# Glycerol monoethers in the scent gland secretions of the western diamondback rattlesnake (*Crotalus atrox*; Serpentes, Crotalinae)

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*Summary.* 1-O-Monoalkylglycerols with $C_{12}$ to $C_{20}$ chains were identified in the scent gland secretions of the western diamondback rattlesnake (*Crotalus atrox*). This is the first documentation of these compounds in the skin secretions of a reptile.
*Key words.* Rattlesnake; *Crotalus atrox;* scent gland secretions; alkylglycerol monoethers.

All snakes possess paired scent glands in the base of the tail from which they typically discharge fluids when molested [1]. Macromolecules [1-3] and lipids [4-7] have been reported in scent gland secretions, but there is little information on the structures of the compounds present. An analysis of the secretions of more than 20 snakes by thin-layer chromatography indicated sterols, free fatty acids, and other compound classes [6]. Lipids, however, were not detected in either of two pit vipers (Crotalinae), the copperhead (*Agkistrodon contortrix*) and the timber rattlesnake (*Crotalus horridus*).

An analysis by tandem mass spectrometry of the scent gland secretions of the mamushi (*Agkistrodon halys*; Crotalinae) indicated free fatty acids ranging in carbon chain length from 20 to 22 [7]. We report here on the chemical composition of the scent gland secretions of another crotaline, the western diamondback rattlesnake (*Crotalus atrox*).

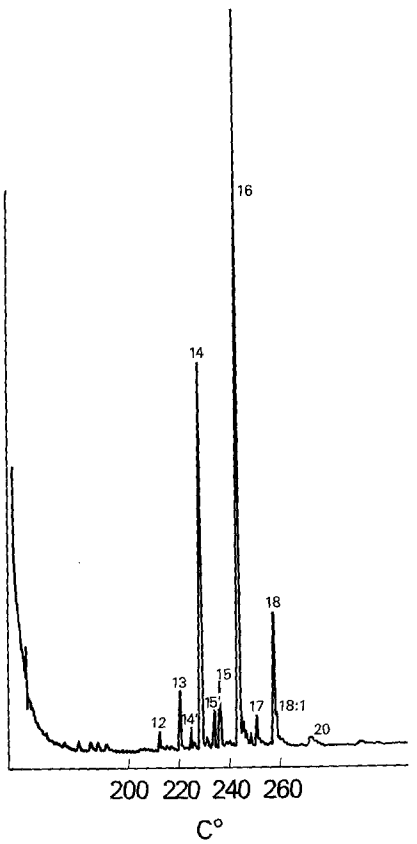
*Material and methods*

Scent gland secretions were obtained from seven male and seven female *C. atrox* from Nolan and Brazos counties, Texas, by manually compressing the base of the tail and collecting the secretions in vials containing methylene chloride. Secretions were pooled accordingly to sex. The vials were packed in dry ice and stored at 0 °C until analysis. The samples were extracted several times with chloroform: methanol (1:1, v/v) and the solvent was removed under reduced pressure. The residues were dissolved in methylene chloride and dried over $MgSO_2$. Secretions from males and females were analyzed separately.

The trimethylsilyl and isopropylidene derivatives of the glandular constituents were prepared by standard methods [8]. Preliminary analyses of underivatized scent gland extracts were performed on an LKB-9000 combined gas chromatograph-mass spectrometer using a 180-cm OV-17 column. Derivatives of the scent gland compounds were examined on the same instrument with a 30-m DB-17 Megabore column. Authentic samples of chimyl alcohol (1-O-hexadecylglycerol) and batyl alcohol (1-O-octadecylglycerol) were obtained commercially (Fluka Chemical Corp., New York)

*Results and discussion*

Three main components of the extracts eluted between 220 and 300 °C. The major compound, which eluted last, was identified as cholesterol. The other two compounds did not exhibit molecular ions, but each contained three fragment ions in the high mass region at m/z 197, 227, 257 and m/z 225, 255, 285, respectively. These ions, 30




Gas chromatography trace of isopropylidene derivatives of glycerol ethers in the scent gland secretions of *Crotalus atrox*. Labels on the peaks denote carbons of the alkyl-chain component of the molecule.

mass units apart, suggested that the compounds were diols (CHOH = 30); consequently, the samples were converted to the trimethysilyl and isopropylidene derivatives.

Both mixtures of derivatives showed similar GC patterns, and the analyses of male and female secretions gave identical results. In the trimethylsilylated sample, with the exception of cholesterol TMS, all components exhibited fragmentation patterns characteristic of bis-TMS ethers of saturated 1-O-monoalkylglycerols, with a base peak at m/z 205 and fragment ions corresponding to M-15, M-90 and M-147[9]. The TMS ether of authentic chimyl alcohol had the same retention time and mass spectrum as the main component of the mixture.

All isopropylidene derivatives exhibited a characteristic base peak at m/z 101, either no parent ion or an extremely weak one, and a fragment ion at M-15[10]. The main constituents of the mixture (fig.), in decreasing order of abundance, corresponded to the $C_{16}$, $C_{14}$, and $C_{18}$ 1-O-n-alkylglycerol ethers. The isopropylidene derivatives of chimyl and batyl alcohols exhibited the same retention times and mass spectra as the $C_{16}$ and $C_{18}$ compounds, respectively. Less abundant components were identified as $C_{12}$, $C_{13}$, $C_{15}$, $C_{17}$, and $C_{20}$ analogs. The mass spectra of minor constituents indicated that they also contained $C_{14}$, $C_{15}$, and $C_{17}$ saturated alkyl chains and probably were the iso or anteiso isomers of the other compounds. A shoulder on the $C_{18}$ peak indicated a glycerol ether with an unsaturated (octadecenyl) chain.

This is the first report of glycerol ethers in reptile skin gland secretions. Similar unesterified alkylglycerol monoethers have only been isolated previously from marine sponges, although more complex esterified glycerol ethers occur widely in marine and terrestrial organism[11]. Antimicrobial and antitumor activities generally have been attributed to these compounds[12]. Their significance in snake scent gland secretions is uncertain.

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