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Review Article

Mesenchymal stem cells as a platform for research on traditional medicine

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ABSTRACT

The translation of Traditional Medicines (TMs) such as Ayurveda, and Traditional Chinese Medicine into clinical practice remains obstructed due to lack of scientific evidence by means of safety, quality, standardization, clinical efficacy, and mode of action. These limitations can be attributed to the lack of synonymous *in vitro* models which reflect *in vivo* features. Human mesenchymal stem cells (hMSCs) have emerged as an efficient cell source for regenerative medicine and tissue engineering. In this review, the authors discuss how hMSCs can be used as an *in vitro* platform to screen herbs described in TMs using modern methods such as evaluation of its potential, safety, quality, mode of action, etc. Integration of traditional knowledge systems like Ayurveda and hMSCs as a platform to screen and study TMs using modern tools will effectively increase the validity of TMs as evidence-based medicine.

1. Traditional medicines and scientific evidence-based evaluation

The survival of human beings against a wide range of diseases is one of the fascinating aspects of human intelligence and its application in medicine development. In evolutionary terms, civilizations around the globe utilized herbs as prophylactic and therapeutic agents. Development of modern medicine started very recently, i.e., from the 17th century, and is more or less based on the principles of traditional knowledge gained by our ancestors [1]. The traditional Indian medicine system, Ayurveda and Traditional Chinese Medicine (TCM), are two ancient yet in practice medicine systems [2]. Ayurveda, one of the oldest traditional systems of medicine, underlies the benefits of a person-centered cure that deals with a healthy lifestyle, health promotion, sustenance, disease prevention, diagnosis, and treatment. Traditional therapies such as *Panchakarma* and *Rasayana* are used to maintain tissue homeostasis, affected by *Dosha (Vat, Kapha, and Pitta)* balance and *Dhatu* nourishment [3]. However, the translation of Ayurveda principles into clinical practice remained obstructed due to lack of scientific evidence in the context of safety, quality, standardization, clinical efficacy, mode of action, etc. [4]. Over time, the acceptance of TCM in therapeutic practice has increased as its safety and efficacy are validated by modern analytical and biological sciences tools. Ayurveda, being holistic, instinctive, and experimental in nature, hasn't undergone the scrutiny of modern analytical and biological sciences tools up to the extent required. This can be attributed to the lack of an appropriate platform/model to study Ayurveda medicines to generate reproducible scientific evidence.

2. Mesenchymal stem cells

Research on human mesenchymal stem cells (hMSCs) in the last decade opened up new dimensions in regenerative medicine [5]. hMSCs reside in numerous tissue sources such as bone marrow, adipose tissue, muscles, peripheral blood, umbilical cord, placenta, fetal and amniotic fluid, dental pulp, etc., which can be isolated and cultured *in vitro* (Fig. 1) [6]. hMSCs express unique cell surface markers such as CD90, CD73, CD105, and lack expression of CD34, CD45, and HLA-DR (MHC class II) [7]. Absence of HLA-DR expression makes MSCs suitable for allogeneic transfusion widening its scope for therapeutic applications [7]. MSCs possess plastic

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adherent properties and tri-lineage differentiation potential, which is used as a standard for characterization (Fig. 1).

3. Why hMSCs?

The human stem cell field is a relative newcomer on the screen of *in vitro* platforms. Earlier studies on the toxicity of herbal extracts or traditional medicines (TMs) have been carried out employing available cell lines from animal sources [8]. Few studies have also used human cancer cell lines for the same purpose [9]. However, none of these reflect the *in vivo* scenario [10–12]. Moreover, it raises doubts whether the results obtained are reliable. Obviously, what is non-toxic in animal cell lines may be harmful in humans for the same concentration. Another important aspect of using cell lines is their heterogeneity and immortality. These cells are not normal in the sense that they are not diploid but heteroploid. Also, they do not show signs of senescence as they are transformed and therefore immortal i.e., mouse 3T3-L1, HeLa, L 929, BHK 21, VERO, etc. In other words, they have an infinite life which itself is abnormal. Therefore, the results obtained cannot be extrapolated to human studies.

