Lobeline and capsaicin cause reflex hypotension, bradycardia, and apnea within two seconds of their injection into the right atrium of the cat anesthetized with α-chloralose. Since the pulmonary circulation time in this animal is 3.3 to 4.4 seconds, these drugs must act on sensory endings in the pulmonary vascular bed. Serotonin and phenylbiguanide also stimulate receptors in the pulmonary circulation of the cat to cause the same reflex responses. These receptors are pharmacologically and probably anatomically distinct from those stimulated by lobeline.

The experiments to be presented in this paper were undertaken to compare the anatomical localization and some of the characteristics of the visceral afferent endings stimulated by lobeline, capsaicin, and phenylbiguanide. They show that the endings stimulated by lobeline and capsaicin have a number of features in common; their distribution within the pulmonary vascular bed, the latent period between the injection of the two drugs and their reflex responses, and the sensitivity of these responses to blocking doses of sodium pentobarbital and procaine hydrochloride are the same. These features are not shared by the phenylbiguanide-sensitive endings.

**Methods**

A total of 22 healthy adult cats of either sex were used. They were anesthetized with α-chloralose (65 mg./Kg.) in propylene glycol, given by intraperitoneal injection. The trachea was cannulated. Arterial pressure was measured from the femoral artery via a polyethylene catheter by a Statham transducer and recorded on an Offner Dynograph. One lead of the electrocardiogram was recorded on one channel of a Grass Polygraph and the exact time of drug injection recorded on another channel, using an automatic signaling device.

In the experiments designed to localize the drug-sensitive area of the pulmonary vessels, a radiopaque catheter was passed, with the aid of fluoroscopy, via the right external jugular vein into the right atrium and the pulmonary arterial tree. In each animal, the catheter tip was positioned in the pulmonary artery, and in the left and, where possible, the right branches just distal to the bifurcation and at the level of the hilum. The catheter was passed with the animal in the postero-anterior position, and the position of the tip, particularly when positioned just distal to the bifurcation, was confirmed by rotating the cat into the right and left posterolateral positions. In the same animal, another catheter was passed via the femoral vein into the right atrium. By this means, drugs could be injected into different positions of the pulmonary arterial tree and the magnitude of the response in each position compared with a subsequent right atrial injection. By this technique, the effect of any drift in the magnitude of the response to a constant dose during the course of the experiment was minimized. Drug dilution was adjusted so that the volume to be injected was always less than the intraluminal volume of the catheter. The drug was washed into the circulation with twice this volume of physiological saline. Injections were repeated every 5 to 10 minutes according to the magnitude and duration of the preceding response. At the termination of the experiments, both vagi were sectioned in the midcervical region, and the highest doses of each of the drugs previously used were injected, in turn, into the right atrium. In five animals, the roots of the great vessels and the heart were dissected to eliminate embryological anomaly, and in particular, to confirm the origin of the coronary vessels from the aorta.

When the actions of pharmacological antagonists on the reflex responses to lobeline and capsaicin were studied, intravenous injections were made through a polyethylene catheter inserted via the femoral vein into the inferior vena cava, and intravenous infusions were simultaneously made into the contralateral femoral vein using a Harvard Infusion Pump. In each animal, the median effective dose of capsaicin and lobeline for the hypotensive response was determined and used as a standard. Commencing with submaximal blocking...
doses of sodium pentobarbital, procaine hydrochloride, and tetraethylammonium chloride, the concentration of these agents was slowly increased until the response to lobeline was approximately reduced to half its original size. The proportional depression of the response to capsaicine was then measured. Only when the response to capsaicine was bracketed by two responses to lobeline of equal magnitude were the results considered acceptable. Graded higher doses of blocking agents were then employed to determine whether one response could be completely eliminated before the other.

All drugs were obtained from commercial sources.

Results

LOCALIZATION OF THE LOBELINE- AND CAPSAICINE-SENSITIVE AFFERENT ENDINGS

Preliminary experiments showed that the marked reflex hypotensive response to an intravenous injection of lobeline was completely, or almost completely, absent when this drug was injected into either branch of the pulmonary artery at the level of the hilum of the lung. The response to phenyldiguanide was unchanged when injected at this position. These results suggested that lobeline stimulated afferent endings within the pulmonary arterial tree proximal to the hila. To test this hypothesis, a definitive series of experiments were carried out in which the magnitude of the hypotensive response to capsaicine, lobeline, and phenyldiguanide, injected at different positions in the pulmonary arterial tree, was compared to that following right atrial injection. The approximate average median effective dose of the drugs for the hypotensive response on right atrial injection (lobeline, 15 μg/Kg.; capsaicine, 20 μg/Kg.; phenyldiguanide, 12 μg/Kg.), found in earlier experiments, was used throughout this study. When a radiopaque catheter was passed into the pulmonary artery of the cat, it followed the natural curve of that vessel into the left main branch. In 4 of the 10 animals in this series, the right, as well as the left, branch was catheterized. In these 4 animals, when the size of the responses to each drug was compared on injection into the right and left pulmonary arteries both distal to the bifurcation and at the hilum, no significant difference was found. In table 1, the results from this series of 10 animals are summarized, and since similar responses were obtained from comparable positions on the right and left sides, these are listed together. In figure 1, part of a tracing from one of these experiments is shown.

