Original Article

# Are Local Anesthesia Vials Similar to Champagne: Do they Lose their Potency Once Opened?: An *In vitro* study


## Abstract

**Background and Objectives:** Epinephrine is volatile and unstable on exposure to air. Addition of sodium metabisulfite as an antioxidant has been effective, but there are no clear-cut consensus on its efficiency in retarding the oxidation that sets in once the anesthetic vials are kept open with a needle *in situ*. There is a paucity of scientific data regarding the risks of reuse of anesthetic vials following the use of cartridges, a common practice in western countries. It is our endeavor to shed light on the constant change in pH, drug deterioration, and instability that sets in an anesthetic vial with a needle *in situ*. **Methods:** Twenty sets of identical local anesthetic vials (lidocaine 2% with 1:80,000 adrenaline) were collected. The samples were divided into four individual groups followed by a segregation of 5 subsamples at varying time intervals from each individual group. pH was measured using pH meter. **Results:** Change in the pH of the solution was noted as days progressed in open local anesthetic vials when compared to closed vials. However, the decreased pH remained constant following a brief time interval after complete conversion of sulfite to sulfate. **Conclusions:** Local anesthetic vials with needle pricked in for a longer duration can alter the pH, concentration of the vasoconstrictor and deteriorate the properties of local anesthetic solution. This can manifest clinically as delayed onset of action, increased burning sensation, and pain on administration.


**Keywords:** *Adrenaline, local anaesthesia, vasoconstrictors*


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## Introduction

Pain has dogged humankind since its inception and wonder drugs such as local anesthetics (LA) have enabled clinicians to perform painful procedures at ease without loss of consciousness. Dental care has been safe and pain free with the use of LA, a class of drugs that has an impressive history of safety and efficacy. LA drugs are a boon for surgeons as they can perform pain-free minor surgeries on an outpatient basis. Most LA solutions hold a preservative agent like 0.1% sodium metabisulfite, with or without thymol, a fungicide. This has extended the shelf life and enabled us to administer LA agents multiple times from a single vial. LA are available as solutions for injection, sprays, creams, and gels. The archaic practice of needle insertion into the LA vial and its deleterious effects following the ingress of air on its potency seem to have been unnoticed so far from the hawk eye of researchers and clinicians. The impetus for the current study was to point the way for clinicians unaware of a faulty practice that has a significant clinical relevance such as altering the onset of anesthesia and producing a burning sensation on administration.

### Aims and objectives

LA containing epinephrine or any other vasoconstrictors are acidified by the manufacturer using sodium metabisulfite to inhibit the oxidation of the vasopressor. The LA vial once pricked to load the anesthetic agent is often left open with the needle *in situ* for weeks to months until the vial has been discarded. There is a need for further studies to evaluate the process of oxidation that sets in on prolonged exposure to air and its impact on the clinical efficacy of LA, level of burning sensation, and degree of pain experienced by the patient. The efficiency of sodium metabisulfite in retarding the oxidation of the vasoconstrictor needs to be further evaluated by chemical tests that can detect the deterioration of adrenaline on exposure to air. The aim of the study is to determine the change in pH of the LA solution in closed vials and vials with needle *in situ* at successive time intervals. Methodology:

