Stability of Suiphacetamide Eye drops at Higher Temperature

Abstract
Suiphacetamide solutions at higher temperature degrade to its hydrolysed product, sulphanilamide, with a first-order rate constant. The degraded products are identified and characterised by chromatography and spectrophotometry. Attempts are made for the evaluation of kinetics, entropy and enthalpy changes activation energy and frequency factors during heating of sulphacetamide solutions. The determination of these thermodynamic elements is useful in prediction of the stability of suiphacetamide eye-drop formulation at elevated temperatures (JPMA 33: 168, 1983).

Introduction
Suiphacetamide solutions in different concentrations ranging from 5 to 30% are used as ophthalmic drops for various eye infections. In the study of the stability of suiphacetamide eye-drops, many workers, Whitter (1949, 1950), Anderson and Maudson (1963), Fletcher and Norton (1963), Dickenson (1963) have studied the effects of temperature during sterilization and autoclaving of ampoules. On heating at 100-120°C, sulphacetamide solutions get hydrolysed into sulphanilamide which crystallise on cooling. The loss 1.5 to 1.0% occurs in suiphacetamide concentration during heating between 100-120°C (Anderson, 1966; Davies et al., 1970). Clarke (1967) suggested the prevention of crystallisation of sulphanilamide during sterilization by buffering the solution of suiphacetamide at pH 9 to 9.5. According to Kulesh and Bugrim (1968) at 100°C showed no colour change even after one year of storage.

In this investigation, stability of sulphacetamide solutions has been checked between pH 1 to 13 at 70°, 80° and 90°C for 12-15 days. The degradation products have been identified by chromatography and spectrophotometry. The kinetics, activation energy, frequency factor, enthalpy and entropy changes are determined during heating of suiphacetamide solutions.

Material and Methods
5ml portions of 10^{-2}M buffered solutions of pure crystalline sulphacetamide (Sigma, USA) of pH 1 to 13 are filled and sealed in amber glass ampoules. These ampoules are boiled twice for one hour in water, before being filled with sulphacetamide solutions, to remove any trace of alkali, and are finally placed in an oven adjusted to 70°, 80 and 90 with ± 1°C fluctuations in temperature. Samples are removed at suitable intervals chilled in ince to stop the reactions are examined by chromatography and spectrophotometry.

Results
(i) Chromatographic Identification: Thermally heated samples are chromatographed on the fluorescent silica gel G 254 and alumina layers. The solvent systems T_1 to T_{10} have been employed (Table I)
and the degradation products are detected either under the UV light at 254nm and 320nm or sprayed
with iodine vapours or Bratton-Marshall reagent.

(ii) Spectrophotometric Determinations:
(a) Synthetic Mixtures and Extinction Coefficients:
Synthetic mixtures of authentic sulphacetamide, sulphanilamide, and azobenzene-4, 4’-disulphonamide are prepared. The concentration of each component in a mixture is measured on a Pye
Unicam SP 500 spectrophotometer and evaluated by solving the simultaneous equations. The % error
of reproducibility of the mixture analysis technique lies within ± 4 ‘%’. After confirming the validity of
Beer’s law, the molar extinction coefficients, $\epsilon$ (Imol$^{-1}$ cm$^{-1}$) of sulphacetamide, sulphanilamide and
azobenzene-4, 4’-disulphonamide at pH 4.0 are determined by using the least square method. The
value of molar extinction coefficients at 258, 268 and 320nm are given below.

Sulphacetamide:
\[ \text{at } 7258 \text{ (max)} = 13.530 \times 10^6; \text{ at } 7265 \]
\[ (max) = 17.132 \times 103; \text{ at } 320 = 0.1 \times 10^{-3} \]

Sulphanilamide:
\[ \text{€ at } 7258 = 15.254 \times 10^0; \text{at } 7268 = 11.632 \times 10^0; \text{at } 7320 = 0.106 \times 10^3 \]

**Azobenzene -4, 4’-disulphonamide:**
\[ \text{€ at } 7258 = 3.128 \times 10^0; \text{at } 7265 = 3.945 \times 10^0; \text{at } 7320 \text{(max)} = 13.15 \times 10^3 \]
Known dilutions of heated solutions of sulphacetamide with buffer of pH 4.0 are made and the absorbance values are measured. The concentrations of sulphacetamide and sulphanilamide in heated solutions have been calculated by solving the simultaneous equations. Analysis is carried out at pH 4.0 because a distinct difference in the absorption maxima of both sulphacetamide, 7% max 268nm, and sulphanilamide, 7max 258nm, exist.

