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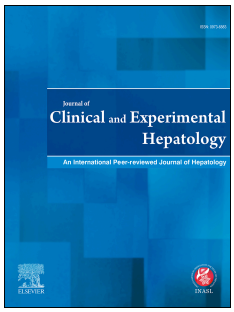

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Immunosuppression In Liver Transplant Recipients In The Setting Of Sepsis

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SUMMARY:

Management of immunosuppression (IS) in liver transplant recipients in the setting of sepsis is an open stage for debate. The age-long practice of reduction or complete cessation of IS during sepsis has been followed by most centres across the world, although, their exact strategies are highly heterogeneous. On the other hand, the emergence of striking new evidence suggesting that there is, in fact, decreased mortality with the continuation of IS in sepsis, has raised doubts about our previously conceived intuitive notion that IS portends increased risk in sepsis. The theory postulated is that IS agents, perhaps reverse the state of dysregulated immune response in sepsis to that of an iatrogenically modulated immune response, thus dimming the inflammatory cascade and preventing its deleterious effects. Of note, none of these studies reported exaggerated rejection-related complications. These contrasting outlooks have made it rather onerous to formulate an evidence-based recommendation for liver transplant recipients afflicted with sepsis. Inclusion of transplanted patients in randomized controlled trials of sepsis-related interventions seems to be the need of the hour.

INTRODUCTION:

The creation of a rejection-free milieu for transplanted solid organs by immunosuppression (IS) was a landmark achievement in the history of transplantation. Calcineurin inhibitors (CNI), steroids, anti-metabolites, and mTOR inhibitors are the IS drugs commonly used following liver transplantation.[1] These marvel drugs are, however, not bereft of adverse effects. The use of immunosuppression drugs is known to increase the chance of acquiring bacterial, viral, fungal and parasitic infections in the immediate as well as long-term post-transplant period.[2] Striking a fine

balance between preventing rejection and having adequate immunological control over infectious complications is of paramount importance with the use of immunosuppressive drugs.

PROBLEM AT HAND:

Sepsis, as defined by the Third International Consensus- SEPSIS 3 is "organ dysfunction caused by a dysregulated host response to infection". Septic shock is a subset of sepsis in which underlying circulatory and cellular/metabolic abnormalities are profound enough to substantially increase mortality. It is characterized by persisting hypotension requiring vasopressors to maintain a mean arterial pressure of 65 mm Hg and having a serum lactate level of >2 mmol/L (18 mg/dL) despite adequate volume resuscitation.[3] Understanding of the core molecular mechanisms behind the transition from localized inflammation to an acute adverse host response in severe sepsis is still incomplete. Despite advances in the management of sepsis, approximately 11 million deaths are caused by sepsis every year.[4] Between 2015 and 2019, nearly 10,000 liver transplants were performed in India.[5,6] The predominant cause of mortality, when extrapolated from western data is sepsis. The mortality amongst solid organ transplant recipients increases to as high as 50% when accompanied by septic shock.[7] Understandably, Sepsis occurring in transplant patients adds another layer of complexity and is the problem at hand in this review.

Withdrawal of IS in the setting of sepsis comes across as an instinctive step in the management of sepsis in liver transplant recipients. Perhaps the basis of this could be the publication by Starzl et al in 1994, where they opined regarding the importance of temporary withdrawal of IS drugs to allow host native response to revert to normal. In the study, 31 patients developed sepsis following liver transplantation, in whom IS was changed for all patients either as complete cessation (20 cases, 64%) or reduction of IS to steroid use only (11 cases, 36%). Mortality due to infection-related complications occurred in 13 (42%). None of the deaths was attributed to rejection although only 8 out of 13 had a liver biopsy. Of the 18 survivors, only 4 developed rejection on follow-up, all of which could easily be salvaged by rescue IS. They reiterated the feasibility of withdrawing IS in the setting of sepsis without the danger of acute rejection and the importance of doing so, in order to gain back the host immunity. The conjecture on the absence of rejection was based on two mechanisms. Firstly, the infection itself could have augmented the iatrogenically prescribed IS. Secondly, the recipient graft acceptance by the donor, secondary to a process of "tolerance induction" could have already been underway. [8]

