

REVIEW**Triterpenoid Saponins from the Genus *Camellia***

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1. Introduction. – Triterpenoid saponins are an important class of natural products, which are distributed widely in plant kingdom [1–3]. The name ‘saponin’ comes from the phenomenon that these molecules form soap-like foams when shaken with water [4]. Triterpenoid saponins consist of one or more monosaccharide moieties combined with non-polar aglycones, termed sapogenin. According to their structures, sapogenins are further classified into two major sub-classes: tetra- and pentacyclic triterpenoid saponins [2][3]. In addition to the surface-active properties, triterpenoid saponins possess numerous pharmacological activities and have been applied widely in beverages, healthy foods, cosmetics, and pharmaceutical products [1–5].

The genus *Camellia* is composed of two subgenera, 14 sections, and ca. 119 species distributed only in the Eastern Asia [6]. *Camellia* species are very popular in China, Japan, India, and many other South-East Asian countries. Among them, *C. sinensis*, *C.*

sinensis var. *assamica*, *C. oleifera*, *C. reticulata*, *C. japonica*, and *C. sasanqua* are of significant economic value. For example, the leaves of *C. sinensis* and *C. sinensis* var. *assamica* are the raw materials for producing tea, one of the most popular beverages consumed in the world. The seeds of *C. oleifera* can be used to produce edible oil, while *C. reticulata*, *C. japonica*, and *C. sasanqua* are famous ornamental plants.

So far, the phytochemical investigations on *Camellia* species have been mainly focused on five species, and saponins, called tea saponins, were recognized as one of the major bioactive constituents accounting for more than 10% per dry weight of the *Camellia* material [7]. To date, only acylated pentacyclic triterpenoid saponins have been reported from the genus *Camellia*, and most of them are oleanane-type triterpenoid saponins. They are characterized by a sugar chain at C(3), and one or more acyl groups located at C(21), C(22), and C(28) of aglycones. Due to their physicochemical properties, tea saponins, which are a new source of saponins in China, have been used widely as an excellent native non-ion surfactant in industry and agriculture where they have been employed as detergent, vesicant, emulsifier, insecticide, and fungicide [8][9]. Recent studies on tea saponins have also focused on the pharmacological properties, *e.g.*, antimicrobial, antiviral, anti-inflammatory, antioxidant, gastroprotective, EtOH-absorption inhibitory, antihyperlipidemic and anti-allergic activities [10–25].

In this review, we compile the phytochemical progress and list all triterpenoid saponins isolated from the genus *Camellia*, also considering the biological activities of isolated individual saponins.

2. Acylated Triterpenoid Saponins. – Tea saponins were first isolated from the tea-seed cake in 1931 as mixtures [26]. In 1952, *Ishidate* and *Ueda* obtained a pure saponin in crystalline form. It contained glucuronic acid, galactose, arabinose, xylose, and angelic acid, but its full structure was not established [27]. From the 1970s, a series of studies on separation, characterization, and utilization of saponins from the plants of genus *Camellia* were conducted in major tea-producing countries. Several extracting methods and products had been developed during this period. To date, 82 acylated saponin constituents, **1–82**, were reported from the genus *Camellia*. Of them, 60 saponins, **1–60**, were isolated from different plant parts of *C. sinensis*, whereas 16 saponins, *i.e.*, **1**, **2**, **29–34**, **36**, and **60–66**, were identified in *C. sinensis* var. *assamica*. Studies on the seeds, leaves, and flower buds of *C. japonica* and *C. oleifera* led to identification of twelve, *i.e.*, **36**, **37**, and **67–76**, and seven, *i.e.*, **37** and **77–82**, saponins, respectively. Only one saponin, **37**, was reported from the seeds of *C. sasanqua*. The structures of saponins **1–82** are shown in *Figs. 1–4*, according to the studied plant parts of *Camellia* plants, and their names and the corresponding plant sources are compiled in the *Table*. Among them, saponin **35** existed in all the seeds, leaves, and flower buds, while **38** and **39** were found in both seeds and leaves of *C. sinensis*. In addition, saponin **60** was found in the flower buds of *C. sinensis* and in the seeds of *C. sinensis* var. *assamica*.

