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Silicone Plastination of a Malpositioned Long-term Formalin-fixed Green Iguana

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Abstract: Plastination of a malpositioned, long term-formalin fixed green iguana *(iguana iguana)* was performed to attempt a repositioning of the specimen once impregnated with silicone. The impregnated specimen was able to be brought into proper anatomical position during the curing period. The iguana was preserved without noticeable shrinkage and appeared very natural. The resulting specimen is now a nice representation of its genus.

Key words: **plastination; silicone; S10; formalin; iguana; reptile**

Introduction

The green iguana *(iguana iguana)* is the most common pet lizard whose popularity as a domestic pet is increasing. Caring for a green iguana is difficult because they have special husbandry considerations such as appropriate humidity, temperature and diet. Iguanas often suffer from metabolic disease due to inadequate husbandry. Preservation of green iguanas using plastination is an excellent method for preparation and storage of a reference collection for and indefinite period.

Often, iguana specimens which have been improperly positioned before fixation with preservatives are donated to herpetological collections. Formalin fixed biological tissues are not very pliable and are resistant to repositioning. Specimens in extreme states of distortion tend to make poor examples of living animals and validates the desirability to prepare specimens appropriately before fixation (Henry et al., 1997).

The iguana cadaver selected for this study had been formalin fixed in a contorted position for a period of more than three years. Its head was ventro-flexed such that it was positioned below and between its forelimbs. The iguana's trunk was wrapped with its tail. The dorsal spinous scales were flattened against the trunk. The challenge was to correct the position of iguana after impregnation with silicone before the curing stage to assess if silicone impregnated-formalin fixed tissue is more pliable to bending and repositioning without disruption of the normal anatomy of the biological specimen.

This work describes the complete plastination and repositioning procedure of a green iguana carried out in the Department of Anatomy, Faculty of Veterinary Medicine in Skopje, Macedonia.

Materials and methods

Fixation

The green iguana was obtained from the Main Veterinary Hospital in Skopje. The fresh specimen was fixed by immersion in 3% formaldehyde solution and was kept in this solution for more than three years. A midventral incision of abdominal wall that had been made previously during an operation had been closed with suture. This port provided entrance for fixative solution into the body cavity and hence good preservation of the tissues.

After this extended period of fixation, the iguana was rinsed with cold tap water for one week to remove the majority of formalin. Before dehydration, the iguana was cooled to 5°C. Table 1 lists the stages of plastination, their durations and the temperature under which the stages occurred.

Step	Time	Temperature
Fixation	>3 years	room temperature
Flushing	1 week	10°C
Cooling	24 hours	5°C
Dehydration I	31 days	-25°C
Defatting/ Dehydration II	5 days	room temperature
Immersion	7 days	-20°C
Forced impregnation	3 weeks	-20°C
Pre-curing	5 days	room temperature
Gas-curing	12 days	room temperature
Post-curing	>3 months	room temperature

Table 1. Steps and timetable of iguana plastination.

Dehydration

The iguana was dehydrated using cold acetone (freeze substitution method) (Tiedemann and Ivic-Matijas, 1988; Weiglein, 1997). The iguana was submerged in the first acetone bath of 100% acetone for two weeks at -25°C. The second acetone bath (100% acetone) was for ten days at -25°C. Finally, the iguana was transferred to the third 100% acetone bath for seven days at -25°C. The purity of each acetone bath was monitored after warming to 20°C using an appropriately calibrated acetonometer. When the acetone purity of the third bath remained over 98%, the iguana was transferred to a fourth acetone bath (100%) at room temperature for five days for defatting and to complete dehydration. The acetone concentration of the acetone following the last acetone bath measured 99.5% at which point dehydration was considered complete.