It can be seen that the hypotensive response to lobeline and capsaicine, although greater on injection into the pulmonary artery than the right atrium, was almost completely absent on injection into the right and left pulmonary hila. The magnitude of the hyper-
tensive response to lobeline was unchanged. In contrast, the hypotensive response to phenyl-
diguanide, which was also greater on injection into the pulmonary artery, was only slightly
reduced on injection at the level of the hilum.
In all these experiments, the hypotensive response to the highest dose of each drug com-
pletely disappeared following bilateral mid-
cervical vagotomy.

A COMPARISON OF SOME OF THE
CHARACTERISTICS OF THE REFLEX
RESPONSES TO CAPSAICINE
AND LOBELINE

Dosage
The median effective dose for the hypoten-
sive response to capsaicine following injection
into the right atrium was found to be 20.4 ±
S.E. 5.0 μg./Kg. (n = 14), that for lobeline
sulphate, 14.3 ± S.E. 1.6 μg./Kg. (n = 10),
and that for phenyl diguanide administered in
the same manner was 12.1 ± 2.8 μg./Kg.
(n = 10).

Latent Period
In nine cats, the latent period between right
atrial injection and reflex bradycardia was
determined for just-maximal doses of cap-
saicine and lobeline. When these latent pe-
riods were grouped in pairs from the same
animal, there was no significant difference
between the two groups (t [n = 9] = 0.014).
The longer latent period for the response to
phenyl diguanide, previously described, was
reconfirmed.

Differential Pharmacological Block
As it has been shown that it is possible to
differentiate among the depressor responses
caused by phenyl diguanide, serotonin, verat-
ridine, and lobeline using pharmacological
antagonists, attempts were made to compare
the responses following capsaicine and lobe-
line by this means.

Sodium Pentobarbital. In five animals, so-
dium pentobarbital in doses ranging between
0.5 and 3 mg./Kg. reduced the standard re-
sponse to capsaicine and lobeline by equal
amounts (t [n = 8] = 1.4). Higher doses of
sodium pentobarbital did not eliminate one
response before the other. The response to
lobeline is known to be more sensitive to the
blocking action of sodium pentobarbital than
that to phenyl diguanide.

Procaine Hydrochloride. After an initial
loading dose of 1 mg./Kg., this drug was ad-
ministered at rates varying from 0.17 to 1.25
mg./Kg./min. in five animals. The propor-
tional reductions in the standard responses
of the two drugs were not significantly dif-
ferent (t [n = 8] = 0.74).

Tetraethylammonium Chloride (TEA). To
each of five animals, an initial loading dose
of TEA (0.3 mg./Kg.) was given, followed
by a slow intravenous infusion. It was found
that this agent could be infused at such a rate
that the response to lobeline was abolished
when that to capsaicine was only slightly re-
duced. The infusion rates varied between
0.013 and 0.17 mg./Kg./min. These doses of
TEA were usually sufficient to cause some
ganglionic blockade as evidenced by a slight
fall in arterial pressure.

Discussion
The results of the catheter studies suggest
that the sensory afferent endings stimulated
by capsaicine and lobeline are situated somewhere in the pulmonary artery and its right and left branches proximal to the level of the hilum. The response from injection into the pulmonary artery was greater than on right atrial administration. It is, therefore, unlikely that the lobeline-sensitive receptors are in the right atrial or ventricular wall. The depressor response was absent on injection at the hilum. In contrast to this, the receptors for phenylbiguanide are situated either at, or distal to, the level of the origin of the pulmonary lobar branches, for the response following injection at the level of the hilum is little smaller than that just distal to the bifurcation. A detailed quantitative comparison of the magnitude of the responses is not possible, since the proportion of the pulmonary vascular bed into which the drug may flow varies according to the site of injection. For instance, drugs injected into the right atrium and pulmonary artery flow into both lungs, but when injected distal to the bifurcation and at the level of the hilum flow only into one lung. Furthermore, the drug concentration at any one position in the pulmonary bed will vary according to the site of injection and the degree of mixing that follows. In these localization studies, only small doses of the exciting agents were employed. It is known\textsuperscript{1, 2, 5} that all these drugs stimulate endings in the coronary vascular bed to cause hypotension, but the doses employed in these experiments were probably too small to stimulate these endings. For example, on no occasion did lobeline, capsaicine, and phenylbiguanide cause a reflex bradycardia that originated more than 1.5 seconds after hilar injection. Since the total volume of each injection varied between 0.35 and 0.45 ml., this response may be partly due to the retrograde flow that may occur when this volume is rapidly injected into the distal part of the pulmonary artery. It is not due to mechanical stimulation of local stretch receptors, for it is not seen following the injection of the same volume of saline.