2.1. From *C. sinensis*. *C. sinensis* is mainly cultivated in the central regions of China, and Japan. Its leaves are mainly used as the raw materials for producing green tea and oolong tea. The benefits of tea drinking for mental as well as physical health have been discussed for thousands of years in China and many other countries where the tea is

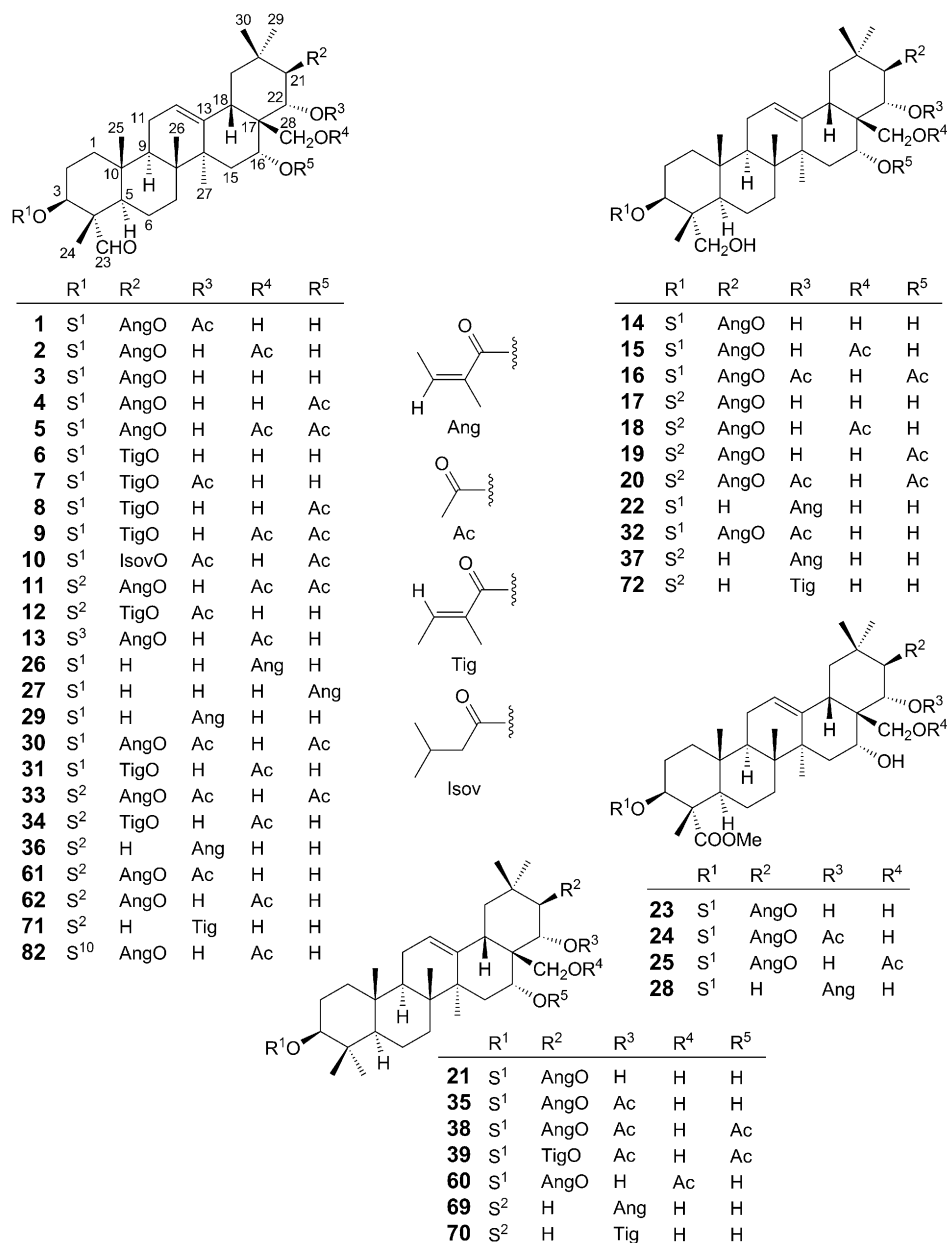


Fig. 1. Saponins isolated from the seeds of *Camellia* plants. For S¹, S², S³, and S¹⁰, cf. Fig. 4.

consumed. It is the most investigated plant with respect to the biological activities of the genus *Camellia*, and saponins were found abundantly in the seeds of *C. sinensis*. In 1998, two new saponins called theasaponins E₁ and E₂ (**1** and **2**, resp.) were first isolated

