Overview on Pathogenesis and Role of Probiotics in Liver Fibrogenesis

Moganthan R[1], Abarnadevika A[2], Ariharasivakumar G[3], Swarnabala S[4], Deepika R[5]
[1]*Department of Pharmacology,KMCH College of pharmacy, Coimbatore, India*
[2]*Assistant Professor, Department of Pharmacology, KMCH College of pharmacy, Coimbatore, India*
[3]*Professsor & Head, Department of Pharmacology, KMCH College of pharmacy, Coimbatore, India*
[4]*Department of Pharmacology, KMCH College of pharmacy, Coimbatore, India*
[5]*Department of Pharmacology, KMCH College of pharmacy, Coimbatore, India*

Abstract-Liver fibrogenesis is generally marked by deposition of Extracellular matrix protein (ECM), which is a reason for majority of the long-term liver diseases. The most common causes of liver fibrogenesis are misuse of alcohol genetic abnormalities, hepatitis virus infections, autoimmunity, and other non-infectious illnesses including fatty liver. A chain of events results in the liver hardening, including the retention of inflammatory cells and the activation of collagen-producing cells inside the injured liver that destroys hepatocytes and ultimately lead to hepatic fibrogenesis, which hardens the liver. Extracellular matrix proteins [ECM], notably collagen type 1, which is largely produced by hepatic stellate cells, are deposited excessively in this condition, making it distinct (HSCs). Probiotics are effective, economical, and have no long-term detrimental effects. Probiotics are the therapeutic option for liver fibrogenesis. Probiotics have antioxidant potency to reduce the elevated degree of (ROS) reactive oxygen species because it is the major cause for HSCs activation and it will introduce the IL-10 which is act as a protective cytokine and it inhibits the inflammatory cytokines and decrease the protein deposition.

Keywords: Liver fibrogenesis, Extracellular matrix proteins [ECM], Hepatic stellate cells (HSC), Probiotics.

I.INTRODUCTION

The majority of chronic liver conditions are accompanied by extracellular matrix protein accumulation, which results in liver fibrogenesis. Major reasons are alcohol abuse, genetic abnormalities, autoimmunity, hepatic viral infections and non-infectious conditions like fatty liver also lead to liver fibrogenesis. [1]

Liver fibrogenesis is chronic disease leads to cirrhosis and finally developed into hepatocellular carcinoma [HCC] Fibrogenesis results from an excessive accumulation of scar tissue that is triggered by inflammation of the liver cells. Dead liver cells are formed into abnormally rounded patches of tissue called nodules, where they will be modified by regenerated cells. A chain of events results in the liver hardening, including retention of inflammatory cells and the activation of collagen-producing cells inside the injured liver that destroys hepatocytes and ultimately lead to hepatic fibrogenesis, which hardens the liver. Extracellular matrix proteins [ECM], notably collagen type-I, which is largely produced by hepatic stellate cells, are deposited excessively in this condition, making it distinct (HSCs). [2-3]

It has been discovered recently that eliminating pathogens or eliminating the aetiology, has the capacity to treat or prevent viral infections, as well as reverse liver fibrogenesis. This is due to increased observation of the prevalence and improvement mechanism of liver fibrogenesis as well as the use of scientific drugs. Yet, internal liver fibrogenesis reversal still has a lot of really good difficulties.

The large number of anti-fibrotic candidate medications has demonstrated accurate outcomes in clinical investigations using experimental animal models and their anti-fibrotic effects are still quite restricted. [4]

2. LIVER FIBROGENESIS-RELATED CELL TYPES

2.1 HSC_s

The activation of HSCs is thought to be an essential step in the development of liver fibrogenesis, and novel research continues to be produced on the molecular processes behind this cellular alteration [5, 6]. From the perspective of cell destiny or lineage control, however, nothing is known about HSC activation [7-9]. Recent research has established that multipotent mesenchymal

progenitor cells (MMPC), which may develop into neural and other types of mesenchymal cells [10, 11]. HSCs also express markers for the neuronal and mesenchymal lineages, validating our findings. It makes that the cell types are formed by MMPC are capable of transdifferentiating within their lineages, HSC transdifferentiation may take place in these mesenchymal lineages. [11]. HSCs are present in quiescent form in healthy liver tissue, where they store the retinoid and glial fibrillary acidic protein (GFAP) was produced.[12-15] After liver damage, HSCs become activated when retinoids and GFAP gradually disappear, which reduces the factors of adipogenic and lipogenic production. Meanwhile, the transdifferentiation of HSCs and Myofibroblastic phenotype is mediated by a complex network of autocrine/paracrine fibrogenic signals.

