

CHAPTER - 4

Materials - Chemicals, Equipments, Software and Animals

4.1 MATERIALS

The list of chemicals, equipment and software were given in Tables, 4.1, 4.2 and 4.3, respectively.

4.1.1 Chemicals

Table 4.1. List of chemicals.

S. NO.	CHEMICAL	SOURCE
1	Ezetimibe	Lupin Laboratories Ltd., Pune, India
2	Nicotinic acid	Avra Labs Pvt. Ltd., Hyderabad, India
3	Nicotinamide	Avra Labs Pvt. Ltd., Hyderabad, India
4	Hydroxypropyl- β -cyclodextrin	Gangwal Chemicals Pvt. Ltd., Mumbai, India
5	D- α -tocopheryl polyethylene glycol 1000 succinate	Antares Health Products, Inc., Illinois, USA
6	L-ascorbic acid-2-glucoside	Nagase Pvt. Ltd., Mumbai, India
7	Methanol	Merck Specialities Pvt. Ltd., Mumbai, India
8	Ethanol	Merck Specialities Pvt. Ltd., Mumbai, India

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9	Acetone	Merck Specialities Pvt. Ltd., Mumbai, India
10	Mannitol	Merck Specialities Pvt. Ltd., Mumbai, India
11	Sodium lauryl sulphate	Merck Specialities Pvt. Ltd., Mumbai, India
12	Glacial acetic acid	Merck Specialities Pvt. Ltd., Mumbai, India
13	Sodium acetate (tri-hydrate)	Merck Specialities Pvt. Ltd., Mumbai, India
14	Hydrochloric acid	Qualigens Fine Chemicals, Mumbai, India
15	Acetonitrile	Merck Specialities Pvt. Ltd., Mumbai, India
16	Ammonium acetate	Merck Specialities Pvt. Ltd., Mumbai, India
17	β -glucuronidase (minimum 100,000 units/mL)	Sigma Aldrich, India
18	Whatman [®] cellulose filter paper	Sigma Aldrich, India
19	Boric acid	Merck Specialities Pvt. Ltd., Mumbai, India
20	Potassium chloride	Merck Specialities Pvt. Ltd., Mumbai, India
21	Sodium hydroxide	Merck Specialities Pvt. Ltd., Mumbai, India
22	Triethylamine	Merck Specialities Pvt. Ltd., Mumbai, India
23	Methyl tertiary butyl ether	Merck Specialities Pvt. Ltd., Mumbai, India
24	Nitrendipine	RA Chem Pharma Pvt. Ltd., Hyderabad, India
25	Dimethyl sulphoxide	Hi Media, Mumbai, India
26	Hard gelatin capsules (size #3)	RA Chem Pharma Pvt. Ltd., Hyderabad, India
27	Membrane filter (0.45 μ m, 0.22 μ m)	Hi Media, Mumbai, India

28	Microcentrifuge tubes	Volex Plasticware, India
29	Sodium carboxymethylcellulose	Hi Media, Mumbai, India
30	Heparin sodium injection	Biological E. Ltd., Hyderabad, India
31	Disposable syringes	Hindustan Syringes & Medical Devices Ltd, Faridabad, India
32	Aluminum foil	Hindalco Industries Ltd., Mumbai, India
33	Foam Tape	Camlin Ltd., Mumbai, India
34	Diethyl ether	SD Fine Chemicals, Mumbai, India
35	Sodium chloride	SD Fine Chemicals, Mumbai, India
36	Rat feed	Lipton feed, Mumbai, India
37	Cogent (<i>in-vitro</i>) diagnostic kits - CHOD-POD - PEG-CHOD-POD - GPO-POD	Span Diagnostics Ltd., Surat, India

4.1.2 Equipments

The following major equipments were used in the various stages of experimental studies.

Table 4.2. List of equipments.

S. NO.	EQUIPMENT	SOURCE
1	Mortar and pestle	Hi Media, Mumbai, India

2	Vacuum desiccator	Hi Media, Mumbai, India
3	Vacuum oven	Cintex, Mumbai, India
4	Digital electronic balance	Shimadzu, Japan
5	Digital pH meter	IKON Instruments, New Delhi, India
6	Digital magnetic stirrer	IKA [®] , C-MAG, HS 7, Germany
7	UV–VIS spectrophotometer	UV–Visible 1800, Shimadzu, Japan
8	HPLC-UV	Shimadzu Corporation, koyoto, Japan
9	Vortex mixer	Fisher Scientific, India
10	Bath sonicator	WUC 1.8L, Fisher Scientific, India
11	Probe ultrasonicator	UP50H, Hielscher, USA
12	Dissolution apparatus	Electrolab India Pvt. Ltd, Mumbai, India
13	High speed homogenizer	IKA [®] T25 digital ULTRA-TURRAX [®] , Germany
14	Lyophilizer	Freezeone, LABCONCO, USA
15	Cooling centrifuge	Refrigerated Centrifuge RC 4100 F, Eltek, Mumbai, India
16	Particle size analyzer and Zeta sizer	DELSA™ NANO C, Beckman Coulter, Inc., USA
17	Stability Chamber	Narang Industries, New Delhi, India
18	Fourier transform infrared spectrophotometer (FTIR)	FTIR-8400S, Shimadzu Co., Kyoto, Japan
19	Differential scanning calorimeter (DSC) and Thermogravimetric analyzer (TGA)	TGA/DSC-1, STAR ^e system, Mettler Toledo, AG, Analytical, Switzerland

20	Powder X-ray diffractometer (PXRD)	PW3050/60 X'pert PRO, PANalytical, Netherlands
21	NMR spectrometer	Joel-FT NMR-AL300, Japan
22	Scanning electron microscope (SEM)	FEI, QUANTA-200, Netherlands
23	Atomic force microscope (AFM)	NT-MDT Solver Next, Limerick, Ireland

4.1.3 Software

Table 4.3. List of software.

S. NO.	SOFTWARE	SOURCE
1	Design-Expert (Version 7.0)	Stat-Ease, Minneapolis
2	Kinetic 5.0 (Trial version)	Fischer scientific, India
3	GraphPad Prism (version 5.03)	GraphPad Prism Inc., USA
4	OriginPro 8	Origin Lab Corp., USA

4.1.4 Animals

Healthy Male Albino Wister rats of 200 – 250 g body weight were obtained from Central Animal Ethical Committee, Institute of Medical Sciences, Banaras Hindu University, Varanasi, India. The study protocol was approved and guided by the same committee. These animals were used for pharmacokinetic and pharmacodynamic studies.

4.1.4.1 Ethics statement

All the animals were obtained and kept in quarantine for at least one week before starting the experiments. All the animals were maintained in accordance with the guidelines suggested by the Council for the Purpose of Control and Supervision of Experiments on Animals, Ministry of Social Justice and Empowerment, Government of India, Delhi, India. The animals were used for the experiments according to the protocol approved and guided by the Central Animal Ethical Committee (CAEC), Institute of Medical Sciences, Banaras Hindu University, Varanasi, India (**CAEC approval number: Dean/2014/CAEC/606**). All the animals were provided with standard laboratory diet (Lipton feed, Mumbai, India) and water *ad libitum*. The animals were housed in polypropylene cages under normal laboratory conditions at ambient temperature and relative humidity (25 ± 2 °C and $55\pm 5\%$ RH). All possible efforts were made to decrease animal distress.

