labelled calysterol, while apparently stigmasterol and β-sitosterol cannot act as precursor of this unique sterol. Purification of calysterol through the diacetates, the diols and again the diacetates in the experiment with [7,7-$^3H_2$]-fucosterol shows that the radioactivity associated with the calysterol derivatives is substantially constant in all the steps of the purification, whilst in the case of the experiments with [7,7-$^3H_2$]-stigmasterol and [7,7-$^3H_2$]-β-sitosterol, the radioactivity decreases to the final values given in table 2.

In a trial experiment, 'cold' calysterol was mixed with labelled fucosterol and acetylated in the hot: The calysterol derivatives, purified as above, were found to be devoid of significant radioactivity.

Although these findings are not conclusive if fucosterol is indeed the true calysterol precursor, nevertheless they add further support to the suggestion that the sponges are unable to synthesize their sterols de novo but they modify sterols taken up from the diet.

Completion of the natural groups of ergot alkaloids: Syntheses and pharmacological profiles of β-ergosine and β-ergoptine*

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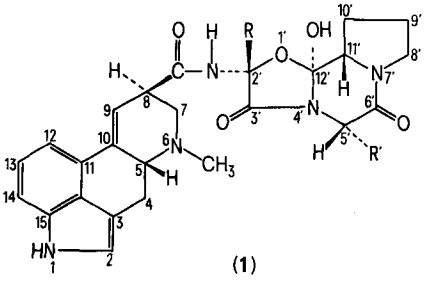
Summary. The syntheses and pharmacological potencies of β-ergosine and β-ergoptine, the missing links in the natural groups of ergot peptide alkaloids are described.

Ergot alkaloids occuring in nature can be divided into 3 groups by their substitution in position 2′ of the peptide moiety (see depicted general structure **1** and corresponding table).



(1)

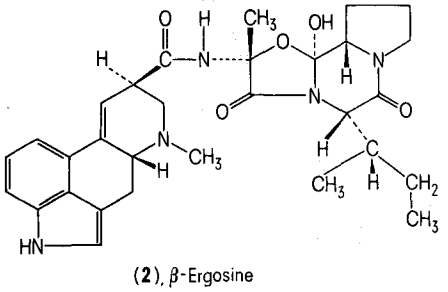
R′	Ergotamine group R=CH_3	Ergoxine group R=C_2H_5	Ergotoxine group R=$CH(CH_3)_2$
CH_2–C_6H_5	Ergotamine	Ergostine	Ergocristine
CH_2–$CH(CH_3)_2$	Ergosine	Ergoptine*	α-Ergokryptine
$CHCH_3$–C_2H_5	β-Ergosine*	β-Ergoptine*	β-Ergokryptine
$CH(CH_3)_2$	Ergovaline*	Ergonine*	Ergocornine

* Not yet found in nature.

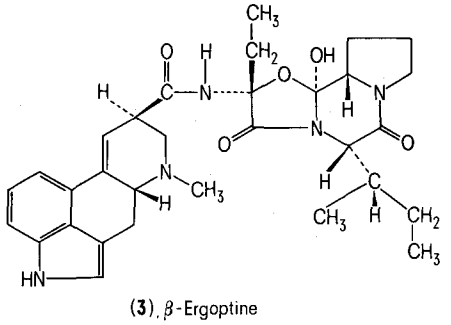
4 natural alkaloids are found in the ergotoxine group characterized by an isopropyl residue in position 2′ of the peptide moiety: ergocristine, α-ergokryptine, β-ergokryptine and ergocornine. In the 2 other groups, smaller numbers of natural alkaloids occur: in the ergotamine group ergotamine and ergosine, in the ergoxine group ergostine only.

In order to complete the 2 latter groups ergovaline[1], ergoptine and ergonine[2] have formerly been synthetized. Discovery[3] and synthesis[4] of the 4th natural member of the ergotoxine group, β-ergokryptine, prompted us to synthetize the corresponding analoga of the ergotamine and ergoxine group and to investigate their pharmacological activities.

In line with the existing nomenclature, the name β-ergosine (**2**) is proposed for the alkaloid of the ergotamine group and the name β-ergoptine (**3**) for the alkaloid of the ergoxine group.



