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# (E)-24-Ethylidene-cholest-7-en-3ß-ol and Other Sterols in Asteroids

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## Abstract

The sterols of the asteroids, *Laiaster leachii* and *Coscinasterias acutispina*, were examined by gas-liquid chromatography (GLC), column chromatography, and infrared absorption (IR), mass and nuclear magnetic resonance (NMR) spectrometries.

The sterol composition of two asteroids was fairly complex, however 7-cholestenol was predominant in these animals. Cholestanol, 7, 22-cholestadienol, stellasterol and 7, 24(28)-ergostadienol were isolated from the asteroids as a pure sterol by column chromatography on a silver nitrate-impregnated silicic acid.

Moreover, a small amount of sterol isolated from *L. leachii* was identified as (E)-24-ethylidene-cholest-7-en-3$\beta$-ol by IR, mass and NMR spectrometries and GLC.

It has been reported by many workers that the asteroids contain a variety of $\Delta^7$-sterols ; stellasterol [1, 17], stellastenol [1, 14], 7-cholestenol [2-4, 8, 11, 14], $\alpha$-spinasterol [5-8, 14], $\alpha$-spinastenol [8, 9, 14], 7, 24(28)-ergostadienol [10, 17], 7, 22-cholestadienol [11, 17], 7, 22-ergostadienol [11, 14], 7-ergostenol [11], and acansterol [11, 12]. The most reasonable conclusion of the sterols from asteroids has been shown to be composed of only $\Delta^7$-sterols but not $\Delta^5$-sterols [11, 13]. In addition to this, the occurrence of cholestanol [15, 16, 18] and cholesterol [16, 18] besides $\Delta^7$-sterols, recently has been recognized in asteroids. Moreover, $C_{26}$ sterol (24-nor-7, 22-cholestadienol) [15, 16, 17], $C_{29}$ sterol (24-ethylidene-7-cholestenol) [11, 16, 18], and $C_{30}$ sterols (24-propyl-7-cholestenol and 24-propylidene-7-cholestenol) [18], including new sterols, have been found by a combined gas-liquid chromatography (GLC)-mass spectrometry. Therefore, a detailed examination of asteroid sterols by using efficient analytical techniques seems to be necessary and attractive.

This study presents some knowledges of the sterol mixtures in two species of the Asteroidae, *Laiaster leachii* and *Coscinasterias acutispina*.

## Materials and Methods

**Asteroids.** The asteroids, *L. leachii* and *C. acutispina* were harvested at the neighborhood of Sakurajima in Kagoshima in August, 1972, and dried under sun-light for several days.

**Isolation of sterols.** From the asteroids, the lipids were extracted by refluxing the dried materials with acetone, and saponified with 10% of ethanolic potassium hydroxide at 80°C for 2 hours. The sterols were isolated from the unsaponifiable matters by using column chromatography on alumina with hexane-benzene [19]. The sterol fraction was purified by crystallization from methanol.

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**Separation of individual sterols.** For the purpose of separating the individual sterols, the sterol mixtures were acetylated with acetic anhydride–dry pyridine (1:1, v/v) and chromatographed on a silver nitrate–impregnated silicic acid ($AgNO_3$–$SiO_2$) with hexane–benzene [20].

**Spectral analyses.** Infrared absorption (IR) spectrum was measured in chloroform with a Shimadzu IR-27G. Mass spectrum was obtained with a Hitachi RM-50GC (chamber voltage 70eV). Nuclear magnetic resonance (NMR) spectrum was measured with a Japan Electron Optics Spectrometer modified to 100-MHz instrument. The signal was analyzed by using tetramethylsilane as an internal standard to determine chemical shifts.

**Other analyses.** Analytical GLC was made on a Shimadzu model GC-3AF chromatograph unit, and the GLC systems used were 1.5% OV-17 on Shimalite W with the coil stainless column (3m×4mm I.D.) and 1.5% QF-1 on Chromosorb W with the glass column (2m×4mm I.D.). Melting point (MP) was determined with a Yanagimoto Melting Point Apparatus and uncorrected. Liebermann–Burchard reaction was performed as a classical check of the position of the double bonds in the sterol nucleus.

## Results

**Sterols of *L. leachii*.** In GLC on 1.5% OV-17, the sterol fraction revealed the presence of at least eleven different compounds as shown in Fig. 1. The steryl acetate mixture could be separated by column chromatography on $AgNO_3$–$SiO_2$ into sixteen components, as shown in Fig. 2 and Table 1. In this chromatography, the six sterols (A-1, 7, 9, 14, 15 and 16) were obtained as highly enriched acetates, and then recrystallized several times from methanol.

The sterol A-1, non-colored with Liebermann–Burchard reaction, corresponded to the peak P-2 and was identified as cholestanol (cholestan-3β-ol) from MP (found acetate 115–118°C; reference 113°C [21]) and relative retention time (RRT) to cholesterol; 1.01 on 1.5% OV-17 and 1.09 on 1.5% QF-1. Further, the identification was confirmed from the relationship

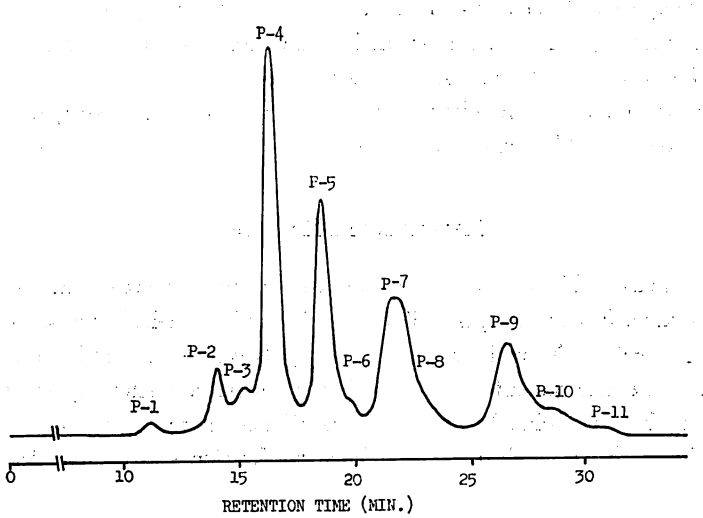


Fig. 1. GLC on 1.5% OV-17 of the sterols isolated from the asteroid, *L. leachii*.

