

MAMMALIAN EXOCRINE SECRETIONS XVI.  
CONSTITUENTS OF SECRETION OF SUPPLEMENTARY  
SACCU LI OF DWARF HAMSTER, *Phodopus sungorus sungorus*

B. V. BURGER,<sup>1,\*</sup> D. SMIT,<sup>1</sup> H. S. C. SPIES,<sup>1</sup> C. SCHMIDT,<sup>2</sup> U. SCHMIDT,<sup>2</sup>  
and A. Y. TELITSINA<sup>3</sup>

<sup>1</sup>Laboratory for Ecological Chemistry  
Department of Chemistry, University of Stellenbosch  
Stellenbosch 7600, South Africa

<sup>2</sup>Zoological Institute, University of Bonn  
Poppelsdorfer Schloss, D-53115 Bonn, Germany

<sup>3</sup>Institute of Animal Evolutionary Morphology and Ecology, Russian Academy of Sciences  
Leninsky 33, Moscow 117071, Russia

**Abstract**—As a first step in a study of the role of the secretion of the supplementary sacculi (buccal secretion) of the dwarf hamster, *Phodopus sungorus sungorus*, almost complete chemical characterization of the secretion was achieved. The 35 compounds identified include carbon dioxide, hydrogen sulfide, a large number of carboxylic acids (representing the bulk of the organic volatile fraction of the secretion), phenol, 2-piperidone, indole, two long-chain hydroxyesters, cholesterol, desmosterol, and lanosterol. The position of the double bonds in  $\gamma$ -icosadienyl- $\gamma$ -butyrolactone and  $\gamma$ -hencosadienyl- $\gamma$ -butyrolactone could not be determined, and these two compounds remained only partially characterized. Large variations were found in the relative concentrations in which the short-chain carboxylic acids are present in the secretions of individual animals, and although this aspect was not investigated in sufficient detail in the present investigation, the difference in the carboxylic acid profiles of the secretions of individual animals could play a role in individual recognition in this animal.

**Key Words**—*Phodopus sungorus sungorus*, dwarf hamster, mammalian semiochemicals, mammalian pheromones, exocrine secretion, buccal gland secretion, supplementary sacculi.

\*To whom correspondence should be addressed.

## INTRODUCTION

Certain aspects of the biology of the dwarf hamster, *Phodopus sungorus sungorus*, were summarized in the previous article in the series on mammalian exocrine secretions (Burger et al., 2001). In addition to the ventral gland, both male and female dwarf hamsters produce a buccal secretion from the supplementary sacculi situated at the opening of their cheek pouches. Very little information is available on this subject. Functions attributed to this secretion include communication of information about sex, identity, female breeding condition, and even regulation of certain physiological functions in *P. s. campbelli* (Vasilieva and Feoktistova, 1994). These authors reported the results of a study on the role of the sacculi in the development of juveniles. It was found that surgical removal of the sacculi did not influence development of the pups when they could consume secretion from parents and littermates but that restriction or inability to consume secretion led to a significant increase in the pup mortality, with conditions such as nonspecific enterocolitis and disbacteriosis, and a significant delay in growth and reproductive development. An increase in adrenal gland weight occurred in such animals, and the effects of deprivation were found to be more pronounced in male pups.

The buccal secretion thus appears to play an important role in the survival and development of juvenile dwarf hamsters. Although polar involatile compounds could be responsible for at least some of the functions attributed to the secretion, it was decided to start with the investigation of the volatile organic fraction as a first step towards the complete chemical characterization of the secretion. We now report the identification of practically all its volatile organic constituents.

## METHODS AND MATERIALS

*General and Analytical Methods.* The procedures and instrumentation used in the identification of the constituents of the buccal secretion of the hamster were described by Burger et al. (2001) in the previous paper in this series. In addition to the column coated with 95%-dimethyl-5%-diphenylsiloxane copolymer used for analysis of the ventral secretion of the male dwarf hamster, a glass capillary column (40 m × 0.3 mm) coated with 0.375 μm of OV-1701 was used for qualitative as well as quantitative analyses of the buccal secretion of male and female dwarf hamsters. The GC and GC-MS parameters used for analysis with the PS-089 column were also used for analyses on the OV-1701 column. These analyses were done on both columns using extracts of the buccal secretions of individual animals and the analyses were repeated using solventless sample introduction techniques. Small quantities of the secretion (ca. 0.2 mg) were, for example, introduced into the injector of the GC and the GC-MS instrument using a sample introduction

probe (Burger et al., 1990), and similar sample sizes were also introduced as a smear on the inside of a clean injector liner. When solventless sample introduction was used, the volatile material was cryotrapped on the column with dry ice. For quantitative analyses, the injector was operated in the splitless mode and the data acquisition and processing software specified in the previous paper in this series (Burger et al., 2001) were used.

