coefficients (3). The following equations will be valid:

$$D_{QHB} = E_{QHB} \times C_B \times \alpha_{HB}^{-1} \times \alpha_Q^{-1} \quad \text{(Eq. 4)}$$

α_Q is given by Eq. 5, where X^- means ClO_4^-:

$$\alpha_Q = ([Q^+] + [QX]_{org.} + [X^-]_{org.})/[Q^+] = 1 + E_{QX} \times C_X + (k_{diss.} \times E_{QX} \times C_X/C_Q)^{1/2} \quad \text{(Eq. 5)}$$

Eq. 5 is valid under the assumption that $[X^-]_{org.} = [Q^+]_{org.}$. If the constants in Table I, $C_X < 10^{-4.5}$, and $C_X/C_Q = 100$ are used, the calculation will give $\alpha_Q = 10^{0.55}$.

The calculation of α_{HB} at pH = 9.0 is easily made from Eq. 6:

$$\alpha_{HB} = C_B/[HB^-] = 10^{pH-pK'_{HB}} = 10^{1.88} \quad \text{(Eq. 6)}$$

When equal phase volumes are used, a percentage extraction of 99.0 is obtained if $D_{QHB} = 100$. If this value, E_{QHB} from Table I, and the found α_Q and α_{HB} are inserted in Eq. 4, a $C_B = 10^{-4.2}$ is obtained. This is the minimum concentration of BTB necessary for exchanging perchlorate for BTB in the emepronium ion pair. In the method, a slightly higher concentration is used to avoid losses due to small changes in the pH of the buffer.

The method was tested repeatedly on urine samples with added known amounts of emepronium bromide giving an initial concentration of the urine of 0.2 mcg./ml. and higher. Including the compensation for the solubility of dichloromethane in the urine, the absolute recovery of the method was 98 ± 3%, which is in good agreement with the theoretical recovery. For some single objects, somewhat lower precision in the determination was obtained due to incomplete phase separation.

Urinary Excretion—The described determination method was applied in some excretion studies; examples of excretion curves after oral administration of therapeutic doses are presented in Fig. 1. The comparative study of the total urinary excretion 12 hr. after oral and rectal administration is presented in Table II, where the mean values and the standard deviation between the subjects are given. A rather small part of the drug was excreted in the urine, the value for the rectal administration being higher than for the oral. In the rectal experiment, about half of the volunteers complained of side effects typical for anticholinergic drugs, *i.e.*, drynes in the mouth and accommodation disturbances.

In another rectal experiment, when the dose was lowered and no premedication was given, no side effects were noted. The four persons participating were identical with those in Fig. 1, and their excretion curves are given in Fig. 2. Maximal excretion in this case occurred within the first 2 hr. after administration.

Further studies of the absorption, metabolism, and excretion of emepronium bromide are in progress.

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Cactus Alkaloids X: Isolation of Hordenine and *N*-Methyltyramine from *Ariocarpus kotschoubeyanus*

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Abstract □ Hordenine HCl and *N*-methyltyramine HCl were crystallized from extracts of the cactus *Ariocarpus kotschoubeyanus* (Lemaire) Schumann. Other alkaloids, which have been found in closely related *Ariocarpus* species, were not detected in this species. Extraction of the isolated alkaloids by percolation gave higher yields than continuous extraction.

Keyphrases □ *Ariocarpus kotschoubeyanus*—isolation of hordenine, *N*-methyltyramine □ Hordenine—isolation from *Ariocarpus kotschoubeyanus* □ *N*-Methyltyramine—isolation from *Ariocarpus kotschoubeyanus* □ TLC—separation

Ariocarpus kotschoubeyanus (Lemaire) Schumann, like some other species of *Ariocarpus*, is commonly called pezuña de venado, pata de venado, chaute, or peyote in areas of northern and central Mexico where the cactus is indigenous (1–5). The common name of peyote and the purported poisonous nature of the plant (2, 4) aroused interest in this laboratory concerning its alkaloid chemistry. No previous phytochemical studies have been reported regarding *A. kotschoubeyanus;* however, *N*-methyl-3,4-dimethoxy-β-phenethylamine, hordenine, and *N*-methyltyramine have been identified and/or isolated from *A. fissuratus* (Engelmann) Schumann (6–8), *A. trigonus* (Weber) Schumann (9), and *A. retusus* Scheidweiler (10, 11), and *N*-methyl-4-methoxy-β-phenethylamine has been isolated from *A. retusus* (11).

