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PHYSALINS E AND H, NEW PHYSALINS FROM *PHYSALIS ANGULATA* AND *P. LANCIFOLIA**

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Key Word Index—*Physalis angulata*; *Physalis lancifolia*; Solanaceae; physalins B, E, F, G, H and I; structural elucidation.

Abstract—From the stems and leaves of *Physalis angulata* and *P. lancifolia*, the isolation of five new physalins E, F, G, H and I is reported. The structures of physalins E and H are established respectively as 5,6-dihydro-5α,7α-dihydroxy and 7β-hydroxy derivatives of physalin B.

INTRODUCTION

During a morphological study of *Physalis* species grown in the Botany Department of Andhra University, we became interested in the chemical composition of *Physalis angulata* and *Physalis lancifolia*. The former were raised from seeds procured from Lucknow and Copenhagen and though there were no marked morphological differences between the two plants, the chemical composition of leaves and stems appeared to differ in both varieties sufficient to merit a full scale investigation of their chemical constituents.

RESULTS AND DISCUSSION

The air-dry green stems and leaves were extracted with hot methanol in a Soxhlet apparatus separately and the extracts fractionated between *n*-hexane and chloroform. The green chloroform extract was freed from chlorophylls by adsorption on alumina and eluting with benzene–ethyl acetate (1:1). The eluate on evaporation in vacuum, furnished a mixture of physalins which were successfully separated on a Si gel column. Table 1 gives the physalins identified in the stems and leaves of the two varieties of *Physalis angulata*. Similar studies were then extended to the stems and leaves of *P. lancifolia*, grown from the seeds collected from Canada (Table 1).

The plants for the Copenhagen variety, were the most prolific and contained as many as four new physalins (E, F, H, and I), hitherto unnoticed in *Physalis* species [1–4]. The Lucknow variety contained only two (E and F) but differed in having another new physalin G. *P. lancifolia* likewise yielded three new physalins E, F, and G. Physalin B (**1**) was the major component in both plants. The new physalins E to I were named by letters in conformity with previous practice in this group.

The structure of physalins E (**2**) *and H* (**3**)

Both physalins E (**2**) and H (**3**) bear a close resemblance to physalin B (**1**), first isolated from *P. alkekangi* [1]. Physalin E (**2**) $C_{28}H_{32}O_{11}$, M^+ 544, mp 305–307°, $[\alpha]_D$ −83°, contained the characteristic methylene oxide bridge between C-14 and C-26 (26-*H*a, δ 4.28, *dd*, J = 14 Hz; 26-*H*b, δ 3.60, *dd*, J = 14, 2 Hz) as found in physalin B and other common functional groups including the tertiary hydroxyl at C-13. However, it lacked the 5:6 double bond and contained in addition a tertiary hydroxyl (δ 4.22, *s*, 5-O*H*) and a secondary hydroxyl (δ 4.86, *d*, J = 4 Hz, 7-O*H*). Physalin E (**2**) furnished a monoacetate (**2a**), $C_{30}H_{34}O_{12}$, M^+ 586, mp 278–279°, $[\alpha]_D$ −75°, which contained two free tertiary hydroxyls, (δ 5.78, *s*, 13-O*H*; 4.76, *s*, 5-O*H*). On oxidation with Jones' reagent, the secondary hydroxyl disappeared and gave rise to the ketone (**4**) in which the tertiary hydroxyls showed a very significant down-field shift (δ 6.01, *s*, 13-O*H*; 5.80, *s*, 5-O*H*) suggesting that the 5-hydroxyl was released from 1:3-interaction, if any, from 7-hydroxyl.

