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Lamination as a method of preserving reptile sloughs

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The sloughed skins of snakes and lizards can sometimes be found in reptiles' habitats. In Europe they are more commonly found during the spring after snakes and lizards have emerged from hibernation than at other times of the year (Bauwens et al., 1989; Nilson, 1980). These can then be used to assist in identification of winter hibernacula locations. Unfortunately sloughs tend to dry out quickly in warm climates and break down readily in the environment. This means that searching for sloughs at a known reptile site may increase the likelihood of encountering freshly sloughed skins. If collected early enough, they can be stored dry for later reference or used as a non-invasive sampling method to extract DNA (Jones et al., 2008).

A number of reptiles are often used as educational tools in outreach aimed at children. During these events, the children may come into close contact with the reptiles and may even get to hold the animals. There is a risk here of the handled reptiles infecting the children with salmonellosis, although this risk is only small. The use of antibacterial hand gel when children handle such animals is vital. We propose a method in which the sloughs of reptiles can be preserved, meaning they can be used as an up-close educational tool during such outreach events. The method of lamination means the slough can be handled without damage and without distorting any detail. This doesn't remove the need for reptiles at such events but instead adds another level of interaction and interest.

An inspection of the slough should be performed to ascertain if there is any damage and if present to what degree. If there are multiple sloughs to choose from then selecting a different one for lamination may be the best choice to ensure the optimum detail is captured and as little wastage of the laminate sheets is achieved. This visual inspection also allows for the selection of potential cut points as well as a visualisation of the final lamination. To prepare the slough for lamination it is advised that it is lightly misted with a spray bottle. If the slough has been in dry storage then leave the skin for a short while after misting (time varies on size, length and ambient room temperature). Ensure that the slough is only lightly rehydrated and isn't brittle or saturated.

Once the slough has been sufficiently rehydrated and is ready to be laminated, it should be cut to size. If the dorsal and ventral surfaces are desired to be fully preserved, then the slough can be cut longitudinally. At this stage a suitable pouch size should be selected, for most snakes

Figure 1. The laminated sloughs from a captive corn snake (*Pantherophis guttatus*).

we recommend A4 pouches. The prepared sections should then be placed on a hard surface such as an ironing board, with a tea towel placed on top of them. This step is taken in case the slough has become warped as part of the storage process. To avoid burning the slough during the ironing, lightly mist the tea towel and then pass a hot household iron over the slough until the tea towel is sufficiently ironed. Care should be taken not to apply too much pressure and ironing should replicate the care one would take with a silk shirt. This not only protects the delicate skin beneath but also acts as an indicator. The process works to both flatten the skin ready for lamination and to fix it into position. The process of ironing the slough sections may stretch them slightly.

Depending on the size and colour of trimmed sloughs, they can either be placed directly into an A4 lamination pouch (for larger and darker sloughs) or be glued onto a sheet of A4 paper. Being glued onto the a sheet of paper

prevents the sloughs from slipping during the lamination process and allows a greater level of clarity of any patterns that may be present on the slough. This is particularly helpful if a slough is light in colour and any patterns present are desired to be displayed. Multiple cut sloughs sections can be fitted within a single lamination pouch; printed labels can also be added before the final process of lamination. If preferred these can be added afterwards with a permanent marker pen. Once the pouch is full, run through a laminator as normal. There is an alternate method to our own which requires more skill than needed for the process described. After the visual inspection, a plastic-backed paper sheet is prepared for the slough to be applied. The backing paper recommended is Super Loxol which is usually used as a freezer wrap. It is then moistened and the slough is applied with liberal amounts of water. When the slough can absorb no more water it is rolled with a heavy instrument to remove the excess water before a cotton towel is used to remove any additional moisture. The slough is then left to dry and the end result is very similar to our own, without the need for adhesives or lamination. For a more detailed methodology please see Kaleta (1979).

The lamination of sloughs may also be a useful tool for a population based study where sloughs of individuals are sometimes found. This could work well for species such as the northern adder (*Vipera berus*) as individuals of the species have unique scale patterns (Sheldon & Bradley, 1989). This novel technique could over time add richness to a population study by way of having a visual database and associated DNA profile of an individual without having to encountering it. The proportion of sloughs compared with animals present is likely to be low and so should only be restricted to a minor role compared with visual surveys. Lamination will likely denature the DNA contained within the laminated sections of the slough and so part of the discarded or excess trim could be used for DNA extraction. This is an area which could be explored further in the future and would work well if partnered with a photographic census of a known population.

Due to the size limitations of standard laminate sheets only smaller skins can be fully laminated, for larger skins a selection of sections can be made to ensure that any identifying features such as dorsal head scales are preserved. Fig. 1 shows three sections of which the left and right have been orientated so that the dorsal surfaces are facing upwards but the centre section is orientated so that the ventral surface faces upwards. Dorsal patterns on either the head or body are easier to capture than those of ventral markings, at least in our limited experimentation with the sloughs of captive corn snakes (Pantherophis guttatus).

Our preferred use for the laminated sloughs is as an educational tool, to educate people about squamates and their life histories. These are often used in outreach events to add another level of intrigue although they are not a substitute for live animals.. Another use for the lamination of sloughs could be as a training aid, helping to train individuals in the identification of varying scalation types across a number of species. Being able to recognise morphological features such as scale groups can be used in identification of visually similar species. We feel that this technique is of so much value we will ensure that each classroom that we exhibit for have at least one lamination for future reference, this we hope will serve to lessen the fear associated with snakes that so many people seem to have and change the public's view towards reptiles.

The method of preservation works better on the sloughs from captive animals which tend to be in a single piece and are encountered more often than those of wild animals. Lamination may not work well for sloughs which are fragmentary to overcome this; they can be prepared by being glued to sheets of paper of the appropriate size. This method is ensures that the sloughs can be handled and studied far past the lifetime of an unpreserved slough.

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