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Full Length Research Paper

Analysis of silymarin in the seeds of white and blue capitulum of *Silybum marianum* plants

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Silybum marianum **is a wild medicinal plant containing silymarin, which is a known drug, used as the hepatoprotective agent.** ***S. marianum*** **growing in different areas of NWFP were collected and analyzed for the silymarin and oil contents. In all the samples analyzed for the silymarin, high yield was found in the sample collected from Swat district (3.85%) while high yield of oil was recorded in the sample collected from Kohat district (27.87%). The rest of samples for silymarin contents were found between 2.98 to 3.85% while the oil contents were found to be between 11 to 27.87%. Both the yield of silymarin and oil contents found in all the samples depend on the environment in which the plants were grown and the environmental conditions affecting both the contents of silymarin and oil of this highly medicinal plant. The analysis is of particular importance for collecting this medicinal plant for silymarin content and utilization in the pharmaceuticals.**

Key words: *Silybum marianum,* silymarin, milk thistle, oil contents.

INTRODUCTION

Silybum marianum commonly known as milk thistle is an annual or biennial herb, with stem (20 to 150 cm high), leaves (25 to 50 cm long, 12 to 25 cm wide) and fruit (15 to 20 mm long) it belongs to the family Asteraceae (Compasittae). The stem branches at the top and reaches a height of 4 to 10 ft. The leaves are wide blotches with veins. The flowers are red purple. The small, hard skinned fruit is brown spotted and shiny. Milk thistle is easy to grow and it matures in less than a year. It is indigenous to North America, Asia minor, Southern Europe, Russian Federation, it naturalized to South and North America, Australia, China, Central Europe and is found wild in North-West Frontier Province (NWFP) and the Punjab areas of Pakistan (Burgess, 2003).

Silymarin is used as a hepatoprotector for oral treatment of toxic liver damage and of chronic inflammatory liver diseases and liver cirrhosis (Valenzuela et al., 1986; Flora et al., 1988). Apart from its antioxidant properties and its role in stimulating protein synthesis and cell regeneration (Katiya et al., 1997). Silymarin may also reduce incidence of certain forms of cancer (Sonnenbichler et al., 1988). The extracts prepared from the roots and seeds have been used for liver disorders of bile duct and spleen (Tawaha et al., 2007). In a recent study, it has been demonstrated that the protective effect of silymarin is most likely due to its free- radical scavenging activity (FRSA) (Burgess, 2003).

Dried extracts of milk thistle seeds contain approximately 60% silymarin. Silymarin consists of four flavonolignans of silybin (50 to 60%), isosilybin (5%), silychristin (20%) and silydianin (10%) (Burgess, 2003). The three isomers, silybin, silychristin and silydianin differ only in the linkage of the taxifolin moiety to coniferyl alcohol (Figure 2). Recently silymarin, the purified compound of the seeds and its major isomer silybin are used in the manufacture of therapeutic products administered against liver diseases, jaundice and gallstones (Vogel, 1968). The liver protective action of silybin and silymarin has been established *in vivo* and in many *in vitro* liver damage models (Schriewer et al., 1973). Silybin has also been found to active on the polymerase A of hepatocytes,

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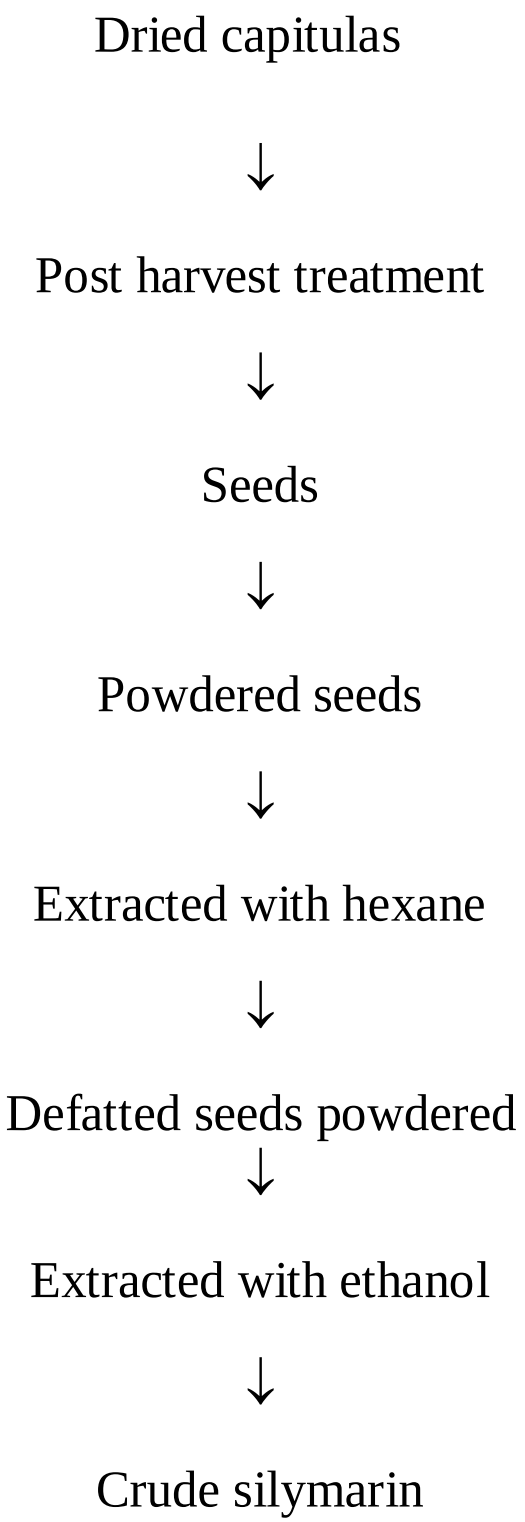

