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Hepatoprotective Effect of Merlot Grape Pomace Extract against Acetaminophen-Induced Damage in Mouse

By Viviane Cassanelli Spader, Diego de Carvalho, Conrado de Oliveira Gamba,
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Abstract- This study investigated the benefits of Merlot grape pomace extract (EBUM) in experimental acetaminophen-induced intoxication. Swiss mice received the respective treatments for 7 days: Control (saline), Silymarin (100 mg/Kg), EBUM: 100 mg/Kg and 500 mg/Kg. On the seventh day, the acetaminophen control, Silymarin and EBUM received a single dose of acetaminophen 500 mg/kg. After 24 hours, the mice were sacrificed, the blood and liver were collected for analysis of serum transaminases (ALT and AST), bilirubin, hepatic markers of oxidative stress and histopathological analysis. EBUM was able to prevent acetaminophen-induced hepatic necrosis with a significant reduction ($p < 0.0001$) in ALT and AST, bilirubin, as well as a significant increase in hepatic GSH and CAT levels. The histological data are consistent with the transaminase results, showing less liver damage in the treated groups when compared to the acetaminophen group. The results allow us to infer that EBUM has a hepatoprotective effect against intoxication caused by acetaminophen in animal model, proving to be a therapeutic alternative in preventing drug-induced liver damage.

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Hepatoprotective Effect of Merlot Grape Pomace Extract against Acetaminophen-Induced Damage in Mouse

Efeito Hepatoprotetor do Extrato de Bagaço de Uva Merlot Contra Danos Induzidos Por Acetaminofeno em Camundongos

Viviane Cassanelli Spader ^α, Diego de Carvalho ^σ, Conrado de Oliveira Gamba ^ρ, Natan Veiga ^ω,
Karine Luz [‡] & Claudriana Locatelli [§]

Abstract- This study investigated the benefits of Merlot grape pomace extract (EBUM) in experimental acetaminophen-induced intoxication. Swiss mice received the respective treatments for 7 days: Control (saline), Silymarin (100 mg/Kg), EBUM: 100 mg/Kg and 500 mg/Kg. On the seventh day, the acetaminophen control, Silymarin and EBUM received a single dose of acetaminophen 500 mg/kg. After 24 hours, the mice were sacrificed, the blood and liver were collected for analysis of serum transaminases (ALT and AST), bilirubin, hepatic markers of oxidative stress and histopathological analysis. EBUM was able to prevent acetaminophen-induced hepatic necrosis with a significant reduction ($p < 0.0001$) in ALT and AST, bilirubin, as well as a significant increase in hepatic GSH and CAT levels. The histological data are consistent with the transaminase results, showing less liver damage in the treated groups when compared to the acetaminophen group. The results allow us to infer that EBUM has a hepatoprotective effect against intoxication caused by acetaminophen in animal model, proving to be a therapeutic alternative in preventing drug-induced liver damage.

1. INTRODUÇÃO

A hepatite medicamentosa, também conhecida como lesão hepática induzida por medicamentos ou *Drug Induced Liver Injury (DILI)*, constitui uma forma de reação adversa a medicamentos (Lunardelli et al., 2020). Podendo se manifestar dentro de um período de um a noventa dias após a administração de um medicamento (Silva et al., 2023).

A avaliação laboratorial da hepatite tóxica é abrangente e diversificada, especialmente em relação ao tipo de medicamento envolvido. Alterações nos níveis séricos das enzimas hepáticas aspartato aminotransferase (ALT) e alanina aminotransferase (AST) são consistentemente detectadas, juntamente com elevações na bilirrubina e modificações no tecido hepático (Mancilha et al., 2020).

O acetaminofeno (APAP) é um fármaco de alta biodisponibilidade, que possui potente ação analgésica

e antipirética e discretos efeitos anti-inflamatórios. Quando biotransformado, origina metabólitos sulfatados e glucuronizados, inativos por conjugação. Além disso, uma pequena fração é excretada inalterada na urina e a fração restante é metabolizada pelo sistema enzimático citocromo *CYP450* com ênfase para as enzimas *CYP3A4* e *CYP1A2*, as quais levam a formação do N-acetil-p-benzoquinoneimina (NAPQI). A glutatona desempenha o papel crucial de conjugação e desintoxicação do NAPQI. Entretanto, em situações de ingestão elevada do medicamento, observa-se uma redução nos níveis de glutatona, resultando na liberação do metabólito para se ligar aos hepatócitos, desencadeando assim, danos ao fígado (Sousa et al., 2023); Ribeiro et al., 2023; Vieira & Franca, 2015).

A ingestão de altas doses de APAP tem sido relatada em diversos estudos que demonstram a correlação da overdose com a insuficiência hepática aguda (IHA). A ingestão máxima diária de APAP é de 4 g/dia, de acordo com o *Food and Drug Administration (FDA)*, órgão regulamentador americano. Nos Estados Unidos e no Reino Unido os casos de IHA e mortes associadas à overdose chegam à 50% e 70% respectivamente. Em contrapartida, países como Alemanha e Espanha aparentam ter comportamento diferente em relação à automedicação, visto que apenas 3 a 9% dos casos de IHA ocorrem por sobredosagem de APAP (Bunchorntavakul & Reddy, 2018; Blieden et al., 2014).

A hepatotoxicidade do APAP vem sendo estudada exaustivamente nos últimos anos e grande parte dos mecanismos envolvidos nesse processo encontram-se elucidados (Torres et al., 2019). Desta forma, é possível utilizar o APAP como indutor de dano hepático em modelo animal com a finalidade de estudar sua toxicidade e de avaliar o uso de potenciais substâncias hepatoprotetoras.

Os antioxidantes são compostos presentes em vários vegetais que fazem parte da dieta humana. Em concentrações inferiores às do substrato oxidável, essas substâncias têm a capacidade de eficazmente inibir ou retardar o processo de oxidação (Macedo et al., 2020).

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Um dos agentes antioxidantes que opera de maneira semelhante à glutatona é a N-acetilcisteína (NAC). Esta substância reage com os radicais livres nos grupos sulfidríla, doando elétrons e se transformando em cistina. Além disso, pode atuar como substrato na síntese de glutatona hepática. Dessa forma, a NAC desempenha um papel significativo no tratamento de lesões hepáticas originadas por radicais livres, como aquelas decorrentes da hepatotoxicidade induzida pelo APAP (Oliveira & Costa, 2021).

A NAC tem sido reconhecida como o antídoto primário para casos de overdose de APAP, sendo administrada tanto por via intravenosa quanto oral. Atualmente, a NAC é considerada o tratamento padrão e essencial para casos de intoxicação por APAP (Moreira, 2016).

Os resíduos resultantes do processamento de frutas constituem uma fonte valiosa de compostos bioativos, que demonstram potencial aplicação na produção de cosméticos e fármacos com propriedades anti-inflamatórias e antioxidantes (Haghighatdoost et al., 2020). O Extrato de Bagaço de Uva (EBU), frequentemente subutilizado, emerge como uma fonte significativa de compostos fenólicos, oferecendo benefícios nas áreas alimentícia, cosmética e farmacológica (Nascimento, 2019). A composição diversificada do EBU, compreendendo sementes, cascas e engaço, revela uma considerável quantidade de componentes bioativos, como óleos essenciais, fibras, proteínas e complexos fenólicos (Bocsan et al., 2022).

O resveratrol é um polifenol natural identificado em diversas espécies de plantas, sendo uma delas a videira (*Vitis vinifera*). É considerado um potente antioxidante e neutralizador de radicais livres. O EBU é uma fonte rica de polifenóis antioxidantes (Chupradit, et, al. 2022), que por sua vez possuem propriedades benéficas para a saúde, prevenindo o câncer e a eliminação de radicais livres, doenças cardiovasculares, inflamações e distúrbios relacionados ao envelhecimento, além de capacidade hepatoprotetora (Dezena et, al. 2023).

O presente estudo objetivou investigar o efeito hepatoprotetor do Extrato de Bagaço de Uva Merlot (EBUM) em lesão hepática induzida experimentalmente por APAP em camundongos Swiss.

II. METODOLOGIA

a) Obtenção do Extrato Hidroalcolico do Bagaço de Uva Merlot (EBUM)

Os EBUM utilizados nesta pesquisa foram obtidos pela Empresa de Pesquisa Agropecuária e Extensão Rural (EPAGRI), o qual foi transportado ao laboratório e secos em estufa de aeração. Para a obtenção do extrato foi pesado 20 g de bagaço de uva Merlot e adicionadas em 100 mL de solvente água e

etanol (50:50 v/v). As soluções foram trituradas em um liquidificador industrial por 3 minutos, após a trituração a solução foi filtrada em bomba de vácuo para a extração do solvente e os filtrados obtidos foram concentrados por destilação a pressão reduzida com auxílio de um rotaevaporador. Após a evaporação do etanol, as amostras foram levadas ao liofilizador. Assim obtendo o EBUM).

b) Animais e Delineamento Experimental

Foram utilizados 70 camundongos albinos, machos, da linhagem Swiss, com 60 dias de idade e pesando aproximadamente 25 g. Os animais não foram submetidos a condições de estresse ou qualquer tipo de sofrimento. Não houve restrição de água ou alimento. O experimento foi conduzido dividindo-se os animais em 7 grupos com 10 animais cada. O grupo 1 foi denominado grupo controle (G1), recebendo apenas solução salina. Os grupos denominados 2 a 5, receberam, durante 7 dias, o seguinte tratamento: grupo 2 (APAP 500 mg/kg, em solução salina, no sétimo dia); grupo 3 (silimarina 100 mg/kg, em solução salina, durante 7 dias e APAP 500 mg/kg, em solução salina, no sétimo dia); grupo 4 (EBUM 100 mg/kg, em solução salina, durante 7 dias e APAP 500 mg/kg em solução salina, no sétimo dia); grupo 5 (EBUM 500 mg/kg, em solução salina, durante 7 dias e APAP 500 mg/kg em solução salina, no sétimo dia); grupo 6 (EBUM 100 mg/kg, em solução salina, durante 7 dias); grupo 7 (EBUM 500 mg/kg, em solução salina, durante 7 dias). No oitavo dia todos os animais foram eutanasiados. A descrição do experimento está demonstrada na Figura 1.

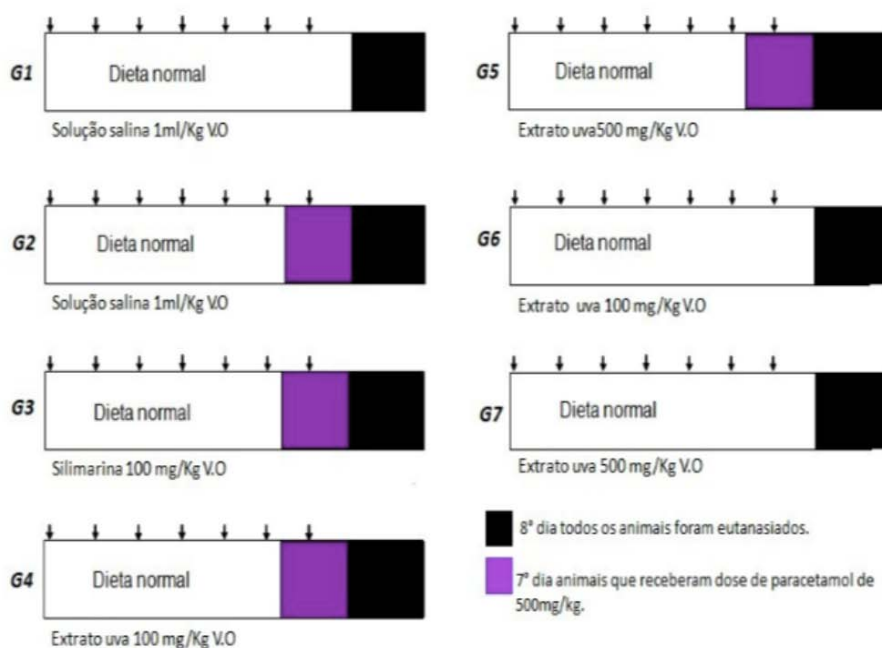


Figura 1: Delineamento experimental representando a divisão de grupos do estudo.

