bar of osmotic pressure were, respectively, 0.576 ± 0.004, 0.570 ± 0.004, 0.568 ± 0.004, and 0.562 ± 0.005. These sensitivity values for the separate couples are averages for determinations at five osmotic pressures. The standard errors measure the accuracy of proportionality between output, in microvolts, and osmotic pressure. The coefficient of variability of the voltage readings for five replicates of each of the KCl concentrations was determined for each of the thermocouples; the over-all average of these 20 coefficients was 0.5 percent. The highest variability occurred at the highest osmotic pressure for which the average coefficient of variability for the four junctions was 1.1 percent.

The calibration of a hygrometer that makes use of evaporative cooling depends on atmospheric pressure. For a given osmotic solution in the sample chamber of the couples here described, the rate of change of sensitivity with change in atmospheric pressure is constant but increases as the osmotic pressure of the sample is increased. The increase in sensitivity for a 10-mbar decrease in barometric pressure is 0.00145, 0.00155, and 0.00160 μv, respectively, per bar of osmotic pressure, for standard osmotic solutions of 5, 10, and 20 bars. Correction for change of atmospheric pressure from the value at calibration will not often be needed, but barometer readings should be taken so that correction can be made if necessary.

The physical condition of water in soil is usually specified in terms of equivalent membrane pressures, largely because measuring techniques employing membranes are available. With thermocouples which can be accurately calibrated in terms of relative pressure near saturation, it seems clear that the thermodynamic functions of free energy and activity will be more generally used for describing soil-water-plant systems.

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Effects of Selenium and Vitamin E on White Muscle Disease

Reports of the pharmacodynamic interrelationship of selenium and vitamin E in liver necrosis of rats (*1*) and "exudative diathesis" of chicks (*2*) prompted the inclusion of this element in an experiment designed to study the factor or factors involved in the cause of white muscle disease, a myopathy in lambs and calves which results when legumes from certain areas are fed to the dams during gestation (*3, 4*). Since reports concerning the role of vitamin E in this disease are somewhat contradictory (*4, 5*), that vitamin was likewise included in this experiment.

Table 1. Scheme and results.

Ration supplement	Lambs	
	Affected	Not affected
Lot 1, control ration		
None	1	17
Lot 2, basal ration		
None	11	4
Lot 3, basal ration		
770 I.U. of α-tocopherol per ewe per week*	11	4
Lot 4, basal ration		
100 I.U. of α-tocopherol per ewe per day†	16	4
Lot 5, basal ration		
0.1 part of Se per million, calculated for the total ration‡	1	15

* Administered parenterally as *d*-α-tocopheryl polyethylene glycol 1000 succinate (*8*).
† Administered orally, as Myvamix (*8*), with the oats.
‡ Administered orally, as Na_2SeO_3, with the oats.

Lots of 12 ewes each were used in this experiment. The ration fed the control lot consisted of Ladino clover and alfalfa hay grown in relatively nonaffected areas, plus 0.25 lb of oats per ewe per day. The basal experimental ration consisted of Ladino clover hay from a severely affected area, plus 0.25 lb of oats per ewe per day. Supplements were given as indicated in Table 1. A preliminary trial indicated that vitamin E as injected maintained satisfactory blood levels in ewes.

The ewes were placed on the experimental regime 50 days after the bucks were placed with them, and the respective rations were continued for approximately 140 days, the termination being governed by the ages of the individual lambs. One of each pair of twins occurring in lots 1 and 2 was sacrificed soon after birth. These, and other lambs that died, in all lots, were necropsied. With a few exceptions, all of the others were necropsied at approximately 6 wk of age.

Analyses of the hays (*6*) fed in this experiment indicate levels of less than 0.1 part of selenium per million, the limit of the analytical method employed. Tocopherol levels in the hays and in the blood of both the dams and the lambs are being determined.

These results appear to indicate that selenium had a definite protective pharmacodynamic effect with respect to white muscle disease under the conditions of the experiment and suggest that a more comprehensive and critical investigation should be made of the role of this element in white muscle disease and other myopathies occurring in animals, including man (*7*).

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6. We acknowledge the assistance of W. W. Ellis, of the University of Wyoming, in determining the selenium content of the hays.
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Anaphylaxis in Passively Sensitized Guinea Pigs after Subcutaneous Eliciting Injection

Lethal anaphylaxis after a subcutaneous eliciting injection of homologous antigen has been reported relatively rarely. Recently it was shown that in actively sensitized guinea pigs an ultimately lethal but protracted anaphylactic shock can be regularly elicited by a subcutaneous injection of relatively large amounts of antigen (*1*). The signs of protracted shock include pruritus, dyspnea, bristling of fur, a fall in body temperature, and prostration. Symptoms first appear a few minutes after the eliciting injection (pruritus), but death may not occur until several hours have elapsed. At necropsy, the most consistent finding is stasis or hemorrhages of the intestine and stomach walls.

In the experiments to be described in this report, guinea pigs were passively sensitized by the intravenous route with graded amounts of guinea pig or rabbit antiovalbumin serum. Table 1 shows the results of subcutaneous injection of 50 mg of ovalbumin 19 to 20 hours after passive sensitization. In view of the lack

of information on the nature of all the types of antibodies capable of causing anaphylactic sensitization (*2*), passive cutaneous anaphylaxis (PCA) was chosen as the method for titration of both rabbit and guinea pig antisera used for passive sensitization. However, precipitin antibody nitrogen determinations were made on the rabbit antiserum for comparison with prior quantitative studies of systemic anaphylaxis (*2, 3*).

