The Evolution of Sodium Hypochlorite as a Useful Antiseptic

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The discovery of sodium hypochlorite is attributed to Berthollet in 1788. The history of this agent includes the names of Labarraque and Javel as well as the more familiar Dakin. The books by Carrel and Dehelly (*1*) and Dakin and Dunham (*2*) provide interesting background about this useful class of compounds.

The drug standards compendia had monographs for Sodium Hypochlorite Solution and Diluted Sodium Hypochlorite Solution until 1975 (*NF XIV*). Since that time there has not been an official monograph for the diluted solution. Presently, a monograph for Sodium Hypochlorite Solution is found in *USP 23*. However, diluted sodium hypochlorite solutions of different compositions are being used as antiseptics.

One of the monographs proposed for introduction as a solution that could be compounded by a pharmacist pursuant to a prescription is a sodium hypochlorite solution. The practice of extemporaneously preparing an article is appropriate when the article is not commercially available or when stability is a concern.

Sodium hypochlorite solutions are valuable antiseptics that must be used with great care. Sodium Hypochlorite Solution USP is a 4.0–6.0% solution of sodium hypochlorite in water and carries the caveat, *Caution—This solution is not suitable for application to wounds*. The pH of this solution is greater than 11 because the hypochlorite ion is very basic, as is shown in equation (1).

$$ClO^- + H_2O = HClO + OH^- \quad (1)$$

Diluted Sodium Hypochlorite Solution *NF XIV* is a solution that contains not less than 450.0 mg and not more than 500.0 mg of NaClO in each 100 mL. There is no admonition about use on wounds. The procedure for preparation of the solution includes use of sodium bicarbonate to buffer the pH to less alkaline values with phenolphthalein as an external acid-base indicator. Unfortunately, the use of any oxidizable compound in the presence of hypochlorite is liable to bring about oxidation by the hypochlorite. This happens to phenolphthalein, thus making its indicator function unreliable (*3*).

Some recent papers raised doubts about the acceptability of sodium hypochlorite solutions as regards safety toward tissues that are attempting to heal. A study by Cotter et al. (*4*) reports that a 0.1% NaClO solution, buffered with sodium dihydrogen phosphate and sodium monohydrogen phosphate to a pH of 7.4 and having a concentration of solutes such that the osmolality is 386 milliosmoles per liter, does not have a significant effect on basal cell viability. Lineaweaver et al. (*5*) make no mention of the chemical or physical properties of the solution used but report that a 0.005% NaClO solution was the most concentrated that could be used that avoided undesirable effects on fibroblasts while still retaining antibacterial activity. These findings introduce uncertainty as to the best composition for a sodium hypochlorite solution that would be suitable for use on wounds.

In this study, work was carried out that involved preparation of sodium hypochlorite solutions of varying concentrations with pH and osmolality that are close to physiological values (pH 7.4, ~300 mOsm/liter) and included the evaluation of important solution attributes. The stability of the solutions was determined by measuring the hypochlorite concentration as a function of time using a stability-indicating titration of the iodine released when potassium iodide is added to hypochlorite under acidic conditions, and may be summarized by equations (2) and (3). Starch is used as the indicator.

$$ClO^- + 2I^- + 2H_3O^+ = Cl^- + I_2 + 3H_2O \quad (2)$$

$$I_2 + 2S_2O_3^{2-} = 2I^- + S_4O_6^{2-} \quad (3)$$

Since both osmolality and pH remained constant during the times of the stability study, neither property was stability-indicating.

Important biological properties of the solutions were determined. Each solution was evaluated as to antimicrobial activity (*S. aureus, E. coli*) and to antiviral activity (Poliovirus Type 1). In addition, the effect of the solutions on tissue cultures of primary and low pass cultured human fibroblasts and HL-60 cells was observed. The fibroblasts were chosen because they are the primary cell type responsible for tissue remodeling and wound healing. HL-60 cells are a model for both neutrophils and monocytes, primary inflammatory cells observed in wounds. The studies started with 0.10% sodium hypochlorite solution, but stability of this solution was not satisfactory because only 87% of the original sodium hypochlorite remained after 3 days at controlled room temperature. The studies continued with 0.05%, 0.025%, and 0.005% sodium hypochlorite solutions. The results of this work are summarized in Table 1.

From the properties reported in Table 1, the 0.025% sodium hypochlorite solution is judged to have the most desirable properties. The solution is stable to the extent that a beyond-use date of 7 days is proper. The biological testing indicates that the solution possesses antimicrobial and antiviral action and that cells exposed to the solution were able to recover.

The Topical Sodium Hypochlorite Solution, 0.025%, monograph appearing at this time is based on the results of these studies. The pharmacist who compounds this solution is reminded respectfully that measurements of solution volumes with a proper volumetric apparatus and determination of weights with a balance of sufficient accuracy, together with proper technique, is essential if a solution that meets the requirements of the monograph specifications is to result.

If the solution is compounded properly, the beyond-use date supplied in the monograph will be correct and will be included on the label.

The laboratory procedure for the assay of the 0.025% solution is the following.

Assay—Measure accurately, into a glass-stoppered flask, exactly 50.00 mL of Solution. Add 0.5 g of potassium iodide and 10 mL of 6 *N* acetic acid, and titrate the liberated iodine with 0.1 *N* sodium thiosulfate solution VS, adding 2 mL of starch TS as the endpoint is approached. Perform a blank determination, and make any necessary correction. Each mL of 0.1 *N* sodium thiosulfate is equivalent to 3.722 mg of NaClO.

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Table 1. Summary of Diluted Sodium Hypochlorite Results.

Strength, pH, Osmolality[a]	Chemical Stability[b]	Antimicrobial Activity[c]	Antiviral Activity[d]	Tissue Culture
0.05, 7.1, 300	5	<4.5, <4.5	+	Attached fibroblasts survived up to 5 minutes contact, but did not grow and died within one week.
0.025, 7.0, 300	NA	<4.8, <4.8	+	After 1 to 2 minutes contact, attached fibroblasts maintained a confluent monolayer but did not increase in number; after 5 or more minutes the cells suffered a loss in viability and growth potential.
0.025, 7.5, 300	NA	<4.8, <4.8	+	Most lethal; attached fibroblasts would not grow after 1 minute contact, extensive cell loss five days later.
0.025, 8.0, 300	>14	11.5, 10.4	+	After 1 to 2 minutes contact, attached fibroblasts recovered immediately; after 5 to 15 minutes they were tattered but recovered in 24 hours; after 1 hour they were less than 50% confluent but recovered within 5 days.
0.005, 7.1, 300	>17	51.9, 21.5	+	Attached fibroblasts survived up to 60 minutes contact, but did not grow and died within one week.

[a] Nominal sodium hypochlorite content in percentage (w/v), nominal pH, and nominal osmolality (mOsm/kg).
[b] Time (days) required for the sodium hypochlorite content to decrease to 90% of its original value at controlled room temperature. NA = not studied.
[c] D-value: time (sec) for 90% reduction in microbial concentration for *S. aureus* and *E. coli*, respectively.
[d] Poliovirus Type 1: "+" complete kill in less than 30 sec contact; "-" complete survival after 120 sec contact.

References

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