# CHAPTER-6

# Summary & Conclusion

## Summary & Conclusion:

As per World Health Organization: Traditional medicine is "the knowledge, skills and practices based on the theories, beliefs and experiences indigenous to different cultures, used in the maintenance of health and in the prevention, diagnosis, improvement or treatment of physical and mental illness". The core use of herbal medicines is for health promotion and treatment for chronic, as different to life-threatening, conditions. However, usage of traditional remedies rises when conventional medicine is ineffective in the treatment of disease like obesity.

Consistent and safe herbal product for weight reduction is a need of developed and developing countries. In our literature survey, herbal plants showed potential effects on weight control. However, for the majority of products, more data are needed to assess the suitability as anti-obesity plants**.**

Everyone knows that exercise with a controlled diet is the only way to keep in shape. However, your aim to be slim is obstructed by your urge to eat more and to snack in between meals. It's difficult for many people to resist food or snacks after a long and tiring work day. It's only natural! Tiredness and fatigue can also make people crave sugary food for energy. That's why many are unable to stick to healthy food choices every day. But now imagine the same situation with a controlled appetite. With your appetite under your control, you can lower the intake of calories.

*To overcome the side effects of the current allopathic treatment, Herbal will be seen as the alternative medication.*

Obese persons are preferred the use of herbal products for weight management because of following probable reasons:

- ✓ Health benefits of weight loss without any side effects,

- ✓ Less demanding than accepted lifestyle changes, such as exercise and diet,

- ✓ Easily available without a prescription,

- ✓ More easily accepted than a professional consultation with a physician or a nutritionist

- ✓ 100% natural origin and perception that natural means safe

- ✓ Herbal plants for weight reduction may be effective in the treatment of obesity and associated disorders.

Herbal medicines are playing a significant role in preservation of health in rural and remote areas, and provide the health for all. Utilization of Herbal medicine along with conventional drug defiantly helps health or cure diseases in the positive way. Herbal Drug contains the lots of Chemical moieties, which will help to inhibit the whole pathway of lipid synthesis.

Herbal plant selected based on their individual targets for reduction of obesity. The pathogenesis of obesity involves regulation of calorie utilization, appetite, and physical activity, but has complex interactions with availability of health-care systems, the role of socio-economic status, and underlying hereditary and environmental factors.

*Achyranthus aspera(*Aghedo seeds)*, Garcinia Indica fruit (Kokam fruits)*, *Murraya koeinigii (*Curry patta)and *Commiphora mukul* (Guggul) extracts having the known Anti-obesity activity.

Individual selected plants are having their own mechanism to treat the obesity.

*Achyranthes aspera* seed can inhibit obesity by reducing the excess accumulation of body fat and altering the serum lipid profile. This is because of marker compound like oleanolic acid. In Ayurveda mention that consumption of seeds of achyranthes aspera is suppress the appetite. And that's how used to treat the obesity.

Extracts of *Murraya koenigii* leaves significantly reduced the body weight gain, plasma total cholesterol (TC) and triglyceride (TG) levels significantly due to presence of various alkaloids.

*Garcinia Indica* Besides its efficacy in the decrease of body weight and food intake, Garcinia has been proven to be beneficial in ameliorating obesity-related problems such as inflammation, oxidative stress, and insulin resistance. Hydro citric acid and other reported phenolic compounds predicted as reduce the obesity.

*Commiphora mukul* has been used in Ayurvedic medicine for many years to treat a variety of ailments like obesity, lipid disorders, rheumatoid arthritis,  bone fracture, cardiovascular disorder disease and antihyperglycemic and antioxidant activities.

Phytochemical screening was done to check the presence of various primary and secondary metabolites likes proteins, carbohydrates, flavonoids, alkaloids and glycosides.

Quantitative analysis was also done to verify the presence of various amounts of flavonoids and phenolics compounds. As we know that this flavonoids and phenolics compounds can play an important role as antioxidant. These present antioxidants having significant role in the treatment of obesity.

Here Tablets were prepared by Direct Compression method. Direct compression method is the best suitable method for developing herbal tablet.

The whole study was focused on the development of the formulation via using DOE with the appropriate concentration of the different excipients which, passes all preliminary and secondary parameters of the formulations like Hardness, Friability and Disintegration time.

Avicel PH 102, Syloid, Cross carmellose sodium, talc, Methyl and propyl paraben were the selected excipients from the trial and error method. Syloid was the majorly focused excipient just because of the moisture sensitivity of the drug.

Physical Stability at room temperature was evaluated as per the ICH Guidelines. But, no significant change was observed.

In wet granulation method the mass became very sticky and in dry granulation the mass became too much hard difficult for disintegrating the tablet.

In herbal tablets more common problems occurs due to the hygroscopic nature of the extract. To overcome this problem we have selected adsorbent material like stylod. Basically stylod is silica. Syloid is having good adsorbent property. Therefore it is widely used in formulating herbal tablets. These prepared tablets having good quality in terms of physical appearance and also passed the parameters which are used for the evaluation of tablets. In future these prepared tablets can be used modify with the film coating or to mask the taste can be sugar coating. To justify the shelf life we had did the three month stability study at room temperature and found no significance change in the quality of the tablets.

