Preservatives in Liquid Pharmaceutical Preparations

Chawki Boukarim,* Sarah Abou Jaoudé, Rita Bahnam, Roula Barada, Soula Kyriacos

Department of Pharmaceutical Sciences, School of Pharmacy, Lebanese American University, Byblos, Lebanon.

KEY WORDS: Preservatives, sodium benzoate, potassium sorbate, methylparaben, quality control.

ABSTRACT

Sodium benzoate, potassium sorbate, and methyl hydroxybenzoate are commonly used as preservatives in liquid pharmaceutical preparations. The purpose of this study is to determine the amount of the aforementioned preservatives in pharmaceutical products, as some recent studies have reported serious side effects associated with the ingestion of these substances. The content of 37 liquid pharmaceutical products were simultaneously determined by high performance liquid chromatography. Preparations were analyzed in triplicate for their preservative content using a sensitive and reproducible HPLC method modified in our laboratory. Preservative levels were found to fall outside the typical allowed concentration range for 70% of the samples, with some exhibiting significantly higher concentrations. The reason behind such findings is unclear, and could be due to poor quality control or to intentionally extend the shelf-life of the products. These findings highlight issues related to quality control and to patient's safety. Consequences on patient health need to be evaluated, especially since most liquid pharmaceutical products are administered to the pediatric population.

INTRODUCTION

Preservatives have been commonly used as additives in pharmaceutical products, cosmetics, and food. Liquid preparations are particularly susceptible to microbial growth because of the nature of their ingredients. Such preparations are protected by the addition of preservatives that prevent the alteration and degradation of the product formulation.[1] Preservatives are mainly effective in controlling mold, inhibiting yeast growth, and protecting against bacterial proliferation. Their antimicrobial and antifungal properties make them an integral part of the product formulation.

Among the most commonly used preservatives in the conservation of liquid pharmaceutical preparations are sodium benzoate, potassium sorbate, and methyl hydroxybenzoate (methylparaben). Their typical allowed concentrations range respectively from 0.1-0.2%, 0.1-0.2%, and 0.1-0.25% (w/w).[1] The purpose of this study is to determine the amount of the former preservatives in liquid pharmaceuticals for quality assurance purposes as well as for patient safety. Accrued interest is given to preservatives as recent studies have reported serious side effects associated with these substances. Skin reac-

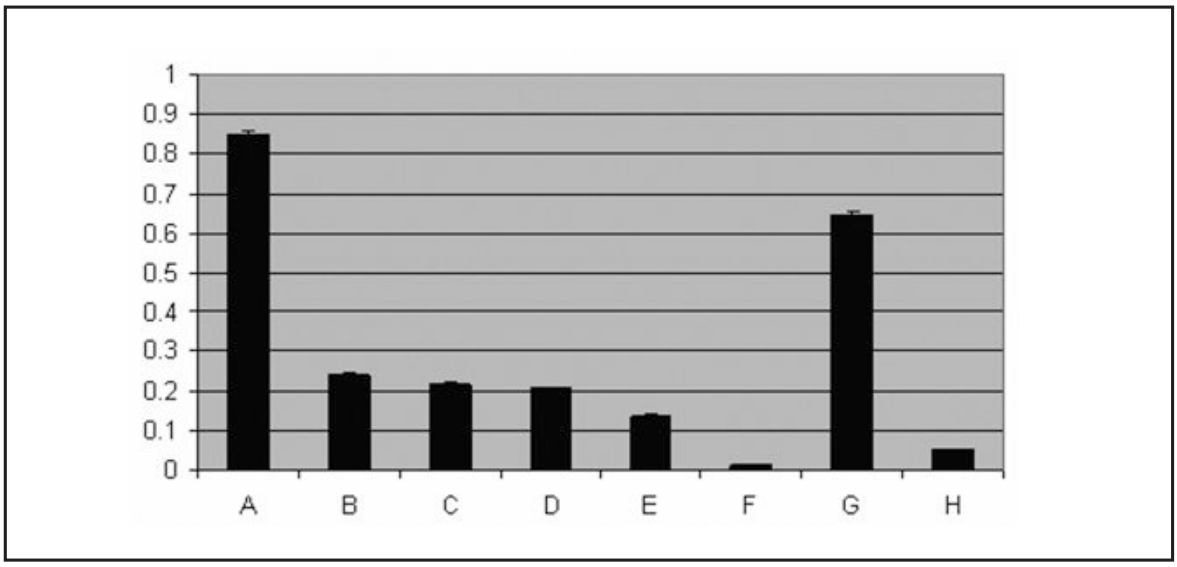

Figure 1: Concentration (%w/w) of Sodium Benzoate in labeled samples.