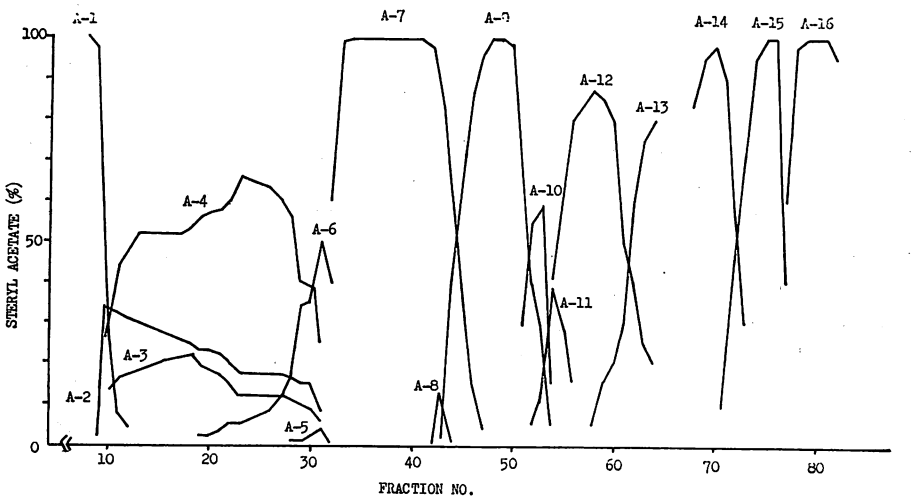


Fig. 2. Column chromatography on a silver nitrate-impregnated silicic acid of the steryl acetates isolated from the asteroid, *L. leachii*.

The steryl acetates detected are as follows : A-1, cholestan-3$\beta$-ol ; A-2, 24R- or 24S-ethyl-cholest-7-en-3$\beta$-ol ; A-3, 24R- or 24S-methyl-cholest-7-en-3$\beta$-ol ; A-4, cholest-7-en-3$\beta$-ol; A-5, cholest-5-en-3$\beta$-ol; A-6, 24R- or 24S-ethyl-cholesta-7, 22-dien-3$\beta$-ol; A-7, 24 R-methyl-cholesta-7, 22-dien-3$\beta$-ol ; A-8, unknown ; A-9, cholesta-7, 22-dien-3$\beta$-ol; A-10, unknown ; A-11, 24-norcholesta-7, 22-dien-3$\beta$-ol; A-12, unknown; A-13, unknown ; A-14, (E)-24-ethylidene-cholest-7-en-3$\beta$-ol ; A-15, 24-methylene-cholest-7-en-3$\beta$-ol ; A-16, unknown

Table 1. The sterol compositions of the asteroid, *L. leachii*, determined by GLC on 1.5% OV-17.

| Peaks | %* | Components | RRT** | Identified as |
|---|---|---|---|---|
| P-1 | 1.0 | A-11 | 0.78 | 24-Norcholesta-7, 22-dien-3$\beta$-ol |
| P-2 | 4.9 | A- 5 | 1.00 | Cholest-5-en-3$\beta$-ol |
| | | A- 1 | 1.01 | Cholestan-3$\beta$-ol |
| P-3 | 3.3 | A- 9 | 1.09 | Cholesta-7, 22-dien-3$\beta$-ol |
| P-4 | 30.2 | A- 4 | 1.16 | Cholest-7-en-3$\beta$-ol |
| P-5 | 20.1 | A- 7 | 1.32 | 24R-Methyl-cholesta-7, 22-dien-3$\beta$-ol |
| P-6 | 2.3 | A-16 | 1.45 | Unknown, triene sterol |
| P-7 | 18.6 | A- 3 | 1.53 | 24R- or 24S-Methyl-cholest-7-en-3$\beta$-ol |
| | | A-15 | 1.57 | 24-Methylene-cholest-7-en-3$\beta$-ol |
| P-8 | 2.2 | A- 6 | 1.66 | 24R- or 24S-Ethyl-cholesta-7, 22-dien-3$\beta$-ol |
| P-9 | 14.1 | A-10 | 1.84 | Unknown |
| | | A- 2 | 1.88 | 24R- or 24S-Ethyl-cholest-7-en-3$\beta$-ol |
| | | A-12 | 1.95 | Unknown |
| | | A-14 | 1.99 | (E)-24-Ethylidene-cholest-7-en-3$\beta$-ol |
| P-10 | 2.8 | A-13 | 2.05 | Unknown |
| P-11 | 0.5 | A- 8 | 2.20 | Unknown |

* Calculated by peak area.

** Relative retention time to cholesterol.

between the chemical structure of steryl acetates and the mobility on $AgNO_3$-$SiO_2$ column chromatography.