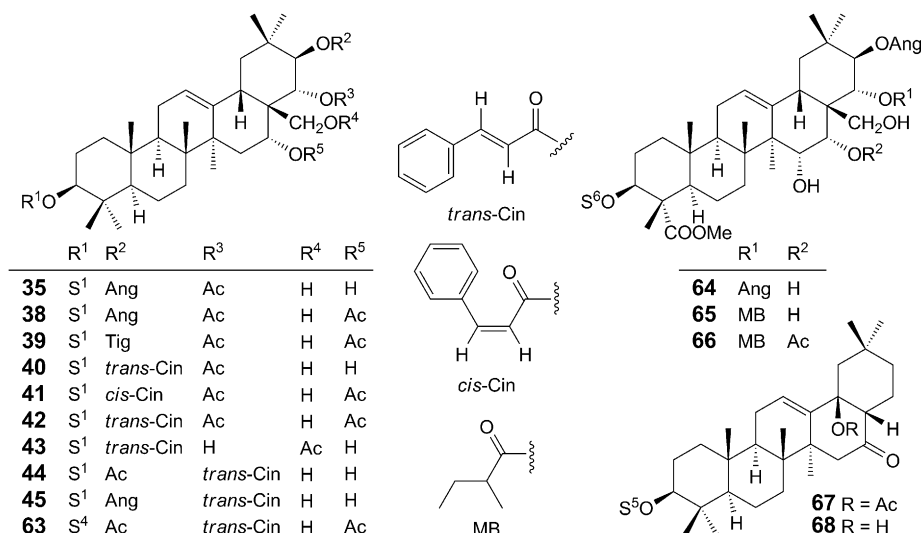


Fig. 2. Saponins isolated from the leaves or roots of *Camellia* plants. For S¹, S⁴, and S⁶, cf. Fig. 4.

from the fresh seeds of *C. sinensis* cultivated in Kyoto Prefecture (Japan) [28]. Then, from 2005 to 2007, 26 new saponins, theasaponins E₃–E₇ (**3**–**7**, resp.) [15], E₈, E₉ [29], E₁₀–E₁₃ [30] (**8**–**13**, resp.), theasaponins A₁–A₃ [16], A₄, A₅ [29], A₆ and A₇ (**14**–**21**, resp.), B₅ [31], theasaponin C₁ [29], theasaponins F₁–F₃ [16] (**23**–**25**, resp.), theasaponins G₁ [29], G₂ [30] (**26** and **27**, resp.), and theasaponin H₁ (**28**) [29] were reported from the seeds collected from the title plant cultivated in Shizuoka Prefecture (Japan), together with twelve known saponins, **1**, **2**, assamsaponins A–D, F, and I (**29**–**34**, resp.), floratheasaponin A (**35**), camelliasaponins B₁ and C₁ [15] **36** and **37**, resp., and foliatheasaponin III (**38**) [31]. In 2008, seven saponins, **29**–**34**, and foliatheasaponin I (**39**) were reported from the seeds collected from the plant cultivated in Hangzhou, Zhejiang Province (P. R. China) [32]. Among them, saponin **39** was reported from the seeds of *C. sinensis* for the first time.

From the leaves of *C. sinensis*, nine saponins, **35**, **38**–**45**, were isolated. Among them, foliatheasaponins III, I, II, and IV [25] **38**–**41**, resp., as well as theasaponin B₁ (**42**) [33] and isotheasaponins B₁ – B₃ (**43**–**45**, resp.) [34] were reported as new saponins.

From the flower buds of *C. sinensis*, three new saponins named floratheasaponins A, B, and C (**35**, **46**, and **47**, resp.), were isolated from samples collected in Shiga Prefecture (Japan) [23], and eleven new saponins identified as florasaponins D–I (**48**–**53**, resp.) [24], floratheasaponin J (**54**) [35], and chakasaponins I–III [19], V, and VI [36] **55**–**59**, resp. were reported from plants cultivated in Anhui, Fujian, and Sichuan Province (P. R. China), respectively, together with a known assamsaponin E (**60**) [24].