**(b) Characteristics of sulphacetamide and its decomposition products:**
Sulphacetamide: m.p. 184°C (lit. Clarke, 1969) m.p. 181-184°C; Rf. O.42 (chloroform : ethyl alcohol : heptane, 1:1:1) (lit. (Klein and Kho, 1962) Rf.O.42); UV aq79 (max) 259nm (log \( \varepsilon \) 4.24) lit. Bohme and Wanger, 1942) max 260nm; IRKBrV max 1320, 1155 (S=O, stretching), 1585 (-NH deformation) and 1690cm' (-CoCH3 stretching). The values are in agreement with those of Clarke (1969) and Bellamy (1964). Sulphacetamide: m.p. 166°C (lit. Clarke, 1969) m.p. 164.5° - 166.50°C; Rf. 0.53 (Chloroform ethyl alcohol: heptane, 1: 1: 1) (lit. Klein and Kho, 1962) Rf. 0.53; Tmax (pH 7) 258 nm (loge 4.18) lit. Elvidge, 1941) 7max 259nm; IR. (KBrv max 1310, 1150 (S=O stretching) and 1591 cm' (-NH deformation). The frequencies values are in agreement with those of Clarke (1969).

Azobenzenes -4, 4’-disulphonamide: m.p.311°C dec. (lit. (Seikel, 1940) 312°C dec.); Rf.O.17 (n-butanol: acetic acid: water, 50:15:35) (lit. (Clarke, 1969) T max336cmn); IRKBrV max 1350, 1170 (S=O stretching Rf. o.17); /max (0.1 N NaOH) 336nm (log C 4.16) (lit. Pondula, 1969) / max 3 36nm ; and 1410cm', N=N stretching).

**Discussion**

It is known that sulphacetamide solutions at high temperatures facilitate hydrolysis leading to the formation of sulphanilamide and the behaviour of these thermo-chemical reactions have been studied by many workers (Whittet, 1949, 1950; Fletcher and Norton, 1963; Anderson, 1966; Davies et al., 1970). The chromatographic examination of the heated solutions of sulphacetamide showed the presence of sulphanilamide -4.0 and azobenzene-4,4’-disulphonamide. The chromatographic separation of the decomposition products of sulphacetamide leads to the development of a multicomponent spectrophotometric analysis. The gradual decrease in the absorption of heated solutions at 268nm of sulphacetamide and simultaneous increase in absorption at 258nm and in the region of 280-300nm is in accordance with the absorption characteristics of sulphanilamide formation and coloured azobenzene -4,4’-disulphonamide. The presence of azo derivative of sulphanilamide is detectable after at least 200 hours of heating between 70° to 90°C of sulphacetamide solutions at pH 5-11. Azo derivative of sulphanilamide is similar to azobenzene in having the band characteristic to it -p transitions. In the light of experimental observations and kinetic data calculated in present study, the following scheme may be proposed for degradation of sulphacetainide (S0) (Ahmäd, 1978).

Sulphacetamide k1 \( \to \) Sulphanilamide (S1) k2 \( \to \) Azo deriv. The hydrolysis of sulphacetamide (S0) to sulphanilamide (S1) is a first-order (k1) reaction and the oxidation of (S1) to azo derivative is a second-order(k2) reaction (Fig. 1).
The overall rate of hydrolysis of sulphacetamide is found independent during heating, some fluctuations in rate values are possible towards strong acidic/alkaline media which may be due to ionic mobilities of the molecules (log-pH profiles in Fig.2).

Fig.1. First-Order plots for $10^{-3}$M Sulphacetamide at 70°, 80° and 90°C.
The activation energy expresses the influence of temperature on reaction velocity. The reacting molecules must acquire this energy in order to undergo degradation. The higher the value for the energy of activation the greater is the stability of the substance to temperature. In addition the frequency factor determination also help in determining the frequency of collisions which is expected between the reacting molecules for a given reaction. The values of activation of energy factors for the sulphacetamide solutions (103M) at pH 1 to 13 have been determined by the use of Arrhenius equation $K = A e^{E/RT}$ where $K$, is specific rate of degradation.

A, is frequency factor (Table II) $E$, is activation energy, also called AF, free energy of activation (Table II), $R$, is gas constant (1.987 calories degree ‘mole l), $T$, is temperature in degree absolute. The entropy and enthalpy changes are also calculated for the sulphacetamide solutions at different temperatures (Table II and Fig. 3,4).
Fig. 3. Arrhenius plots for the determination of activation energy for the $10^{-3}$ M sulphaectamide solutions.
The values calculated for activation of energy (E) for suiphacetamide solution of pH 1 to 13 lie in the range of 8 to 15 K. cal. mole'. The lower values of activation energy (Relatively) of suiphacetamide are obtained at pH 3 and pH 11, showing relatively less stable species of molecules which may be due to catalytic effect of buffer ions at these particular pH values.

References