The sterols A-7 and A-9 corresponded to the peaks P-5 and P-3, respectively. The two sterols were assumed to be diene sterols with a double bond in side chain at C-22 besides in ring B, by the mobility on $AgNO_3$-$SiO_2$ column chromatography. Moreover, the sterols A-7 and A-9 were identified as stellasterol (24R-methyl-cholesta-7, 22-dien-3$\beta$-ol) and 7, 22-cholesta dienol (cholesta-7, 22-dien-3$\beta$-ol), respectively, by mass, NMR and IR spectra, and MP. Sterol A-7 : mass (acetate) (Fig. 3) several highly intense peaks at m/e 440 ($M^+$, $\Delta^7$), 342 ($M^+ - C_7H_{14}$, $\Delta^{22}$), 312 ($M^+$ - side chain+2H), and 255 ($M^+$ - side chain+60); NMR (free) (Fig. 4) $\tau$4. 84-4. 94 (3H, multiplet, $\Delta^7$ and $\Delta^{22}$), $\tau$6. 30-6. 60 (1H, multiplet, OH), $\tau$9. 35 (3H, singlet, C-18 methyl), $\tau$9. 20 (3H, singlet, C-19 methyl), $\tau$9. 00 (3H, doublet, J = 7Hz, C-21 methyl), $\tau$9. 14 (6H, doublet, J = 6Hz, C-26 and C-27 methyl) and $\tau$ 9. 08 (3H, doublet, J = 6Hz, C-28 methyl) ; IR (free) (Fig. 5) 3550-3400cm$^{-1}$ (OH), 970cm$^{-1}$ (trans $\Delta^{22}$), and 830cm$^{-1}$ ($\Delta^7$). The above spectral data

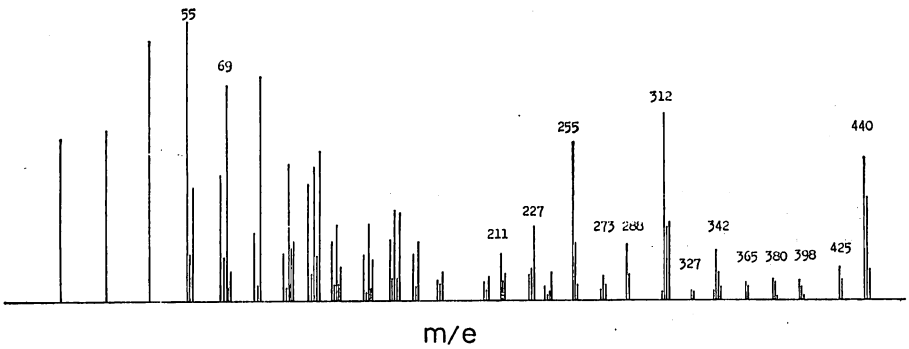


Fig. 3. Mass spectrum of A7, 24R-methyl-cholesta-7, 22-dien-3$\beta$-ol (acetate) isolated from the asteroid, *L. leachii*.

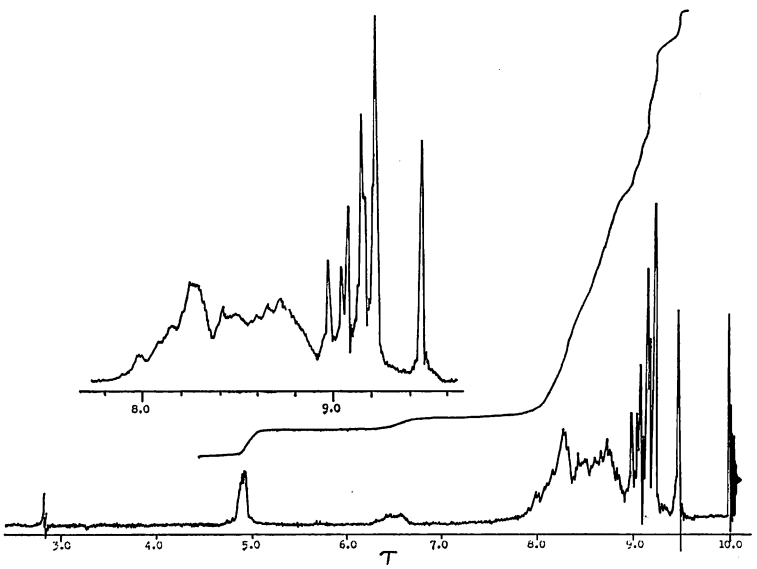


Fig. 4. NMR spectrum of A-7, 24R-methyl-cholesta-7, 22-dien-3$\beta$-ol.

suggested that the sterol A-7 is 24ξ-methyl-cholesta-7, 22-dien-3β-ol. The MP of the sterol (acetate 163-166°C, free 154-157°C) was fairly low and clearly distinguishable from that of 24S-methyl-cholesta-7, 22-dien-3β-ol (acetate 181°C, free 173°C), therefore the sterol was identified as 24R-methyl-cholesta-7, 22-dien-3β-ol. Sterol A-9: mass (acetate) m/e 426 ($M^+$, $\Delta^7$), 342 ($M^+-C_6H_{12}$, $\Delta^{22}$), 313 ($M^+$—side chain+2H, diene), 255 ($M^+$—side chain+60) and 228 ($M^+$—side chain+60+27); IR (free) (Fig. 5), 3550-3400cm$^{-1}$ (OH), 970cm$^{-1}$ ($\Delta^{22}$), and 830cm$^{-1}$ ($\Delta^7$); MP, found acetate 136-138°C, free 115-118°C.

