ARE DEHYDRATED SPECIMENS A LOST CAUSE? A CASE STUDY TO RECLAIM DEHYDRATED FLUID-PRESERVED SPECIMENS

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Abstract.—Fluid-preserved specimens in collections persist only as long as their preservative is maintained. When preservatives evaporate due to neglect or container malfunction, collection managers are often forced to discard the specimens. Subjecting specimens to a rehydration process can be both time consuming and hazardous. A recent development in vertebrate specimen rehydration that mitigates these hazards and is relatively simple to conduct is discussed. Through the use of concentrated water vapor, and gradual staging in various concentrations of preservative, dehydrated museum specimens can be rehydrated. Similar techniques have been applied to invertebrates for decades, and more recently to herpetofauna. Herein a new technique is applied to both fishes and mammals and its efficacy for most other groups is indicated.

Introduction

Curation and collections management require attention to detail and endless vigilance. As stewards of natural history collections it is important to have a keen eye and a particularly meticulous nature. Nothing is more disheartening to a collection manager than discovering a jar on a shelf from which all of the alcohol has evaporated leaving its contents desiccated and apparently ruined. Keeping specimens hydrated is crucial in preventing damage (through changes in internal and external morphology), stopping microorganism growth, and keeping specimens malleable for use in subsequent research.

The use of caustic or hazardous chemicals such as potassium hydroxide, ethylene glycol, trisodium phosphate, and lactic acid have all been used with marginal results that often damage the tissues of specimens (Jeppesen 1988). Certain techniques work for some taxa but not others, leaving no room for a "standard" technique (van Cleave and Ross 1947). Recently, safer techniques have been developed by collection managers John Simmons and Simon Moore that have successfully returned integrity to vertebrate specimens (Moore 1999, Simmons pers. comm.). Such techniques use deionized water vapor (Moore 1999 via Jeppesen 1988) or Decon 90® (Banks et al. 1972, Simon Moore via Waterhouse and Graner 2009). The technique described herein developed by Simmons (1987 and 1991) has been previously used only on herpetofauna (Simmons pers. comm.). The intention of the following study was to review the applicability of this method across several fluid-preserved taxa.

Materials and Methods

All specimen measurements were taken as either standard length (SL) for fish or total length (TL) for mammals. The subjects of this rehydration included five specimens of ceratioid anglerfishes (*Himantolophus spp*. 21.06–35.06 mm SL: UF 40805, UF 23788, UF 23789, and UF 40805, where UF refers to the Florida Museum of Natural History). The specimens had originally been fixed in 10% formalin and then stored in 70% ethanol. These specimens had been sealed in plastic bags for several years in anticipation of being sent to Florida Museum of Natural History (UF) as returned loan material, but were forgotten. Eventually the alcohol evaporated, and they became desiccated. Due to the

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rarity of these species in collections and the fact that some of these specimens were types, an effort was made to rehydrate all of them. In addition, shrews (*Cryptotis parva.* 55.30–56.76 mm TL: UF 25148, UF 16277) were rehydrated. Two specimens were obtained from a collection made by the US Fish and Wildlife Service in the early 1980s. The specimens were improperly preserved and then placed in substandard containers (glass food containers) with 70% ethanol for storage; eventually the alcohol evaporated, which resulted in the dehydration of the specimens. The specimens had been desiccated for over 10 years before being donated to the museum. One specimen was subjected to the same rehydration process as the anglerfish specimens, and for comparison one was left dehydrated.

This study followed the procedure described by Simmons (pers. comm.). The process involved the use of deionized (DI) water in a sealed container to create a high humidity environment in which the dehydrated specimens could absorb water vapor. DI water was used to reduce the presence of impurities (mainly salts). The dried specimens were placed on a perforated rubber platform (Fig. 1) situated just above the water for several days until improvement was observed. Following this treatment they were placed into pure DI water. They were then staged through increasing concentrations of ethanol by increments of 20% for several days until the desired alcohol concentration was obtained. The materials used in this type of rehydration are easy to procure and should be present in most collections.

Materials

The following materials were used:

- gal. (1 gallon=3.79 liters) wide-mouthed jar (or similar large container) with a tightly sealing lid
- Thymol crystals (antifungal agent) (care should be used in the handling and disposal of this substance because it has a long residence time in water)
- Staging platform for specimens to rest above liquid. This can be any material not affected by high humidity and can rest either above or in liquid for an extended period of time. A 12.7×12.7 cm perforated rubber square was used in this treatment.
- DI water (H_2O) (enough to fill containers adequately)
- Ethanol diluted to varying concentrations (e.g., 20%, 40%, and 60%) with DI water.