Similar information was provided by S. Raia et al who analyzed 18 patients with 20 episodes of rejection, divided into a group with opportunistic infections (n=14) and a group with bacterial sepsis (n=5). Opportunistic infection referred to those due to viruses (cytomegalovirus, herpes

simplex), pneumocystis or tuberculosis occurring in iatrogenically over-immunosuppressed individuals. IS was discontinued in all cases, although steroid was discontinued after tapering. The median time to discontinuation was 20.5 days. Survival and rejection rates were respectively 50% and 25% in those with opportunistic infections as opposed to 17% and 50%in those with bacterial sepsis. Complications of infections were the cause of mortality in 90%. The authors noted the disparate outcomes of cessation of immunosuppression in opportunistic versus bacterial infections. In those with an opportunistic infection, rejection was lower, occurred later and resulted in lesser mortality whilst in recipients suffering from bacterial sepsis, rejection occurred earlier with more severity and resulted in overall much higher mortality. They concluded that the strategy of IS withdrawal in patients with bacterial sepsis may not be altogether beneficial, as they have a higher risk of graft rejection than those with opportunistic infections. [9]

CURRENT PRACTICES:

The real-world practices concerning the management of IS in transplant recipients with sepsis were evaluated by Daniel Shepshelovich et al, in a multinational survey with 33% response (124/381) from liver and kidney transplant centres. In the event of suspected bacterial sepsis, only 28% continued with CNI, 14% with anti-metabolite, 25% with mTOR inhibitors and in the case of steroids, 27% made no change while 8% increased the dose. On the other hand, re-initiation of CNI and anti-metabolite following sepsis reversal were carried out by >90% of liver centres.[10] While this data highlights the heterogeneity in the management of sepsis, it is striking that majority of transplant professionals are uniformly reluctant to continue IS in this scenario. The lack of head-to-head trials and high grade of evidence left us with a presumptive notion that withdrawal of IS is the apt strategy to adopt in the setting of sepsis in immunocompromised transplant recipients. Interestingly, there are new studies which go against this perceived wisdom and convention. Perhaps it is now time to re-visit the management of IS in liver transplant patients afflicted with sepsis.

NEWER CONCEPTS: (Table no. 1)

On a counter-intuitive note, authors, Kalil et al published a report comparing the outcomes of sepsis between solid organ transplant (SOT) recipients versus non-transplant patients. This was a novel venture since it was considered that patients on IS with sepsis perform worse than non-transplant patients with sepsis. Indeed, for this reason, almost all trials conducted for various aspects of sepsis would exclude septic transplant recipients. In

this matched, case-controlled, propensity-adjusted study, authors compared 123 transplant patients with 246 age, gender and hospital location matched controls. The outstanding finding was that after propensity-matched analysis, SOT patients had 78% lower 28-day mortality and 57% lower 90-day mortality than non-SOT patients. **To reiterate, transplant patients with blood culture-proven sepsis had significantly higher survival outcomes than identical non-transplant patients.** It is also worth noting that in this study, the transplant group of patients were comparatively sicker with a higher percentage of organ failure, septic shock and higher sequential organ failure assessment (SOFA) scores. The explanation provided by the authors was that in a state of sepsis which comes with gross dysregulated immune response, immunosuppressive agents bring about immune modulation and ameliorates the inflammatory response related organ failure. The authors call for the uniform inclusion of transplanted individuals in future trials of sepsis.[11] Information regarding modulation/discontinuation/reduction of immunosuppressant agents during periods of sepsis is lacking in the study. Therefore, direct extrapolation with regards to IS management in septic patients can only be presumptive.

John P Donnelly et al published a study along similar lines, comparing the outcomes of sepsis and severe sepsis between transplant recipients and non- transplanted individuals. This large retrospective cohort study using data from a consortium of academic medical centres compared 39,618 cases of SOT with 8,64,198 cases of non-SOT having severe sepsis. **SOT recipients hospitalized with severe sepsis had lower mortality rates compared to non-SOT patients (5.5% vs 9.4%).** These findings were restricted to liver and kidney recipients. Heart transplant patients showed no difference while increased mortality was seen in lung transplant recipients. This was despite the SOT group of patients having a higher incidence of diabetes mellitus, hypertension, renal failure, liver disease and deficiency anaemia. The authors postulate that, IS agents used at induction and maintenance interfere with the harmful inflammatory cascade that is known to be associated with the pathophysiology of sepsis. Blunting of this response in the hyper-inflammatory phase also brings about a lessened reciprocal anti-inflammatory response in the late phase of sepsis. This may lead to improved survival by stopping progression towards major inflammatory/coagulation responses[12]. Unlike Kalil et al study, this report doesn't provide matched analysis and may be biased by factors like the possibility of milder sepsis in SOT recipients or non-specialists care of the non-SOT group of patients. Akin to Kalil's paper, no mention is made with regards to IS therapy offered during periods of sepsis hence readers are left to assume the exact strategy of immunosuppression to be used in such scenarios. The authors mention the need for identifying the exact mechanisms involved in achieving these counter-intuitive results.

