Results. The calibration scale for Figure 1 is 20 mV per horizontal division and 100 msec per vertical division. The upper portion of the trace in A, prior to impalement is O mV and the upper trace in B is a marker at O mV. Figure 1A shows a typical of a cell in a preparation superfused with 95% N_2, 5% CO_2 in buffer; the resting potential is –90 mV. In Figure 1B, with the electrode in the same cell, the buffer was changed to95% air, 5% CO_2 and after 15 min a new resting potential of –60 mV was reached.

The results of experiments on 2 separate ducti are shown in the Table. The mean resting potential in the presence of 95% N_2, 5% CO_2 was 81.7 ± 3.1 mV while in the presence of 95% air, 5% CO_2 the potential was 50.8 ± 7.8 mV. The mean attenuation of 30.9 mV in the air mixture was statistically significant using the student's *t*-test at $P < 0.001$.

Experiment	No. of cells impaled	Cell letter	Membrane potential in 95% N_2, 5% CO_2 (mV)	Membrane potential in 95% air, 5% CO_2 (mV)
1	2	a	−80	−40
		b	−70	−20
2	4	c	−80	−50
		d	−80	−60
		e	−90	−75
		f	−90	−60
		$\bar{X} \pm$ SEM	81.7 ± 3.1	50.8 ± 7.8

Significant difference $P < 0.001$

Discussion. The results show that ductus cells depolarize when exposed to oxygen concentrations which cause contraction of normal ductus cells. Whether this depolarization is necessary for producing ductus contraction is not yet known.

SOMLYO and SOMLYO[10] divided vascular smooth muscle into 2 electrophysiological classes based on drug-induced contractions. The classes were: 1. single unit vascular smooth muscle; and 2. multi-unit smooth muscle. Action potentials accompany contraction in single unit smooth muscle (e.g., canine and rabbit mesenteric veins). Multi-unit vascular smooth muscle (e.g., aorta and pulmonary artery) does not exhibit spike action potentials but depolarizes to a degree commensurate with the magnitude of contraction. The fact that action potentials were not found in the presence of oxygen levels which normally cause contraction suggests that ductus smooth muscle may be classified as multi-unit vascular smooth muscle. However, more data are needed to confirm this. Future impalements must be studied at higher amplification (10× to 100×) to see if there are low amplitude spikes (1–3 mV) riding on the initial depolarization, as was shown by KAJIMOTO et al.[11] in their study on the smooth muscle cells of guinea-pig seminal vesicle.

Further work must also be done to study the relationship between the separately recorded electrical and mechanical events.

Zusammenfassung. Membran-Potentiale vom Ductus arteriosus der Ziege wurden im ruhenden Zustand aufgezeichnet. Die Zellen depolarisieren, sobald sie mit einer Kontraktion auslösenden Konzentration Sauerstoff in Berührung kommen.

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Inhibitory Influence of Ligation of the Small Intestine on Gastric Secretion in the Pylorus-Ligated Rats

It has been well known and repeatedly confirmed that the pylorus ligation causes a gastric hypersecretion in rats[1–3]. The mechanisms are not fully known but the vago-vagal reflex elicited by the stimulation of pressure receptor in the antrum mucosa was postulated as an important factor by BRODIE[2]. Certainly, central nervous system depressants, ganglionic blockers, anticholinergic agents, and vagotomy could well inhibit the secretion in pylorus-ligated rats[4–6]. As the other inhibitory procedures, duodenal souring[7], presence of fat in the duodenum[8], ligation of the common bile duct[9], adrenalectomy[10] and hypophysectomy[11] have been also known to suppress the gastric secretion. These facts indicate rather complex mechanisms involved in the secretion process. The present study will deal with the extensive inhibitory influence of a ligation of the small intestine on the gastric secretion in pylorus-ligated rats, with or without acute fistula.

Materials and methods. Male Donryu rats (195–220 g) were used. Following 24 h fast, while given water ad libitum, the animals were subjected to simultaneous ligations of the pylorus[1] and several parts of the small intestine under ether anesthesia, as shown in the Figure. Part A is an upper part of the duodenum (about 2.0 cm distal to the pylorus, i.e., just orad to the entry of the common bile duct). Part B is a lower part of the duodenum (around the ligamentum of Treitz). Part C is a middle part of the jejunum (about 15–20 cm distal to the pylorus). 7 h later, the animals were sacrificed by an overdose of ether, the stomach being removed and gastric contents collected and centrifuged. Titratable acid output was calculated by multiplying the volume and acidity which was measured by titrating a 1 ml sample with 0.01 *N* NaOH to pH 7.4 on a Hitachi pH meter. Average values were given for data