Physalin E (**2**) was inert to aqueous periodate in neutral or acid medium; but lost a molecule of water when refluxed in acetic acid with or without a trace of H_2SO_4 giving rise to anhydrophysalin E (**5**), $C_{28}H_{30}O_{10}$, M^+ 526, mp 280–281°. Its UV spectrum, λ_{max}^{EtOH} 229 nm

Table 1. Physalins isolated from *Physalis* species

Compound	R_f* value	mp	*Yield* ($\times 10^{-3}$ %) *P. angulata* Copenhagen Leaves	*P. angulata* Copenhagen Stems	*P. lancifolia* Lucknow Stems	*P. lancifolia* Lucknow Stems
Physalin B	0.87	264–266°	30	8	9	10
Physalin F	0.77	294–296°	20	5	6	5
Physalin G	0.65	295–297°	—	—	2	3
Physalin H	0.73	234–236°	—	2	—	—
Physalin I	0.69	294–295°	—	2	—	—
Physalin E	0.45	305–307°	30	9	9	5

* Si gel TLC; EtOAc–C_6H_6 (7.3).

* Part 1 in the series 'New Physalins from *Physalis angulata* and *P. lancifolia*'.

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Table 2. NMR chemical shifts of relevant protons in new physalins and their derivatives

Compound	2-H	3-H	6-H	7-H	22-H	26-H_a	26-H_b	13-OH	5-OH	7-OH	Additional groups
2	5.70*dd* (10, 2)	6.61*dm* (10)	—	3.52*m*	4.56*m*	4.27*dd* (14, 4)	3.60*dd* (14, 2)	5.66*s*	4.22*s*	4.86*d* (4)	—
2a	5.74*dd* (10, 2)	6.57*dm* (10)	—	4.76*m*	5.56*m*	4.25*dd* (14, 4)	3.57*d* (14)	5.78*s*	4.76*s*	—	—
3	5.84*dd* (12, 2)	6.78*dm* (12)	5.65*m*	3.88*m*	4.58*m*	4.29*dd* (14, 4)	3.61*d* (14)	5.96*s*	—	4.94*d* (4)	
4	5.76*dd* (10, 3)	6.67*dm* (10)	—	—	4.60*m*	4.28*dd* (12, 4)	3.78*d* (13)	6.01*s*	5.80*s*	—	—
5	5.70*dd* (10, 3)	6.59*dm* (10)	4.78*m*	*	4.56*m*	4.26*dd* (14, 5)	3.57*d* (14)	5.68*s*	—	4.78*m*	—
6	5.75*dd* (10, 2)	6.61*dm* (10)	—	4.12*m*	4.56*m*	4.29*dd* (14, 4)	3.60*d* (13)	5.95*s*	4.94*s*	—	—
7	—	—	—	*	4.58*m*	4.20*dd* (13, 4)	3.58*d* (13)	5.52*s*	4.06*s*	4.73*d* (4)	—
8	—	—	—	*	4.46*d*	—	—	5.67*s*	3.98*s*	4.66*d* (4)	14-OH, 6.12*s* 25-CH_3, 1.15*d* (6)

* merged with *H*DO of DMSO-d_6. Spectra were taken in DMSO-d_6 solution; chemical shifts are in δ units; coupling constant (*J*) in parentheses are given in Hz.

(ε 6300), suggested no formation of extended conjugation by dehydration and in its ^{1}H NMR spectrum, the δ 4.22 *s* signal for the tertiary hydroxyl disappeared. The broad multiplet at δ 4.78 integrated for 2 H, one of which exchanged with D_2O and the signal was reduced to half its intensity, exposing the new olefinic proton. The new double bond was, therefore, placed at the 5:6 position supporting the earlier placement of the tertiary hydroxyl at C-5. Further dehydration of anhydrophysalin E (**5**) with sulphuric acid gave the known dehydrophysalin B (**6**) [1], which was also obtained from physalin E (**2**) in a similar manner, confirming the same skeletal arrangement of physalin E (**2**) as in physalin B (**1**).

When physalin E (**2**) was treated with $POCl_3$–pyridine, an isomeric anhydrophysalin E (**6**), $C_{28}H_{30}O_{10}$, mp 278–280°, was formed whose ^{1}H NMR spectrum contained no secondary hydroxyl, and a single olefinic proton at δ 4.12, *m*, was readily identified. The new double bond was, therefore, located at the 7:8 position. The upfield signal for C-6H (δ 4.78 *m*) in anhydrophysalin E (**5**) and that of 7-H (δ 4.12, *m*) in *iso*-anhydrophysalin E (**7**) are not easy to explain. But the 26-14 methylene oxide bridge and the neighbouring 7α-hydroxyl in anhydrophysalin E (**5**) and 5α-hydroxyl in *iso*-anhydrophysalin E (**7**) may be responsible for this upfield shift.

