Biodiversity Journal, 2022,13 (3): 685–693 https://doi.org/10.31396/Biodiv.Jour.2022.13.3.685.693

Can a leopard seal, *Hydrurga leptonyx* de Blainville, 1820 (Mammalia Phocidae), change its spots? (Spoiler alert: at least one adult female can)

Ingrid N. Visser[1,2,3,A], Krista van der Linde[1,3,4,B], Sarah E. Richard[1,2,C], Tracy E. Cooper[1,2,D], Terry M. Hardie[1,2,E] & Rick Bout[1,F]

[1]LeopardSeals.org, www.LeopardSeals.org, New Zealand
[2]Orca Research Trust, P.O. Box 1233, Whangarei, New Zealand
[3]School of Biological Sciences, University of Canterbury, Christchurch, New Zealand, 8041
[4]World Wildlife Fund for Nature New Zealand (WWF-NZ), Level 2/77 Thorndon Quay, Pipitea, Wellington New Zealand, 6011
Corresponding author: ingrid@orca.org.nz
ORCID: [A]https://orcid.org/0000-0001-8613-6598 - [B]https://orcid.org/0000-0002-3757-2200 - [C]https://orcid.org/0000-0002-9358-3174 - [D]https://orcid.org/0000-0001-6713-0502 - [E]https://orcid.org/0000-0002-7643-2729 - [F]https://orcid.org/0000-0002-5230-057X

ABSTRACT Leopard seals, *Hydrurga leptonyx* de Blainville, 1820 (Mammalia Phocidae) have variable spots on their pelage, allowing for identification of individuals. We monitored a subset of spots (*n*=40) on the face and neck of an adult female leopard seal residing in New Zealand. We compared images that were 1,701 days (~4 years and 8 months) apart. The use of scars, acquired from wounds, allowed for cross-matching and confirmation that this was the same individual. We investigated if the spots were more visible when the animal was wet or dry. We found that all 40 spots were visible during this time period and when the animal was both wet and dry. However, they were better defined, and therefore more visible, when the pelage was wet. Additionally, we identified a number of new and emerging spots, none of which masked or obscured the 40 aforementioned spots. These changes illustrate that diligence must be applied when matching individuals over long periods, to ensure that mis-matches and missed matches do not occur. Our findings do not invalidate photo-ID studies for leopard seals, rather they show that this is a robust system of identification, as spots were not lost over time and spot patterns were an effective tool for both individual identification and observing pigmentation change.

KEY WORDS Capture-mark-recapture; leopard seal; photo-identification; pinniped; pelage; resident; scar longevity.

Received 13.05.2022; accepted 08.07.2022; published online 18.09.2022

INTRODUCTION

Identification of individuals is one of the fundamental baselines for understanding *inter alia*; following focal animals, behavioural complexities in social groups, unravelling the ecology of a species, improving management plans when cohabitation conflicts occur and correct identification of potential nuisance individuals (e.g., Altmann, 1974; Fraker, 1994; Visser, 2000; Boyd et al., 2010; Clutton-Brock & Sheldon, 2010; Ford, 2010; Massei et al., 2010; van der Linde & Visser, 2020).

Historically, identification included capture-mark-recapture (CMR) methods such as tags or marking (e.g., a range of methods used on pinnipeds is given in Siniff & DeMaster, 1979), but more recently, acquired and naturally occurring scars and pigmentation patterns on marine mammals have also been utilized and incorporated with photo-identification (photo-ID), removing the need for physically capturing (or even marking) the individual (e.g., Hammond et al., 1990; Urian et al., 2014; Copello et al., 2021). When matching individuals, typically, a small range of unique features are utilized to confirm identification e.g., for orca (*Orcinus orca* Linnaeus, 1758) this may be a series of notches in the dorsal fin combined with pigmentation on the body (Visser & Cooper, 2020), for sea otters (*Enhydra lutris* Linnaeus, 1758) it may be nose scars (Gilkinson et al., 2007) and for polar bears (*Ursus maritimus* Phipps, 1774) whisker patterns (Anderson et al., 2010).

