Meta-analysis of 50 Phase III clinical trials in evaluation of efficacy and safety of Liv.52 in infective hepatitis

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ABSTRACT
Hepatitis A (HA) has a worldwide distribution occurring in epidemic and sporadic patterns. Hepatitis A is an acute, but benign form of viral hepatitis and early renormalizations of hepatic functions with symptomatic and clinical recovery are the objectives in the clinical management of HA. meta-analysis is the term used to describe quantitative methods for combining information across different studies and this study was planned for meta-analysis of the efficacy and safety of Liv.52 tablet and syrup, in HA, as reported in 50 published study reports.

All study reports evaluating efficacy and safety of Liv.52, were included for the meta-analysis, regardless of the study design, but Phase I and II clinical, experimental and preclinical studies were excluded from the meta-analysis. Each study was abstracted for the number and ages of enrolled patients, changes in the biochemical parameters [serum bilirubin (SB), serum glutamic oxaloacetic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT), serum alkaline phosphatase (SAP), serum albumin (SA) and serum globulin (SG), and prothrombin time (PT)] from baseline to values at the end of study and total duration of clinical recovery were recorded. Incidence of adverse events during the study period and patient compliance to the drug treatment was noted. The predefined primary endpoints were to determine level of statistical significance for symptomatic improvement, renormalization of biochemical parameters and total duration of clinical recovery. The predefined secondary endpoints were incidence of adverse events during the study period and compliance to the drug treatment.

Total 50 clinical studies conducted over a span of 30 years were considered for this meta-analysis and the mean duration of these studies was 6.62 months. Out of total fifty studies, 3 were double-blind placebo-controlled studies, 21 were placebo-controlled studies, 22 were non-comparative studies and 4 studies were case reports. Total 4490 patients were enrolled in these studies and 233 children were part of study population. Cumulative data analysis showed a significant reduction in the mean SB, SGOT, SGPT, AP levels, PT and mean period required for total (symptomatic, clinical and biochemical) recovery. The decreased SA and SG levels were also increased significantly, when compared to the pre-treatment values, in all studies. There were no reported or observed significant adverse events in all trials and the overall drug compliance was excellent. Therefore, this metaanalysis concludes that, Liv.52 tablets and syrup are effective and safe in the management of hepatitis.

INTRODUCTION
Meta-analysis (also referred as ‘quantitative synthesis’ or ‘overview analysis’) is the term used to describe quantitative methods for combining information across different studies. Glass coined the term 'meta-analysis' in 1976\(^1\), to describe this idea of utilizing information in many studies of the same effect, although the concept itself is much older (dating back to the 1930s studies by Fisher and Pearson). Today, the term 'meta-analysis' is reserved for a
situation where one combines numerical effects from a collection of studies, rather than giving a more general non-quantitative overview.

The term “Hepatitis A (HA)” has replaced all previous designations like ‘type A viral hepatitis’, ‘infectious hepatitis’, ‘epidemic hepatitis’, ‘epidemic jaundice’, ‘catarrhal jaundice’, ‘infectious icterus’, ‘Botkin’s disease’, and ‘MS-1 hepatitis’. HA has a worldwide distribution and occurs in both epidemic and sporadic patterns. In developing countries, the incidence of HA in adults is relatively low due to the exposure to the virus in childhood and most adults demonstrate an immunity that provides lifelong protection against reinfection.

Hepatitis A is an acute but benign form of viral hepatitis caused by an RNA virus that does not persist in the blood serum. Hepatitis A virus (HAV) is of the enterovirus group of the Picornaviridae family and HAV has a single molecule of RNA surrounded by a small (27 nm diameter) protein capsid. HA is a food/waterborne disease, with feco-oral transmission and the incubation period is variable (from 15 to 25 days). Incubation period is inversely proportional to infective dose and the infectious dose presumably is 10-100 virus particles.