2.2 Portal fibroblast

Portal fibroblasts are known as mesenchymal spindle-shaped cells that differentiate into myofibroblasts particularly in biliary and cholestatic liver injury [16]. Portal fibroblasts vary from activated HSCs in terms of their genetic profiles and signalling behaviours, albeit sharing some basic characteristics with them [17, 18]. The latter might make it possible to create antifibrotic treatments that are tailored to a given illness and target these cells.

2.3 Fibrocytes

Hematopoietic stem cells give rise to fibrocytes, which can develop into MFs. Fibrocytes multiply, move to the wounded tissues and produce growth factors that encourage ECM production in situations of tissue damage [19-21].The kind of damage and the organ determine how far fibrocytes differentiate into MFs. Additional investigations have shown that liver damage causes fibrocytes to migrate to lymphoid tissues, indicating that the role of these cells may be more diverse than just ECM deposition. [20, 21]

2.4 Bone marrow derived myofibroblast

Mesenchymal stem cells (MSCs), multipotent progenitor cells with the tendency to develop into cells of certain cellular lineages [20, 21] and can also give rise to a portion of hepatic MFs. 3.2 Various factors activates the hepatic stellate cells

Circulating MSCs most possibly constitute a population that is different from hematopoietic-derived fibrocytes, albeit it is unclear at this time whether they significantly contribute to ECM deposition during liver fibrogenesis or not [21].

2.5 Epithelial mesenchymal transition (EMT)

Fully developed epithelial cells transform phenotypically into completely differentiated mesenchymal cells through a process known as EMT. Hepatocytes and cholangiocytes have been demonstrated in experiments using liver cell cultures to go through EMT and FSP-1 expression and other mesenchymal features are acquired[22,23] The majority of current research, however, strongly affirm that ECM-producing cells have an epithelial origin and provide compelling evidence that the liver is not a source of MFs through EMT.[24,25]

3.PATHOGENESIS OF LIVER FIBRO GENESIS

ECM is an essential substrate in a healthy liver that has a properly regulated balance between production to breakdown. Yet, as a result of the development of thick fibrotic septae and chemical cross-linking of collagen, hepatic fibrogenesis results from ECM synthesis exceeding ECM breakdown after chronic liver damage. Moreover, these modifications to the ECM composition directly promote fibrogenesis [26],

The liver contains ECM, which includes collagens (I, III, IV), undulin, fibronectin, laminin, elastin, hyaluronan, and proteoglycans, is approximately 6 times more abundant in advanced stages than it is normal. ECM builds up as a result of both enhanced production and reduced deterioration[27].

A significant cause of the ECM-removing MMPs' reduced activity is an overexpression of their particular inhibitors (TIMPs).

Due to reactive oxygen species generation also activates the quiescent hepatic stellate cells into activated hepatic stellate cells and it further activates the myofibroblasts also activates the MMP-3 & 9 it leads to deposition of extracellular matrix protein. The pathogenesis of fibrogenesis was described in (Figure: 3.1) and various factors that activates the hepatic stelate cells were described in (figure:3.2.1)