Michelle Bartoletti et al, in a retrospective single-centre observational cohort study described, IS management in transplant recipients with bloodstream sepsis at their centre over 10 years. Out of 209 episodes of bloodstream infections (BSI) in 157 liver recipients, 119 had no change of IS while 90 had either reduction or discontinuation. **Once again, the surprising finding was the significantly lower mortality in the group**

without any change in IS (22% to 7%; p=0.002) compared to the group with either reduction or discontinuation. On multivariate analysis, the only risk factor for mortality other than septic shock was the reduction in IS with a hazard ratio of 3.18, (95% CI 1.38-7.32; p=0.006). The theory that the authors put forth for these results is similar to the previously mentioned studies, that being, IS aids in the reversal from a dysregulated hyper-immune response to a state of regulated response. The authors conclude that the "habit" of reducing IS in BSI cases should be put into urgent scrutiny as the practice seems to be associated with worse outcomes.[13]

Authors Fang Chen et al conducted a retrospective cohort study in 2020, aimed at understanding the effect of temporary IS withdrawal in liver transplant recipients with severe infection. Out of 74 episodes of BSI in 70 recipients, IS reduction (at least 50% dose reduction or cessation of one or more immunosuppressive agents) was made in 41.2% of cases. **The authors on cox regression analysis showed that rejection and complete IS withdrawal were independent risk factors for 30-day mortality, especially for patients with Gram-negative bacterial (GNB) sepsis**. It must be mentioned that the data on mortality in this paper lacks granularity as head to head comparative data between IS withdrawal and IS continuation was presumptive.[14]

In a retrospective analysis by Maricar F. Malinis et al, it was found that the **all-cause mortality following Staphylococcus Aureus bacteremia was found to be less in solid organ transplant recipients in comparison to non- organ transplant patients (risk ratio: 0.37; p=0.02), despite having higher numbers of Methicillin resistance (86% vs. 52%, p<0.0001) and longer duration of bacteremia (mean 3.8 vs. 1.6 days, p<0.01).** The authors propose possible theories for the occurrence of such a phenomenon as earlier antibiotic administration in the SOT cohort, involvement of infectious disease specialists and the possibility of immunomodulatory effects of anti-rejection therapies. [15]

These findings mirror some of the recent evidence gathered from Covid-19 infection in liver transplant recipients. Not only were liver transplant recipients devoid of increased risk of mortality from covid-19 infection, compared to normal population, but immunosuppression ostensibly executed a protective effect on them and conferred a survival advantage. [16,17]

PATHOLOGICAL AND MOLECULAR BASIS OF SEPSIS:

Sepsis for many years has been considered to be the result of an uncontrolled inflammatory response to infection. The discovery of various cytokines such as interleukins and Tumour Necrosis Factors (TNF) led to the concept of "cytokine storm". Cytokines can be either pro-inflammatory or anti-inflammatory. Typically, sepsis has an initial hyper-inflammatory response characterised by fever, tachycardia and hypotension followed by a hypo-inflammatory state. While the pro-inflammatory state is primarily aimed at microbial destruction by innate and adaptive immunity, an overdose of this may lead to self-destruction of host tissues. Therefore, the hypo-inflammatory state, termed as Compensatory Anti-inflammatory Response Syndrome (CARS), functions as an "off-switch" to the hyper-inflammation and is equally vital to the survival of the host.

Significant scientific advance has transpired over the last decade in the understanding of the molecular factors driving sepsis. Bacteria, viruses, parasites, and fungi all possess unique cellular constituents *not found in humans*, referred to as Pathogen Associated Molecular Patterns (PAMPs). Non-infectious injuries such as trauma, ischemia or reperfusion release self-antigens termed Damage Associated Molecular Patterns (DAMPs) and include Heat shock protein, fibronectin, Hyluran, S 100 etc. Both exogenous PAMPs and endogenous DAMPs are recognised by specialised family of receptors called as Pattern Recognition Receptors (PRRs) such as Toll Like Receptors (TLRs). When TLRs are activated by PAMPS, the innate immune cells such as neutrophils, dendritic cells, monocytes and macrophages recognise the "non-human" pattern of the pathogen and phagocytose them. Subsequently two courses of actions take place. First, the pathogens are killed by lysozymes or reactive oxygen species(ROS)/reactive nitrogen species (RNS). This action does not require prior exposure and is of the same magnitude, irrespective of the number of prior exposures. Second, the antigen presenting cells (APC) which comprise the dendritic cells, monocytes and macrophages, after phagocytosis, convert the proteins of the digested pathogens into antigens and load them on their HLA class 2 Major Histocompatibility Complex (MHC). These are recognised by the T-cell receptors of CD4+T lymphocytes and trigger the highly specific adaptive immune response. Eventually, B cells are stimulated and converted to plasma cells which produce the precise antibody for the antigen. The adaptive response is not only explicitly specific but occurs with higher magnitude and rapidity, with each subsequent exposures of the antigen. The extent of pro-inflammatory response is plausibly driven by pathogen factors such as virulence and load as well as host factors that may include age, co-morbidity, genetic factors, and immune responsiveness. The pro-inflammatory response, when overwhelmed, leads to the familiar "cytokine storm" and culminates in early death, often within a week, due to cardiovascular collapse, metabolic dysfunction and multi-organ failure.

