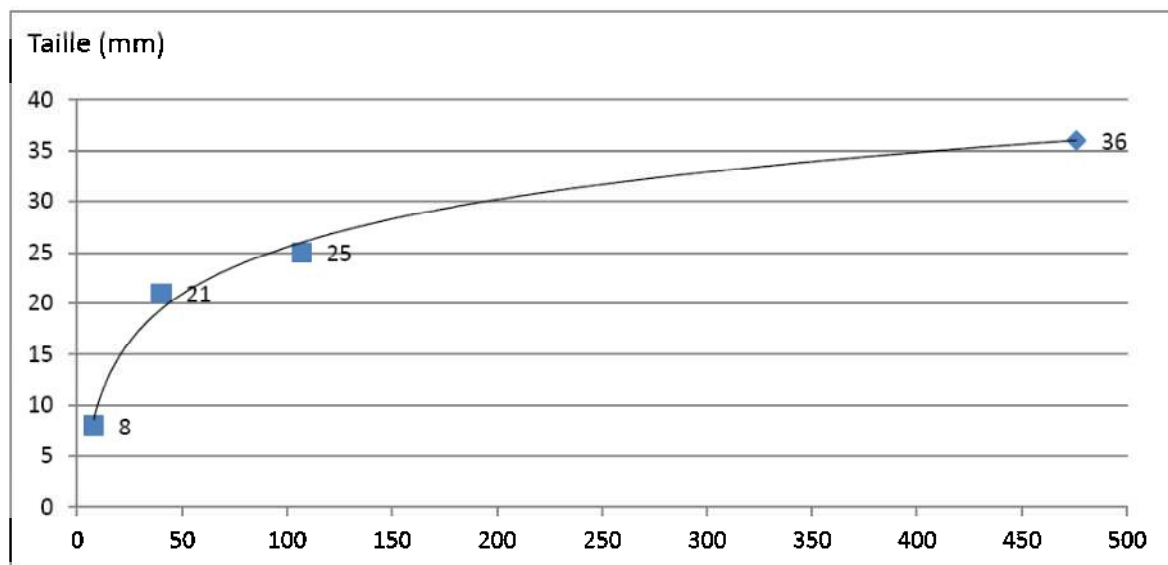


Figure 6 - Increase in size of the capitulum of a *Lepas anserifera* barnacle depending on the number of days (days 0-500). The first three points (squares) correspond to the literature data (see Evans, 1958). The last point (diamond) is extrapolated from the first three items of data by estimating logarithmic growth. The size of 36 mm corresponds to a growth period of 476 days, or 15-16 months.

The growth of the specimens stopped on the day of the discovery of the flaperon on Réunion Island, on 29/7/2015. The initial colonization therefore dates back to 10/04/2014, or 33-34 days after the disappearance of the Boeing 777-200 on flight MH370.

This estimate, although very approximate and based on limited data, reinforces the idea that this part could belong to the missing aeroplane.

Possible additional examinations

As part of this expert appraisal, I consulted my colleague and crustacean expert at the Western Australian Museum, whose contact details are as follows:

Diana S Jones

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Western Australian Museum

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Ms Jones is a recognized specialist for the barnacle group. Incidentally, she was chosen as the expert by the Australian Transport Safety Bureau in the case of flight MH370. She kindly provided me with her own expert appraisal, based on press photos.

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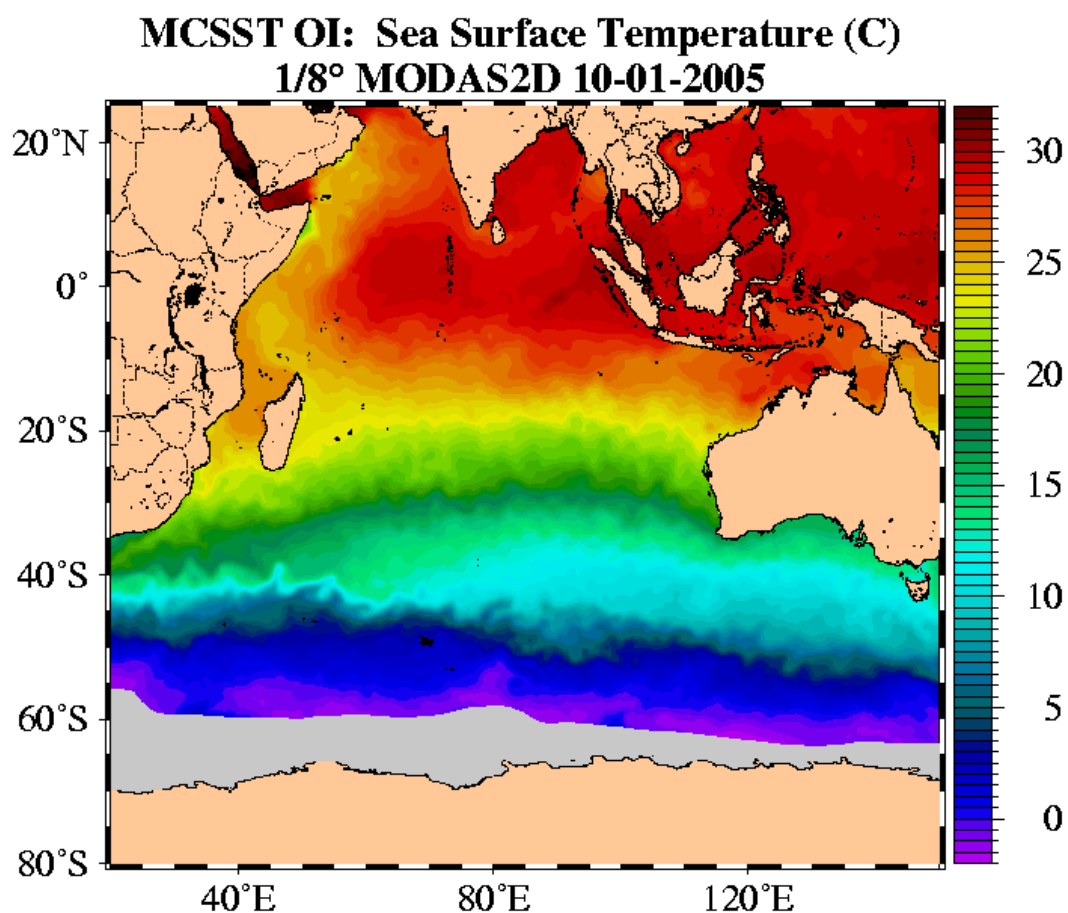
In broad terms, with the exception of the growth analysis in this report, her conclusions are similar to mine.

In addition, D. Jones suggested an analysis of the calcium deposits in order to establish the ratios of oxygen isotopes. This technique would make it possible to identify the temperature of the water after the barnacles became attached.

However, given the distributions of the surface temperature in the Indian Ocean (Figure 7), even if knowledge of the water temperature at the time of growth of the deposits were accurately known, it would help to identify the latitude with greater certainty but would provide little information about the longitude.

Not being an expert on this issue, I have forwarded her suggestion for further analysis. For information, a French laboratory that could perform this type of analysis is the 'PALEOCEAN' group of the LSCE in Gif-sur-Yvette

[http://www.lsce.ipsl.fr/Phoce/Vie des labos/Ast/ast groupe.php?id groupe=13](http://www.lsce.ipsl.fr/Phoce/Vie%20des%20labos/Ast/ast%20groupe.php?id%20groupe=13)



Naval Research Laboratory MODAS 2.1

Figure 7 – An example of the distribution of surface temperatures in the Indian Ocean

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