weight proteins. However even after injury by freezing, the protein concentration in lymph did not reach that of plasma.

After these immediate changes additional proteins appeared in the lymph which seemed to originate in the damaged tissue. One major protein band was found in lymph collected after injury, which migrated faster than α_2 macroglobulin but slower than the haptoglobins. It could not be detected in control lymph or in plasma collected after injury. The density of this protein band increased with increasing severity of injury. After a 60°C burn its appearance was delayed for about one hour after reaching a maximum at 2–3 h; whereas after an 80°C burn it appeared within the first hour.

This component was eluted from Sephadex G-150 superfine with proteins of molecular weight 70,000.

When lymph collected after injury was chromatographed on Whatman DEAE-cellulose, the additional protein band separated into three components. One contained 5–10% of the total lactate dehydrogenase activity and a second contained about 60% of the total creatinine kinase activity of the lymph. The third component was not associated with these enzyme activities.

Homogenates of muscle but not of skin, contained a major protein component having the same electrophoretic mobility as the extra protein in lymph collected after injury.

Chromatography of this muscle homogenate on DEAE cellulose and Sephadex G-150 superfine resulted in the separation of a protein band with the same elution characteristics as that found in lymph after inury.

The protein bands in lymph and in muscle homogenate are therefore indistinguishable on these criteria and their presence in the lymph is indicative of muscle damage.

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A quantitative assessment of tissue changes accompanying homograft reaction: changes in tissue dry weight, DNA and moisture content in rabbit skin homografts

E. BITTERLI and M. K. JASANI*

CIBA Laboratories, Horsham, Sussex

Until recently the influence exerted by various chemical substances and immune sera upon the strength and rapidity of onset of skin homograft rejection has been evaluated chiefly on the basis of the prolongation of graft survival, and this has been judged either as a change in the outward appearance of the graft indicative of cessation of blood flow through it or as the actual shedding of the graft itself.

In the present study experiments were made on Norfolk White rabbits using skin removed from New Zealand Whites. Six homografts were made on one leg or both legs of each recipient as described previously (Jasani & Lewis, 1971). Each graft was weighed prior to grafting and subsquently on removal postoperatively. Then it was subjected to dry weight analysis using a modification of the method of Schneider (1945) and DNA estimation (Burton, 1956). Moisture content was estimated from the difference between the fresh weight and the total dry weight of each graft. The final values for each parameter were compared with those obtained from a similar analysis of a portion of non-grafted donor skin.

In the first series of experiments all the six grafts were removed at the same time interval after grafting. Although there was a statistically significant increase in all three parameters in each of the six grafts from the fourth day onwards, the extent of the increases in dry weight and moisture content of grafts on the medial aspect were significantly lower than those in the lateral grafts. These findings indicated that these two parameters and to a lesser extent DNA content, were significantly influenced by the anatomical position of the graft.

This was shown more clearly in a second series of experiments in which pairs of grafts from the same anatomical site on both legs were removed at daily intervals after grafting. When these were compared there was no statistically significant difference between the dry weight, moisture content or DNA content of homologous pairs.

Since changes in the three parameters did not parallel one another, even in the grafts taken from the same site, it is concluded that each index may represent a separate tissue event. Changes in dry weight may parallel the increases in vascularity of the graft, an event which comes to a halt with the onset of rejection; increases in the moisture content may reflect changes in vascular permeability and those in the DNA content may indicate increases in the cellularity of the graft.

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Effects of frusemide on cyclic AMP binding in the toad bladder

D. R. FERGUSON and B. R. TWITE*

Department of Pharmacology, Medical School, University of Bristol

Frusemide, a rapidly acting diuretic, is known to antagonize the stimulation of active sodium transport produced by vasopressin in toad bladders (Ferguson, 1966).

Using a modification of Gilman's (1970) isotope displacement assay for cyclic AMP, we have investigated the effect of frusemide on cyclic AMP binding to rabbit muscle cyclic binding protein. There was a significant reduction in cyclic AMP binding in the presence of $2{\cdot}5 \times 10^{-3}$M frusemide ($P<0{\cdot}01$).

When the experiments were repeated using toad bladder epithelium cyclic binding protein, we found a significant displacement of cyclic AMP at a lower frusemide concentration.