Some species of pinnipeds exhibit spots on their pelage allowing for identification of individuals e.g., grey (*Halichoerus grypus* Fabricius, 1791) (Hiby & Lovell, 1990) and harbour seals (*P. v. vitulina* Linnaeus, 1758) (Langley et al., 2021). This technique has also been successfully applied to leopard seals (*Hydrurga leptonyx*, de Blainville, 1820) at South Georgia Island (Walker et al., 1998; Forcada & Robinson, 2006) and in New Zealand (NZ) waters (Hupman et al., 2020; van der Linde et al., 2021). Herein, we investigated if there were any changes of the spot patterns during the monitoring of one individual over a number of years.

MATERIAL AND METHODS

Using the database developed by Leopard-Seals.org (a non-profit organization based in NZ, run by researchers who volunteer their time; see www.leopardseals.org for details), we extracted high-resolution images from days where researchers attended sightings of a female leopard seal (HLNZ-001) and compared them over time.

As a result of HLNZ-001 showing long-term residence in NZ waters the local Māori hapu, Ngati Whatua ki Orakei, named this female leopard seal "*He owha nā ōku tūpuna*" meaning "treasured gift from our ancestors". The shortened version, "*Owha*" has become her colloquial name. To ensure that we were examining images of the same individual, we cross-matched images from each day that included acquired marks such as a 'crescent shaped' scar with two parallel scars on her right-side flank, and a forward-facing V-shaped scar posterior to the gape on the left-side of her face, all of which were visible for the duration of the study (van der Linde et al., 2022). For this current study, the V-shaped scar was used as an orientation reference point (supplemented by the position of the mouth gape, eye and the external opening to the ear canal) for mapping of the spots.

From our extracted images, we then selected a subset which were similar in orientation (i.e., perpendicular to the camera and with the head and body in a similar position). These images were; left full-body, left face/neck, right full-body, right face/neck. To establish if a particular body zone was more suitable for matching spots on this individual, we selected a high-resolution full-body left-side image from 2022 and marked as many darker pigmentation spots and shapes (hereafter referred to as spots) as was feasible. This mapping also gave us a minimum number of spots that would be available for matching. Although many studies use a relatively low number of unique features to identify individuals of various species (as outlined in the introduction), we wanted to increase the probability of documenting any changes in the pigmentation pattern between years (i.e., not just identify the individual), therefore we used 40 unique spots in each image we compared.

Due to the technology available at the time, older images were lower resolution and smaller in size. Therefore, we post-processed those using Photoshop 2022, TopazLabs Stabilize AI and Gigapixel AI software to improve photo quality (i.e., contrast, sharpness) and to resize the images to be similar to the larger, more recent, images for better comparison. To ensure that the enhanced images were not impacted by any machine learning algorithm artefacts, they were compared side by side to the original (unedited) images (see Supplemental Material S-1 for an example).

We hypothesized it would be easiest to identify markings of both scars and spots when the individual was wet. To examine if such states of the pelage would impact the results, we next extracted paired images for each body zone from the subset, which were taken on the same day and where the seal was both 'wet' and 'dry'. We duplicated this process for as many days and years as possible to create a chronological sequence of similar images.

Finally, for the spot analysis, we extracted two pairs of images for the face/neck; the earliest and the most recent pairs of both dry and wet (i.e., two images per day, with a total of four images), ensuring that they were a minimum of three years apart (this timeframe was used based on published photo-ID studies of leopard seals and erring on the side of inclusion - see Discussion for details).

We mapped the spots (using numbers) on the earliest wet and dry pair of photographs and then cross-matched the spots to the most recent photographs, noting any changes (using letters) in number, shape or spot density that may have occurred over time.