The peak corresponding to the sterol A-14 was hid by the peaks P-9 and P-10 in GLC on 1.5% OV-17. The mass, NMR and IR spectra of the acetate of A-14 are shown in Figs. 6, 7, and 8, respectively. The mass spectrum yielded significant peaks at m/e 313 ($M^+$—side chain+2H, double bond at side chain, relative intensity 100%), 356 ($M^+-C_7H_{14}$, $\Delta^{24(28)}$, 64%), 255 ($M^+$—side chain+60, 25%) and 454 ($M^+$, $\Delta^7$ $C_{29}$-diene sterol, 29%) in close resemblance to (E)-24-ethylidene sterol [22]. From these relative intensity data, the sterol may be considered as

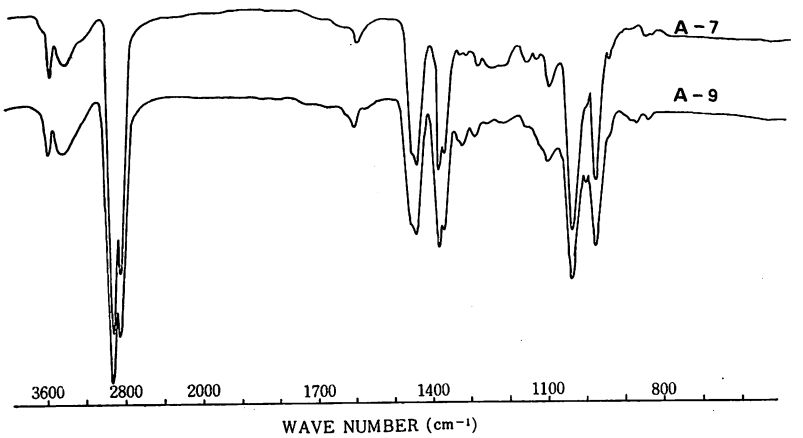


Fig. 5. IR spectra of A-7, 24R-methyl-cholesta-7, 22-dien-3β-ol and A-9, cholesta-7, 22-dien-3β-ol.

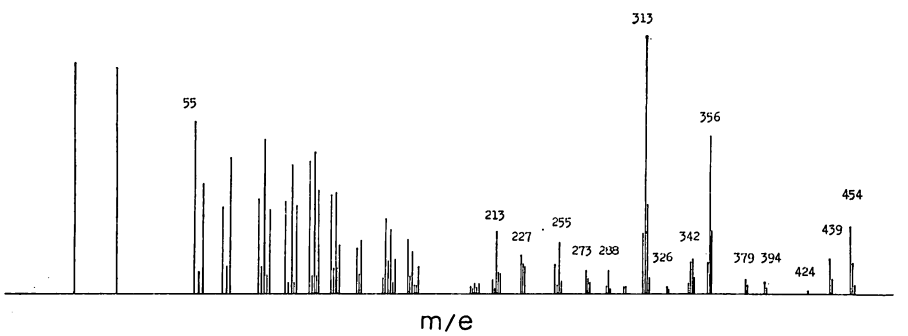


Fig. 6. Mass spectrum of A-14, (E)-24-ethylidene-cholest-7-en-3β-ol (acetate) isolated from the asteroid, *L. leachii*.

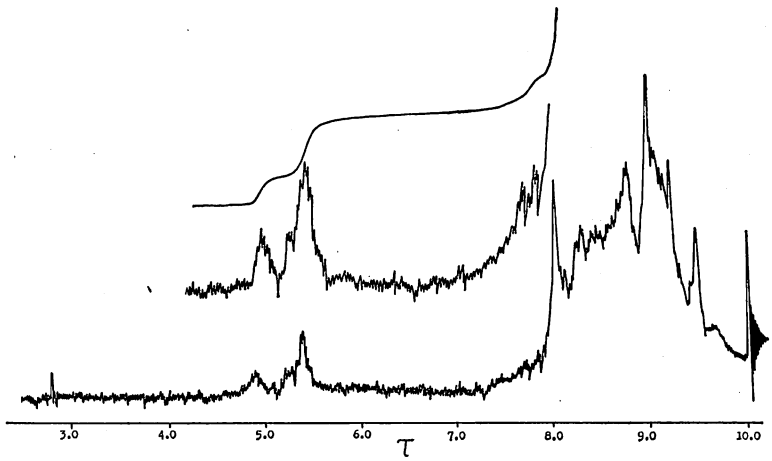


Fig. 7. NMR spectrum of A-14, (E)-24-ethylidene-cholest-7-en-3$\beta$-ol (acetate).

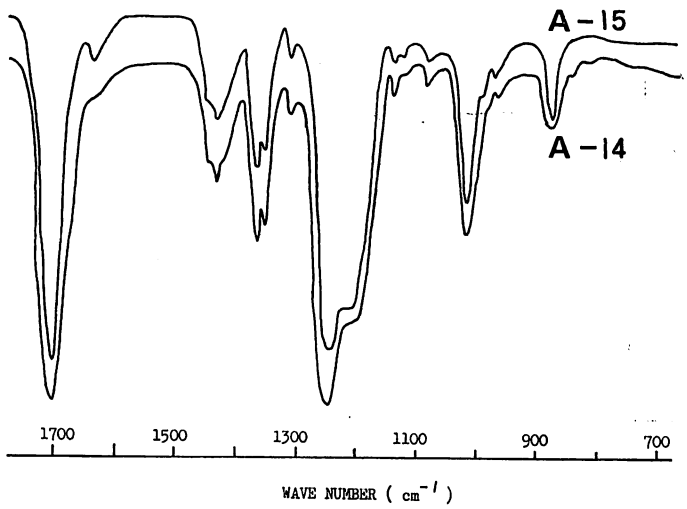


Fig. 8. IR spectra of A-14, (E)-24-ethylidene-cholest-7-en-3$\beta$-ol (acetate) and A-15, 24-methylene-cholest-7-en-3$\beta$-ol (acetate).