To prevent the cytokine storm, the CARS "switches off" the pro-inflammatory response by several mechanisms. These include a) down regulation of the antigen expression by APCs, especially monocyte, b) neutrophil cell death by necrosis c) lymphocyte cell death by apoptosis and d) secretion of anti-inflammatory cytokines such as IL-10, TGF beta. Often, the secondary hypo-inflammatory response may also become uncontrolled and may not "switch off", leading to a state of monocyte and lymphocyte anergy or "immunoparalysis". Now the patient becomes an easy prey for secondary infection and then succumbs to a repeat of hyper- hypo inflammatory cycle.

The receptors of PAMPs and DAMPs overlap spatially and temporally and exert several feedback loops that may send positive or negative signals to TLRs. There is a fine balance required at the molecular level, between up-regulatory signalling for transcription of pro-inflammatory cytokines and down-regulatory signalling to limit the endogenous apoptoptic pathways, for ultimately salvaging the patient from septic cascade. Recent studies have suggested that a subtype of lymphocyte population called T regulatory-cell (Tregs) may have the capacity to actively suppress an adaptive immune response and alter the dynamics of sepsis. Tregs appear to be involved with inducing lymphocyte anergy. Likewise, neutrophils and monocyte/macrophage elements play a dual role. While in the defensive mode, they engulf the pathogens and lyse them with proteolytic enzymes, inappropriate activation of these cells may result in dangerous extracellular release of cytotoxic molecules and self-tissue damage. Tight regulation of neutrophil and macrophage activation is, therefore, vital to maximise host defence and minimise its damage.

In transplant recipients who are on anti-rejection medications, the immunosuppressants may exert a protective effect against sepsis by two plausible mechanisms, which however have eluded confirmation.

1. The immunosuppressant drugs may limit the initial hyper-inflammatory response from being overactive and catastrophic to the host.
2. The regulated immunosuppression by the drugs may help in controlled apoptosis of the neutrophils and elements of monocyte/macrophage systems. This may partly be due to lower expression of Tregs in transplant recipients.

The mechanism of action of various IS agents and their corresponding role in the pathophysiology of sepsis has been explained in Table no. 2.

Of course, whether immunosuppressed transplant recipients do have a survival advantage over non transplanted patients and if so, what are the mechanisms underlying this curious outcome require further scientific illumination and validation.[4,18,19.]

OUR PRACTICE:

In the absence of rigorously evaluated evidence, our practice like most others' is based on conjecture and is perhaps, still old-fashioned. As lack of evidence prevents the recommendation of firm protocols, we practice an a-la-carte approach to individual patients' needs. If the patient is diagnosed with severe infection/sepsis **without alteration in the haemodynamic status**, we refrain from any obvious change in the ongoing IS strategy. We aim at maintaining a CNI trough level of 3 to 5ng/ml. In the event of renal dysfunction, we tend to withdraw tacrolimus completely and subsequently restart it once renal function improves. Likewise, in the presence of leukopenia (total count <3000cells/ml) and thrombocytopenia (platelets <40,000cells/ml), we stop the anti-metabolite agents. Concerning mTOR inhibitors, the drug is discontinued if there is bone marrow

depression or significant proteinuria. **In the presence of septic shock**, we withdraw tacrolimus and anti-metabolites completely and give hydrocortisone in doses of 50-100mg, 8th hourly. However, as soon as ionotropic supports come off, we re-start the previous IS. If there is suspicion of biochemical rejection during recovery, we give methylprednisolone pulses usually at the dose of 5mg/kg/body weight for 3 days.