Este estudo foi encaminhado ao Comitê de Ética para Uso de Animais CEUA-UNOESC para realização dos ensaios com camundongos, obtendo parecer favorável sob número 75/2018.

c) Coleta do Material Biológico e Processamento Histológico

Após o final do tratamento os animais foram eutanasiados, segundo protocolo do comitê de ética e pesquisa com animais. A eutanásia deu-se após utilização de quetamina injetável acrescido de lidocaína. O sangue foi coletado em tubo seco através de punção cardíaca, e as amostras de soro foram armazenadas a -20°C até a análise. Imediatamente após, o fígado foi coletado, pesado e dividido em duas seções, uma para análise histopatológica e outra para avaliação do estresse oxidativo.

d) Análise de Biomarcadores Séricos de Dano Hepático

A função hepática foi avaliada por meio das determinações dos níveis séricos de transaminases (Aspartato aminotransferase (AST), Alanina aminotransferase (ALT)) e bilirrubina total (BT). Em tais determinações, foram utilizados kits reagentes da Labtest Diagnóstica S.A., e as análises foram realizadas conforme instruções do fabricante (Labtest, Lagoa Santa-MG).

e) Preparo dos Homogeneizados Para Avaliação do Estresse Oxidativo

O tecido hepático foi retirado para avaliação dos parâmetros oxidativos, Substâncias Reativas ao Ácido Tiobarbitúrico (TBARS), Glutathiona Reduzida (GSH) e Catalase (CAT). Para determinação de TBARS e CAT foi preparado um homogeneizado de fígado em

tampão fosfato 0,2 M pH 7,4, e para determinação de GSH foi preparado um extrato ácido com ácido tricloroacético 12%. As amostras foram misturadas em homogeneizador e centrifugados a 12000 rpm por 4 min à 4°C. As alíquotas necessárias foram utilizadas nos testes propostos e o restante foi armazenado congelado.

f) Avaliação dos Parâmetros Oxidativos

A avaliação da peroxidação lipídica foi realizada através da determinação dos níveis hepáticos de TBARS. Este método avalia o dano celular, como descrito por Bird e Draper (1984), e baseia-se na oxidação provocada por espécies reativas de oxigênio em biomoléculas tais como lipídios, carboidratos, ácidos nucleicos, levando à formação de substâncias que reagem com o ácido tiobarbitúrico (TBARS). O nível da lipoperoxidação é indicado pela formação de malondialdeído (MDA) e outras substâncias que reagem ao TBARS. A absorvância foi monitorada a 535nm. A concentração de TBARS na amostra foi calculada a partir da curva analítica de MDA e os resultados foram expressos como μmol de MDA/mg de proteína.

A avaliação da concentração de GSH foi realizada pelo método de Tietze (1969). Grupamentos sulfidrilas da GSH interagem com DTNB (ácido 5,5'-ditiobis (ácido 2-nitrobenzóico), obtendo-se o GSTNB (forma oxidada da GSH), há liberação de TNB (ácido 5-tio-2-nitrobenzóico). Sendo assim, a intensidade de cor produzida pelo TNB é diretamente proporcional à atividade da glutathiona redutase sobre a GSTNB e dos níveis de GSH intracelular. A absorvância foi monitorada a 412 nm. Os resultados foram expressos em mM/mg proteína.

Atividade da enzima antioxidante CAT foi avaliada pelo método descrito por Aebi (1984), que quantifica a velocidade de decomposição do peróxido de hidrogênio (10mM) em 240nm, pela enzima presente nas amostras. Os resultados foram expressos em mol H₂O₂/min/mg de proteína.

A determinação do teor proteico das amostras foi realizada por dosagem espectrofotométrica através da curva padrão de albumina bovina pela técnica de Lowry (1951).

g) Processamento Histológico

As amostras para análise histológica foram fixadas em formalina tamponada neutra a 10%, embebida em parafina, cortadas na espessura de 4 µm, coradas com hematoxilina e eosina; posteriormente avaliadas por microscopia óptica e classificadas conforme os seguintes escores: escore 0 (sem necrose); escore 1 (necrose <10% do fígado); escore 2 (necrose entre 10-25% do fígado); escore 3 (necrose entre 25-50% do fígado); escore 4 (necrose >50% do fígado) (LIU et al., 2018).

h) Análise Estatística

Para análise estatística os resultados da dosagem da AST, ALT, BT, TBARS, GSH e CAT, foram tabulados utilizando o software Microsoft Excel® e analisados estatisticamente no programa Prisma 9.0.0.

A diferença entre os grupos foi comparada pelo teste ANOVA ONE-WAY seguido pelo teste de Turkey de múltiplas comparações. As diferenças observadas durante a análise foram consideradas estatisticamente significativas quando a probabilidade foi menor que 0,05 (5%).

III. RESULTADOS

a) Biomarcadores Séricos de Dano Hepático

A administração de uma dose tóxica de 500 mg/Kg de APAP em camundongos Swiss (G2) aumentou significativamente os níveis séricos de AST, ALT e BT (Figura 2), quando comparado ao grupo controle (G1). Os grupos tratados com EBUM 100 (G4) e 500 mg/Kg (G5) preveniram significativamente (p< 0,0001) o dano hepático induzido por APAP, quando comparado ao grupo G2. O grupo G3 tratado com silimarina 100 mg/Kg foi capaz de prevenir significativamente (p< 0,0001) o aumento dos níveis séricos de BT e AST induzido pelo APAP sem, no entanto, reduzir os níveis de ALT.

Conforme pode ser observado na Figura 2, os animais tratados somente com EBUM (G6 e G7) não apresentaram modificações no nível sérico de ALT, AST e BT quando comparado ao grupo controle (G1) que recebeu somente salina.

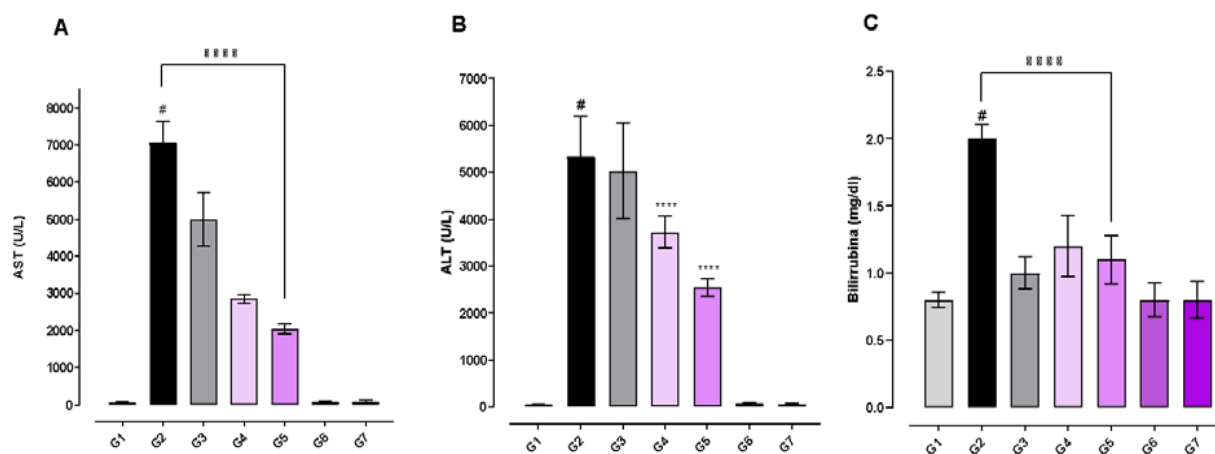


Figura 2: Análise de biomarcadores séricos de dano hepático em camundongos Swiss intoxicados com dose única de acetaminofeno (500 mg/Kg) e pré-tratados com silimarina ou Extrato de Bagaço de Uva Merlot (EBUM). Aspartato amino transferase (AST) e Alanina aminotransferase (ALT). Cada barra representa a média ± o desvio padrão da média. # (p< 0,0001) comparado com G1, * (p< 0,0001) comparado com G2.

b) Biomarcadores Hepáticos de Estresse Oxidativo

Os resultados mostrados na Figura 3 indicam que o tratamento dos camundongos Swiss com APAP na dose 500 mg/Kg foi capaz de induzir um estresse oxidativo, aumentando significativamente os níveis hepáticos de TBARS e reduzindo os níveis de GSH (Figura 3A e 3B). Quando os animais foram submetidos ao pré-tratamento durante 7 dias com silimarina ou

EBUM o dano hepático oxidativo induzido por APAP foi prevenido (Figuras 3A, 3B e 3C).

Os animais submetidos ao tratamento somente com EBUM (G6 e G7), melhoram as defesas antioxidantes com redução significativa (G7) nos níveis de TBARS (Figura 3A).

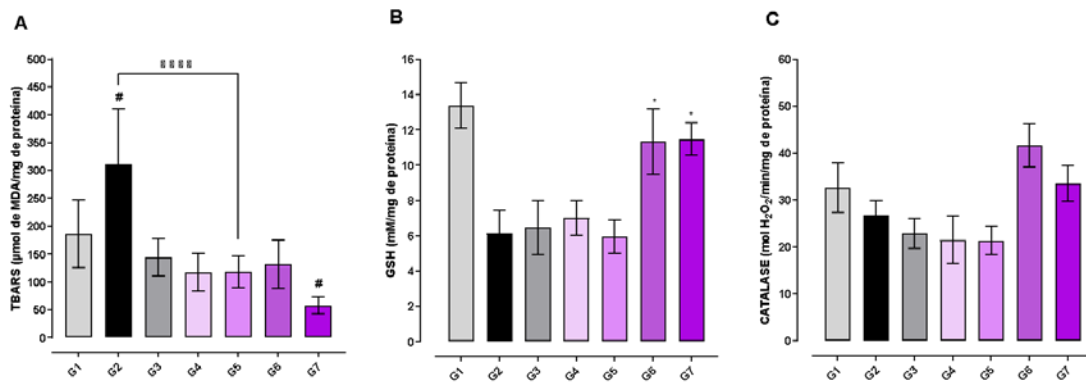


Figura 3: Efeito do Extrato de Bagaço de Uva Merlot (EBU) nos parâmetros de estresse oxidativo em camundongos Swiss intoxicados com dose única de acetaminofeno (500 mg/Kg). Ácido Tiobarbitúrico (TBARS). Glutathiona reduzida (GSH). Catalase (CAT). Cada barra representa a média \pm o desvio padrão da média. # ($p < 0,0001$) comparado com grupo controle G1; **** ($p < 0,0001$) comparado com G2; * ($p < 0,05$) comparado com G1.

c) Análise Histológica

Na análise histológica, observou-se que o G2 (Tabela 1 e Figura 4B), que recebeu uma dose de APAP (500 mg/kg), apresentou necrose centrolobular multifocal, com predominância no escore 3 (3/6, 50%) ou seja, apresentando um nível de necrose de até 50% do fígado. O G3 (Tabela 1 e Figura 4C) que recebeu pré-tratamento com silimarina (100mg/ Kg), demonstrou predominância nos escores 3 (2/6, 33,3%) e 4 (2/6, 33,3%), apresentando um nível de necrose hepático de

50% na maioria dos animais. O G4 (Tabela 1 e Figura 4D), que recebeu a dose de APAP (500 mg/kg) e pré-tratamento com EBUM (100 mg/Kg), exibiu predominância do escore 3 (4/6, 66,6%), mostrando um nível de necrose de até 50% do fígado. O G5, que recebeu a dose de APAP (500mg/kg) e pré-tratamento com EBUM (500 mg/Kg), houve predominância do escore 2 (4/6, 66,6%), (Tabela 1 e Figura 4E), com um nível de necrose hepática igual ou inferior a 25%.

O G6 e G7 não apresentaram desenvolvimento de necrose, mostrando morfologia hepática normal similar ao G1, conforme a Tabela 1 e Figuras 4A, 4F e 4G.

Tabela 1: Análise Histológica do tecido hepático de camundongos tratados com EBUM, classificadas por escore.

Grupos	APAP	N	Escore [n (%)]				
			0	1	2	3	4
Grupo1	-	6	6 (100)	0 (0)	0 (0)	0 (0)	0 (0)
Grupo 2	500 mg/kg	6	0 (0)	1 (16,7)	1 (16,7)	3 (50)	1 (16,7)
Grupo 3	500 mg/kg	6	0 (100)	1 (16,7)	1 (16,7)	2 (33,3)	2 (33,3)
Grupo 4	500 mg/kg	6	1 (16,7)	1 (16,7)	0 (0)	4 (66,6)	0 (0)
Grupo 5	500 mg/kg	6	0 (0)	0 (0)	4 (66,6)	2 (33,3)	0 (0)
Grupo 6		6	6 (100)	0 (0)	0 (0)	0 (0)	0 (0)
Grupo 7		6	6 (100)	0 (0)	0 (0)	0 (0)	0 (0)

Escore 0 (sem necrose); escore 1 (necrose <10% do fígado); escore 2 (necrose entre 10-25% do fígado); escore 3 (necrose entre 25-50% do fígado); escore 4 (necrose >50% do fígado). Esses resultados podem ser visualizados na Figura 4, que inclui fotomicrografias representativas e a porcentagem de área de necrose hepática.