E-isomer rather than Z-isomer of 24-ethylidene sterol. The NMR spectrum showed 3$\alpha$-proton and C-28 methin proton at $\tau$ 5.38, and C-7 olefin proton at $\tau$ 4.92. The spectrum also contained singlets (each 3H) at $\tau$ 8.02 (acetyl), $\tau$ 9.47 (C-18 methyl) and $\tau$ 9.20 (C-19 methyl), and doublets at $\tau$ 8.97 (6H, isopropyl), $\tau$ 9.12 (3H, C-21 methyl) and $\tau$ 8.41 (3H, C-29 methyl). The absence of signal at $\tau$ 7.20 eliminated the possibility of Z-isomer of 24-ethylidene sterol, and the presence of a heptet centered at $\tau$ 7.80 characteristic of E-isomer of 24-ethylidene sterol, was observed [23]. In the IR spectrum, a strong region at 880cm$^{-1}$ and a weakly bond at 1640cm$^{-1}$ indicated that the acetate possesses an ethylidene group instead of terminal methylene grou pshowing a fairly strong bond at 1640cm$^{-1}$ besides a strong bond at 880cm$^{-1}$. Moreover, RRT of this sterol (1.68) to 7-cholestenol was equal to that of fucosterol (1.68) (E-isomer) to cholesterol but not 28-isofucosterol (1.78) (Z-isomer) in GLC on 1.5% OV-17. On the basis of the

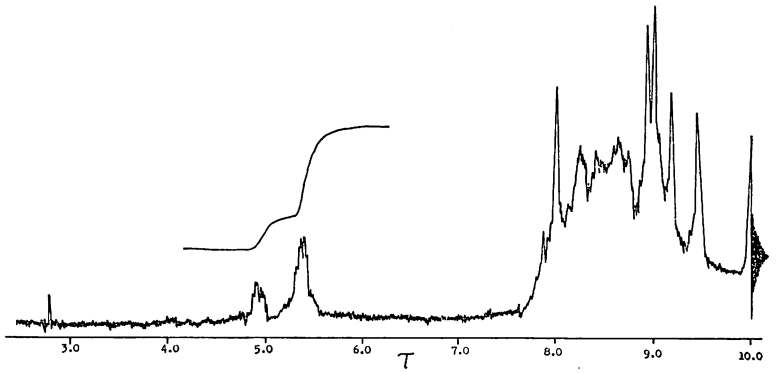


Fig. 9. NMR spectrum of A-15, 24-methylene-cholest-7-en-3$\beta$-ol (acetate).

above data, the sterol A-14 was identified as (E)-24-ethylidene-cholest-7-en-3$\beta$-ol.

The sterol A-15 was identified as 7, 24(28)-ergostadienol (24-methylene-cholest-7-en-3$\beta$-ol) from the following data: Mass (acetate) m/e 440 ($M^+$, molecular weight, $\Delta^7$), 356 ($M^+-C_8H_{12}$, fucosterol type), 313 ($M^+$—side chain+2H, diene sterol), 255 ($M^+$—side chain+60), and 213 ($M^+$—side chain+60+42); NMR (acetate) (Fig. 9) $\tau$ 5.24—$\tau$ 5.60 (1H, multiplet, 3$\alpha$-proton), $\tau$ 5.38 (2H, doublet, terminal methylene), $\tau$ 4.93 (1H, multiplet, C-7 proton), $\tau$ 8.02 (3H, singlet, acetyl), $\tau$ 9.47(3H, singlet, C-18 methyl), $\tau$ 9.20 (3H, singlet, C-19 methyl), $\tau$ 8.96 (3H, doublet, J = 7Hz, C-21 methyl), $\tau$ 9.03 (6H, doublet, J = 6Hz, C-26 and C-27 isopropyl); IR (acetate) (Fig. 8) 1640cm$^{-1}$, 890cm$^{-1}$ (terminal methylene), 1730cm$^{-1}$ (C=O); MP (acetate) found 137-140°C, reference 138°C.

Elucidation of the chemical structure of the sterol A-16 ($C_{28}H_{46}O$), probably $C_{28}$ triene sterol, will be performed in this laboratory.

The separation of other acetates besides the above six acetates was unsufficient under the conditions of $AgNO_3$-$SiO_2$ column chromatography adopted in this study. A part of the chemical structures of these steryl acetates could be confirmed tentatively from the relationship between the eluting order on $AgNO_3$-$SiO_2$ column chromatography and analytical GLC. With this technique, the sterols A-2A, -3, A-4, and A-5 were presumed to be $\alpha$-spinastenol or chondrillastenol (24R- or 24S-ethyl-cholest-7-en-3$\beta$-ol), stellastenol or 7-ergostenol (24R- or 24S-methyl-cholest-7-en-3$\beta$-ol), 7-cholestenol (cholest-7-en-3$\beta$-ol) and cholesterol, respectively, which are all monoene sterols.

The sterols A-6 and A-11 may be diene sterols with double bonds at C-7 and C-22. The RRTs of sterols A-6 (1.42) and A-11 (0.66) to 7-cholestenol were equal to those of stigmasterol and 22-*trans*-24-norcholesta-5, 22-dien-3$\beta$-ol to cholesterol in GLC on 1.5% OV-17, respectively. Therefore, the sterols A-6 and A-11 were assumed to be $\alpha$-spinasterol or chondrillasterol (24R- or 24S-ethyl-cholesta-7, 22-dien-3$\beta$-ol) and 22-*trans*-24-norcholesta-7, 22-dien-3$\beta$-ol, respectively.

