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STRUCTURE-ACTIVITY RELATIONSHIP OF STEROIDAL INSECTICIDES. IX. CHOLESTANE AND STIGMASTANE 5- AND 7-BROMO-6-KETOSTEROIDS

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*Steroids **1-16**, which are intermediates in the synthesis of ecdysteroids, have been shown to possess insecticidal activity for larvae of the Colorado beetle* Leptinotarsa decemlineata. *Active growth- and development-regulators were found among them.*

Key words: 5- and 7-bromo-6-ketosteroids, ecdysteroids, insecticidal activity.

We previously prepared several structural analogs of ecdysteroid insect hormones [1-6]. Biological tests revealed among these compounds active growth- and development-regulators of the Colorado beetle [7]. However, it should be noted that the chemical synthesis of such compounds is complicated and requires many steps. For this reason, it seemed interesting to seek active insecticides among compounds of simpler structure, for example, **1-16**, which we obtained earlier [1-6] and used as intermediates in the chemical synthesis of ecdysteroids. We checked **1-16** for insecticidal activity for larvae of the Colorado beetle *Leptinotarsa decemlineata* Say. (Coleoptera), the most harmful potato pest.

We screened **1-16** using larvae and a contact-feed method of treatment. We collected under field conditions egg sacs of Colorado beetle and grew larvae in the laboratory. Second-growth larvae and their feed, potato leaves, were sprayed once with a 0.01% suspension of **1-16** in water containing the surfactant OP-10. Larvae were fed during the first day by food treated with the tested compounds. Then, the potato leaves were replaced with fresh ones. The number of dead larvae was counted after one, three, and five days. The natural insect-molting hormone 20-hydroxyecdysone (**17**) was selected as the standard and had in the same test the highest insecticidal activity compared with other ecdysteroids [8]. Then, the insecticidal activities of the most active compounds were studied at a concentration of 0.001%.

Table 1 lists the results for the activities of the steroids on Colorado beetle larvae and indicates that at least half of the 22 compounds had at a concentration of 0.01% significant toxicity. The most active were **4b** and **16b**, which killed all insects in their experiments.

It should be noted that the active compounds exhibited various toxicities. In particular, the dynamics of insect death depended on the steroid structure. Compounds **7**, **12**, and **16b** killed the most larvae immediately during the first day after treatment. On the other hand, 3α-5-cyclo-6β-hydroxysteroids **1a** and **2** or 6-ketosteroids **3b**, **5**, **8**, and **10** did not lead to insect death during the first day. The maximum was observed only after three days. Also, larvae died more or less evenly during the first three days after treatment of **13b**.

Using the active compounds at a concentration of 0.001% caused insignificant insect death. As a rule, larvae stopped feeding, became immobile, and remained in this state until molting. Whereas molting of larvae was observed in the control, a few of the experimental larvae did not molt, darkened, and died. Larvae that did manage to complete molting then developed normally.

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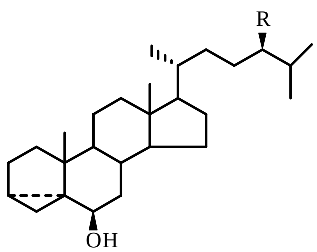
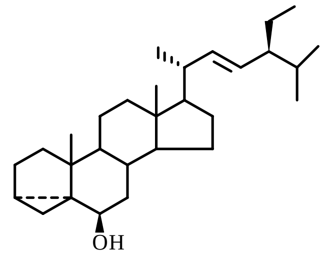
TABLE 1. Toxicity of Steroids for Colorado Beetle Larvae

