SUMMARY

This project was designed to develop a tissue culture technique for gallbladder epithelium to study gallbladder function. Guinea pig gallbladder cells were cultured in eight culture media supplemented with foetal calf serum for up to seven days. Gallbladder epithelium survived in all media with no difference in growth assessed by light microscopy. Two media were further evaluated by electron microscopy after culture for 14 days. Ultrastructural preservation of epithelial cell morphology was maintained with both media. Mucous secretory vesicles disappeared from cell apices together with increased cellular glycogen content and the appearance of lipid droplets in the cytoplasm and lamina propria. Medium renewal, addition of insulin, hydrocortisone or collagenase to the medium, and treatment with trypsin before culture, were without effect.

Gallbladder epithelial cells cultured for ten days responded to cationized ferritin in the same way as gallbladder *in vivo*. Cells cultured for two days failed to bind tripotassium dicitrato-bismuthate to the apical surface. Instead, this marker permeated through the basal lamina and appeared in the intercellular space and at the periphery of lipid droplets. This may indicate a secretory function of the cultured cells.

Selected enzyme activities showed marked day to day variations with an overall decrease throughout

culture. Incorporation of $[^3H]$ thymidine into DNA was maximal after 2.5-3 days and then remained constant up to seven days. $[^3H]$ Leucine incorporation was maximal after one day and remained constant up to seven days. These data suggest biochemical stability between days 3-7 but further studies are required.

After one day of culture in the presence of 1% bile (V:V) cells became oedematous with damaged organelles. It is particularly important to wash human gallbladder epithelium thoroughly to remove all traces of viscous gallbladder bile.

Human gallbladder cells cultured for seven days retained the ultrastructural features of epithelial cells with increased glycogen content and the appearance of lipid droplets. They also responded to cationized ferritin in the same way as guinea pig but differed in the presence of secretory vesicles throughout culture.

This procedure offers a valuable method for study of human gallbladder function.