hMSCs obtained from adipose tissue, bone marrow, dental pulp, umbilical cord etc., are normal cells. They exhibit normal diploid chromosome numbers characteristic of *Homo sapiens*. They have a finite life and exhibit signs of senescence as the passage number increases [13]. These hMSCs also secrete growth factors and cytokines, indicating their functionality [14,15]. Hence, these hMSCs are closer to human cells *in vivo*. Therefore, they are attractive and suitable candidates for screening TMs. Their ability to proliferate, differentiate into multiple lineages, immune modulation in response to specific stimuli and senescence make them ideal for testing activity of Ayurvedic compounds, providing evidence-based knowledge. The embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs) are also suitable candidates for the investigations on the TMs, although ethical issues associated with their isolation and the level of expertise required for their culture limit their application as a screening tool in general cell culture laboratories [16]. On the other hand, hMSCs can be isolated from discarded tissues post surgeries and cultured by explant/enzymatic method with a minimal laboratory set-up without genetic manipulations [17].

4. Integration of TMs and hMSCs as an *in vitro* study platform to generate scientific evidence

Recent translational research on TMs and stem cell properties is focused on investigating their proliferative and differentiation potential. MSCs as a platform can be employed to study the ability of TMs to study proliferation, cytotoxicity, senescence, lineage-specific differentiation, immune modulation, etc.

4.1. Proliferation

hMSCs maintain their cell number through somatic division despite their unique potential to differentiate into various lineages under the influence of intracellular and extracellular signals. Cell proliferation becomes essential as the cell number of MSCs declines with increasing age, affecting its concomitant differentiation with downstream fall into tissue maintenance and regeneration. Cell survival and proliferation are necessary to maintain the requisite amount of cell pool to maintain tissue homeostasis. Naturally obtained herbs contain phytochemicals, such as polyphenols, flavonoids, and many other undiscovered and undescribed chemical substances that promote the proliferation of MSCs. In recent years,