*Sample Collection and Preparation.* The sticky, whitish secretion was collected separately from the supplementary sacculi of males and females in their summer state, i.e., with the day length exceeding 10 hr. Samples were collected on a weekly basis. Grasping a hamster firmly by the scruff of the neck caused the animal to open its mouth, and the waxy secretion was collected by scooping it from the glandular area with the device used for the collection of the animal's ventral secretion (Burger et al., 2001). The material was transferred from the wire loops to a Reacti-Vial containing dichloromethane using another piece of stainless-steel wire, after which the contents of the vial were thoroughly mixed using a thin glass rod. This caused the secretion to be spread out on the wall of the vial. The material in the vial was sonicated for 10 min, centrifuged for 15 min at 2500 rpm, and the dichloromethane extract containing the organic material transferred to a clean Reacti-Vial with a 100- $\mu$ l syringe. The extract was concentrated in an inert atmosphere (Burger et al., 1999a) for GC and GC-MS analysis. Secretion intended for quantitative analyses was collected on a wire loop as described above. A small sample (ca. 0.2 mg) of the material was accurately weighed into a sample introduction probe (Burger et al., 1990) or a clean glass liner and immediately analyzed to avoid losing the more volatile constituents of the secretion. Hexadecanoic acid was used as external standard in the quantitative analyses.

*Reference Compounds.* Some of the compounds identified in the secretion are commercially available, while others were available from previous research projects. The following compounds were synthesized during the present study.

*3-Phenylpropanoic acid (19).* Cinnamic acid (3.06 g, 20 mmol), dissolved in degassed methanol (25 ml), was hydrogenated at room temperature and atmospheric pressure in the presence of Pd on activated carbon (5%, 300 mg) until 1.02 equivalents of hydrogen had been taken up. The catalyst was filtered off and the methanol removed on a rotary evaporator. The product containing some residual catalyst was taken up in dichloromethane, the solution filtered through a layer of silica gel, and the solvent evaporated to give 3-phenylpropanoic acid (2.66g, 85.8%).  $^{13}\text{C}$  NMR ( $\text{CDCl}_3$ ):  $\delta = 179.36$  (s, C-1), 140.14 (s, C-1'), 128.56 (d, C-3 and 5), 128.26 (d, C-2 and 6), 126.38 (d, C-4), 35.63 (t, C-2), 30.57 (t, C-3).

*2-Hydroxyoctadec-1-yl acetate (31).* A mixture of the hydroxyester (**31**) and 1-hydroxyoctadec-1-yl acetate was synthesized by the  $\text{Al}_2\text{O}_3$ -catalyzed reaction of 1,2-epoxyoctadecane with acetic acid as described by Burger et al. (1999a). The title compound (**27**) and 1-hydroxyoctadec-1-yl acetate were formed in a ratio of 1 : 3 (GC-MS). HR-MS (mixture of the two isomers):  $m/z$   $\text{M}^+$  255.262, calcd. for

$C_{20}H_{40}O_3$  255.269. *Major component*:  $^{13}C$  NMR ( $CDCl_3$ ):  $\delta = 171.48$  (s, C-1), 75.74 (d, C-2'), 64.82 (t, C-1'), 31.95 (t, C-16'), 30.51 (t, C-3'), 29.30–30.5 (t, C-5'–C-15'), 25.33 (t, C-4'), 22.71 (t, C-17'), 21.20 (q, C-2), 14.12 (q, C-18'). *Minor component (31)*:  $^{13}C$  NMR ( $CDCl_3$ ):  $\delta = 171.20$  (s, C-1), 69.99 (d, C-2'), 68.81 (t, C-1'), 33.37 (t, C-3'), 25.38 (t, C-4') 29.30 (t, C-5'–C-15'), 31.95 (t, C-16'), 22.71 (t, C-17'), 20.89 (q, C-2), 14.12 (q, C-18').

*2-Hydroxyicos-1-yl acetate (32)*. A mixture of the hydroxyester (**32**) and 1-hydroxyicos-2-yl acetate was prepared from 1,2-epoxyicosane as described above. The two isomers were present in the product in a ratio of 1 : 2 (GC-MS). HR-MS (mixture of the two isomers):  $m/z$   $M^+$  356.332, calcd. for  $C_{22}H_{44}O_3$  356.329. *Major component*:  $^{13}C$  NMR ( $CDCl_3$ ):  $\delta = 171.55$  (s, C-1), 75.75 (d, C-2'), 64.81 (t, C-1'), 31.95 (t, C-18'), 30.52 (t, C-3'), 29.3–30.5 (t, C-5'–C-17'), 25.33 (t, C-4'), 22.71 (t, C-19'), 21.22 (q, C-2), 14.13 (t, C-20'). *Minor component (32)*:  $^{13}C$  NMR ( $CDCl_3$ ):  $\delta = 171.27$  (s, C-1), 70.00 (d, C-2'), 68.80 (t, C-1'), 33.38 (t, C-3'), 31.95 (t, C-18'), 29.3–31.0 (t, C-5'–C-16'), 29.18 (t, C-17'), 25.39 (t, C-4'), 22.71 (t, C-19'), 20.90 (q, C-2), 14.13 (q, C-20').

