In all animals subjected to PCS, the liver weight decreased markedly (Table). This liver 'atrophy' corresponded to the general experience in this and other laboratories[1-4] and implied a well functioning PCS. 10 days after PCS, GGTP activity had increased tenfold and remained at this level throughout the period of observation. In the control group, the activity was almost undectable (Table).

Our findings of very low activity of GGTP in normal adult rat livers are in agreement with earlier observations[5,8-10]. In these animals GGTP activity may be found histochemically in the endothelial cells of periportal vessels, in bile ducts and in Kupffer cells, whereas virtually no activity can be detected in hepatocytes[11]. The high enzyme activity in certain rat hepatomas which contain glycogen, produce bile and have a high level of glucose-6-phosphatase[7], suggests that GGTP may be activated in parenchymatous liver cells during the carcinogenic process. This has been interpreted as a re-acquirement of a biochemical feature which predominates in the fetus, but is repressed in the adult liver[7].

The microscopical anatomy of the liver of shunted rats looks grossly normal[2,12]. Measurements of DNA content have suggested that the initial loss of liver mass is due more to a reduction in cell size rather than in cell number[13]. Some observers noted a slight increase in the number of Kupffer cells[14]. Even though we have no information about the histochemical distribution of GGTP activity in shunted rats, the generally recognized minor morphological alterations do not appear sufficient to explain the tenfold increase in enzyme activity after PCS. The impressive elevation in hepatic GGTP-content could therefore be interpreted more reasonably in the context of the similar findings in neonatal liver and in chemically induced rat hepatomas. According to this view, the shunt consequences might be associated with derepression of an enzyme, normally present only in the embryonal liver. Further studies are required to demonstrate whether such a mechanism is limited to a few specific enzymes or represents a form of generalized hepatic immaturity after PCS.

Zusammenfassung. In Leberhomogenaten von Ratten mit einem portocavalen «Shunt» wurde gegenüber unbehandelten Kontrolltieren eine zehnfache Vermehrung der Aktivität der γ-Glutamyltranspeptidase festgestellt. Dieser Befund könnte als Derepression dieses Enzyms gedeutet werden, da ähnlich gesteigerte Enzymaktivitäten bisher nur bei embryonalen Lebern und bei chemisch induzierten Hepatomen gemessen worden sind.

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The Use of Alfalfa Residual Juice for Production of Single-Cell Protein

One of the most serious problems facing the world nowadays is the provision of an adequate diet for a rapidly increasing population. This can be especially observed in Latin America, where the protein shortage constitutes the main nutritional deficiency[1]. Under these circumstances, the developing of processes for conversion of waste materials into edible microbial foods is of paramount importance.

Several processes for the obtention of leaf protein concentrate have been reported in the literature[2-4]. During these processes, alfalfa residual juice is obtained as a byproduct and the feasibility of its use for biomass production was the aim of the present investigation. Earlier results have been published elsewhere[5,6].

Materials and methods. The culture selected for its highest growth rate was *Candida sp.*[5]. The medium used in fermentation contained 0.5 g of K_2HPO_4/l of alfalfa residual juice. Fermentation was conducted at 30 °C and at a controlled pH of 5.0. The fermentor used throughout this study was a 14 l total capacity (Fermentation Design Inc.) with a working volume of 9 l. Stirrer speed was of 400 rpm. Simple on-off control was used, arranged so that the dissolved oxygen partial pressure did not fall below 0.10 atm. Inoculum was grown in 250 ml Erlenmeyer flasks with 50 ml of medium, and incubation was for 1 day. Dry weight was determined gravimetrically after drying to constant weight the cell material at 100 °C.

Approximately 33 mg of dried biomass was subjected to acid hydrolysis with 6*N* HCl at 110 °C for 48 h. The hydrolysate was evaporated under vaccum to dryness and then resuspended in 20 ml of distilled water. This step was repeated 3 times and then the hydrolysate was taken up in a pH 3.1 sodium acetate-acetic acid buffer. The analysis for amino acid was carried out upon a 0.5 ml sample using a Hitachi Perkin-Elmer amino acid analyzer (Model KLA-3B). Tryptophane was determined by a colorimetric method[7]. The amino acid content of soybean meal was also determined by the procedures mentioned above.

Results and discussion. The stationary phase was practically attained after the 50th h of cultivation, as is shown in Figure 1a. The maximum productivity p was

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0.38 g/l^{-1}h^{-1}, which was reached at the 45 th h (Figure 1b). In Figure 1c a graphic representation of the specific growth rate and generation time is shown. The maximum specific growth rate (μm) was of 0.069 h^{-1}, corresponding to a generation time (g) of 10 h.