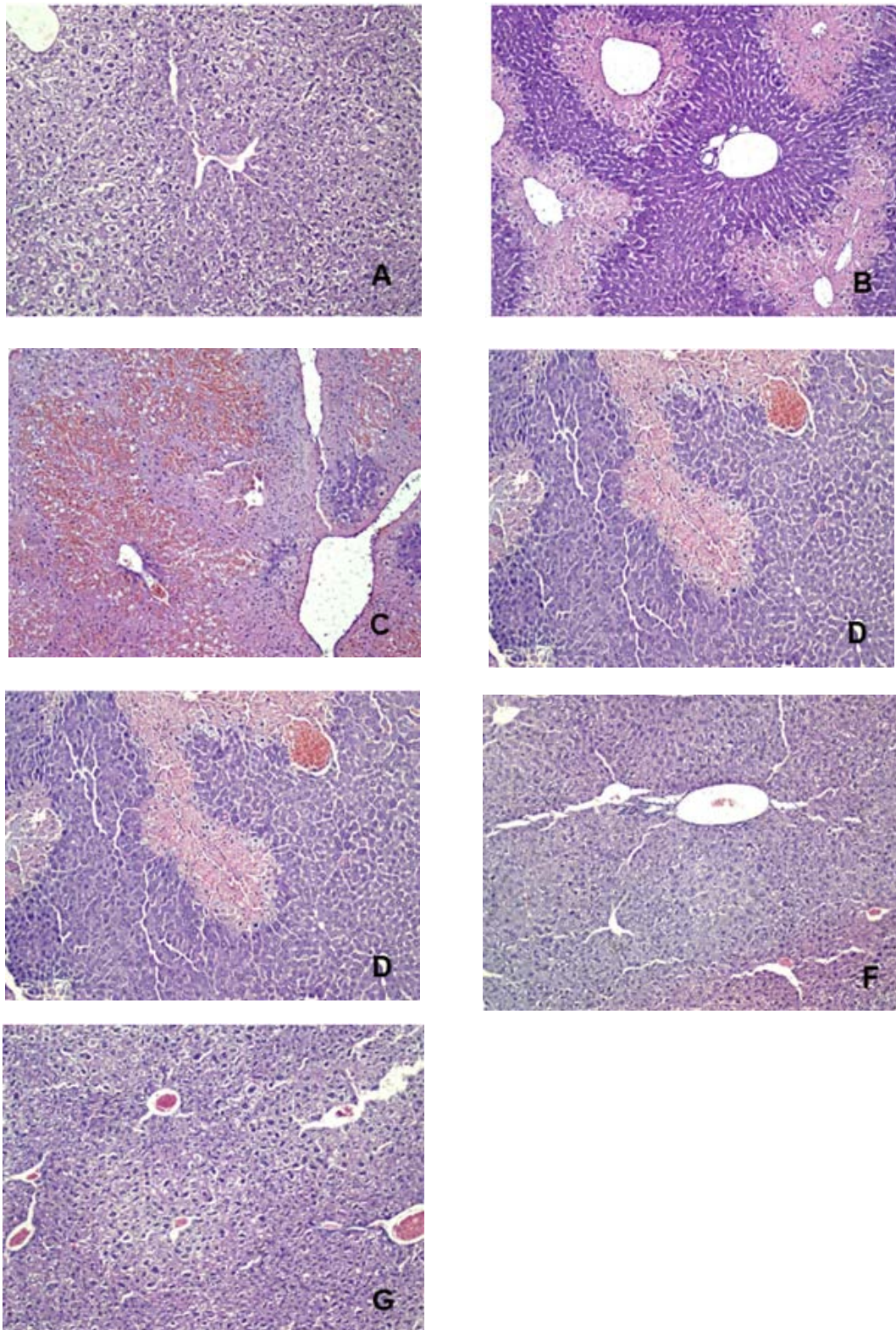


Figura 4: Análise histopatológica do tecido hepático em camundongos Swiss intoxicados com dose única de acetaminofeno (500 mg/Kg). As fotomicrografias representativas e a porcentagem da área de necrose hepática foram obtidas a partir de seções de fígado conforme demonstrado nas Figuras 4A, 4F e 4G, que retratam a histologia hepática normal para os Grupos 1, 6 e 7, respectivamente. A Figura 4B ilustra a histologia hepática para o Grupo 2, que não recebeu nenhum pré-tratamento, enquanto as Figuras 4C, 4D e 4E representam os Grupos 3, 4 e 5, respectivamente. Todas as imagens foram coradas com hematoxilina e eosina (HE) e capturadas com um aumento de 100 x.

IV. DISCUSSÃO

Estudos demonstraram toxicidade e injúria no fígado, induzidos por doses elevadas de APAP (S. Torres et al., 2019; Chao et al., 2018; Ramachandran & Jaeschke, 2019), assim como a elevação dos níveis séricos das transaminases e BT, importantes indicadores de lesão hepática. Essas transformações levam a uma alteração da homeostase, elevando a permeabilidade da membrana celular, resultando em edema, perda de conteúdo celular e dano aos hepatócitos (Ahmed et al., 2019).

Neste estudo foi observado que o G2, o qual não recebeu nenhum pré-tratamento, apresentou um aumento significativo das atividades da AST, ALT e BT quando comparado ao grupo G1 que recebeu somente solução salina, indicando que a dose de 500 mg/Kg foi capaz de induzir o dano hepático. O decréscimo da atividade da AST pelo G3, G4 e G5, quando comparado ao G2 mostrou que a silimarina (G3), assim como o EBUM (G4 e G5) possuem efeito hepatoprotetor frente ao APAP. Outros estudos corroboram com estes achados quando o dano hepático foi induzido por tetracloreto de carbono e os animais tratados com silimarina ou extrato de bagaço de uva (Papackova et al. 2018, Miao et al. 2023).

A atividade da BT demonstrou significativo decréscimo frente ao uso do bagaço de uva, quando comparados os grupos G3, G4 e G5 com o G2. Este resultado se mostra similar ao de Chupradit et al. (2022) que através da ingestão do resveratrol, polifenol encontrado no bagaço da uva, demonstrou melhora na atividade da BT e das enzimas hepáticas. Resultado de estudo de Andrade et al. (2021) também evidenciam que o teor de polifenóis e flavonóides contidos no EBUM possuem alta capacidade antioxidante e com isso possível efeito hepatoprotetor frente a indutores de dano ocasionados pelo estresse oxidativo.

A elevação significativa na concentração de TBARS nos camundongos que receberam overdose de APAP reflete a peroxidação lipídica, um marcador de dano celular. O pré-tratamento com extrato de bagaço de uva resultou em uma redução significativa nos níveis de TBARS nos grupos G4, G5, G6 e G7, indicando uma capacidade eficaz de reduzir a peroxidação lipídica associada ao estresse oxidativo. Esses resultados corroboram com as descobertas de Li et al. (2018), que, ao avaliar os efeitos do extrato de semente de uva em ratos submetidos a uma dieta rica em gordura, observaram uma supressão significativa nos níveis de TBARS, indicando um impacto positivo na redução da peroxidação lipídica hepática. A união desses achados reforça a eficácia do extrato de bagaço de uva na proteção contra danos oxidativos em diferentes contextos experimentais.

O estudo de Choleva et al. (2022) concentrou-se no potencial antioxidante do EBU, com ênfase em participantes submetidos a uma refeição padrão com alto teor de gordura. Notavelmente o EBU demonstrou uma redução significativa nos níveis de TBARS, indicando uma capacidade eficaz de diminuir a peroxidação lipídica associada ao estresse oxidativo. Essa diminuição persistente sugere um impacto antioxidante consistente do EBUM nesse grupo específico.

A GSH desempenha um papel fundamental na neutralização de espécies reativas de oxigênio e na detoxificação de compostos prejudiciais. Este achado é consistente com estudos anteriores, como o de Queiroz (2022), que destacou a importância da glutatona na detoxificação de substâncias tóxicas, incluindo o APAP. O aumento nos níveis de GSH sugere uma capacidade do EBUM, 100 mg/kg e 500 mg/kg em fortalecer a propriedade antioxidante endógena do fígado. O estudo de Ugan et al. (2023), corrobora com o presente estudo mostrando que o aumento de GSH é importante para prevenir danos hepáticos induzidos pelo APAP.

Papackova et al. (2018), em condições semelhantes a este estudo, verificaram uma diminuição acentuada nos níveis de GSH após a administração de APAP, assim como demonstrando nos grupos que receberam o pré-tratamento com silimarina ou EBUM (G3, G4 e G5) quando comparado ao grupo que recebeu somente o APAP (G2).

O pré-tratamento com EBUM foi capaz de reduzir o dano hepático oxidativo induzido pelo APAP, diminuindo a peroxidação lipídica sem, no entanto, aumentar os níveis hepáticos de GSH ou a atividade da CAT, como observado na Figura 3B e 3C. Estes achados sugerem que a presença de fenóis no extrato de bagaço de uva pode ter atuado como um hepatoprotetor uma vez que, a peroxidação lipídica foi reduzida. Esses resultados alinham-se com os achados de Chedea et al. (2019), que também observaram um aumento na atividade da CAT em suínos submetidos a uma dieta enriquecida com bagaço de uva, sugerindo uma resposta similar em diferentes modelos animais. Li et al. (2019) em seu estudo observaram que ratos alimentados com uma dieta rica em gordura apresentaram níveis diminuídos de CAT, entre outras enzimas antioxidantes. No entanto, a suplementação com extrato de semente de uva, especialmente na concentração mais elevada, resultou em uma recuperação significativa dos níveis de CAT, indicando uma resposta positiva do antioxidante frente ao estresse oxidativo provocado pela dieta rica em gordura. Esses achados complementam os resultados obtidos no estudo atual, reforçando a capacidade do EBUM, especialmente em concentrações específicas, de modular a atividade da CAT em situações de estresse oxidativo.

O EBUM quando administrado nas concentrações de 100 mg/Kg ou 500 mg/Kg (G6 e G7) mostram-se seguros visto não ter alterado os níveis séricos das enzimas hepáticas AST, ALT e BT, além de ter melhores desempenhos frente à parâmetros analisados de estresse oxidativo, evidenciado pela determinação da GSH (Figura 3B). A morfologia das células hepáticas também não foi alterada, conforme demonstrado nas Figuras 4F e 4G.

Os resultados relatados nos grupos G6 e G7 atestaram ainda a segurança e a eficácia do uso do EBUM, nas concentrações de 100 mg/kg e 500 mg/kg quando analisados os parâmetros de estresse oxidativo induzido pelo APAP. Este achado pode estar relacionado aos teores de polifenóis e flavonóides encontrados no EBUM. O teor desses compostos já foi retratado no estudo de Andrade et al. (2021) o qual afirmou sobre os efeitos benéficos do EBU Bordeaux, na redução do estresse oxidativo.

A análise histológica fornece uma perspectiva morfológica das alterações hepáticas, corroborando os achados dos marcadores séricos. O estudo de S. Torres et al., (2019) mostraram que a administração combinada de ácido valpróico (VPA) e APAP em camundongos resultou em lesão hepática marcante e necrose centrolobular, em acordo com o presente estudo, que evidenciou a predominância de necrose centrolobular no G2, o qual recebeu overdose de APAP, dado consistente com a toxicidade hepática induzida pela mesma substância.

O G5, que recebeu a dose de 500 mg/kg de APAP e pré-tratamento com 500 mg/kg de EBUM demonstrou predominância do escore 2, ou seja, com nível de necrose igual ou inferior à 25%, indicando que esta concentração foi hepatoprotetora, quando comparada ao G4 e G3. Essa relação dose-resposta destaca a eficácia do extrato em preservar a integridade do tecido hepático.

Os grupos G6 e G7, que receberam apenas o EBUM, não apresentaram desenvolvimento de necrose, indicando um perfil de segurança e ausência de toxicidade associada ao EBUM. Ugan et al. (2023) em estudo recente ratifica o uso de fisetina, um flavonóide encontrado em diversas frutas, em promover a proteção dos danos hepáticos induzidos por APAP.

V. CONCLUSÃO

Os compostos presentes no bagaço de uva mostraram resultados positivos quanto a atividade hepatoprotetora e antioxidante. Diante do que foi apresentado, evidenciou-se a atividade hepatoprotetora do extrato de bagaço de uva, nas concentrações de 100 mg/kg e 500 mg/kg, como demonstrado na redução significativa das enzimas hepáticas AST e ALT e da bilirrubina, redução do estresse oxidativo (TBARS e GSH) e da necrose hepática induzida pelo APA.

Conforme demonstrado, as atividades antioxidantes e hepatoprotetoras dos compostos presentes no EBUM poderiam gerar novas alternativas em fármacos que atuassem na proteção das células hepáticas, associados à medicamentos agressores ou na prevenção de doenças como na esteatose hepática, visto que uso de medicamentos naturais com propriedades antioxidantes vem sendo alvo de consumidores.