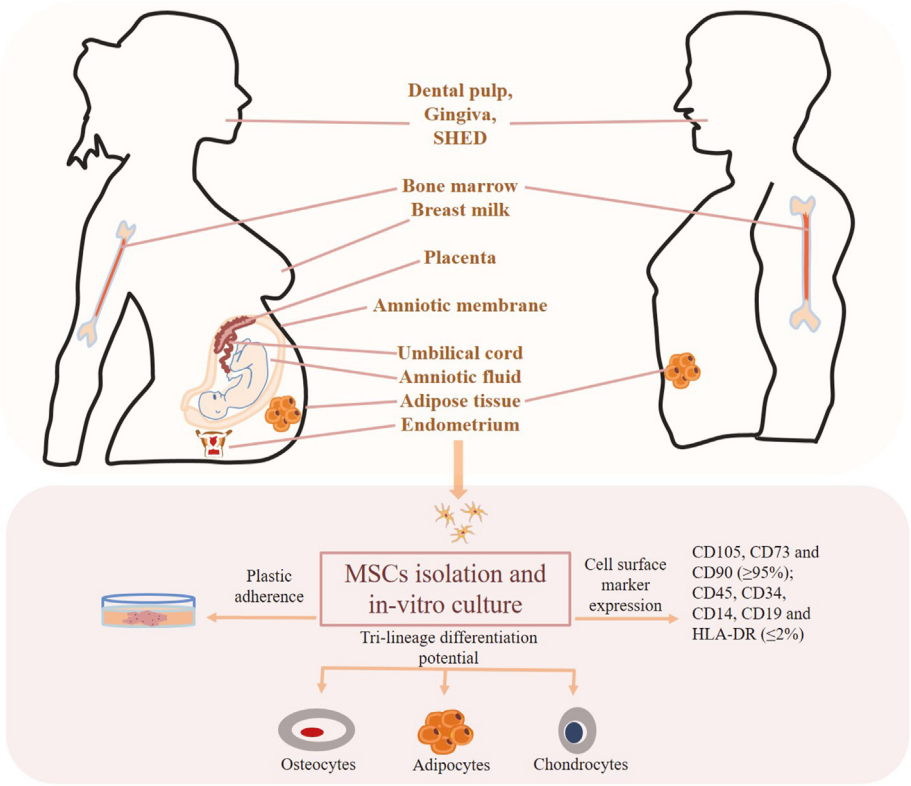

Fig. 1. Potential sources of hMSCs and their characterization criteria: hMSCs can be isolated and cultured *in vitro* from dental pulp, gingiva, human exfoliated deciduous teeth (SHED), bone marrow, and adipose tissue from both sexes. hMSCs can also be obtained from female origin tissues such as breast milk, placenta, amniotic membrane and fluid, umbilical cord, and endometrium.

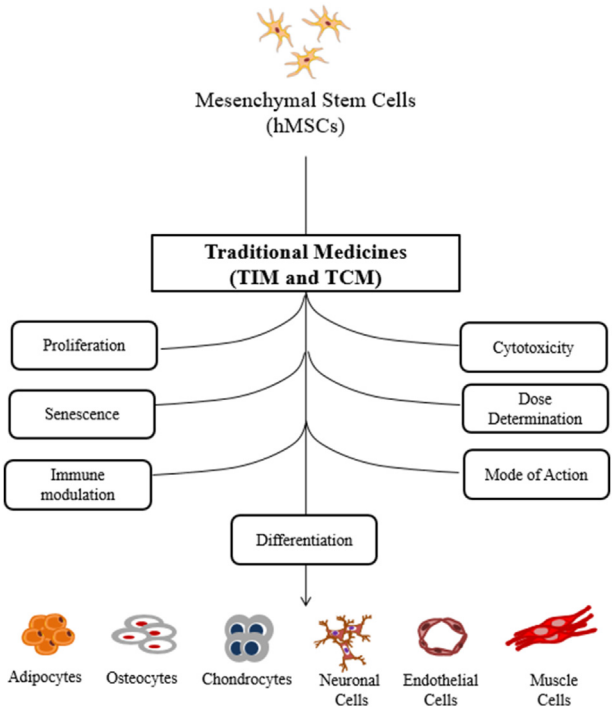

Fig. 2. hMSCs as a platform to screen and investigate TMs for their proliferation, cytotoxicity, senescence, dose determination, immune modulation, mode of action, and differentiation potential.

several studies have demonstrated the effect of herbs on the proliferation rate of MSCs (Table 1). An increase in the proliferation rate of MSCs has been reported by the treatment of *Viscum album* L [18]., *Dhanwantharam kashaya* [19], *Carica papaya* [20], *Tinospora cordifolia* [21], *Withania somnifera* [21], *Cissus quadrangularis* [22], *Terminalia bellirica, Glycine max* var. [23], *Ocimum basilicum* [24], *Paullinia cupana (guaraná)* [25], *Glycyrrhiza glabra* [26], *Thymbraspicata* var. *intricata* [27], TCM formula *ZD-I* [28], *Rhizoma drynariae* [29], *Foeniculum vulgare* [30], *Maluspumila Mill* [31], *Ferula gummosa* [32], *Ginkgo biloba* [33], *Laminaria japonica* [34]. These evidences have been obtained from the bone marrow MSCs (BM-MSCs) [24,26,27,29,30,32,33], wharton's jelly MSCs (WJ-MSCs) [19,21], adipose tissue-derived MSCs (AD-MSCs) [23,25,31], human exfoliated deciduous teeth derived MSCs (SHED) [20], cord blood-derived MSCs (CB-MSCs) [23,31], dental pulp MSCs (DP-MSCs) [24,27], placenta-derived MSCs (PD-MSCs) [18] and telomerised hMSCs [28]. Several plant-derived bioactive compounds have been studied for their proliferative potential, which include phloridzin (flavonoid) [31], naringin (flavonoid) [29], fucoidan (*Laminaria japonica*) [34], amphirionin-4 and 5 (Polyketides) [35].

