Zagazig J. Pharm. Sci., December 2007
Vol. 16, No. 2, pp. 53-62

ISSN 1110-5089

QUALTY CONTROL OF FOUR FINISHED HERBAL DRUG FORMULATIONS MARKEDTED IN SAUDIA ARABIA

Fawkia A. Abbas
Department of Pharmacognosy, Faculty of Pharmacy, Zagazig University, Zagazig, Egypt

ABSTRACT

Quality control of the main constituents was conducted on four finished herbal drug products, one-batch each in soft gelatin capsules manufactured by an American Company including Devil's claw, Valerian, Twinlab powerman Yohimbe power and Korean white Ginseng root. Pharmacopoeial constants and elemental analysis were determined. The results obtained revealed that there is no significant difference from the constants published in different Compendia. The results indicated no adulteration or substitution in these herbal preparations, and no pesticidal residues were detected in all samples under investigation. The results of viable count revealed that all the four herbal preparations were free from pathogenic organisms and the limits of aerobic and yeast not exceed the acceptable limits. The results obtained for the detection of aflatoxins revealed that the fungi isolated from the four herbal preparations showed no fluorescence in UV after growing on Sabauraud medium, the chloroform extracts of the four preparations when subjected to HPLC analysis, confirmed absence of aflatoxins. The obtained Pharmacopoeial values are around the recorded values. HPLC, qualitative and quantitative determinations of the major constituents were in agreement with the reported data.

INTRODUCTION

The quality criteria for herbal drugs are based on a clear scientific definition of the raw material. Depending on the type of preparation, sensory features, moisture, ash, physical constants, solvent residues, and adulterations have to be checked to prove identity and purity. Microbiological contamination and foreign materials, such as heavy metals, pesticide residues and aflatoxins are further criteria to be tested. To prove the constant composition of herbal preparations, adequate analytical methods such as TLC and HPLC have to be applied[1].

The present study dealt with four finished herbal drug products one- batch each in soft gelatin capsules manufactured by an American Company including Nature's Way Standardized Devil's claw Extract (*Harpagophytum procumbens* DC.), Concentrated Valerian Root (*Valeriana officinalis* L.), Twinlab powerman Yohimbe power (*Pausinystalia yohimbe* K. Schum.) and Korean White Ginseng Root (*Panax ginseng* C.A. Meyer.) as examples for finished herbal drug preparations in Saudi Market.

Devil's claw (*Harpagophytum procumbens* DC. Family Pedaliaceae) has a large claw-like fruit and also called Grapple plant..The major uses of Devil's claw are as anti-inflammatory and pain reliever for joint diseases, back pain and headache[2, 3]. Devil's claw is considered medicinal and is preferred by herbalists because of its high concentration of the beneficial component harpagoside[4].

The root of valerian (*Valeriana officinalis* L. Family Valerianceae), a perennial herb native to North America, Asia, and Europe, is used most commonly for its sedative and hypnotic properties in patients with insomnia. Although valerenic acid is responsible for most of valerian's biologic effects, it is likely that all of the active constituents of valerian act in a synergistic manner to produce a clinical response[5].

Yohimbe bark (*Pausinystalia yohimbe* K. Schum. Family Rubiaceae) is name of bark of a tall evergreen tree in Western Africa for fever, leprosy, and coughs[6]. It is used to dilate pupils, for heart disease, and as a local anesthetic. It has a more recent history of use as aphrodisiac and treats male sexual dysfunction[7]. Alkaloid yohimbine is primary active constituent in yohimbe. Yohimbine is shown in several double blind studies to treat men with impotence[8, 9].

Ginseng refers to species within *Panax*. (*Panax ginseng* C.A. Meyer. Family Araliaceae) is used by elderly persons in the Orient to improve mental and physical vitality. Ginseng's action in body are due to a complex interplay of constituents[10]. Primary group is ginsenosides, which are believed to increase energy, counter effects of stress and enhance intellectual and physical performance. Thirteen ginsenosides Rg_1 and Rb_1 have received the most attention[10]. Other constituents include panaxans, which help lower blood sugar, and the polysaccharides which support immune function[11, 12, 13]. Thus, the present study included detection and determination of chemical contaminants, microbial contaminants (viable count, detection of aflatoxins), determination of certain Pharmacopoeial constants, general monitoring for pesticide residues, physical properties, qualitative and quantitative investigations of the major constituents in the four products using TLC and HPLC were undertaken.

EXPERIMENTAL

I- Material:

1- *Pharmaceutical preparations:*

Preparations used in this study were manufactured by an American Company including Devil's claw, Valerian, Yohimbe and Korean white Ginseng , collected from local market in Riyadh city, Saudi Arabia. Sample of one batch number in soft gelatin capsules was used for each of the following preparations:

Standardized Devil's claw extract:

Each capsule contains 350 mg Devil's claw (*Harpagophytum procumbens* DC.) root extract and 130 mg Devil's claw root. Standardized to 1.5 % harpagoside. Manufacture date: 3/1999, Expiry date: 3/2003, Batch number #953106.

Concentrated valerian Root 1000 mg capsules:

Each capsule contains 1000 mg Valerian root (*Valeriana officinalis* L.) (from 125 mg of Valerian root extract 8:1). Manufacture date: 6/1999, Expiry date:7/2003, Batch number # 354536.

Twinlab powerman Yohimbe power:

Each capsule contains 225 mg Yohimbe bark (*Pausinystalia yohimbe* K. Schum.) extract standardized to contain 2% yohimbine alkaloid. Manufacture date : 4/2000, Expiry date: 6/2003, Batch number # 868211.

Ginseng gold, Korean white ginseng root, 500 mg capsules:

Each capsule contains 500 mg powdered *Panax ginseng* C.A. Meyer. standardized to contain 4% ginsenosides. Manufacture date : 3/2000, Expiry date : 5/2003, Batch number # 645335.

2- *Material for microbiological investigation*[14]:

Media for microbiological study:

Mueller Hinton agar, sheep blood agar, Eugongar: reconstituted and sterilized, Sabauraud agar and broth, sterile plastic loop for taking up pure culture, nutrient agar: reconstituted and sterilized, sterile filter paper disc, peptone water: reconstituted and sterilized, Sabauraud yeast extract agar: 20 g glucose, 10 g peptone, 10 g yeast extract, 20 g agar and 1 liter of distilled water.

Microorganism:

Gram positive bacteria: *Staphylococcus aureus* ATCC 6538 , *Bacillus* cereus CECT 232 .

Gram negative bacteria: *Escherichia coli* CECT 99 , *Proteus mirabilis* CECT 170 and *Salmonella veneziana* CECT 450.

Yeast: *Candida albicans* UBC 1.

All these were supplied from Department of Microbiology, Central Laboratory, Riyadh Medical Complex, Riyadh, Saudi Arabia.