## RESULTS AND DISCUSSION

A typical total ion chromatogram of an extract of the buccal secretion of a male dwarf hamster is given in Figure 1. Tentative identification of the constituents of the secretion was based on low-resolution mass spectral data, supported by information obtained by their chemical ionization mass spectra, generated with methane as reactant gas. Final confirmation of most of the proposed structures was obtained by coinjection of an extract of the secretion with authentic synthetic material.

Solventless sample introduction made it possible to identify even highly volatile constituents of the secretion such as carbon dioxide (which was to be expected in the material and is probably extracted from the animal's breath), hydrogen sulfide, and trimethylamine. The bulk of the volatile organic fraction of the secretion consists of short-chain carboxylic acids. These compounds together with the hydrogen sulfide and trimethyl amine are responsible for the unpleasant odor of the animal's breath.

A few diunsaturated compounds could not be fully characterized because dimethyl disulfide (DMDS) derivatization did not yield informative mass spectral data. This was probably due to the high molecular masses of the derivatives, which are expected to elute as flat peaks in the upper isothermal part of the chromatogram of the material subjected to DMDS derivatization. The interpretation of the mass spectra of long-chain hydroxyesters has been discussed by Burger et al. (1996). In the present study the identification of the hydroxyesters **31** and **32** was based

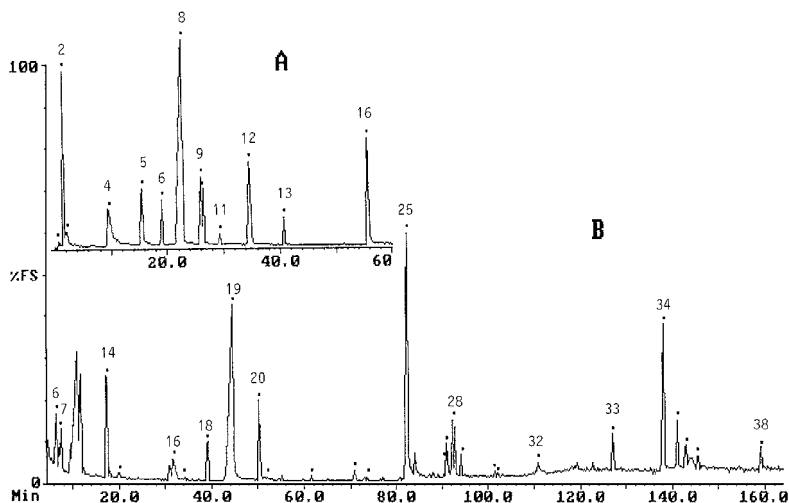


FIG. 1. Total ion chromatograms of the secretion of the supplementary sacculi (buccal secretion) of the dwarf hamster, *Phodopus sungorus sungorus*: (A) Analysis of unprocessed secretion on a relatively polar column (OV-1701) using solventless sample introduction; (B) Extract of the secretion analyzed on an apolar column (PS-089).

mainly on their coelution with the synthetic hydroxyesters because they are present in the secretion in such low concentrations that some of the less abundant ions were not visible in the high-molecular-weight range of their mass spectra. The two unsaturated  $\gamma$ -lactones, **33** and **36**, which are also present in the ventral secretion of the dwarf hamster (Burger et al., 2001), remained incompletely characterized in the present investigation. The compounds identified in the buccal secretion are listed in Table 1 together with information on the analytical techniques employed in their identification, and some quantitative data on the constituents present in male and female secretions. Several compounds identified in the animal's buccal secretion are also present in its ventral secretion. In addition to the structures of representative examples of these compounds given in the paper on the ventral secretion (Burger et al., 2001), the structures of some of the compounds that are not common to both secretions are given in Figure 2.

Typically about 1–4 mg of secretion could be collected from one animal, but some of the animals from our colony consistently produced almost no secretion. The quantitative determination of the volatile organic constituents of the secretion posed a problem because polar compounds such as the short-chain carboxylic acids were not quantitatively extracted with small quantities of dichloromethane from the mucoid material in which they are produced. The highly volatile constituents are lost during concentration of extracts when larger quantities of solvent are used.