Hepatitis A is usually a mild illness characterized by sudden onset of fever, malaise, nausea, anorexia and abdominal discomfort, followed after several days by jaundice. The dark urine (which precedes jaundice by 2/3 days), indicates the onset of the disease. The risk of transmission is generally low amongst household contacts, but outbreaks occur in nurseries and institutions with attack rates around 10%-15%. The duration of viral shedding is up to 14 days; with a sharp fall off after 5 days, but the period of infectiousness is about 8 to 17 days (the stool remains infectious up to 15 days). Asymptomatic infections are frequent. The serial interval between index and secondary cases is either shorter or equal to the incubation period, indicating that transmission usually occurs before or around the onset of jaundice.

Hepatitis A is diagnosed by finding IgM anti-HAV in serum during the acute or early convalescent phases of disease. A moderate increase in conjugated bilirubin, with marked elevation of SGOT, SGPT and Lactate Dehydrogenase isoenzymes (LDH) are noted in HA (low Na+ and Cl- levels may be seen due to severe vomiting). Marginal elevation of alkaline phosphatase is seen due to compression of intrahepatic biliary canaliculi by swollen parenchymatous cells. Triglycerides and cholesterol may be temporarily and marginally lowered.

Hepatitis A is the least serious of the hepatitis viruses, as it does not kill liver cells and also, there is no risk for a chronic form. Hepatitis A is a self-limiting disease and no specific treatment is available. The primary goals for managing acute viral hepatitis are to provide adequate nutrition (with restricted protein and fat intake), to prevent further damage to the liver, and transmission of infection to others. Therefore, early renormalizations of hepatic functions with symptomatic and clinical recovery are the objectives in the clinical management of HA.

Liv.52 tablet and syrup are polyherbal formulations, used extensively in the management of HA. Liv.52 syrup contains powders of Capparis spinosa, Cichorium intybus, Solanum nigrum, Terminalia arjuna, Cassia occidentalis, Achillea millefolium and Tamarix gallica (in Liv.52 tablet, Mandur bhasma is added). This study was planned for meta-analysis of the efficacy and safety of Liv.52 tablet and syrup, in HA, as reported in 50 published study reports.
AIM OF STUDY
The aim of the study was to meta-analyze the efficacy and short- and long-term safety of Liv.52 in HA, as reported in fifty published study reports.

STUDY DESIGN
This study was a cumulative meta-analysis of fifty published study reports of Liv.52 in HA.

MATERIALS AND METHODS

Inclusion criteria
All published study reports evaluating efficacy and safety of Liv.52 were included for the meta-analysis, regardless of the study design (either double-blind placebo-controlled studies, placebo-controlled studies, open, non-comparative studies, or case reports with less than ten patients).

Exclusion criteria
Phases I and II clinical, experimental and preclinical studies were excluded from the meta-analysis.

Study procedures
All studies were categorized into four subgroups as per the design of the study. Group I included all double-blind placebo-controlled studies, group II included placebo-controlled studies, group III included all open (non-comparative) studies and group IV included case reports, in which less than 10 patients were enrolled.

Each study was abstracted with emphasis on the number and ages of enrolled patients. Changes in the biochemical parameters (serum bilirubin (SB), serum glutamic oxaloacetic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT), serum alkaline phosphatase (SAP), serum albumin (SA), serum globulin (SG), and prothrombin time (PT)) from baseline to values at the end of study, and total duration of clinical recovery were recorded. Incidence of adverse events during the study period and patient compliance to the drug treatment was noted.

Adverse events
All adverse events either reported or observed by patients were recorded with information about severity, duration and action taken regarding the study drug. Relation of adverse events to study medication was predefined as “Unrelated” (a reaction that does not follow a reasonable temporal sequence from the administration of the drug), “Possible” (follows a known response pattern to the suspected drug, but could have been produced by the patient’s clinical state or other modes of therapy administered to the patient), and “Probable” (follows a known response pattern to the suspected drug that could not be reasonably explained by the known characteristics of the patient’s clinical state).

For patients recorded as withdrawing from the study, efforts were made to ascertain the reason for dropout. Non-compliance (defined as failure to take less than 80% of the medication) was not regarded as treatment failure, and reasons for non-compliance were recorded.
Primary and secondary endpoints
The predefined primary endpoints were to determine level of statistical significance for the following parameters: symptomatic improvement, renormalization of biochemical parameters and total duration of clinical recovery. The predefined secondary endpoints were incidence of adverse events during the study period and compliance to the drug treatment.