2.2. From *C. sinensis* var. *assamica*. Besides *C. sinensis*, *C. sinensis* var. *assamica* is another important source for tea. This plant is widely cultivated in Sri Lanka, India, Indonesia, and Yunnan Province of P. R. China. The saponin fraction from the seeds of *C. sinensis* var. *assamica* cultivated in Sri Lanka, showed potent protective effect on

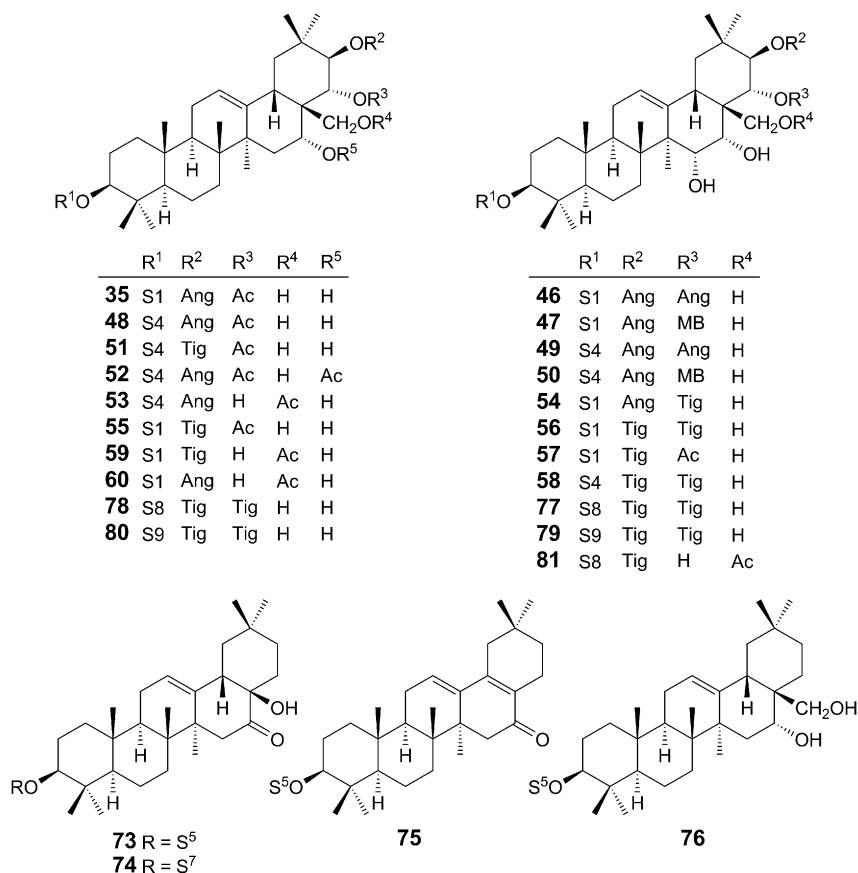


Fig. 3. Saponins isolated from the flower buds of *Camellia* plants. For S¹, S⁴, S⁵, S⁷, S⁸, and S⁹, cf. Fig. 4.

gastric mucosal lesions induced by EtOH in rats [13][14]. From this fraction, nine new acylated triterpenoid saponins, named assamsaponins A–D, E [13], and F–I [14] (**29–32**, **60**, **33**, **61**, **62**, and **34**, resp.), were isolated, together with three known saponins, **1**, **2**, and **36** [13]. From the leaves of the same plants, assamsaponin J (**63**) was reported [14].

Three new saponins, named TR-saponins A–C (**64–66**, resp.), were isolated as methyl esters from the roots of *C. sinensis* var. *assamica* cultivated in India after treatment with CH₂N₂. This was the only investigation on saponins from the roots of tea plants up to now [37]. Differing from the saponins from the other parts of tea plants, the tea root saponins have an aglycone with 23-oic acid, and a disaccharide instead of a tetrasaccharide as sugar residue.

2.3. From *C. japonica*. *C. japonica* is one of the best known species of *Camellia* native to Japan, Korea, and China. The seeds have been medicinally used for their stomachic and anti-inflammatory effects in traditional Chinese medicine (TCM) and Japanese folk medicine, while the flowers have been prescribed in the Chinese traditional preparations for the treatment of hematemesis and blood stagnation. In