It is essential to proliferate hMSCs without compromising their stem cell properties to meet the requisite number of cells for therapeutic use. Several recombinant factors, cytokines, and proteins are used to enhance the proliferation of hMSCs, although they possess several limitations and are costly [36]. These studies reveal that hMSCs are a promising platform to screen novel, cost-effective, and non-toxic herbs and their bioactive compounds and to test their potential to enhance cell proliferation.

4.2. Cytotoxicity

Cytotoxicity and alteration of cell morphology were observed upon treatment of *Cimicifugae Rhizoma* [39], *Ligustrum lucidum* [40], *Naringin* [29], and *Asiasari radix* [41] against BM-MSCs and G-MSCs at a concentration range of 100–1000 μg/ml. TMs are used as a medicine for centuries, although not all are safe for human consumption. Many herbs produce secondary metabolites as a defense against predators, which may have disastrous effects upon human consumption [42]. As hMSCs are of human origin, results obtained in the cytotoxicity screening can be highly reliable and reproducible. Furthermore, tissue-specific cytotoxicity can be assessed in the differentiated progenies of hMSCs such as islet of Langerhans, liver and lung organoids [43,44] (Table 2).

4.3. Senescence

hMSCs undergo replicative senescence upon repetitive passaging in *in vitro* culture and limit their therapeutic value for clinical applications [45]. Our studies have reported delay in the senescence of hMSCs by the treatment of *Dhanwantharam kashaya* [19], *T. cordifolia* [21], *W. somnifera* [21]. Studies on herb-treated MSCs have surfaced underlying molecular mechanisms implicated in the senescence, which includes induction of autophagy [18], upregulated synthesis of IL-6 and SCF [20], reduction in apoptosis through restriction on caspase 3 activation [46], activation of mammalian target of rapamycin (mTOR), extracellular signal-regulated kinase (ERK) signaling [31], and activation of Akt signaling pathway [34] (Table 3). Ideally, hMSCs exhibit signs of senescence after passage 5–7. Therefore, hMSCs can be an appropriate cellular model to investigate the anti-aging and anti-senescence herbal formulations from TMs. hMSCs can be a potent tool to unravel the mechanisms implicated in cellular senescence.

4.4. Differentiation

4.4.1. Osteogenic differentiation

hMSCs strengthen and enhance bone formation by differentiating into osteocytes and paracrine secretion of growth factors that promote resident cells to repair the degenerated tissue [47]. Herb-mediated induction/enhancement of osteogenesis holds immense translational potential in age-related bone disorders. Recent studies have highlighted the hidden potential of TMs to drive the osteogenic differentiation of MSCs (Table 4). *F. gummosa,* a herb used in Indian traditional medicine, augments differentiation of BM-MSCs into osteocytes through enhanced alkaline phosphatase activity. *Dipsaci Radix*, a Korean herbal medicine, promotes osteoblastic differentiation of bone marrow-derived MSCs through up-regulation of expression of BSP and OC [48]. Gu-Sui-Bu (*Drynaria fortunei (kunze) J. Sm*), a TCM, enhanced the osteoclast formation through increased expression of alkaline phosphatase (ALP), acid phosphatase titer, and prostaglandin E2 titer [49]. *Herba epimedii* extract augments osteogenesis of MSCs through up-regulation of BMP or Wnt-signaling pathway [50]. A TCM formula extract (ZD-I) promoted proliferation and reversed mineralization of hMSCs through down-regulation osteocalcin, bone morphogenetic protein 2, and osteopontin [28]. A Naringin-containing herb, *R. drynariae*, promoted the proliferation and osteogenic differentiation of human BM-MSCs [29]. Furthermore, *Fructus Ligustri Lucidi* enhances osteogenic differentiation of BM-MSCs through increased ALP activity [40]. An ethanol extract of *F. vulgare* augments differentiation of BM-derived hMSC into osteoblasts [30].