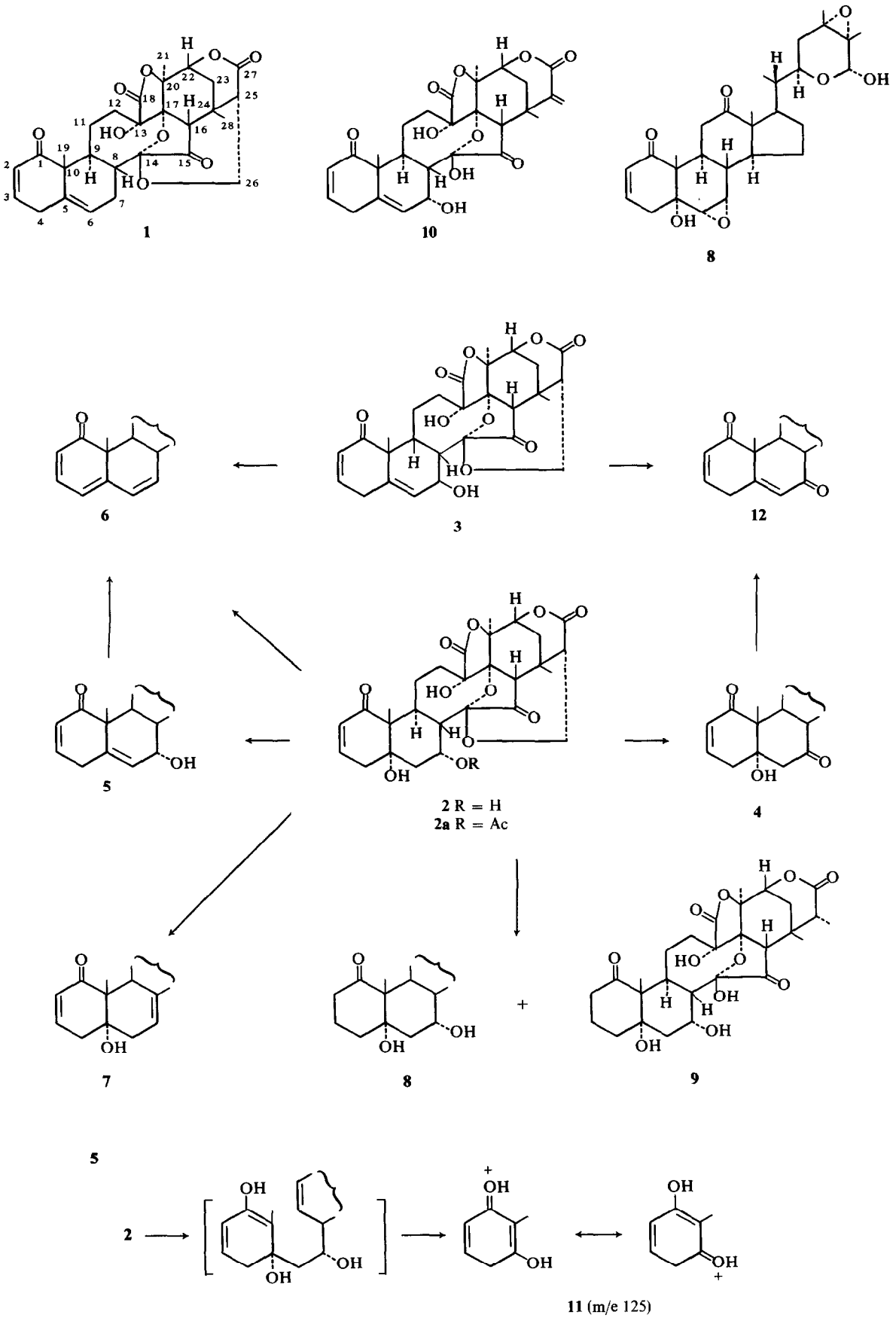
Further chemical study of these two anhydrophysalins (**5**, **7**) emphasized this point. The 7α-hydroxyl in anhydrophysalin E (**5**) was resistant to acetylation and also to oxidation, unlike its susceptibility in the parent physalin E (**2**). This change in behaviour can be explained only by assigning a 7α-axial configuration (7-O*H*, δ 4.86, *d*, J = 4 Hz) to this hydroxyl whose NMR signals resemble those of the 7α-hydroxyl in physalin A (**10**) (7-O*H*, δ 4.97, *d*, J = 4 Hz).