TABLE 1. COMPOUNDS IDENTIFIED IN SUPPLEMENTARY SACCVLI SECRETION (BUCCAL SECRETION) OF DWARF HAMSTER

Peak (Figure 1)	Compounds	EI Mass spectral data <i>m/z</i> (%)	Quantity (ng/animal) <sup>a</sup>	
			Male	Female
1	Carbon dioxide <sup>b,c</sup>	44(100), 28(7)	7	43
2	Hydrogen sulfide <sup>b,c</sup>	36(5), 35(3), 34(100), 33(38)	6	15
3	Trimethylamine <sup>b,c</sup>	59(46), 58(100), 57(8), 56(4), 43(5), 42(26), 41(5)	17	27
4	Ethanoic acid <sup>b-d</sup>	61(3), 60(100), 45(85), 43(92), 42(16)	569	682
5	Propanoic acid <sup>b-d</sup>	74(76), 73(5), 57(33), 55(19), 45(62)	30	376
6	2-Methylpropanoic acid <sup>b-d</sup>	88(7), 73(30), 55(8), 45(17), 43(100), 41(50)	65	163
7	Hexanal <sup>b-d</sup>	100(3), 99(4), 82(12), 72(19), 71(8), 67(11), 57(54), 56(78), 44(100), 43(60), 41(75)		9
8	Butanoic acid <sup>b-d</sup>	88(2), 73(30), 60(100), 55(9), 45(21), 43(21), 42(28), 41(25)	840	119
9	3-Methylbutanoic acid <sup>b-d</sup>	87(20), 74(4), 60(100), 45(30), 43(52), 41(40)	205	433
10	2-Methylbutanoic acid <sup>b,d</sup>	87(22), 74(100), 57(60), 45(19), 41(54)	140	143
11	Pentanoic acid <sup>b-d</sup>	87(3), 74(10), 73(36), 60(100), 57(12), 55(14), 45(19), 43(25), 41(28)	35	34
12	4-Methylpentanoic acid <sup>b-d</sup>	101(3), 87(6), 83(12), 74(48), 73(50), 60(35), 57(100), 55(57), 45(16), 43(40), 41(42), 39(20)	315	4
13	Phenol <sup>b-d</sup>	94(100), 66(44), 65(36), 55(15), 51(8), 50(10)	4	4
14	Hexanoic acid <sup>b-d</sup>	87(11), 73(40), 60(100), 56(11), 55(19), 45(20), 43(27), 41(36)	1	
15	1-Acetylimidazole <sup>b-d</sup>	110(6), 82(1), 68(100), 43(39), 41(9)		
16	2-Piperidone <sup>b-d</sup>	99(100), 70(30), 58(9), 56(20), 55(50), 43(78), 42(95), 41(69), 30(69)	367	290
17	1-Octyl acetate <sup>b-d</sup>	116(5), 112(7), 107(2), 101(5), 84(15), 83(17), 70(28), 69(20), 61(22), 57(12), 56(31), 55(30), 43(100), 41(27)		
18	Indole <sup>b-e</sup>	117(100), 90(49), 89(37), 63(22), 59(16), 51(7), 50(7)	18	3
19	3-Phenylpropanoic acid <sup>b-e</sup>	150(33), 105(18), 104(52), 91(100)	270	194
20	<i>m</i> -Hydroxyacetophenone <sup>b-d</sup>	136(38), 121(100), 93(38), 65(40), 53(10), 51(8), 43(18)		
21	Oxindole (1,3-Dihydro-2 <i>H</i> -indol-2-one) <sup>b-d</sup>	133(100), 105(34), 104(90), 78(48), 77(22), 52(30), 51(33), 50(19)		

