International Journal of Health Sciences and Research
Vol.10; Issue: 8; August 2020
Website: www.ijhsr.org
Short Communication *ISSN: 2249-9571*

Evaluation and Comparison of Buffering Capacity of *Apamarga* and *Phalasha Ksharas*

Premalatha Shetty[1], Seema Mandi B[2], Pampanna Gouda H[3]

[1]Scientist, Department of Biotechnology & Microbiology, Sri Dharmasthala Manjunatheshwara Centre for Research in Ayurveda and Allied Sciences, Udupi-574118, Karnataka, India
[2]Professor, Department of Rasashastra and Bhaishajya Kalpana, Sri Dharmasthala Manjunatheshwara College of Ayurveda and Hospital, Udupi-574118 Karnataka, India;
[3]Professor, Department of Samhita, SDM Institute of Ayurveda, Bangalore, Karnataka, India;

Corresponding Author: Premalatha Shetty

ABSTRACT

Background and Aim: Apamarga (Achyranthes aspera) and Phalasha (Butea monosperma) ksharas are two of the extensively employed Ayurvedic medications in treatment skin diseases. Alkaline nature of Kshara is believed destroy the vitiated tissues. Ability of these preparations to resist pH may give insight to the characteristics of Ksharas.
Methodology: Mrudu ksharas of Apamarga and Phalasha were prepared through series of stages as recommended by Acharya Sushruta. The ability of the ash and Kshara suspensions, and Kshara jalas to resist changes in pH was assessed by acidification with 5N HCl and compared with the buffers used in bioscience laboratories.
Results: All the preparations were found to be highly alkaline in nature exhibiting pH values in the range of 10-12. Apamarga preparations obtained at all the three stages namely the ash, Kshara jala and the final product Kshara were characterized by high buffering capacity. Phalasha ash exhibited strong buffering ability. The proton neutralizing ability of Phalash kshara jala and kshara preparations however, were significantly lower than the respective Apamarga preparations. It appears that significant fraction of components responsible for the buffering ability in Phalasha ash are not carried over into its soluble fraction (Jala). Although the pH of the preparations remained high, the buffering ability of the Phalasha jala and Kshara preparations were significantly lower than the respective Apamarga preparations, indicating that buffering ability of a Kshara can be an interesting characteristic to be considered in the study of Ksharas.

Key words: Phalasha Kshara, Apamarga Kshara, pH, Buffering capacity

INTRODUCTION

Acharyas have employed Kshara karma since ancient times believably for destruction of vitiated tissues (kshapana). Being an alkaline preparation, Kshara is prepared by extracting the ash of dried plant materials. The unique protocol for preparation of ksharas is well described by Sushruta in Ayurvedic literature. This modality of treatment is believed to scrape away unhealthy tissues even from deep rooted locations, thereby reducing the recurrence of the disease. Therefore, Kshara karma holds a position of significance in treatment of kshudra rogas (small diseases) such as warts, corns, facial melanosis. Kshara has also attained a dominant position in surgical and parasurgical procedures. [1,2]

Major ingredient of Kshara is the ash of dried plant parts which is processed through series of filtration steps followed by heat treatment. Acharya Shushrutha mentions set of herbal drugs to be used for kshara preparation such as Apamarga, Kutaja, Paribhadra, Ashwakarna, Bibitaki, Tilvaka, Arka, Phalasha etc. The drugs possess the qualities of sampath veerya as they are balavan (potent) and kriya Samarth

(efficient). The herbal drug for Kshara preparation must be collected in summer from Ushara Bhumi (alkaline soil) [3] Of the various Ksharas cited in literature, Apamarga and Phalasha are the two widely applied medications. Although used mainly for topical applications (Pratisaraniya Kshara), paaniya form of kshara (mrudu/ mild form) is often prescribed for oral administration in treatment of certain abdominal and urogenital ailments. Mrudu kshara is the mild form of this medicament. Alkalinity is believably responsible for the therapeutic effect of kshara. The major mineral components reportedly present in ksharas are potassium, sodium, chlorides, carbonates and sulphates. [2,4]

In the present study, the ability of the ash, kshara jala and kshara samples to resist change in pH was explored and compared.