tions such as rash, urticaria,,and contact dermatitis have been reported after topical application of potassium sorbate,-methylparaben,- and sodium benzoate-containing products.[1,2,3] Other side effects have been reported after ingestion of medications containing these preservatives such as the allergic potential and estrogenic potential of parabens,[2,4] or the genotoxic activity potential of sodium benzoate.[3]

MATERIALS AND METHODS

Thirty seven liquid pharmaceutical preparations were purchased from randomly selected pharmacies in Lebanon. Not all preparations were labeled with respect to their preservative content: 19, 8, and 1 preparation(s) were labeled for their methylparaben, sodium benzoate, and potassium sorbate content respectively, while 14 preparations were unlabeled with respect to one or more preservatives. Commercial brands of the liquid preparations were coded using letters (A to AL).

Acetonitrile (HPLC grade) and sodium acetate (analytical grade) were purchased from Sigma, Germany. Sodium benzoate, potassium sorbate, and methylparaben standards were purchased from Fluka, UK. Standards were used to prepare standard working solutions using distilled water, to obtain stock solutions of 1 g/L concentration. Stock solutions were used to prepare solutions of lower concentrations (50, 100, 200, and 400 mg/L) to build the calibration curve. The correlation coefficients were above 0.999 in all cases.

Preparations were analyzed in triplicate for their preservative content using a sensitive and reproducible HPLC method [5-8] modified in our laboratory. The HPLC system consisted of LC-10 pump (Shimadzu), a variable ultraviolet detector monitor set at 229 nm (Shimadzu, SPD-10), and a Chromatopac Shimadzu (C-R8A) integrator. Separation was done using a pre-packed stainless steel column (15 cm x 0.46 cm) filled with Shimpack C18 10 µm Silica (Waters, Germany), and the flow rate was set to 1.5 mL per minute for sodium benzoate and potassium sorbate and 2 mL/min for methylparaben. The precision of the assay method was determined by calculating the relative standard deviation (inter- and intra-days) of the peak areas obtained after repeated injections (n=3) of all standard solutions. The relative standard deviations of the areas were found to be less than 3.5%, which confirms the precision of the method.

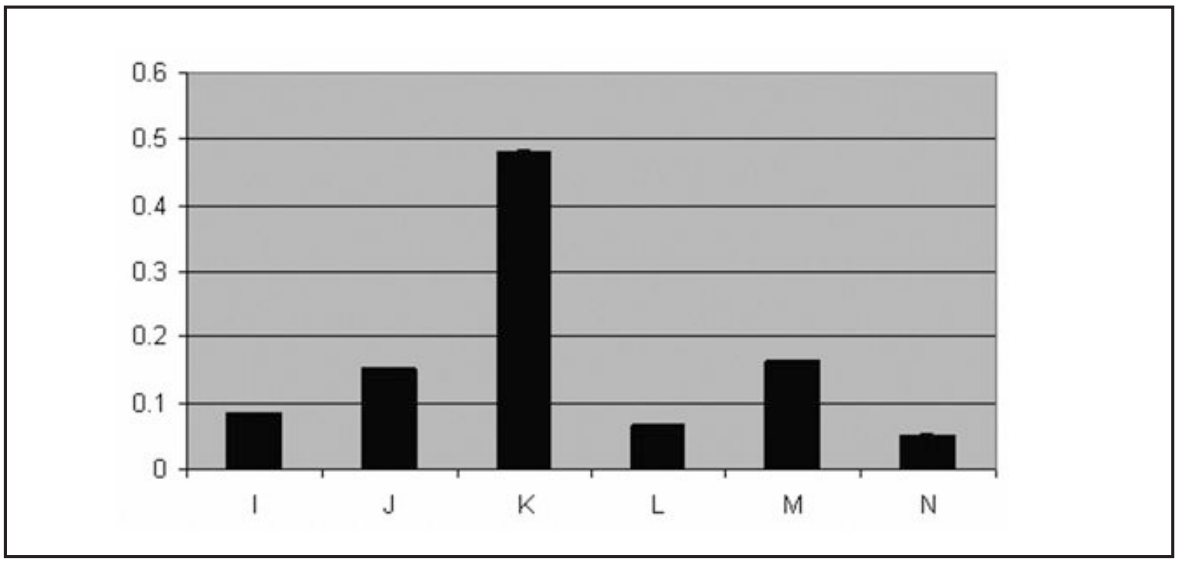

Figure 2: Concentration (%w/w) of Sodium Benzoate in unlabeled samples.

