SARSASAPOGENIN IN CALLUS CULTURE OF *ASPARAGUS RACEMOSUS*

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REMARKABLE success has been achieved in the production of secondary metabolites by plant cell cultures in the past decade[1,2]. Over fifty different steroids have been isolated from various plant tissue cultures[3]. *Asparagus racemosus* Willd., an indigenous medicinal species[4] of the family Liliaceae, contains sarsasapogenin[5]. Due to the importance of this steroid in pharmaceutical industry *A. racemosus* was successfully grown *in vitro* and the ability of such cultured cells to produce sarsasapogenin was assessed.

The callus was initiated from shoot segments of *A. racemosus* grown on Murashige and Skoog's basal medium[6] supplemented with 2,4-D and kinetin, 1 mg/l each[7]. The callus tissue has been maintained on the same medium at $25 \pm 1°C$ with 16 hr of light by subculture every 4 to 6 weeks.

The callus was harvested, dried and hydrolysed with 30% v/v hydrochloric acid for 4 hr, filtered and neutralized. The residue was then dried at 60°C and Soxhlet extracted with chloroform for 30 hr. The chloroform extract was concentrated and analyzed for sapogenin by chromatography.

The extract was applied to thin-layer plates precoated with silica gel G and developed with chloroform-acetone (80:20). Liebermann-Burchard reagent[8] was used as the detection reagent. Standard sapogenin samples were used along with the extracts.

A Hewlett Packard Model 5730A gas chromatograph containing 'U' shaped steel column (20" × 3/16") with the stationary phase 10% UCW-982, 80-100 WAW-DMCS was used for sapogenin analysis. The column temperature was 240°C and N_2 was the carrier gas at a pressure of 3 kg/cm^2.

Synthesis of sarsasapogenin in the callus tissue of *A. racemosus* as evidenced from TLC (R_f 0.63) and GLC (RT 12.10') is now established as reported in this note. Previously sarsasapogenin has been identified from tissue culture of *Yucca*.[9]

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KARSCHIA NIGERRIMA SACC. (PATELLARIACEAE)—A NEW REPORT FROM INDIA

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So far only 3 species of *Karschia* Korb., a lichenicolous genus, are known from India: *K. prinsipiae* Chona *et al*[1], *K. lignyota* (Fr) Sacc and *K. buelloides* (Korb) Sacc by few workers[1-3]. However, *K. prinsipiae* has been transferred to *Gibbera* Fr-Dothideales by Arx and Mueller[4] in 1975. In the present note *K. nigerrima* Sacc is recorded for the first time from India.

Karschia nigerrima Sacc Syll. Fung, **8**, 780, 1889. Collection examined: 3021 (PAN, K) on dead wood, Narkanda, (alt. 2,400 m) Simla, H. P. August 6, 1965. Leg H. Singh.

In the present collection the asci are larger than reported by Saccardo[4] in the original description of the taxon. The other recorded species from India *K. buelloides* differ in having larger 100 × 18 μm asci and

K. lignyota has smaller (10–) 11–14 × 4–5 μm oblong–ellipsoid ascospores.

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ROLE OF CYTOMIXIS IN THE SPECIATION OF *JASMINUM*

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JASMINES are highly domesticated ornamental plants grown mainly for their fragrant flowers and propagated entirely by vegetative means. Sexual reproduction is obsolete and almost absent in many of the *Jasminum* species being either pollen sterile or seed sterile. An earlier study by the authors shows[1] that the sterility of their pollen is ultimately due to some defective gene function in their meiotic cells causing various meiotic abnormalities, including cytomixis which occur spontaneously and abundantly in their pollen mother cells. Variation of hereditary characters by gene mutation and recombination, which form the raw materials of evolution, and operative through the sexual reproduction can be ruled out. At the same time, it is interesting to note that there are about five hundred species of *Jasminum* and several varieties of these exist today[2]. The question, as to how speciation takes place in this type of sexually sterile but vegetatively reproducing plants has been the subject of the authors' investigation for quite some time. These investigations on the hereditary variation through alteration in the somatic chromosome complement of *Jasminum* species offer a clue to solving the problem of the mechanism of speciation in vegetatively reproducing plants. The results of the method of speciation through the process of cytomixis in the shoot meristems of *Jasminum* spp. are presented.

For studying the somatic chromosome number and behaviour in several species of *Jasminum*, samples of *J. angustifolium*, *J. rigidum*, *J. malabaricum*, *J. revolutum*, *J. grandiflorum*, *J. sambac* var. 'sujimallikae', 'gundumalli', 'iruvachy', 'adukkumalli' etc. collected from different parts of South India and grown in the Kerala University Botanic Gardens were used. Shoot tips of these plants were fixed at 4.30 p.m. in 1:1:3 chlor-acetic-ethanol and stained by lactopropionic orcein[3]. The squash preparations were examined immediately after staining.

Shoot meristem cells at various stages of mitosis showed cytomixis. The cytoplasmic communicating channels between adjoining cells were observed to have partial or complete chromosome complement migrating through them (figure 1). Another interesting observation was the occurrence of cells with differing chromosome numbers in the same tissue (figure 2). For instance, in *J. revolutum* (2n = 65) cells with 59, 65 and 78 chromosomes or in *J. sambac* var. 'adukkumalli' (2n = 26) cells with 22, 23, 26 and 27 chromosomes or in *J. angustifolium* (2n = 52) cells with 33, 46 and 52 chromosomes were encountered in the same slide. In other words, a myxoploid condition was existing with no fixity of the chromosome numbers in the shoot meristem cells of the plants investigated. Similar condition with no fixity of chromosome number exists in the root tip cells of *J. humile*, *J. arborescens* and *J. angustifolium* also[4].

Cytomixis as originally defined[5] involves the migration of part or whole complement of chromosomes from one PMC to another. In the PMCs since there is no other division other than the meiotic division, further shuffling of chromosomes could not be expected. Here the meiotic products either degenerate or become nonfunctional in fertilization as indicated by the sterility of pollen and seeds of the jasmines. On the other hand, in the meristematic tissue of the shoot bud, repeated mitotic divisions are likely to involve a series of cytomixis, shuffling of chromosomes and consequent stable genetic constitution. It results in cells with different levels of ploidy but more commonly aneuploidy as observed. When one of these cells with altered chromosome number undergoes normal mitotic divisions, it forms a homogeneous but heteroploid tissue. From this tissue a new strain can be generated in the form of a shoot bud as in a chimera. This sort of chimeral variation spontaneously occurring in other plants has been reported[6]. A shoot arising from such a tissue when propagated vegetatively after