Statistical analysis
Statistical analysis was done according to intention-to-treat principles. The data was evaluated for normality by the Shapiro-Wilk normality test, while Gaussian approximation and status of exactness was evaluated by Wilcoxon Signed-Rank Test. Changes in various parameters from baseline values and values at the end of the study were pooled and analyzed cumulatively by “paired ‘t’ test”. The minimum level of significance was fixed at 99% confidence limit and a 2-sided $p$ value of $<0.001$ was considered significant.

RESULTS
Total fifty clinical studies that evaluated the efficacy and safety of Liv.52 in HA, conducted over a span of 30 years (from 1967 to 1997) were considered for this meta-analysis. The mean duration of these studies was 6.62 months and the total study duration of all trials was 331 months (minimum period=1.0 month, maximum period=36.00 months, mean of means (M)=6.62, Std. deviation (SD)=6.645, Std. error of mean (SEM)=0.9398, lower 99% confidence interval (CI) of mean=4.097, upper 99% CI of mean=9.143, $W=0.6497$ (Shapiro-Wilk normality test), $p<0.0001$, significant (S)) (Table 1). Out of fifty studies, three were double-blind placebo-controlled studies, twenty-one were placebo-controlled studies, twenty-two were open (non-comparative...
studies and four studies were case reports with less than ten enrolled patients (Figure 1).

Total 4490 patients were enrolled in these studies and 3007 patients received Liv.52 for a mean period of six months. From the control group, 785 patients consumed placebo and other patients consumed corticosteroids, multivitamins, or other treatment (Figure 2).

Two hundred and thirty-three children were part of a study population classified as per their age: 97 children below age of 5 years, 117 children between of 6 to 10 years and 19 children between 11 to 15 years (Figure 3).

The mean serum bilirubin values were elevated at the time of the enrolment, in all patients in these studies. Cumulative data analysis showed a significant reduction in the mean serum bilirubin level, at the end of all studies (parameter, value before treatment, value after treatment: minimum=2.660 and 0.5000, maximum=12.80 and 8.050, mean of means =7.030 and 2.014, SD=2.965 and 1.840, mean of means =7.030 and 2.014, SEM=0.6630 and 0.4115, lower 95% CI of mean=5.642 and 1.153, upper 95% CI of mean=8.418 and 2.875, p<0.0001 (Gaussian approximation by Wilcoxon Signed-Rank Test), S; and t=8.255, p<0.0001, S, mean of differences = 5.016, 95% confidence interval =3.744 to 6.288 (paired t test) (Figure 4).

Similarly, the elevated serum glutamic oxaloacetic transaminase (SGOT) levels were reduced significantly, when compared to the pre-treatment values, in all the above studies. (minimum=24.00 and 0.0, maximum=140.0 and 39.40, mean of means=78.37 and 21.60, SD=37.47 and 13.47,
SEM=12.49 and 4.488, lower 99% CI of mean=36.47 and 6.539, upper 99% CI of mean=120.3 and 36.66, \( p=0.0020 \) and \( p=0.0039 \) (Wilcoxon Signed-Rank Test for exactness), S; and \( t=5.083, p=0.0009 \), mean of differences=56.78, 95% confidence interval=31.02 to 82.53 (Paired ‘t’ test) (Figure 5).

The elevated serum glutamic pyruvic transaminase (SGPT) levels were also reduced significantly, when compared to the pre-treatment values, in all of the above studies (minimum=30.00 and 0.0, maximum=344.0 and 91.59, mean of means =133.4 and 33.59, SD=94.22 and 23.48, \( \text{SEM}=27.20 \) and 6.779, lower 95% CI of mean=73.55 and 18.67, upper 95% CI of mean=193.3 and 48.51, \( p=0.0002 \) and \( p=0.0005 \) (Wilcoxon Signed-Rank Test for exactness), S; and \( t=4.051, p=0.0019 \), mean of differences=99.84, 95% confidence interval=45.59 to 154.1 (Paired ‘t’ test) (Figure 6).

