sera prepared with residue RC (a residue deprived of its free lipids), the cells sensitized with the phosphatide antigens did not. Besides, as is clearly shown in Table 2, the phosphatide hemagglutination reaction was not at all inhibited by the tuberculin polysaccharides, and vice versa (*3*).

The foregoing two facts clearly indicate that, in the animal body, the formation of antibodies to the bacillary phosphatide takes place independently of the formation of polysaccharide antibodies, at least so far as tuberculin polysaccharide is concerned.

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References and Notes

1. L. Nègre, *Les lipoïdes dans les bacilles tuberculeux et la tuberculose* (Masson, Paris, 1950).
2. R. J. Anderson, *Fortschr. Chem. org. Naturstoffe* 3, 145 (1939).
3. The mechanism of phosphatide hemagglutination and its clinical meaning in tuberculosis are under investigation.

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Pharmacology and Toxicology of Nicotine with Special Reference to Species Variation

During the brief history of modern pharmacology few chemical entities have received such intensive investigation as nicotine. Langley's demonstration (*1*) in 1889 on the ability of nicotine to inhibit transmissions at synaptic junctions initiated a deluge of literally thousands of articles in the scientific literature.

Several researchers have published on the physiological response of animals to nicotine as an index of neural development in the phylogenetic scale. Greenwood (*2*) reported that the toxic effect of nicotine on invertebrate organisms is determined by the degree of development of the nervous system but that vertebrate animals which have enough in common to stand near each other in the phylogenetic order may react differently to nicotine.

Our interest in this alkaloid results from a screening program designed for the selection of a drug to immobilize wild deer for restocking wildlife conservation areas. The subsequent successful capture and translocation of over two hundred wild white-tailed deer (*Odocileus virginianus*), with nicotine administered by the dart-gun technique previously reported (*3*), warrants the reappraisal of reported toxicity of this chemical.

Gause and Smaragdova (*4*) have compared toxicity of synthetic D-nicotine with natural L-nicotine and have postulated that the variance of toxicity between the two isomers is a function of development of spatially specific receptors at the neuroeffector synapse of voluntary muscles, that it is proportionate to the development of the acetylcholine system.

Pharmacologically, nicotine is considered to be an autonomic active drug with curariformic activity. The classical biphasic action, consisting of evanescent stimulation followed by paralysis of all autonomic ganglia, is generally conceded to be a function of dose, with increasing concentrations involving the central nervous system and myoneural junctions.

The ability of nicotine to induce experimental catatonia, similar to the cataleptoid action of bulbocapnine, was demonstrated by Gutierrez-Noriega (*5*). The maintenance of abnormal or bizzare body positions typical of catalepsy was observed in dogs injected with relatively small doses of nicotine tartrate.

Repeated administration of small or sublethal quantities of the alkaloid, or its salts, results in a rapidly diminishing pharmacological response. Ruppert (*6*) reports that isolobinine and nicotine produce mutual tachyphylaxis in rats and that one-tenth of the effective dose for either of these alkaloids will confer protection against convulsive doses; however, tachyphylaxis is less pronounced in cats and rabbits.

Species variation in ability to eliminate nicotine and in susceptibility to toxic effects was reported by Larson *et al.* (*7*). The median lethal dose, by intravenous injection, was established for cats as 2.0 mg/kg; for dogs, as 5.0 mg/kg; for mice, as 7.1 mg/kg; and for rabbits, as 9.4 mg/kg. However, when nicotine was perfused slowly, intravenously, over an 8-hour period, dogs could tolerate 2.0 times the lethal dose, rabbits 3.3 times, mice 4.6 times, and cats, 10.0 times the lethal dose; this indicates that the ability to metabolize or excrete nicotine is independent of species susceptibility.

Aqueous solutions of chemically pure, natural L-nicotine, unbuffered, of *p*H 8.4, or the equivalent amount of nicotine salt, were administered by intramuscular routes, to all experimental animals. Injection of wild or feral animals was accomplished with the projectile type automatic injecting hypodermic syringe, delivered by a 50-caliber pneumatic rifle (*8*). To avoid development of tachyphylaxis, animals were used only once, or were allowed an interval of not less than 48 hours for eliminating all traces of the drug. The end point for determining paralysis of animals was loss of locomotor function.

The minimal paralytic dose of nicotine and the approximate lethal dose for 14 species are listed in Table 1.