Methods

The process began with the addition of room temperature DI water to the gallon jar. Warm water may be used, as it seemed to have quicker results. Cold or boiling water is not recommended because cold water will take longer to evaporate and boiling water may damage the specimens. To prevent the growth of microorganisms, a few thymol crystals were added to the DI water. These can be predissolved or partially dissolved in a small amount (10 ml or so) of DI water. The platform was placed in the water such that the specimens rested on the platform suspended just above (but not touching) the water. The jar was then sealed to prevent the evaporation of the DI water and to facilitate the formation of humidity. Condensation formed almost immediately. The specimens were left this way until they appeared to be moist and pliable. This was visibly achieved when limbs, skin, or hair appear to be moist and can be easily manipulated. This was most noticeable in the extremities. For example, the fish had dry, brittle fins when dehydrated, but the fins appeared to be normal once rehydrated. The water was changed as needed (e.g., if discoloration or cloudiness formed). This process took anywhere from one week to a month depending on the size of the organism. Larger specimens took more time on

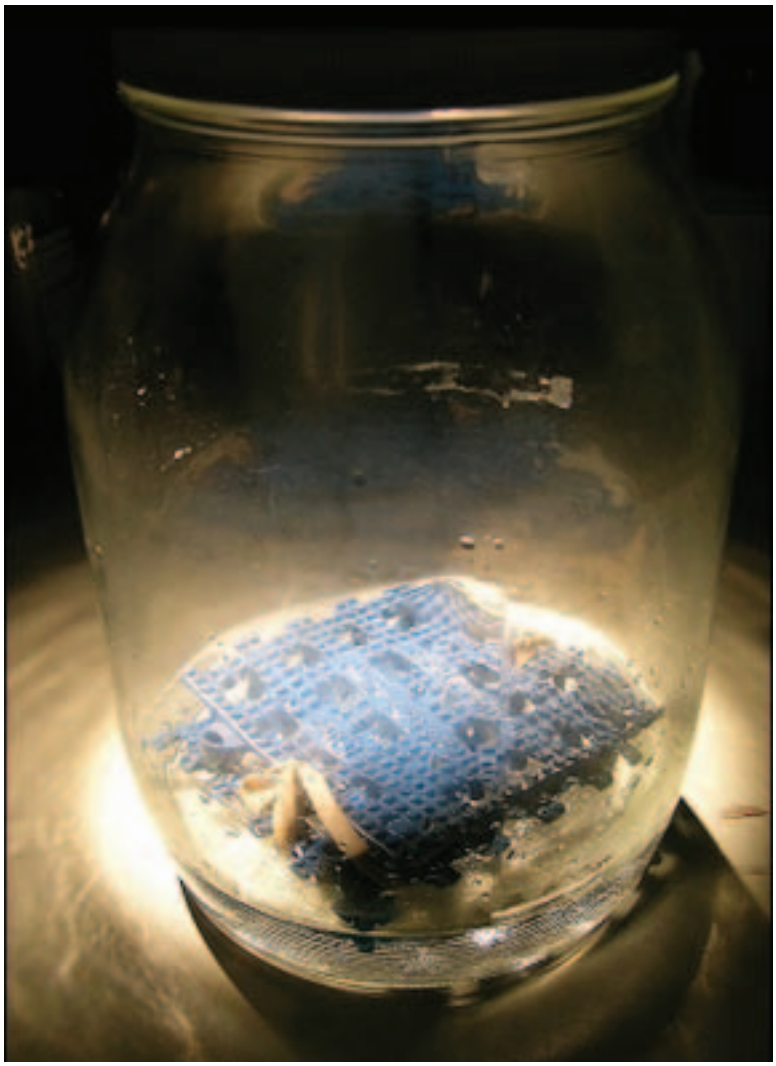

Figure 1. Rehydration chamber with rubber staging platform. Evaporated DI water can be observed in a fog layer above the rubber platform.

average. Once moist, the specimens were removed from the chamber and placed into 100% DI water. After 1 to 2 days, the water became cloudy or brown and was changed as needed. The specimens were then staged through increasing concentrations of ethanol, going from pure DI water to 60% ethanol, in increments of 20%. More delicate specimens may require increments of 10% to avoid dehydration. The specimen remained in each step for no more than 2 days. Once sufficiently rehydrated, the specimens were placed in the desired concentration of preservative (70% ethanol). The total process took on average 20 days to successfully rehydrate the specimens.

Results

The results observed were successful with both taxa. Changes in relative hydration were noticed 5 days after they were placed in the DI ''hydration chamber.'' The