In Figure 2, the essential amino acid content of the biomass with 49% of protein was compared with the standard set-up by FAO[8], and the soybean meal protein. The amino acid composition of the protein of *Candida sp.* grown on alfalfa residual juice, is favorably compared to the FAO standard as well as to soybean meal protein. Approximately 38.8% of the amino acids in the single-cell protein obtained were found to be essential, while in soybean meal this Figure was 31.4%.

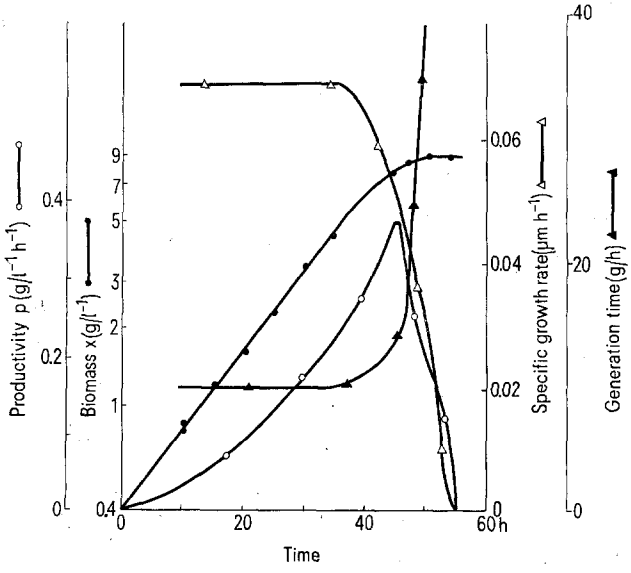

Fig. 1a–1c. Cultivation of *Candida sp.* on alfalfa residual juice. Biomass x (g/l^{-1}), productivity p (g/l^{-1}h^{-1}), and specific growth rate (μm/h^{-1}) and generation time (g/h) respectively, in relation to cultivation time.

The production of single-cell protein from alfalfa residual juice seems to be feasible. Since the use of continuous culture would increase the productivity and the obtention of leaf protein concentrate would be more economical, such studies are being carried out.

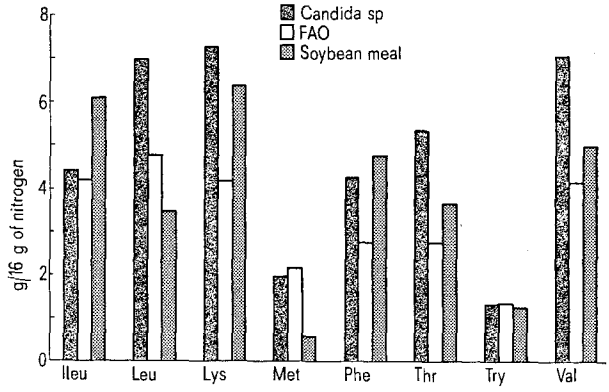

Fig. 2. Essential amino acid distribution in *Candida sp.* grown on alfalfa residual juice, FAO standard-reference protein, and soybean meal. Abbreviations: ileu, isoleucine; leu, leucine; lys, lysine; met, methionine; phe, phenylalanine; thr, threonine; try, tryptophane; val, valine.

Resumen. Jugo residual de alfalfa, obtenido como subproducto durante la elaboración de concentrado proteico de hojas, fue utilizado como sustrato para la producción de proteína unicelular.

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Effect of Various Prostaglandins and Serotonin on Protein Secretion from Rat Exocrine Pancreas

A number of vasoactive substances have recently been demonstrated in pancreatic tissue. Prostaglandins (PG) E_2 and $F_{2\alpha}$ are found in bovine pancreas[1]; their distribution in various cell types, however, is still unknown. Mouse exocrine pancreas can take up 5-hydroxytryptophan and apparently store the decarboxylated amine, serotonin (5-HT) in zymogen granules[2]. The release of these agents during 'stimulus-secretion coupling'[3] is a distinct possibility, 5-HT because of its subcellular distribution and the PG's perhaps in an analogous fashion to their observed release by catecholamines and adrenocorticotrophic hormone[4]. To test if these substances play any regulatory role in secretory events their direct actions on basal and stimulated release of secretory protein was monitored in rat exocrine pancreas in vitro.

Materials and methods. Female Wistar rats, 180–225 g, were decapitated; pancreata were excised, and trimmed of adherent mesentary and fat in chilled incubation medium. Pancreatic fragments (10–20 mg) were 'Pulse-labelled' with ^{3}H-leucine according to the technique of JAMIESON and PALADE[5] in 10 ml Krebs-Ringer bicarbonate buffer supplemented with 10 m*M* glucose and L-amino acids (without L-leucine) in concentrations suggested by EAGLE[6]. Following a 1 h 'chase period' (1 m*M* L-leucine present) tissue samples were distributed in 2 ml of buffer and incubated an additional 60 min in the presence or absence of added stimuli. Termination of incubation and measurement of secreted ^{3}H-labelled protein were as previously described[7]. Release of protein from the tissue

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