After minimal paralytic doses of nicotine an average of 3 minutes is required for development of symptoms of polyuria, locomotor ataxia, lethargy, and catatonia or flaccid paralysis. Large sublethal amounts of nicotine precipitate severe convulsive seizures, followed by flaccid paralysis and recovery within three hours. Death generally occurs during the convulsive seizure from lethal doses of nicotine; however, monkeys and cattle may succumb to latent effects of minimal lethal doses many hours after their recovery from paralytic effects. Necroscopy studies of these animals reveal pronounced urinary retention, distended ureters, and acute hydronephrosis with petechiasis, indicative of acute uremia. In subparalytic doses, nicotine is an effective emetic in those animals capable of emesis and promotes lethargy or somnolence in all species studied.

The paralytic dose of nicotine administered by intramuscular injection and the approximate lethal dose have been established for 14 species of animals (*9*).

The therapeutic index for nicotine is sufficiently great to allow the use of

Table 1. Paralytic and approximate lethal doses of nicotine, administered by intramuscular injection.

Animal	No. of observations	Minimal effective dose (mg/kg)	Approximate lethal dose (mg/kg)	Therapeutic index
Pigeons	20	5.0	9.0	1.8
Mice	100	5.0	8.0	1.6
Rats	50	10.0	15.0	1.5
Rabbits	30	18.0	30.0	1.6
Guinea pigs	15	8.0	15.0	1.8
Chinchillas	18	10.0	18.0	1.8
Dogs	24	4.5	15.0	3.3
Cats	10	3.5	9.0	2.5
Cattle	38	4.5	9.0	2.0
Horses	6	4.0	8.8	2.2
Swine	4	6.6	> 14.0	not determined
Goats	200	3.0	13.0	4.3
Deer	205	3.0	9.0	3.0
Monkeys	8	4.0	6.0	1.5

careful estimates of body weight in calculating paralytic doses to be administered (by means of the projectile hypodermic syringe) for the purpose of safely inactivating wild, feral, or dangerous and unmanageable animals.

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References and Notes

1. J. N. Langley and W. L. Dickinson, *Proc. Roy. Soc. (London)* 46, 423 (1889).
2. M. Greenwood, *J. Physiol. (London)* 11, 573 (1890).
3. J. H. Jenkins *et al., Proc. Southeastern Assoc. Game and Fish Comm.* (19[illegible] H. A. Crockford *et al., Wildlife Management* 21, 213 (1957); F. A. Hayes *et al., J. Am. Vet. Med. Assoc.* 130, 479 (1957).
4. G. F. Gause and N. P. Smaragdova, *Physiol. Zoöl.* 12, 238 (1939).
5. Gutierrez-Noriega, *Rev. Neuro-Psiquiat.* 5, 323 (1942).
6. H. Ruppert, *Arch. Exptl. Pathol. Pharmakol. Naunyn-Schmiedeberg's* 199, 497 (1942).
7. P. S. Larson, J. K. Finnigan, H. B. Hoag, *J. Pharmacol. Exptl. Therap.* 95, 506 (1949).
8. The projectile-type hypodermic syringe and pneumatic rifle used in this study were kindly supplied by Palmer Chemical and Equipment Company, Inc., Atlanta, Ga.
9. This study was supported by the University of Georgia Research Budget, Athens, and the Palmer Chemical and Equipment Company, Inc., Atlanta, Ga. This report was presented, in part, before the Fourth Pan-American Congress of Pharmacy and Biochemistry, Washington, D.C., November 1957.

11 December 1957

Number of Species of Black-Widow Spiders (Theridiidae: Latrodectus)

The last revision of the black-widow genus *Latrodectus* was that of O. P.-Cambridge in 1902 (*1*). Chamberlin and Ivie in 1935 (*2*) made a study of the black-widow spiders north of Mexico, establishing several subspecies names subsequently used by nonarachnologists in this country. Although there is a steady flow of papers on the toxicology and distribution of black-widow spiders, the taxonomists have not kept pace with their colleagues. The common black-widow spider of the United States is now revealed to include two separate species. As a result of this, the specific toxicological properties of the two have been confused consistently in the literature (a further example of the dependence of physiological research upon accurate determination of the experimental animal).

There are two catalogs listing all the species of spiders known. Roewer's *Katalog der Araneae* (*3*) lists 21 species of *Latrodectus*. Bonnet, who covered the arachnological literature to the year 1938 in his *Bibliographia Araneorum* (*4*), also lists 21 species. Since the completion of these works, *Latrodectus rivivensis* Shulov, 1948, has been described, from Palestine.

In my systematic revision of the comb-footed spiders (Theridiidae), in progress for the last 6 years, I have now included the genus *Latrodectus*. The anatomy of all species was studied in some detail, and for the first time the many names created by Dahl (*5*) and Badcock (*6*) could be evaluated.