MATERIALS AND METHODS

Preparation of the Ksharas:

Dried forms of panchanga (leaf, root, stem, fruit and flower) were converted into ash in iron pan. Apamarga (AP) ash was then suspended in six parts of water and filtered 21 times through thick white clean cotton cloth as recommended by Acharya Sushruta [5] to obtain clean carbon less Apamarga Kshara jala. The kshara jala thus obtained was subjected to mandagni till it reached semi solid consistency. Phalasha (PL) Kshara was prepared in a similar manner using Phalasha Bark as the raw material.

Parameters	Apamarga	Phalsha
Weight of dried plant parts, Kg	12.5	6
Weight of ash obtained, Kg	1.10	1.3
Volume of ash obtained L	2.1	2.6
Volume of ash taken, L	2	2.5
Volume of water taken, L	12	15.3
Ksharajala obtained, L	10	14.8
Ksharajala taken ,L	9.900	14.7
Kshara obtained, g	285.0	140
% Kshara obtained w/w	2.28	2.3

Pharmaceutical

The ash samples, kshara jalas and the kshara preparations were stored in air-tight glass containers.

Preparation of the biological buffer systems:

Glycine (0.2M) and Tris-glycine (0.025M Tris and 0.25M glycine) buffer systems were prepared and the pH was adjusted to a value of around 10.25 with NaOH. Borate and Carbonate buffer systems at pH values of ≈10.25 were prepared at a concentration of 0.2M.

Determination of pH:

The pH of the Jala was measured using digital pH meter (Systronics). The ash and Kshara suspensions (10% w/v) were prepared in D/W and the pH was measured.

Buffering capacity:

To fifteen mililitres of the ash/kshara preparations (10% in D/W), 5N HCl was added in increments of 0.1ml. After each addition, the suspension was stirred well and the pH was noted after stabilization.

The proton scavenging ability of the Jala/ buffers was tested by adding 100µl increments of 5N HCl to 15ml of the sample. The pH was noted each time after the addition of acid.

RESULTS

The ash obtained by direct ignition of the raw material, was further roasted to burn the unburnt residues ensuring maximum recovery of the drug. The pH of all the ayurvedic preparations used in the study was in the range of 10.0 to 12. The pH values of the ash, jala and mrudu kshara prepared from Apamarga were 11.88, 10.88 and 10.9 respectively (figure 1A). The pH values of PL ash, jala and mrudu kshara were 12.03, 10.09 and 11.98 respectively (figure 1B). The ability of these ayurvedic preparations to neutralize protons was determined. As shown in figure 1, AP ash, jala and kshara were neutralized (pH reduced to around 6.9-7.1) by addition of 0.9, 0.7 and 0.6ml of 5N HCl respectively. Although the buffering ability of PL ash was similar to AP ash requiring 0.9-1ml for neutralization, its jala and kshara samples exhibited significant reduction in their ability to resist changes in the pH on acidification requiring around addition of

approximately 0.25ml and 0.4ml of the acid respectively.