The elevated alkaline phosphatase levels were also reduced significantly, when compared to the pre-treatment values, in all of the above studies (minimum=9.540 and 0.0, maximum=32.96 and 16.70, mean of means=19.09 and 10.72, SD=7.761 and 4.722, \( \text{SEM}=2.074 \) and 1.262, lower 95% CI of mean=14.61 and 7.996, upper 95% CI of mean=23.57 and 13.45, \( p=0.0001 \) and \( p=0.0001 \) (Wilcoxon Signed-Rank Test for exactness), S; and \( t=3.970, p=0.0016 \), mean of differences=8.369, 95% confidence interval=3.816 to 12.92 (Paired ‘t’ test) (Figure 7).

Decreased serum albumin levels also increased significantly compared to pre-treatment values in all studies (minimum=2.100 and 3.010, maximum=4.300 and 4.500, mean of means=3.405 and 3.843, SD=0.7928 and 0.6042, \( \text{SEM}=0.3237 \) and
0.2467, lower 95% CI of
mean=2.573 and 3.209, upper
95% CI of mean=4.237 and
4.477, \( p = 0.0156 \) and \( p = 0.0156 \)
(Wilcoxon Signed-Rank Test for
exactness), S; and \( t = 2.758, \)
\( p = 0.0399 \), mean of differences=
0.4383, 95% confidence interval
= 0.8469 to 0.02975 (Paired ‘t’
test) (Figure 8).

Similarly, the decreased serum
globulin levels were also
increased significantly, when
compared to the pre-treatment
values, in all of the above
studies. (minimum=2.300 and
3.580, maximum=3.160 and
4.100, mean of means=2.662 and
3.696, SD=0.4519 and 0.2260,
SEM=0.2021 and 0.1011, lower
95% CI of mean=2.101 and
3.415, upper 95% CI of
mean=3.223 and 3.977, \( p = 0.0031 \)
and \( p = 0.0031 \) (Wilcoxon Signed-
Rank Test for exactness), S; and
\( t = 3.904, \ p = 0.0175 \), mean of
differences=1.034,
95% confidence interval=0.2987 to
1.769 (Paired ‘t’ test) (Figure 9).

Raised prothrombin time levels
also reduced significantly
compared to pre-treatment values
in all studies (minimum=17.00 and 15.00,
maximum=26.00 and 20.00, mean of means=20.75
and 16.25, SD=4.113 and 2.500, SEM=2.056 and 1.250, lower 95% CI of mean=14.21 and
12.27, upper 95% CI of mean=27.29 and 20.23, \( p = 0.0062 \) and \( p = 0.0062 \) (Wilcoxon Signed-
Rank Test for exactness), S; and
\( t = 3.781, \ p = 0.0324 \), mean of differences=4.500, 95% confidence interval=0.7127 to 8.287 (Paired ‘t’ test) (Figure 10).

There was highly significant reduction in the mean period required for total (symptomatic, clinical and biochemical) recovery, as compared to placebo (minimum=9.000 and 14.00,
maximum=17.00 and 26.60, mean of means=11.84 and 19.56, SD=3.189 and 5.764,
SEM=1.426 and 2.578, lower 95% CI of mean=7.882 and 12.40, upper 95% CI of
mean=15.80 and 26.72, \( p = 0.0031 \) and \( p = 0.0031 \) (Wilcoxon Signed-Rank Test for exactness), S; and \( t = 2.620, \ p = 0.0307 \), difference between means = 7.718 ± 2.946, 95% confidence interval=14.51 to 0.9241 (Paired ‘t’ test) (Figure 11).