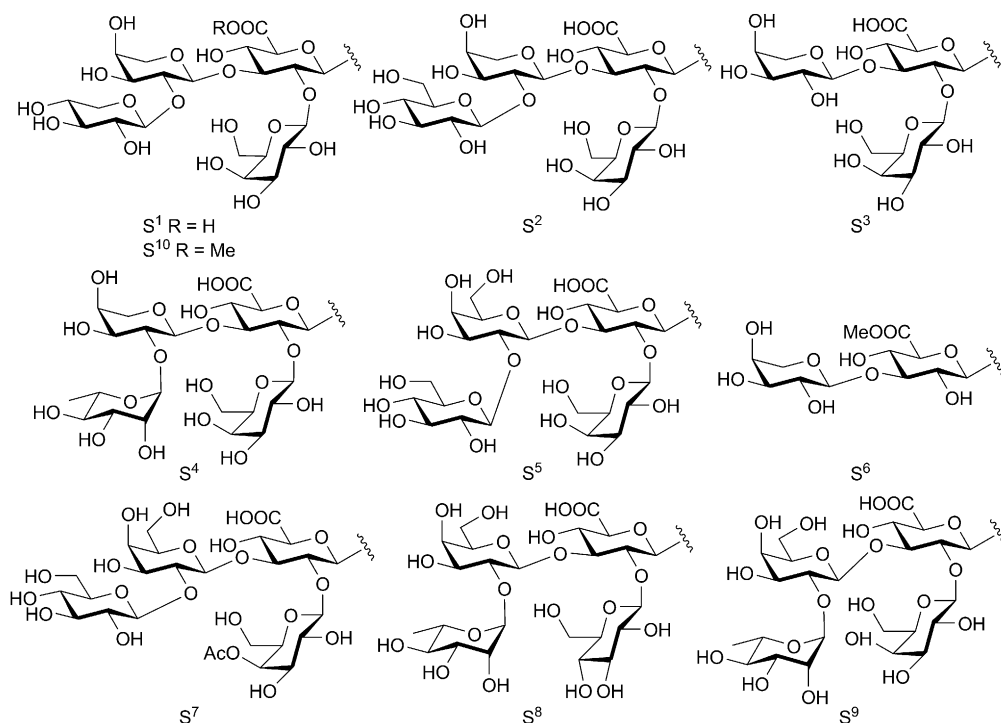


Fig. 4. The sugar residues in saponins isolated from the genus *Camellia*

1985, two saponins, camellidins I (**67**) and II, were first isolated from the fresh leaves of *C. japonica* cv. 'Benikarako' and 'Otometsubaki' cultivated in Japan, which showed antifungal activities characterized by abnormal germination of conidia [38]. The structure of camellidin II was revised as **68** by Numata *et al.* in 1987 [39].

From the seeds of *C. japonica* cultivated in Kyoto Prefecture (Japan), six new saponins, named camelliasaponins A₁, A₂ [22], B₁, B₂, C₁, and C₂ [21][22] (**69**, **70**, **36**, **71**, **37**, and **72**, resp.) were isolated.

Three new noroleanane-type saponins, named camelliosides A–C (**73**–**75**, resp.), and a new oleanane-type saponin, camelloside D (**76**), were isolated from the flower buds of the plant cultivated in Tokyo (Japan) [17][40].

2.4. From *C. sasanqua*. *C. sasanqua* is originated in Asia, particularly in Japan and China. Gardeners throughout the world cultivate this plant because of its brightly colored flowers. In 1970, one saponin named sasanquasaponin was first reported from the defatted seed cake [41], the structure of which was determined later as camelliasaponin C₁ (**37**) [15][21][22].

2.5. From *C. oleifera*. *C. oleifera* native to China is notable as an important source of edible oil obtained from its seeds. As the by-product of oil production, defatted seed cakes are used as a detergent or organic fertilizer with low economic value. The flower buds of this plant have been used for the treatment of blood vomiting and bleeding due to internal and external injuries in TCM. The MeOH extract and its BuOH-soluble