3- *Material for chromatography:*

Thin layer chromatography was performed using precoated silica gel G 60 F_{254} plates 20x20 cm (E. Merck, Darmstadt, Germany).

4- *Reference materials:*

Harpagoside, valerenic acid, acetoxyvalerenic acid, hydroxyvalerenic acid, yohimbine HCl, ginsenoside Rb_1 and ginsenoside Rg_1 (Phytolab GmbH & Co-KG, Labor, Addipharma, Wandalenweg 24, 20097-Hamburg, Germany).

II- Apparatus:

1- **Soxhlet apparatus:** For continuous extraction.

2- **Atomic absorption spectrometric method**[15]: Two g of the powder of each of the four herbal pharmaceutical preparations were separately weighed in porcelain crucibles and incinerated for 2 hours at 500 °C, then let to cool . Ash was wet with 10 drops of water and 3-4 ml 50% nitric acid were added. The excess nitric acid was evaporated on a hot plate at 100-120 °C. The crucibles were incinerated an additional one hour at 500 °C, cooled and the ash was dissolved in 10 ml of 50% hydrochloric acid and quantitatively transferred to 100 ml volumetric flasks.

Atomic Absorption Spectrometer Model 680 (Shimadzu, Japan) with flameless and HVG-Option, was used with the following conditions:

For arsenic (As): Lamp current: 6 mA, Slit: 6 nm, Wavelength : 193.7 nm , Mode : (BGC) Back Ground Correction Mode HVG-1 , Model for Hydride Vapor Generator Unit from Shimadzu , Working standard : 1 , 2 & 3 ppb.

For lead (Pb) : Lamp current : 7mA , Slit: 0.3 nm , Wavelength : 217 nm , Mode : (BGC) Back Ground Correction Mode HVG-1 , Flame's Condition : Furnace GFA 4B , Shimadzu , plain or uncoated graphite tube , Temp. Condition : Atomization temperature 1200° C .

For Mercury (Hg) : Lamp current : 2 mA , Slit : 0.7 nm , Wavelength : 253.7 nm , Mode : (BGC) Back Ground Correction Mode HVG-1, Model for Hydride Vapor Generator Unit from Shimadzu , Working standard : 10 , 20 &30 ppb .

For Cadmium (Cd) : Lamp currernt : 3 mA , Slit : 0.2 nm , Wavelength : 326 nm , Mode : (BGC) Back Ground Correction Mode HVG-1, Flame's Condition : Furance GFA 4B , Shimadzu , plain or uncoated graphite tube, Temp. Condition : Atomization temperature 1200° C .

For Nickel (Ni) : Lamp current : 4 mA , Slit : 0.2 nm , Wavelength : 341.5 nm , Mode : (BGC) Back Ground Correction Mode HVG-1, Flame's Condition : Furance GFA 4B , Shimadzu , plain or uncoated graphite tube, Temp. Condition: Atomization Temperature 1200 ° C.

3- Apparatus and condition for HPLC analysis:

High Performance Liquid Model SCL-10 AD *VP*- Shimadzu Auto-controller, SCL-10A *VP* – Shimadzu System Controller, CTO-10A *VP*- Shimadzu Column Oven LC- 10A *VP*- Shimadzu, Liquid Chromatography, DGU-12A Shimadzu Degasser. SPD-10A *VP*- Shimadzu UV-Vis Detector. Water Corp., Millford Massachusetts (1996), using Photodiode detector at 220, 240, 565 and 590 nm. Each determination was repeated three times.

For harpagoside: isocratic elution at 30° C . using methanol- water (60-40) as mobile phase with flow rate 1ml /min using octadecylsilyl silica gel (5 µm), type HYPERSIL BDS (0.25 m x 4 mm) as column and a spectrophotometeric detector at 278nm.

For Ginsenosides (Rb_1+Rg_1): gradient elution at room temperature using acetonitrile – water (90:10, 80:20) as mobile phase with flow rate 1ml/min using Aminopropylsilyl silica gel (0.1 m x 4.5 mm) as column and UV detector at 203 nm.

For sesquiterpene acids (calculated as Valerenic acid): isocratic elution at 30 °C using a mixture of a 5 g/L solution of phosphoric acid-methanol (30:70) as a mobile phase with flow rate 1ml /min using octadecylsilyl silica gel (5 µm), type SPHERISORB ODS2 (0.25 m x 4 mm) as a column and a spectrophotometer detector at 225 nm.

For yohimbine HCl : isocratic elution at room temperature using methanol - 0.005M octan sulfonic acid salt (50:50) as a mobile phase with flow rate 1ml /min using SUPEKOSIL LC-18 (0.25m x4mm) as a column and a UV detector at 254 nm.

III- Method:

Preparation of sample for total aerobic bacterial and fungal count:

These samples were prepared according to methods reported in B.P.[16].

Zagazig J. Pharm. Sci., December 2007
Vol. 16, No. 2, pp. 53-62
ISSN 1110-5089

Preliminary detection of aflatoxins - producing fungi:

Isolated fungi from each product (on the Sabauraud dextrose agar in the step of fungal count determination) were screened for their ability to produce aflatoxin on Sabauraud yeast extract agar plates. Each of the isolated molds was inoculated as a single short streak at the center of the surface of the plate The plates were then incubated 7 days at 25-28 °C and examined under UV light (366 nm), the presence of any fluorescence in the medium surrounding the fungal growth was recorded.

Quantitative estimation of aflatoxins[17]:

50 g from each herbal preparation were separately extracted into 500 ml Erlenmeyer flask by adding 25 ml water, 25 g diatomaceous earth and 250 ml chloroform, and secure stopper with masking tape. Shaked and filtered. The chloroform filtrate was subjected to HPLC analysis using suitable conditions.

HPLC condition for detection of aflatoxins:

High Performance Liquid Chromatography Model SCL-10 AD *VP*-Shimadzu, Japan, Autocontroller, SCL-10A *VP*-Shimadzu Japan, System Controller,CTO-10A *VP*-Shimadzu Column Oven LC-10 A *VP*- ,Shimadzu Liquid Chromatography DGU-12A Shimadzu Degasser.

Stationary support: Silica size 5µM, Bonded phase: Octadecylsilyl, proper size 100A, Frit pore size: 2µM. Isocratic elution at room temperature using methanol – acetonitrile – water (17.5 : 17.5 : 65) as mobile phase. The flow rate was 1 ml/min and injection volume 20 µl. Aflatoxins were detected using scanning fluorescence detector.

Monitoring for pesticide residues:

A weight of 1g of each sample under investigation was extracted separately in a Soxhlet using acetone: hexane mixture (1:1) at 60 °C for 6 hours. Each solution was filtered, dried (anhydrous sodium sulphate). and evaporated under reduced pressure) to 1 ml. The extracts of the four preparations were subjected to GC/MS analysis under the following conditions: Column used: Rtx®-1 MS (Crossbond® 100% dimethyl polysiloxane) 30 m, 0.25 mm ID (Internal diameter), 0.25 µm df (diameter of film around the crystal), cat.# 11623 serial # 20412, RESTEK, UK Ltd.