Figure 1. Flow chart diagram of *S. marianum* seeds processing.

which corresponds with a stimulation of protein synthesis in liver cell (Machicao et al., 1977).

Despite the wide spread distribution of *S. marianum* in Pakistan as indigenous wild specie, the seeds have not been studied as a source of silymarin and oil.

In the present study, seeds of the white and blue capitulum of *S. marianum* were collected from different natural environmental zones of NWFP exposed to variable environmental conditions for the analysis of crude contents of silymarin and oil. The study is of particular importance for the investigation of silymarin contents, which could be a good source for pharmaceutical utilization.

EXPERIMENTAL

Ripe fruits of white and blue capitulum of *S.marianum* were collected from different regions of NWFP.

Dried capitulas of *S. marianum* were thrashed (Khan et al., 2003). The materials were crushed into powder. The powdered material (10 g) was extracted three times with Hexane (250 ml) through solvent recycling at room temperature for 12 h. The combined extracts were concentrated under reduced pressure to yield the yellow oil (Table 1).

The defatted material was then extracted four times with ethanol (250 ml) by percolation and recycling at room temperature for 12 h. The combined ethanolic extracts was filtered, and concentrated under reduced pressure and gradually added to water under intensive stirring. The product was salted out from the aqueous extract with sodium chloride (25%). The precipitated material was then filtered, washed with water, dried in an electric oven and powdered (Figure 1). The yield of the crude material for each of the sample is shown in Table 1.

RESULTS AND DISCUSSION

As can be seen from Table 1, high yield of silymarin was found in sample collected from Swat district (3.87%) which was followed by Charsadda (3.75%), Nowshehra (3.72%), Khyber agency (3.70%), Kohat (3.64%) and Swabi district (3.41%). However, the least amount of silymarin was recorded from Bajawar agency (2.09%).

The percentage yield of silymarin of the white capitulum collected from Peshawar area is 3.61% while in the blue capitulum its concentration was 3.53%. The plant sample of white capitulum from Nowshera zone has relatively lower concentration (3.49%) of silymarin than the blue capitulum seeds (3.66%). Similarly both the white and blue capitulum seeds collected from Charsadda area have closely the same concentration 3.74 and 3.70% of crude silymarin. In a similar way both the white and blue capitulum seeds obtained from Khyber agency has the concentration of 3.78 and 3.80% respectively. The concentration of white capitulum seeds plant sample secured from Mardan area has 3.78% while in the blue capitulum its concentration is relatively different (3.58%). The concentration of silymarin in both the samples found closely the same 3.71 and 3.68%. A similar amount of silymarin was recorded in the white 3.79% and blue 3.76% capitulum plant sample. A slightly variable