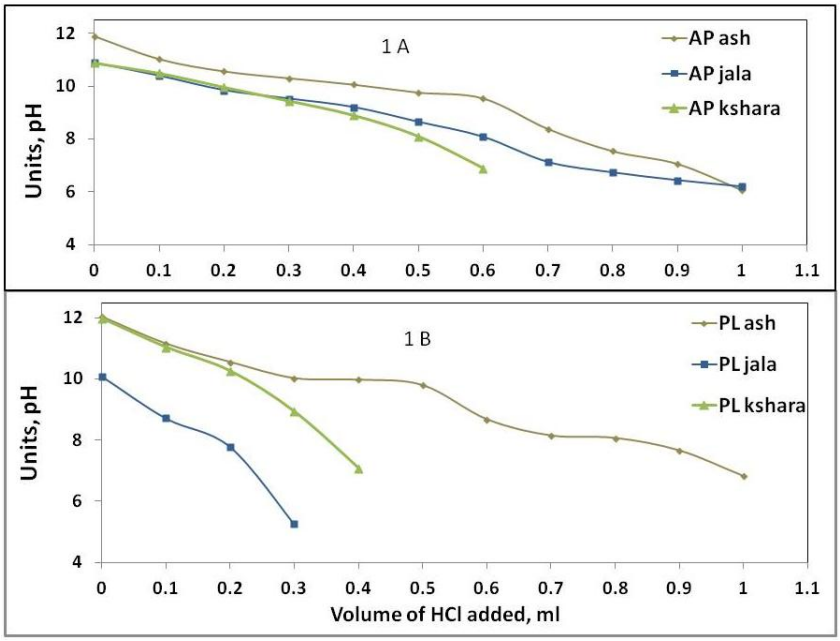

Figure 1: Acid neutralizing ability of the Apamarga and Phalasha preparations

The buffering capacity of the samples was compared to that of buffers generally used in bioscience laboratories (figures 1 and 2). Tris-glycine (TG), Borate, Carbonate and Glycinate (Gly) buffers are the most commonly used alkaline buffer systems. These buffers were assessed for their ability to resist changes in pH at concentrations as high as 0.2/0.25M. As shown in figure 2, all the four buffer systems exhibited buffering capacity in the alkaline range. TG and Borate buffer systems exhibited strong buffering capacity over the broad range of pH, requiring around 0.65-0.75ml of 5N HCl for reduction pH from initial pH of ≈10.25 to neutral pH, under the defined experimental conditions.

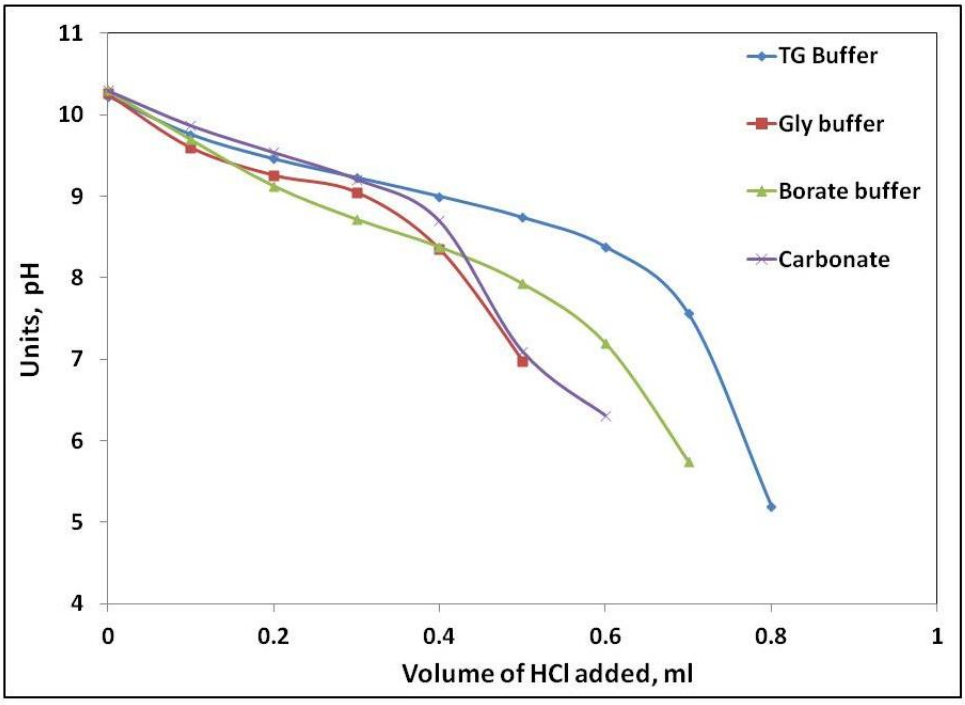

Figure 2: Buffering capacity of the standard buffer systems

DISCUSSION

The pH of the AP jala and kshara was lower by one unit in comparison to its ash. The pH of PL jala was reduced by two units in comparison to its ash.

