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Formulation and stability test of Benjakul extract tablets: a preliminary study

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Abstract

Benjakul is a Thai Traditional medicine preparation, used for balanced health. In our previous study, the ethanolic extract of Benjakul preparation exhibited high cytotoxic activity against COR-L23. In this study, we formulated a Benjakul extract tablet and tested product stability under accelerated condition. A wet granulation method was used in developing the tablets. The suitable excipients were lactose, starch, Explotab® and magnesium stearate. The physical properties of tablets were evaluated following by the USP25 requirements. The results of stability testing found that stability of Benjakul extract tablet related with stability of Benjakul extract from our previous studied. It was indicated that plumbagin is unstable and the cytotoxic activity depend on plumbagin content. Thus, the cytotoxic activity against COR-L23 was also reduced.

Keywords: tablet formulation, stability test, Benjakul extract

Introduction

Benjakul is a Thai traditional medicine preparation which composed of five plants; *Piper longum* fruit, *Piper samentosum* root, *Piper interuptum* stem, *Plumbago indica* root and *Zingiber officinale* rhizome, in equal proportions (1). In our previous study, we found that the ethanolic extract of Bejakul preparation exhibited high cytotoxicity against lung cancer cell lines (COR-L23) with IC_{50} value of 19.80 μg/ml. Two compounds, piperine and plumbagin, were isolated from the extract. Plumbagin exhibited the highest cytotoxic activity against COR-L23 with IC_{50} value of 2.55 μM. The results from stability testing under accelerated condition of Benjakul extract, indicating that plumbagin was unstable (2).

Tablets are solid dosage forms containing medicinal substances that widely used. Plant extracts are often poorly flow ability, low compressible and very hygroscopic. In addition, tablets are appropriately containing high amount of extract show prolong disintegration times, affecting the release of active constituents (3). Thus, wet granulation method often used to improve the properties of products from plant extracts.

From this result, the development of appropriate dosage forms is necessary to increase the stability and to make them suitable for use. Thus, the aims of this study were preliminary study formulation of tablets from Benjakul extract using wet granulation method and investigation the tablet stability under accelerated condition.

Methods

Preparation of Benjakul extract

Plant materials were dried at 50°C and powdered. All plants in equal portion were mixed and extracted by macerated with 95% ethanol for 3 days, filtered and concentrated to dryness under pressure. The marc was macerated 2 times and dried by evaporator.

Tablet formulation

The tablet, containing 50% of extract, was prepared by conventional wet granulation method. Various excipients were screened before the tablet formulation study. According to quality control of product development, the physical properties that consisted of shape, color, weight variation, hardness, disintegration and friability were identified in accordance with the USP25 requirements.

Stability testing

The stability testing was performed according to Thai FDA guideline on stability testing of drug product (4). The tablets, packed in close amber glass containers, were stored under accelerated conditions (45 ± 2°C with 75 ± 5% RH) for 4 months and randomly sampled every 4 weeks interval for analyzing the percent remaining of pipierine and plumbagin contents by HPLC (2) and determining its cytotoxic activity against COR-L23 by SRB assay (5).

Results and discussion

Benjakul extract tablet formulation

From excipients screening, the suitable excipients were used: lactose as a diluent, starch as a binder, Explotab® (sodium starch glycolate and sodium carboxymethyl starch) as a disintegrant and magnesium stearate as a lubricant. Tablets were compressed by single punch machine and adjust weight about 500 mg/tab. Percentages of each ingredient showed in Table 1.

Table 1 Tablet formulation of Benjakul extract by wet granulation.

Ingredients	Percent (%)
Benjakul ethanolic extract	50
Lactose	q.s.
Starch	q.s.
Starch for paste	q.s.
Explotab®	4
Magnesium stearate	1.2

The physical properties of Benjakul extract tablet

Benjakul extract tablets were analyzed by physical properties, the data showed in Table 2. The tablet characteristics were smooth, shiny surface and round in shape. Because of the extracted was dark brown in color, so the tablet was darker brown in color. This prepared tablet had a weight variation of 491.1 ± 9.5 mg, a hardness of 6.7 ± 0.6 kg, a percentage friability of 0.02% and a disintegration of 10.7 ± 1.2 minutes. All the physical properties were allowed in the requirements of the USP 25 standard.