As we know the major issues faced in the herbal formulation is the quality of formulation and its shelf life. For that purpose suitable analytical methods can be used. That can do the quantitative evaluation of herbs and their formulation with respect to their marker compound. There are two types of marker compound are present in herbs. One is analytical marker and another is biological marker. Analytical markers are generally used for the standardization of herbs, extract or herbal formulation. And biological markers are used to assess the different bioactivity.

To justify this in this present work we had develop the analytical methods by using modern methods like HPLC and HPTLC.

All the methods were validated with respect to parameters including Linearity, Precision, Accuracy, Limit of Detection (LOD), Limit of Quantification (LOQ), System suitability and Robustness.

**HPLC method was used for simultaneous estimation of gallicacid and Oleanolic acid. Following are the parameters were used.**

Column was Hyperchrom ODS BP C18 (Size: 250*4.6 mm,5μ). Flow rate was 1.0 ml/min. Detection wavelength was 222 nm. Mobile Phase was Ortho Phosphoric acid 0.1 % in Water: Methanol (5:95) It was filtered through 0.45 μm Nylon filter and sonicated for 5 min. Injection Volume was 20 μl through rheodyne manual injector. Temperature was Ambient. Retention Time was 2.8 min for Gallic acid and 9.9 min for Oleanolicacid.

**HPTLC method was used for Simultaneous estimation of Gallicacid, Oleanolicacid And E-Guggulsterone**

HPTLC Plate  Aluminium plates pre coated with silica gel 60 F 254 (10 x 10 cm) was used. .The plates were activated at 110 0 C for 30 minutes Prior to chromatography. Development chamber Camag twin through glass chamber (20x20 cm) was used. The optimized chamber saturation time for mobile phase was 45 Min at room temp. Mobile Phase  Toluene:     Ethyl Acetate: Formic acid (7:6:1) Injection volume was   20 μl. Detection wavelength was UV at 270 nm and 368 nm after spraying with Methanolic Sulphuric acid.

**In silico approach was used to check the anti-obesity potential of different active markers in the extract used in the tablet formulation.**

Different target receptor was selected to check the binding affinity of the active markers to predict the anti-obesity activity.

The PDB file (1HLG) of receptor was downloaded from the PDB databank and ligands were downloaded from the ZINC database. Autodock Vina software was used to predict the binding interactions of markers and standard drugs with protein.

Following proteins ware selected for in silico approach

Pancreatic α-Amylase, Pancreatic Lipase, PPARs (peroxisome proliferator activated receptor) (PPARalpha), Leptin, Cannabinoid receptor type 1(CB1) and HMG CoA Reductase

Oleanolic acid bound efficiently to the active site of alpha-amylase with good complementarity, and the docking score is -9.0 kcal/mol. which is far more compared with standard drug acarbose (docking score -7.6 kcal/mol). These binding interactions present a clear view that Oleanolic acid can have good interactions with alpha-amylase.

Garcinia showed some interactions with human gastric lipase protein through hydrogen bonding and other nonbonding interactions but the interactions are not strong enough as the docking score is -4.7 kcal/mol while Oleanolic acid shoed good binding interaction with docking score of -8.4 kcal/mol. These binding interactions present a clear view that Oleanolic acid interact with human gastric lipase stronger then Garcinia and Orlistat.

Guggulsterone, Oleanolic acid, Mahanine, and Quercetin shown good binding interactions with the human PPAR alpha efficiently as the docking score is -7.9, -8.0, -8.5, and -9.5 kcal/mol, respectively. The reported drug Clofibrate has showed docking score of only -6.2 kcal/mol indicating poor interactions compared to marker compounds.

Mahanine and Oleanolic acid are showing interactions with the active site of human obesity protein, leptin with good complementarity, and the docking score is -6.4 and -7.9 kcal/mol, respectively for 1AX8 whereas -6.4 and -7.5 kcal/mol, respectively for 3V6O. The pose and

interaction suggested that the oleanolic acid binds strongly with the leptin in all the protein structures.

Quercetin, and oleanolic acid are bound efficiently to the active site of cannabinoid receptor type 1 with good complementarity, and the docking score is -8.1 and -8.2 kcal/mol, respectively. These binding interactions (Figure 7) present a clear view that quercetin, and oleanolic acid can irreversibly interacted cannabinoid receptor type 1. Binding interaction of quercetin, and oleanolic acid can irreversibly interact cannabinoid receptor type 1.

Quercetin, oleanolic acid, guggulsterone, and mahanine, are bound efficiently to the active site of human HMG-CoA reductase with good complementarity, and the docking score is -8.2, -8.9, -8.1 and -8.3 kcal/mol respectively. The standard drug atorvastatin formed many H-bonds but has less nonbonding interactions. All the interactions can be visualized.

From the above docking score and binding affinity towards the receptor it can be predicted the selected combination of markers (herbs) having anti-obesity potential.