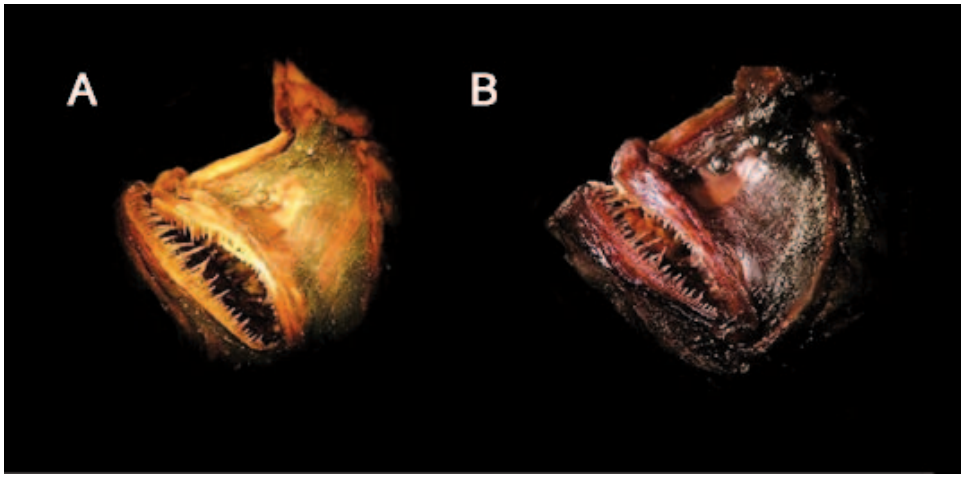

Figure 2. UF 23790 *Himantolophus paucifilosus* (35.1 mm SL) before (A) and after (B) rehydration.

anglerfishes changed from material unusable for research to perfectly acceptable specimens within the span of 1 month (Fig. 2). Change was particularly noticeable in the softer areas of the anatomy (e.g., eyes, gill filaments, maxillary tissue, fin rays). The specimens had some damage from dehydration to their fins (e.g., broken rays, tears), and the overall texture was not as smooth as that of an undamaged specimen. Although sufficiently rehydrated, the specimen was still more ridged than a specimen that had not experienced desiccation. The entire process took 20 days.

The shrew specimens were originally white due to fungal growth, and over the course of the treatment the natural black color returned (Fig. 3). The snout, feet, and tail became pink 4 days after being exposed to the water vapor. Also, the texture of the fur became softer. The After 6 days the nares opened, and the flesh on the limbs became engorged. The specimen also became much more pliable. The entire process took 21 days.

Discussion

The process reversed some damage due to dehydration, rendering the specimens suitable for use in research. Particular success was noted in the shrews. Before

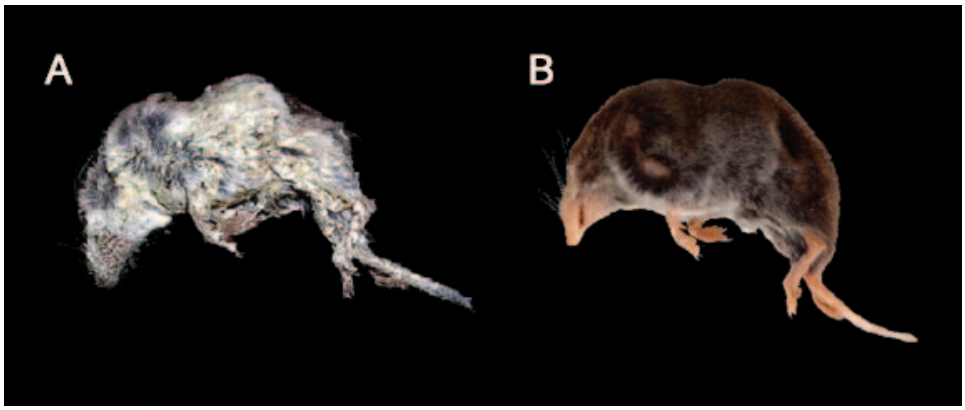

Figure 3. UF 25148 *Cryptotis parva.* (55.30 mm TL) before (A) and after (B) rehydration.

rehydration, the identification of the specimen was not possible due to changes to internal and external morphology brought on by desiccation and extreme brittleness. Rehydration is crucial in examining specimens that require meticulous inspection or manipulation (such as examination of dentition in angler fishes and shrews). Overall, this process was observed to be effective in the rehydration of dehydrated fluid–preserved fish and mammals. All specimens were returned to an acceptable state of preservation.

This process can be useful for recovering valuable specimens that have been improperly curated as is often seen in orphaned and abandoned collections. In addition, it can be valuable to large collections where the status of each jar cannot be individually monitored and problem lots or faulty jars may be overlooked leading to specimen desiccation. The applications are extensive. This method is extremely simple and can be conducted by any collection worker with little supervision. One main consideration is allowing for a block of uninterrupted time to monitor the specimen. The materials used in the process are cost effective and nonhazardous. It is well known that DNA extraction from specimens exposed to formalin is rarely possible. By not using caustic chemicals, this technique can possibly be applied to low alcohol preserved, non-formalin-fixed specimens such as vertebrate specimens preserved in 95% ethanol. DNA could potentially be extracted from rehydrated specimens such as marine invertebrates and insects that, while dehydrated, prove difficult to extract viable tissue samples. This remains to be examined further.

A small sample size was used for this study, and future studies should consider the use of multiple specimens of the same taxa of different sizes. Several replications using a random dehydrated control group could be used to quantify the success of rehydration beyond the initial visible results. In addition, it is important to document the success of the rehydration over a longer timescale. The preliminary work done using this method by Simmons (2002, pers. comm.) will allow collection managers and technicians to save a number of specimens that would have previously been discarded or left dehydrated for lack of a reliable method of recovering desiccated specimens.

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