4.4.2. Adipogenesis

Quzhisu, a Chinese herb compound, inhibits the adipogenic differentiation of hMSCs derived from aplastic anemia patient's bone marrow [51]. *Tithonia diversifolia* (Hemsl.) A. Gray (Asteraceae) exhibits reduced adipogenesis of ADMSCs by protecting against oxidative stress and upregulation of pAMPK (phosphorylated 5′-adenosine monophosphate-activated protein kinase) levels

Table 1
The potential of hMSCs platform to screen herbal medicines for their proliferative action.

Name of the herb/herbal formulation	Source of the extract/ solvent used	Stem cells source	Significant outcome	Reference
Korean mistletoe lectin (*Viscum album* L. var. coloratum agglutinin, VCA)	Natural product isolated from semi-parasitic plant	PMSCs	Increase proliferation of PD-MSCs; Cell-specific toxicity to HepG2 cells and immortalized PDMSCs	[18]
Dhanwantram kashaya	Combination of 20 herbs	WJMSCs	Increase proliferation; decrease turnover time	[19]
Carica papaya	Fruit	SHED	Promotes gap closure *in vitro* SHED scratch culture enhances *in vitro* IL-6 synthesis	[20]
Tinospora cordifolia	Leaf	WJMSCs	Increase proliferation of WJMSCs; increase in G2/M phase and decrease in apoptotic cells; upregulation of proliferation marker ki67	[21]
Withania somnifera	Root	WJMSCs	Increase proliferation of WJMSCs; increase in G2/M phase and decrease in apoptotic cells; upregulation of proliferation marker ki67	[21]
Cissus quadrangularis	Stem	UCMSCs	Increase proliferation	[22]
Vegetable soy peptides (*Glycine max* var.)	Soy beans	ADMSCs and CBMSCs	Increase proliferation	[23]
Ocimum basilicum	Flower buds	DPMSCs and BMMSCs	Increase proliferation	[24]
Paullinia cupana	Hydroalcoholic extract	ADMSCs	Increase proliferation	[25]
Glycyrrhiza glabra	Ethyl Acetate Extract of root	BMMSCs	Increase proliferation	[26]
Thymbra spicata var. *intricata*	Ethanolic extract of flower buds	DPMSCs	Increase proliferation	[27]
TCM formula extract (ZD-I)	Water-soluble fractions of the formula	hMSC-TERT cells	Stimulate proliferation	[28]
Rhizoma drynariae (Naringin)	Commercially procured Naringin	BMMSCs	Enhance proliferation	[29]
Foeniculum vulgare	Ethanol	BMMSCs	Increase proliferation	[30]
Apple	Ethanol extract	ADMSCs and CB-MSCs	Promote proliferation	[31]
Ferula gummosa	Ethanol extract	BM-MSCs	Increase proliferation	[32]
Ginko biloba (ginkgolide and bilobalide)	Leaves	BM-MSCs	Increase proliferation	[33]
Fucoidan	Derived from brown algae and seaweed	BM-MSCs	Induce proliferation	[34]
Cissus quadrangularis (Linn.)	Petroleum ether extract	BM-MSCs	Enhance proliferation	[37]
Piper longum L.	Aqueous fruit extract	UC-MSCs	Enhance proliferation	[38]

Table 2
The potential of hMSCs platform to screen herbal medicines for cytotoxicity.