Embora o estudo tenha constatado resultados positivos frente à necrose hepática, algumas limitações podem ter contribuído para um desfecho mais assertivo. O uso de apenas um tipo de uva, assim como a dose de EBUM estudado; utilização de apenas um agente agressor (APAP); o número limitado de camundongos; análise de outras enzimas associadas à biotransformação, como do citocromo P450 e de marcadores inflamatórios poderiam trazer resultados mais evidentes.

Sugere-se que investigações futuras explorem diferentes doses do extrato, variedades de uvas ou métodos de administração para elucidar melhor os efeitos potenciais do bagaço de uva. Estudos voltados para alvos moleculares específicos podem proporcionar uma compreensão mais aprofundada dos mecanismos envolvidos na atividade hepatoprotetora e antioxidante desses compostos.

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Challenges of Access to Human Rights in the Amazon Context: Ecosystem Insights and Identifying Key Local Players and Initiatives for Legal and Policy Frameworks

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Abstract- This comprehensive study delves into the multifaceted human rights challenges in the Amazon region, focusing on ecosystem insights and identifying key local stakeholders. It explores the intricate interplay between environmental concerns and human rights, emphasizing the role of indigenous communities, NGOs, and government bodies. The paper proposes innovative legal and policy frameworks to address these issues effectively, advocating for integrated strategies that respect the unique socio-cultural and ecological fabric of the Amazon.

Keywords: amazon region, human rights, indigenous communities, environmental challenges, ecosystem analysis.

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Challenges of Access to Human Rights in the Amazon Context: Ecosystem Insights and Identifying Key Local Players and Initiatives for Legal and Policy Frameworks

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Abstract- This comprehensive study delves into the multifaceted human rights challenges in the Amazon region, focusing on ecosystem insights and identifying key local stakeholders. It explores the intricate interplay between environmental concerns and human rights, emphasizing the role of indigenous communities, NGOs, and government bodies. The paper proposes innovative legal and policy frameworks to address these issues effectively, advocating for integrated strategies that respect the unique socio-cultural and ecological fabric of the Amazon.

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INTRODUCTION

The Amazon rainforest, a bastion of unparalleled biodiversity and a crucial component of the global ecological system confronts an array of challenges that intricately intertwine human rights with environmental sustainability. This scholarly exploration seeks to dissect and understand the complex human rights landscape within the Amazon, where ecological concerns, indigenous heritage, and socio-political dynamics converge.

Delving into the roles and interactions of key local stakeholders – indigenous communities, non-governmental organizations, government entities, and private sectors – the paper endeavors to shed light on the multifaceted nature of human rights issues in this region. It critically examines the effectiveness of existing legal and policy frameworks, whilst proposing future strategies for sustainable development and conservation.

This comprehensive analysis aims not only to articulate the challenges but also to propose actionable solutions, encapsulating a holistic view that is essential for the protection and advancement of human rights in the Amazon. Through this discourse, the paper aspires to contribute to the evolving legal scholarship, offering nuanced perspectives on ensuring the preservation of the Amazon's ecological integrity and the dignity of its diverse inhabitants.

I. OVERVIEW OF THE AMAZON REGION AND ITS GLOBAL SIGNIFICANCE, CONTEXT, AND CURRENT CHALLENGES

The Amazon rainforest colloquially termed the "lungs of the Earth," represents a vast and biologically diverse expanse that stretches across multiple South American nations, predominantly within Brazilian territory. Beyond Brazil, the Amazon extends into Peru, where about 13% of the forest is found, making it the second-largest portion. Colombia holds the third-largest share, with about 10% of the rainforest within its borders.

Additionally, the Amazon rainforest also stretches into smaller parts of several other South American countries, including Venezuela, Ecuador, Bolivia, Guyana, Suriname, and French Guiana. Each of these nations holds a portion of the rainforest, contributing to the Amazon's immense biodiversity and ecological significance.

This ecologically vital region encompasses the Brazilian state of Amazonas, a substantial segment of the Amazon rainforest, integral to the wider Amazonian sphere. In Brazil, the "Brazilian Legal Amazon" (BLA) is designated as an administrative area, not limited to the state of Amazonas alone but including additional states and territories. These collectively constitute a region of paramount importance for its environmental, economic, and cultural attributes. The significance of the BLA transcends its physical confines, exerting a profound influence on global climate regulation, water cycle dynamics, and the preservation of biodiversity (Montibeller et al., 2020).

This region is not only a repository of unparalleled biological diversity, hosting a wide array of aquatic and terrestrial life forms, but also acts as a critical reserve of organic carbon. Its integration into the global water and energy cycles is indispensable (Gonçalves et al., 2013). The Amazon's intricate relationship with the global climate system is further highlighted by its dual function as both a sink for atmospheric carbon dioxide and a source of methane and nitrous oxide, both potent greenhouse gases (Davidson & Artaxo, 2004). The resources within this region are pivotal to Brazil's energy landscape and provide a model for balancing economic growth with sustainable natural resource management (Redclift, 1994).

Home to up to one-fifth of the world's freshwater reserves and a substantial portion of the remaining rainforests, the Amazon's ecological and hydrological significance is profound (Garcia, 2011). The Amazon basin, renowned for holding the world's largest hydric potential, plays a crucial role in global climate regulation. Its significant water cycles contribute immensely to the formation and stability of weather patterns and climate across the globe, emphasizing its ecological importance (Ishihara et al., 2014). Furthermore, the Amazon's extensive rainforests play a vital role in establishing global climate patterns, influencing the hydrological cycle, and contributing to the global carbon budget (Behling, 2012).

The Legal Amazon, spanning almost half of Brazil's territory, is divided into the Western Amazon, encompassing the states of Amazonas, Acre, Rondônia, and Roraima, and the Eastern Amazon, including Pará, Maranhão, Amapá, Tocantins, and Mato Grosso. Since the 1970s, the Brazilian Legal Amazon (BLA) has been the site of intense land use and rapid land cover change. This has occurred despite the forest's significant roles as a carbon sink and a biodiversity hotspot. Key drivers of deforestation in this region include cattle farming and soybean production, which have led to an increase in the region's carbon emissions and a reduction in its ability to provide ecosystem services. Deforestation has also resulted in forest fragmentation and edge effects, exacerbating environmental challenges (Montibeller et al., 2020).

Efforts to combat deforestation in the BLA have seen some success, with annual deforestation rates decreasing from 27.8 km² in 2004 to 4.6 km² in 2012. This reduction can be attributed to a combination of factors such as changes in commodity and land prices, as well as government policies. Notably, the Action Plan for the Prevention and Control of Deforestation in the Legal Amazon (PPCDAm) and the Soy Moratorium, launched in 2004 and 2006 respectively, played significant roles in this reduction. The effectiveness of these programs is evidenced by data from Brazil's national deforestation monitoring system (PRODES), which uses satellite imagery to monitor deforestation in primary forests in the BLA and provides annual reports on its findings (Montibeller et al., 2020).

In considering the broader context of sustainability, particularly concerning the Amazon, it is essential to move beyond focusing solely on the forest and the region itself. The conceptual framework of sustainability theory rests fundamentally on three pillars: ecological, social, and economic. Within the social dimension, it is imperative to comprehensively understand the Amazon in terms of its societal impacts and implications. This encompasses a thorough exploration of the human rights challenges prevalent in the Amazon, which includes the socio-economic conditions of the region's inhabitants, and the impact of

development policies on these communities. A holistic approach to sustainability in the Amazon thus necessitates a nuanced consideration of these social factors, alongside ecological and economic concerns, to ensure a balanced and just approach to conservation and development in the region.

In the Amazon region, a complex tapestry of human rights issues emerges, spanning from indigenous rights to environmental degradation, each intersecting with the other in a multifaceted manner. This overview seeks to elucidate these various dimensions, highlighting the challenges and potential solutions identified in recent scholarship.

Indigenous communities in the Amazon confront formidable challenges in asserting their rights, grappling with identity, cultural integrity, and external pressures and demands (Nagan, 2013). Their plight is further compounded by resistance to land demarcation, leading to internationally condemnable human rights violations (Valenta, 2003). These issues underscore the crucial need for robust legal frameworks and policies that prioritize indigenous rights, religious freedom rights, and heritage preservation.

The Amazon also contends with the illegal occupation of public lands and irrational resource usage, exacerbated by a lack of governmental oversight and inconsistent public policies (Velasquez, Bôas, & Schwartzman, 2006). This situation is further aggravated by property rights issues and rampant deforestation, especially in smaller properties, calling for enhanced enforcement and inspection mechanisms (Sasaki Okida et al., 2021).

A backdrop of inequality and violence characterizes the region, with the impoverished facing high levels of violence, health crises, and limited access to healthcare and social rights (da Silva, 2017). These social inequities are often intensified by development projects that, while intended to boost regional economies, frequently lead to human rights infringements (Linton, 2010). Moreover, the region's healthcare challenges, particularly in rural areas, impede the global goals within proposed timeframes (Silva, 2006).

Environmental concerns, such as the impact of small hydropower plants, threaten ecosystem services, biodiversity, and the livelihoods of indigenous and traditional communities (Athayde et al., 2019). This situation necessitates initiatives that mitigate environmental degradation and promote environmentally sensitive economic activities (Castro et al., 2019). Agroforestry practices in the Amazon exemplify a blend of productivity and sustainability, thereby enhancing the livelihoods of smallholders and indigenous communities (Porro et al., 2012).

Institutional strength and innovation are pivotal in addressing these challenges. Advancements in science, technology, and regionalization play a

significant role in safeguarding human dignity in the Amazon (Becker, 2016). Social organization, awareness, training, and agricultural diversification are effective in supporting the dignity of Amazon residents (Szlafrstein, 2014). Additionally, programs like the Indigenous Amazonian REDD (RIA) and the National Forest Conservation Program (PNCB) in Peru focus on strengthening property rights for native peoples, essential for promoting human dignity (Dupuits & Cronkleton, 2020).

Smallholder management systems that maintain environmental stability and contribute to local well-being are instrumental in supporting human dignity (Pokorny et al., 2013). Community empowerment for conservation, employing an assets-based approach, aids in sustainable natural resource management by local communities (Wali et al., 2017). Furthermore, transnational social movements play a critical role in protecting the Amazon rainforest, focusing on environmental, cultural, and human rights issues (Schittecatte, 1999). Place-based sustainability initiatives enhance living standards and environmental sustainability in the Brazilian Amazon through individual and collective action (Brondízio et al., 2021). Lastly, the implementation of effective public policies in health and environment can improve the health of Amazonian populations and promote environmental protection (Silva, 2006).

The complex interplay of these issues and initiatives in the Amazon region illustrates the multifaceted nature of human rights concerns and the necessity of integrated, comprehensive strategies to address them effectively.

This section critically examines the global significance and ecological diversity of the Amazon region, along with its current environmental and socio-political challenges. We shall now progress to a detailed exploration of Amazon's ecosystem, focusing on how its complex dynamics are interwoven with pertinent human rights issues and the broader socio-political fabric of the region.

II. ECOSYSTEM INSIGHTS

In the Amazon region, a non-biological ecosystem comprised of organizations, players, and stakeholders forms a complex and dynamic network. This network is characterized by interdependent elements, where the actions of one entity significantly impact others. These diverse entities coexist, collaborate, and compete, shaping the dynamics and outcomes in areas such as business, technology, social sectors, and public policy. This ecosystem metaphor aptly describes the intricate interplay between various actors in a shared environment.

Within this ecosystem, the Amazon exerts a notable influence on education, with Amazon's business

models and technologies increasingly permeating educational institutions and shaping their interaction with digital platforms (Williamson et al., 2022). Concurrently, the Amazon faces urban infrastructure deficiencies, leading to limited improvements in urban conditions and quality of life, thereby impacting social dynamics and development opportunities (Costa & Brondízio, 2009).

In the Peruvian Amazon, interlinked networks and institutions strengthen conservationist groups and indigenous peoples' organizations, countering the social forces of commodification and supporting indigenous rights (Orihuela, 2020). The region also exhibits varying relationships between connectivity and resilience, affecting the impact of infrastructure on local communities (Perz et al., 2012). This complexity is further evidenced in community connectivity, where greater connectivity corresponds to greater resilience in some aspects but not others, highlighting the nuanced nature of social and ecological interactions in the southwestern Amazon (Perz et al., 2012).

Furthermore, entrepreneurship and social change are facilitated through direct sales networks in remote areas, influencing local-global value reconciliation on beauty and fashion, and leading to stronger brand relationships and social network dynamics (Chelekis & Mudambi, 2010). In environmental governance, social network analysis reveals potential for communication and organization among actors, which is crucial for addressing environmental and social issues (Mertens et al., 2011).

In the realm of food security, strong-tie social networks contribute to the availability, access, utilization, and stability of fish resources, demonstrating the interplay between social relationships and resource management (Mertens et al., 2015). Participatory stakeholder workshops in infrastructure projects enhance community engagement and problem-solving, particularly related to road paving in the southwestern Amazon (Mendoza et al., 2007).