There were no reported or observed significant adverse events in any trial and the overall drug
compliance was excellent.
DISCUSSION
The number of papers published on meta-analysis in medical research has increased sharply in the past decade, however the merits and perils of the meta-analysis continue to be debated in the medical community.\textsuperscript{53,54} A useful definition of meta-analysis was given by Huque as: "A statistical analysis that combines or integrates the results of several independent clinical trials considered by the analyst to be 'combinable'".\textsuperscript{55} The terminology, however, is still debated, and expressions used concurrently include "overview," "pooling," and "quantitative synthesis." The present consensus is that the term meta-analysis should be used to describe the statistical integration of separate studies, whereas "systematic review" is appropriate for denoting any review of a body of data that uses clearly defined methods and criteria. Systematic reviews can include meta-analyses, appraisals of single trials, and other sources of evidence.\textsuperscript{56} "Meta-analysis" has recently been included as a Medical Subject Heading (MeSH) and publication type within the Medline indexing system of the National Library of Medicine.\textsuperscript{57}

A single study often cannot detect or exclude with certainty clinically relevant differences in the effects of two treatments. A trial may thus show no significant treatment effect when in reality such an effect exists (false negative result - type II error). Generally better recognized is the type I error (significant difference due to chance and the probability that corresponds to the \(p\) value).\textsuperscript{58} Cumulative meta-analysis is defined as the repeated

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<td>Vijaykumar et al.\textsuperscript{49}</td>
<td>1988</td>
<td>OP</td>
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<td>48</td>
<td>Patney et al.\textsuperscript{50}</td>
<td>1988</td>
<td>C</td>
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<td>325</td>
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<td>49</td>
<td>Dange et al.\textsuperscript{51}</td>
<td>1989</td>
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<td>50</td>
<td>Kalab et al.\textsuperscript{52}</td>
<td>1997</td>
<td>OP</td>
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C: placebo-controlled; DB: Double-blind placebo-controlled; OP: Open-non-comparative; CR: Case reports
Meta-analysis thus not only consists of the combination of data but also includes the epidemiological exploration and evaluation of results ("epidemiology of results")\(^{60}\). Therefore, new hypotheses that were not posed in single studies can be tested in meta-analyses\(^{61}\). The number of patients included in clinical trials is often inadequate as in some cases the required sample size may be difficult to achieve\(^{62}\). Meta-analysis can, nevertheless, lead to the identification of the most promising or urgent research question, and may permit a more accurate calculation of the sample sizes needed in future studies\(^{63}\). The goals of meta-analysis are to enable the overall significance of an effect to be evaluated, based on the multiple studies available; to estimate an overall effect size by combining the individual estimates in multiple studies. The problem of comparability of different study designs and effects of a difference in quality in multiple studies is still not resolved and there are no guidelines for the same\(^{64}\).

While evaluating the efficacy of Liv.52 in HA, the primary endpoints (quantitative comparison of pooled data for biochemical parameters by ‘before’ and ‘after’ tests) were selected by their specific significance in natural history of HA. Most adult-acquired liver disease causes impairment in bilirubin secretion from the liver, which causes elevation of SB in the blood. In acute liver disease, SB is usually elevated relative to the severity of the acute process. Blood levels of SGOT (also referred as aspartate aminotransferase (AST)) and SGPT (also referred as Alanine aminotransferase (ALT)) are elevated in all types of hepatitis (viral, alcoholic, drug-induced, etc.). An elevation in the level of SAP suggests disease of the bile ducts (bile duct obstruction, primary biliary cirrhosis or primary sclerosing cholangitis). Albumin is the major protein synthesized in the liver, which circulates in the bloodstream and low SA concentrations indicate poor liver function. The PT is prolonged when the blood concentrations of some of the clotting factors made by the liver are low and in acute liver diseases, the PT can be prolonged.

This meta-analysis included data of 50 clinical studies (24 placebo-controlled studies and 26 open non-comparative studies) conducted over a 30 years, in 4490 patients (including 117 children) and this sample size is large enough for calculating the intervention (drug) effect. The recommended method for calculating sample size is by the formula: \( n=4pq/L^2 \) (\( p=\)prevalence, \( q=100 – p, L=1.96 \)) and by this formula, this sample size is large enough for a disease, with a very high prevalence. There was significant symptomatic control observed in a week’s time. The cumulative analysis revealed significant reduction in the levels of mean SB, SGOT, SGPT and SAP and there was renormalization of protein levels and PT. There was significant reduction in the mean period required for total recovery, as compared to placebo. There were no reported or observed significant adverse events in all trials and the overall patient compliance to the treatment was excellent. Therefore, as per the predefined primary and secondary endpoints the efficacy and safety of Liv.52 in HA is clinically, biochemically and statistically well proved. These significant effects might be due to the synergistic properties and actions of the ingredients of Liv.52.