Name of the herb/herbal formulation	Source of the extract	Stem Cells Source	Significant outcome	Reference
Cimicifugae Rhizoma	Aqueous	G-MSCs	Significant reduction in cellular viability at 100 and 1000 μg/ml	[39]
Asiasari radix	Aqueous	G-MSCs	Significant reductions in cell viability at 100 and 1000 μg/ml	[41]
Rhizoma drynariae (Naringin)	Commercially procured Naringin	BMMSCs	Exhibit cytotoxicity at 200 μg/ml or higher dose	[29]
Fructus Ligustri Lucidi	Ethanol extract	BM-MSCs	Inhibit cell proliferation in dose dependent manner	[40]

[52]. Aloe-emodin (AE), a constituent of aloe vera and roots, and rhizomes of *Rheum palmatum* L. inhibited adipogenesis by dowregulating mRNA expression of resistin, adiponectin, aP2, lipoprotein lipase, PPARγ, and tumor necrosis factor-α.

4.4.3. Neurogenesis

Salvia miltiorrhiza extract induces WJMSCs differentiation into neural-like cells as characterized by upregulated expression of nestin, β-tubulin, neurofilament, and glial fibrillary acidic protein [53]. A seed extract of *Mucuna gigantea*, which contains L- DOPA promotes neurogenesis of BM-MSCs through upregulation of nestin and β-III tubulin mRNA [54]. A Chinese herb, *Radix Angelicae Sinensis,* exhibited positive expression of neuron-specific enolase and differentiated AD-MSCs into neuron-like cells [55].

4.4.4. Endothelial differentiation

Extract of *Curcuma longa* L. and (−)-Epigallo Catechin-3-Gallate enhances differentiation of AD-MSCs into endothelial progenitor cells by increasing expression of CD34, CD133, and vascular endothelial growth factor receptor (VEGFR)-2 [56]. Olive leaf extract induced differentiation of hMSCs into endothelial cells via upregulation of vascular endothelial growth factor, PCAM, platelet-derived growth factor receptor, and VEGFR-1 [57].

Research on TMs has unfolded the hidden potential of naturally occurring herbs and their bioactive compounds in the hMSC

Table 3
The potential of hMSCs platform to investigate the anti-senescence properties of the TMs.

Name of the herb/herbal formulation	Source of the extract	Stem Cells Source	Significant outcome	Reference
Dhanwantram kashaya	Combination of more than 20 herbs	WJMSCs	Delay senescence	[19]
Tinospora cordifolia	Leaf	WJMSCs	Delay senescence	[21]
Withania somnifera	Root	WJMSCs	Delays senescence	[21]
Guaraná *(Paullinia cupana)*	Hydroalcoholic extract	ADMSCs	Reduce senescence	[25]

Table 4
The potential of hMSCs platform to investigate the multi-lineage differentiation of the TMs.