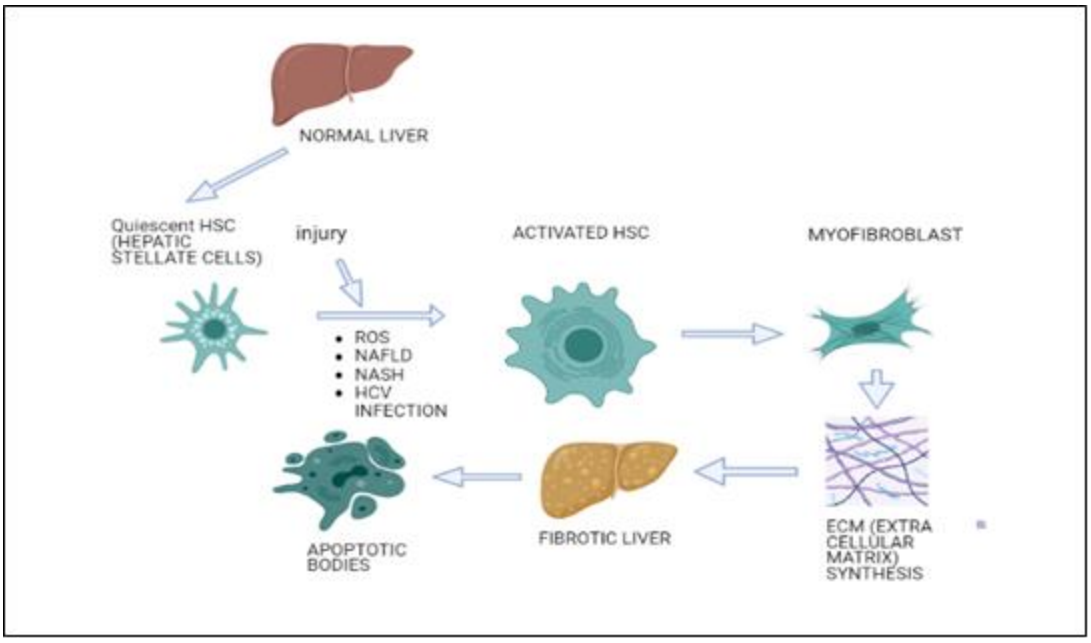

Figure 3.2.1: Various factors that activates hepatic stellate cells

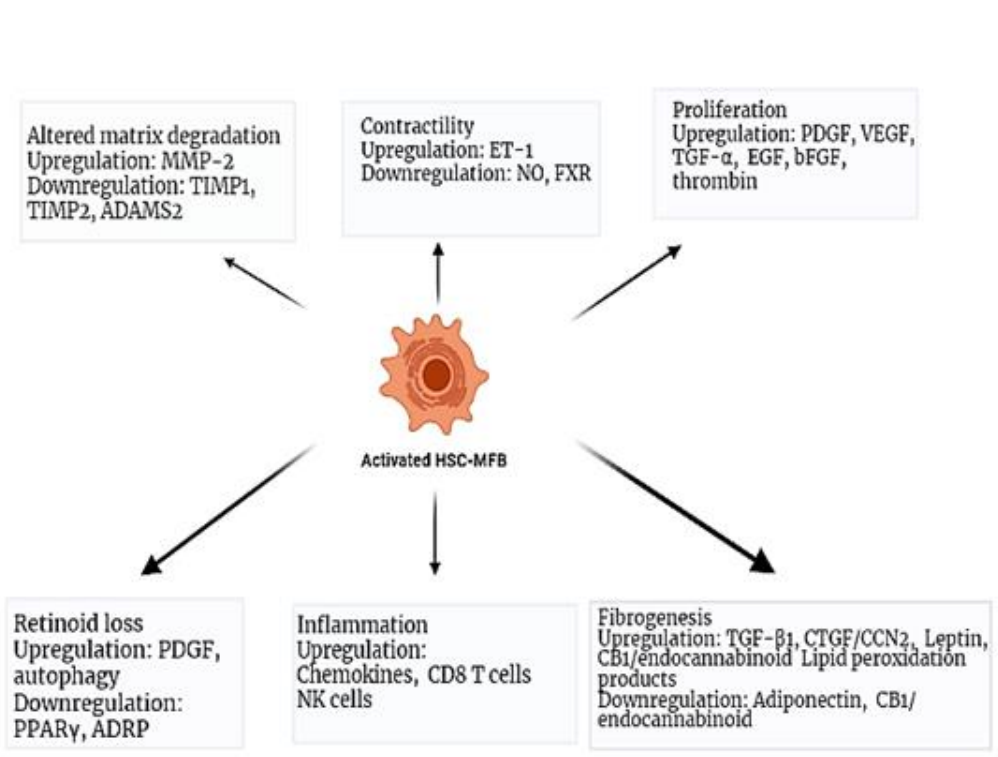

Figure 3.1: Pathogenesis of liver fibrogenesis

4. PROBIOTICS

Originally defined as "microbes inflicting growth of various organisms," probiotics are now understood to be "live microorganisms that, when taken in sufficient concentrations, result in beneficial effects to the host." [28] The following criteria must be met for a microorganism to be considered probiotic: it must be of human origins, non-pathogenic, extremely resistant to passing through the intestine, flexible enough to stick to