Other sterols A-8, A-10, A-12 and A-13 were not identified, however the sterols were assumed to have some double bonds at side chain, considering their eluting order on $AgNO_3$-$SiO_2$ column chromatography.

**Sterols of *C. acutispina*.** The identification of the sterols of the asteroid, *C. acutispina* was performed by comparing the RRT on GLC and the mobility on $AgNO_3$-$SiO_2$ column

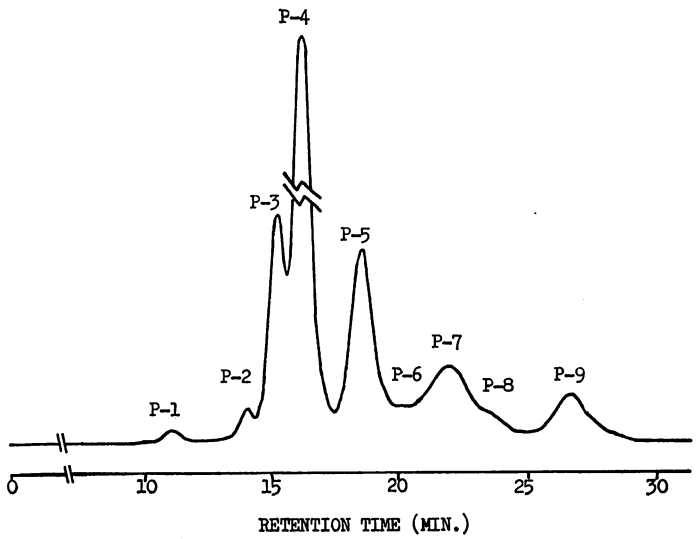


Fig. 10. GLC on 1.5% OV-17 of the sterols isolated from the asteroid, *C. acutispina*.

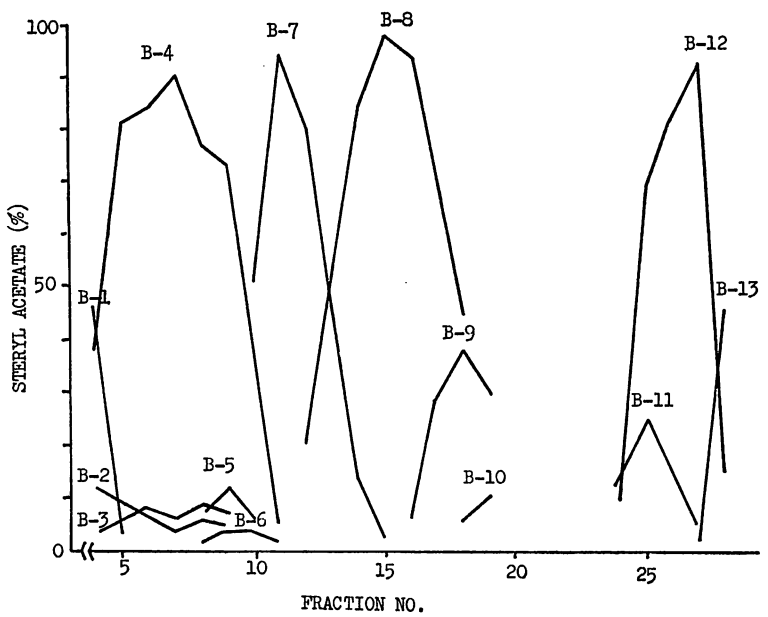


Fig. 11. Column chromatography on a silver nitrate-impregnated silicic acid of the steryl acetates isolated from the asteroid, *C. acutispina*.

The steryl acetates detected are as follows : B-1, cholestan-3$\beta$-ol ; B-2, 24R- or 24S-ethyl-cholest-7-en-3$\beta$-ol ; B-3, 24R- or 24S-methyl-cholest-7-en-3$\beta$-ol; B-4, cholest-7-en-3$\beta$-ol; B-5, cholest-5-en-3$\beta$-ol; B-6, 24R- or 24S-ethyl-cholesta-7, 22-dien-3$\beta$-ol ; B-7, 24R-methyl-cholesta-7, 22-dien-3$\beta$-ol ; B-8, cholesta-7, 22-dien-3$\beta$-ol ; B-9, 24-norcholesta-7, 22-dien-3$\beta$-ol; B-10, unknown; B-11, unknown; B-12, 24-methylene-cholest-7-en-3$\beta$-ol; B-13, unknown

chromatography with those of the sterols of the preceding asteroid, *L. leachii*.

The sterols isolated from the asteroid, *C. acutispina* were subjected to GLC on 1.5% OV-17, and were found to be composed of nine components as shown in Fig. 10. The steryl acetates were chromatographed on $AgNO_3$–$SiO_2$ in order to isolate the individual sterols and to know the eluting position of those. The results are as shown in Fig. 11 and Table 2. From these analytical procedures, the peak P-2 was composed of four components, B-1, B-5, B-10 and B-11. Similarly the peak P-7 was composed of two components, B-3 and B-12. Other peaks,

Table 2. The sterol compositions of the asteroid, *C. acutispina*, determined by GLC on 1.5% OV-17.