CONCLUSION:

That, sepsis-afflicted transplanted patients on IS have higher mortality than non-IS patients, maybe a myth constructed on our intuitive beliefs. Current evidence points towards a protective effect of IS on the beneficial outcomes of sepsis. Therefore, it may not be necessary to withdraw IS in liver transplant recipients in this scenario. In fact, there is recent strong evidence in the literature to suggest that the continuation of IS in the face of sepsis may be the appropriate strategy. However, the tactic in presence of septic shock and organ failure will have to be modified on a case-by-case basis till firm evidence, preferably by randomized control trials become available.

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ABBREVIATION LIST:

IS - immunosuppression

CNI - calcinuerin inhibitors

MAP - mean arterial pressure

SOT - solid organ transplant

SOFA - sequential organ failure assessment

BSI - blood stream infections

GNB - Gram negative bacterial

PAMPs - pathogen associated molecular patterns

DAMPs- damage associated molecular patterns

TLR - toll like receptors

PRR – patterns recognition receptors

T regs – T-regulatory cells

APC - antigen presenting cells

HLA DR – human leukocyte antigen DR

IL-10 – interleukin 10

TGF- B – transforming growth factor - B

PD -1 – programmed death protein 1

ROS – reactive oxygen species

RNS – reactive nitrogen species

KEYWORDS:

Immunosuppression in liver transplant; Immunosuppression in sepsis; Immunomodulation; Immune dysregulation; Cytokine storm.

Table no. 1: List of comparative studies on sepsis afflicted transplant patients versus non-transplant patients.

Author	Type Of Study	Patients With Sepsis Included		Mortality		IS Modulation	Risk Factors Of Mortality	Rejection	Recommendation
		SOT	**non-SOT**	**SOT**	**Non-SOT**				
Kalil et al [11]	Matched, case-controlled propensity-adjusted study.	n = 123 Kidney-36.6% Liver-34.1% PK – 13% SB – 7.3% Heart/lung – 5.7% MV-3.3%	n= 246	**28-day crude mortality:** 8.1% (HR-0.22[95%CI; 0.09-0.54]; p=0.001). **90-day crude mortality:** 14.6% (HR-0.43[95% CI; 0.20-0.89]; p=0.025)	8.9% 9.8%	Not mentioned	SOT patients had 78% decreased 28day mortality and 57% decreased 90-day mortality	Not mentioned	IS associated with transplants may provide a survival advantage to recipients with sepsis.
John P Donelly et al [12]	Large, retrospective, cohort study.	n= 39,618 (4.4%) Kidney – 54.8%	n= 8,64,198 (95.6%)	**Severe sepsis:** 5.5%	9.4%	Not mentioned	**SOT patients had lower mortality.**	Not mentioned	SOT patients had decreased mortality with severe sepsis and sepsis.

		Liver – 18.2% Lung – 10% Heart – 7.8% Co-Transplant – 8.2% Other – 0.9%		[aOR = 0.83(0.79-0.87)] **Sepsis:** 8.3% [aOR – 0.78 90.73-0.84)]	12.7%		[OR -0.83; 95% CI (0.79-0.87)]		Need for identifying the specific mechanism.
Maricar F Malinis et al [15]	Single-centre, retrospective study.	n*= 70 Liver – 19 Lung – 26 Kidney – 18 Heart – 7 *patients infected with SAB	n*= 2889 *patients infected with SAB	**30- day mortality:** 6% (p= 0.002) **1-year mortality:** 28%	21% 44%	Not mentioned	**SOT status was associated with decreased mortality**. [RR – 0.37; 95%CI 0.12-0.88; (p=0.02)]	Not mentioned	30-day all-cause mortality decreased in SOT patients. IS drugs might have a role to play.
		No IS change	**IS changed**	**No IS change**	**IS changed**				
Michelle bartoletti et al [13]	Single centre, retrospective, observational cohort study.	n= 119 (all LT recipients with BSI)	n= 90 (all LT recipients with BSI)	7% (p= 0.002)	22%	**Any reduction** = 90 (43%). **Dose reduction** = 31 cases (11 Tac, 8 steroids, 7 others, 5 Tac + steroids). **Discontinuation** = 28 cases	**Septic shock** [aHR- 2.42; 95%CI 1.05-5.56; (p=0.04)] **Reduction in IS** [aHR 3.18; 95%CI	n=21 (10%) overall rate. No difference between groups.	IS reduction leads to worse outcomes in LT patients with BSI. Need for “sharp discussion” with regards to IS management in sepsis.