GC Condition: Carrier gas helium, at flow rate 0.5 ml/min., injection temp. 280 °C., interface temp. 230 °C, linear velocity 25 cm/sec, total flow 25.7 ml/min.

Preparation of sample for TLC:

Methanol (50ml) has been used for extraction of the corresponding constituents of the four herbal preparations (1g each) at room temperature for six hours. Each extract was evaporated separately under reduced pressure (40 °C),each residue was re-dissolved in methanol and kept at 4 °C in sealed vials for TLC analysis.

Thin-layer chromatography:

Each extract and reference solutions were applied as bands on precoated 20 x20 TLC silica gel 60 F_{254} plates. Developments were performed by ascending mode at room temperature in different mobile phases. Visualisation of the compounds was achieved by spraying the plates with vanillin- sulphuric acid [18], anisaldehyde sulphuric acid [19] followed by heating at 110 °C for ten minutes in both cases .

Standards preparation and calibration curves:

About 10mg of each harpagoside, ginsenoside Rb_1, ginsenoside Rg_1, valerenic acid , acetoxyvalerenic acid , hydroxyvalerenic acid and yohimbine- HCl reference standards were accurately weighed into a 10-ml volumetric flasks , add HPLC grade methanol (10ml) to each reference and sonicate until dissolve and mix. These1000 ppm standards (i.e. 1mg/ml) were stored at 4 °C and used for the calibration curves. Suggested standard dilution of each stock are 1:10, 1.5:10, 2:10, 2.5:10 using methanol. For each standard, 10 µl from each dilution were injected and the concentration were plotted against the resulted corresponding area.

Sample preparation for HPLC analysis:

1- ***For harpagoside in Devil's claw soft gelatin capsules,*** the content of two capsules were macerated in 200ml methanol (HPLC) for 24 hours, then filtered. A volume of 100 µl of the prepared extract was injected into HPLC.
2- ***For ginsenosides (Rb_1&Rg_1) in Korean white Ginseng soft gelatin capsules,*** the sample was prepared by refluxing 2 gm of powdered Korean White Ginseng capsulated herbal preparation with 200 ml methanol (HPLC) for one hour. The methanolic solution was filtered and concentrated to 20 ml volume under reduced pressure at a temperature not exceeding 60 °C. A volume of 20 µl was injected into HPLC.
3- ***For valerenic acid in Valerian soft gelatin capsules,*** the sample was prepared by macerating the content of two capsules in 100 ml of methanol (HPLC) for 24 hours, then filtered, concentrated under reduced pressure and 10 µl was injected into HPLC.
4- ***For yohimbine HCl in Twinlab Powerman Yohimbe Power capsules,*** the sample was prepared by macerating the content of two capsules in 50 ml methanol for 30 minutes, shake then filtered and inject 20 µl into HPLC.

RESULTS AND DISCUSSION

1. Detection of chemical contaminants:

Certain microelements of known reported toxicity (As, Hg, Ni, Pb, Cd) were determined in the four herbal preparations under investigation, using atomic absorption method[20]. From table (1), it could be concluded that the level of As, Hg, Pb, Cd are within the allowed levels[20].

Table (1): The concentration of microelements in the four herbal preparations (ppm)

Herbal Product	Arsenic (As)	Mercury (Hg)	Nickel (Ni)	Lead (Pb)	Cadmium (Cd)
Max. Allowed[14]	1-5 PPm	0.03 PPm	1 ppm	1.2 ppm	0.05 ppm
Devil's claw	0.0436	0.013	0.012	0.412	0.002
Valerian	0.015	0.018	0.032	0.02	0.001
Yohimbe	0.0951	0.02	0.016	0.32	0.008
Korean white Ginseng	0.0375	0.015	0.071	0.14	0.004

2. **Detection of microbial contaminants by viable count[21]:**

The results obtained (table 2) revealed that all the four preparations were free from pathogenic organisms and the limits of aerobic and yeast did not exceed the acceptable limit.

Table (2): Viable count of the four herbal preparations

Sample	Bacterial Count (Cfu/g)		Fungal Count (Cfu /g)	
	Total count	Potential pathogen	Yeast	Total viable count
Accepted limits[22]	$<10^2$*	Nd*	10	10
Devil's claw	10	negative	10	10
Valerian	10	negative	10	10
Yohimbe	10	negative	10	10
Korean white Ginseng	10	negative	10	10

cfu = culture forming unit, nd = not detectable
potential pathogen = *E. coli* and *Salmonella veneziana*
* = per g or ml

3. **Detection of aflatoxins:**

The chloroform extracts of the four herbal preparations were subjected to HPLC analysis, which confirmed the absence of aflatoxins compared with standard aflatoxins.

4. **Determination of certain pharmacopoeial constants:**

Total ash, water-soluble ash, acid-insoluble ash as well as crude fibers, moisture contents, alcohol-soluble and water soluble extractive values were determined in the four herbal preparations according to the British Pharmacopoeia[16].

The obtained values (table 3) were found in accordance with the recorded values in different compendia[23, 24], which indicate that there is no adulteration or substitution in these herbal preparations.

Table (3): Results of certain pharmacopoeial Constants of the four herbal preparations

Material	Total ash	Water soluble ash	Water insoluble ash	Acid soluble ash	Acid insoluble ash	Crude fibers	Moisture content	Alcohol soluble extractive value	Water soluble extractive value
Devil's claw	4.6%	1.6%	3 %	3.7%	0.9%	0.9%	6.6%	4%	13.5%
Valerian	7.8%	1.9%	5.9%	6.4%	1.4%	1.03%	1.2%	3.1%	6.01%
Yohimbe	10.2%	1.6%	8.6%	7.8%	3.1%	2.3%	4.2%	3.1%	8.3%
Korean white Ginseng	6.9%	2.1%	4.8%	4.4%	1.1%	2.2%	5.2%	2.98%	11.4%
Max. Allowed[22]	10% in Devil's claw , 12 % in Valerian & 7 % in Korean white Ginseng				5% in Valerian & 1 % in Korean white Ginseng		12 % in both Devil's claw and Valerian but 10 % in Korean white Ginseng		

ISSN 1110-5089

5. **General monitoring for pesticide residues:**

In the present study, GC-MS analysis indicated no pesticidal residues[17] [Methane, bromotrifluoro, 2-pentanone, 4-hydroxy-4-methyl, Di-n-octylphythalate, 1,2-benzendicarboxylic acid , ditridecyl ester and diisooctyl ester, Bis(2-ethylhexyl) phethalat, cycloheptane, 1,3,5,-tri-(methylene), 2H-pyrane-2-one ,5,6-dihydro-4-methoxy-6-(2-phenyl) , 1,3 cyclohexadiene, 5 (1,5-dimethyl-4-hexenyl)-2-methyl undecane, 1-bromo) in all samples under investigation].

