above freezing to 90° F and never noted any significant change in the pattern of the markings. At low temperatures the color of the animal as a whole is quite dark, but the pigment spots are still clearly out-

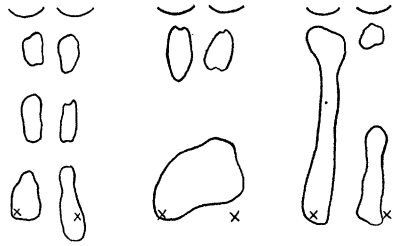

FIG. 2

lined. We have used this method of identification for three years in several hundred frogs and have found it to be consistently satisfactory.

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MONOTHIOGLYCOL

MONOTHIOGLYCOL,[1] CH_2OHCH_2SH (also designated thioglycol, monothioethylene glycol and β- or 2-mercaptoethanol), is a useful non-nitrogenous sulfhydryl reagent for protein investigations. It is a colorless liquid (b.p. 69–70°(28 mm)),[2] completely soluble in water and most organic solvents. Smythe[3] showed that monothioglycol resembled cysteine and glutathione in its reaction with iodoacetate and iodoacetamide, although the reaction time for monothioglycol was slower. Fischer[2] measured the normal oxidation potential and found it approximately equal to that of cysteine (0.44 volt).

The sulfhydryl content of monothioglycol solutions can be determined readily by iodine titration. As with other SH compounds, dilute solutions in acid are more stable than those in the neutral or alkaline pH range (Table I). Monothioglycol can be added to buffer

TABLE I

PERCENTAGE LOSS OF SH FROM 0.1 M MONOTHIOGLYCOL SOLUTIONS IN BUFFER SOLUTIONS[a]

Days	Citrate (0.1 M) pH 3.92	Phosphate (0.067 M) pH 6.74	Phosphate (0.067 M) pH 7.89
1	0.4	3.3	4.6
2	1.5	5.6	7.3
3	2.1	7.7	10.6
5	3.1	13.0	18.7
14	9.0	46.0	51.0

[a] In half-filled glass-stoppered clear glass bottles, diffuse light, 25–30°.

solutions without appreciably changing pH or ionic strength (Table II). It gives a deep red nitroprusside test. The disulfide oxidation product, in contrast to cystine, is soluble in water in all proportions.

TABLE II

CONDUCTANCE OF MONOTHIOGLYCOL

Solute	Solvent	Specific Conductance[a] reciprocal ohms
0.1 M Monothioglycol	0.001 M Acetate buffer (pH 6.5)	5.2×10^{-5}
0.1 M Thioglycolic Acid	0.001 M Acetate buffer (pH 6.5)	3.4×10^{-3}
None	0.001 M Acetate buffer (pH 6.5)	4.4×10^{-5}
0.125 M Monothioglycol	0.1 M Acetate buffer (pH 6.5)	3.4×10^{-3}
None	0.1 M Acetate buffer (pH 6.5)	3.5×10^{-3}
0.125 M Monothioglycol	0.1 M Glycine–NaOH buffer (pH 11.8)	3.6×10^{-3}
None	0.1 M Glycine–NaOH buffer (pH 11.8)	3.8×10^{-3}

[a] Measured at 0.8° C. with freshly prepared solutions.

That monothioglycol is a reducing agent for the disulfide linkages in proteins is indicated by its effect in increasing the solubility of keratins, decreasing the viscosity of enzyme-free wheat gluten dispersions and activating papain. It should be found useful in providing a suitable environment for preventing the air oxidation of reduced proteins during dialysis without interfering with subsequent mobility determinations. Details of the experiments mentioned will be described in later publications.

Dr. C. B. Jones studied the keratin solubilities and Dr. Hans Lineweaver performed the papain assays.

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[1] The Carbide and Carbon Chemicals Corporation kindly furnished a generous sample.

[2] E. K. Fischer, *Jour. Biol. Chem.*, 89: 753, 1930.

[3] C. V. Smythe, *Jour. Biol. Chem.*, 114: 601, 1936.

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