TABLE V. Effect of Cortisone on Weights of Spleen, Liver and Adrenals.

No. of pigs	Treatment	Wt of following organs in g, Mean ± S.E. Spleen	Liver	Adrenals
17	Cortisone, 5 mg/day, 0-18th day; 10 mg/day, 18-30th day	.77±.05	27.7±1	.20±.01
26	0	1.22±.05	24.4±1.1	.27±.01

sions of the treated animals were smaller and less severe than those of the controls. Furthermore, none of the lesions of the treated animals became necrotic, whereas one-third of the control lesions showed necrotic centers.

The organ changes produced by cortisone (Table V) were similar to but less extensive than those which have been previously observed in the rabbit(8). The spleen and adrenals were distinctly smaller than those of the controls, weighing respectively two-thirds and three-fourths as much. The liver was normal in size and appearance and no lipemia was visible.

Summary. 1. Large doses of cortisone failed to protect guinea pigs against active or passive anaphylactic shock. These results are in agreement with those of other investigators(3,4). 2. Treatment with this hormone during period of active sensitization diminished the intensity of the Arthus reaction and the quantity of circulating antibody produced. Neither of these effects was as extensive as those observed in rabbits following the administration of comparatively smaller doses of cortisone(1,2). Similarly, the anatomic changes produced by cortisone were less striking in the guinea pig than in the rabbit (8). These findings suggest that the guinea pig is considerably more resistant to the action of cortisone. The failure of cortisone to abolish the active Arthus reaction and possibly its failure to alter active anaphylactic shock might have been related to the incomplete suppression of circulating antibody under the experimental conditions employed.

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A Phylogenetic Study of the Milk "Let-Down" Hormone. (19566)

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Mammals are divided into 3 divisions. The monotremata or egg-laying mammals are the most primitive derivatives from reptilian stock. Their mammary glands open on the skin in association with hairs but are without

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nipples. The eutherian mammals include all those in which the young are born alive (viviparous). This group includes both marsupial and placental mammals. The former group are without a true placenta and usually possess a pouch in which the offspring are kept after birth. Nipples are always present. The young become attached to a nipple for a considerable period during early pouch life. Only later do they become detached from the nipple during the interval between nursing periods. In placental mammals which are more mature when born, the young generally nurse only at intervals. (During the interval between nursing the young are not attached to the nipples.) In certain placental mammals, it has been shown that the milk secreted in the mammary glands only becomes available to the nursling or the milker upon the nervous stimulation of the nipples. This causes the discharge of the posterior pituitary milk let-down factor which is thought to cause the contraction of the myoepithelial cells surrounding the alveoli and ducts, so releasing the milk.

Extensive assays of the amount of milk "let-down" hormone in the pituitaries of dairy cattle have been made possible by the use of the lactating sow(1). In connection with these studies, it seemed of interest to determine if milk "let-down" hormone was secreted by all types of mammals. Such information would throw some light on the milk "let-down" mechanism of primitive mammals. The question of whether the mechanism for the release of the "let-down" hormone is the same in monotremes without nipples as in the more developed mammals is unanswered. This problem is also unsolved in the case of the marsupials whose young are attached continuously to the nipples for a period. The first point to be cleared up is whether or not these animals have a posterior pituitary milk "let-down" hormone. Further, it seemed of interest to determine the amount of milk "let-down" hormone in a series of placental experimental animals with the view of determining the species and sex variation. The object of this paper is to present the results of a phylogenetic study of mammalia in regard to the presence of the milk "let-down" hormone in the neuro-hypophysis of representative species, and if present to estimate the amounts present in widely differing forms.

Material and methods. As a representative of the monotremata, the pituitary of a single female echidna, the Australian ant-eater, was obtained from the Auckland (N.Z.) Zoo. The animal choked to death on a piece of ground meat. The pituitary was removed immediately the accident was discovered and placed in acetone. The animal was not lactating, nor did the ovary appear active at the time of death. Two representative marsupials were obtained. The Australian phalanger (*Trichosurus vulpecula*) has been introduced into New Zealand, where it is commonly called an opossum. These animals were trapped around Palmerston North, N.Z. The pituitaries of males, non-lactating and lactating females were dissected out and immediately dropped into acetone. This was done as soon as the animal was taken from the trap. Representative samples of each type of pituitary were obtained. A wallaroo in the Auckland Zoo was found dead in its paddock after appearing in perfect health the day before. Three of the 4 mammary glands were inactive but one teat was greatly elongated and the gland was much enlarged. The exact "pouch" age of the young was not known but since the non-used mammary glands were considerably smaller, some time must have elapsed for the involution to have proceeded so extensively. The time elapsing from its death until the pituitary was placed in acetone would undoubtedly decrease the amount of hormone. Of the placental mammals, posterior pituitary lobes were obtained from the following: Normal male and non-lactating female albino rats of the Wistar strain, normal male and non-lactating female rabbits, Romney Marsh ewes in mid-lactation, a large white sow at end of lactation, and dairy calves, heifers and lactating mature cows. The rat and rabbit pituitaries were obtained in Missouri. The other animals were Ruakura stock. For comparison a marlin (sword fish) pituitary was obtained. A period of about 24 hours elapsed between the time of capture and placing the pituitary into acetone. The resulting assay can only be qualitative due to the loss of potency caused by enzyme activity.

TABLE I. The Milk "Let-Down" Hormone in the Posterior Lobes of Representative Mammals.

