to enhance the linearity of the response and the total acid output was determined from the area under the curve of pH plotted against time.

An integrated response is obtained if the acid is accumulated in the measuring chamber and the total output is then given by linear measurement from the pH versus time curve. In the present experiments this was achieved by recirculating the effluent through the stomach in a closed perfusion system.

Acid concentration can be determined by titration or by pH measurement, but we have found it more convenient to use a conductimetric system. The equivalent conductances of the ions found in the stomach are H^+ 350, Cl^- 70, Na^+ 50.9, K^+ 74.5 and HCO_3^- 50 approximately. Changes in conductivity will therefore be due predominantly to HCl secretion, as neutral salts will give relatively small effects. Figure 1 shows the results obtained in such a system when isotonic glucose

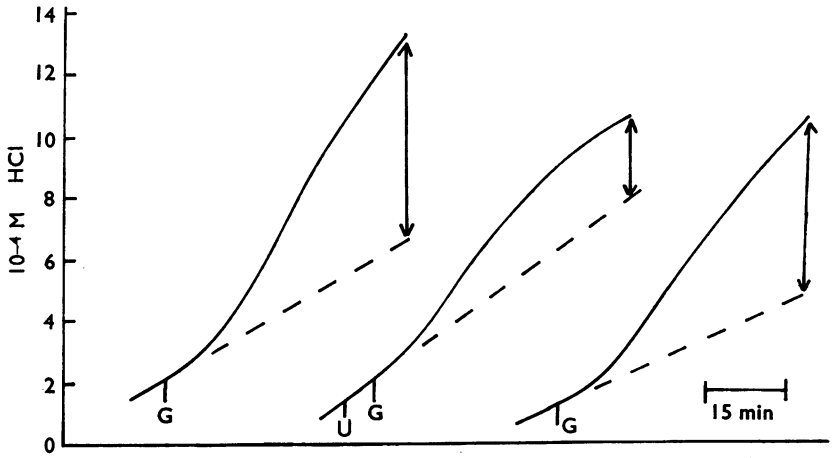

FIG. 1. Reperfusion of urethanized rat stomach. Conductivity changes due to three successive injections (at 1–2 hr intervals) of 0·1 μg gastrin II (G). Middle tracing: inhibitory effect of a dose of urogastrone (U) extracted from 200 ml. of human urine. Ordinate: conductivity in terms of equivalent concentration of HCl; abscissa: time; — — —, basal secretion.

solution was reperfused through the stomach of an anaesthetized rat. The conductivity meter (designed in conjunction with T. Sales) gave a linear output over the acid concentration range 0–10^{-3}M HCl. The effects of 100 ng of gastrin II and the inhibitory effect of a dose of urogastrone extracted from human urine, are shown.

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The storage and binding of gastrin

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The distribution of gastrin in subcellular fractions prepared by differential centrifugation of sucrose homogenates from antral mucosa of guinea-pig was studied. The fractions were characterized by estimation of RNA, DNA, succinic dehydro-

genase, acid phosphatase, protein and hexosamine. Gastrin content was determined by biological assay using the perfused stomach of the anaesthetized rat (Amure & Ginsburg, 1964). In preliminary experiments the conditions of homogenization and centrifugation were used which, when applied to liver, would have yielded nuclear, mitochondrial plus microsomal and supernatant fractions. Although the mitochondrial plus microsomal fraction contained 20–33% of the total gastrin, large amounts of all the biochemical markers and of gastrin were found in the first sediment; this is probably due to the presence of mucus causing heavy cross-contamination of the fractions and incomplete cell disruption (Hubscher, West & Brindley, 1965). A gastrin-rich sediment was prepared by centrifugation of the "nuclei-free" supernatant at 20,000 *g* for 20 min which, after resuspension, was subjected to centrifugation in a modified density gradient (Baker, 1959). These experiments showed that the gastrin particle was distinct from mitochrondria (succinic dehydrogenase activity) but was not clearly separated from lysosomes (acid phosphatase activity).

Evidence for the occurrence in the gastrin-rich granules of a macro-molecule capable of forming complexes with gastrin was obtained from experiments in which the elution in molecular sieve chromatography (Sephadex G-50) of free gastrin and the gastrin activity of osmotically disrupted granules was compared.

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The role of protein synthesis inhibition in the prevention of morphine tolerance

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We have previously reported that tolerance rapidly develops to the analgesic effects of morphine-like drugs when these are infused intravenously into conscious rats at selected rates. We also showed that concurrent administration of actinomycin-D reduced or prevented the development of tolerance (Cox, Ginsburg & Osman, 1968). Since actinomycin-D inhibits DNA directed RNA synthesis and hence *de novo* protein synthesis, the effects on tolerance development of some other drugs which inhibit protein synthesis have been studied.

Cycloheximide, puromycin, 6-mercaptopurine (6MP) and 5-fluorouracil (5FU) all reduced the degree of tolerance produced by intravenous infusion of morphine HCl (7.5 mg/kg per hr), although it was necessary to inject 6MP and 5FU intracerebrally to demonstrate this effect. The effect of these drugs (in doses shown to be capable of preventing morphine tolerance) on the incorporation of ^{14}C-lysine into brain proteins has also been measured. Cycloheximide, which inhibits protein synthesis by hindering the movement of ribosomes along messenger RNA, produced an 18% inhibition of ^{14}C-lysine incorporation at a dose (50 μg/kg per hr, intravenously) which markedly reduced tolerance development. Cycloheximide at