Compound	Concen-tration, %	No. of larvae	Larvae mortality after							
			1 day		3 days		5 days		Total	
			number	%	number	%	number	%	number	%
1a. 3α,5-Cyclo-5α-cholestan-6β-ol	0.01	30	2	6.7	15	50.0	7	23.3	24	80.0
	0.001	30	7	23.3	2	6.7	4	13.3	13	43.3
1b. (24*R*)-3α,5-Cyclo-5α-stigmastan-6β-ol	0.01	30	2	6.7	0	0	0	0	2	6.7
2. (24*S*)-3α,5-Cyclo-5α-stigmast-22-en-6β-ol	0.01	33	0	0	27	81.8	1	3.0	28	84.8
	0.001	30	0	0	1	3.3	1	3.3	2	6.7
3a. (22*E*,24*S*)-3β-Acetoxy-5-bromo-5α-stigmast-22-en-6-one	0.01	30	9	30.0	1	3.3	0	0	10	33.3
3b. (22*E*,24*S*)-3β-Acetoxy-5α-stigmast-22-en-6-one	0.01	30	3	10.0	19	63.3	0	0	22	73.3
	0.001	30	5	16.7	4	13.3	5	16.7	14	46.7
4a. 3β-Chloro-5-bromo-5α-cholestan-6-one	0.01	30	9	30.0	1	3.3	1	3.3	11	36.7
4b. (24*R*)-3β-Chloro-5-bromo-5α-stigmastan-6-one	0.01	28	0	0	24	85.7	4	14.3	28	100
	0.001	30	2	6.7	2	6.7	0	0	4	13.3
5. (22*R*,23*S*,24*S*)-2β,3β-Diacetoxy-5,22,23-tribromo-5α-stigmastan-6-one	0.01	30	0	0	21	70.0	2	6.7	23	76.7
	0.001	29	4	13.8	7	24.1	1	3.4	12	41.4
6. (22*E*,24*S*)-2β,3β-Diacetoxy-5α-stigmast-22-en-6-one	0.01	31	0	0	12	38.7	0	0	12	38.7
7. (24*R*)-3β-Chloro-5-hydroxy-5α-stigmastan-6-one	0.01	30	22	73.3	5	16.7	0	0	27	90.0
	0.001	29	0	0	0	0	1	3.4	1	3.4
8. (22*E*,24*S*)-3β-Acetoxy-7α-bromo-5α-stigmast-22-en-6-one	0.01	29	0	0	15	51.7	6	20.7	21	72.4
	0.001	30	0	0	1	3.3	1	3.3	2	6.7
9. 3β-Chloro-7α-bromo-5α-cholestan-6-one	0.01	30	0	0	0	0	0	0	0	0
10. (22*E*,24*S*)-3β-Chloro-7α-bromo-5α-stigmast-22-en-6-one	0.01	29	0	0	18	62.1	4	13.8	22	75.9
	0.001	31	3	9.7	2	6.5	1	3.2	6	19.4
11. 7α-Bromo-5α-cholest-2-en-6-one	0.01	30	2	6.7	0	0	0	0	2	6.7
	0.001	30	0	0	2	6.7	7	23.3	9	30.0
12. (22*E*,24*S*)-7α-Bromo-5α-stigmasta-2,22-dien-6-one	0.01	30	18	60.0	2	6.7	3	10.0	23	76.7
	0.001	30	0	0	2	6.7	7	23.3	9	30.0
13a. 7α-Bromocholesta-2,4-dien-6-one	0.01	28	5	17.9	0	0	0	0	5	17.9
13b. Cholesta-2,4-dien-6-one	0.01	28	11	39.3	7	25.0	0	0	18	64.9
14a. (24*R*)-7α-Bromostigmasta-2,4-dien-6-one	0.01	30	2	6.7	7	23.3	1	3.3	10	33.3
14b. (24*R*)-Stigmasta-2,4-dien-6-one	0.01	30	6	20.0	1	3.3	7	23.3	14	46.7
15. (24*R*)-2α,3α-Diacetoxy-7α-bromostigmast-4-en-6-one	0.01	29	1	3.4	3	10.3	9	31.0	13	44.8
16a. 3β-Chloro-5α,7α-dibromo-5α-cholestan-6-one	0.01	30	0	0	0	0	5	16.7	5	16.7
16b. (24*R*)-3β-Chloro-5α,7α-dibromo-5α-stigmastan-6-one	0.01	30	30	-	-	-	-	-	30	100
	0.001	30	0	0	2	6.7	4	13.3	6	20.0
17. 20-Hydroxyecdysone	0.01	30	21	70.0	4	13.3	1	3.3	26	86.7
	0.001	30	8	26.7	4	13.3	4	13.3	16	53.3
Control		30	0	0	0	0	0	0	0	0