| Peaks | %* | Components | RRT** | Identified as |
|---|---|---|---|---|
| P-1 | 1.0 | B- 9 | 0.78 | 24-Norcholesta-7, 22-dien-3β-ol |
| P-2 | 1.6 | B-10 | 0.94 | Unknown |
| | | B-11 | 0.99 | Unknown |
| | | B- 5 | 1.00 | Cholest-5-en-3β-ol |
| | | B- 1 | 1.01 | Cholestan-3β-ol |
| P-3 | 14.2 | B- 8 | 1.09 | Cholesta-7, 22-dien-3β-ol |
| P-4 | 55.9 | B- 4 | 1.16 | Cholest-7-en-3β-ol |
| P-5 | 13.1 | B- 7 | 1.32 | 24 R-Methyl-cholesta-7, 22-dien-3β-ol |
| P-6 | 0.6 | B-13 | 1.45 | Unknown |
| P-7 | 6.3 | B- 3 | 1.53 | 24 R- or 24 S-Methyl-cholest-7-en-3β-ol |
| | | B-12 | 1.57 | 24-Methylene-cholest-7-en-3β-ol |
| P-8 | 2.7 | B- 6 | 1.66 | 24 R- or 24 S-Ethyl-cholesta-7, 22-dien-3β-ol |
| P-9 | 4.8 | B- 2 | 1.88 | 24 R- or 24 S-Ethyl-cholest-7-en-3β-ol |

* Calculated by peak area.

** Relative retention time to cholesterol.

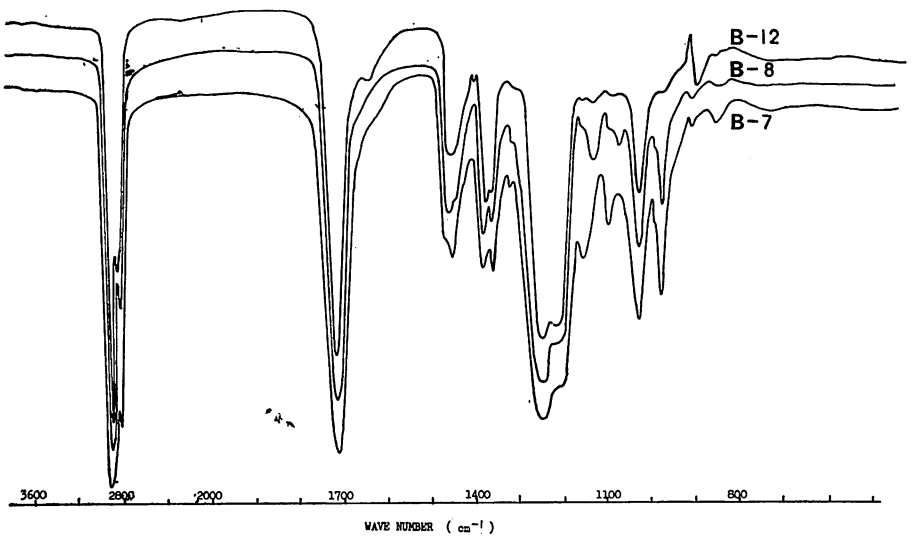


Fig. 12. IR spectra of B-7, 24 R-methyl-cholesta-7, 22-dien-3β-ol (acetate), B-8, cholesta-7, 22-dien-3β-ol (acetate), and B-12, 24-methylene-cholest-7-en-3β-ol (acetate).

P-1, P-3, P-4, P-5, P-6, P-8 and P-9 corresponded to the components, B-9, B-8, B-4, B-7, B-13, B-6 and B-2, respectively. As the result of the column chromatography, the four sterols (B-4, B-7, B-8 and B-12) were obtained as highly enriched acetates, and then subjected to thin-layer chromatography on a silver nitrate-impregnated Kieselgel $HF_{254+366}$ in order to purify each of them.

The sterol B-4 was identified as 7-cholestenol, on the basis of the MP (found acetate 116-118°C; reference 119°C) and RRT in GLC.

IR spectra of the acetates of B-7, B-8 and B-12 are shown in Fig. 12. The sterols B-7, B-8 and B-12 were identified as stellasterol, 7, 22-cholestadienol and 7, 24(28)-ergostadienol, respectively, basing on the eluting position in $AgNO_3$-$SiO_2$ column chromatography and on the following data. B-7 (acetate): MP found 162-166°C, reference 181°C (24S-methyl-cholesta-7, 22-dien-3$\beta$-ol); IR 970cm$^{-1}$ ($\Delta^{22}$) and 830cm$^{-1}$ ($\Delta^{7}$). B-8 (acetate): MP found 135-139°C; IR 970cm$^{-1}$ ($\Delta^{22}$) and 830cm$^{-1}$ ($\Delta^{7}$). B-12 (acetate): MP found 137-139°C, reference 138°C; IR 1640cm$^{-1}$, 890cm$^{-1}$ (terminal methylene) and 830cm$^{-1}$ ($\Delta^{7}$).

Other sterols except the above four sterols were not sufficiently isolated on $AgNO_3$-$SiO_2$ column chromatography. The mobility on $AgNO_3$-$SiO_2$ column chromatography and RRT on analytical GLC of the components B-1, B-2, B-3, B-5, B-6, B-9 and B-13 agreed with that of the components A-1, A-2, A-3, A-5, A-6, A-11 and A-16 in *L. leachii*. Therefore sterols B-1, B-2, B-3, B-5, B-6 and B-9 were tentatively identified as cholestanol, $\alpha$-spinastenol or chondrillastenol, stellastenol or 7-ergostenol, cholesterol, $\alpha$-spinasterol or chondrillasterol and 24-norcholesta-7, 22-dien-3$\beta$-ol, respectively.

The sterols B-10, B-11 and B-13 were not identified in this study.