Moreover, the discussion network in the Brazilian Amazon is characterized by both resilience and vulnerability, necessitating actions to stimulate opinion leaders and increase redundancy in discussion channels for effective communication and organization (Mertens et al., 2008).

In the Amazon region of Northern Brazil, access to justice is significantly impeded by a confluence of environmental, socio-political, and infrastructural challenges. Environmental degradation, particularly in the Brazilian Amazon, coupled with the emergence of vector-borne diseases, directly impacts local populations, hindering their access to justice. These environmental challenges have broader implications, affecting adjacent areas and complicating the pursuit of justice (Castro et al., 2019).

Land conflicts between small-farmer migrants and large estate owners further exacerbate access to justice. Government interventions in these disputes often have political and ideological underpinnings, adding complexity to the legal landscape and impeding justice for affected communities (Schmink, 1982). Additionally, riverine communities in Northern Brazil, such as those in the Bailique Archipelago in Amapá State, face distinct challenges in accessing labor justice due to factors like limited access to capital, poor state services, and the absence of specialized courts (Furlan & Pires, 2017).

Moreover, issues related to late birth registration and unregistered births in remote, rural, and indigenous areas of Northern Brazil, including the Amazon region, present multifaceted challenges. Children without official birth certificates often lack access to essential social services, such as schooling and healthcare, especially in areas with high under-registration of births (Duryea, Olgati, & Stone, 2006). Similar to Northeast Brazil, the Amazon region likely faces significant quality issues in birth registration, with considerable regional variations (Paes & Dos Santos, 2010).

The impact of unregistered births and lack of personal identification is particularly pronounced among Indigenous people. This scenario, evident in northern Ontario, Canada, parallels the Amazon region, where remote indigenous communities encounter similar obstacles (Sanders & Burnett, 2019). Healthcare-related challenges, such as those associated with preterm birth in the Western Brazilian Amazon, underscore the importance of healthcare access and quality in these remote areas (Ramos de Oliveira et al., 2021). Traditional birth methods among Indigenous women in the Amazon also indicate a gap in integrating traditional and modern healthcare practices (de Lima, Santana, & Ferreira, 2020).

Effective law enforcement, crucial for ensuring proper birth registration and access to rights, remains a challenge in Brazil's remote regions, including the Amazon (Schneider, de Marques, & Peres, 2021). Access to healthcare in these extremely remote areas is critical for ensuring that births are registered and that mothers and children receive the necessary care (Tobias, Schweickardt, & Harris, 2020). The COVID-19 crisis has exacerbated these challenges for Brazil's indigenous groups, especially in remote areas like the Amazon, due to reduced budgets for inspection bodies and inadequate public health systems (Emerald Expert Briefings, 2020).

These points underscore the intricate relationship between social networks and infrastructure in the Amazon, where connectivity, resilience, conservation, entrepreneurship, and stakeholder engagement are crucial in shaping local problems and opportunities, which include multifaceted impacts on education, access to justice, social development, and the broader context of sustainability.

After this analysis of Amazon's diverse ecosystem and its environmental challenges, the discourse shifts to identifying and scrutinizing the roles of key stakeholders. The forthcoming section delves into the impacts and contributions of indigenous leaders, non-governmental organizations, and government entities in shaping the region's future and addressing its multifaceted challenges.

III. KEY LOCAL PLAYERS

In the Amazon region, particularly in Northern Brazil, research underscores the pivotal roles of indigenous leaders, NGOs, and government entities in managing and protecting this ecologically critical area. The complex interplay of these stakeholders forms the cornerstone of environmental and social governance in the Amazon.

The role of indigenous leaders and communities is crucial in preserving the ecological integrity of the Amazon. Studies such as those by Schwartzman et al. (2013) demonstrate how NGOs and government support have enabled indigenous and traditional communities to effectively organize and administer their institutions in areas like the Xingu Indigenous Lands and Protected Areas Corridor (ILPAs). These initiatives focus on scalable resource management and income generation, highlighting the collaboration between different stakeholders. These efforts contribute significantly to reducing deforestation rates and preserving ecological intactness in the region.

Dall'Orso (2020) discusses the importance of policies for the protection of Indigenous Peoples in Voluntary Isolation and Initial Contact (IPVILC) in Amazonian countries. The review highlights the leading roles played by national Amazonian indigenous people's organizations and Amazonian governments, emphasizing their implications for territorial dynamics and indigenous peoples' development in Amazonia.

The work of Schwartzman and Zimmerman (2005) explores the contribution of indigenous understandings of their resource base to long-term conservation and sustainability in Amazonian Indigenous Reserves. The study emphasizes the vital role of indigenous leaders in environmental management and conservation efforts in the Amazon.

The intricate network of stakeholders in the Amazon region, particularly in Northern Brazil, exemplifies a critical interplay among indigenous communities, non-governmental organizations (NGOs), and government entities. This interplay is pivotal in addressing the multifaceted environmental, social, and economic challenges faced in the region. The initiatives undertaken by these diverse groups are fundamental to the long-term sustainability and conservation of the Amazon, necessitating a comprehensive and integrated approach.

Within this framework, the Tribunal de Justiça de Roraima (TJRR) and the Escola Judicial de Roraima (EJURR) have been instrumental in enhancing access to justice, particularly for indigenous communities and remote populations. The Vara da Justiça Itinerante, established in 1997, plays a crucial role in promoting citizenship among indigenous peoples and those in remote regions of the state. This initiative acknowledges the unique cultural and legal specificities of the original communities of the region. By bringing justice directly to indigenous communities and overcoming geographical barriers, the Justiça Itinerante has evolved to address not only judicial issues but also challenges such as indigenous under-registration.

Another noteworthy initiative is the Pólo Indígena de Conciliação e Mediação "Elias Souza" in the Maturuca community, established in 2015. This project, developed in collaboration with regional leaders, focuses on alternative conflict resolution methods in traditional communities, emphasizing restorative justice practices. It aims to empower community members with mediation and conciliation skills, strengthening community relations and reducing the need for external judicial intervention.

The Programa Justiça Cidadã, pioneered by the Poder Judiciário de Roraima, marks a transformative step in the Brazilian justice system. It aims to provide widespread access to judicial services, particularly in areas without local court jurisdictions. This program has established physical representations of the judiciary in all municipalities of Roraima, making it the first in the country to achieve such coverage. The initiative has been recognized nationally by the Conselho Nacional de Justiça (CNJ) as a successful model in improving access to justice.

Meanwhile, the Tribunal de Justiça do Amazonas and the Escola da Magistratura do Amazonas have made significant strides in legal research and education. The Escola Superior da Magistratura do Amazonas (ESMAM) recently achieved accreditation as a research institution by the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPQ). This accreditation enables ESMAM to conduct extensive legal research, with a focus on human rights issues in the Amazon context and criminal justice processes in the legal Amazon region. These research initiatives aim to explore the unique challenges and dynamics of human rights and legal processes in the Amazon, contributing significantly to the broader discourse in Brazilian legal scholarship.

These initiatives collectively demonstrate a proactive approach to legal and judicial innovation in the Amazon region, focusing on the unique challenges faced by indigenous and remote communities. They serve as models for similar initiatives in other regions of Brazil, highlighting the importance of adapting legal

services to the specific needs and contexts of traditionally marginalized populations.

In this context, the role of private organizations is increasingly pivotal in addressing environmental, social, and economic challenges. These challenges necessitate a comprehensive approach that intertwines respect for indigenous rights, and other laws, with the strategic involvement of various stakeholders.

The entry of private organizations like Starlink, a satellite internet company led by Elon Musk, exemplifies this dynamic. Starlink's rapid expansion in the Amazon has brought high-speed internet access to remote communities, enabling significant advancements in communication, financial transactions, and educational opportunities. The company's services have reached about 90% of municipalities in the Amazon region as of mid-2023, primarily serving areas with limited traditional internet infrastructure. While this expansion has been instrumental in bridging the digital divide, it has also raised concerns about its unintended consequences, such as potential facilitation of illegal activities and implications for national security and sovereignty (BBC, 2023).

Moreover, the Starlink initiative was announced with the intent to provide internet connectivity to 19,000 unconnected schools in rural areas and assist in environmental monitoring of the Amazon. This endeavor reflects the growing recognition of the role of private organizations in supporting vital infrastructure and services in remote regions (BBC, 2023).

Such international involvement is further underlined by the presence of different NGOs in the region managing different kinds of projects, on such example is the "Science Panel for the Amazon" (SPA), a high-level scientific initiative sponsored by the United Nations Sustainable Development Solutions Network (UNSDSN).

Launched by a group of 150 scientists, the SPA aims to deliver the first scientific assessment of the state of the Amazon Basin. This assessment is expected to offer a blueprint for policymaking, addressing the urgent environmental threats to the Amazon. The SPA underscores the value of integrating scientific research, indigenous knowledge, and public-private partnerships in formulating policies for the sustainable development of the Amazon.

These examples underscore the critical role of private organizations in the Amazon, demonstrating their potential to complement the efforts of public entities and indigenous communities. However, their involvement must be balanced with due consideration for environmental preservation, social equity, and respect for human rights. The challenge lies in harnessing private sector capabilities and resources in a manner that aligns with the broader goals of sustainable development and conservation in the Amazon region.

Highlighting the crucial roles of indigenous communities, NGOs, and governmental organizations in the Amazon, this section forms the foundation for our concluding arguments. It underscores the necessity for a collaborative approach, integrating the insights gathered to advocate for concerted and holistic strategies in safeguarding the Amazon and its inhabitants.

IV. CONCLUSION

The paper culminates with a comprehensive synthesis of the Amazon region's overview, its ecosystem, and the pivotal roles of key local players. In conclusion, the paper underscores the imperative for multifaceted strategies and policies that effectively confront the Amazon's complex challenges, ensuring its preservation and the welfare of its diverse communities.

In the context of the Amazon region, the formulation of future strategies and policies must prioritize a collaborative approach among indigenous communities, NGOs, government bodies, diplomatic missions, and private entities. This collaboration should leverage technology to bridge infrastructural gaps, while being mindful not to exacerbate existing environmental or social challenges. Policies must promote the responsible use of technology, respecting cultural and ecological sensitivities.

Developing comprehensive policies that address indigenous rights and environmental conservation is essential. These policies should be formulated in consultation with indigenous communities to ensure the preservation of their cultural heritage and the respectful integration of their traditional knowledge in resource management. Legal recognition of indigenous lands and resources is critical, alongside the promotion of sustainable development practices.

The involvement of the private sector in the Amazon, particularly in resource and infrastructure development, must be regulated to safeguard the environment and local communities. Policies should encourage corporate responsibility and sustainable practices, offering incentives to companies that positively contribute to the region's ecological and social fabric.

A more active role for the international community is imperative in supporting conservation efforts in the Amazon. This could involve funding research, supporting local NGOs, and endorsing international agreements aimed at preserving the Amazon's biodiversity and the rights of its indigenous peoples.

Legal reforms are necessary to enhance access to justice in remote and indigenous areas. Initiatives like the Justiça Itinerante and the Pólo Indígena de Conciliação e Mediação "Elias Souza" underscore the importance of establishing itinerant courts and legal

services. The adoption of technology in legal processes can improve accessibility for remote communities.

Future policies should also encourage sustainable economic development in the Amazon. This includes supporting local industries that are environmentally friendly and providing indigenous communities with economic alternatives that align with their traditional practices and lifestyles. Ongoing research and environmental monitoring are vital to understanding Amazon's dynamics and in formulating evidence-based policies for its sustainable management.

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Organizations in Social Media Environments: Strategy, Communication, and Mediatization

By Fabio Frá Fernandes

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Abstract- Our text explores the mediatization phenomenon in smaller organizations in geographically remote towns. Through a theoretical-practical study of an applied nature and with an exploratory-descriptive objective, we aim to highlight how the current media ecosystem is used and appropriated while also describing the communicative strategies employed by a group of thirteen organizations situated in the northwest region of the Brazilian state of Rio Grande do Sul. For our methodology, we utilized participant observation, employing techniques to analyze the social media presence of each organization over thirty days. Additionally, we conducted a strategic communication audit targeting specific audiences and indepth interviews with professionals responsible for their communication. The outcome of our efforts is a comprehensive mapping that delves into and describes the mediatization phenomenon within these organizations, which, in our perspective, is in the process of establishment.

Keywords: *mediatization. media and communication strategies. media ecology. public relations. organizations.*

GJHSS-H Classification: *FOR Code: 2001*



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1. INTRODUCTION

Historically, transformations in media environments¹ have redefined the forms and processes of human sociability, impacting both individuals and organizations. This redefinition involves the interplay between changes in culture, society, and organizations on the one hand and transformations in media, communications, and socio-organizational relationships on the other. In a context where the interdependence of individuals with the media is becoming increasingly profound, the mediatization of their social, cultural, and political practices and processes becomes more evident.

