Spitze des Wolframdrahtes nicht in tiefere Schichten des Mesenchyms eindringt, damit Blutungen vermieden werden. Das mit einer Spemann-Pipette applizierte Transplantat wird nun mit Hilfe einer Glasnadel auf dem freiliegenden Extremitätenmesenchym ausgebreitet. Anschliessend wird mit einer feinen Pinzette durch Aneinanderdrücken der Wundränder der Amnionsack wieder verschlossen. Durch das über der Extremität nun wieder verspannte Amnion wird das Transplantat an das unterlagernde Mesenchym angedrückt. Nach der Operation werden 2 ml Eiweiss abgesaugt. Das Fenster in der Eischale wird mit wasserfestem Leukoplast verschlossen.

Nach mehrtägiger Wiederbebrütung bereitet die Identifizierung des Transplantats keinerlei Schwierigkeiten (Figur 1). Die Figur 2 lässt die innige Verbindung zwischen Transplantat einerseits und Wirtsmesenchym andererseits erkennen.

Über Ergebnisse, die mit dieser Transplantationstechnik erzielt wurden, wird zu einem späteren Zeitpunkt berichtet werden.

Summary. The right wing bud of 4-day-old chick embryos is suggested as a cultivation site for embryonic epithelial membranes.

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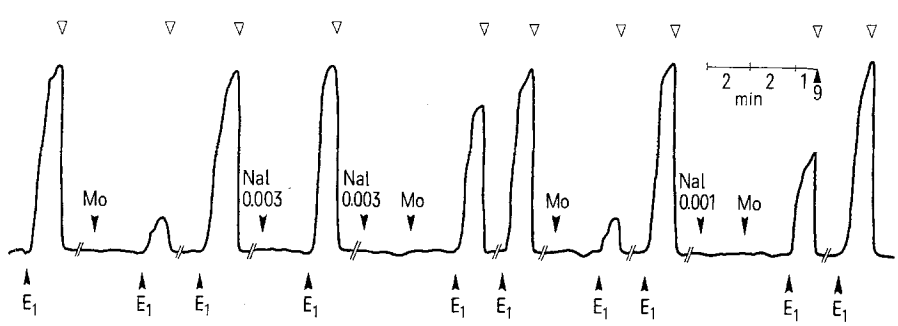
A Simple in vitro Method of Characterizing Narcotic Antagonists

In previous communications it has been reported that non-steroidal compounds with anti-inflammatory or analgesic properties, or both, counteract contractions elicited in the isolated guinea-pig ileum by arachidonic acid peroxide (AAP). This antagonism therefore affords a means of screening new substances for anti-inflammatory and analgesic activity[1,2].

Recently, it was shown that as a rule only narcotic analgesics also antagonize contractions produced in the same organ by prostaglandin E_1 (PGE_1)[3], whereas non-steroidal, anti-inflammatory agents are generally inactive against such contractions. The only non-steroidal, anti-inflammatory compound that antagonizes PGE_1-induced contractions of the guinea-pig ileum at concentrations in which it also counteracts those elicited by AAP is tribenoside[4]; this substance also displays an unusually broad spectrum of antagonistic activity in a variety of smooth-muscle organs, including the intestine, the uterus[5,6] and the veins[7,8], whereas it is typically inactive in arterial preparations[9]. The findings made with narcotic analgesics in regard to PGE_1-induced contractions of the guinea-pig small intestine have now been shown to apply equally to narcotic antagonists. It will be demonstrated that with the aid of this test model narcotic antagonists can easily be divided into 'true' antagonists and partial agonists, depending on whether or not they are capable of reversing the inhibitory effect of morphine, or drugs with a morphine-like action, on contractions induced by PGE_1 or AAP. The experiments in question were performed on the isolated guinea-pig jejunum, suspended in Tryode, and not, as on previous occasions, on the ileum[1].

Maximal to submaximal contractions of the jejunum were elicited by adding AAP and PGE_1 (or PGE_2) to the bath fluid at final concentrations of 0.3 and 0.1 µg/ml, respectively. The substances whose capacity to counteract these contractions was to be tested had been introduced 2 min beforehand. Narcotic antagonists devoid of PGE_1-antagonistic activity that were to be tested for anti-morphine effects were added to the organ bath 2 min before morphine. The results obtained with a selection of non-steroidal, anti-inflammatory agents, and narcotic and non-narcotic analgesics are summarized in the Table. It is evident from these data that all 3 categories of pharmacological agents are – as has already been demonstrated in previous papers[1-3] – capable of counteracting the contractions elicited by AAP in the isolated guinea-pig intestine. On the other hand, only narcotic analgesics and some narcotic antagonists inhibit the contractions induced by PGE_1, but, as a rule, they have no effect on

[1] R. Jaques, Helv. physiol. Acta *17*, 255 (1959).
[2] R. Jaques, Helv. physiol. Acta *23*, 156 (1965).
[3] R. Jaques, Experientia *25*, 1059 (1969).
[4] Generic name of ethyl-3,5,6-tri-*O*-benzyl-D-glucofuranoside (Glyvenol®).
[5] R. Jaques, G. Huber, L. Neipp, A. Rossi, B. Schär and R. Meier, Experientia *23*, 149 (1967).
[6] R. Jaques and B. Schär, Schweiz. med. Wschr. *17*, 553 (1967).
[7] H. Helfer and R. Jaques, Pharmacology *5*, 23 (1971).
[8] R. Jaques, Pharmacology *4*, 193 (1970).
[9] R. Jaques, in *Current Aspects of Chronic Venous Insufficiency;* Int. Symposium, Porto Cervo 1970, p. 120.



