

Although it appears reasonable to assume that the measured volume corresponds to biliary dead space, it is not possible to define this space in strict anatomical terms. Presumably, the measured volume represents all or a large part of the bile duct system. Further investigations combined with stereological studies of the structures involved will be needed for a better definition of this space. In the absence of precise methods to determine prebiliary delay, ^{14}C -taurocholate may represent a relatively ideal marker substance for measurement of the biliary dead space; firstly it exhibits a much shorter prebiliary transit time than BSP and secondly it signals its appearance in the bile canaliculi by its potent choleretic effect.

Zusammenfassung. Mit der von BARBER-RILEY angegebenen Methode wird das Gallengangsvolumen auf Grund der unbekannten Transitzeit der verwendeten Testsubstanz vom Ort der Injektion bis zum Gallencanaliculus

(präbiliäre Transitzeit) überschätzt. Es wird eine Methode angegeben, bei der ^{14}C -Taurocholat als Testsubstanz benützt wird. Auf Grund der choleretischen Wirkung dieses Gallensalzes ist es möglich, den Zeitpunkt des ersten Erscheinens von ^{14}C -Taurocholat in den Gallencanaliculi zu ermitteln und damit den bisher durch die unbekannte präbiliäre Transitzeit gegebenen Fehler zu eliminieren. Der mit dieser Methode bestimmte Gallengangstotraum der Ratte betrug $2.3 \pm \text{SEM } 0.11 \mu\text{l/g Leber}$.

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Transmembrane Potentials of the Ductus Arteriosus

Using intracellular microelectrodes in taenia coli, BÜLBRING¹ recorded reductions in membrane potentials as well as action potentials in the presence of either stretch or acetylcholine-induced contraction. In addition, electrical recordings²⁻⁴ have provided evidence of attenuation of membrane potentials associated with contraction of vascular smooth muscle. Since reduction in transmembrane potential has been shown to accompany contraction in vascular smooth muscle, it was of interest to determine if the ductus arteriosus fit the pattern, of electrical and mechanical response to the same stimulus, seen in other vascular smooth muscle. Physiological closure of the ductus arteriosus is initiated by an increase in blood oxygen tension which occurs at birth⁵⁻⁸. Events occurring at the cell membrane of smooth muscle cells of the ductus arteriosus which lead to its closure remain unknown. The purpose of this study is to describe the state of the transmembrane potential during exposure to a level of oxygen which stimulates contraction in a normal ductus.

Materials and methods. Ducti were obtained from goat fetuses used in another laboratory involved in fetal pulmonary research. The ducti were surgically removed by cutting the ductal insertions into the pulmonary artery and aorta. The ducti were then cut into longitudinal strips and placed in Krebs-Henseleit buffer bubbled with 95% N_2 and 5% CO_2 , at 37°C, for at least 1 h. After this period of equilibration, a strip of ductus was anchored in the recording chamber of the bath with stainless steel pins. To record

transmembrane potential changes in response to oxygen, the perfusing gas was switched from 95% N_2 , 5% CO_2 to 95% air, 5% CO_2 .

Capillary microelectrodes similar to those described by LING and GERARD⁹ were used to record transmembrane potentials. Only electrodes with a tip diameter too small to be resolved by a dissecting microscope were used (average tip diameter = 0.9 μm). A Grass P-18 D.C. amplifier was used as a cathode-follower stage to a Tektronix 564 B storage oscilloscope. Records were photographed with a PC-2A Nihon Kohden continuous recording camera. Criteria for evaluating an impalement were: 1. clean, rapid shift of the potential; 2. stability at the new level, and 3. clean, rapid return to baseline on withdrawal. Transmembrane potentials were measured in tissue superfused by oxygenated Krebs-Henseleit buffer (95% air, 5% CO_2) or deoxygenated Krebs-Henseleit buffer (95% N_2 , 5% CO_2).

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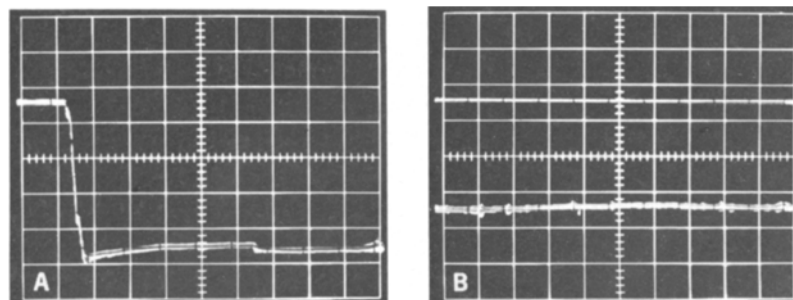


Fig. 1. Potentials obtained on a typical impalement of a cell in a fetal goat ductus arteriosus. In 1A the preparation is super-perfused with 95% nitrogen, 5% CO_2 and buffer; resting potential is -90 mV . In 1B, the buffer was gassed with 95% air, 5% CO_2 ; new resting potential is -60 mV . Calibration is 20 mV per horizontal division and 100 msec per vertical vision.

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 0 mV and the upper trace in B is a marker at 0 mV. Figure 1A shows a typical of a cell in a preparation superfused with 95% N₂, 5% CO₂ in buffer; the resting potential is -90 mV. In Figure 1B, with the electrode in the same cell, the buffer was changed to 95% air, 5% CO₂ and after 15 min a new resting potential of -60 mV was reached.

The results of experiments on 2 separate ducts are shown in the Table. The mean resting potential in the presence of 95% N₂, 5% CO₂ was 81.7 ± 3.1 mV while in the presence of 95% air, 5% CO₂ 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
		$\bar{X} \pm \text{SEM}$	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 SOMLYO¹⁰ 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 ($10\times$ to $100\times$) 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 vagovagal reflex elicited by the stimulation of pressure receptor in the antrum mucosa was postulated as an important factor by BRODIE². 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 souring⁷, presence of fat in the duodenum⁸, ligation of the common bile duct⁹, adrenalectomy¹⁰ and hypophysectomy¹¹ 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 pylorus¹ 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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