RESULTS

We documented >300 spots on the full-body left-side of this leopard seal. We found that the face (anterior from the mouth-gape/eye) of HLNZ-001 had very few spots (Figs. 1–4) and therefore we excluded that zone for matching. However, posterior from the eye (to the tip of the hind flippers), all other areas that exhibited paler pigmentation (and some darker pigmentation areas) had a proliferation of dark spots that were unique, clearly distinguishable and suitable for photo-ID matching. We also noted some paler spots in the dark pelage areas. We therefore ascertained that on this individual no particular body zone (other than the anterior section of her face) was more suitable than another for matching spots.

From the subset of images, only two pairs fulfilled all the selection criteria for our dataset (i.e., pairs of images taken on the same day where the seal was both wet and dry, which were also similar in orientation and at least three years apart). These chronological pairs were the left and right face/neck images from 10 August 2017 and 07 April 2022 (i.e., 1,701 days, or 4 years, 7 months, 28 days apart), as calculated by the online 'Time and Date' calculator (https://www.timeanddate.com).

Although both left and right-side image sequences were suitable on both of these dates, we analysed only the left face images because the V-shaped scar provided a mark that we could use to cross-match and it was not influenced by any potential variation in pigmentation.

We first marked the wet image of the left face of HLNZ-001 from 2017. We found there were at least 80 spots visible, of which we marked 40 that were; in the paler pelage zone, darker in colour, and either (i) in a pattern that might facilitate matching to the remaining three images (e.g., see the line of spots 25–29 in Fig. 1) or, (ii) of a size that they were not obscured by the number we placed on the image (e.g., see the spot marked with the number 30 in Fig. 1). We then marked corresponding spots in the remaining three images (i.e., the wet 2022 image, Fig. 2, as well as the pair of dry 2017, Fig. 3, and 2022, Fig. 4, images).

We found all of the 40 marked spots were visible in each of the four images. Additionally, we found 10 areas (arrows A–J) which were zones where the pigmentation changed (Figs. 1–4). Nine of those (arrows A, B, D–J, Figs. 1, 2) exhibited areas where the pigmentation was either completely absent or not clearly visible in the 2017 images, but which subsequently developed and were visible in the 2022 images. These new spots were either just beginning to emerge (i.e., these were a pale pigmentation) or obviously visible and varied in size and shape. A tenth region (Fig. 1 and 2, arrow C) exhibited an area of pigmentation in 2017 which developed darker pigmentation by 2022.