Table. *Acylated Triterpenoid Saponins from the Plants of the Genus Camellia*

	Name	Source ^{a)}	Part	Ref.
1	Theasaponin E ₁	A, B	Seed	[13][15][28]
2	Theasaponin E ₂	A, B	Seed	[13][15][28]
3	Theasaponin E ₃	A	Seed	[15]
4	Theasaponin E ₄	A	Seed	[15]
5	Theasaponin E ₅	A	Seed	[15]
6	Theasaponin E ₆	A	Seed	[15]
7	Theasaponin E ₇	A	Seed	[15]
8	Theasaponin E ₈	A	Seed	[29]
9	Theasaponin E ₉	A	Seed	[29]
10	Theasaponin E ₁₀	A	Seed	[30]
11	Theasaponin E ₁₁	A	Seed	[30]
12	Theasaponin E ₁₂	A	Seed	[30]
13	Theasaponin E ₁₃	A	Seed	[30]
14	Theasaponin A ₁	A	Seed	[16]
15	Theasaponin A ₂	A	Seed	[16]
16	Theasaponin A ₃	A	Seed	[16]
17	Theasaponin A ₄	A	Seed	[29]
18	Theasaponin A ₅	A	Seed	[29]
19	Theasaponin A ₆	A	Seed	[31]
20	Theasaponin A ₇	A	Seed	[31]
21	Theasaponin B ₅	A	Seed	[31]
22	Theasaponin C ₁	A	Seed	[29]
23	Theasaponin F ₁	A	Seed	[16]
24	Theasaponin F ₂	A	Seed	[16]
25	Theasaponin F ₃	A	Seed	[16]
26	Theasaponin G ₁	A	Seed	[29]
27	Theasaponin G ₂	A	Seed	[30]
28	Theasaponin H ₁	A	Seed	[29]
29	Assamsaponin A	A, B	Seed	[13][15][32]
30	Assamsaponin B	A, B	Seed	[13][15][32]
31	Assamsaponin C	A, B	Seed	[13][15][32]
32	Assamsaponin D	A, B	Seed	[13][15][32]
33	Assamsaponin F	A, B	Seed	[15][32]
34	Assamsaponin I	A, B	Seed	[14][15][32]
35	Floratheasaponin A	A	Seed, leaf, flower bud	[15][23][25]
36	Camelliasaponin B ₁	A, B, C	Seed	[13][15][21][22][44]
37	Camelliasaponin C ₁	A, C, D, E	Seed	[15][21][22][41][42]
38	Foliatheasaponin III	A	Seed, leaf	[25][31]
39	Foliatheasaponin I	A	Seed, leaf	[25][32]
40	Foliatheasaponin II	A	Leaf	[25]
41	Foliatheasaponin IV	A	Leaf	[25]
42	Theasaponin B ₁	A	Leaf	[25][33]
43	Isotheasaponin B ₁	A	Leaf	[25][34]
44	Isotheasaponin B ₂	A	Leaf	[34]
45	Isotheasaponin B ₃	A	Leaf	[34]
46	Floratheasaponin B	A	Flower bud	[23]
47	Floratheasaponin C	A	Flower bud	[23]
48	Floratheasaponin D	A	Flower bud	[24]
49	Floratheasaponin E	A	Flower bud	[24]
50	Floratheasaponin F	A	Flower bud	[24]

Table (cont.)

	Name	Source ^{a)}	Part	Ref.
51	Floratheasaponin G	A	Flower bud	[24]
52	Floratheasaponin H	A	Flower bud	[24]
53	Floratheasaponin I	A	Flower bud	[24]
54	Floratheasaponin J	A	Flower bud	[35]
55	Chakasaponin I	A	Flower bud	[19]
56	Chakasaponin II	A	Flower bud	[19]
57	Chakasaponin III	A	Flower bud	[19]
58	Chakasaponin V	A	Flower bud	[36]
59	Chakasaponin VI	A	Flower bud	[36]
60	Assamsaponin E	A	Flower bud	[13]
		B	Seed	[24]
61	Assamsaponin G	B	Seed	[14]
62	Assamsaponin H	B	Seed	[14]
63	Assamsaponin J	B	Leaf	[14]
64	TR-Saponin A	B	Root	[37]
65	TR-Saponin B	B	Root	[37]
66	TR-Saponin C	B	Root	[37]
67	Camellidin I	C	Leaf	[38]
68	Camellidin II	C	Leaf	[38][39]
69	Camelliasaponin A ₁	C	Seed	[22]
70	Camelliasaponin A ₂	C	Seed	[22]
71	Camelliasaponin B ₂	C	Seed	[21][22]
72	Camelliasaponin C ₂	C	Seed	[21][22]
73	Camellioside A	C	Flower bud	[17][40]
74	Camellioside B	C	Flower bud	[17][40]
75	Camellioside C	C	Flower bud	[17][40]
76	Camellioside D	C	Flower bud	[17][40]
77	Yuchasaponin A	D	Flower bud	[20]
78	Yuchasaponin B	D	Flower bud	[20]
79	Yuchasaponin C	D	Flower bud	[20]
80	Yuchasaponin D	D	Flower bud	[20]
81	Jegosaponin B	D	Flower bud	[20]
82	Theasaponin E ₂ methyl ester	D	Seed	[43]

^{a)} A: *C. sinensis*; B: *C. sinensis* var. *assamica*; C: *C. japonica*; D: *C. oleifera*; E: *C. sasanqua*.

fraction from the flower buds of this plant cultivated in Fujian Province (P. R. China) exhibited inhibitory effects on EtOH- and indomethacin-induced gastric mucosal lesions in rats [20]. Four new saponins, yuchasaponins A–D (**77–80**, resp.), together with jegosaponin B (**81**), were isolated from the flower buds [20]. From the defatted seed cake, sasanquasaponin (**37**) [42], theasaponin E₂ methyl ester (**82**), [43], and a saponin mixture containing **36** [44] were isolated.