mucous secretions while possibly advantageous to the system and overall human health, it also serves to prevent the adhesion of other pathogenic microbes. [29] Since they reduce bacterial tissue layer adhesion, probiotics are widely recommended as a therapy for preventing chronic liver damage. This prevents the production of antimicrobial peptides, as well as microorganism translocation and epithelial invasion.They also reduce inflammation and enhance host immunity. [30]

4.1. Mechanism

Probiotic advantages' underlying mechanisms are not entirely understood. Four broad advantages were described, though:

(1) Pathogenic bacterial invasion or growth inhibition by epithelial binding [31]

(2) Enhanced intestinal barrier performance [32]

(3) Immune system modulation, which includes suppressing the migration of

T-coadjutor 1 cells and inducing protective cytokines like IL-10 and inhibiting pro-inflammatory cytokines like TNF-α [33]

(4) The dominant expression of micro-opioid and cannabinoid receptors modulates the experience of intestinal discomfort.

4.2. Probiotics and inflammation

The most necessary causes of inflammation are when bacteria are present where they shouldn't be. Inflammation will developed as a result of both internal and exterior influences. As the morbific bacteria are aggressive, this can make the membrane's barrier less effective, allowing the microorganism to enter the body. After their termination within the liver, these components cause inflammation in the liver. [34] Probiotics provide health benefits to the host through a variety of possible processes, including as reducing the encroachment of harmful agents from the stomach into the body. Most of the liver's microbial activity and clearance takes place there, where kupffer cells, or macrophages, are present [35]

These Kupffer cells trigger an inflammatory response by producing chemokines, pro-inflammatory cytokines, nitrogen species and reactive oxygen species all of which injure the liver tissue when they are exposed to LPS or other microbial products such unmethylated DNA, lipopeptides, and double stranded RNA. [35]

These microbic substances originate injurious effects by acting through a specific class of receptors known as Toll-like receptors (TLRs). TLRs identify pathogen-derived signature molecules and alter the host to regulate innate immune responses. [36]

Kupffer cells, hepatocytes, and liver radial cells are just a few of the different types of alternative liver cells that express TLRs (HSCs). The extremely potent anti-inflammatory properties of TLRs, their expression in the liver, as well as the hepatic exposure to TLR ligands from the gut, suggest that TLRs play a key role in mediating hepatic inflammation, damage, and fibrogenesis. A significant relationship between inflammation and wound-healing responses in the liver is the enteric microbiota and TLRs.

TLR4 plays a particularly important role in the promotion of inflammation and injury in conditions like alcoholic disease and NASH [36] by interacting with CD_{14} on the cell surface to start LPS-induced signal transduction, particularly the activation of nuclear factor κB (NF-κB) and the subsequent production of inflammatory mediators. TLR4 binds to CD14 on the cell surface to initiates LPS-induced signal transduction, which includes the activation of nuclear factor κb (NF- κB), which leads to the synthesis of disruptive cytokines similar to normal cytokines and enzymes. [36].

An significant intracellular inflammatory cascade, including the NF-κB pathway, c-Jun-N-terminal kinase, stress- and mitogen-activated super molecule enzymes, and TLR4 is triggered by LPS. IKK- (NF-κB kinase monetary unit) removal enables NF-κB to translocate to the nucleus, where it activates genes involved in constrained inflammatory pathways, producing TNF-α and IL-1β.