Table 2 Physical properties of Bejakul extract tablet formulation.

Physical properties	Benjakul extract tablet (500 mg/tab)
Shape	Round
Color	Dark brown
Weight variation (mg)	491.1 ± 9.5
Hardness (kg)	6.7 ± 0.6
Friability (%)	0.02
Disintegration time (min)	10.7 ± 1.2

All data are mean±SD as obtained by triplicate analyses.

Stability of Benjakul extract tablet

The results of piperine and plumbagin contenting and cytotoxic activity against human lung cancer cell (COR-L23) were showed in Table 3 and Figure 1. Piperine was slightly reduced (about 15.09% from day 0) and it remained to be 27.71 mg/g after day 120. By the contrast of piperine, plumbagin content was reduced quickly and it could not be detected after day 120. In addition, the IC_{50} value of Benjakul extract tablet was also change from 39.77 µg/ml to be 61.66 and 91.56 µg/ml after day 60 and 120. Its results indicated that cytotoxic activity of this preparation depend on plumbagin content which this compound can evaporate in high temperature.

This result related with stability of Benjakul extract from our previous studied which indicating that plumbagin was unstable but piperine exhibited as a stable compound. It's illustrated that the amount of plumbagin was significantly reduced under high temperature due to its low melting point of 78-79 °C and can be sublimated easily. Therefore, the cytotoxic activity was also reduced due to increasing of IC_{50} value. Thus, the uncoated tablet formulation cannot improve the stability of Benjakul extract.

Table 3 Piperine and plumbagin content (mean ± SD) of Benjakul extract tablet after stored under accelerated condition (n=3)

Day	Piperine content (mg/g)[a]	Plumbagin content (mg/g)[b]
0	30.04 ± 0.17	2.77 ± 0.03
30	29.22 ± 0.70	1.02 ± 0.09
60	26.86 ± 1.58	0.76 ± 0.03
90	28.08 ± 0.14	0.47 ± 0.01
120	27.71 ± 0.57	ND

ND = cannot detected

[a] Calculated as the linear equation: $y = 19388x - 5076.5$, $r^2 = 0.9999$ (**y** = peak area, x = conc. of sample)

[b] Calculated as the linear equation: $y = 28888x - 39265$, $r^2 = 0.999$ (**y** = peak area, x = conc. of sample)

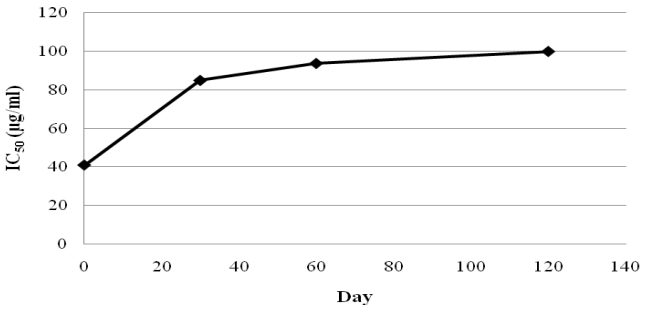

Figure 2 Cytotoxic activity (IC_{50}) against COR-L23 of Benjakul extract tablet after stability testing.

Conclusion

In summary, Benjakul extract tablet was prepared by wet granulation and the suitable excipients were used lactose, starch, Explotab® and magnesium stearate. All the physical properties of tablet were accepted by the requirements of the USP 25 standard. For stability testing, the Benjakul extract tablet was unstable both chemical and biological activities.

Because of its unstable, Benjakul extract tablet should be further developed by using another tabletting technique, such as film coated tablet, to improve the stability of product or avoid the method of preparing tablets which had to used high temperature.

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