In this investigation, alkaloids were initially extracted from defatted and basified plant material, using chloroform in large continuous extraction apparatuses; an anion-exchange resin was used to separate the purified alkaloids into phenolic and nonphenolic fractions (12). As evidenced by TLC, the phenolic fraction contained hordenine and *N*-methyltyramine, which were resolved by preparative TLC and crystallized as the hydro-

chlorides. Unlike the other *Ariocarpus* species investigated, the nonphenolic fraction was surprisingly devoid of any alkaloids in isolable quantity.

A second portion of plant material was extracted identically, but using cold percolation with chloroform rather than continuous extraction, to be assured that some thermolabile alkaloids had not been destroyed during the continuous extraction. Again, only minute amounts of nonphenolic alkaloids were detected; however, hordenine and *N*-methyltyramine were isolated in higher yields from the percolated extract. These alkaloids are weak sympathomimetics (13, 14) and might be responsible for some stimulatory effects upon ingestion of the plant.

EXPERIMENTAL

Plant Material—A total of 185 plants of *A. kotschoubeyanus* were purchased[1] and several are being maintained as greenhouse specimens[2]. The remaining plants were sliced, dried in a forced-air oven at 48° (78% moisture), and ground through a 3-mm. screen in a Wiley mill.

Extraction of Crude Alkaloids—Using large continuous extraction apparatuses, 1.5 kg. of the dried plant material was defatted, basified, and extracted with chloroform as previously described (12). A second portion of the plant material (1.2 kg.) was defatted in the large continuous extraction apparatuses, basified, and extracted with chloroform, using two large (10 × 50 cm.) percolators and adjusting the combined flow rate to about 1 l./hr. for 40 hr. The residues from the two chloroform extractions were processed identically, with one significant variation from a published procedure (10), to yield the crude alkaloids; the variation involved an additional chloroform extraction after adjusting the extracted aqueous solution at pH 9.5 to pH 10.5. The crude alkaloids were then resolved into phenolic and nonphenolic fractions with an anion-exchange column as previously described (12).

Preparative TLC—Each of the four resulting extracts was assayed by TLC, using previously described methods (11, 12). The nonphenolic extracts both contained scarcely detectable quantities of several unidentifiable alkaloids, while the phenolic fractions both contained appreciable quantities of compounds corresponding in R_f values and color reactions to hordenine and *N*-methyltyramine. These alkaloids were subsequently isolated from both phenolic fractions, using preparative TLC. The methods for the separation and preparation of the hydrochlorides were as previously described (11), with the exceptions that SGPF-254[3] was used as the adsorbant and the acid–base partitioning steps were omitted.

The continuous extraction method yielded 455 mg. (0.030%) of hordenine HCl (m.p. 181.5–183°)[4]; the percolation extract yielded 702 mg. (0.059%) of hordenine HCl which melted at the same temperature. The mixed melting points with authentic hordenine HCl[5] (m.p. 182–183°) were not depressed, and IR spectra[6] of the reference and natural compounds were essentially identical.

N-Methyltyramine HCl (55 mg., yield 0.004%), melting at 149–151°, was obtained from the continuous extraction method; 176 mg. (yield 0.015%) of this compound, melting at 148.5–150°, was obtained from the percolation extract. The two yields were combined and recrystallized twice from absolute ethanol–ethyl ether to afford 148 mg. of material melting at 151–152°. Synthetic *N*-methyltyramine HCl[7] also melted at 151–152°, as did a mixture of the synthetic and isolated material. IR spectra of the *N*-methyltyramine HCl from the two sources were indistinguishable.

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[1] Southwest Cactus Co., Alpine, TX 79830

[2] Identification confirmed by Dr. Edward F. Anderson, Department of Biology, Whitman College, Walla Walla, WA 99362

[3] Brinkmann Instruments, Inc.

[4] Fisher-Johns melting-point apparatus, uncorrected.

[5] Prepared from hordenine sulfate from Mann Research Labs.

[6] KBr pellets, Beckman IR-5A.

[7] Prepared from *N*-methyltyramine HBr supplied by Dr. A. Brossi, Hoffmann-La Roche, Inc.