These kinds of consequences include the conflict for limited nutrients with pathogenic bacteria [38] and the modification of inflammatory pathways brought on by gut bacterial overgrowth via cytokine signalling. The clinical benefits of probiotics in treating liver illnesses may possibly be attributed to their numerous anti-inflammatory capabilities. [39]

4.3. Probiotics as a potential anti-fibrotic therapeutic alternative

As recurrent and ongoing liver cell damage results in HSC activation, which is known as the primary matrix-producing cell in liver fibrogenesis, chronic liver injury is defined by the development of hepatic fibrogenesis. [40]

Intestinal microbiome appears to be able to cause fibrotic liver disease by activating Kuppfer cells, inducing c synthesis, and activating HSCs through an increase in portal delivery of endotoxins. The majority of myofibroblast progenitors as well as the "predominant targets via which TLR4 ligands stimulate fibrogenesis" is HSCs." [41]

HSCs may play a significant part in the hepatic inflammatory cascade linked to endotoxemia. Kupffer cells contribute to fibrogenesis by producing unfavourable and pro-fibrogenic mediators, but HSCs are the primary source of the extracellular matrix deposited throughout the fibrotic process. Both Kupffer cells and HSCs express TLR4. [40]

Probiotics also show metal chelating exertion. Thus, probiotics are suggested to be a implicit source of antioxidant compounds so that it can reduce the elevated degree of reactive oxygen species (ROS) and it also reduce the TGF-β and TLR4 expression. [43]

Table: 1 Treatment of Probiotics in various conditions in animals

Conditions	Treatment	Results
CCl_4 (Carbon tetrachloride)	Mixture of *Saccharomyces cerevisiae* + *Lactobacillus acidophilus*	Decrease the levels of hepatic oxidative stress, inflammation, ER stress, MAPK signaling, , ALT AST, α-SMA ,Col1agen [44]
	Lactobacillus fermentum	Decrease the levels of AST , ALT, MDA , SOD, GSH ,IL -1β, TNF-α, Caspase 3↓, Bax, NF-κB, p65 Inflammation, and increase the level of Bcl-2 [45]
	Lactobacillus plantarum	Decrease the levels of Inflammation MDA, SOD, GSH, AST, ALT, IL-1β, TNF-α, NF-κB, Bax, p65, Caspase increase the level of Bcl-2 [45]
	Lactobacillus salivarius LI01	Decrease the levels of ALT, AST, TLR2,4,5,9, GGT, intestinal barrier integrity, Col1agen, TGF-β, Timp1 [46]
	Pediococcus pentosaceus LI05	Decrease the levels of AST, ALT, GGT, TLR2,4,5,9, Col1agen, Timp1, TGF-β [46]
TAA (Thioacetamide)	Mixture of *Weissella confusa* + *Lactobacillus casei* + *Lactobacillus paracasei*	Decrease the levels of serum enzyme levels, inflammation, fibrogenesis [47]
Ethanol	*Lactobacillus fermentum*	Decrease the levels of steatosis score, iNOS, AST, ALT [48]
BDL (Bile Duct Ligation)	*Lactobacillus rhamnosus GG*	Decrease the levels of AST, ALT, ALP, Col1agen, α-SMA, Col3, TGF-β, Mmp2, Timp1, F4/80, TNF-α, IL-1β ,IL-6 [49,50]

Table 2: Various probiotic treatments in humans

Treatment	Strains	Dose	Outcomes
Probiotics	*L. rhamnosus* CBT LR5 *L. acidophilus* CBT LA1 *P.pentosaceus* CBT SL4 *L.paracasei* CBT LPC5 *B. breve* CBT BR3 *B. lactis* CBT BL3	10^9 Colony forming unit per day of probiotic strains, for 12 weeks	Increase the levels of *L. rhamnosus, L. acidophilus,+ B. lactis P. pentosaceus,* and *B. breve* ↑ Agathobaculum, Dorea (OTU 527923), Dorea (OTU 195044), Blautia, Ruminococcus, and Dorea (OTU 470168). Decrease the levels of Intrahepatic fat fraction , % total body fat and visceral fat TNF-α [51]
	S. thermophilus *L. delbrueckii* subsp. bulgaricus	220g of of yogurt per day, For 24 weeks.	Decrease the levels of LPS , ALT , Fat mass TNF-α [52]