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¹ In our current research, we regard social media environments as platforms such as Facebook, Instagram, LinkedIn, Twitter, Snapchat, and TikTok; streaming platforms like Spotify, YouTube, and others; mobile applications exclusively developed for organizational use; instant messengers such as WhatsApp; in addition to their Institutional Portals.

With mediatization, socio-organizational potentials and vulnerabilities emerge across any territory, prompting professionals and researchers to understand their configuration, causes, and effects in all aspects of society. Furthermore, concerning communication practices and processes, media environments, with digitalization, enable the reversal of roles between emission and reception. It also allows different individuals to be consumers and producers of content simultaneously. The transformation in communication paradigms affects large media conglomerates and individuals and organizations that now need to manage their audiences' movements in media environments strategically.

From this context, it is essential to note that organizations in various sectors located in major urban centers or near industries such as technology, innovation, media, entertainment, or foreign trade, initially have well-established communication strategies. Subsequently, these same organizations have qualified professionals with theoretical-practical expertise in media communication, especially in the digital realm. However, in cities far from major population centers, smaller organizations primarily focusing on the local market often find themselves relatively distant from strategic media appropriation, as commonly reported in specialized articles, university studies, and success stories.

Our focus in this presentation is to discuss the mediatization movement in smaller organizations in rural cities. Through a theoretical-practical study of an applied nature and with an exploratory-descriptive objective, our narrative attempts to highlight how a group of thirteen organizations representing the microregions shaping the northwest region of the Brazilian state of Rio Grande do Sul configures the uses and appropriations of the current media ecosystem.

To accomplish this, our narrative about the research development unfolds throughout the text, presented in four sections. The first section is dedicated to methodological characterization, the second to theoretical-epistemological dialogue, and the third is employed to scrutinize the reality revealed by our empirical object while allowing the creation of two maps illustrating the mediatization phenomenon in the organizations of the concerned territory. Finally, in the fourth and last section, we conclude our account by highlighting other possible studies to be generated from our research.

II. METHODOLOGICAL APPROACH AND RESEARCH DESIGN

For methodological, cultural, and professional reasons rooted in extensive academic and professional experience in municipalities located in the state's interior, we defined this research's empirical object as small and medium-sized organizations. These organizations are situated in one of the thirteen microregions forming the Northwest region² of the state of Rio Grande do Sul, Brazil. These microregions include cities such as Carazinho, Cerro Largo, Cruz Alta, Erechim, Frederico Westphalen, Ijuí, Não-Me-Toque, Passo Fundo, Sananduva, Santa Rosa, Santo Ângelo, Soledade, and Três Passos. We selected one organization from each municipality based on the criteria for the sample, requiring them to use at least two social media platforms in their communication and relationship strategies. Additionally, they needed a communication professional or someone with marketing expertise within their functional structure, whether institutionally affiliated or outsourced. Organizations meeting these criteria were contacted via email, and upon confirming their participation, their representatives recorded their acceptance in a Free and Informed Consent Form.

The economic sectors of the researched organizations primarily encompass retail e-commerce; sale and distribution of motor vehicles; industry, production, and commercialization of implements for agribusiness; production and commercialization of agricultural machinery; cereal production with biodiesel manufacturing; affiliate of a television station; credit cooperative; furniture and decoration trade; professional development agency; professional education school; community-university; and web solutions development agency³.

Considering the context and the anticipations in the initial considerations, this research engages in a dialogue and scrutiny of how mediatization manifests in medium and small-sized organizations outside major urban centers. This exploration seeks to understand their institutional positioning amid individuals' deepening interrelation and interdependence with the media. Additionally, we describe the media uses the apps, operations, and communication strategies while highlighting the processes and practices adopted by these organizations.

We constructed the mediatization phenomenon to achieve this, drawing on media ecology and organizational communication studies. This approach allowed us to present the communicational reality of our empirical object. Through a theoretical-epistemological dialogue, we reflected on the interrelation and interdependence of media within organizations and their audiences. Subsequently, we read the situational and cultural reality of the field (Santos, 2015). To operationalize this reflection, we engaged the thirteen organizations through three data collection approaches: participant observation (Peruzzo, 2012), strategic communication audit with specific audiences (Kunsch, 2012), and in-depth individual interviews (Gaskell, 2017). Over thirty days, we closely monitored the digital media of each of the thirteen organizations. This process involved qualitatively analyzing their communicational and strategic construction within the current media ecosystem. We considered [A] informational flows, [B] the relevance of information, [C] levels of interaction with user audiences, [D] aesthetic and graphic patterns, and [E] processes of media convergence.

Simultaneously, we audited the communication strategies identified in the observation based on the perceptions of internal and external audiences. The audits were conducted through structured questionnaires sent via email to groups of people with and without institutional affiliation, using mailing lists provided by each organization and randomized by specific software. Two hundred sixty questionnaires were sent, organized into subgroups of 10 employees and ten clients for each organization. Of these, 189 were completed and returned, 23 were incomplete, and 48 needed to respond. Moreover, 137 audits correspond to employees' and 52 to clients' perceptions.

Following the initial approaches, we conducted in-depth interviews with communication professionals designated by each organization. Seven of these professionals are outsourced, with functional ties to communication agencies. The remaining interviewed professionals are employees with institutional affiliations. It is worth noting that among the thirteen professionals interviewed, five are journalists, four are advertisers, three are public relations specialists, and one is an administrator with expertise in digital marketing. Only three interviewed individuals with institutional ties are Communication managers; the other four collaborate with other areas in their organizations.

For each interview, we conducted meetings via Google Meet, lasting an average of one to two hours, structured by a semi-structured script. In these meetings, we juxtaposed the institutional discourse of these professionals with the information obtained and developed through the previous approaches.

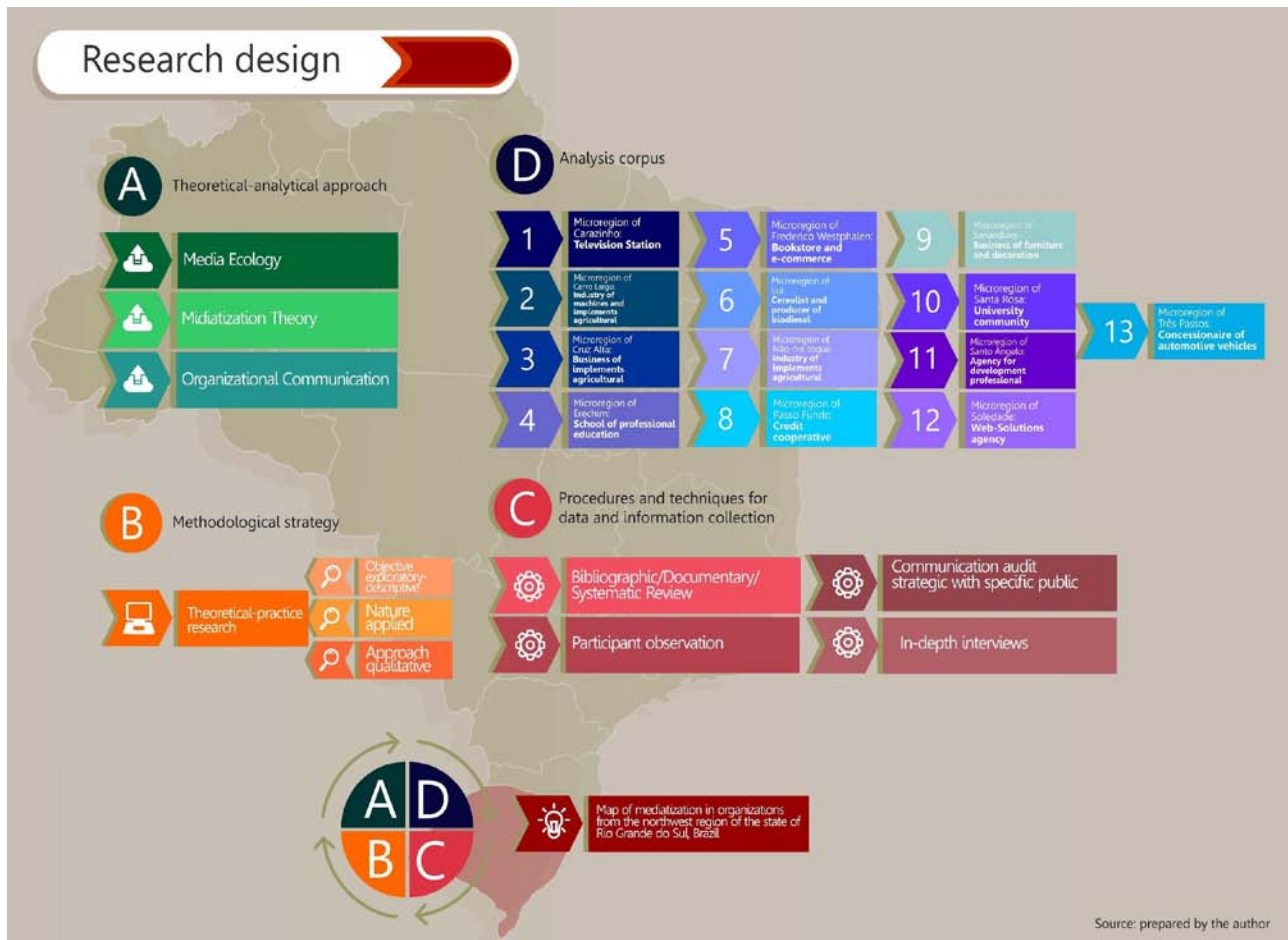
Following this, we present the overall plan applied to develop the research, emphasizing the analytical approaches we employed on the collected

² The northwest region constitutes one of the seven mesoregions in the Brazilian state of Rio Grande do Sul. It consists of a union of 216 municipalities, grouped and organized into thirteen microregions, with the municipalities above serving as their headquarters.

³ We have omitted the identification of the organizations comprising the research corpus to respect the privacy of their representatives and align with ethical norms for conducting research in the Social and Applied Sciences field.

data and information, the procedures and techniques used to obtain such data and information, our empirical

object, and finally, the achievement of our exploratory-descriptive objective.



III. THE MEDIATIZED ORGANIZATIONAL CONTEXT

With the growing mediation of social practices, communication processes are also affected, especially concerning organizations, as radio, television, press, and other media converge into the digital realm and are reconfigured to generate interactivity. This perspective also applies to other technologies that operate as suitable media environments for mediating different socio-organizational interactions, even if not initially designed for this purpose. Examples include social media platforms, messaging apps, platforms for virtual games, and various other technological resources available in the current media ecosystem.

In the contemporary era, we have moved beyond the sender/receiver and medium/message dichotomy. With the phenomenon of mediation, we witness the inversion of the sender and receiver poles. In other words, in traditional broadcast communications, these poles had well-defined positions, but with the evolution of the media ecosystem, we observe a particular shift in their positions. Now, anyone can become both a sender and receiver of information,

reflecting the democratization of technologies and their extensive reach through the intertwining of physical devices (hardware) and virtual environments (software). This context allows individuals with internet access to consume content and produce it, creating broad and multicultural networks of relationships. Media communication in today's society is no longer confined to media institutions; it is accessible to anyone proficient in digital technologies.

As a result, developing a networked society and consolidating digital social media favors the deepening of mediation in social and organizational practices. In this context, social media emerges as a strategic resource, becoming allies for organizations by facilitating real-time multi-level interaction. This situation can potentially engage organizations with their target audiences while fostering the circulation and feedback of collective knowledge and intelligence.

Social media platforms constitute spaces for the virtualization of sociability. Initially designed for network interconnection free from territorial barriers, these platforms have evolved into strategic resources for organizations to gain visibility and public legitimacy through qualified relationships with their audiences.

According to Recuero, Bastos, and Zagors (2018), this configuration of social media consists of two central elements: social actors (individuals, institutions, groups) and connections (interactions and social ties). These platforms, therefore, present themselves as intricate networks of mathematically configured nodes, creating dense media environments whose underlying logic and operations often go unnoticed in everyday life. Media permeates all processes and practices of human sociability, living in what Sodr  (2012) calls the technointeraction and the bios mediaticus.

The bios mediaticus is the territory of media, where individuals accessing this environment are never the same when they leave, having transformed. This process extends to society and its organizations, as individuals who experience changes within the mediatic bios subsequently transform others in their social and organizational relationships.

The most significant change brought about by social media, the primary resource of the mediatic bios, is the ability to do almost anything within its environments. Beyond enabling individuals to be consumers, producers, and transmitters of information, social media platforms create spaces where physical reality is ubiquitous with mediatic reality. This characteristic favors the development of omnichannel communication within organizations - a communication strategy integrated, simultaneous, and symmetrical across all media environments an organization uses to enhance its relationships, with mediation as a potentializing meta-process (Fernandes & Silva, 2020).