Name of the herb/herbal formulation	Source of the extract	Stem Cells Source	Significant outcome	Reference
Ocimum basilicum	Flower buds	DPMSCs and BMMSCs	Increase osteogenic differentiation	[24]
Glycyrrhiza glabra	Ethyl Acetate Extract of root	BMMSCs	Increase osteogenic differentiation	[26]
Thymbraspicata var. *intricata*	Ethanolic extract of flower buds	DPMSCs	Increase osteogenic differentiation	[27]
TCM formula extract (ZD-I)	Water-soluble fractions of the formula	hMSC-TERT	Decrease the bone mineral deposition of hMSCs	[28]
Rhizoma drynariae (Naringin)	Commercially procured Naringin	BMMSCs	Enhance osteogenic differentiation	[29]
Foeniculum vulgare	Ethanol	BMMSCs	Increase osteogenic differentiation	[30]
Ferula gummosa	Ethanol extract	BM-MSCs	Osteoprotective	[32]
Ginko biloba (ginkgolide and bilobalide)	Leaves	BM-MSCs	Increase osteogenic differentiation	[33]
Fucoidan	Derived from brown algae and seaweed	BM-MSCs	Induce osteogenic differentiation	[34]
Cissus quadrangularis (Linn.)	Petroleum ether extract	BM-MSCs	Stimulate osteoblastogenesis	[37]
Piper longum L.	Aqueous	UC-MSCs	Enhance osteogenic differentiation	[38]
Fructus Ligustri Lucidi	Ethanol extract	BM-MSCs	Improve osteogenic differentiation	[40]
Dipsaci Radix (hederagenin 3-O-(2-O-acetyl)-α-L-arabinopyranoside)	Dichloromethane Fraction	BM-MSCs	Improve osteogenesis	[48]
Herba Epimedii (flavonoids)	Flavonoids	BM-MSCs	Promote osteogenic differentiation	[50]
Quzhisu	Wulingzhi, Kuihuami and Shanzha	BM-MSCs	Inhibit the adipogenic differentiation	[51]
Tithonia diversifolia (Hemsl.) A. Gray	Leaves	AD-MSCs	Inhibit adipogenesis	[52]
Mucuna gigantea	Seed aqueous acetic acid	BM-MSCs	Induce expression of neural protein and gene markers	[54]
Radix Angelicae Sinensis (Angelica sinensis) (Danggui)	Dried root	AD-MSCs	Induce differentiation into neuron-like cells	[55]
Curcuma longa L. (ECL) and (−)-Epigallo catechin-3-gallate (EGCG)	Dried rhizomes; Ethanol	AD-MSCs	Enhanced differentiation into EPCs	[56]

differentiation into multiple lineages (Table 4). The hMSC culture allows simultaneous evaluation of the herbal compounds at a particular concentration for their guided lineage-specific differentiation (Fig. 2). Therefore, naturally occurring herbal bioactive compounds with differentiation potential can be exploited to replace the synthetic cocktails conventionally used for *in vitro* differentiation studies. It will be an inexpensive and non-toxic substitute whose potential can be encashed by their hidden economic potential. Furthermore, a fixed-dose/concentration of TM can be tested for its adipogenic, osteogenic, chondrogenic, and neuronal differentiation potential. It can provide us a clue whether the same concentration of any drug triggers only one differentiation pathway or stimulate/or inhibit another differentiation lineage. This is of significance in deciding dose and time course. Such studies will also caution us about the adverse effects of compound if any, or side-effects of the dose chosen and will help us choose compounds with uni-differentiation potential. It is not expected that the drug used for chondrogenesis should not promote adipogenesis or that an anti-adipogenic drug is not inducing osteoporosis. hMSC platform can resolve many unanswered questions when used judiciously.

5. Scope and significance of hMSC platform for screening TMs

hMSCs have their own merit as a human brand. Firstly, these are derived from the human medical waste tissue supporting the trash to treasure concept. Secondly, the tissue availability is abundant, and there are fewer ethical issues as cultures are derived from biomedical waste. This very fact makes them an appropriate platform for testing TMs and other allopathic medicines to get comparative analysis. The results obtained would be consistent and reproducible as one can use cells from the same early passage before showing signs of senescence. It is also possible to synchronize cells in one phase of the cell cycle to get reproducible and meaningful results. Being tested on normal human cells, the data can be extrapolated to human cases without any doubt. Moreover, it is also possible to use stem cells and their differentiated progeny for studying the effect on specific cells such as hepatocytes, islets of Langerhans, neural cells, cardiomyocytes, adipocytes, etc. To investigate tissue-specific effects of bioactive compounds, researchers need to perform a primary culture of the tissue or obtain organ-specific cell lines that are costly, time-consuming, and still may not reflect *in vivo* scenarios. In contrast, hMSCs can be differentiated into the multiple lineages of interest, and the cytotoxicity or the biological activity of the TMs can be assessed by using hMSCs and differentiated progeny as an in-house *in vitro* model.