One gram of each of the samples was accurately weighed and transferred to a 100 mL volumetric flask. Mobile phase was added to volume and mixed. The mobile phase was an aqueous solution of acetonitrile 0.02 M sodium acetate buffer (20:80, v/v), adjusted to a pH 4.3 with acetic acid. The mobile phase was filtered through a 0.45 Ìm filter and degassed before use. Twenty microliters of the filtrate were injected into the HPLC.

The retention times for sodium benzoate, potassium sorbate and methylparaben were 5.2, 6.7, and 7.6 minutes, respectively.

RESULTS AND DISCUSSION

In the eight samples labeled as containing sodium benzoate as a preservative, concentrations of the latter ranged between 0.012 - 0.85 % (w/w). Two samples (A and G) included sodium benzoate in amounts significantly exceeding the typical allowed concentrations (up to four-fold difference) whereas two other products had a sodium benzoate concentration slightly exceeding that range (Figure 1).

After screening the 12 unlabeled samples, six were found to contain sodium benzoate in concentrations ranging from 0.05-0.48 % (w/w). Only Product K contained the preservative in concentrations above the typical allowed concentrations (Figure 2). Two products exhibited sodium benzoate concentrations below typical allowed concentrations. Additionally, as much as 50% of the pharmaceutical products containing sodium benzoate are formulated at a pH>5, at which the preservative is ineffective.

As for potassium sorbate, the only labeled product included a higher amount of preservative. Many unlabeled samples were found to contain potassium sorbate as a preservative. As shown in Figure 3, products E and O exceeded the typical allowed concentrations for its use.

Nineteen samples were labeled as having methylparaben as a preservative. It was found in concentrations ranging from 0.03-0.55 % (w/w). Only one labeled product included methylparaben in concentration exceeding the maximal allowed amount and 14 out of the 19 samples had concentrations below the allowed range (Figure 4). None of the unlabeled samples exhibited concentrations exceeding the typical allowed range (Figure 5). All, however, had their methylparaben concentration below the allowed range.

CONCLUSION

Preservatives levels were found to fall outside the typical allowed concentration range for 70% of the samples, with some exhibiting significant higher concentrations. The present study highlights the high amount of preservatives that may be found in some liquid pharmaceutical preparations. The reason behind such finding is unclear and could be due to poor quality control or to intentionally extend the shelf life of the products. Consequences to patients' health need to be evaluated, especially since most liquid pharmaceutical products are administered to the pediatric population.

ACKNOWLEDGEMENTS

This work was supported by a grant from the Research Council at the Lebanese American University.

REFERENCES

1. P. Beringer, A. derMarderosian, L. Felton, et al. Remington: *The Science and Practice of Pharmacy*, 21st edition, Lippincott Williams & Wilkins, Philadelphia, 2006.
2. M.G. Soni, I.G. Carabin, G.A. Burdock. Safety Assessment of esters of p-hydroxybenzoic acid (parabens). *Food.Chem Toxicol.* 43(7): 985-1015 (2005).
3. B. Nair. Final Report on the Safety Assessment of Benzyl Alcohol, Benzoic Acid, and Sodium Benzoate. *Int.J.Toxicol.* 20 (Suppl 3): 23-50 (2001).
4. M.G. Soni, S.L. Taylor, N.A. Greenberg, G.A. Burdock. Evaluation of the health aspects of methyl paraben: a review of the published literature. *Food Chem Toxicol.* 40(10): 1335-73 (2002).
5. G. Shabir. Determination of combined p-hydroxy benzoic acid preservatives in a liquid pharmaceutical formulation by HPLC. *J Pharm Biomed. Anal.* 34:207-213 (2004).
6. D. Satinsky, J. Huclova, R.L.C. Ferreira, et al. Determination of ambroxol hydrochloride, methylparaben and benzoic acid in pharmaceutical preparations based on sequential injection technique coupled with monolithic column. *J Pharm Biomed. Anal.* 40:287-293 (2006).
7. H.M. Pylypiw Jr., M.T. Grether. Rapid high-performance liquid chromatography method for the analysis of sodium benzoate and potassium sorbate in foods. *Journal* of Chromatography A. 883: 299-304 (2000).
8. K. Kuprovskyt?, B. Pranaityt?, and A. Padarauskas. Isocratic HPLC determination of preservatives in beverages. *Chemija (Vilnius).* T. 13, Nr. 3 (2002).