However, the 7β-hydrogen in physalin E (**2**) appeared at considerably higher field at δ 3.52, *m* (δ 4.76, *m*, in its acetate) than that of physalin A (**10**) (7-H, δ 4.58, *m*; physalin A acetate, 7-H, 5.60, *m*). Obviously, this proton might have come into the shielding influence of the 26-14 oxide bridge in physalin E.

When physalin E (**2**) was hydrogenated over Pd/C in ethanol at room temp. and atmospheric pressure, two products were formed which, from their molecular ions, were shown to be dihydrophysalin E (**8**), M^+ 546, mp 274–276°, and tetrahydrophysalin E (**9**), M^+ 548, mp 288–289°. Their structures were readily assigned from a study of their ^{1}H NMR spectra. In dihydrophysalin E (**8**), the disappearance of olefinic proton signals confirmed the reduction of the 2:3 double bond in physalin E, while in tetrahydrophysalin E, besides the disappearance of these two olefinic protons and 26-CH_2 resonances, there appeared an additional tertiary hydroxyl at δ 6.12, *s*, and an additional secondary methyl at δ 1.15, *d*, J = 6 Hz, which closely resemble those found in physallin A (**10**) (14β-OH, δ 6.20, *s*) and tetrahydrophysalin A (25α-C*H*$_3$, δ 1.15, *d*, J = 6 Hz). This indicated that the 26-methylene oxide bridge suffered a reductive cleavage and the tetrahydrophysalin E resembled closely tetrahydrophysalin A in its NMR spectrum.

Under the electron impact at 75 eV (230°), physalin E suffered extensive fragmentation such that no meaningful deductions can be drawn. However, the fragment (**11**) at *m/e* 125 might arise out of McLafferty rearrangement as in withanolides [5] providing support for the presence of 5α-hydroxyl in physalin E (**2**). The α-configuration of the 5-hydroxyl was finally confirmed by the ORD and CD spectra of physalin E (**2**) and its acetate (**2a**). Both compounds showed the strong negative Cotton effect of the cyclohexenone $n \rightarrow \pi^*$ band near 340 nm in accordance with those of the steroidal constituents of *Nicandra physaloides*, for example, Ni -7 [6], and also showed two negative CD bands near 340 nm attributable to the $n \rightarrow \pi^*$ bands of 1- and 15-carbonyls in accordance with those of 5α,6α-epoxyphysalin B [7]. Other Cotton effects and a negative CD band were observed at shorter wavelengths for both **2** and **2a**, possibly attributable to the lactone $n \rightarrow \pi^*$ and enone $\pi \rightarrow \pi^*$ bands.

Physalin H (**3**) is a minor steroid, $C_{28}H_{30}O_{10}$, M^+ 526, mp 238–240° isomeric with anhydrophysalin E (**5**). Besides an olefinic proton (δ 5.65, *m*), it contains a secondary hydroxyl (δ 4.94, *d*, J = 4 Hz) (monoacetate).


1
10
8
6
3
12
5
2 R = H
2a R = Ac
4
7
8
9
5
2
11 (m/e 125)