In the expansive networked society, understanding communication within the scope of mediation has become a constant challenge for organizations, particularly in the last decade. They have had to envision new strategies or renew existing ones to engage with their audiences in the various organisms that compose the current media ecosystem. Today, organizations need to synchronize and make every communication and relational strategy inseparable, not only converging and adapting broadcast communication processes with directed communication practices but also crafting discursive narratives tailored to each media environment. In communication, there remains a need to align and adapt organizational culture to messages, language, resources, time, visual, and graphic typology to ensure that mediatic processes focused on socio-organizational interaction occur effectively and reach their target audiences.

Observing the historical trajectory of communication within the context of mediation, especially in recent years, we perceive the emergence of more participatory social actors. These individuals with social media technologies hold influences that ultimately redefine organizational culture. Thus, mediatic environments contribute to these actors increased social and organizational participation, enabling dialogical and

proximity relationships between internal and external actors associated with organizations (Fernandes & Silva, 2020a).

Therefore, communication in the mediated organizational context involves the interaction between what the organization communicates and the communication of its audiences. Being present in these environments does not make an organization part of the mediatic bios. Still, by developing a robust action plan to be present in these environments, considering internal and external organizational realms, and appropriating the logic of media, the organization becomes a mediated socio-organizational environment.

IV. MEDIA AND COMMUNICATION STRATEGIES AMID THE MEDIATIZATION OF ORGANIZATIONS

Despite the redefinition of interactional paradigms stemming from the mediated social experience, the emergence of social media, and the knowledge they provide to their users, communication within organizations, especially those considered in this research, remains instrumental rather than embracing the strategic potential offered by the current media ecosystem. When we scrutinize the technointeraction in the mediatic bios, the communication and relational strategies of these organizations, the processes and practices fall short of the recursive possibilities of mediation. Particularly in the digital realm, media environments still need to be fully integrated into the culture of many organizations despite being present and highly palpable in the daily lives of their organizational and social actors.

We understand that mediated communication alters habits and shapes culture and social dynamics. These changes also impact organizations, presenting a significant challenge in understanding the phenomenon of mediation, especially concerning the formation of virtual communities (social networks), which are mediated by technical devices with redefined logic and affordances⁴, have already become integral to contemporary society.

People and organizations can and should use social media in a plural, accessible manner, expanding various categories of interaction, reflection, participation, or communication in general. Individuals now engage in activities once limited to professionals, reshaping communication's linear informational pathways. The opening up of media, even informally, affects and alters

⁴ According to Gibson (2014) and Hjarvard (2014), in this research, we interpret affordances as the standard and potential uses that a given object possesses. When applied to understanding the field of media and communications, our concept of affordances of technological artifacts allows various actions to be performed, organized, or even excluded based on their appropriation in mediated interaction processes.

organizational practices or the social construction of reality.

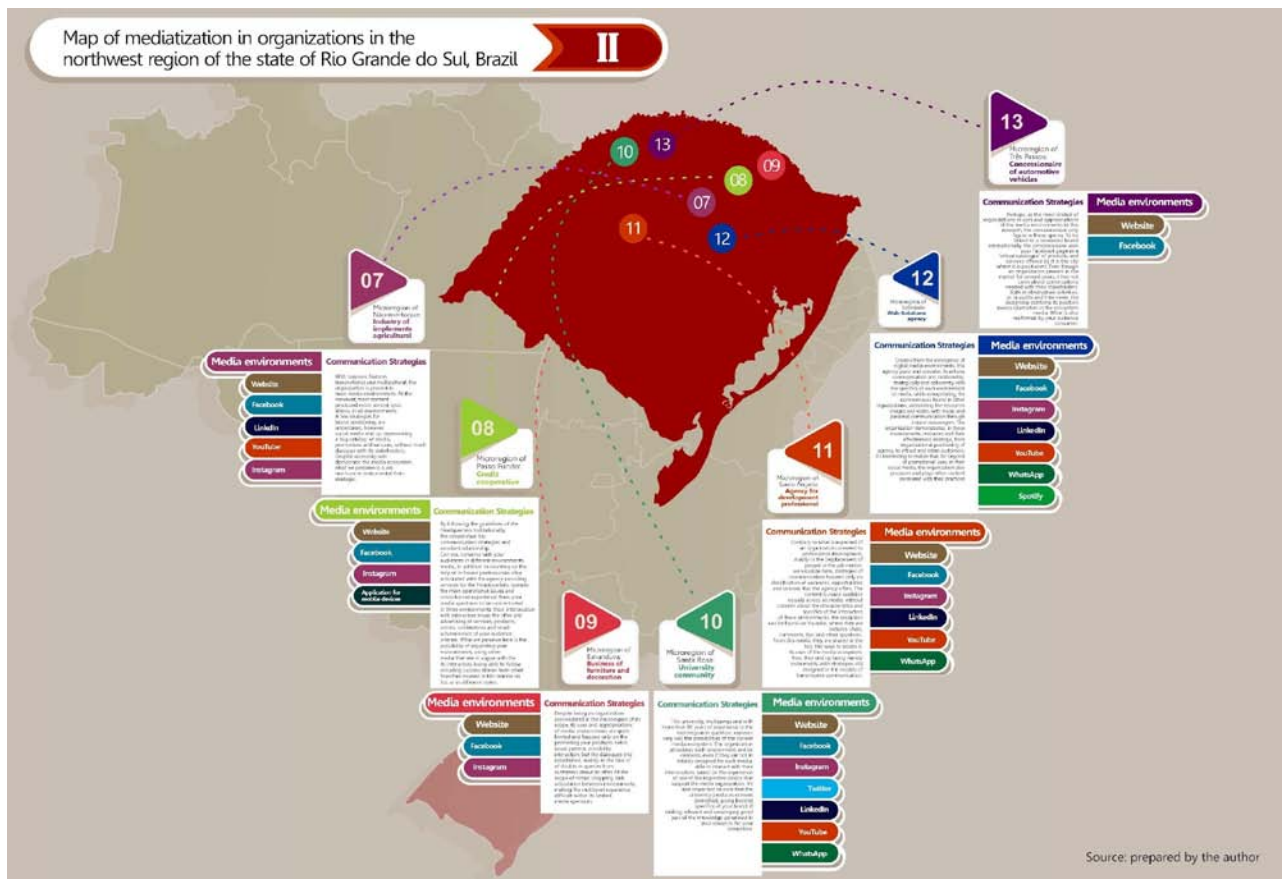
With the shift in cultural communication paradigms—where media operate as socio-organizational extensions—users realize the symbolic power they hold. The transition from mere spectators to opinion formers or, as the current hype defines it, digital influencers (Terra, 2021) for brands and organizations. From this characteristic, problems or opportunities arise. The outcome depends on the culture adopted and implemented within their framework and the conversion strategies between what the organization communicates and what is communicated about the organization.

Examining the data and information collected from the study, we observe that the communicative

reality of these organizations is still predominantly instrumental. Despite the resources available to mediate culture and society, these organizations are still in their early stages. Thus, it can be asserted that mediation in the researched organizations is still in the establishment process, quite different from other regions and countries where mediation has already deepened or become entrenched (Couldry & Hepp, 2020).

Continuing our narrative, we present two maps that succinctly depict the context of mediation in organizations located in the northwest region of Rio Grande do Sul. In these maps, we explore and describe the mediatic configurations and communication strategies.





As we can see from the maps, even though the researched organizations navigate through the primary media environments, most still need to learn about the resources these technologies offer to make their communication and relationship strategies more aligned with the interests of their audiences. This situation worsens the further their core activities are from communication or technology. In this context, there is a behavioral pattern or a particular reproduction of marketable content and social interaction formats. Due to the lack of appropriation of these environments, these organizations produce meanings that must align with their communicational reality and, more importantly, their audiences' reality.

They merely replicate strategies that are more common to broadcast transmission communication in media environments, where communicational culture is still centered on past habits, beliefs, and views, overlooking mediation's multifaceted and influential connections. Even in transmission media environments like radio and television, strategies need to be adapted according to the logic of each medium. With mobility and hyperconnectivity, the need to transverse communicational processes and practices, converging content and organizational objectives and policies for each new environment the organization accesses, becomes even more urgent.

By not embracing these characteristics, organizational actors, increasingly interdependent on

media, have limited access to the new meanings that organizational mediation can offer. They seek the strategic relationships they desire and imagine in other brands in these environments.

The organizational discourse presented by most researched organizations needs to be more consistent and different from their practice. Their representatives claim to understand and be familiar with the current spectrum of the media ecosystem. They say they research, identify, and monitor their target audiences and other users. However, in practice, this only happens in part. There are common characteristics present throughout the sample, both positive and negative. The organizational and relational presence on social media, for the most part, functions as a mere extension of broadcast communication strategies. However, even with a technical and instrumental discourse, organizations understand and perceive that there is still much to be explored in the media field. According to their managers, the time and resources for qualification need to be improved regarding the constant and accelerated technological development and mediation methods to mediate organizations.

It is important to note that courses and events that address this theme or similar topics are usually held outside the regional axis, which also complicates and increases the cost of access. Thus, there is the infrastructure for the operation and appropriation of the media ecosystem, and organizations and their

professionals know the potential for its adoption. However, the use based on similarity (from the experience of established brands) determines the mediatized organizational communication of these organizations.

Considering this, we must remember that historically, media was intuitively integrated into society's daily life. In other words, we learned to use each resource based on daily experience or the already consolidated experience of others. However, in the context of social media, even though the intuitive method is a constant in the daily lives of many, the accelerated technological development and media education that younger generations are perfecting require organizations and their professionals to have a more agile and specialized appropriation of the current media ecosystem.

V. SOME CONCLUSIONS

This text explores and describes the media configurations and communication strategies of small and medium-sized organizations from various sectors, geographically located in regions distant from major urban centers, through the lenses of mediatization, illuminated by media ecology and organizational communication.

Through applied research, we observed, for thirty days, the media environments accessed by thirteen organizations, attempting to highlight their positioning in the interrelation and interdependence of media organisms in the current socio-organizational context. This context, by extension, aligns with the understanding of the conversion of communication strategies in the face of the mediatization of culture and society.

We encountered organizations with a mediatized organizational profile, equipped with technical resources and efficient knowledge for an ideal⁵ appropriation of media environments. However, a significant portion of the sample needs to explore these resources, making their experience within the media bios fragmented and with limited strategic impact. This demonstrates a need for more understanding of the deepening interrelation between technologies, society, and their organizations.

In cases where there are resources but no interest or limited knowledge about the mediatized socio-organizational context, even when expressing concern about the ubiquity of face-to-face and mediated sociability, communication ends up losing value. In

other words, dialogues with their audiences become deficient, fail to generate brand experiences, and do not add value to institutional image. Despite potential recursiveness, the apparent limitations in management or professional specialization result in the communication of these organizations in media environments happening instrumentally.

Indeed, there are processes in mediatization. This is evident when we revisit the information presented in Maps I and II, noting that eight researched organizations cover a broad and varied spectrum of media, compared to five that only use more conventional media. This situation supports our understanding that some uses, and communication strategies found in various media environments can often be carried out by these organizations intuitively. Thus, the logic of each media ends up being experimented with and tested without genuinely understanding and comprehending this logic from a cultural perspective, with causes and consequences of the uses and misuses of the media ecosystem in the communication of these organizations.

This is one of the potential issues to reflect on the media field and its relationship with organizations, operating from twisted logics of the mediated, its more visible elements, and ongoing experiments in society. In this perspective, technologies are more thoroughly investigated, not as determinants of processes but as an environment in experimentation in the face of what we tentatively define as an organizational phenomenon in mediatization.

By no means do we intend to belittle or criticize the behavior of these organizations or establish models and standards facing the mediatization of the current socio-organizational praxis? We intend to counter what theories teach us with the discourse and practice of the market. This is pertinent to favor both professional formation and qualification and to encourage the pursuit of expansion of studies on this theme, object, and reality. We have yet to expose the identity of the sample, including for the preservation of its professionals, managers, and other audiences.

In concluding our report, mediatization is still a developing process in organizations in cities distant from major urban centers. However, this research is far from determining that the media configurations and organizational strategies of the researched empirical object are an axiom or a closed field of ideas. On the contrary, it presents itself as an open field, an instrument to guide other approaches and broaden the scope of discussion about the mediatized social context of organizations.

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⁵ This discusses the uses and appropriations of media in a way that goes beyond their inherent logic, adapting communication processes based on the interests and behavior of their audiences. Primarily, it aims to converge each experience in alignment with the configurations of media environments, strategically positioning organizational relationships.

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Acknowledgments

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The following is the official style and template developed for publication of a research paper. Authors are not required to follow this style during the submission of the paper. It is just for reference purposes.