The buffering ability of the ash and kshara preparations was compared to the biological buffers. Borate buffer is reportedly effective in the range of pH 8.5-10.2 with a pKa at around pH 9.2. The pKa value is the pH value at which the acid and conjugate base forms of the buffer are present at equal concentrations and hence are able to resist the change in pH around its pKa. Carbonate buffer system is reportedly effective in the range of 9.2-10.6. The pKa of glycine in the alkaline side of the pH is reported to be around 9.8 and resists changes in pH in the range of pH 8.2-10.6. Tris exhibits its buffering ability in the range of pH 7.2-9 with a pKa of around 8.3 (https://www.interchim.fr/ft/B/BC002b.pdf). Thus, TG buffer system contends as stronger buffer system effective over a broader pH range. The buffer systems adjusted to a pH of 10.23-10.3 were employed for comparative studies. We found that distilled water (15ml) adjusted to a pH of 10.26 could be neutralized by addition of 5.5 microlitres of 5N HCl. Around 0.65 and 0.75ml of 5N HCl was however, required for neutralization of 15ml of borate and TG buffer systems respectively. Addition of 0.5ml of acid resulted in neutralization of Gly and carbonate buffer systems. As shown in figure 1, around 0.6ml and 0.7ml of the acid was required to neutralize the AP ash and PL ash suspensions respectively, from the pH of 10.25-10.3. It is interesting to note that the AP jala and kshara preparations used up around 0.45-0.55 ml of the acid for reduction of the pH from 10.25 to neutral pH. Thus, the buffering power of the ash suspensions was higher in comparison to jala and kshara preparations. The ash suspensions exhibited strong buffering power approximating the acid neutralizing ability of borate or TG buffers. AP jala and kshara preparations approximated the acid neutralizing ability of 0.2M carbonate and glycinate buffers. The buffering ability of PL jala and Kshara was comparatively weaker than rest of the preparations used in the study (figure 1). Thus, both the ash suspensions showed strong buffering capacity in comparison to their respective jala and mrudu form of ksharas. As the ash is processed through series of filtration steps and evaporation stages for preparation of Kshara, buffering ability of the PL kshara jala and kshara was found to reduce to a significant extent. In comparison, AP jala and kshara preparations were found to retain their buffering capacity to a significant extent. It must also be noted that pH of PL jala was lower than its respective ash form by two units. The water insoluble minerals of Phalasha may be contributing to the buffering capacity of the ash to a significant extent. During the neutralization process, the added acid may be aiding in solubilisation of water insoluble mineral components of the ash which possess the proton neutralizing capacity. The acid soluble components in the ash of Phalasha, therefore appears to be responsible for scavenging of protons. HCl is known to extract metals in bound forms such as iron-manganese oxides/hydroxides, insoluble phosphates and carbonates. It is also reported to solubilise sulphides to a limited extent. The practice of addition of powdered sea shell to the jala while preparing the madhyama/teekshna (strong) kshara may help in improvement of the buffering capacity due to the added carbonate present in the shell powder. The inflections in the pH curve for ash suspensions (especially of Phalasha) indicate that mixture of components present in the ash, are perhaps responsible for the buffering effect over the broad range of pH. Similar studies with various ash, jala and kshara samples may give insight to the characteristics of these preparations and the possible co-relation of buffering capacity to the efficacy of ksharas.

The ash and Kshara samples were characterized by production of effervescence during the neutralization

process. Sudden drop in pH accompanied by effervescence and then increasing of pH to a stable value was observed. Studies have reported that Ksharas are rich in carbonates. [6] Carbonates are known to breakdown and release carbon dioxide during the neutralization (by acidification) process. When Kshara is applied at the wound site the possibility of slow release of CO_2 due to the biochemical reactions and buffering mechanism at the wound site cannot be ruled out. If CO_2 is generated, the pasty consistency of the Kshara is likely to entrap and hamper the escape of CO_2. Transdermal application of CO_2 is reported to accelerate chronic and incisional wound healing processes by improving granulation. The therapeutic effect of Kshara could be through a complex network of events and therefore, it would be interesting to explore the mechanistic of Kshara therapy.

CONCLUSION

All the three apamarga preparations were found to possess strong buffering capacity. In comparison, Phalasha jala and kshara were relatively weak at resisting the change in pH on acidification. Albeit retaining high alkalinity, the Ksharas differed in their buffering ability to a significant extent. Buffering ability can be included as a parameter in characterization of the Ksharas. Apamarga kshara is considered to be highly potent. Ksharasutra with apamarga kshara is reportedly more effective than treatment with Phalasha kshara. [7-9] At above the pH of 8, the fungi and most of the bacteria will not be able to survive and hence, the ksharas will prevent proliferation of microbes at the site of application. The buffering capacity of the apamarga kshara can therefore be studied as a characteristic of ksharas and needs to be probed in relation to its efficacy as a medicament. Although it is reported that acidic environment facilitates wound healing, [10] in Ayurvedic practice however, Kshara therapy is being employed extensively in treatment of skin diseases which includes treatment of chronic wounds. Wound healing is a complex process involving sequence of events. The pH of above 7.4 is reported to aid in granulation and tissue replacement strategies. [11,12] Thus it would be interesting to study the mechanism by which the ksharas exert their effect.

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How to cite this article: Shetty P, Mandi BS, Gouda HP. Evaluation and comparison of buffering capacity of apamarga and phalasha ksharas. Int J Health Sci Res. 2020; 10(8):90-95.

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