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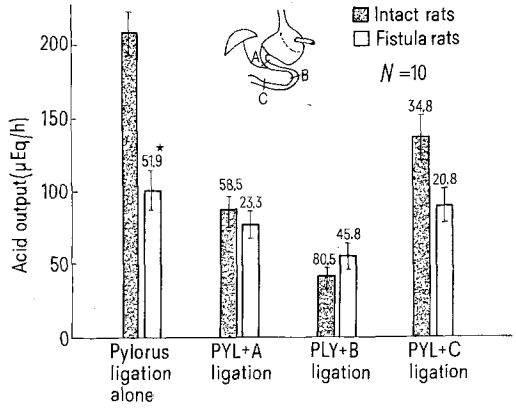
Intestinal contents retained by the simultaneous ligations of the pylorus and several parts of the small intestine in rats

Procedures	Intact rats			Fistula rats		
	N	Volume (ml)	pH	*N*	Volume (ml)	pH
1. Pylorus plus A ligation	10	0.8 ± 0.2	7.43 ± 0.06	10	0.7 ± 0.1	7.44 ± 0.07
2. Pylorus plus B ligation	10	1.6 ± 0.1	6.60 ± 0.17	10	2.0 ± 0.2	6.81 ± 0.04
3. Pylorus plus C ligation	10	0.5 ± 0.1	6.76 ± 0.03	10	$1.6 + 0.3$	6.68 ± 0.04
		1:2 < 0.005	1:2 < 0.001		1:2 < 0.001	1:2 < 0.001
P value		1:3 NS	1:3 < 0.001		1:3 < 0.025	1:3 < 0.001
		2:3 < 0.001	2:3 NS		2:3 NS	2:3 < 0.05

All values represent the mean $\pm$ standard error of the mean; NS, non-significant.

obtained from groups of 10 rats. In another experiment, after the concomitant ligations of the pylorus and small intestine, an acute fistula was implanted into the rumen portion of the stomach and brought out through midline incision. A plastic test tube was attached to the cannula, and the rat was placed in an individual cage with a slotted bottom. The stomach was washed with saline several times through the cannula until the solution became clean. Gastric juice obtained for the first 30 min was discarged, then was collected at 3, 2, and 2 h intervals and stored at 4 °C, in order to minimize the change of gastric contents by exposing to air. 7 h later, the gastric juices were pooled together and their contents were analyzed as described above. Both in non-fistula and fistula rats, the duodenal contents, which were retained in the lumen by the ligations, were also examined quantitatively with regard to the volume and its pH. Level of significance was calculated by using Student's *t*-test.

Results. Intact rats. As shown in the Figure, simultaneous ligation of the pylorus and A, B or C significantly reduced the acid output by 58.5% ($P < 0.001$), 80.5% ($P < 0.001$) or 34.8% ($P < 0.01$) respectively, as compared with the pylorus ligation alone. Moreover, the suppression by the ligation of B was found to be significantly greater than that of A ligation ($P < 0.001$). It should be noted that the ligation of C showed a tendency of restoration of the suppression to the control level as compared with B ligation ($P < 0.001$). The intestinal contents in each part of ligation are shown in the Table. In 2 of 10 animals less duodenal juice was found in A ligation group. The ligation of the pylorus and B resulted in a significant increase ($P < 0.001$) of the amount of intestinal juice as compared with A, presumable by secreted bile. However, the ligation of C significantly reduced the contents in comparison with B ligation group ($P < 0.001$). The pH of the juice obtained by B and C ligation showed a significant lowering in comparison with A ligation ($P < 0.001$).



Gastric secretion in the simultaneous ligation of the pylorus and the small intestine in rats with or without acute fistula. Numbers on the column mean the percent inhibition from the corresponding control (pylorus ligation alone). *, Percent inhibition from the intact rats.

Fistula rats. In fistula rats subjected to pylorus ligation, the acid output was reduced by 51.9% as compared with the non-fistula rats as shown in the Figure. However, the suppression rate induced by placing a ligature on the pylorus plus A or C was slight in comparison with the pylorus ligation alone, 23.3% or 20.8%, respectively, being not significant ($P > 0.05$). The ligation of the pylorus and B, in contrast, resulted in a significant suppression to 45.8% ($P < 0.025$). The ligation of C showed a tendency for the restoration of the suppression compared with the B ligation, but not significant ($P > 0.05$). Amount of the intestinal contents by ligation of the pylorus plus B or C was found to increase significantly compared with that of A ligation and their pH significantly lowered ($P < 0.001$), presumably by the contamination of bile acids.

Discussion. As an interesting finding, a ligation of the small intestine, especially the duodenum, was found to have intensive inhibitory influence on the gastric secretion in pylorus-ligated rats. In particular, the degree of the inhibition on the gastric secretion was far greater in intact rats (non-fistula) than in fustula rats. As to the mechanisms of the gastric hypersecretion caused by pylorus ligation in intact rats, gastric distention elicited by a secreted gastric juice was envisaged to be an important factor[2,3]. Even in the present experiment, it was strongly suggested that retention of gastric juice might stimulate the basal secretion. Actually, when the gastric juice was collected through a fistula after the pylorus ligation, the acid output was halved in comparison with that of pylorus ligation alone (non-fistula rats).