The single olefinic proton, however, differed in its chemical shift from that of anhydrophysalin E (δ 4.78, *m*, 6-C*H*). Treatment of physalin H (**3**) with sulphuric acid gave dehydrophysalin B (**6**), which was obtained from anhydrophysalin E (**5**) in a similar manner, indicating the epimeric relation between **3** and **5** with respect to the 7-hydroxyl. Furthermore, **3** was directly correlated to physalin E (**2**) as follows. The hydroxyl of **3** could be oxidized with Jones' reagent and the resulting product $C_{28}H_{28}O_{10}$, mp 284° was found identical with physalin-B-7-one (**12**) by mixed mp and IR. The latter was secured from physalin E (**2**) in a two-step process by first oxidizing with Jones' reagent to give physalin-E-7-one (**4**) followed by dehydration with glacial acetic acid containing a trace of H_2SO_4. Physalin-B-7-one (**12**) had the expected UV absorption, at 232 nm (ε 13200). These experiments confirm C-7 for the hydroxyl in physalin H (**3**). However, the olefinic proton in physalin H (**3**) (δ 5.65, *m*, 6-C*H*) differs significantly from that of anhydrophysalin E (δ 4.78, *m*, 6-C*H*) but corresponds very well with that of Δ^5-steroids [7]. This difference is probably due to β-equatorial configuration of the 7-hydroxyl in physalin H.

Physalins E and H are closely related to physalin B, the major component of *P. angulata* and *P. lancifolia*. Their formation in the same plant has some biogenetic as well as taxonomic significance.

EXPERIMENTAL

The seeds of *Physalis angulata* (Solanaceae) were secured from Copenhagen (Denmark) and Lucknow (India) and the plants grown in the Botanic Farm, Andhra University. Each group of plants were examined separately.

Extraction of the stems of Physalis angulata *(Copenhagen).* Powdered air-dried stems (12 kg) of *Physalis angulata* (Copenhagen) were extracted with hot EtOH (50 l.). The inorganic salts (yield: 350 g) were filtered off and the filtrate concd (5 l.). Additional inorganic salts separated out (250 g) were filtered off. The filtrate was concd (2 l.), adsorbed on spent powder (1.5 kg) and re-extracted with *n*-hexane and $CHCl_3$ successively. The dark green hexane extract (4 litres) was concd (0.5 litres) and the residual solvent removed *in vacuo*. On keeping for a month, a solid compound separated which was collected and recrystallized from MeOH to give shining needles (1.5 g), mp 135° (compound A). As the mother liquors did not give any crystals even after several attempts it was not examined further.

The green $CHCl_3$ extract (4 l.) was evapd to a dark green residue (45 g). No pure compound could be separated by fractional crystallization of this residue. Hence, it was adsorbed on neutral alumina (100 g) and after drying placed on the top of a column (95 × 6 cm) of alumina (150 g) in C_6H_6. The column was developed with C_6H_6–EtOAc (1:1) until no more compound was eluted (4 litres). The eluant, on evapn yielded a pale green residue (9.5 g) which did not yield any pure compound on fractional crystallization from MeOH, Me_2CO or EtOAc. The residue was dissolved in Me_2CO (40 ml) and adsorbed on Si gel (30 g). The dry powder was placed on to a column (90 × 6 cm) of Si gel (100 g, 100–200 mesh) in C_6H_6. The column was eluated with C_6H_6–EtOAc, 250 ml fractions being collected. Table 3 lists the fractions collected and the compounds isolated from them.

Table 3. Separation of physalins from the stems of *Physalis angulata* (Copenhagen)

Fraction	Eluant (C_6H_6–EtOAc)	Compound	R_f Value*	Yield (g)†
19–31	82:18	Physalin B	0.87	1.5
49–64	82:18	Physalin F	0.77	1.2
65–73	82:18	Physalin H	0.73	1.15
78–88	75:25	Physalin I	0.69	0.22
97–112	65:35	Physalin E	0.45	1.5

* Si gel TLC, benzene–ethyl acetate (7:3).
† Yield after crystallization.

Table 4. Separation of physalins from the leaves of *Physalis angulata* (Copenhagen)

Fraction	Eluant (C_6H_6–EtOAc)	Compound	R_f Value*	Yield (g)†
13–15	85:15	Physalin B	0.87	0.25
18–32	85:15	Physalin F	0.77	0.42
61–75	65:35	Physalin E	0.45	0.13

* Si gel TLC, EtOAc–C_6H_6 (7:3).
† Yield after crystallization.