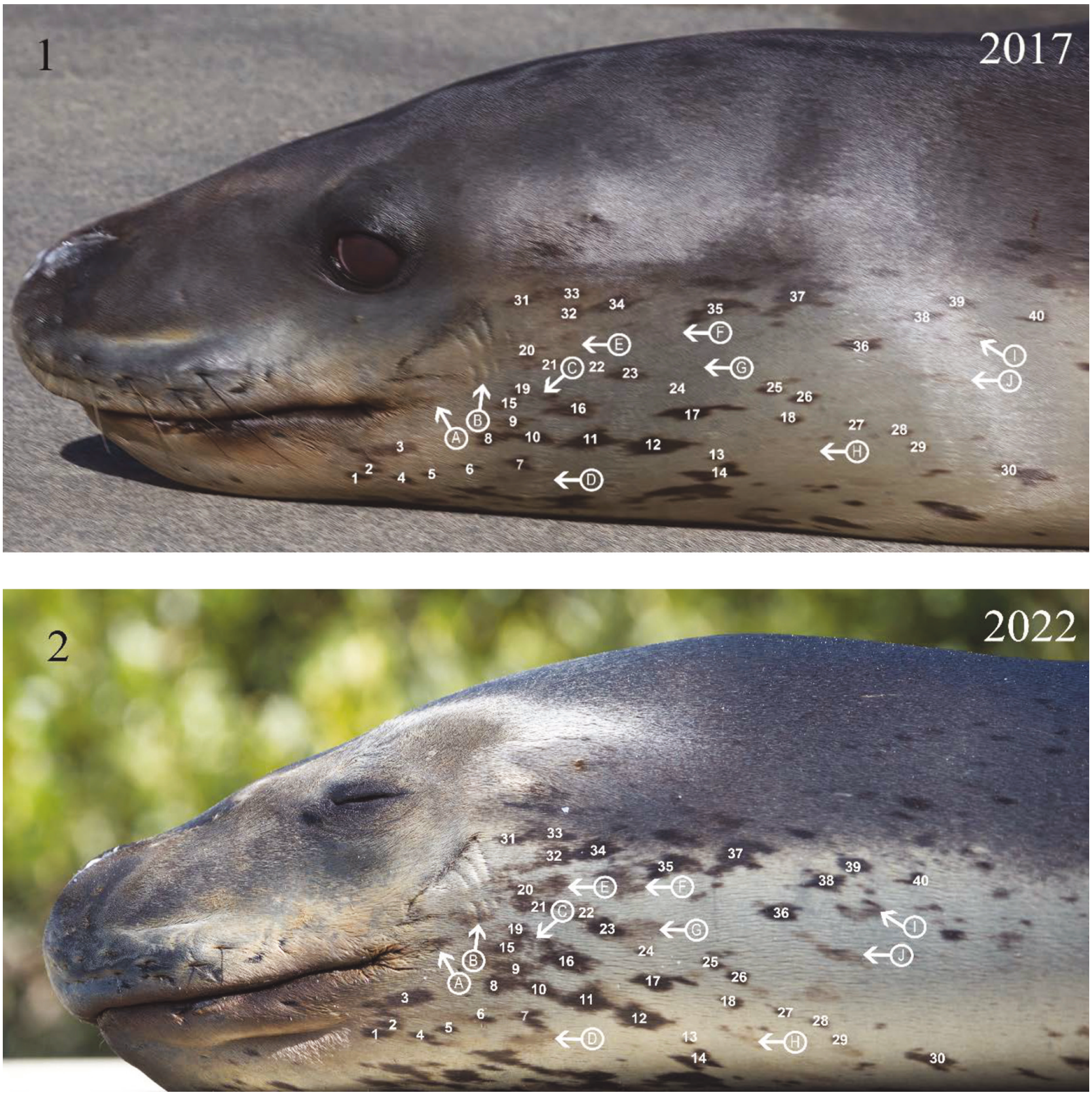
In all 10 instances, none of the new or darker spots obscured, modified or masked previous spots. The meta-comparison of the spots, across the wet and dry images, as well as between years illustrated that she did not lose any of the 40 spots we marked.

DISCUSSION

The use of spots and scars for matching leopard seals has been well established with durations between matches including "*about three weeks*" (King, 1975) and ~150 days (Acevedo & Martinez, 2013).

Hiruki et al. (1999) photographed or tagged (with bleach or flipper tags), 44 leopard seals and stated "*five seals were seen in at least two consecutive years…, and one was seen for four consecutive* *years*", however they do not state if this was a tagged or photographed individual. Walker et al. (1998) photographed or tagged (with flipper tags) 129 leopard seals and mentiond "*150 days*" as the "*longest recorded residence*" and that this individual was "*also present in three consecutive years*", however they do not state if this was a tagged or photographed individual.