## Discussion

It has been succeeded to isolate each component from the steryl acetates of two asteroids by using column chromatography on $AgNO_3$-$SiO_2$ modified by TESHIMA *et al.* [20]. As the result, eleven sterols were identified from the asteroids, *L. leachii* and *C. acutispina*. The composition of these sterols was essentially similar to those reported by MATSUNO *et al.*[14), 15)], KOBAYASHI *et al.*[16, 17)], and GROSSERT *et al.*[18)]. The presence of small amounts of cholestanol and $\Delta^5$-sterol besides $\Delta^7$-sterols was also suggested in two asteroids. Since the marine invertebrates such as mollusks are conceivable to be utilized as a diet for asteroids, it seems reasonable to consider that the $\Delta^5$-sterols present in asteroids may be derived from the exogenous sources. FAGERLUND and IDLER have demonstrated the conversion of dietary cholesterol to 7-cholestenol in an asteroid[24)]. Recently, SMITH *et al.* have reported that the asteroid can convert injected cholesterol into cholestanol, and that cholestanol can be fruther metabolized to give 7-cholestenol [25, 26)]. In the present study, (E)-24-ethylidene-cholest-7-en-3$\beta$-ol was identified in *L. leachii*. In animals, it is generally accepted that 24-ethylidene sterol is a Z-isomer. For example, 28-isofucosterol ((Z)-24-ethylidene-cholest-5-en-3$\beta$-ol) has been isolated from the mollusks [20, 27, 28)]. Accordingly, by the occurrence of E-isomer of 24-ethylidene sterol in asteroid, some of the information may be given for the comparative biochemistry of marine invertebrates.

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## References

1) BERGMANN, W. and H. A. STANSBURY (1944): *J. Org. Chem.*, **9**, 281–289.
2) TOYAMA, Y. and T. TAKAGI (1954): *Bull. Chem. Soc. Japan*, **27**, 39–41.
3) TOYAMA, Y. and T. TAKAGI (1954): *Bull. Chem. Soc. Japan*, **27**, 421–423.
4) MATSUMOTO, T. and T. WAINAI (1954): *J. Chem. Soc. Japan*, **75**, 756–758.
5) MATSUMOTO, T. and T. WAINAI (1954): *J. Chem. Soc. Japan*, **75**, 1147–1149.
6) MATSUMOTO, T. and T. WAINAI (1955): *Bull. Chem. Soc. Japan*, **28**, 447.
7) TOYAMA, Y. and T. TAKAGI (1955): *Bull. Chem. Soc. Japan*, **28**, 469–473.
8) TOYAMA, Y. and T. TAKAGI (1956): *Bull. Chem. Soc. Japan*, **29**, 317–319.
9) MATSUMOTO, T., T. WAINAI, and C. HIRAI (1956): *J. Chem. Soc. Japan*, **77**, 531–533.
10) FAGERLUND, U. H. M. and D. R. IDLER (1957): *J. Am. Chem. Soc.*, **81**, 401–405.
11) GUPTA, K. C. and P. J. SCHEUER (1968): *Tetrahedron*, **24**, 5831–5837.
12) SHEIKH, Y. M., C. DJERASSI, and B. M. TURSCH (1971): *Chem. Comm.*, 217–218.
13) BERGMANN, W. (1962): In" Comparative Biochemistry" Vol. 3, p. 144. Ed. by FLORKIN, M. and H. S. MASON (Academic Press, New York and London).
14) MATSUNO, T., S. NAGATA, and K. MIZUTANI (1972): *Bull. Jap. Soc. Sci. Fish.*, **38**, 144–147.
15) MATSUNO, T., S. NAGATA, and K. HASHIMOTO (1972): *Bull. Jap. Soc. Sci. Fish.*, **38**, 1261–1265.
16) KOBAYASHI, M., R. TSURU, K. TODO, and H. MITSUHASHI (1972): *Tetrahedron Letters*, 2935–2938.
17) KOBAYASHI, M., R. TSURU, K. TODO, and H. MITSUHASHI (1973): *Tetrahedron*, **29**, 1193–1196.
18) GROSSERT, J. S., P. MATHIAPARANAM, G. D. HEBB, P. PRICE, and I. M. CAMPBELL (1973): *Experientia*, **29**. 258–259.
19) TESHIMA, S. and A. KANAZAWA (1971): *Bull. Jap. Soc. Sci. Fish.*, **37**. 63–67.
20) TESHIMA, S., A. KANAZAWA, and T. ANDO (1973): *Mem. Fac. Fish. Kagoshima Univ.*, **22**, 7～13
21) ERDMAN, T. R. and R. H. THOMSON (1972): *Tetrahedron*, **28**, 5163–5173.
22) BROOKS, C. J. W., B. A. KNIGHTS, W. SUCROW, and B. RADÜCHEL (1972): *Steroids*, **20**, 487–497.
23) BATES, R. B., A. D. BREWER, B. A. KNIGHTS, and J. W. ROWE (1968): *Tetrahedron Letters*, 6163–6167.
24) FAGERLUND, U. H. M. and D. R. IDLER (1960): *Can. J. Biochem. Physiol.*, **38**, 997–1002.
25) SMITH, A. G. and L. J. GOAD (1971): *Febs Lett.*, **12**, 233–235.
26) SMITH, A. G., R. GOODFELLOW, and L. J. GOAD (1972): *Biochem. J.*, **128**, 1371–1372.
27) YASUDA, S. (1970): *Yukagaku*,**19**, 1014–1018.
28) IDLER, D. R. and P. WISEMAN (1971).: *Comp. Biochem. Physiol.*, **38**A, 581–590.