Table 5. Separation of physalins from the stems of *Physalis angulata* (Lucknow)

Fraction	Eluant (C_6H_6–EtOAc)	Compound	R_f* Value	Yield (g)†
9–12	82:18	Physalin B	0.87	0.32
18–25	82:18	Physalin F	0.77	0.05
32–49	82:18	Physalin G	0.65	0.26
73–88	70:30	Physalin E	0.45	0.37

* Si gel TLC, EtOAc–C_6H_6 (7:3).
† Yield after crystallization.

Table 6. Separation of physalins from *Physalis lancifolia*

Fraction	Eluant (C_6H_6–EtOAc)	Compound	mp	R_f* Value	Yield (g)
14–20	82:18	Physalin B	262–264°	0.87	1.1
27–32	82:18	Physalin F	295–296°	0.77	0.35
50–62	82:18	Physalin G	295–296°	0.65	0.22
91–105	70:30	Physalin E	305–307°	0.45	0.9

* Si gel TLC, EtOAc–C_6H_6(7:3).

Extraction of the leaves of Physalis angulata *(Copenhagen).* The leaves (1.2 kg) of *Physalis angulata* (Copenhagen) were similarly treated by the above isolation procedure. The residue (9 g) from the $CHCl_3$ extract was adsorbed on Si gel G (20 g) and after drying placed on the top of a column (90 × 6 cm) of Si gel (100 g). The column was eluted with C_6H_6–EtOAc, 250 ml fractions being collected. Table 4 presents the compounds isolated

Extraction of the stems of Physalis angulata *(Lucknow).* The stems (6.0 kg) of *Physalis angulata* (Lucknow) were extracted in a similar way. The residue (15 g) from the $CHCl_3$ extract was adsorbed on Si gel G (35 g) and after drying placed on top of a column (90 × 6 cm) of Si gel (170 g). Elution with C_6H_6–EtOAc collecting 250 ml fractions gave compounds shown in Table 5.

Extraction of the aerial parts of Physalis lancifolia. The fresh air-dried aerial parts (2.0 kg) of *Physalis lancifolia* were successively extracted with hexane (6.0 litres) and $CHCl_3$ (6.0 litres). The $CHCl_3$ extract was evapd and the residue (18 g) was adsorbed on Si gel G (40 g) and after drying placed on a column (60 × 6 cm) of Si gel G in C_6H_6. The column was eluted with C_6H_6–EtOAc collecting 250 ml fractions. Table 6 gives the compounds isolated from the chromatographic fractions.

Identification of compound A as sitosterol. Compound A crystallized from MeOH as colourless prisms, mp 135°, undepressed by an authetic sample of sitosterol, $[\alpha]_D$ − 36°, (*c*, 0.125, $CHCl_3$).

Identification of physalin B (**1**). Compound **1** crystallized from Me_2CO–MeOH (1:1) as short colourless needles, mp 264–66°, $(\alpha)_D$ − 149° (*c*, 0.9, $CHCl_3$) (Found: C, 65.13, H, 6.01; M^+ 510. $C_{28}H_{30}O_9$ requires: C, 65.87; H, 5.92%); λ_{max}^{EtOH} 228 nm (ε 10000); ν_{max}^{Nujol} cm^{-1} 3400, 1790, 1757, 1732, 1665. The IR and NMR spectra of **1** were identical with those of authentic physalin B [1].