3. Biological Activities. – 3.1. *Gastroprotective Activity.* The MeOH extract and its BuOH-soluble fraction from the seeds of *C. sinensis* [15][16] and *C. sinensis* var. *assamica* [13][14], the flower buds of *C. sinensis* [18][19], *C. japonica* [17], and *C. oleifera* [20] were found to exhibit potent gastroprotective effects on EtOH- and

indomethacin-induced gastric mucosal lesions in rats. Furthermore, the saponin mixture was found to exhibit an inhibitory effect on gastric emptying and an accelerating effect on gastrointestinal transit in mice [14]. It is noteworthy that no drug having both activities has been identified so far.

With regard to pure saponins, theasaponin E₁ (**1**) exhibited more potent gastro-protective activity (inhibition: 94.3%) at a dose of 10 mg/kg than the reference drug omeprazole (inhibition: 63.4% at 30 mg/kg) [13]. Saponin **1** was found to inhibit gastric emptying and accelerate gastrointestinal transit, while theasaponin E₂ (**2**) showed no such activities. These findings indicate that the location of the AcO group is the key to the activity [14].

Saponins **1** and **2**, theasaponins E₅, A₁, A₂, and F₃ (**5**, **14**, **15**, and **25**, resp.), and assamsaponins A–D (**29–32**, resp.) isolated from the seeds of *C. sinensis* (Japan) showed potent protective effects on EtOH-induced gastric lesions in rats at a dose of 5.0 mg/kg (inhibition: 71.4, 77.6, 45.4, 37.0, 54.7, 42.1, 61.0, 39.7, 64.4, and 47.9%, resp.), and their activities were more potent than that of omeprazole (inhibition: 43.1% at 10 mg/kg) [15][16].

Camelliosides A and B (**72** and **73**, resp.) from the flower buds of *C. japonica* showed protective effects on both EtOH- and indomethacin-induced gastric lesions (EtOH-induced: **72**, ED₅₀ 9.7 mg/kg; **73**, 7.4 mg/kg; indomethacin-induced: **72**, 21 mg/kg, **73**, 13 mg/kg), and their effects were equivalent or stronger than those of the reference compounds, omeprazole (EtOH-induced: 10 mg/kg; indomethacin-induced: 4.7 mg/kg) and cimetidine (EtOH-induced: 69 mg/kg; indomethacin-induced: 21 mg/kg) [17]. Floratheasaponins A–C (**35**, **46**, and **47**, resp.) from the flower buds of *C. sinensis* also exhibited potent inhibitory effects on EtOH- and indomethacin-induced gastric mucosal lesions in rats [18]. Chakasaponins I–III (**54–56**, resp.) from the flower buds of *C. sinensis* showed an accelerating effect on gastrointestinal transit in mice at a dose of 100 mg/kg [19].

With regard to structure–activity relationships between the saponin structures and gastroprotective activities, the following structural requirements were suggested: 1) the acyl groups at C(21) and/or C(22) were essential for the activity, 2) acetylation of the OH group at C(16) reduced the activity [15], 3) the Ac moiety at C(28) enhances the activity, and 4) theasaponins with a CH(23)=O group exhibit more potent activities than those with CH₂(23)OH or C(23)(=O)OMe groups [16]. Since the structures of yuchasaponins A–D (**77–80**, resp.) with two acyl groups at C(21) and C(22) are similar to those of gastroprotective saponins, yuchasaponins might be the gastroprotective principles of the flower buds of *C. oleifera* [20].

3.2. EtOH-Absorption Inhibitory Activity. The MeOH extract of the defatted seeds of *C. japonica* has inhibitory effect on the EtOH absorption in rats. Camelliasaponins B₁, B₂, C₁, and C₂ (**36**, **70**, **37**, and **71**, resp.) isolated from the seeds showed inhibitory effect on EtOH absorption after a single oral administration at the dose of 100 mg/kg. Saponin **36** exhibited the most potent inhibitory activity. This result also indicated that the acyl group in camelliasaponins was essential for the activity [21][22].