The structures of compounds **1-16** differ markedly. Therefore, only tentative conclusions about the importance of certain functional groups for insecticidal activity can be drawn. In particular, it can be supposed that **4b**, **7**, and **16b** have significant insecticidal activity owing to a large extent to the presence in them of a 3β-Cl. This supports an analogous conclusion that was drawn previously [10] using other 3β-chlorosteroids as examples. The observance of insecticidal activity for 3α,5-cyclo-6β-hydroxysteroids **1a** and **2** is also important. These compounds are rather easily prepared from cholesterol and stigmasterol, respectively, which may provide a basis for defining the direction of further research.



1a: R = H
b: R = Et



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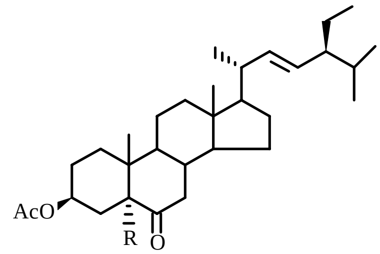
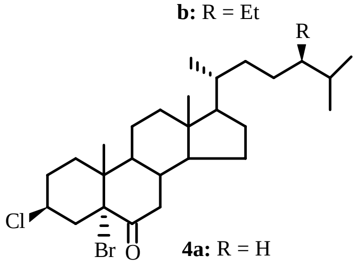


3a: R = Br
b: R = H



4a: R = H
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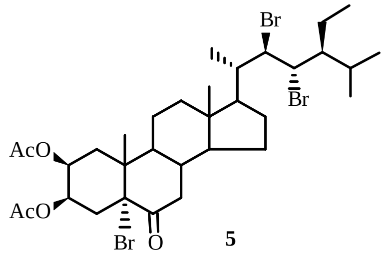

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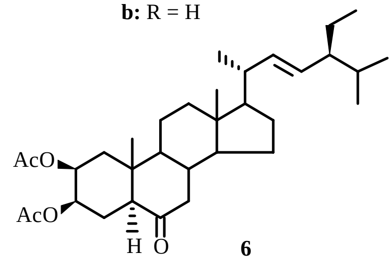

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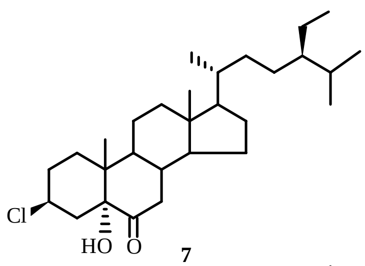

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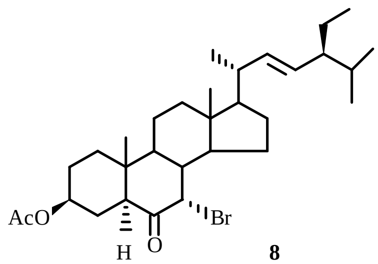

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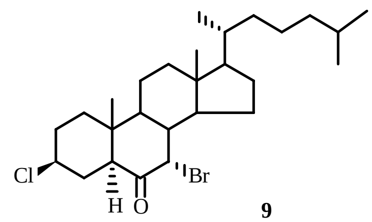

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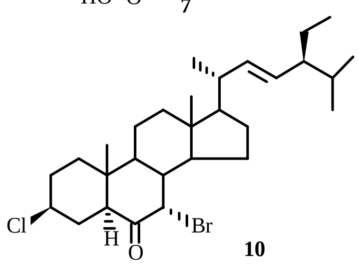