Khanfar \textit{et al.} isolated and identified active ingredients of \textit{Capparis spinosa} as betasitosterylglucoside-6'-octadecanoate and 3-methyl-2-butenyl-beta-glucoside\(^{65}\). p-Methoxy benzoic acid isolated from \textit{Capparis spinosa} was found to possess potent hepatoprotective activity against \textit{CCl}_4, paracetamol (\textit{in vivo}) and in thioacetamide, galactosamine (\textit{in vitro})-induced hepatotoxicity\(^{66}\). Al-Said \textit{et al.} demonstrated strong anti-inflammatory activity...
of *Capparis spinosa*, which was comparable to oxyphenbutazone. Bonina *et al.* documented significant antioxidant activity of *Capparis spinosa* and also identified flavonols (Kaempferol and Quercetin derivatives) and hydroxycinnamic acids (caffeic acid, ferulic acid, p-cumaric acid, and cinnamic acid) as major antioxidants from *Capparis spinosa*. In another study, Germano *et al.* observed the antioxidant activity of *Capparis spinosa* using tests like lipid peroxidation, bleaching of free radicals and autoxidation of iron ions. Mahasneh *et al.* observed potent antimicrobial and antifungal activity of *Capparis spinosa*.

He *et al.* isolated 2,3,4,9-tetrahydro-1H-pyrido-(3,4-b) indole-3-carboxylic acid, azelaic acid and daucosterol as the major constituents of *Cichorium intybus* and Du *et al.* identified the other chemical constituents as alpha-amyrin, taraxerone, baurenyl acetate and betasitosterol. Aktay *et al.* and Zafar *et al.* observed hepatoprotective effect (confirmed by histopathological examination) of *Cichorium intybus* against CCl4-induced hepatotoxicity and reported significant prevention of the elevation of malondialdehyde formation (plasma and hepatic) and enzyme levels (AST and ALT). Ahmed *et al.* screened *Cichorium intybus* for antihepatotoxic activity and measured the degree of protection using biochemical parameters (AST, ALT, ALKP and TP). Potent antihepatotoxic activity (comparable to Silymarin) was observed with almost complete normalization of the tissues (as neither fatty accumulation nor necrosis was observed on histopathological study). Kim *et al.* studied the effects of *Cichorium intybus* on the immunotoxicity of ethanol and reported significant increase in the number of circulating leukocytes, the weights of concerned organs (liver, spleen and thymus), number of splenic plaque forming cells, hemagglutination titeres and the secondary IgG antibody response. There were also significant increases in delayed-type hypersensitivity reaction, phagocytic activity, natural killer cell activity, cell proliferation and interferon gamma secretion. Sultana *et al.* reported that, the presence of *Cichorium intybus* in the reaction mixture (containing calf thymus DNA and free radical generating system) protects DNA against oxidative damage to its deoxyribose sugar moiety. All these studies suggest that the observed hepatoprotective effect of *Cichorium intybus* might be due to its ability to suppress the oxidative degradation of DNA in the tissue debris. El *et al.* and Papetti *et al.* documented antioxidative activity (radical scavenging effects, inhibition of hydrogen peroxide, and iron chelation) of *Cichorium intybus*. Gurbuz *et al.* observed significant cytprotection against ethanol-induced damage and these results were further confirmed by using histopathological techniques. Amirghofran *et al.* reported the capacity of *Cichorium intybus* to enhance the proliferation of lymphocytes after stimulation with the allogenic cells. Kim *et al.* investigated the effect of *Cichorium intybus* on mast cell-mediated immediate type allergic reactions and observed inhibition of systemic anaphylactic reaction, reduction in the plasma histamine level.