22	Hexadecane <sup>b-d</sup>	226(5), 155(3), 141(4), 127(6), 113(7), 99(9), 85(30), 71(52), 57(100), 43(81), 41(37)	1	3
23	Tetradecanoic acid <sup>b-d</sup>	228(7), 199(4), 185(14), 171(8), 157(6), 143(9), 129(28), 115(9), 101(8), 87(17), 73(87), 69(33), 60(97), 57(70), 55(80), 43(100), 41(78)	9	10
24	Octadecane <sup>b-d</sup>	155(1), 141(5), 127(6), 113(8), 99(9), 85(32), 71(50), 57(100), 43(80), 41(35)		3
25	Hexadecanoic acid <sup>b-e</sup>	256(7), 227(2), 213(8), 189(3), 185(7), 171(8), 157(9), 143(4), 129(27), 115(13), 101(8), 87(16), 73(96), 69(42), 60(99), 57(87), 55(89), 43(100), 41(75)	133	217
26	(Z)-9-Octadecenoic acid (Oleic acid) <sup>b-e</sup>	264(7), 185(3), 152(1), 151(1), 138(2), 137(2), 129(4), 125(5), 124(5), 123(6), 111(11), 97(30), 84(22), 83(36), 73(22), 69(55), 60(23), 57(41), 55(100), 43(68), 41(73)	32	30
27	(E)-9-Octadecenoic acid <sup>b-e</sup>	264(7), 185(3), 151(1), 138(2), 137(2), 125(3), 123(3), 111(10), 97(28), 84(20), 83(32), 73(20), 69(54), 60(25), 57(28), 55(100), 43(57), 41(65)	18	16
28	Octadecanoic acid <sup>b-e</sup>	284(4), 241(5), 199(3), 185(7), 171(4), 143(4), 129(20), 115(8), 101(7), 97(20), 87(12), 83(23), 73(68), 69(38), 60(73), 57(70), 55(80), 43(100), 41(70)	87	118
29	9,12-Octadienyl acetate <sup>b,c,f,g</sup>	308(0.1), 265(0.2), 248(0.3), 220(0.2), 149(4), 135(8), 121(15), 109(14), 107(10), 95(35), 81(54), 79(47), 67(88), 55(69), 43(100), 41(56)		
30	Icosanoic acid <sup>b-d</sup>	312(12), 284(2), 283(2), 269(10), 255(5), 227(3), 199(2), 185(3), 171(3), 157(2), 143(3), 129(11), 115(7), 101(5), 97(15), 87(8), 85(13), 83(18), 73(39), 69(26), 60(40), 57(53), 55(55), 43(100), 41(50)	1	4
31	2-Hydroxyoctadec-1-yl acetate <sup>b,d</sup>	225(3), 155(2), 125(2), 111(6), 103(10), 97(12), 83(15), 74(29), 71(12), 69(16), 57(27), 55(29), 43(100), 41(27)		
32	2-Hydroxyicos-1-yl acetate <sup>b,d</sup>	283(1), 111(6), 103(10), 97(13), 83(16), 74(28), 71(15), 69(17), 57(28), 55(25), 43(100), 41(23)	1	

(Continued)

TABLE 1. (Continued)

Peak (Figure 1)	Compounds	EI Mass spectral data <i>m/z</i> (%)	Quantity (ng/animal) <sup>a</sup>	
			Male	Female
<b>33</b>	Tetracosadien-4-olide ( $\gamma$ -Icosadienyl- $\gamma$ -butyrolactone) <sup>b,f</sup>	362(7), 251(6), 197(6), 155(8), 141(9), 125(7), 111(18), 97(38), 85(100), 83(43), 71(28), 69(63), 57(77), 55(88), 43(95), 41(73)	94	161
<b>34</b>	Cholesterol <sup>b-d</sup>	386(6), 368(5), 353(4), 326(1), 301(7), 275(10), 255(6), 231(4), 213(10), 199(5), 173(7), 159(16), 145(28), 133(17), 119(20), 107(17), 105(36), 91(40), 81(48), 79(37), 67(33), 57(54), 55(62), 43(100), 41(54)	143	93
<b>35</b>	Unidentified steroid <sup>b</sup>	384(2), 369(3), 351(4), 325(1), 300(4), 271(16), 253(7), 213(7), 173(6), 159(12), 145(19), 133(13), 119(15), 109(18), 107(23), 105(28), 94(28), 93(23), 91(32), 81(37), 69(100), 67(36), 55(70), 43(30), 41(75)	18	41
<b>36</b>	Pentacosadien-4-olide ( $\gamma$ -Henicosadienyl- $\gamma$ -butyrolactone) <sup>b,f</sup>	376(2), 125(8), 111(19), 97(40), 85(95), 83(45), 71(35), 69(65), 57(80), 55(86), 43(100), 41(65)	97	299
<b>37</b>	Desmosterol <sup>b-d</sup>	385(3), 384(2), 370(7), 369(6), 352(4), 351(6), 329(3), 300(7), 271(33), 253(10), 231(8), 213(9), 191(4), 159(10), 145(13), 133(15), 119(23), 107(26), 105(32), 91(40), 81(41), 69(100), 67(42), 55(80), 43(48), 41(98)	19	53
<b>38</b>	Lanosterol <sup>b-d</sup>	426(2), 411(6), 393(4), 259(2), 229(3), 189(5), 187(6), 175(6), 173(5), 161(7), 159(8), 147(8), 145(8), 135(9), 133(9), 123(9), 121(13), 119(18), 109(27), 107(14), 105(16), 95(26), 93(16), 91(16), 83(13), 81(25), 79(15), 69(100), 55(6), 43(35), 41(60)	93	47

<sup>a</sup>Quantities lower than 1 ng/animal are not given.