						(7 Tac, 9 steroids, 12 others). **Reduction + discontinuation of any 1 drug** = 13 cases. **Complete withdrawal** = 18 cases.	1.38-7.32; (p=0.006)]		A multidisciplinary approach, timely antibiotics and source control are more important.
		LT recipients with GPC	**LT recipients with GNB**	**GPC group**	**GNB group**				
Fang Chen et al [14]	Single centre, retrospective cohort study.	n= 42 (Total BSI 74 out of 70 LT recipients)	n= 28	n=2 (4.8%) (Cause - Graft versus host disease)	n= 11 (39.3%) (Cause - worsening infection associated with IS withdrawal)	**IS reduction** = 28 cases (41.2%) **GPC** – 5 cases (17.9%) **Dose reduction** = 2 (Tac) **Complete withdrawal =** 3 **GNB –** 23 cases. **Dose reduction** = 6 (Tac, Steroid , Cyclosporine, MMF).	**Rejection within 90 days of BSI** (p= 0.01) **Complete IS withdrawal** (p=0.019).	GPC group = 3 (7.1%) GNB group = 7 (25%).	Higher tendency to reduce IS in GNB infections more than GPC infections. Increased mortality with IS withdrawal in GNB infections.

						Single drug discontinuation = 1 **Both dose reduction and discontinuation of one drug** = 1 **Complete withdrawal** = 15.			

ABBREVIATIONS:
IS – immunosuppression; SOT – solid organ transplant; PK – pancreas-kidney; SB –small bowel; MV – multi-visceral; HR – hazard ratio; CI – confidence interval; aOR – adjusted Odds ratio; SAB – Staphylococcus Aureus Bacteremia; RR – risk ratio; BSI – blood stream infection; Tac-Tacrolimus; MMF – Mycofenolate Mofetil; LT – liver transplantation; GPC – Gram positive cocci; GNB – Gram negative bacilli.

Table no. 2: Mechanism of IS and role in sepsis pathophysiology.[20-22]

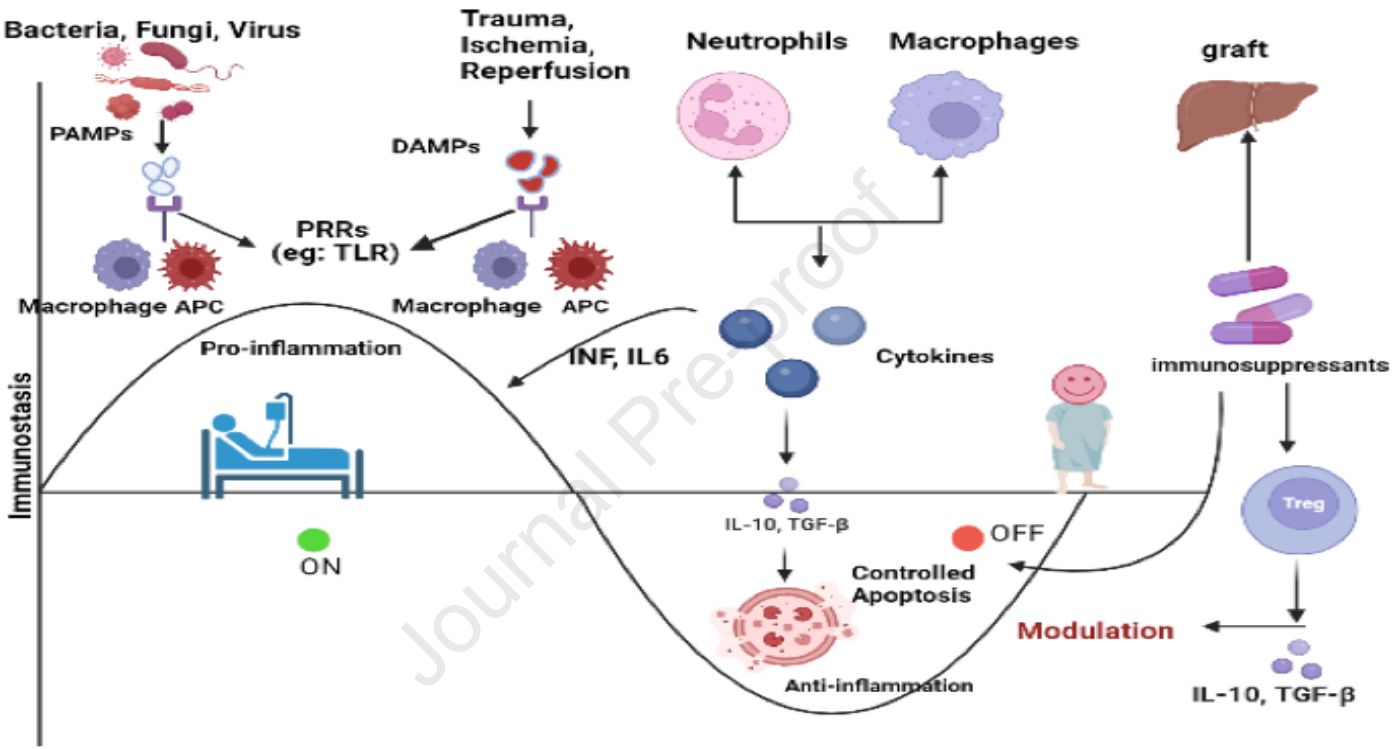
TYPE OF IS	MECHANISM OF ACTION	MODULATION IN SEPSIS
Calcineurin Inhibitors • Cyclysporine • Tacrolimus	Inhibit calcineurin which results in re-phosphorylation of nFAT (nuclear factor for activated T cells). This prevents translocation of nFAT from cytoplasm to nucleus and	IL2 has a dual role in sepsis- while in the initial stage, it promotes the hyper-inflammation, later in septic cascade, it has a pro-apoptotic effect. Additionally, IL-2, prompts division of Tregs when their numbers become critical and may provoke lymphocyte anergy. Perhaps tacrolimus may blunt these effects of IL-2 and smoothen both the hyper and hypo inflammatory cascade of sepsis.