(**2**), β-Ergosine



(**3**), β-Ergoptine

The syntheses of the 2 alkaloids have been accomplished in strict analogy to those of ergosine[1], respectively ergoptine[2], with the only difference that L-isoleucyl-L-proline-lactam[4] was used as essential building block instead of L-leucyl-L-proline-lactam. The syntheses and absolute configurations of S-(+)-methyl-benzyloxy-malonic-acid-monoethylester-chloride[5] used for the synthesis of β-ergosine, respectively S-(+)-ethyl-benzyloxy-malonic-acid-monoethylester-chloride[6] used for the synthesis of β-ergoptine were known. Hence follow automatically the absolute configurations of the 2 new alkaloids given in formulas **2** and **3**, since it is known that the syntheses

Table 1

Name of intermediate or alkaloid	Synthesis of β-ergosine m.p.	$(\alpha)_D^{20}$	Synthesis of β-ergoptine m.p.	$(\alpha)_D^{20}$
Cyclolcarbonic acid-ethylester	124–125 °C	+ 11.2°, c = 3, ethanol	84–85 °C	+ 18.0°, c = 1, ethanol
Cyclolcarbonic acid	173–174 °C (dec.)	+ 16.6°, c = 1, ethanol	164–167 °C (dec.)	+ 25.4°, c = 1, ethanol
Benzyloxycarbonyl-aminocyclol	188–189 °C	+ 26.6°, c = 1, ethanol	180.5–181.5 °C (dec.)	+ 33.7°, c = 1, ethanol
Aminocyclol-hydrochloride	122–123 °C (dec.) contains 1 mole DMF	+ 62.5°, c = 1, dimethyl-formamide	167–169 °C (dec.)	+ 59.4°, c = 1, dimethylformamide
β-Ergosine (**2**)	203–205 °C (dec.)	+ 5.2°, c = 1, pyridine −153.0°, c = 0.5, chloroform		
β-Ergosinine	206–208 °C (dec.)	+443.0°, c = 1, chloroform +446.0°, c = 2, pyridine		
β-Ergoptine (**3**)			201–203 °C (dec.)	−163°, c = 1, chloroform − 22.6°, c = 0.5, pyridine
β-Ergoptinine			205–206 °C (dec.)	+421°, c = 0.5, chloroform +488°, c = 0.5, pyridine

of the ergot-peptide alkaloids proceed with complete steric control[7].

In table 1, melting points and optical rotations of the most important intermediates of both syntheses are given, in addition to those of β-ergosine (**2**) and β-ergoptine (**3**) and their iso-forms. For all compounds, correct elemental analyses and spectral data which are in agreement with the corresponding structures are available.

6 main activities of ergot alkaloids were investigated pharmacologically: a) The α-adrenoceptor blocking activity in the adrenaline-stimulated guinea-pig seminal vesicle in vitro[8]. b) The serotonin antagonism in the serotonin-stimulated rat uterus in oestrus[9,10]. c) The vaso-

* 86th communication on ergot alkaloids.

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Table 2

	α-Adrenoceptor-blockade isolated guinea-pig seminal vesicle		5HT Receptor blockade isolated rat uterus		Pressor activity spinal cat i.v. injection		Oxytocic activity rabbit uterus in situ i.v. injection		Inhibition of fertility in rats s.c. injection		Acute toxicity (LD_{59}) rabbits, i.v. injection	
Ergotamine	100 $EC_{50}1.4 \cdot 10^{-8}$ g/ml		100 $EC_{50}5.8 \cdot 10^{-8}$ g/ml		100 ED 1–3 µg/kg		100 ED 0.1–0.3 mg/kg		100 ED_{50} 15.8 mg/kg		100 ED_{50} 1.26 mg/kg	
Ergosine	230	100	90	100	100	100	220	100	280	100	100	100
β-Ergosine	175	75	125	145	155	155	130	60	250	90	90	90
Ergoptine	350	100	20	100	100	100	80	100	750	100	130	100
β-Ergoptine	350	100	65	325	100	100	10	13	830	110	115	90
α-Ergokryptine	210	100	40	100	25	100	19	100	1600	100	130	100
β-Ergokryptine	320	150	50	125	50	200	22	115	570	35	160	125
	a	b	a	b	a	b	a	b	a	b	a	b

Pharmacological activities of β-(5′α-sec.butyl)-ergot peptide alkaloids compared with those of the corresponding α-(5′α-isobutyl)-ergot peptide alkaloids (= 100; columns b) and those of ergotamine (= 100, columns a) in 6 different experiments. For ergotamine absolute values are given additionally.