It was well demonstrated in several experimental animals that distention of the stomach releases the hormone gastrin by the antrum and elicits vagal reflex leading to the increased gastric secretion[12]. Therefore, the ligation of the duodenum in rats seems to exert its inhibitory influence greatly on the stimulated gastric secretion by antagonizing related humoral and/or neural factors and to a lesser extent on basal secretion. In contrast, the ligation of the jejunum both in intact and fistula rats showed a rather weak suppression, suggesting that the jejunum has less influence on the inhibition in comparison with the upper or lower part of the duodenum. As to the duodenal contents retained in the ligated intestinal lumen, their pH values were nearly neutral, even though the pH values in B and C ligation were lowered as compared with A ligation.

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Thus, the participation of secretin or enterogastron, which is assumed to be released by duodenal souring (around pH 1.5–2.0) or presence of fat in the duodenum[13], appeared to be dismissible from the inhibitory factors. The distention of the duodenum by the secreted juice, as well as the ligation procedure itself, may influence the gastric secretory conditions, by mechanisms so far unknown. However, in dogs distention of the duodenum per se was reported to increase the gastric secretion, because of the act of the so-called 'intestinal phase'[14]. In addition, 2 of 10 animals subjected to A ligation had almost no juice in the duodenum. Thus, the participation of distention of the duodenum to the inhibitory mechanisms may be negligible. Further study concerning the detailed mechanisms seems to be worthwhile.

Résumé. La ligature de l'intestin, particulièrement du duodénum, gêne la sécrétion gastrique chez le Rat à pylore lié avec ou sans fistule fine.

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Do Chickens have Gastrin-Like Compounds?

It has been reported that in chickens gastrin is located in the mitochondrial fraction of the duodenum[1]. Recently, using BLAIR's extraction procedure for gastrin[2], we found no gastrinlike activity in any parts of the chicken's upper gastrointestinal tract, bioassaying it either in an anaesthetized rat[3] or in conscious gastric fistula chickens[4]. However, in the experiments with anaesthetized rats, a very low gastrin-like activity in duodenal extracts could not be completely excluded[3]. It was therefore of interest to reinvestigate the problem radioimmunologically.

From 10 chickens venous blood samples were withdrawn after 18 h of starvation and after subsequent refeeding for 30 min. Serum was obtained by spinning down the clot formed within a few minutes. Another group of 10 chickens was killed by cutting the throat. The upper gastrointestinal tract was dissected into oesophagus crop, glandular stomach, gizzard, and duodenum (the first 10 cm distally from the gizzard). The mucosa was scraped from the organs, homogenized in 4 volumes of ice-cold 1/15 *M* phosphate buffer (pH 7.0), and boiled for 5 min. The pancreas in total was treated in the same way. The boiled homogenates were filled up to the original volume with distilled water and were then frozen. The frozen material was taken by car to one of the authors (H. K.) for the radioimmunoassay of gastrin. The experimental procedure was in principle that described by MCGUIGAN[5] as modified by FEURLE et al.[6] using synthetic human gastrin 2–17 (SHG 2–17) as antigen. The investigator identified the samples only by numbers.

The following concentrations of immunoreactive gastrin-like material (IGM) were found:

Serum	a) after starvation	0 pg/ml
	b) after refeeding	0 pg/ml
Oesophagus		0 pg/g
Crop		0 pg/g
Glandular stomach		0 pg/g
Gizzard		0 pg/g
Duodenum		19 ± 8 pg/g
Pancreas		0 pg/g

The only organ cross-reacting with antibodies against SHG 2–17 was the duodenum. Although the assay was done under blind conditions, none of the duodenal extracts was found to be free of IGM. These results are in agreement with those of BLAIR et al.[1]. However, the concentration of IGM we found was much less than the gastrin-like biological activity found by these authors. This may be due to the fact that chicken's gastrin cross-reacts only very poorly with antibodies against SHG 2–17. If so, it must differ chemically from human gastrin. This hypothesis is supported by the fact that dilution curves of the duodenal extracts are much flatter than those obtained with human gastrin. Presumably, if only 1% or less of the IGM in the duodenal mucosa was detectable by this method, even then the concentration is very much lower than that found in the mammalian antrum. This finding, together with the poor responsiveness of chickens to pentagastrin[4], and the fact that no rise in serum IGM in response to feeding could be observed, makes it unlikely that this IGM is the main source for the humoral control of gastric acid secretion in chickens. To speculate: There is either another big pool of gastrin in chickens hitherto undetected or the chicken represents a species in which the humoral control of gastric acid secretion by gastrin-like compounds is not a very important factor.

Zusammenfassung. Bei Hühnern wurden in Extrakten der Schleimhaut des Duodenums geringe Mengen einer Substanz nachgewiesen, die radioimmunologisch mit Antikörpern gegen synthetisches Humangastrin 2–17 reagiert. Serum, Oesophagus, Kropf, Drüsenmagen, Hornmagen und Pankreas enthielten keinerlei Aktivität. Die gastrinähnliche Substanz spielt vermutlich für die Steuerung der Magensekretion bei Hühnern keine wesentliche Rolle.

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