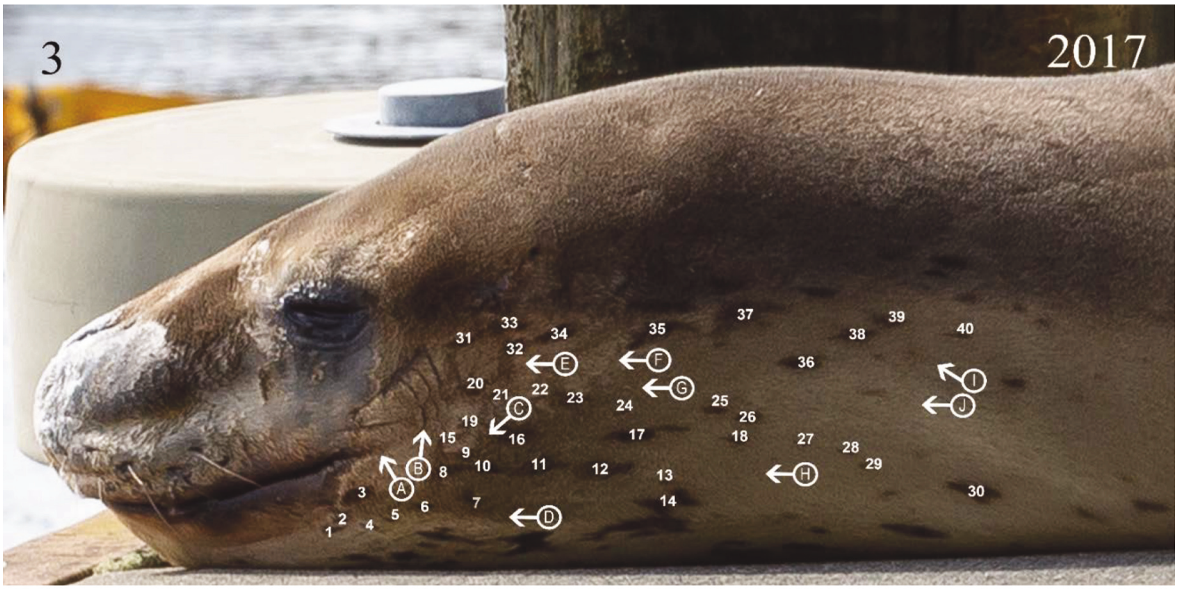
However, these studies did not discuss or document changes in the spots and we believe this is the first study that illustrates changes in leopard seal pelage patterns over time. Although this leopard seal residing in NZ has been documented through photo-ID for more than nine years (LeopardSeals.org, unpublished data), we used a smaller dataset which was limited in time, due to our strict criteria (i.e., images