Abbreviations: EC_{50} (ED_{50}), Effective concentration (dose) to generate 50% of the maximal effect. ED, effective dose. LD_{50}, Dose killing 50% of the animals within 24 h.

constrictor activity in the spinal cat preparation[11] (i.v. injection). d) The oxytocic activity using the uterus of non-pregnant rabbit in spontaneous oestrus[12,13]. e) The antifertile activity in rats after s.c. injection on day 5 post coitum (spermatozoa control; autopsy at day 12 p.c.). f) The acute toxicity in rabbits after i.v. injection (LD_{50} Probit-method[14]).

The results are given in table 2. 2 comparisons were calculated: 1. Activities in percent of those of ergotamine = 100. 2. Activities of the β-alkaloid in percent of those of the α-alkaloid = 100. Additionally absolute values are given for ergotamine.

Table 2 shows that the activity profiles of the ergot alkaloids under investigation are qualitatively similar but differ quantitatively. No systematic difference exists between the α- and β-alkaloids with the exception of serotonin-antagonism which is more potent in the β-alkaloids. The greatest quantitative difference was found between α- and β-ergoptine concerning uterotonic activity (factor 8) followed by the serotonin-antagonism (factor 3), and between α- and β-ergokryptine in the antifertile activity (factor 3). All other differences in activity are smaller.

The results compare well with those reported earlier for α- and β-ergokryptine[15].

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New tigliane and daphnane derivatives from *Pimelea prostrata* and *Pimelea simplex*

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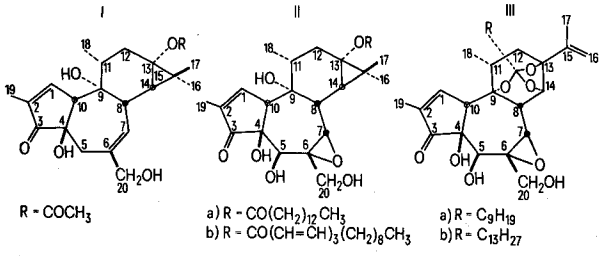
Summary. From the methanol extract of *Pimelea prostrata*, prostratin (**I**) and 2 autoxidation products have been isolated. They are tigliane derivatives and relatively nonirritant on the mouse ear. The irritant pimelea factor P_5 (**IIa**) also with a tigliane skeleton and related to mancinellin (**IIb**), as well as the irritant diterpene ester pimelea factor P_1 (**IIa**, simplexin) with daphnane skeleton, were found to be present in both *P. prostrata* and *P. simplex*. Further the irritant homologue of simplexin, pimela factor **IIIb** was detected in *P. prostrata*. Some biogenetic consequences of these findings are discussed.

A large number of species of the Euphorbiaceae are known to contain toxic, irritant and cocarcinogenic diterpene esters of the tigliane and/or daphnane as well as of the ingenane type[4]. From species of the Thymelaeaceae, the toxic and irritant diterpene esters isolated until recently[4a] were of the daphnane type, e.g. mezerein[5] from *Daphne mezereum* L. (spurge laurel) and simplexin[6] from *Pimelea simplex* F. Muell. (desert rice flower). Both toxins are cocarcinogenic[4,7,8] in mouse skin. Moreover, mezerein[9] and crude extracts of *P. simplex*[10] were shown to exhibit antileucemic activity. The first tigliane derivative from the Thymelaeaceae family, prostratin [13-O-acetyl-12-deoxyphorbol (**I**)], was isolated recently from the strathmore weed *Pimelea prostrata* Willd.[11]. This plant is a small endemic New Zealand shrub known to be toxic to livestock[12], extracts of which were reported to exhibit antitumor activity[11]. Now we wish to report on further new tigliane and daphnane derivatives isolated from *P. prostrata* and *P. simplex* (figure).

From methanol extracts of different air-dried parts of *P. prostrata*, by a combination of counter current distribution and chromatographic methods, besides the main constituent prostratin (**I**)[11], some oxidation products derived from it, as well as the new pimelea factors P_1, P_4 and P_5, were obtained (for irritancy and some other data see table).

$\Delta^{5,6}$-7-Hydroperoxide of **I** (table): IR (CH_2Cl_2): 3380, 3560 (OH); 1705 (CO); 1620 cm^{-1} (C=C); UV (MeOH):



Structures of prostratin (**I**), mancinellin (**IIb**) and simplexin (pimelea factor P_1, **IIIa**) together with the new pimelea factors P_5 (**IIa**) and P_4 (**IIIb**) isolated from *Pimelea prostrata*.

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3 Acknowledgment: The authors are deeply indebted to Dr T. Cashmore (Palmerton North, New Zealand) and Dr H. B. Roberts (Sydney, Australia) for kindly supplying plant materials.

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