Isolated guinea-pig jejunum. Dose-dependent reversal by naloxone (NaI) of the inhibitory effect of morphine (Mo, 0.03 µg/ml) on contractions elicited by PGE_1 (E_1 0.1 µg/ml). Bath fluid changed at points marked ▽.

Antagonistic actions of various agents on contractions induced by arachidonic acid peroxide (AAP), prostaglandin E_1 (PGE_1) and acetylcholine (Ach) in the isolated guinea-pig jejunum

Group	Name	AAP ($n \geq 2$)	PGE_1 ($n \geq 3$)	Ach ($n \geq 2$)
Narcotic analgesics	Morphine[a]	0.02	0.02	∅ 100
	Meperidine	0.03	0.6	3
	Codeine[b]	0.6	1	∅ 100
	Etonitazene[a]	0.0003	0.00002	3
Non-narcotic analgesics	Pentazocine	0.03	1	10
	Dextro-propoxyphene[a]	0.03	0.8	3
	Cyclazocine	0.03	0.03	∅ 10
Narcotic antagonists	Nalorphine[c]	0.01	0.02	∅ 10
	Levallorphane[d]	0.003	2↑[f]	∅ 10
	Naloxone[a]	30	∅ 30	∅ 30
Non-steroidal anti-inflammatory compounds	Aminopyrine	0.1	∅ 100	> 100
	Phenylbutazone[e]	1	∅ 100	> 100
	Noramidopyrine-methanesulphate[e]	7	∅ 100	∅ 100
	Salicylic acid[e]	∅ 100	∅ 100	∅ 100
	Acetylsalicyclic acid[e]	3	∅ 100	∅ 100
	Mefenamic acid[e]	0.03	30	∅ 100
	Flufenamic acid[e]	0.02	30	∅ 100
	Ibufenac[e]	0.3	> 100	∅ 10
	Indomethacin[e]	0.1	60	∅ 10
	Tribenoside	3	3	3

ED_{50} in μg/ml: all concentrations refer to the salt. ∅, denotes inactive at the concentration indicated. [a] Hydrochloride. [b] Phosphate. [c] Hydrobromide. [d] Tartrate. [e] Sodium salt. [f] Slight enhancement at concentration shown.

contractions caused by PGE_2. In contrast, with the exception of tribenoside, non-steroidal anti-inflammatory agents are inactive against PGE_1-induced contractions. Of the narcotic antagonists tested so far, only naloxone and levallorphane have proved inactive as antagonists of PGE_1-elicited contractions of the guinea-pig ileum. These compounds, however, can reverse the effect of morphine on contractions induced by PGE_1 or AAP. When naloxone, for instance, is added to the bath before morphine, it suppresses the inhibitory action of the latter drug in a dose-dependent fashion (see Figure). Naloxone also reverses the PGE_1-inhibitory action of etonitazene[10] or nalorphine.

With regard to the site at which 'true' narcotic antagonists such as naloxone exert their morphine-antagonistic effect in the in vitro system mentioned above, some earlier findings reported by other authors and certain observations made in our laboratories are of importance. PATON[11] has shown that morphine depresses the release of acetylcholine resulting from coaxial stimulation of the guinea-pig intestine, and according to SCHAUMANN[12] the release of acetylcholine following distension of the isolated guinea-pig ileum is inhibited by morphine.

The inhibitory effect of morphine on induced acetylcholine release is thought to take place at the nerve endings rather than in the ganglionic cells[12], since according to FELDBERG and LIN[13] morphine does not inhibit ganglionic transmission in the intestine. In the first paper dealing with the mechanism of the slow-contraction-inducing effect of arachidonic acid in the isolated guinea-pig intestine, its highly specific susceptibility to inhibition by morphine and its potentiation by cholinesterase inhibitors and potassium, it was concluded that arachidonic acid owes its ability to induce contractions in this organ to the fact that it releases acetylcholine[1]. Later on, DAKHIL and VOGT[14] very convincingly showed that arachidonic acid as such is devoid of intrinsic contractile activity in the guinea-pig small intestine and that contractions of the guinea-pig gut such as we observed[1,2] are due to an apparent artefact: arachidonic acid peroxide. The finding would, however, seem to be hardly unexpected, since both arachidonic acid and lipoxidase occur in almost every tissue. Nevertheless, in the in vitro test system mentioned above morphine antagonism has been shown to take place in peripheral structures presumably at the endings.

The fact that in general only contractions induced by PGE_1 and not by those elicited by PGE_2 are inhibited by morphine and other narcotic analgesics may serve as the basis for a pharmacological test for detecting minute quantities of PGE_1 in biological fluids and distinguishing between PGE_1- and PGE_2- like material present in extracts of biological origin.

Zusammenfassung. «Narcotic analgesics» und einzelne «narcotic antagonists» hemmen am isolierten Meerschweinchen-Jejunum durch PGE_1 ausgelöste Kontraktionen. Die PGE_1-antagonistische Wirkung der «narcotic analgesics» kann durch echte «narcotic antagonists» aufgehoben werden.

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[10] Generic name of *p*-ethoxybenzyl-2-dimethylaminoethyl-1-nitro-5-benzimidazole[9].

[11] W. D. M. PATON, Br. J. Pharmac. *12*, 119 (1957).
[12] W. SCHAUMANN, Nature, Lond. *178*, 1121 (1956).
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