Physalin E and acetate **2** *and* **2a** (*carried out by K. S. Reddy*) Physalin E crystallized from MeOH as white shining plates, mp 305–307°; $[\alpha]_D$ − 83°; (*c*, 0.5, $CHCl_3$) (Found: C, 61.07; H, 6.09%; M^+ 544. $C_{28}H_{32}O_{11}$ requires: C, 61.82; H, 5.92%): λ_{max}^{EtOH} 229 nm (ε 6800); ν_{max}^{Nujol} cm^{-1} 3400, 1780, 1767, 1730, 1665; ORD (dioxan), $[M]_{356\ nm}$ − 5700° (min), $[M]_{312}$ + 3700° (max), $[M]_{246}$ − 8500° (min), $[M]_{216}$ + 30400° (max), $[M]_{205}$ + 25300° (min); CD (dioxan) λ_{max} 344 nm (Δε − 2.38), 338 (−2.35), 333 (−2.41), 270 (−0.05), 231 (−7.76) (strongly positive at shorter wavelengths). The acetate (Py–Ac_2O at 100° for 3 hr) of physalin E crystallized from MeOH as colourless shining needles, mp 278–79°; $[\alpha]_D$ − 75° (*c*, 0.79, $CHCl_3$); (Found: C, 61.07; H, 6.1; M^+ 586. $C_{30}H_{34}O_{12}$ requires: C, 61.43; H, 5.85%); λ_{max}^{EtOH} 228 nm (ε 8900); λ_{max}^{Nujol} cm^{-1}: 3400, 1790, 1760, 1735, 1728, 1670; ORD (dioxan), $[M]_{352\ nm}$ − 4700° (min), $[M]_{312}$ + 4100°(min), $[M]_{240}$ − 6700°(min), $[M]_{206}$ + 34900° (max); CD (dioxan) λ_{max} 344 nm (Δε − 2.35), 340 (−2.33), 333 (−2.49), 266 (−0.04), 228 (−5.87) (strongly positive at shorter wavelengths).

Dehydration of physalin E (**2**) *to anhydrophysalin E* (**5**). Physalin E (50 mg) in glacial HOAc (10 ml) was refluxed on an oil bath for 12 hr. The product was worked up and the residue crystallized from MeOH as shining needles (45 mg), mp 280–281°; (Found: C, 63.71; H, 5.85; M^+ 526. $C_{28}H_{30}O_{10}$ requires C, 63.87; H, 5.74%); λ_{max}^{EtOH} 228 nm (ε 6500); ν_{max}^{KBr} cm^{-1}: 3400, 1780, 1767, 1730, 1665.

Dehydrophysalin B (**6**) (*carried out by K. Sambi Reddy*). Anhydrophysalin E (**5**) (35 mg) was triturated with two drops of conc H_2SO_4, and after 10 min at room temp. the mixture was diluted with ice-cold H_2O (50 ml). The resulting yellow solid was extracted with $CHCl_3$, and after the usual workup, it crystallized from $CHCl_3$ as bright yellow glistening plates (10 mg), mp 230–231°, which were identified as dehydrophysalin B (**6**) [1] by IR and TLC. A similar treatment of physalin H and physalin E (10 mg) gave also **6** (5 mg).

Dehydration of physalin E to isoanhydrophysalin E (**7**). Physalin E (50 mg) in dry Py (1 ml) was treated with freshly distilled $POCl_3$ (4 drops) for 15 min on a steam bath. The product was worked up and the residue crystallized from MeOH as shining colourless needles (40 mg), mp 287–89°; (Found: C, 63.77, H, 5.71, $C_{28}H_{30}O_{10}$ requires: C, 63.87, H, 5.74%) λ_{max}^{EtOH} 232 nm (ε 8900); ν_{max}^{KBr} cm^{-1}: 3400, 1800, 1770, 1730, 1670.

Oxidation of physalin E to physalin E-7-one (**4**). Physalin E (70 mg) in aldehyde free Me_2CO (40 ml) was treated with Jones' reagent (1 g CrO_3 + 7 ml H_2O + 0.9 ml conc H_2SO_4, 8 drops) for 15 min. Excess reagent was destroyed using MeOH (1 ml). The mixture was concd, poured into cold H_2O and extracted with $CHCl_3$ (30 ml × 3). The combined $CHCl_3$ extracts were washed with 1 N HCl followed by H_2O, dried over dry Mg_2SO_4 and evapd. The residue crystallized from EtOAc as shining needles, mp > 300° (50 mg); (Found: C, 61.92; H, 5.43; M^+ 542. $C_{28}H_{30}O_{11}$ requires: C, 62.06; H, 5.43%); λ_{max}^{EtOH} 228 nm (ε 7200); ν_{max}^{KBr} cm^{-1}: 3400, 1780, 1767, 1730, 1713, 1665.