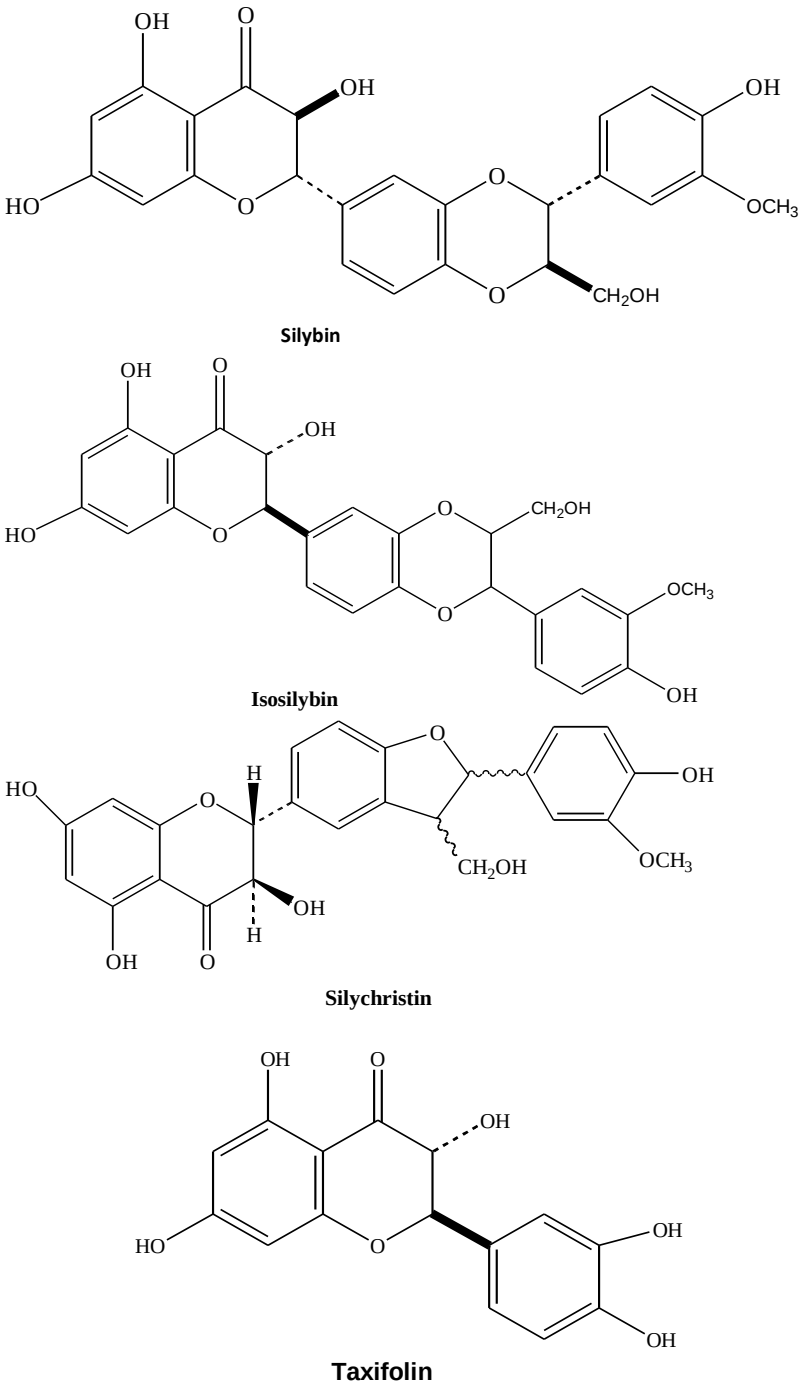

Figure 2. Structures of the isomers of silymarin.

concentration of silymarin was obtained in the white 3.45% and blue 3.56% from Karak region.

The concentration of oil in both the white and blue capitulum plant samples was found as the same as those collected from different areas including Peshawar, Nowshera, and Mardan (22.40, 22.21, 22.8, 22.5, 22.3 and 22.5%). However a little high percentage (23.6%) of oil was found in the blue capitulum and 22% in the white capitulum seeds sample of *S. marianum* collected from Charsadda area. A relatively different amount of oil was recorded in the white (22.2%) and blue (20.9%) capitulum sample from Khyber agency. The rest of the white and blue capitulum plant samples collected from Swabi, Kohat and Karak areas have the oil contents (21.60, 23.05, 20.8, 22.07, 20.06, 22.16%) respectively.

The silymarin compounds in milk thistle actually accelerate protein synthesis in the liver, which stimulates the production of new, healthy liver cells. The flavonoids

Table 1. Crude yield of Silymarin and oil collected from different environmental zones of NWFP.