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This was an *in vitro* study carried out in St. Aloysius Research laboratory Mangalore (India) for a period of 1 month. Ethical approval was not required as there were no human subjects involved. Previous studies have designated lignocaine as the least irritating local anesthetic in comparison to the other LA solutions; hence, we decided to explore only lignocaine in our study. Good efficacy, low allergenicity, and minimal toxicity facilitate its safe use.[1,2] Five vials of 2% lidocaine with 1:80,000 adrenaline from the same manufacturer (Warrens co) with analogous expiry dates were chosen for this study. pH of the LA vial while freshly opened was determined using a pH meter. pH meter is an instrument used to measure pH It has a platinum electrode which is dipped in LA solution; it is also called a standard electrode as it measures the pH of the unknown solution.[3] This in turn is connected to a circuit to calibrate the readings. The samples were then divided into four groups and further categorized into five subsamples from each group to assess the pH at varying time intervals. Group 1: 2% lidocaine with 1:80,000 adrenaline pricked with a 25-gauge needle and kept *in situ* calculation of the pH was done at regular time intervals of 1, 3, 7, 15, and 30 days, respectively. Group 2: 2% lidocaine with 1:80,000 adrenaline pricked with an 18-gauge needle and kept *in situ*. pH was determined at regular time intervals as mentioned in Group 1. Group 3: 2% lidocaine with 1:80,000 adrenaline, LA bottle was kept open. Group 4: 2% lidocaine with 1:80,000 adrenaline, pH was determined in a closed LA bottle with a threaded lid. The antioxidant used for vasoconstrictor is sodium metabisulfite which on exposure to air is converted to sodium metabisulfate. A confirmatory test to validate our hypothesis of change in pH and oxidation of the vasoconstrictor on exposure to air was carried out in a fresh solution by the addition of barium chloride. The absence of precipitation by adding barium chloride confirmed the absence of sodium metabisulfate in a fresh solution. A chemical reaction between manganese dioxide and hydrogen peroxide yields oxygen as a by-product that is made to react with the fresh solution. This results in conversion of sodium metabisulfite to sodium metabisulfate. Further, the oxidized solution is made to react with barium chloride that forms a precipitate. This is a confirmatory test of conversion of sodium metabisulfite to sodium metabisulfate on exposure to air [Figure 1]. The series of chemical reactions that set in a local anesthetic vial on exposure to air at varying time intervals resulted in pH changes which were detected in our study with the help of pH meter using platinum electrode [Figure 2]. These pH readings are subjected to change from one electrode to another depending on the surface area of the electrode.[3-5]

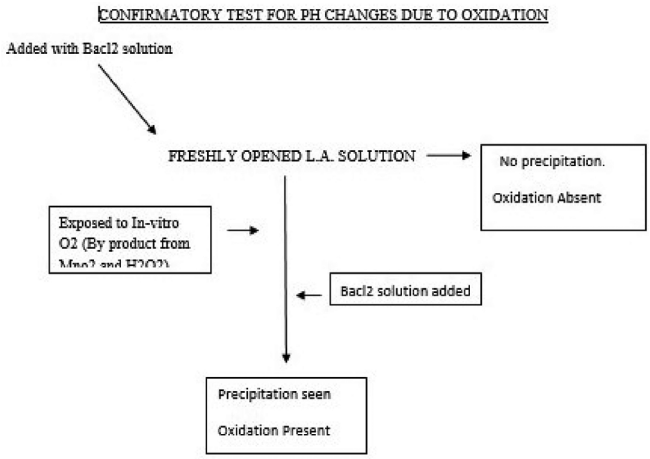


**Figure 1: Flow chart confirming the presence of oxidation by a series of chemical reactions**

## Results

Intergroup comparison was done using one-way ANOVA and Tukey *post hoc* test. Intragroup comparison was done using repeated measures ANOVA with Bonferroni *post hoc* test.

$P < 0.05$ was considered statistically significant [Table 1]. The collected data were subjected to statistical analysis using SPSS software version 19 (Warrens co). The study group included open vials and vials pricked with 25- and 18-gauge needle. The control group included closed vials. A steady change in pH was noted at varying time intervals in vials pricked with 25-, 18-gauge needle; however with a duration of 30 days, there was a substantial change in pH as seen in the graph. The open local anesthetic vials had a constant change in pH level at varying time intervals.

In the control group, in spite of being a closed vial, there were changes in the pH noted. This can be attributed to the fact of inevitable ingress of air while transferring the solution from the closed vial to the platinum electrode to determine the pH. Significant differences were observed in the mean scores of test and control group and $P = 0.001$ which is statistically significant. The study demonstrates a steady decrease in the pH with the ingress of air into the vial; however, once the conversion of sulfite to sulfate takes place, the pH remains at a constant level. Local anesthetic vials with needle *in situ* will lead to decrease in the pH of the solution. This can manifest clinically as delayed action of onset of anesthesia and discomfort experienced by the patient due to the acidic pH.