Ikeda *et al.* identified saponins (nigrumnins I and II) as the active ingredients of *Solanum nigrum*. *Solanum nigrum* was investigated for its hepatoprotective activity against CCl4-induced hepatic damage and Raju *et al.* observed remarkable hepatoprotective activity confirmed by evaluated biochemical parameters (AST, ALT, ALP and TB). Sultana *et al.* demonstrated that *Solanum nigrum* protect DNA against oxidative damage and the results suggest that the observed hepatoprotective effect of *Solanum nigrum* might be due to the ability to suppress the oxidative degradation of DNA in the tissue debris. Moundipa *et al.* studied the effects of *Solanum nigrum* on hepatotoxicity and reported increased activity of aminopyrine N-demethylase, uridine diphosphate glucuronyltransferase and glutathione S-transferase, without any alteration in levels of alkaline phosphatase, aspartate aminotransferase and gamma-glutamyltransferase levels in the serum. Son *et al.* reported *Solanum nigrum* as a potent scavenger of hydroxyl radicals and DPPH radicals. Prashanth Kumar *et al.* tested *in vitro* *Solanum nigrum* for its cytoprotection (against gentamicin-
induced toxicity) and observed significant inhibition of cytotoxicity, alongwith hydroxyl radical scavenging potential, which might be the mechanism of cytoprotection. Similarly, Akhtar et al. observed gastric mucosal cytoprotection offered by Solanum nigrum against aspirin-induced gastric ulcers. Qureshi et al. reported antifungal activity of Solanum nigrum.

Upadhyay et al. identified arjunetoside, oleanolic and arjunic acids as active ingredients from Terminalia arjuna. Munasinghe et al. reported potent antioxidant activity of Terminalia arjuna, which might be due to its effects on lipid peroxidation. Ali et al. demonstrated that arjunapthanoloside from Terminalia arjuna inhibits nitric oxide (NO) production and terminoside A isolated from Terminalia arjuna, decreases inducible nitric oxide synthase (iNOS) levels in LPS-stimulated peritoneal macrophages. Cheng et al. observed potent antiviral activity by virtue of inhibition of viral attachment and penetration by Terminalia arjuna. Samy et al. demonstrated potent antibacterial activity of Terminalia arjuna.

Jafri et al. reported significant hepatoprotective effects of Cassia occidentalis in chemically induced liver damage. Bin-Hafeez et al. showed that Cassia occidentalis modulates hepatic enzymes and provides hepatoprotection against induced immunosuppression. Samy et al. reported antimicrobial properties of Cassia occidentalis comparable with standard reference antibiotics. Perez et al. reported strong antibacterial activity of Cassia occidentalis against Salmonella typhi. Tona et al. reported inhibitory effect of Cassia occidentalis on P. falciparum growth. Caceres et al. and Graham et al. observed antifungal activity of Cassia occidentalis.

Harnyk et al. documented clinically beneficial effects of Achillea millefolium in the treatment of chronic hepatitis. Krivenko et al. reported similar clinical improvements in chronic hepatocleocystitis and angiocholitis with Achillea millefolium. Lin et al. observed anti-hepatoma activity of Achillea millefolium. Candan et al. and Bezic et al. reported antioxidant and antimicrobial activities of Achillea millefolium.

Devarshi et al. studied Mandur bhasma for the hepatoprotective property in hepatitis induced by CCl₄ and observed prevention of CCl₄ mediated changes in the enzyme activities, which suggests the hepatoprotective role of Mandur bhasma.

Therefore, as discussed above these synergistic actions (hepatoprotective, antimicrobial, antioxidant and anti-inflammatory) exhibited by the ingredients of Liv.52 might provide the mechanism of action of Liv.52 in hepatitis.

CONCLUSION

In this study, meta-analysis of 50 clinical studies conducted over 30 years in 4490 patients, was performed to evaluate the efficacy and short- and long-term safety of Liv.52 in HA. The cumulative data analysis revealed clinical and biochemical improvements with significant symptomatic control. In addition, there was highly significant reduction in the mean recovery period. There were no reported or observed significant adverse events in all trials and the overall drug compliance was excellent. Therefore, it may be concluded that Liv.52 tablets and syrup are effective and safe in the management of hepatitis A.
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