The current methods of cytotoxicity testing involve the testing of herbal extracts against the homogenous culture of human cells in a 2D platform which do not mimic the *in vivo* environment of complex organ systems and their interactions. With advancements in cell culture techniques, it is possible to culture hMSCs in 3D platform which mimics the *in vivo* environment. hMSCs can be cultured in scaffold-based hydrogel-based support, polymeric hard material-based support, hydrophilic glass fiber, and organoid cultures. To overcome this limitation, hMSCs can be differentiated into 3D organ culture of choice (intestine, lung, stomach, liver, Islet-like clusters, etc.), and cytotoxic effects of TMs can be assessed, which will improve the translation of TMs from the laboratory into clinical practice [43,44]. Therefore, a 3D *in vitro* hMSCs platform can provide more accurate and reliable data on the cytotoxicity, drug screening, and its mode of action [58]. The additional benefit would be a reduction in animal experimentation, supporting the “3 R” principle of Reduction, Refinement, and Replacement in animal experimentation. hMSC platform will be economical in terms of money spent and time consumed. Since the cell culture system is clean and not influenced by systemic factors like anxietyand stress, the results obtained are pure and accurate to the model chosen. The toxicity data is likely to shed light on the effect of TMs at the cellular level. Even the type of cell death pathway can be traced, indicating apoptosis, necrosis, or autophagy. In a nutshell, this hMSC platform will resolve several issues related to heavy metal toxicity present in Ayurvedic preparations; irrespective of whether they are toxic or mode preparation has detoxified, their toxicity can be studied.

hMSCs as an *in vitro* platform to screen and investigate the TMs for therapeutic application may possess few limitations. The outcomes from the *in vitro* studies may vary with donor characteristics and increasing passage number. Also, results may vary with the

tissue source of hMSCs as they tend to possess niche-specific properties of proliferation, differentiation, and paracrine secretion. The influence of donor's age, sex, genetics, environmental conditions, and epigenetic modifications cannot be overruled and may influence the outcomes [59]. Source-specific hMSCs properties such as proliferation, differentiation, and immune modulation may also result in varied outcomes. Therefore, selecting the ideal source of hMSCs and the dosage of TMs are crucial factors in achieving consistent and reproducible results. Furthermore, TMs encompass a wide range of health practices, including intervention through herbal medicines and spiritual therapies, manual techniques, and exercises to prevent and treat diseases. Several herbal, non-herbal origin TMs and homeopathic formulations possess solubility issues which may further limit their accessibility towards screening in the hMSCs as an *in vitro* platform. Different parts of the plant (roots, stem, leaf, and fruit) may possess a varied concentration of the bioactive compounds. Therefore, it is necessary to select the appropriate part of the herbs for extracting bioactive components. Moreover, naturally occurring phytochemicals such as alkaloids, polyphenols, flavonoids, and other plant-derived chemicals exhibit a varied range of solubility in different solvents. As per Ayurveda, traditionally, extracts and formulations like *asavas* and *arishtas* are prepared in solvents like water or self-generated alchohol (water-alcohol); hence, the choice of solvent is limited while employing hMSCs for screening TMs. Organic solvents such as acetone, ethyl ether, ethanol, dichloromethane, ethyl acetate, hexane, benzene, toluene, etc., may possess intrinsic cytotoxicity and may exert undesired cellular effects on hMSCs. This solubility issue is a limitation of all cell culture models.

Needless to say that in the era of reverse engineering in the case of natural compounds and TMs, the hMSC platform will provide scientific evidence to the action of these compounds, the probable mode or mechanism of action, and enhance their use globally for a good cause (Fig. 2). It will give rebirth, name, and fame to the TMs as it will be tested on modern medicine platforms, which is a need of time.

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Conflict of interest

None.

Author contributions

Ramesh Bhonde: Conceptualization, Writing- Reviewing and Editing; **Avinash Sanap:** Data curation, Writing- Original draft preparation; **Kalpana Joshi:** Writing- Reviewing and Editing.

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