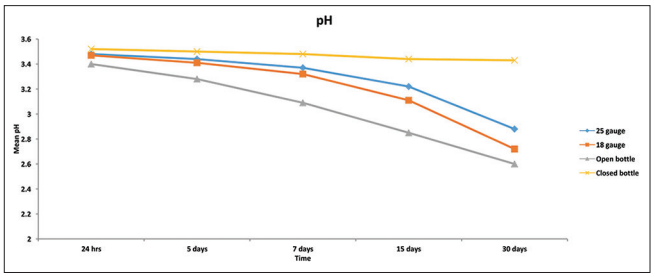


**Figure 2: Line diagram depicting variation in the pH level at different time intervals in each study group**

**Table 1: Intergroup comparison - pairwise comparison with similar uppercase superscript - statistically NS ($P > 0.05$). Intragroup comparison - pairwise comparison with similar lowercase superscript - statistically NS ($P > 0.05$). All other intergroup and intragroup pairwise comparison - statistically significant, $P < 0.05$. NS: Nonsignificant**

| Groups | *N* | 24 hrs | 5 days | 7 days | 15 days | 30 days | Repeated measure ANOVA | |
|---|---|---|---|---|---|---|---|---|
| | | | | | | | *F* - value | *P* |
| 25 gauge | 5 | 3.48 (0.01)$^{A}$ | 3.44 (0.01) | 3.37 (0.02) | 3.22 (0.02) | 2.88 (0.02) | 1216.38 | <0.001* |
| 18 gauge | 5 | 3.47 (0.01)$^{A}$ | 3.41 (0.01) | 3.32 (0.01) | 3.11 (0.01) | 2.72 (0.03) | 2300.99 | <0.001* |
| Open bottle | 5 | 3.40 (0.02) | 3.28 (0.01) | 3.09 (0.02) | 2.85 (0.03) | 2.60 (0.03) | 1133.29 | <0.001* |
| Closed bottle | 5 | 3.52 (0.02)$^{a,b}$ | 3.50 (0.01)$^{a,c,d}$ | 3.48 (0.01)$^{b,c,e}$ | 3.44 (0.01)$^{f}$ | 3.43 (0.03)$^{d,e,f}$ | 31.65 | <0.001* |
| ANOVA | *F* - value | 72.41 | 526.56 | 734.99 | 856.15 | 1153.05 | | |
| | *P* | <0.001* | <0.001* | <0.001* | <0.001* | <0.001* | | |