6. **Qualitative microscopy:**

Physically, each capsule was examined separately for its colour, odour and taste. The results obtained indicate normal colour, odour and taste for each capsule, which indicate that there is no adulteration or substitution in these herbal preparations.

Table (4): Results of qualitative microscopy of the four herbal preparations

Tests	Devil's claw	Valerian	Yohimbe	Korean White Ginseng
Appearance	Translucent capsules	Translucent capsules	Translucent capsules	Oblong soft gelatin capsules
Colour	Caramel brown	Caramel	Dark brown	Yellowish-brown
Content	Beige fine powder	Pale beige fine powder	Dark brown powder	Brown powder
Odour	Slight characteristic	Strong characteristic	Strong characteristic	Strong characteristic

7. **Thin-layer chromatography:**

Devil's claw:

Glucoiridoids of Devil's claw were developed with glacial acetic acid: water: butanol (10:10:40) and examined in ultraviolet light at 254 nm , it shows 3 quenching zones , one of which is similar in position to the zone of reference harpagoside solution . Spraying the chromatogram with vanillin reagent followed by heating at 110 °C for 10 min., shows 3 purplish- pink zones, one of which is similar in position and colour to harpagoside reference solution.

Valerian:

Ethyl acetate – hexane (30:70), gave separation of alcoholic extract of valerian. The chromatogram when sprayed with anisaldehyde solution followed by heating at 110°C for 10 min., shows a dark –purple zone similar in position and colour to the zone obtained with valerenic acid , above this zone a grey-brown zone is present (hydroxy valerenic acid) . It also shows a purple zone (acetoxyvalerenic acid).

Korean white Ginseng:

Ethyl acetate –butanol - water (1:4:5) was used to identify the presence of Rb_1 and Rg_1 in Korean White Ginseng . When the chromatogram was sprayed with anisaldehyde solution followed by heating at 110 ^{o}C for 10 min., shows a violet zone similar in position and colour to the zone of Rb_1 reference ,above this zone another violet zone similar in position and colour to Rg_1 reference was also present.

Yohimbe:

Chloroform- acetone–diethylamine (5:4:1), gave separation of alcoholic extract of yohimbe. The chromatogram when sprayed with anisaldehyde solution followed by heating at 110°C for 10 min, shows a violet zone similar in position and colour to the zone of yohimbine-HCl reference solution.

8. **HPLC determination of the main active constituents of the four herbal preparations[23]:**

HPLC analysis of the methanol extract of Devil's claw on octadecylsilyl silica gel column is shown in figure (2). Identification of harpagoside in the extract was confirmed by comparing with authentic harpagoside (figure 1). Quantitative content of harpagoside in the sample was obtained by applying the following equation :

$$X = \frac{Y - A}{B} = \frac{205549 - 2091.5}{119539} = 1.7\%$$

Where: X = conc., Y = peak area, A = intercept and B = slope.

The harpagoside content of Devil's claw capsules (1.7%) was higher than the given specification (1.5%).