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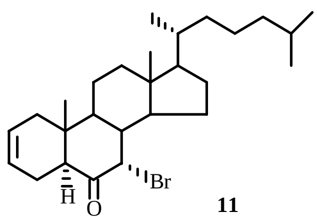

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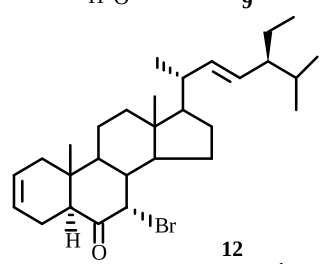

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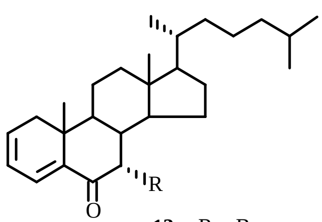
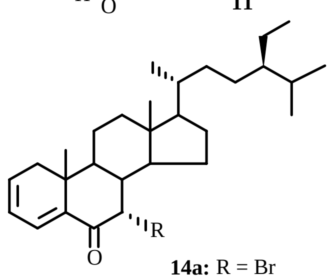
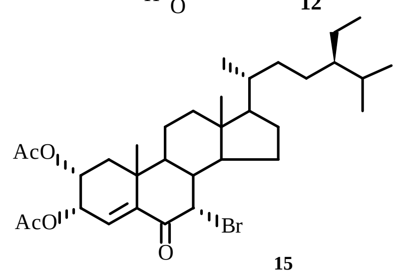


13a: R = Br
b: R = H



14a: R = Br
b: R = H



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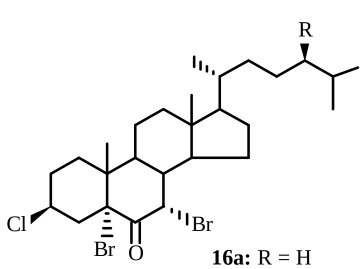
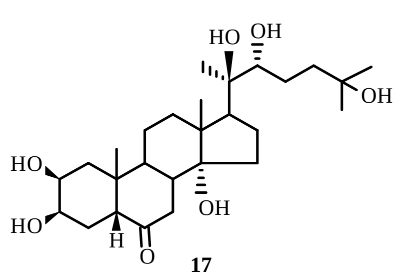


16a: R = H
b: R = Et



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EXPERIMENTAL

The syntheses of steroids **1-16** have been reported [1-6].

Details of the experiments for determining the insecticidal activity of compounds **1-17** for second-growth Colorado beetle larvae have been described [9].

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REFERENCES

1. N. V. Kovganko and V. L. Survilo, *Khim. Prir. Soedin.*, 359 (1999).
2. N. V. Kovganko and V. L. Survilo, *Khim. Prir. Soedin.*, 309 (2000).
3. N. V. Kovganko and V. L. Survilo, *Khim. Prir. Soedin.*, 402 (2000).
4. N. V. Kovganko and V. L. Survilo, *Khim. Prir. Soedin.*, 135 (2001).
5. N. V. Kovganko and V. L. Survilo, *Khim. Prir. Soedin.*, 216 (2002).
6. N. V. Kovganko and V. L. Survilo, *Khim. Prir. Soedin.*, 284 (2002).
7. R. M. Zolotar′, A. I. Bykhovets, V. L. Survilo, and N. V. Kovganko, *Khim. Prir. Soedin.*, 358 (2002) (previous article).
8. R. M. Zolotar′, A. I. Bykhovets, and N. V. Kovganko, *Khim. Prir. Soedin.*, 460 (2001).
9. R. M. Zolotar′, A. I. Bykhovets, S. N. Sokolov, and N. V. Kovganko, *Khim. Prir. Soedin.*, 462 (2001).
10. R. M. Zolotar′, A. I. Bykhovets, S. N. Sokolov, and N. V. Kovganko, *Khim. Prir. Soedin.*, 57 (2002).