INTRODUCTION

Curing of the S10 polymer presents special challenges. The polymer reaction mixture of S10 + S3 (the impregnation bath) must remain liquid until the specimen is impregnated and preferably longer so that the polymer may be utilized to impregnate more specimens. However, it is desirable for the final curing to occur rapidly within the specimen once the specimen is removed from the plastination kettle to prevent polymer loss from the specimen. Initially rapid cure was achieved by immersing the impregnated specimen into a liquid curing bath. This was messy and not totally acceptable as uncured surface polymer would be dissolved from the specimen. In 1981, gas curing was introduced and is technically a much easier method to achieve polymerization of the polymer in the impregnated specimen (von Hagens, 1984).

After the specimen has been impregnated with the reaction mixture (S10-S3), the polymer must be hardened to keep the polymer inside the specimen and to obtain a durable preparation. First, the excess polymer is allowed to drain from the specimen and the collected polymer is returned and stored in the cold plastination kettle. Curing of the polymer in the impregnated specimen is accomplished using two curing agents: 1. BIODUR S3 and 2. BIODUR S6 known as Gas Cure (von Hagens, 1984; 1985). Curing or polymerization of the polymer begins when the silicone polymer (BIODUR S10) and hardener (BIODUR S3) are mixed together. However, cold temperature «0°C) retards polymerization of the reaction mixture so that the reaction mixture remains fluid and impregnation can take place. The S3 commences end to end-linkage and hence elongation of the silicone molecules, which produces increased viscosity of the reaction mixture. This linkage is reported to enhance flexibility of the impregnated specimen (von Hagens, 1984; 1985; 1986; Bickley, 1987). Theoretically, after many months at room temperature, the reaction mixture in the impregnated specimen should polymerize. However, experientially after impregnation, the specimen will continue to ooze for many months when left at room temperature. Therefore, the second curing agent (BIODUR S6, Gas Cure) is necessary to complete the polymerization and to assure a durable product.

BIODUR S6 (Gas Cure) is a liquid containing silicate with a high saturated vapor pressure (boiling point 70 - 80°C). To react with the polymer of the impregnated specimen, the S6 must vaporize and the active vapor reacts with the silicone on the surface of the specimen. S6 commences a three-dimensional cross-linking (side to side-linkage) of the molecules and hence imparts toughness or firmness to the specimen (von Hagens, 1984). The surface molecules cure first, thus sealing the surface of the specimen and decreasing oozing of polymer from the specimen. Over a period of time, the S6 diffuses into the specimen and polymerization proceeds deeper, toward the center of the specimen until the specimen is totally cured.

Two general categories of curing have been described and used over the years (von Hagens, 1984; 1985; 1986; Bickley,
1987):  1. Slow Cure and  2. Fast Cure. Both the slow and the fast curing method utilize both of the hardening agents [Biodur S3 and S6 (Gas Cure)].

SLOW CURE was the original curing method for the S10 procedure. It is based on the premise that the specimen will be kept at room temperature for several weeks prior to exposure to the final curing agent (S6). Slow cure has two segments:  
1. Precuring and 2. Gas curing. The initial period of time with the specimen kept at room temperature is called precuring. Hence, precuring can be defined as the time interval which allows the S3 component of the reaction mixture to propagate and maximize end-to-end-linkage of the silicone molecules in the impregnated specimen. Hence, the reaction mixture in the specimen becomes very viscous before exposure to the second curing agent, S6. In the past, after a few weeks of room temperature, an increase in temperature to 40 - 50°C was suggested to enhance the end to end-linkage of the silicone molecules. After a prescribed period of time at room temperature and/or oven temperature, the reaction mixture will thicken enough so that the specimen feels tacky. Precuring is finished when the specimen feels tacky, then it is ready to be exposed to the second curing agent, S6. In the past, after a few weeks of room temperature, an increase in temperature to 40 - 50°C was suggested to enhance the end to end-linkage of the silicone molecules. After a prescribed period of time at room temperature and/or oven temperature, the reaction mixture will thicken enough so that the specimen feels tacky. Precuring is finished when the specimen feels tacky, then it is ready to be exposed to the second curing agent, S6. To prevent oozing polymer from hardening on the surface of the specimen, the specimen must be manicured or wiped off frequently during the first 24 to 48 hours of gas curing and then as needed. The final step is final cure. Final curing is the period of time necessary for the S6 vapor to diffuse throughout the depths of the specimen to complete polymerization. This is accomplished by sealing the specimen, which is no longer oozing, in a container or plastic bag with a desiccant. Length of final cure is dependent on the thickness of the specimen. Thicker specimens require more time than do thin specimens.

USAGE of S6

S6 or Gas Cure should be used in a closed environment (gas chamber). A medium to large shallow container (dish) will provide a large surface area for maximal vaporization of the S6. A high vapor concentration will enhance polymerization and hence seal the surface of the specimen and prevent oozing of the polymer. To minimize atmospheric contamination with S6 and decrease the quantity of S6 used, only small volumes (10 - 100 ml) of S6 are necessary. A desiccant should be present in the chamber to absorb excess moisture. The specimens should be placed on a grid and/or absorbent
toweling to permit excess polymer to drip free from the specimen or be absorbed from the surface.