Since the hypotensive response to the doses used completely disappeared following vagotom y, the observed effects must be reflex in origin and are not due to the actions of this alkaloid on the brain\textsuperscript{6} or ganglia.\textsuperscript{7} It might be argued that since the arterial pressure response to lobeline is biphasic, the hypotensive component may become smaller as the catheter tip is advanced towards the hilum, not because it passes beyond the "hypotensive receptors," but because it approaches the "hypertensive receptors." However, this is unlikely since the magnitude of the hypertensive response is unchanged in any position of the catheter. Furthermore, the hypotensive response disappears following the movement of the catheter tip only 1 to 2 cm. along the artery in which the peak stream velocity is 50 to 80 cm./sec.\textsuperscript{8} The nearest sensory endings which, on stimulation, lead to hypertension are in the aortic body. Since the pulmonary circulation time is three to four seconds,\textsuperscript{3} the movement of the catheter tip from the pulmonary bifurcation to the hilum will not appreciably advance the tip towards these receptors and, therefore, cannot account for the observed phenomena. It is possible that in some cats, a coronary artery may arise from the pulmonary artery and the location of the lobeline receptors may, therefore, be in the heart rather than the pulmonary vessels. This rare anomaly is well recognized in the human subject.\textsuperscript{9} In the five cats dissected, the origin of the coronary vessels from the aorta was confirmed. As the distribution of the lobeline responses was essentially the same in all the cats studied, this abnormality would not account for the results obtained.

Bianconi and Green,\textsuperscript{10} recording from the vagus nerve, found evidence of massive pressoreceptor activity originating from the pulmonary artery and its main branches in the cat. This activity is increased by lobeline in the absence of pulmonary artery pressure changes.\textsuperscript{11} Coleridge et al.\textsuperscript{12} showed that in the dog these afferent endings are found mainly in the region of the bifurcation and in the walls of the main branches of the pulmonary artery, and that pulsatile distention of the right pulmonary artery will cause hypotension and a brief bradycardia.\textsuperscript{13} Since
the distribution of these pulmonary pressoreceptors and the capsaicin- and lobeline-sensitive afferent endings is similar, the reflex bradycardia, caused by these drugs, may be due to their action on hypotension pulmonary pressoreceptors. Although it has not been shown in vitro that lobeline will stimulate pressoreceptors, nicotine, a drug with many actions, like lobeline, will stimulate carotid sinus pressoreceptors. However, Aviado et al. showed that an increased pressure in the pulmonary bifurcation produced bradycardia, hypotension, but also hyperpnea. Recently, Salisbury and his co-workers have described a reflex systemic arterial hypertension in the dog following occlusion of the main pulmonary artery proximal to the bifurcation.

The sensory endings stimulated by lobeline and capsaicin appear to possess a number of common features. They are localized in the same part of the pulmonary vascular bed and the latent period between injection and response is the same for both drugs. The reflex responses to these agents may not be differentiated by the blocking action of procaine or sodium pentobarbital. Since these blocking agents do differentiate between the reflex responses to lobeline and phenylisoguanide, it seems unlikely that these antagonists act on any part of the reflex pathways that are common to these drugs. They, therefore, probably act on the afferent sensory neurones or their central connections. Barbiturate derivatives do not appear to depress carotid chemoreceptor activity by a peripheral action. As the responses to capsaicin and lobeline are antagonized to a similar degree, it is therefore probable that the groups of afferent neurones stimulated by capsaicin and lobeline are the same. The observation that TEA will differentiate between the response to capsaicin and lobeline is not necessarily opposed to this hypothesis. The same sensory ending could be stimulated in two pharmacologically distinct ways. It is highly likely that lobeline, like nicotine, is almost completely ionized at physiological pH and would therefore act by membrane depolarization. TEA as an antidepolarizing agent probably blocks the reflex action of lobeline by such an action on the pulmonary afferent sensory endings. It is very unlikely that capsaicin would act in the same manner as lobeline, and therefore be susceptible to the blocking agent of TEA.

Summary
A comparison has been made of the anatomical localization and some of the characteristics of the visceral afferent endings stimulated by capsaicin, lobeline, and phenylisoguanide. The reflex hypotensive responses to these drugs have been measured following injection into the right atrium and different positions of the pulmonary artery tree in cats anesthetized with a-chloralose. In comparison with the magnitude of the hypotensive responses to capsaicin, lobeline, and phenylisoguanide from the right atrium, the response to capsaicin and lobeline was small or absent on injection into the right or left branch of the pulmonary artery at the level of the hilum. The response to phenylisoguanide was the same on injection at these two positions. The latent period between the injection and the reflex responses to capsaicin and lobeline, and the sensitivity of these responses to blocking doses of sodium pentobarbital and procaine hydrochloride were the same. Tetraethylammonium chloride will block the reflex action of lobeline in doses that have little effect on the reflex action of capsaicin. It is concluded that, in the cat, the capsaicin- and lobeline-sensitive receptors have a similar distribution and are probably situated in the pulmonary artery and the extrapulmonary part of its main branches and that these are anatomically separate from those stimulated by phenylisoguanide.

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