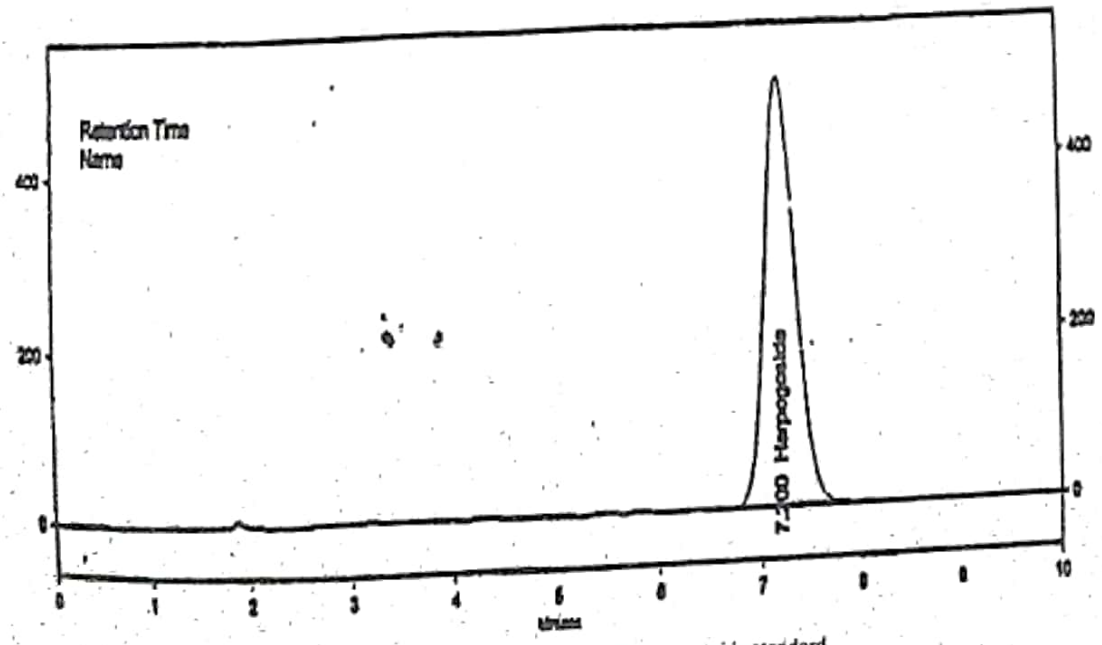

Figure (1): HPLC chromatogram of Harpagoside standard

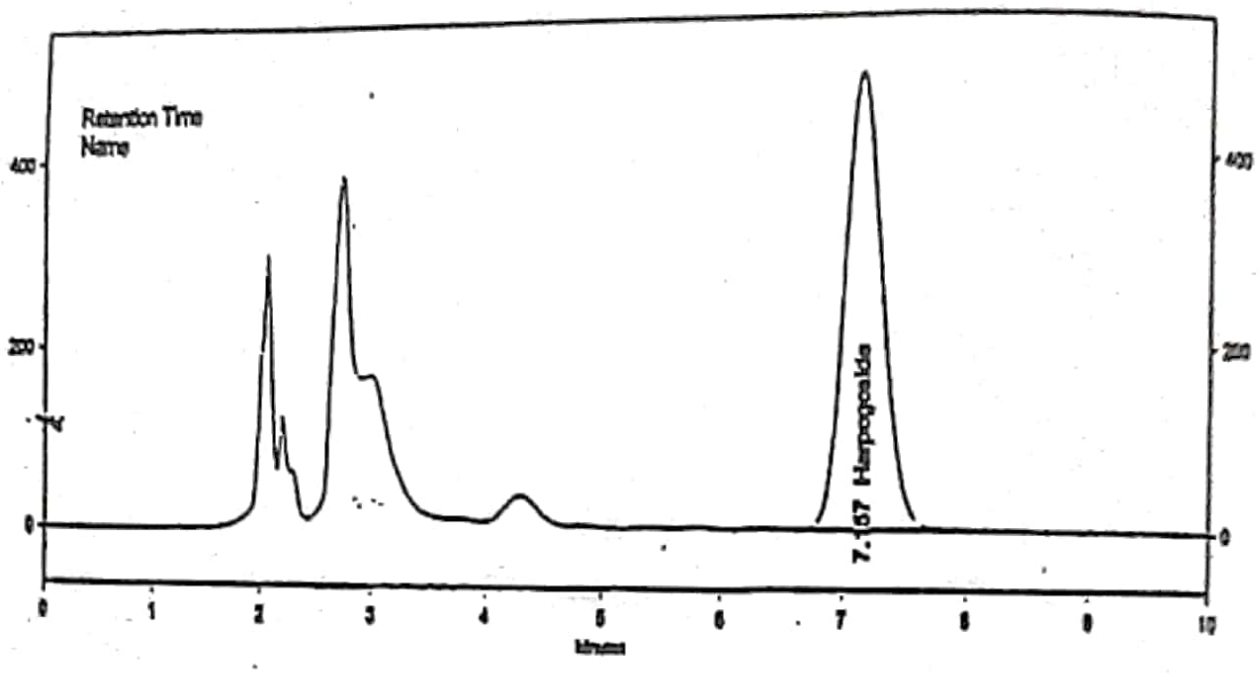

Figure (2): HPLC chromatogram of Devil's claw capsules

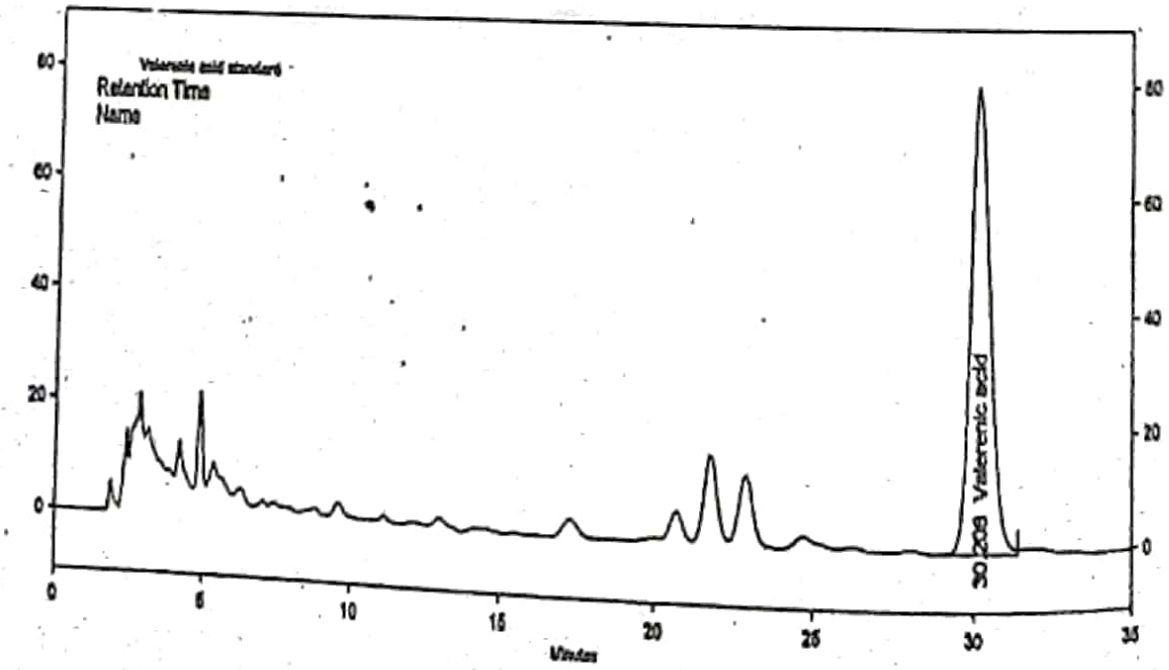

Figure (3): HPLC chromatogram of Valerenic acid standard

ISSN 1110-5089

HPLC analysis of sesquiterpene acids (expressed as valerenic acid) in Valerian herbal preparation (figure 4) shows peaks corresponding to valerenic acid, acetoxy valerenic acid and hydroxyvalerenic acid standards. The content of sesquiterpene acids expressed as valerenic acid was calculated from the formula:

$$\frac{\sum A2 \times m1 \times 100}{A1 \times m2} = \frac{10779095 \times 10 \times 100}{1000 \times 5046369 \times 2} = 1.06$$

M_1: Mass of the valerenic acid in the reference solution, in grams

M_2: Mass of the capsule contents sample in the test solution in grams

A_1: Area corresponding to valerenic acid in the chromatogram obtained with the reference solution

$\sum A_2$: Sum of the areas of the three peaks corresponding to valerenic acid, acetoxyvalerenic acid and hydroxyvalerenic acid.

The calculated percentage of the sesquiterpene acids in the sample (expressed as valerenic acid) was 1.06% and there is no specified value for sesquiterpene acid on the product for comparison.