<sup>b</sup>Low-resolution mass spectrum.

<sup>c</sup>Published data.

<sup>d</sup>Retention time comparison with authentic material.

<sup>e</sup>Cl(CH<sub>3</sub>) mass spectral data.

<sup>f</sup>Position and configuration of double bond(s) uncertain.

<sup>g</sup>Tentative identification.



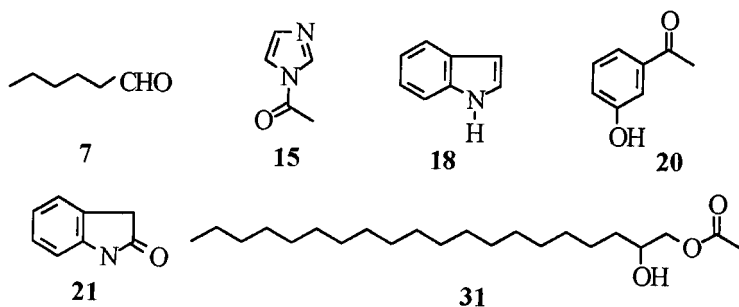


FIG. 2. Structures of some representative examples of the compounds identified in the buccal secretion of the dwarf hamster, *P. s. sungorus*.

Solventless sample introduction, on the other hand, discriminates to a certain extent against the heavy constituents of the secretion. Furthermore, the FID in GC analyses and the mass spectrometer in GC-MS analyses both have different quantitative responses to small and large molecules, while CO<sub>2</sub> and H<sub>2</sub>S are not detected by FID. Ideally the volatile organic constituents present in the headspace gas of the secretion should be determined at temperatures corresponding to the hamster's body temperature in their summer and winter states. Furthermore, the extreme polarity differences between highly polar compounds such as ethanoic acid and the less polar long-chain constituents of the secretion, make it difficult to suggest an universal stationary phase for sample enrichment. Headspace analysis of the buccal secretion was not done in the present investigation. The quantitative data in Table 1 were obtained by combining the results of analyses using different capillary columns and sample introduction techniques in GC as well as GC-MS analyses. Although the quantitative results probably represent a relatively accurate picture of the quantitative composition of the secretion analyzed, the data are considered to be somewhat preliminary at this stage and are given merely as a guideline for biologists planning behavioral studies on the dwarf hamster. Nevertheless, the results do give quite a reliable picture of the ratio in which compounds within limited volatility ranges are present in the secretion. Some of the largest quantitative differences among the buccal secretions from individual animals were found among the short-chain carboxylic acids, and the role of these compounds in individual and sexual recognition in *P. sungorus* should be investigated in more detail.

The highly volatile compounds, carbon dioxide (1), hydrogen sulfide (2), and trimethylamine (3) were detected in the secretion because solventless sample introduction techniques were employed in GC and GC-MS analyses in addition to conventional analysis of extracts. These compounds are almost completely lost from an extract if the solvent has to be evaporated to concentrate an extract for analysis. The presence of carbon dioxide in the secretion is to be expected because it is

extracted from expired breath of the animal by the moisture-containing secretion. Hydrogen sulfide does not feature in reports on mammalian exocrine secretions, probably because it is not detected by FID. It will also not be registered in the total ion chromatogram if the mass spectra are scanned from a mass higher than  $m/z$  34. Hydrogen sulfide, trimethylamine, and the short-chain fatty acids, of which almost all the unbranched, iso- and anteiso-branched members from  $C_2$  to  $C_6$  are present in the buccal secretion, are probably responsible for the obnoxious smell of the dwarf hamster's breath and the secretion. A mixture of similar volatile fatty acids has been identified in many mammalian secretions, such as, for example, the interdigital secretion of the reindeer (Brundin et al., 1978), the perineal scent gland of guinea pigs (Wellington et al., 1979), and human vaginal secretion (Huggins and Preti, 1976). The microbial production of trimethylamine, indole (**18**), other amines, organosulfur compounds, and fatty acids from amino acids in mammalian exocrine secretions has been reviewed and discussed by Albone (1984). The occurrence of long-chain saturated and unsaturated fatty acids, 2-piperidone (**16**), and 3-phenylpropanoic acid (**19**) in mammalian exocrine secretions was discussed briefly in the previous paper in this series (Burger et al., 2001).

Long-chain acetates are not common in mammalian secretions. Several have, however, been identified in the male abdominal scent gland of the jird, *Meriones tristrami* (Kagan et al., 1983), the preputial secretion of the mouse, *Mus musculus* (Spener et al., 1969), and the preorbital secretion of the male oribi, *Ourebia orebi* (Mo et al., 1995).