S/No.	Name of area	White capitulum seeds silymarin yield (%)	Blue capitulum seeds silymarin yield (%)	White capitulum seeds oil yield (%)	Blue capitulum seeds oil yield (%)
1	Peshawar	3.61	3.53	22.40	22.21
2	Nowshehra	3.49	3.66	22.80	22.50
3	Charsadda	3.74	3.70	22.00	23.6
4	Khyber Agency	3.78	3.80	22.20	20.9
5	Mardan	3.78	3.58	22.30	22.5
6	Swabi	3.71	3.68	21.60	23.05
7	Kohat	3.79	3.76	20.8	22.07
8	Karak	3.45	3.56	20.06	22.16

contained in milk thistle act as effective free radical scavengers, which also protect the liver from damage. Silymarin is considered a spleen, and gallbladder tonic. The therapeutic activity of silymarin is based on two sites or mechanisms of action:

(a) It alters the structure of the outer cell membrane of the hepatocytes in such a way as to prevent penetration of the liver toxin into the interior of the cell.
(b) It stimulates the action of Nucleolar polymerase A, resulting in an increase in ribosomal protein synthesis, and thus stimulates the regenerative ability of the liver and the formation of new hepatocytes.

Silymarin binds hepatocyte (liver cell) membranes, which protects them from the potential damage of environmental toxins, (such as the death cap mushroom), foreign chemicals, endogenous poisons and free radicals.

Milk thistle is believed to protect the cells of the liver by blocking the entrance of harmful toxins and helping to remove these toxins from the liver cells. As with other bioflavonoids, silymarin is a powerful antioxidant. Milk thistle also regenerates injured liver cells.

Silymarin enters the liver cells and promotes their ability to produce certain enzymes, which are vital to liver health. This action can speed the healing of liver cells from injury or disease. By enhancing the production of these enzymes, liver cell regeneration is also stimulated.

Thus the above analysis showed that both the white and blue capitulum seed samples of S*. marianum* plant obtained from different environmental zones yielded almost the same amount of silymarin and a slightly different amount of oil contents. The slight difference in the contents of oil may be due to different environmental conditions in which the *S. marianum* plants are grown.

The physiochemical properties of *S. marianum* were studied by Khan et al. (2003). The above analysis of silymarin in the seed of *S. marianum* is of particular importance for pharmaceutical utilization and the plants containing high percentage of silymarin, should be collected from environmentally pure area where no other possibilities of contamination of toxic materials nor having week seeds that will have lower contents of the active components. The collection of the *S. marianum* seeds from area yielding economically high percentage of silymarin will have effect on the marketed product.

Moreover economically poor people can get benefited using this active component for curing various type of hepatitis.

Conclusions

The study regarding the silymarin and oil contents showed that the seeds should be collected from environmentally pure area where it has no possibilities of contamination of toxic materials. However before collection and utilization, the seeds must be checked for other ingredients including bacteria, fungus, heavy metals, and other toxic contaminants in order to make it safe for human consumption.

REFEENCES

Burgess CA (2003). *Silybum marianum* (Milk Thistle). J. Pharm. Soc. Wincons, pp. 38-40.
Flora K, Hahn M, Rosen H, Benner K (1998). Milk Thistle (*Silybum marianum*) for therapy of liver disease. Am. J. Gastroenterol., 93: 139-143.
Khan L, Shafi N, Farooq S, Gilani, SN, Mahmood T, Ahmad N (2003). Pakistan. J. Sci. Ind. Res. 46(4): 307.
Katiyar SK, Korman NJ, Mukhtar H, Agrawal R (1997). Protective effects of silymarin against photocarcinogenesis in a mouse skin model. J. Natl. Cancer Inst., 89: 556-566.
Machicao F, Sonnenbichler JZ (1977). Hoppe-Seyler' Physiololgical. Chem., 358: 141.
Schriewer H, Rauen R, Roth G, Rauen HM (1973). Arzneimittel-Foeschung. Drug-Res., 23: 160.
Sonnenbichler J. Sonnnenbichler, I. Sealkera F (1998). Influence of the flavonolignan silybinin of milk thistle on hepatocytes and kidney cells. ACS Symp. Ser. Phytomed. Eur., 691: 263.
Tawaha K, Alali FQ, Gharaibeh M, Mohammad M, El-Elimat T (2007). Antioxidant activity and total phenolic content of selected Jordanian plant species. Food Chem., 104: 1372-1378.
Valenzuela A, Guerra R, Videla LA (1986). Antioxidant properties of flavonoids silybin and silibin, an active constituent of milk thistle: comparison with silymarin. Cancer Lett., 147: 77–84.
Vogel G (1968). Arzneimittel-Foeschung. Drug-Res., 18: 1063.