Animal	Sex	No. tested	Avg wt of dried glands (mg)	Avg total activity (I.U.)	Specific activity (I.U./mg)	Body wt (lb)	Activity/lb body wt
			Posterior lobe				
Rabbit	♀	6	1	.22	.2	6.45	.03
	♂	3	1	.27	.3	8.71	.03
Rat	♀	22	.27	.22	.8	.35	.6
	♂	20	.25	.18	.7	.4	.4
			Whole pituitary				
Phalanger	♀	17	4.7	.21		5†	.04
	♀ L*	14	5.3	1.1		5†	.2
	♂	27	5	1.1		5†	.2
			Posterior lobe				
Walleroo	♀	1	60	2.7			
Echidna	♀	1		2.3			
			Whole pituitary				
Marlin	♂	1	24	.7		260	.003‡
			Posterior lobe				
Cows	L	9	63	31	.5	766	.04
Calves	♀	14	32	51	1.6	184	.3
	♂	1	116	20		82	.2
			Whole pituitary				
Sheep	♀ L	5	257	4.6		120†	.04
			Posterior lobe				
Pig	♀ L	1	16	13	.8	270	.05

* L = Lactating. † Approx. ‡ Gland removed 24 hr after death.

Except where otherwise indicated, within 30 minutes after the animals were killed the entire neurohypophysis was dissected out, dropped into cold acetone and stored at —12°C. Usually the next day in the case of cattle the median eminence, the stalk and the posterior lobe were separated, further defatted and dehydrated in changes of acetone, dried *in vacuo* and weighed. The anterior lobe of the ewes could not be separated from the posterior lobe. The rat and rabbit posterior lobes only were dropped into acetone and dehydrated.

The milk "let-down" hormone was extracted and assayed by methods previously described (Whittlestone)(2). All assays are expressed as the milk "let-down" equivalent of a standard oxytocic solution.

Discussion. A factor or factors which will stimulate the "let-down" of milk in the sow has been shown to be present in the posterior pituitary of all classes of mammals (Table I). The echidna, a representative of the most primitive type of mammal, the monotremes, showed a very definite response (2.3 I.U.) even though some time elapsed after its death before the pituitary was removed. Since in this form teats are not present, the nuzzling of the gland areas must be sufficient to cause discharge of the milk "let-down" hormone.

The pituitaries of two types of marsupials showed responses. The pituitary of a large lactating wallaroo, a type of kangaroo, showed the presence of only slightly more hormone than that of the echidna. It is suspected that this is due to enzymatic losses before the gland was placed into acetone. The phalanger pituitaries are of especial interest since they were collected quickly following death. It is notable that the lactating females contained five times more hormone than the non-lactating females. The high level of hormone in the male pituitary is interesting.

A large representative group of placental mammals has now been studied. In each of them the presence of the factor causing milk "let-down" has been demonstrated. The amount of "let-down" hormone in terms of body weight shows quite large variations between species. The adult rat shows about 10 times the relative activity found in the rabbit, sheep, pig, and mature cow.

The difference in relative activity between the glands of the calf and the mature cow is

very great. The calf shows an amount of milk "let-down" hormone per unit of body weight which is almost 10 times as great as that shown by the older cows. Work in progress may throw light on the factors influencing the amount of hormone in the dairy cow (Whittlestone *et al.*)(1).

All assays of milk "let-down" hormone were carried out on the lactating sow. The duration of the activity was used as a basis for comparison. It is interesting to note that despite their widely different sources all preparations appeared to exert the same action on the sow mammary gland. The changes in the tone of the gland as detected by the milker appeared to be the same regardless of the origin of the hormone. Similarly the rate at which the milk was ejected was not influenced by the source of the milk "let-down" substance. There is no reason to suspect that the active principle or principles from the posterior pituitary gland is different even in such widely separated creatures as the pig and the echidna.

It is worth noting that the pituitary gland of the fetus of one of the cows studied in another project. to be reported later, was examined for milk "let-down" activity and found to be inactive. This fetus, which was 12 weeks old, had not reached a stage of development at which milk "let-down" hormone was present. The dry weight of the whole posterior pituitary gland was 90 mg.

In addition to the mammalian pituitaries, the presence of a milk "let-down" factor was also demonstrated in the pituitary of a 260 lb marlin or sword fish. While this might appear surprising, it should be pointed out that both the oxytocic and vasopressor (antidiuretic) hormones cause the milk "let-down" phenomena (Whittlestone, 1952)(2). It is possible that the "let-down" effect observed might be due to the antidiuretic factor.

These data on the widespread presence of the factor influencing milk "let-down" may be compared with the assays of mammalian pituitaries for the presence of oxytocic and vasopressor factors by Geiling(3), who has shown that both hormones occur in whales, porpoise, armadillo, and even in chickens.

Summary. 1. A factor or factors which will stimulate the "let-down" of milk in the sow has been shown to be present in the posterior pituitary gland of representatives of the 3 divisions of mammals, the monotremes, the marsupials, and the placentals. 2. The pituitary of a marlin or sword fish also caused milk "let-down" in a sow.

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Effect of Acetylcholine on Mammary Gland of the Lactating Sow. (19567)

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The role of the neurohypophysis as the source of the hormone controlling milk let-down has been clearly established by many workers since the first observations of Ott and Scott(1) on the effect of pituitary extracts on the mammary gland. Cross and Harris(2) have recently shown that the electrical stimulation of the pituitary stalk of the rabbit will result in the let-down of milk. There is evidence that both of the posterior pituitary hormones will cause milk let-down(3). Following milk let-down, it has been shown by Cross(4), and Peeters and Coussens(5) that antidiuresis occurs. Despite the clearly hormonal aspects of the control of milk let-down,

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