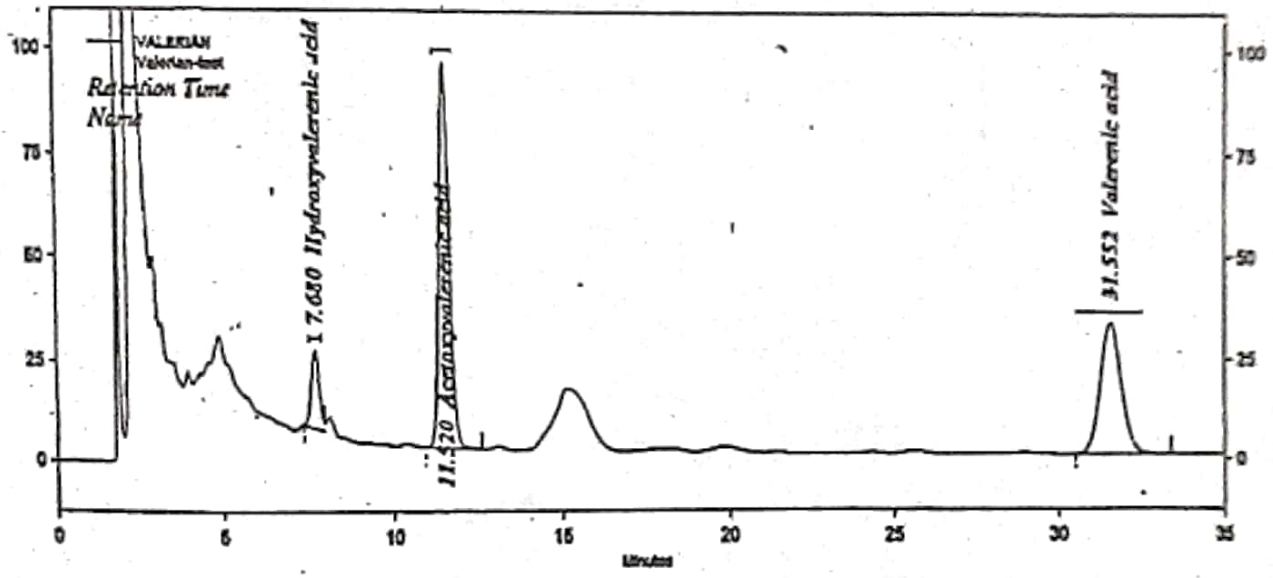

Figure (4): HPLC chromatogram of valerian capsules

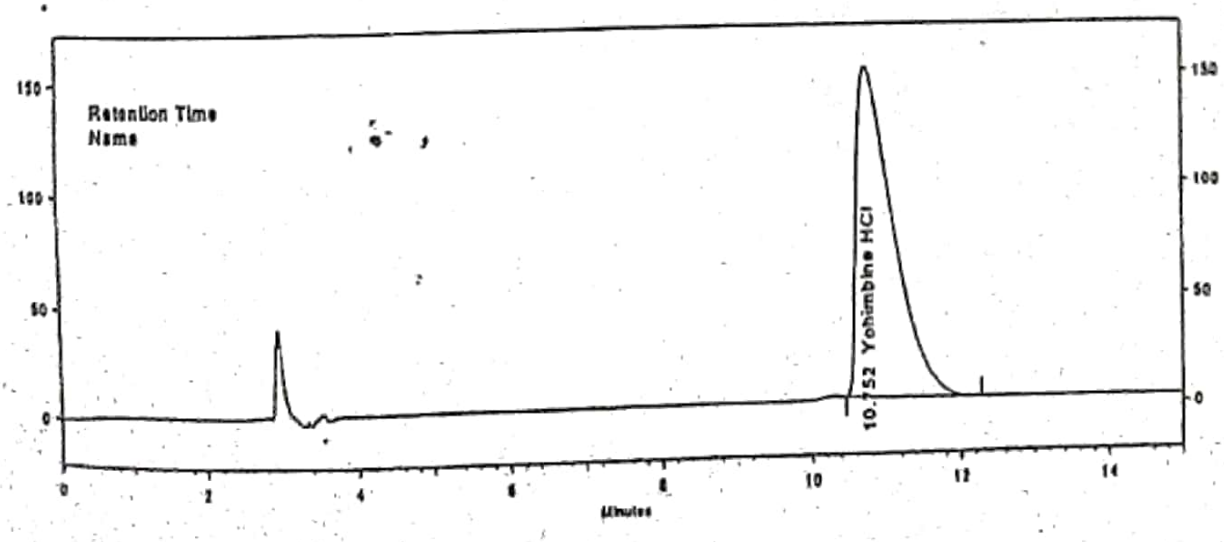

Figure (5): HPLC chromatogram of Yohimbine HCl standard

HPLC analysis for Twinlab powerman Yohimbe power (figure 6) shows peak corresponding to standard yohimbine HCl . The calculated percentage of yohimbine HCl in the sample was calculated from the formula:

$$X = \frac{Y - A}{B} = \frac{7262 - 2091.5}{2189} = 2.3\%$$

The calculated percentage of yohimbine HCl in the sample was 2.3% which complies with the manufacturer's specifications.