	inhibits transcription of IL2. Thus final action is through their inhibition of IL2 production.	
Steroids	Has wide ranging effects on innate and adaptive immunity. Steroids depress innate immunity by activating receptors that induce anti-inflammatory mediator genes (trans activation) and represses factors that incite prolonged inflammation such as nf Kappa b (transrepression). They also inhibit adaptive immunity by deactivating T cell stimulation via multiple pathways.	Steroids are thought to ultimately reprogram the hyper- hypo inflammatory cycle such that there is more regulated immune response by host. Additionally, steroids mitigate monocyte anergy and prevent adrenal insufficiency, both of which accompany severe sepsis. There is evidence of restoration of cardiovascular and other organ function by steroids although precise mechanism is as yet unknown, but may include its effect on sodium retention, increasing catecholamine sensitivity and augmenting microcirculatory perfusion.
Target of rapamycin inhibitors • Everolimus • Sirolimus	Binds to the cytosolic immunophilin FK-binding protein 12 (FKBP12). This complex binds to and inhibits the activation of the mammalian target of rapamycin (mTOR), a key regulatory kinase. This inhibition suppresses cytokine-mediated T-cell proliferation inhibiting the progression from the G1 to the S-phase of the cell cycle.	mTor may accelerate CD4+ apoptosis in sepsis and lead to immunoparalysis. This maybe ameliorated by mTOR inhibitors and result in a more organized cellular homeostasis, whereby the molecules and organelles are recycled rather than degraded. This has been recently termed as “autophagy” as opposed to apoptosis and purportedly has a protective effect in sepsis.
Antimetabolites • Azathioprine	Metabolized to 6-MP which interferes with normal purine	Not known.

	synthesis by affecting DNA and RNA synthesis thus suppressing the proliferation of B & T lymphocytes. Also reduces the number of circulating monocytes.	
Purine synthesis inhibitors • Mycofenolate Mofetil	Impairs lymphocyte function by blocking purine biosynthesis via inhibition of the enzyme inosine monophosphate dehydrogenase. MPA impairs the ability of dendritic cells to present antigen, suppresses the recruitment of monocyte lineage cells, suppresses the glycosylation of adhesion molecules, inhibits vascular smooth muscle proliferation, improves endothelial function, and inhibits mononuclear cell recruitment into allografts	Not known, but may have a deleterious effect.

ABBREVIATIONS:
IL 2 – Interleukin 2; T-regs - T-regulatory cells; 6-MP – 6- Mercaptopurine.