## Discussion

Local anesthetic drugs are used worldwide to induce anesthesia and analgesia intraoperative and postoperative. Understanding the basic pharmacology, drug interactions, and safe handling enables the anesthetist to use them judiciously to the maximum effect. LA are basic compounds, poorly soluble in water; the addition of hydrochloride forms a stable salt. Adrenaline also known as epinephrine has a dual role of a drug and as a hormone produced by the adrenal glands and certain neurons. It shares its place in the World Health Organization's List of Essential Medicines and is certainly a doctor's friend for its diverse application in emergency situations. Adrenaline is available in an ampule as concentration of 1:1000, 1:10,000 used in anaphylactic shock. It is also added to LA solutions as a vasoconstrictor in diluted concentrations of 1:80,000–1:200,000. Lignocaine with adrenaline combination is favored routinely for easy dissection, pain relief postsurgery, and reduction in the degree of edema and ecchymosis. LA are used to block neural impulse traffic; its rapid absorption is a nuisance as it shortens block and heightens blood level. Unintentionally, high concentration of local anesthetic in the blood stream can cause unwelcome side effects in distant organ system: heart and brain in particular. These deleterious effects are avoided with the addition of adrenaline that maintains the depth and duration of the local anesthetic agent; however, some of the adverse effects of local anesthesia such as tachycardia and tremors are caused by adrenaline.[6,7] The lowest dose possible to produce the desired action is used, and minimal toxicity is an age-old adage, this should be applied to the concentration of vasoconstrictors used in local anesthetic agents. The concentration of vasoconstrictors will vary depending on the class of local anesthetic agent used, duration of the surgical procedure anatomy and vascularity of the injection site, need and extent of intraoperative hemostasis desired. Carbonated LA in comparison to lignocaine hydrochloride has a reduced onset time and increase in the duration of anesthesia. Carbon dioxide enhances the diffusion of local anesthetic through nerve membranes providing a more rapid onset of nerve block. Diffusion of $CO_2$ through the nerve membrane decreases the intracellular pH resulting in increased intracellular concentration of charged cations. As the cationic form of the drug does not readily diffuse out of the nerve, the anesthetic becomes concentrated with nerve trunk (ion trapping) providing a longer duration of anesthesia.[6-8] Our faulty practice also reduces the efficacy of carbonated LA as on exposure to air, the $CO_2$ will diffuse out of the vial easily. The study highlights the gradual reduction in the pH at varying time intervals. The greater the time interval of exposure of the solution to air or moisture the greater is the oxidation with a steady drop in the pH. The gauge of the needle inserted correlates to the volume of air that seeps in which in turn has an effect on pH of the LA agent. Nonetheless, following the complete conversion of sulfite to sulfate, the low pH remains constant as seen in our study. Lidocaine in clinical doses has anti-inflammatory actions. It prevents leukocyte adhesion, migration and phagocytosis, as well as synthesis of inflammatory mediators, vascular hyper permeability, and edema formation.[9,10] The shelf life of a drug also depends on the environmental temperature, effective handling, and storage of the drug. The low pH due to faulty storage conditions could negate its anti-inflammatory activity. The change in pH is brought about by the oxidation of the sulfites into sulfates resulting in liberation of protons.[11] A similar study on local anesthetic solution exposed to sunlight inferred similar findings.[12,13] Cold storage of the local anesthetic solution retains a stable pH for a year,[14] could be a suitable alternative until we discard the age-old practice of reuse of the vial, and espouse the cartridge system used widely in western countries. To date, there has been a dearth of information about the ideal way to store and handle local anesthetic solution, one of the most common drug used in dentistry.

In a developing country like India, there is no oversight by regulatory bodies of the overzealous multiple use of needles from a single vial. A positive aspirated needle can have traces of blood, if not discarded and reloaded can risk the transmission of blood borne infection. Sterility of the multidose vials can be questionable

as there are no studies so far that have assessed the bacteriostatic activity of methyl paraben in vials with needle *in situ*.

Considering the sample size, *in vitro* study conducted using basic instruments, with barely any studies of similar nature to associate with, only gross conclusions could be made after the analysis of the results. A little has been explored on safe handling and effective delivery of the local anesthetic drugs; a study of this sort could alarm the clinicians of the innocuous faulty practice that has been followed blindly since years. The results obtained from this study concluded that the faulty practice of inserting a needle to draw LA solution can leak atmospheric air into the vial, deteriorate its stability, properties, and alter the pH. This can render local anesthetic being ineffective to deliver complete pain-free fruitful fruition of the dental procedure; also, the burning sensation with the change in pH can be a cause of patient anxiety and discomfort.

After a long hiatus, the field of LA is revolutionizing towards the development of drugs with safer predictable effects, rapid onset, and short duration of action that is apt enough to carry out any minor surgical procedures. The future is going to see developments in anesthesia delivery systems, different routes of drug delivery, use of threaded sealed caps of LA vials, autoinjectors, inhalational LA, oral LA, and varying class of local anesthetic agents that is apt for the surgical procedures being carried out.

## Conclusion

Local anesthetic vials with needle pricked in for a longer duration can alter the pH, concentration of the vasoconstrictor and deteriorate the properties of local anesthetic solution. This can manifest clinically as delayed onset of action, increased burning sensation, and pain on administration. The article aims to adopt the cartridge system, while administering local anaesthesia and necessitates the need to discard the age old practice for a safe and effective delivery of local anaesthesia.

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### Conflicts of interest

There are no conflicts of interest.

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