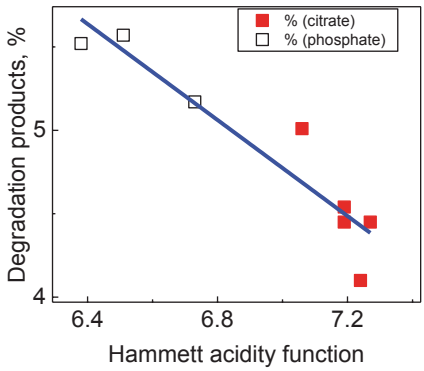

Fig. 6 Total degradation products detected in freeze-dried formulations after storage of lyophilized API with either citrate or phosphate buffer as a function of the Hammett acidity function determined in lyophiles using phenol red *API* active pharmaceutical ingredient

changes from the relatively dilute aqueous solution to an organic (sugar) matrix with low water content and lower polarity. Finally, preferential inclusion of a basic component (disodium phosphate) by ice crystals and resulting acidification of the remaining freeze-concentrate can also be proposed, as discussed above. In either case, the use of pH indicators allowed detection of the difference in the apparent acidity of formulations prepared with citrate versus phosphate buffer. The acidic shift observed with phosphate buffer after freeze-drying correlated the higher degradation rate of the acid-sensitive active ingredient in the phosphate formulations. It means that, in order to achieve a comparable stability with phosphate buffer, phosphate formulations should be lyophilized from a solution with a higher pH than citrate formulations.

Conclusion

Lyophilization of biologics is the most common approach to achieving a robust and stable drug product. The design of a formulated biologic consists of careful selection of excipients and a lyophilization process that maintains biologic activity, promote stability on storage and achieve pharmaceutical elegance and patient acceptability. A critical aspect of this process is knowing how acid–base relationships play a role in the physical and chemical integrity of the therapeutic agent, thus, the selection of a buffer system that maintains optimal pH is paramount. The complex relationships between the extent of proton transfer and temperature, the effect of temperature and dielectric constant on the apparent pK_a and effective ionization states of acids and bases complicate this seemingly simple principle. Furthermore, the potential for crystallization of buffers during cooling and freeze concentration may lead to unexpected changes in the apparent pH leading to irreversible physical or chemical changes that lower therapeutic activity. An understanding of how the therapeutic agent contributes to solution pH, as well as how the addition of stabilizers such as sugars can alter water activity and media polarity is essential to designing buffer system that achieves the desired control of proton transfer and ionization extent. Some buffers, such as phosphate buffers can have a greater propensity to