Forced impregnation

Forced impregnation was by continuous impregnation (von Hagens, 1986; Henry and Nel, 1993). However, at the end of each working day, the vacuum pump was turned off and the vacuum sealed such that the pressure inside the vacuum chamber remained constant. The iguana was submerged in a mixture of silicone polymer (S10) and catalyst/chain extender (S3) combined at a ratio of 100:0.5 for one week at -20°C. After one week of immersion, vacuum was applied for three weeks and pressure was slowly decreased to 3mbar. Vacuum was controlled via a vacuum controller and monitored via formation of acetone bubbles at the surface of polymer. The impregnation process was considered complete when bubbles ceased and the pressure stabilized at 3mbar. During the next three days, the pressure was slowly increased to atmospheric pressure. The iguana was then placed at room temperature and left submerged in silicone for an additional day.

Figure 1. Final cosmetic repositioning using nails, tacks and towels of silicone impregnated green iguana prior to curing.

Figure 2. Gas curing of green iguana. S6 was introduced via tube into the iguana's body cavity.

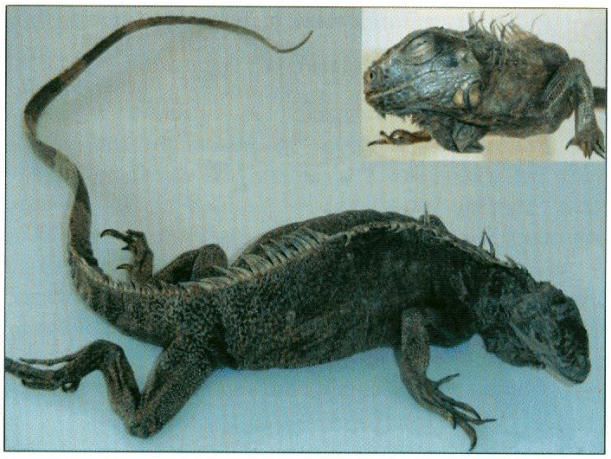

Figure 3. Views of plastinated green iguana.

Curing

In preparation for curing, the iguana was removed from the silicone and placed on a grid to drain excess

polymer from its surface for five days at room temperature. Following draining, the iguana was posed in a desirable position using large nails and a supporting board to fix the tail, legs and head (Fig. 1).

To keep the spines of the back and tail upright, towels were placed adjacent to the spines and fixed by pins to the body of iguana to support the spines. Little pieces of towel were placed between the eyelids to keep the eyes open. Finally, the fast gas curing method was used for curing the specimen.

The specimen was placed in a gas curing chamber and exposed to S6 vapors for five days at room temperature. To prevent white precipitate from forming on the surface of the iguana, $CaCl_2$ was added to the gas curing chamber. A small membrane pump was used to bubble air through the S6 to accelerate the curing of the surface of the iguana. After five days, the surface of the iguana was dry (cured) but the silicone inside the body wasn't polymerized. Therefore, a tube with attached needle was attached to the S6 vapor bottle and inserted into the body cavity of the iguana (Fig. 2). S6 vapor was introduced into the abdomen for seven days. Following gas curing, the iguana was stored in a sealed plastic bag for three months to assure curing of the entire specimen.

Results

The green iguana was preserved well with the slightly altered S10 standard plastination technique. A lower concentration of S3 was chosen in an attempt to obtain a more flexible specimen. No noticeable shrinkage was observed in the plastinated iguana. The position into which the iguana was placed was retained following the removal of the specimen from the board (Fig. 3).

Discussion

The plastinated iguana looked natural. The color of the skin was darker than usual but was likely due to the very long period of fixation in formalin. Slow increases of vacuum were used to prevent shrinkage of the iguana. When the vacuum pump was turned off at the end of the day, the vacuum was sealed and held constant inside the vacuum chamber. This also helped to prevent shrinkage of the specimen. Repositioning the iguana prior to the curing stage was the crucial step in plastination of the iguana because the impregnated, twisted specimen needed to be manipulated and fastened to a retaining board. Following impregnation, the twisted shape of the iguana was reformed even after having been fixed for three years in an inappropriate position. This suggests that poorly shaped formalin fixed specimens can be reshaped with proper impregnation and gentle manipulation.

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