	L. plantarum DSM 24730 *L. paracasei* DSM 24733 *L. delbrueckii* subsp *L. acidophilus* DSM 24735 *B. longum* DSM 24736 bulgaricus DSM 24734 *B. breve* DSM 24732 *S. thermophilus* DSM 24731 *B. infantis* DSM 24737	2×10^{11} Colony forming unit (CFU) Per day of probiotic strains, For 12 months.	Decrease the levels of Endotoxins ,Steatohepatitis ALP, AST, ALT ,TNF-α,IL-1β, IL-6 [53]
	Bifidobacterium *Lactobacillus* *Lactococcus Acetobacter* *Propionibacterium*	1×10^{10} CFU/day of *Bifidobacterium* 6×10^{10} Colony forming unit (CFU)/day of *Lactobacillus* and *Lactococcus* 1×10^{6} CFU/day of *Acetobacter*, for 8 weeks 3×10^{10} CFU/day of *Propionibacterium*	Decrease the levels of AST, TG, LDL-C TNF-α, IL-6 [54]
	L. acidophilus *B. lactis* *S. thermophilus* *L. bulgaricus*	300 g of yogurt per day of probiotic strains, For 8 weeks.	Decrease the levels of BMI, weight [55]
	L. acidophilus *L. casei* *L. bulgaricus* *L. rhamnosus* *B. longum* *S. thermophilus* *B. breve*	3×10^{10} Colony forming unit (CFU)day of *L. acidophilus* 3×10^{9} Colony forming unit (CFU)/day of *L. casei* 5×10^{8} Colony forming unit (CFU)/day of *L. bulgaricus* 7×10^{9} Colony forming unit (CFU)/day of *L. rhamnosus* 1×10^{9} Colony forming unit (CFU)/day of *B. longum* 2×10^{10} Colony forming unit (CFU)/day of *B. breve* 3×10^{8} CFU/day of *S. thermophilus*, for 8 weeks	Decrease the levels of TNF-α, IL-6 [56]

5. CONCLUSION

Some factors such as oxidative stress, HCV infection and Non-alcoholic steatohepatitis (NASH), alcohol abuse etc. That cause liver fibrogenesis in many ways and gut microbiota also cause hepatic steatosis and fibrogenesis. The beneficial effect may achieved by alteration and modification of gut micro biota in pathological condition. Probiotics are effective, economical, and absence of over time serious damage. The probiotics are therapeutic option for liver fibrogenesis.

The given evidences shows that Probiotics have antioxidant potency to reduce the elevated degree of reactive oxygen species (ROS) because it is responsible for HSCs activation and it will introducing the protective cytokines such as IL-10 and inhibiting the inflammatory cytokines and decrease the protein deposition.

Probiotics will have potential to reduce the fibrogensis in liver and it will be used for various diseases in future world.

6. FUTURE PERSPECTIVE

Probiotics have anti fibrotic potential in liver fibrogenesis which is approved as a drug of choice for liver fibrogenesis in future.

7. ABBREVIATIONS

ADM- Acellular dermal matrix
ALP-Alkaline phosphatase

ALT-Alanine aminotransferase
AST- Aspartate aminotransferase
CTGF-Connective Tissue growth factor
ECM- Extracellular Matrix
EMT- Epithelial mesenchymal transition
ET-Endothelin
FXR- Farnesoid X receptor
GFAP- Glial fibrillary acidic protein
GSH- Glutathione
HCC- Hepatocellular carcinoma
HSC- Hepatic stellate cells
HCV- Hepatitis C virus
IL- Interleukin
MDA- Malondialdehyde
MF-Myofibroblast

MMP-Matrix metallo proteinase
MMPC- Multipotent mesenchymal progenitor cells
MSC- Mesenchymal stem cells
NAFLD- Non-alcoholic fatty liver disease
NASH - Non-alcoholic steatohepatitis
NK Cells- Natural killer cells
NO-Nitric oxide
NF-κB- nuclear factor kappa B
PDGF-Platelet-derived growth factor
PPAR-γ-Peroxisome proliferate activated receptor-γ
ROS- Reactive oxygen species
SOD- Superoxide dismutase
TGF-β -Transforming Growth Factor-β
TIMP-Tissue inhibitors of metalloproteinases
TLR- Toll-like receptors
TNF- Tumor necrosis factor
VEGF-Vascular endothelial growth factor

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