Although the structure of the genitalia is the usual criterion for separating spider species, some authors (mostly those otherwise unacquainted with spiders) state that genitalia are not useful in differentiating species of *Latrodectus*. My researches indicate that wherever two forms seem to occur in the same locality there are also differences in the genitalia of these two. Coloration and spines, however, are variable characters.

From a study of large series of specimens it can be concluded that there are three species in America: *Latrodectus geometricus* C. L. Koch, 1841, the cosmotropical brown widow; *Latrodectus mactans* (Fabricius, 1775), also limited to the warmer regions and apparently found in all continents; and *Latrodectus curacaviensis* (Müller, 1776) [= *L. bishopi* Kaston, 1938], endemic in America from Argentina to Canada but possibly more common in the temperate zones of North and South America. The last-named species has been confused with *L. mactans,* and much of the *L. "mactans"* literature of the United States may refer to either or both of the species. No conspicuous morphological differences could be found between *L. hasselti* (New Zealand to India), *L. indistinctus* (Africa), *L. tredecimguttatus* (Mediterranean region), and *L. mactans*. It is possible that they all represent one species. *Latrodectus hystrix* Simon, 1890, from Yemen, and *L. pallidus* O. P.-Cambridge, 1872, from Palestine, Asia Minor, and southern Russia, seem to be distinct. All other names in use appear to be synonyms of the names listed above. It is possible, thus, that the 21 nominal species cited in the literature may have to be reduced to five species.

In the males of *L. curacaviensis,* confused with *L. mactans* in this country, the palpal embolus is wider than and about three-quarters as long as the embolus in males of *L. mactans*. The connecting ducts of the female genitalia have three outside coils in dorsal view, while in *L. mactans* there are four or five. The legs of females of the former species are longer, although the coloration and spines are similar. There are differences in habitat: *L. curacaviensis* lives in trees and shrubs, above ground, in Florida; *L. mactans* lives on the ground. In northern states, *L. curacaviensis* lives under logs and stones in woods and fields and probably gets into urban surroundings only rarely, while *L. mactans* is usually found in trash and near dwellings. The northernmost localities where *L. mactans* is found are Maryland, Indiana, Wyoming, Utah, and central California, although this species may be found also in houses in the larger northern cities. The common black-widow spider of New England, most northern states, and, probably, southern Canada is *L. curacaviensis*.

Latrodectus geometricus has only occasionally been found in this country, in cities of Florida. The females are usually gray in color. The palpal embolus is narrower and about one-quarter longer than that of *L. mactans* (*7*).

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References and Notes

1. O. P.-Cambridge, *Proc. London Zool. Soc.* 1, 247 (1902).
2. R. V. Chamberlin and W. Ivie, *Bull. Univ. Utah.* 25, No. 8, 1 (1935).
3. C. F. Roewer, *Katalog der Araneae* 1, 424 (1942).
4. P. Bonnet, *Bibliographia Araneorum* 3, 2364 (1957).
5. F. Dahl, *Sitzber. Ges. naturforsch. Freunde Berlin* 1902, 36 (1902).
6. H. D. Badcock, *J. Linnean Soc. London Zool.* 38, 12 (1932).
7. A more extensive paper, demonstrating, with figures, the differences between species and mapping their distribution, is in press (*Trans. Am. Microscop. Soc.*). This taxonomic study was made possible by the generous cooperation of the individuals and museums who have loaned collections of *Latrodectus* from various parts of the world. The work is supported by a grant from the National Institutes of Health (No. E-1944).

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New Absorption Peak of Tyrosine

While we were investigating the possible adaptation of the Holiday (*1*) method for the determination of small amounts of tyrosine in sea water, we observed a hitherto unreported absorption peak of tyrosine. This peak, at 330 mμ, arose when dilute solutions of tyrosine (1 to 100 mg/lit) in artificial sea water were autoclaved at relatively high pressures (70 to 90 lb/in.^{2}) in the presence of alkali concentrations ranging from 0.12 to 5.0*N* (Fig. 1, curves 1 and 2).

A similar peak, displaced 10 mμ toward the ultraviolet, was found when tyrosine solutions were autoclaved either in artificial sea water or in distilled water alone. Tryamine, 3,5-diiodotyrosine, and *p*-hydroxybenzoic acid behaved similarly (Fig. 1, curves 3, 5, 7), while other amino acids tested, including phenylalanine, proline, hydroxyproline, histidine, and tryptophan showed no such spectral change. Both crystalline albumin and plasma albumin solutions produced the