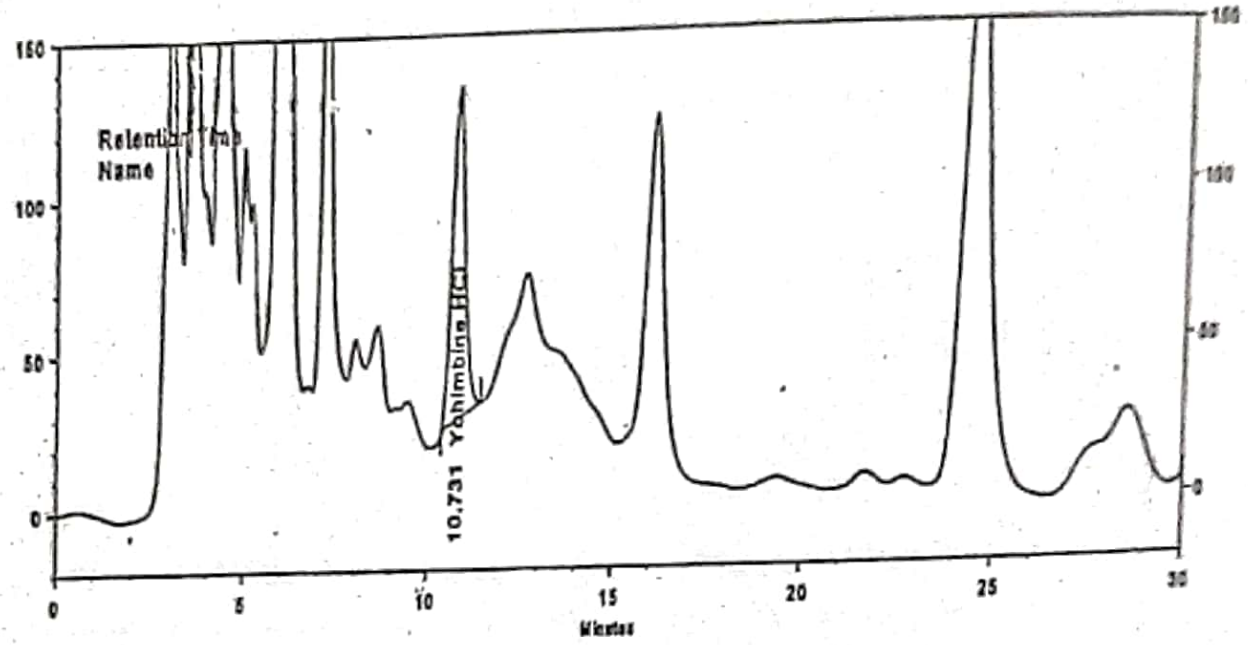

Figure (6): HPLC chromatogram of Twinlab powerman Yohimbine power capsules

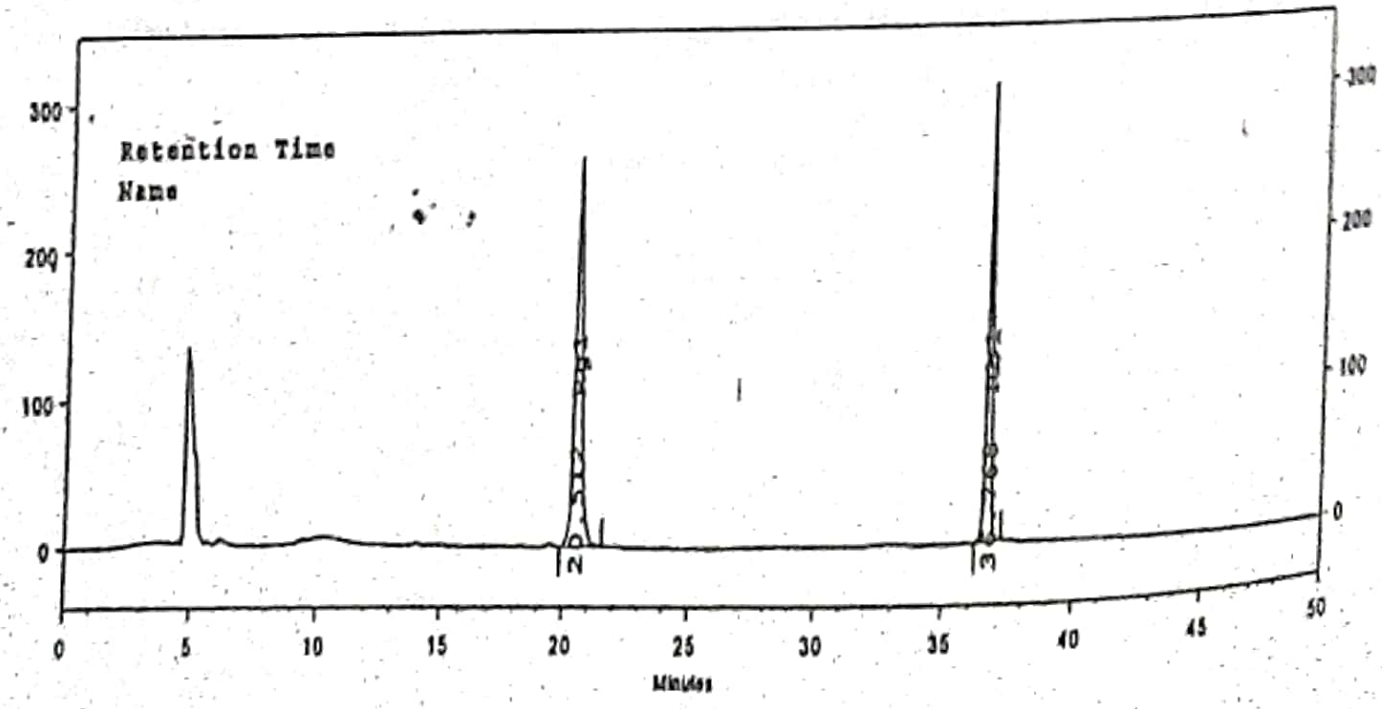

Figure (7): HPLC chromatogram of Ginsenoside Rb_1 and Rg_1 standards

ISSN 1110-5089

HPLC analysis of ginsenosides Rb_1 and Rg_1 in Korean white Ginseng (figure 8) was performed using UV detector at 203 nm.

The % content of Rg_1 + Rb_1=

$$\frac{A1 \times m2 \times 40}{ARb1 \times m1} + \frac{A2 \times m3 \times 40}{ARg1 \times m1}$$

$$= \frac{642720 \times 10 \times 40}{1000 \times 1 \times 82685} + \frac{100550 \times 10 \times 40}{1000 \times 1 \times 18990} = 4.21\%$$

Where: A_1 is Area of the peak due to ginsenoside Rb_1 in the chromatogram of test solution, A_2 is Area of the peak due to ginsenoside Rg_1 in the chromatogram of the test solution, $A\ Rb_1$ is Area of the peak due to ginsenoside Rb_1 in the chromatogram of the reference solution, $A\ Rg_1$ is Area of the peak due to ginsenoside Rg_1 in the chromatogram of the reference solution, m_1 is Mass of the drug to be examined , in grams, m_2 is Mass of ginsenoside Rb_1, in grams and m_3 is Mass of ginsenoside Rg_1 , in grams.

The calculated percentage of the total ginsenosides in the sample was 4.21 % which is close to the recorded value (4%).

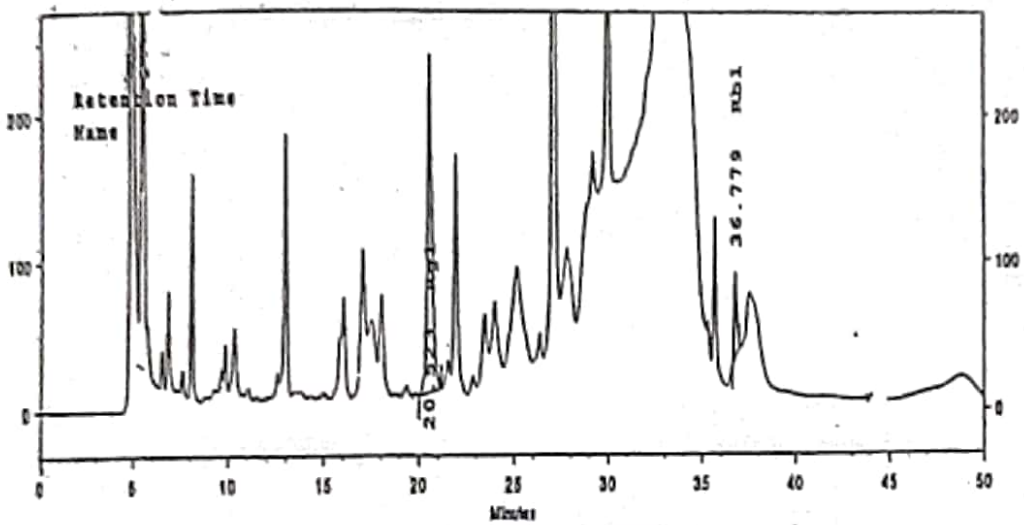

Figure (8): HPLC chromatogram of Korean White Ginseng capsules

CONCLUSION

The result obtained in the four herbal preparations were in agreement with the reported specified data, so these herbal preparations can be used safely in Saudi market.