Vaporization of S6 will occur at room temperature. However, vaporization can be enhanced by bubbling air through an aliquot of S6 (von Hagens, 1986; Henry, 1987; Oostrom, 1987; Tiedemann, 1988). A membrane (aquarium) pump is inexpensive, and will supply adequate air flow for vaporization. 20 to 30 ml of S6 is generally an adequate volume of S6 to produce sufficient vapor for an average size gas chamber, and 5 to 10 minutes of aeration is sufficient time to vaporize the necessary quantity of S6. A narrow container may be used and will provide adequate aeration and hence vaporization of the S6. As S6 vaporizes, a white precipitate remains. Hence, using a smaller volume of S6, at a given time, produces less precipitate and hence less contamination of the S6 container.

Because S6 is volatile, it is wise to have the gas chamber in a well-ventilated area or near a fume hood. Wearing a gas mask is prudent.

When the specimen is no longer oozing polymer, it may be removed from the gas chamber and used. It usually takes 2 to 4 days of intense exposure via vaporization with a membrane pump or 4 to 8 days of self vaporization to seal the surface. However, beware, the center of the specimen is not cured since polymerization commences on the surface and proceeds to the center. Therefore, for a few weeks it maybe necessary to store these specimens in a closed container or plastic bag to assure that the S6 penetrates the depths of the specimen.

The final step of S6 usage is to allow any remaining S6 to evaporate from the specimen. Simply, lay the cured specimen out in the atmosphere for a few days until no S6 odor is detected. It is important to rid the specimen of excess S6, as any excess may cause a white precipitate to appear on the surface of the specimen. If, at a later date, polymer oozes to the surface of the specimen, repeat exposure to the S6 vapor in the gas chamber for a few days.

The more common curing method used today is fast cure. Both curing methods have potential problems and/or disadvantages. The following may occur during or after gas curing: 1. A white precipitate may appear on the specimen. 2. The specimen may shrink. 3. Oozing polymer may coat the specimen.

White precipitations are caused by:
1. Specimen contact with water.
2. Specimen contact with the gas cure fluid.
3. Too long of exposure to the gas cure vapor (von Hagens, 1986). To avoid precipitations:
   1. Use a desiccant, e.g. calcium chloride, especially with fast curing.
   2. Pour the fluid gas cure into the dish and then place the specimens into the gas chamber.
   3. Use slow curing, because precipitates hardly ever form.
   4. Decrease exposure time to the S6 vapor and/or allow the excess S6 to evaporate from the cured specimen.

Shrinkage is caused by polymer oozing out of the specimen (von Hagens, 1986). To avoid shrinkage:
1. Smear cut surfaces with S49 or old viscous S10/S3-solution.
2. Place cut surfaces of the specimen to the top and/or turn over frequently.
3. Use fast curing especially when you have cut surfaces, slices, or soft tissue like fat.
4. Wrap the specimen with thin foil which will adhere to the surface of the specimen.
5. Do not use heat during procure.
6. Use slow cure only on specimens which have been formalin-fixed for a prolonged period.

Curing of hollow organs requires special techniques to obtain the best specimen. For either slow or fast cure, hollow organs should be inflated with air (Henry, 1987; Tiedemann, 1988; Holladay, 1989) or stuffed with paper toweling. Inflation via an air source is done by cannulating a natural opening or suturing a cannula into an incision. Close all other openings with cork or suture to minimize air leakage. To precure, simply allow the appropriate air flow dilate the organ to the desired volume and maintain that flow for a few days or for several weeks. Place the organ in a gas chamber or in a plastic bag to serve as a gas chamber. Insert a S6 source in the air line and bubble air through a small quantity of S6 (10 - 20 ml) to produce S6 vapor.
which is channeled into the hollow organ. Each day or twice a day 10 - 20 ml of S6 may be added to the system until the organ is cured. As an alternative, towelining may be placed inside hollow organs to cure them in an expanded state (Henry, 1987; Oostrom, 1987).

Curing of cut sections offers a unique challenge, to keep the polymer in the slice. Gravity allows the polymer to run out and turning the specimens frequently decreases polymer loss (von Hagens, 1986). An alternative is to start curing in a gas chamber placed in the deep freezer. Cold polymer is more viscous and hence will drain out less. The specimens are placed in the cold gas chamber and exposed to S6. The S6 must be vaporized via a membrane pump to provide adequate vapor. After at least one week of cold curing, the specimens are brought to the room temperature gas chamber and exposed to room temperature rapid vaporized S6. Hardening of the surface polymer usually takes place in 12 to 24 hours.

Remember, once the specimen is cured it will remain in that position for the rest of time. Therefore, imagine how that you can make the specimen look its best before you harden it.

For other special curing techniques, especially for curing and grinding body slices, and unique problems, please refer to the "Plastination Folder", von Hagens, 1985 and 1986.

REFERENCES