Figure 1: Usual response to sepsis

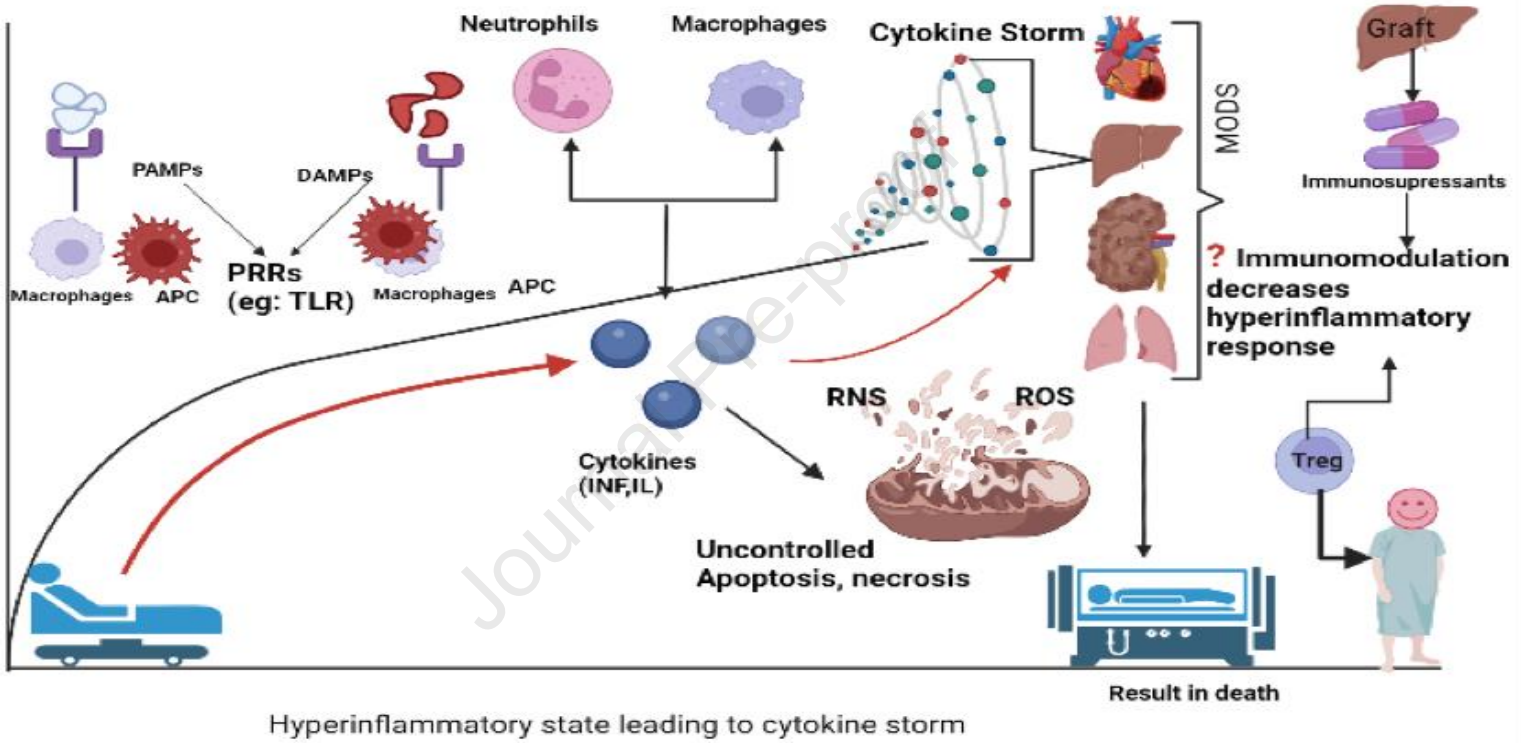


Typically, following sepsis, a hyper-inflammatory response characterized by fever, tachycardia and hypotension occurs. This may be due to exogenous bacteria/virus/fungi releasing PAMPs or trauma, ischaemia reperfusion releasing DAMPs. PAMPs and DAMPs are recognized by PRRs like TLR which leads to pro-inflammatory cytokine release. This phase is aided by innate immune cells like neutrophils and macrophages causing phagocytosis. All these elements additionally have a down-regulatory signaling pathway as well. This is particularly seen during the controlled apoptosis of neutrophils and macrophages. Simplistically, the "on" switch begins the pro-

inflammatory phase while the "off" switch shuts it down. IS perhaps modulate both these phases, particularly the anti-inflammatory phase.

Abbreviations: PAMPs - pathogen associated molecular patterns; DAMPS- damage associated molecular patterns; PRR – patterns recognition receptors; TLR - toll like receptors; IS – immunosuppressants.

Figure 2: Hyper-inflammatory state leading to cytokine storm.



Hyperinflammatory state leading to cytokine storm

In this state, the anti-inflammatory phase does not occur at all. Instead of controlled apoptosis of the innate cells there is uncontrolled

apoptosis and necrosis of neutrophils and macrophages thus releasing ROS and RNS, resulting in multi-organ failure and death. Whether immunosuppressant drugs ameliorate this response and leads to better survival needs further scientific rigor.

Abbreviations: ROS – reactive oxygen species; RNS – reactive nitrogen species.