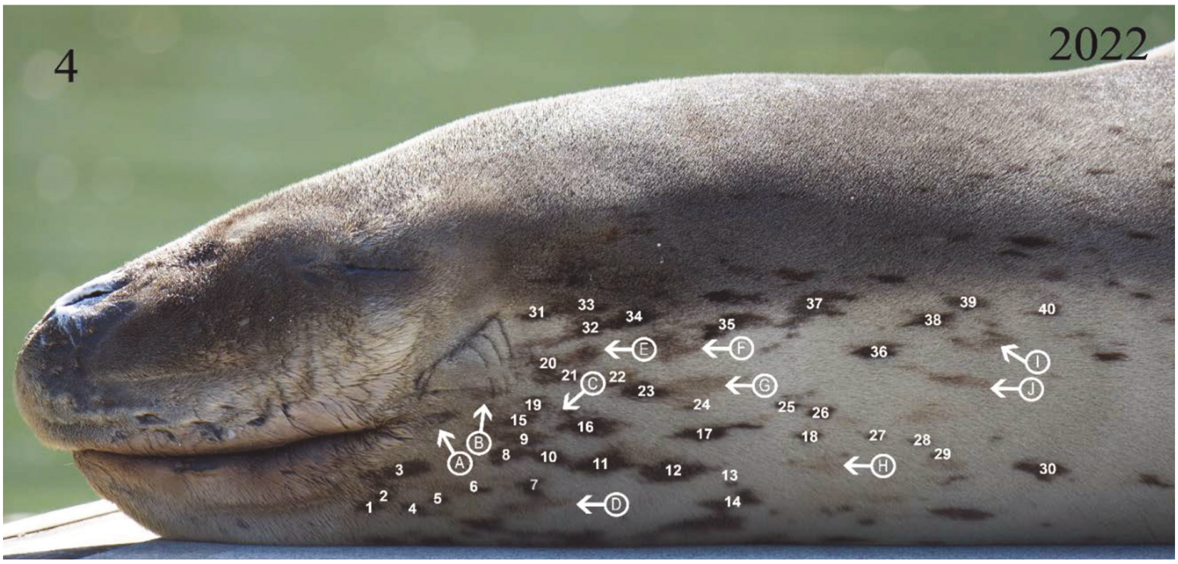


Figures 1, 2. Left-side of the face and neck of leopard seal HLNZ-001, when wet. Note the V-shaped scar just above and posterior to the mouth gape (Fig. 1). When photographed on 10 August 2017, at least 40 spots (indicated by numbers) were clearly visible. All 40 were still visible approximately four years and eight months later 07 April 2022 (Fig. 2). The 10 white arrows in both figures indicate pigmentation that was either barely or not visible in the 2017 image but was clearly visible in the 2022 image. Photographs © Fig. 1: INV, Fig. 2: TMH.

with the same orientation and position of the head). By applying these body position criteria we expected to reduce any potential errors. The additional criteria of having both wet and dry images taken on the same day further reduced our potential sample size, but in this case it ensured integrity of the method across both of the pelage states. Despite these limitations we have verified that in a period of approximately 4 years and 8 months, additional pigmentation was visible in her pelage.

We acknowledge that not all of these additional spots are the same density of pigmentation as the marked 40 spots (e.g., the spots marked by arrows A and B, compared to the spots indicated by numbers 1, 2 and 3). However, they are still clearly visible in 2022 and there are at least 10 examples of this increase in pigmentation, in some cases arising from a complete absence of a spot in previous photographs (e.g., arrow D). Furthermore, none of the new spots masked any previous spots, although some of the





Figures 3, 4. The V-shaped scar just above and posterior to the mouth gape is clearly visible on both the 10 August 2017 (Fig. 3, dry) and 07 April 2022 (Fig. 4, dry). The same 40 spots as indicated in Figs. 1, 2 are indicated here, as are the areas which changed in pigmentation (indicated by the white arrows). Photographs © Fig. 3, INV, Fig. 4, TMH.

emerging spots (e.g., the area indicated by arrow C), should they continue to darken, may merge or obscure the existing spots, however we did not document this. For female grey seals, pigmentation has been documented as darkening as the animal ages, particularly in the first years of life (Vincent et al., 2001). Such findings indicate that when attempting to match leopard seals over extended periods of time, caution should be applied to avoid mismatching as well as missed matching individuals.

Identifying both scars and spots was easier when the individual was wet and as a result, quicker to map than when the animal was dry. When the animal was wet, occasionally the reflection of light was troublesome as it impeded identification, as was found for applying photo-ID to Saimaa ringed seals, *Phoca hispida saimensis* Nordquist, 1899 (Koivuniemi et al., 2016).

In the current study, it was challenging to observe some spots when the pelage was dry, as the pattern shape, size and density of the spots became less defined where the fur was raised. Conversely, the V-shaped scar, used as an orientation reference point, was at least as prominent when the individual was dry. We therefore recommend that where feasible, photographs of leopard seals that are both wet and dry are used for matching an individual to increase the likelihood of matching spots. However, if available, the use of images when the seal is wet may be more productive and yield more robust results when monitoring pigment development or matching for photo-ID. In our dry image from 2017 (Fig. 3) HLNZ-001 appears to be moulting. Stages of moult could provide an additional challenge because they create inconsistencies in pelage texture and that may mask the pigmentation pattern, as has been found in other pinnipeds (Cunningham, 2009).

We found that there were few spots forward of the eyes on this individual and a preliminary assessment of leopard seals randomly selected from the NZ Leopard Seal Database (held by LeopardSeals.org) suggests that this is the norm. Likewise spots on both the neck and pale areas along the flanks seem prevalent on all individuals who were preliminarily assessed, although the quantity varied.

Retrospectively, we found that Langley et al. (2021) used "*extractable areas*", comparable to our "zones", when assessing spot patterns on harbour seals, and that harbour seals had more spots on their faces (including on the forehead) than we found on any leopard seals we assessed. However, their methods may be applicable to leopard seals and we have begun to investigate the use of software for matching leopard seals (LeopardSeals.org, unpublished data).

