

was reached at 100 gm. of protein or 1.8 gm. per kg. and higher protein feeding failed to promote further gain. With the caloric intake constant at 3250 calories and protein intake at 100 gm., the percentage of animal protein was varied from zero to 100%. In this instance 75% animal protein seemed to give better nitrogen gain than any other percentages. There was an increase of albumin from 2.3 to 3.0% and total proteins from 4.0 to 5.2% during the course of first 2 months of study, but after that they stayed fairly constant at these levels. The albuminuria, aside from its steady tendency to diminish as the study progressed, showed no significant variations associated with dietary changes.

In conclusion there seems to be an optimum level both for total calories and protein intake in order to secure maximum nitrogen gain. For an adult 1.8 gm. of protein and 60 calories per kg. seem optimal. The requirements are higher with children. The relative proportion of animal and vegetable protein in the diet does not seem important when both calories and protein are fed at optimal levels.

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Effects of Snake Venoms on Plants.

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With the exception of Mitchell¹ and Salisbury,² who made sporadic observations on the subject in nature, investigators have given very little attention to the effects of snake venoms on plants. In connection with phytopharmacological studies regarding the effect of various animal poisons on living seedlings, the writer made experiments with suspensions or solutions of snake venoms by dissolving small quantities of the dry scales in physiological saline. The method employed has been fully described.^{3, 4, 5} Seeds of *Lupinus albus* are soaked for 24 hours in water and then planted in finely divided sphagnum moss. When seedlings have germinated and

¹ Mitchell, Smithsonian Institute, Contributions to Knowledge, 1860, **12**, 145.

² Salisbury, *J. Med.*, 1854, **13**, 837.

³ Macht and Livingston, *J. Gen. Physiol.*, 1922, **4**, 573.

⁴ Macht, *J. Pharmacol. and Exp. Therap.*, 1926, **29**, 461.

⁵ Macht and Pels, *Arch. Dermatol. and Syphilol.*, 1931, **28**, 601.

borne roots from 30 to 40 mm. long, they are carefully selected, "matched," and placed in upright, hard-glass test tubes containing a plant-physiological solution made up of equal parts of Shive's mixture and distilled water. The average growth of roots of 10 seedlings (in the dark at 20° C. for 24 hours) is determined. Dry scales of snake venoms were rubbed up with small quantities of physiological saline and then diluted with the plant-physiological solution mentioned above in various concentrations; and the growth of the *Lupinus albus* seedlings in such solutions was also determined. The relative inhibition of growth of seedlings in the solutions of venom, compared with growth of the controls, is the phytotoxic index and expressed in percentages. The control experiments, as well as the experiments with solutions of venoms, were all made with seedlings from the same crop, grown under exactly the same conditions of temperature, light, weather, etc. Such sets of experiments were made repeatedly so that the conclusions were definite, as will be seen from the figures in Table I. The venoms of the *Crotalus atrox*, *Crotalus exsul*, *Bothrops atrox*, *Ankistrodon piscivorus* and *Naja* (cobra) were studied.*

It was found that all these venoms markedly *inhibited* the growth of the roots, thus indicating that they were toxic for plant protoplasm. Moreover, it was found that the emulsions of venoms rapidly decomposed on standing at room temperature (24° C.). Study of the table reveals the much lower toxicity of such emulsions after standing for 24 and 48 hours, respectively. The greatest toxicity for plants was exerted by the freshly prepared venom emulsions in which the seedlings were allowed to grow at a rather low temperature, 19° C. Another interesting finding was the considerable decrease in toxicity of the venom scales kept over a period of 5 years. These specimens were kept in small glass tubes in a laboratory cupboard at room temperature but were not made moisture-proof by being placed in a desiccator. Experiments made on *Lupinus albus* in 1933 show much less toxicity than those made with the same material in 1928.

Inasmuch as most snake venoms effect hemolytic changes, it was interesting to inquire into the phytotoxic properties of a number of hemolytic drugs obtained from the vegetable kingdom. Experiments were therefore made with solutions of solanin, saponin and

* The cobra venom used in these experiments was obtained from Professor R. N. Chopra, School of Tropical Medicine, Calcutta; the other venoms were acquired through the courtesy of Dr. T. S. Githens, of the Mulford Biological Laboratories.

TABLE I.

	1928 Experiments				1933 Experiments	
	1:50,000 (Fresh emulsion at 19°C.)	1:25,000 (Fresh emulsion at 19°C.)	1:10,000 (Fresh emulsion at 19°C.)	1:25,000 (After standing 24 hours at 24°C.)	1:10,000 (After standing 48 hours at 24°C.)	1:10,000 (Fresh emulsion at 19°C.)
<i>Crotalus atrox</i>	86	64	44	88	71	70
<i>Crotalus exsul</i>	86	60	30	83	60	36
<i>Bothrops atrox</i>	82	64	18	88	63	55
<i>Ankistrodon piscivorus</i>	90	70	12	—	78	56
<i>Naja</i> (cobra)	—	53	32	—	—	—
<i>Solanin</i>	128	117	110	—	—	—
<i>Saponin</i>	127	120	120	—	110	—
<i>Digitonin</i>	120	100	98	—	93	—

digitonin. All the vegetable toxins hemolyzed blood corpuscles to a greater extent than did the snake venoms. When solutions of these vegetable drugs were tested on the growth of *Lupinus* seedlings, however, it was found that they caused practically no inhibition in growth. These findings agree with those from previous studies by the writer, which indicated that poisons obtained from the animal world are relatively non-toxic for plant protoplasm; and, vice versa, that poisons coming from the vegetable kingdom are usually more poisonous for animals than for plants.⁶

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Effect of Ultraviolet Rays and of Methylthionine Chloride and Heparin on Snake Venoms.

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It is well known that snake venoms in solution or suspension rapidly decompose on standing at room temperature, but few investigators have studied the effect of light on such solutions. Faust, in

⁶ Macht, *Science*, 1930, **71**, 302.