REFERENCES

1- Bauer R: Drug Inform. J.; 32. 101-110 (1998).
2- Grant L, McBean DC and Fyfe L: Phytother Res; 21(3): 199-209 (2007).
3- Chrubasik S, Model A and Black A: Rheumatology; 42: 141-148 (2003).
4- Chrubasik S, Sporer F and Wink M: Forsch Komplmentarmed; 3: 6-11 (1996).
5- Houghton PJ: J. Pharm. Pharmacol.; 51: 505-12 (1999).
6- Duke J: CRC Handbook of Medicinal Herbs. Boca Raton, FL, CRC Press,; 351 (1985).
7- Adeniyi AA, Brindley GS and Pryor JP: Asian J. Androl.; 9(3): 403-7 (2007).
8- Ernst E and Pittler, MH: J. Urol.; 159(20): 433-36 (1998).
9- Carey MP and Johnson BT: Arch. Sex. Behav.; 25(4): 341-360 (1996).
10- Shibata S and Tanaka O: Chemistry and Pharmacology of *Panax* in Economic and Medicinal Plant Research, ed. Wagner H, Hikino, Farnsworth NR. London, Academic Press; 1: 217-84 (1985).
11- Tomoda M and Hirabayashi K: Biol. Pharm. Bull., 16: 1087-90 (1993).
12- See DM, Broumand N and Sahi L: Immunopharmacol.; 35: 229-35 (1997).
13- Yun TK, Lee YS and Lee YH et al.: Journal of Korean Medical Science; 16(5): 6-18 (2001).
14- Merck E: Culture Media. Merck E. Handbook, 1600-Darmstadt, Germany (1981).
15- Official Method of Analysis: 14th ed. Association of Official Analytical Chemists, Inc., Arlington, Virginia, USA; 40 (1984).
16- British Pharmacopoeia: HMSO. London, UK; 351-751 (2000).
17- Official Methods of Analysis .Association of Official Analytical Chemists. Inc., Arlington, Virginia, USA, 15th ed.; 11: 283-89 (1990).
18- Stahl E: Thin layer chromatography. 2nd ed. Springer-Verlag, Berlin; 856-904 (1969).
19- Kricheusky D and Tropper SA: J. Chromatography; 37: 361-76 (1968).
20- Phytochemeco database-USDA-ARS-NGR. www.arsgrin.gov ( ).
21- Baker FJ and Breach MR: Medical microbiology techniques. Butterworts, London, Boston, Sydney, Wellington, Durban, Torento; 423 (1980).
22- British Herbal Pharmacopoeia, Exeter, UK; 87: 118-204 (1996).
23- British Pharmacopoeia: HMSO. London; 639: 952-2134 (2007).
24- Gruenwold J, Brendler and Jaenicke C.: PDR for herbal medicine. 2nd ed. Medical Economics Company. Montvale, NJ, USA; 331-925 (2000).

Received: August 01, 2007
Accepted: September 17, 2007

دراسة الجودة النوعية لأربعة مستحضرات عشبية في السوق السعودي

فوقية عبد الله عباس

قسم العقاقير – كلية الصيدلة – جامعة الزقازيق – الزقازيق – مصر

تم اختيار أربعة من المستحضرات العشبية المعبأة في كبسولات جيلاتينية رخوة كأمثلة على المستحضرات ذات الأصل النباتي الموجودة في السوق السعودي ، و هذه المستحضرات منتجة بواسطة مركز الفيتامينات و الأعشاب الطبيعية (مقرها الولايات المتحدة الأمريكية) ، و هي على النحو التالي : كبسولات ديفلز كلو و كبسولات فاليريان و كبسولات يوهيمب و كبسولات جنسينج . و قد تم إجراء اختبارات الرقابة على الجودة على هذه المستحضرات ، و فيما يلي ملخص لهذه الاختبارات و نتائجها :

1. **تعيين المعدنات الكيميائية :**

تم تعيين بعض المعادن الثقيلة باستخدام جهاز الامتصاص الذري الطيفي ، و أظهرت النتائج أن نسبة الزرنيخ و الرصاص والنيكل و الكادميوم في جميع المستحضرات في حدود النسبة المسموح بها عالميا .

2. **تعيين التلوث الميكروبي عن طريق عد الأحياء الدقيقة :**

تم تعيين عدد البكتيريا و الفطريات المسببة للأمراض في جميع المستحضرات المختارة . و قد أثبتت النتائج خلو جميع المستحضرات من البكتيريا المسببة للأمراض و أن مستوى البكتيريا الهوائية و الخميرة لم تتعد القيم المسموح بها .

3. **تعيين الافلاتوكسينات :**

تم اختبار قدرة الفطريات على إنتاج الافلاتوكسينات و تعيين المحتوى الفطري للأعشاب ، و قد تبين خلو جميع المستحضرات العشبية الأربعة من الأفلاتوكسينات .

4. **تعيين بعض القيم الدستورية :**

تم تعيين القيم الدستورية (نسبة الرماد ، الرماد الذائب في الماء ، و الرماد غير الذائب في الماء ، و الرماد الذائب في الحمض ، و الرماد غير الذائب في الحمض ، تعيين الألياف ، تعيين نسبة الرطوبة ، تعيين نسبة المواد الذائبة في الكحول ، و تعيين نسبة المواد الذائبة في الماء) في المستحضرات العشبية الاربعة طبقا للطريقة الموجودة في دستور الأدوية البريطاني ، و كانت النتائج مماثلة للقيم الموجودة في الحيد من الابحاث المنشورة المختلفة ، و التي أثبتت عدم وجود غش تجاري أو استبدال في هذه المستحضرات ذات الأصل النباتي .

5. **تعيين بقايا المبيدات الحشرية :**

وجد أن العينات خالية من المبيدات تحت الدراسة ، و لكن ليس معنى هذا أن الشركة الصانعة لم تستخدم مبيدات في الزراعة، و لكن ربما استخدمت مبيدات ذات نصف عمر قصير ، فتحللت بسرعة و لم تظهر في التحليل .

6. **التقييم الكيفي المجهري :**

تم فحص لون و طعم و رائحة جميع المستحضرات ، و أوضحت النتائج أن لون و طعم و رائحة جميع المستحضرات المختارة طبيعية، مما يدل على عدم وجود غش تجاري أو استبدال في هذه المستحضرات .

7. **الكشف عن المواد الفعالة الموجودة في المستحضرات الأربعة باستخدام كروماتوجرافيا الطبقة الرقيقة :**

تم إجراء كروماتوجرافيا الطبقة الرقيقة للمكونات الاساسية الموجودة في المستحضرات الأربعة بعد استخلاصها بالمذيبات المختلفة ، و أثبتت النتائج وجود المكونات الاساسية للمستحضرات بعد مقارنتها مع عينات مرجعية أصيلة .

8. **تعيين المواد الفعالة للمستحضرات الأربعة باستخدام كروماتوجرافيا السائل ذي الأداء العالي :**

تم استخدام كروماتوجرافيا السائل ذي الأداء العالي للتقييم الكمي للمواد الفعالة الأساسية في المستحضرات الاربعة مثل تعيين نسبة الهربلجوزيد في ديفلز كلو ، و تعيين نسبة حمض الفاليرينك في الفاليريان ، و تعيين نسبة الجنسينوزيد Rb_1 ، Rg_1 في الجنسينج ، و تعيين نسبة اليوهيمبين-هيدروكلوريد في كبسولات اليوهيمب ، و أوضحت النتائج أن نسبة المواد الفعالة مطابقة للنسب المحددة للشركة الصانعة في جميع المستحضرات المختارة .

و استنادا إلى النتائج المختلفة التي تمت في هذا البحث يمكن استخدام هذه المستحضرات العشبية في السوق السعودي .