Given that the tendency, when photographing wildlife, is to hone-in on the face of the animal (Excell, 2021), it is likely that images collected by citizen scientists would include the face and neck. If one was to utilize images collected by the public, as has been done for leopard seals in NZ (e.g., van der Linde et al., 2021), it would be logical to concentrate on this zone of the animal for spot-matching as this is likely to result in the highest number of available images. Other body zones (as well as scars) would still be of value for cross-matching and confirming identification.

Our investigation sought to explore if leopard seal spot patterns changed over time and we found that they did. However, we found that none disappeared and all ten white arrows (indicating areas of change documented in subsequent images) could be placed accurately using the surrounding 40 marked spots and unique acquired marks. Therefore, the mapping of spot patterns for effective long-term photo-ID, and the monitoring of any continued development of pigmentation density and area, is feasible.

We suggest that long-term studies of leopard seals (preferably from when they are as young as pups, but certainly once they are juveniles), may provide researchers with data as to when spotting is most likely to change e.g., as a general ageing process or perhaps linked to hormonal changes as the animal reaches sexual maturity. Regardless of the time-span between images, in order to monitor any potential changes in spots, we recommend that a minimum of 20 spots (as well as any other natural or anthropogenic scars and markings), are used for matching leopard seals. Given that opportunistically collected images, such as those from citizen scientists, rarely have the animal in the same position, this relatively high number of spots should elevate the probability of matching an individual. We conclude that although changes occurred, and no spots were lost over time, spot patterns were an effective tool for both individual identification and observing pigment change. Therefore, our findings do not invalidate photo-ID studies for leopard seals, rather they show that this is a robust system for identification of individuals that can be utilized over extended time frames.

Supplemental Material S-1

Comparison of 2017 images before (left column) and after (right column) application of software algorithms Photoshop 2022, TopazLabs Stabilize AI and Gigapixel AI. These were applied to improve photo quality (i.e., contrast, sharpness) and to produce images of the same size for better comparison. We could find no difference in the spot patterns between the originals and those with the algorithms applied.

However, we suggest caution when applying such machine learning tools, especially for any features that are low in pixel size or in an image with poor contrast, low light, or poorly focused, which could result in spots being 'created' or small features being removed (e.g., if the algorithms identify an actual spot as potential 'noise').

Pelage State	**ORIGINAL & CROPPED** (file names/size)	**ALGORITHMS APPLIED to CROPPED** (file names/size)
Dry	 20170810-NZE-INV-D1-108 (5.6 MB)  20170810-NZE-INV-D1-108 (cropped to head only) (166 kb)	 20170810-NZE-INV-D1-108 (6.5 MB)
Wet	 20170810-NZE-INV-D1-145 (4.6 MB)  20170810-NZE-INV-D1-145 (cropped to head only) (1.2 MB)	 20170810-NZE-INV-D1-145 (6.9 MB)

Table 1. Comparison of 2017 images before (left column) and after (right column) application of software algorithms Photoshop 2022, TopazLabs Stabilize AI and Gigapixel AI. All images by INV.

ACKNOWLEDGEMENTS

LeopardSeals.org continues to be grateful to those in the NZ community who provide sightings and photographs of leopard seals around our coastline, in particular of Owha (HLNZ-001) who has become an iconic poster child for this species, through her residency within an urban environment. We acknowledge Māori as tangata whenua and Treaty of Waitangi partners in Aotearoa New Zealand and thank Ngati Whatua ki Orakei for naming Owha and for their continued dedication to her protection. Thank you to Dr David Hocking who provided helpful feedback towards improving this paper. This work was conducted under permit numbers 63877-MAR (Visser) and 63499-MAR (van der Linde) issued by the NZ Government (Department of Conservation). Ingrid Visser thanks her Patreon supporters for their financial assistance during the preparation of this publication. We thank Dodoland, the Encounter Foundation and the National Institute of Water and Atmospheric Research (under the MBIE Endeavour Programme C01X1710 - Ross-RAMP - and MBIE NIWA SSIF) for their financial support of LeopardSeals.org.

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