Phenol (**13**) and its derivatives, such as the three isomeric cresols, other alkyl-substituted phenols, and phenolic acids, are present in many mammalian secretions. Phenol and *p*-cresol were found in human vaginal secretions (Huggins and Preti, 1976); *p*-ethylphenol, *p*-propylphenol, and pyrocatechol in castoreum of the beaver, *Castor fiber*, (Lederer, 1946, 1949); phenol, *m*- and *p*-cresol, and *m*- and *p*-propylphenol in aged urine of some African Bovidae (Madubunyi et al., 1996); and phenol, *m*-cresol, *m*-ethylphenol, and *m*-propylphenol in the interdigital secretions of the bontebok, *Damaliscus dorcas dorcas*, and the blesbok, *D. d. phillipsi* (Burger et al., 1999a). The preorbital secretions of the antelope we have so far investigated do not contain phenols and phenolic compounds. As far as mammalian exocrine secretions are concerned, phenol and phenolic compounds are apparently found in glandular structures in which the conditions are favorable for survival of microorganisms and the accumulation of the secretions of the gland and/or the products of microbial activity.

Oxindole (**21**) as its steroid conjugate has been identified in the urine of children (Voeltler et al., 1971). Acetophenone has been identified in the urine of the red fox, *Vulpes vulpes* (Jorgenson et al., 1978), and *p*-hydroxyacetophenone in castoreum of the beaver (Lederer, 1946, 1949).

Hexanal (**7**) is a potent odor with a threshold value to the human nose slightly lower than that of limonene (Ohloff, 1978), which has been identified in human

breast milk (Stafford et al., 1976). It has also been found in interdigital and pre-orbital secretions of the bontebok, *Damaliscus dorcas dorcas* and blesbok, *D. d. phillipsi* (Burger et al., 1999a,b), and in the preorbital secretions of the steenbok, *Raphicerus campestris* (Burger et al., 1999c). In the interdigital secretions of the bontebok and blesbok, this aldehyde is accompanied by another nine saturated and unsaturated C<sub>7</sub>–C<sub>10</sub> aldehydes. The origin of aldehydes in mammalian secretions has not been established, but in addition to the possibility that they are produced by the exocrine glands of the mammals or by microbial action, short- to medium-chain aldehydes are also produced by the autoxidation of unsaturated lipids such as, for example, unsaturated triglycerides.

Unbranched short- to intermediate-chain alkanes have been identified in human effluvia (Ellin et al., 1974). The alkanes found in some mammalian exocrine secretions are mostly unbranched with intermediate to long chain lengths consisting of even as well as odd numbers of carbon atoms (Burger et al., 1990, 1999a–c). It has to be accepted that these compounds are produced by the glands or by microflora present in the glandular structures because there does not seem to be evidence that their presence can be ascribed to contamination of the secretions with, for example, petroleum products. The two  $\gamma$ -lactones (**33** and **36**) that are also present in the ventral secretion of the male dwarf hamster remained unidentified in this investigation. 2-Hydroxyicos-1-yl acetate (**32**) was also found in the male ventral secretion, but instead of 2-hydroxyoctadec-1-yl acetate (**31**), present in the buccal secretion, the ventral secretion contains the 1-hydroxyhexadec-2-yl and 2-hydroxyhexadec-1-yl pentanoates and their hydroxyheptadecyl analogues. The presence of these  $\gamma$ -lactones, hydroxyesters, and the steroidal compounds cholesterol (**34**), desmosterol (**37**), and lanosterol (**38**) in the dwarf hamster was briefly referred to in the article on the ventral secretion of the animal (Burger et al., 2001).

#### REFERENCES

- ALBONE, E. S. 1984. Mammalian Semiochemistry, The Investigation of Chemical Signals Between Mammals. Wiley-Interscience, Chichester, UK, pp. 135–162.
- BRUNDIN, A., ANDERSSON, G., ANDERSSON, K., MOSSING, T., and KÄLLQUIST, L. 1978. Short-chain aliphatic acids in the interdigital gland secretion of reindeer (*Rangifer tarandus*), and their discrimination by reindeer. *J. Chem. Ecol.* 4:613–622.
- BURGER, B. V., MUNRO, Z., SMIT, D., SCHMIDT, U., WU, C.-L., and TIEN, F.-C. 1990. Sample introduction in gas chromatography: Simple method for the solventless introduction of crude samples of biological origin. *J. Chromatogr.* 518:207–214.
- BURGER, B. V., TIEN, F.-C., le ROUX, M., and MO, W.-P. 1996. Mammalian exocrine secretions X. Constituents of the preorbital secretion of grysbok, *Raphicerus melanotis*. *J. Chem. Ecol.* 22:739–764.
- BURGER, B. V., NELL, A. E., SPIES, H. S. C., LE ROUX, M., BIGALKE, R. C., and BRAND, P. A. J. 1999a. Mammalian exocrine secretions XII: Constituents of interdigital secretions of

