

Effect of Lancing on Alkaloids Profile of Capsules in Opium Poppy

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The commercially important crop of opium poppy is grown for production of alkaloids. These alkaloids are now favoured to be extracted from dry capsules due to constraint of manual labour rather than from latex as practised earlier. However, due to presence of bound form of alkaloids in capsules, the process of extraction complicates. Hence, a method has been developed for the extraction of alkaloids from the poppy straw and analysed over HPLC. The lanced and unlanced dried capsules of four promising varieties alongwith two control varieties, were analysed for the spectrum of five major alkaloids by the method developed by us. It was observed that unlanced capsules have greater proportion of alkaloid contents in comparison to lanced capsules. Hence, unlanced capsules of opium poppy can be used for better production of alkaloids.

The opium alkaloids are obtained from poppy plants in two ways : (a) from the latex which on drying is called opium-gum, and (b) from the plant organs specially capsules.

In India, mainly the latex is used for the production of opium alkaloids. However, due to presence of bound form of alkaloids in capsules, the process of extraction is complicated. Hence, an attempt has been made to develop a method for releasing the bound alkaloids in capsules and their analysis on HPLC. The major alkaloids in both lanced and unlanced capsules of the promising varieties and controls were also studied.

MATERIALS AND METHODS

In this study, promising varieties of opium poppy (IC-30, IC-42, IC-88 and IC-128) collected from different agro-climatic zones alongwith two established varieties as controls were grown at NBPGR Research Farm at Issapur, New Delhi, in 1985-86. The composite samples of fully matured lanced and unlanced capsules were collected from the plants of each variety for analysing five major alkaloids.

The dried, mature, terminal and lateral capsules with 2-3 cm stem attached, collected from ten plants of each variety were powdered and used as composite samples for analysis. The dried powder (1 g) was shaken with 25 ml of 5% glacial acetic acid for 20 minutes using a mechanical shaker. The extract was filtered and residue was extracted similarly three times. Residue was rejected and the pH of the total extract was adjusted to 9.0-9.5 with concentrated ammonia solution (25% NH₃). The liberated alkaloids were repeatedly extracted

(4 × 15 ml) with chloroform; isopropanol (3 : 1) solvent system. The extract was dried over anhydrous sodium sulphate and evaporated to dryness under vacuo at 50°C. The residue was redissolved in 5 ml of methanol (HPLC grade) and injected into HPLC after making it free of any particulate matter by passing through sample clarification kit. Peak heights obtained on the plotter were compared with those of standard compounds and the percentage of each alkaloid in the sample calculated.

RESULTS AND DISCUSSION

Unlike in latex, opium alkaloids are present in bound forms in capsules of *Papaver somniferum*. The complexity and molecular weight of these bound alkaloids seem to increase during ripening of capsules and these substances represent transitional forms in the metabolism and translocation of morphine from latex to seed (Fairbairn and Steele, 1980). To overcome this practical difficulty, a new method was developed in which finely powdered poppy capsules (without seeds) were repeatedly extracted with 5% acetic acid. Four extractions of 5% acetic acid were enough to extract optimal amount of alkaloids. The total extract was adjusted to different pH for the liberation of alkaloids and optimum pH was 9.0 to 9.5. The free alkaloids were extracted from the aqueous solution by chloroform; isopropanol (3 : 1) solvent system. The residue left after removal of solvent was subjected to 'High Pressure Liquid Chromatographic' analysis for five major alkaloids under conditions developed earlier (Srivastava and Maheshwari, 1985).

The comparison of alkaloids of the poppy capsules of different varieties tested showed that the percentage of each alkaloid varied greatly between lanced and unlanced capsules (Table 1). Unlanced capsules possessed more morphinane and

TABLE 1. ALKALOIDS PROFILE OF THE POPPY CAPSULE OF PROMISING VARIETIES

Varieties	Percentage of Morphinane alkaloids				Percentage of Isoquinoline alkaloids		
	Morphine	Codeine	Thebaine	Total	Narcotine	Papaverine	Total
<i>In lanced capsules</i>							
IC-30	0.32	0.14	0.02	0.48	0.06	0.01	0.07
IC-42	0.12	0.24	0.12	0.48	0.01	0.06	0.07
IC-88	0.12	0.14	0.02	0.28	0.01	0.03	0.04
IC-128	0.12	0.16	0.02	0.30	0.02	0.02	0.04
Ranjhatak (control)	0.14	0.24	0.04	0.44	0.02	0.02	0.04
Jawahar afeem (control)	0.08	0.26	0.10	0.44	0.02	0.16	0.18
<i>In unlanced capsules</i>							
IC-30	0.40	0.20	0.06	0.66	0.14	0.01	0.15
IC-42	0.30	0.14	0.03	0.47	0.22	0.07	0.29
IC-88	0.32	0.12	0.03	0.47	0.10	0.08	0.18
IC-128	0.72	0.10	0.03	0.85	0.18	0.16	0.34
Ranjhatak (control)	0.44	0.20	0.12	0.76	0.14	0.08	0.22
Jawahar afem (control)	0.38	0.12	0.04	0.54	0.24	0.36	0.60

isoquinoline alkaloids as compared to the lanced capsules. Amongst morphinane alkaloids, morphine content was greatly reduced by lancing. Similarly, thebaine, narcotine and papaverine contents also reduced in lanced capsules. The only exception was codeine content which was found to be unaffected in lanced capsules. It rather increased in some cases.

The opium alkaloids are biosynthesised in the opium poppy through shikimate pathway (Jindra *et al.*, 1966). Synthesis of morphine takes place by successive demethylation process, i.e. from thebaine to codeine and then to morphine. In lanced capsules, all the alkaloids except codeine were reduced in quantity. It has been noticed earlier that in contrast to the alkaloids distribution found in the plant of *papaver somniferum*, callus-cultures accumulated a predominance of codeine and in some cases also thebaine (Hodges and Rapoport, 1982).

Similarly, in cell suspension culture, codeine was not metabolised to morphine whereas codeinone (a precursor of codeine) was transformed to codeine indicating that cell line lacks the ability to demethylate codeine to morphine (Tam *et al.*, 1982). On the other hand, it was observed that latex is capable of synthesising morphine in *in vitro* conditions (Fairbairn and Djote, 1970). This indicates that the metabolic system(s) necessary for biosynthesis of morphine, were mainly present in the latex. Hence, it could be inferred that whenever latex was drawn from the capsules, the conversion of codeine to morphine was inhibited due to inhibition of specific metabolic system(s). This may probably create the situation where codeine accumulates at the cost of morphine. It further suggests the possible application of lanced capsules for tapping the codeine alkaloid. However, if only capsules are to be used for the production of alkaloids, then unlanced capsules will be preferable due to higher content of alkaloids present in them. This can overcome the constraint of skilled labour required for collection of latex and also help in preventing clandestine activities in opium-gum trade.

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