The results obtained with both guinea pig and rabbit serum show that when antiserum is injected 19 to 20 hours prior to the subcutaneous eliciting injection of antigen, the anaphylactic shock observed is of the acute type—that is, there is a swift lethal outcome in respiratory difficulty, and emphysema of the lungs is the striking observation at necropsy. Furthermore, over a limited range of sensitizing dosages, death occurs more rapidly with smaller amounts of antiserum used to prepare the guinea pig; thus within this range, the degree of prolongation is proportional to the amount of serum antibody previously injected. At the effective threshold, the guinea pig may die in acute shock or show acute symptoms of anaphylaxis and recover relatively swiftly or survive a severe acute reaction to suffer protracted anaphylactic shock.

When the antibody is injected intravenously just prior to the subcutaneous eliciting injection, the shock is protracted and usually mild. When, after a preparatory injection of antiserum 19 to 20 hours earlier, an additional amount of antiserum is injected just prior to the subcutaneous eliciting injection, prolongation of the anaphylactic shock may occur. This is probably different from the nonspecific protective effect of foreign sera (*4*) which was observed in our controls. (These controls were injected with 1 ml of normal rabbit or guinea pig serum immediately before the eliciting dose. Rabbit serum regularly provided a true protective effect; the sample of guinea pig serum was much less active.) The type of anaphylaxis observed is dependent on the amount of antiserum given 19 to 20 hours before and the amount injected just before the eliciting dose of antigen. With relatively large amounts of antibody injected the day before, additional amounts of antibody injected just prior to the eliciting injection do not affect the outcome, which is death in 25 to 45 minutes. In these animals, both emphysema of the lungs and stasis in the gastrointestinal tract are usually observed.

The protracted anaphylactic shock of guinea pigs is not a separate form of reaction but that which occurs in a sensitized animal shielded from the full extent of explosive early respiratory effects. It is similar to the anaphylactic shock observed in rats (*5*) and in some aspects

Table 1. Passive anaphylaxis in guinea pigs elicited by subcutaneous injection of 50 mg of egg albumin. Equal numbers of male and female Hartley guinea pigs, weighing from 450 to 550 g, were used. Responses in males and females were similar.

No. of guinea pigs	Amt. of antibody injected before eliciting injection*			Findings at necropsy‡			Deaths			Degree of shock in survivors§
	19–20 hr before		Just before (PCA units)†	Acute	Complex	Protracted	No.	Time (min)		
	PCA units†	Antibody N (μg)						Max.	Av.	
				Guinea pig pool I						
3	10,000		0	0	3	0	3/3	20	17	
5	5,000		0	0	2	1	3/5	25	18	M,S
5	2,000		0	2	3	0	5/5	20	14	
4	1,000		0	2	0	0	2/4	8	7	M,M
5	500		0	1	0	0	1/5	5	5	M,M,M,M
3	100		0	0	0	0	0/3			M,M,M
				Guinea pig pool II						
4	10,000		0	0	4	0	4/4	33	31	
5	2,000		0	2	0	0	2/5	6	6	S,S,S
4	500		0	2	0	0	2/4	5	5	M,M
4	10,000		10,000	1	3	0	4/4	47	39	
4	2,000		10,000	1	2	0	3/4	43	34	M
4	500		10,000	0	0	1	1/4	65	65	M,S,S
				Rabbit pool I						
2	200,000	750	0	2	0	0	2/2	25	23	
4	40,000	150	0	4	0	0	4/4	20	14	
4	10,000	37	0	3	0	0	3/4	8	7	M
3	2,000	7.5	0	0	0	0	0/3			M,M,M

* Antiserum was injected intravenously.
† A PCA unit is the amount of serum capable of sensitization for a threshold response (blueing 8 to 10 mm) unitage determined by twofold dilution titration with guinea pigs as test animals. In rabbit pool I, 1 unit = 0.0038 μg of antibody N, 200,000 units = 1 ml of serum. In both guinea pig pools, 10,000 units = 1 ml of serum.
‡ Acute, inflated lungs; Complex, inflated lungs and stasis in gastrointestinal tract; Protracted, stasis in gastrointestinal tract.
§ M, mild to moderate anaphylactic shock; S, severe anaphylactic shock.

resembles tuberculin shock. The guinea pig survives to undergo the whole syndrome—that is, pruritus, respiratory difficulty, bristling of fur, fall in temperature, and damage to gastrointestinal blood vessels. For example, it was found that sensitized guinea pigs which would die in acute anaphylaxis after a subcutaneous challenge will undergo severe or lethal protracted anaphylaxis if they are protected from the respiratory crisis by Pyribenzamine (*6*). It seems evident that the "protective" factor in guinea pigs challenged by the subcutaneous route is an "excess" of serum antibody presumably free in the circulation and rendered capable of such "protection" by the fact of slow absorption of antigen from a subcutaneous injection site. It is of additional significance that Opie found absorption of egg albumin from the local site of injection slower in the immune than in the normal animal (*7*).

Some unification of cellular and humoral theories of the mechanism of anaphylaxis (*8*) is possible on the basis of the data obtained. It is probable that circulating antibody is not a "protective" factor as much as it is a "prolonging factor." Its protection diverts the blow from the guinea pig "shock organ," and does so only when the challenging route is other than intravascular or when there is little antibody in the issues of the guinea pig (passive sensitization and immediate challenge).

The significance of these findings with regard to the problem of anaphylaxis in man caused by subcutaneous or intramuscular injection of antigenic material and the possible role of serotonin in protracted anaphylaxis of guinea pigs is currently under investigation.

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