3.3. Antihyperlipidemic Activity. The MeOH extract and its BuOH-soluble fraction from the flowers of *C. sinensis* (Japan) were found to suppress serum triglyceride elevation in olive oil-treated mice. Isolated floratheasaponins A, B, and C (**35**, **46**, **47**, resp.) significantly suppressed the increase in serum triglyceride levels 2 h after

administration of olive oil at doses of 25–100 mg/kg. Their activities were more potent than those of theasaponins E₁ and E₂ (**1** and **2**, resp.). It was found that the acyl groups at C(21) and C(22) are essential and the C(23)HO function is not preferable for activity [23].

3.4. Anti-Allergic Activity. The MeOH extract and floratheasaponins A–F (**35** and **46–50**, resp.) from the flower buds of *C. sinensis* (Anhui Province, P. R. China) were found to show inhibitory activities on the release of β -hexosaminidase from RBL-2 H3 cells [24]. Particularly, floratheasaponins B and E (**49**) displayed strong activities (inhibition: 59.8 and 52.3% at 3 μ M, resp.), and their activities were stronger than those of two antiallergic compounds, tranilast and ketotifen fumarate (inhibition: 8.2 and 7.7% at 30 μ M, resp.).

Foliatheasaponin II (**40**; inhibition: 46.3% at 3 μ M; 55.7% at 6 μ M) and foliatheasaponin III (**38**; inhibition: 22.8% at 6 μ M; 47.0% at 10 μ M) isolated from the leaves of *C. sinensis* (Japan) also inhibited the release of β -hexosaminidase, and their activities were stronger than those of tranilast (inhibition: 22.4% at 100 μ M) and ketotifen fumarate (inhibition: 27.6% at 100 μ M) [25].

3.5. Other Activities. Camellidins I and II (**66** and **67**, resp.) from the fresh leaves of *C. japonica* showed antifungal activities characterized by abnormal germination of conidia [38]. Han *et al.* reported that a mixture of teasaponins (**1/2**: 88%/12%) obtained from the leaves of oolong tea (*C. sinensis*; Fujian Province, P. R. China) displayed anti-obese effect in high-fat-diet-treated mice and pancreatic lipase inhibitory effect [45]. The MeOH extract from the flower buds inhibited the enzyme activity of the porcine pancreatic lipase. Chakasaponins I–III (**54–56**, resp.) also inhibited the pancreatic lipase activity with IC_{50} values of 170–530 μ M, although their activities were much weaker than that of orlistat, a lipase inhibitor as positive control (IC_{50} 36 nM) [19].

Floratheasaponins A–C (**35**, **46**, and **47**, resp.) from the flower buds of *C. sinensis* were found to exhibit potent inhibitory effects on serum glucose elevation in sucrose-loaded rats [18]. A mixture of tea-seed saponins from *C. sinensis* was reported to inactivate human type A and B influenza viruses. However, these saponins were also toxic to the host cells, and further studies are needed [46].

On application of 0.5 mM solutions of saponins **1** and **2** isolated from the seeds of *C. sinensis*, **2** was found to suppress the sweet taste of 0.1M sucrose, while **1** showed no antisweet activity [28]. Saponin **1** showed a high activity in killing cells of *Zygosaccharomyces rouxii* (MIC: 2.5 μ g/ml), while **2** did not have any effect on the salt tolerance of *Z. rouxii* or *Saccharomyces cerevisiae* [47]. Camelliosides A and B (**72** and **73**, resp.) from the flower buds of *C. japonica* were found to cause the aggregation of the rabbit platelets (10–100 μ g/ml) [17] in a concentration-dependent manner.

Sasanquasaponin (**37**) inhibited ischemic/reperfusion-induced cardiac arrhythmias by modulating intracellular Cl[–] homeostasis in mouse hearts [48]. Saponin **37** protected cardiomyocytes against oxidative stress induced by anoxia-reoxygenation by attenuating reactive oxygen species generation and increasing activities of endogenous antioxidants [49].

4. Conclusions. – Saponins are widely distributed amongst plants and have a wide range of biological properties. Phytochemical investigations of *Camellia* species have

revealed that many constituents characterized as acylated triterpenoid saponins from this genus exhibit significant biological and pharmacological activities. However, among *ca.* 119 species in genus *Camellia*, only five species have been investigated for their saponin constituents, and many *Camellia* species have received no or only little attention. Further phytochemical and biological studies are necessary to focus on these plants to search for further potentially bioactive components.

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