- bontebok, *damaliscus dorcas dorcas*, and blesbok, *D. d. phillipsi*. *J. Chem. Ecol.* 25:2057–2084.
- BURGER, B. V., NELL, A. E., SPIES, H. S. C., LE ROUX, M., and BIGALKE, R. C. 1999b. Mammalian exocrine secretions XIII: Constituents of preorbital secretions of bontebok, *Damaliscus dorcas dorcas*, and blesbok, *D. d. phillipsi*. *J. Chem. Ecol.* 25:2085–2097.
- BURGER, B. V., GREYLING, J., and SPIES, H. S. C. 1999c. Mammalian exocrine secretions XIV: Constituents of preorbital secretion of steenbok, *Raphicerus campestris*. *J. Chem. Ecol.* 25:2099–2108.
- BURGER, B. V., SMIT, D., SPIES, H. S. C., SCHMIDT, C., SCHMIDT, U., TELITSINA, A. Y., and GRIERSON, G. R. 2001. Mammalian exocrine secretions XV. Constituents of secretion of ventral gland of male dwarf hamster, *Phodopus sungorus sungorus*. *J. Chem. Ecol.* 27:1259–1276.
- ELLIN, R. I., FARRAND, R. L., OBERST, F. W., CRAUSE, C. L., BILLUPS, N. B., KOON, W. S., MUSSELMAN, N. P., and SIDELL, F. R. 1974. An apparatus for the detection and quantitation of volatile human effluents. *J. Chromatogr.* 100:137–152.
- HUGGINS, G. R., and PRETI, G. 1976. Volatile constituents of human vaginal secretions. *Am. J. Obstet. Gynecol.* 126:129–136.
- JORGENSEN, J. W., NOVOTNY, M., CARMACK, M., COPLAND, G. B., WILSON, S. R., KATONA, S., and WHITTEN, W. K. 1978. Chemical constituents in the urine of the red fox (*Vulpes vulpes L.*) during the winter season. *Science* 199:796–798.
- KAGAN, R., IKAN, R., and HABER, O. 1983. Characterization of a sex pheromone in the jird (*Meriones tristrami*). *J. Chem. Ecol.* 9:775–783.
- LEDERER, E. 1946. Chemistry and biochemistry of the scent glands of the beaver, *Castor fiber*. *Nature* 157:231–232.
- LEDERER, E. 1949. Chemistry and biochemistry of some mammalian secretions and excretions. *J. Chem. Soc.* 1949:2115–2125.
- MADUBUNYI, L. C., HASSANALI, A., OUMA, W., NYARANGO, D., and KABII, J. 1996. Chemoeological role of mammalian urine in host location by tsetse, *Glossina spp.* (Diptera Glossinidae). *J. Chem. Ecol.* 22:1187–1199.
- MO, W.-P., BURGER, B. V., LE ROUX, M., and SPIES, H. S. C. 1995. Mammalian exocrine secretions IX: Constituents of preorbital secretion of oribi, *Ourebia orebi*. *J. Chem. Ecol.* 21:1191–1215.
- OHLOFF, G. 1987. Resent developments in the field of naturally occurring aroma compounds. *Fortsch. Chem. Organ. Naturst.* 35:431–527.
- SPENER, F., MANGOLD, H. K., SANSONE, G., and HAMILTON, J. G. 1969. Long-chain alkyl acetates in the preputial gland of the mouse. *Biochim. Biophys. Acta* 192:516–521.
- STAFFORD, M., HORNING, M. G., and ZLATKIS, A. 1976. Profiles of volatile metabolites in body fluids. *J. Chromatogr.* 126:495–502.
- VASILIEVA, N. Y., and FEOKTISTOVA, N. Y. 1994. Unusual function of integumentary glandular complex in Djungarian hamsters (*Phodopus campbelli*). *Chemical Signals in Vertebrates VII*, Tübingen, Germany, Proceedings, p. 49.
- VOELTLER, D., JUNG, G., BREITMAIER, E., KÖNIG, W., GUPTA, D., and BREITMAIER, G. 1971. Isolierung von Oxindol aus Urin von Kindern. *Z. Naturforsch.* 26b:1380–1381.
- WELLINGTON, J. L., BYRNE, K. J., PRETI, G., BEAUCHAMP, G. K., and SMITH, A. B. 1979. Perineal scent gland of wild and domestic guinea pigs. A comparative chemical and behavioral study. *J. Chem. Ecol.* 5:737–751.