Hydrogenation of physalin E to dihydrophysalin E (**8**) *and tetrahydrophysalin E* (**9**). Physalin E (50 mg) was dissolved in absolute EtOH (70 ml) and was hydrogenated in the presence of Pd/C (50 mg). After 30 min, the reaction was stopped and the product worked up. No pure compound crystallized out either from Me_2CO or MeOH or other solvents. The crude mixture was adsorbed on Si gel G (0.1 g) and after drying applied on the top of a narrow column of Si gel G (15 g). Elution with C_6H_6–EtOAc (85:15) (500 ml) gave dihydrophysalin E (**8**), crystallized from MeOH into colourless needles (20 mg), mp 274–276°; (Found: C, 61.45; H, 6.42; M^+ 546. $C_{28}H_{34}O_{11}$ requires: C, 61.60; H, 6.27%); no absorption above 210 nm; ν_{max}^{KBr} cm^{-1}: 3400, 1790, 1770, 1734, 1693. Elution with C_6H_6–EtOAc (1:1, 500 ml) gave tetrahydrophysalin E (**9**), which crystallized from MeOH as colourless shining micro-needles (20 mg) mp 288–289°; (Found: C, 61.27; H, 6.80, M^+ 548. $C_{28}H_{36}O_{11}$: requires C, 61.36; H, 6.62%); ν_{max}^{KBr} cm^{-1}: 3380, 1790, 1760, 1740, 1690; no absorption above 210 nm.

Physalin H (**3**). Physalin H crystallized from MeOH as colourless buttons, mp 238–240°; (Found: C, 63.05; H, 5.92; M^+ 526; $C_{28}H_{30}O_{10}$ requires: C, 63.87; H, 5.74%), λ_{max}^{EtOH} 230 nm (ε 6500); ν_{max}^{KBr} cm^{-1}: 3400, 1795, 1760, 1730, 1670. The acetate (Ac_2O–Py at 100° for 12 hr) of physalin H crystallized from MeOH as colourless needles, mp 243–244°; (Found: C, 63.0; H, 5.80. $C_{30}H_{32}O_{11}$ requires C, 63.42; H, 5.68%; λ_{max}^{EtOH} 229 nm (ε 7200) ν_{max}^{KBr} cm^{-1}: 3400, 1790, 1760, 1740, 1720, 1665.

Oxidation of physalin H to Physalin B-7-one (**12**). Physalin H (50 mg) in aldehyde-free Me_2CO (20 ml) was heated with Jones' reagent (4 drops) for 15 min. After usual work up, physalin B-7-one (**12**) was crystallized from MeOH as colourless needles (25 mg), mp 282–284°; (Found: C, 63.5; H, 5.5. $C_{28}H_{28}O_{10}$ requires: C, 64.12; H, 5.38%) λ_{max}^{EtOH} 232 nm (ε 13200) ν_{max}^{KBr} cm^{-1}: 3400, 1780, 1757, 1730, 1660.

Dehydration of physalin E-7-one to Physalin B-7-one (**12**). Physalin E-7-one (70 mg) in glacial HOAc (20 ml) and a drop of conc H_2SO_4 were refluxed on an oil bath for 22 h. The residue obtained after removing solvent *in vacuo* was worked up and crystallized from MeOH as colourless needles (25 mg), mp 280–282°, found identical with physalin B-7-one obtained above.

Physalin I. Physalin I crystallized from MeOH as short needles, mp 305–6°; $[\alpha]_D$ + 12° (*c*, 0.5, CH_3COCH_3); (Found: C, 61.23; H, 5.99; M^+ 558. $C_{29}H_{34}O_{11}$ requires: C, 61.82; H, 5.92%): λ_{max}^{EtOH} 226–228 nm (ε 7200); ν_{max}^{Nujol} cm^{-1}: 3400, 2820, 1790, 1772, 1745, 1662, 1642; NMR (δ Values): 5.91(*dd*, *J* = 10, 3 Hz, C-2); 6.65 (*dm*, *J* = 10, C-3), 3.84 (*m*, CHOH), 4.58 (*m*, C-22), 4.29 (*dd*, *J* = 14, 4, *C*-26-H_a), 3.60 (*d*, *J* = 13, C-26-H_b), 5.70 (*s*, C-13-O*H*), 4.94 (*d*, *J* = 4, C-3-O*H*), 2.94 (*s*, OCH_3); 1.94 (*s*) 1.10 (*s*) (three methyls).

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