Manuscript Style Instruction (Optional)

- Microsoft Word Document Setting Instructions.
- Font type of all text should be Swis721 Lt BT.
- Page size: 8.27" x 11", left margin: 0.65, right margin: 0.65, bottom margin: 0.75.
- Paper title should be in one column of font size 24.
- Author name in font size of 11 in one column.
- Abstract: font size 9 with the word "Abstract" in bold italics.
- Main text: font size 10 with two justified columns.
- Two columns with equal column width of 3.38 and spacing of 0.2.
- First character must be three lines drop-capped.
- The paragraph before spacing of 1 pt and after of 0 pt.
- Line spacing of 1 pt.
- Large images must be in one column.
- The names of first main headings (Heading 1) must be in Roman font, capital letters, and font size of 10.
- The names of second main headings (Heading 2) must not include numbers and must be in italics with a font size of 10.

Structure and Format of Manuscript

The recommended size of an original research paper is under 15,000 words and review papers under 7,000 words. Research articles should be less than 10,000 words. Research papers are usually longer than review papers. Review papers are reports of significant research (typically less than 7,000 words, including tables, figures, and references)

A research paper must include:

- a) A title which should be relevant to the theme of the paper.
- b) A summary, known as an abstract (less than 150 words), containing the major results and conclusions.
- c) Up to 10 keywords that precisely identify the paper's subject, purpose, and focus.
- d) An introduction, giving fundamental background objectives.
- e) Resources and techniques with sufficient complete experimental details (wherever possible by reference) to permit repetition, sources of information must be given, and numerical methods must be specified by reference.
- f) Results which should be presented concisely by well-designed tables and figures.
- g) Suitable statistical data should also be given.
- h) All data must have been gathered with attention to numerical detail in the planning stage.

Design has been recognized to be essential to experiments for a considerable time, and the editor has decided that any paper that appears not to have adequate numerical treatments of the data will be returned unrefereed.

- i) Discussion should cover implications and consequences and not just recapitulate the results; conclusions should also be summarized.
- j) There should be brief acknowledgments.
- k) There ought to be references in the conventional format. Global Journals recommends APA format.

Authors should carefully consider the preparation of papers to ensure that they communicate effectively. Papers are much more likely to be accepted if they are carefully designed and laid out, contain few or no errors, are summarizing, and follow instructions. They will also be published with much fewer delays than those that require much technical and editorial correction.

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Author details

The full postal address of any related author(s) must be specified.

Abstract

The abstract is the foundation of the research paper. It should be clear and concise and must contain the objective of the paper and inferences drawn. It is advised to not include big mathematical equations or complicated jargon.

Many researchers searching for information online will use search engines such as Google, Yahoo or others. By optimizing your paper for search engines, you will amplify the chance of someone finding it. In turn, this will make it more likely to be viewed and cited in further works. Global Journals has compiled these guidelines to facilitate you to maximize the web-friendliness of the most public part of your paper.

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A major lynchpin of research work for the writing of research papers is the keyword search, which one will employ to find both library and internet resources. Up to eleven keywords or very brief phrases have to be given to help data retrieval, mining, and indexing.

One must be persistent and creative in using keywords. An effective keyword search requires a strategy: planning of a list of possible keywords and phrases to try.

Choice of the main keywords is the first tool of writing a research paper. Research paper writing is an art. Keyword search should be as strategic as possible.

One should start brainstorming lists of potential keywords before even beginning searching. Think about the most important concepts related to research work. Ask, "What words would a source have to include to be truly valuable in a research paper?" Then consider synonyms for the important words.

It may take the discovery of only one important paper to steer in the right keyword direction because, in most databases, the keywords under which a research paper is abstracted are listed with the paper.

Numerical Methods

Numerical methods used should be transparent and, where appropriate, supported by references.

Abbreviations

Authors must list all the abbreviations used in the paper at the end of the paper or in a separate table before using them.

Formulas and equations

Authors are advised to submit any mathematical equation using either MathJax, KaTeX, or LaTeX, or in a very high-quality image.

Tables, Figures, and Figure Legends

Tables: Tables should be cautiously designed, uncrowned, and include only essential data. Each must have an Arabic number, e.g., Table 4, a self-explanatory caption, and be on a separate sheet. Authors must submit tables in an editable format and not as images. References to these tables (if any) must be mentioned accurately.



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Figures are supposed to be submitted as separate files. Always include a citation in the text for each figure using Arabic numbers, e.g., Fig. 4. Artwork must be submitted online in vector electronic form or by emailing it.

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TIPS FOR WRITING A GOOD QUALITY SOCIAL SCIENCE RESEARCH PAPER

Techniques for writing a good quality human social science research paper:

1. Choosing the topic: In most cases, the topic is selected by the interests of the author, but it can also be suggested by the guides. You can have several topics, and then judge which you are most comfortable with. This may be done by asking several questions of yourself, like "Will I be able to carry out a search in this area? Will I find all necessary resources to accomplish the search? Will I be able to find all information in this field area?" If the answer to this type of question is "yes," then you ought to choose that topic. In most cases, you may have to conduct surveys and visit several places. Also, you might have to do a lot of work to find all the rises and falls of the various data on that subject. Sometimes, detailed information plays a vital role, instead of short information. Evaluators are human: The first thing to remember is that evaluators are also human beings. They are not only meant for rejecting a paper. They are here to evaluate your paper. So present your best aspect.

2. Think like evaluators: If you are in confusion or getting demotivated because your paper may not be accepted by the evaluators, then think, and try to evaluate your paper like an evaluator. Try to understand what an evaluator wants in your research paper, and you will automatically have your answer. Make blueprints of paper: The outline is the plan or framework that will help you to arrange your thoughts. It will make your paper logical. But remember that all points of your outline must be related to the topic you have chosen.

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14. Arrangement of information: Each section of the main body should start with an opening sentence, and there should be a changeover at the end of the section. Give only valid and powerful arguments for your topic. You may also maintain your arguments with records.

15. Never start at the last minute: Always allow enough time for research work. Leaving everything to the last minute will degrade your paper and spoil your work.

16. Multitasking in research is not good: Doing several things at the same time is a bad habit in the case of research activity. Research is an area where everything has a particular time slot. Divide your research work into parts, and do a particular part in a particular time slot.

17. Never copy others' work: Never copy others' work and give it your name because if the evaluator has seen it anywhere, you will be in trouble. Take proper rest and food: No matter how many hours you spend on your research activity, if you are not taking care of your health, then all your efforts will have been in vain. For quality research, take proper rest and food.

18. Go to seminars: Attend seminars if the topic is relevant to your research area. Utilize all your resources.

Refresh your mind after intervals: Try to give your mind a rest by listening to soft music or sleeping in intervals. This will also improve your memory. Acquire colleagues: Always try to acquire colleagues. No matter how sharp you are, if you acquire colleagues, they can give you ideas which will be helpful to your research.

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21. Report concluded results: Use concluded results. From raw data, filter the results, and then conclude your studies based on measurements and observations taken. An appropriate number of decimal places should be used. Parenthetical remarks are prohibited here. Proofread carefully at the final stage. At the end, give an outline to your arguments. Spot perspectives of further study of the subject. Justify your conclusion at the bottom sufficiently, which will probably include examples.

22. Upon conclusion: Once you have concluded your research, the next most important step is to present your findings. Presentation is extremely important as it is the definite medium through which your research is going to be in print for the rest of the crowd. Care should be taken to categorize your thoughts well and present them in a logical and neat manner. A good quality research paper format is essential because it serves to highlight your research paper and bring to light all necessary aspects of your research.

INFORMAL GUIDELINES OF RESEARCH PAPER WRITING

Key points to remember:

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- Write your paper in the form which is presented in the guidelines using the template.
- Please note the criteria peer reviewers will use for grading the final paper.

Final points:

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This will provide understanding of the data and projections as to the implications of the results. The use of good quality references throughout the paper will give the effort trustworthiness by representing an alertness to prior workings.

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- Submitting a manuscript with pages out of sequence.
- In every section of your document, use standard writing style, including articles ("a" and "the").
- Keep paying attention to the topic of the paper.
- Use paragraphs to split each significant point (excluding the abstract).
- Align the primary line of each section.
- Present your points in sound order.
- Use present tense to report well-accepted matters.
- Use past tense to describe specific results.
- Do not use familiar wording; don't address the reviewer directly. Don't use slang or superlatives.
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Abstract: This summary should be two hundred words or less. It should clearly and briefly explain the key findings reported in the manuscript and must have precise statistics. It should not have acronyms or abbreviations. It should be logical in itself. Do not cite references at this point.

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Write your summary when your paper is completed because how can you write the summary of anything which is not yet written? Wealth of terminology is very essential in abstract. Use comprehensive sentences, and do not sacrifice readability for brevity; you can maintain it succinctly by phrasing sentences so that they provide more than a lone rationale. The author can at this moment go straight to shortening the outcome. Sum up the study with the subsequent elements in any summary. Try to limit the initial two items to no more than one line each.

Reason for writing the article—theory, overall issue, purpose.

- Fundamental goal.
- To-the-point depiction of the research.
- Consequences, including definite statistics—if the consequences are quantitative in nature, account for this; results of any numerical analysis should be reported. Significant conclusions or questions that emerge from the research.

Approach:

- Single section and succinct.
- An outline of the job done is always written in past tense.
- Concentrate on shortening results—limit background information to a verdict or two.
- Exact spelling, clarity of sentences and phrases, and appropriate reporting of quantities (proper units, important statistics) are just as significant in an abstract as they are anywhere else.

Introduction:

The introduction should "introduce" the manuscript. The reviewer should be presented with sufficient background information to be capable of comprehending and calculating the purpose of your study without having to refer to other works. The basis for the study should be offered. Give the most important references, but avoid making a comprehensive appraisal of the topic. Describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will give no attention to your results. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here.



The following approach can create a valuable beginning:

- Explain the value (significance) of the study.
- Defend the model—why did you employ this particular system or method? What is its compensation? Remark upon its appropriateness from an abstract point of view as well as pointing out sensible reasons for using it.
- Present a justification. State your particular theory(-ies) or aim(s), and describe the logic that led you to choose them.
- Briefly explain the study's tentative purpose and how it meets the declared objectives.

Approach:

Use past tense except for when referring to recognized facts. After all, the manuscript will be submitted after the entire job is done. Sort out your thoughts; manufacture one key point for every section. If you make the four points listed above, you will need at least four paragraphs. Present surrounding information only when it is necessary to support a situation. The reviewer does not desire to read everything you know about a topic. Shape the theory specifically—do not take a broad view.

As always, give awareness to spelling, simplicity, and correctness of sentences and phrases.

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Materials may be reported in part of a section or else they may be recognized along with your measures.

Methods:

- Report the method and not the particulars of each process that engaged the same methodology.
- Describe the method entirely.
- To be succinct, present methods under headings dedicated to specific dealings or groups of measures.
- Simplify—detail how procedures were completed, not how they were performed on a particular day.
- If well-known procedures were used, account for the procedure by name, possibly with a reference, and that's all.

Approach:

It is embarrassing to use vigorous voice when documenting methods without using first person, which would focus the reviewer's interest on the researcher rather than the job. As a result, when writing up the methods, most authors use third person passive voice.

Use standard style in this and every other part of the paper—avoid familiar lists, and use full sentences.

What to keep away from:

- Resources and methods are not a set of information.
- Skip all descriptive information and surroundings—save it for the argument.
- Leave out information that is immaterial to a third party.



Results:

The principle of a results segment is to present and demonstrate your conclusion. Create this part as entirely objective details of the outcome, and save all understanding for the discussion.

The page length of this segment is set by the sum and types of data to be reported. Use statistics and tables, if suitable, to present consequences most efficiently.

You must clearly differentiate material which would usually be incorporated in a study editorial from any unprocessed data or additional appendix matter that would not be available. In fact, such matters should not be submitted at all except if requested by the instructor.

Content:

- Sum up your conclusions in text and demonstrate them, if suitable, with figures and tables.
- In the manuscript, explain each of your consequences, and point the reader to remarks that are most appropriate.
- Present a background, such as by describing the question that was addressed by creation of an exacting study.
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- Do not include raw data or intermediate calculations in a research manuscript.
- Do not present similar data more than once.
- A manuscript should complement any figures or tables, not duplicate information.
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Approach:

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Put figures and tables, appropriately numbered, in order at the end of the report.

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Research papers are not acknowledged if the work is imperfect. Draw what conclusions you can based upon the results that you have, and take care of the study as a finished work.

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- Give details of all of your remarks as much as possible, focusing on mechanisms.
- Make a decision as to whether the tentative design sufficiently addressed the theory and whether or not it was correctly restricted. Try to present substitute explanations if they are sensible alternatives.
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- Recommendations for detailed papers will offer supplementary suggestions.

Approach:

When you refer to information, differentiate data generated by your own studies from other available information. Present work done by specific persons (including you) in past tense.

Describe generally acknowledged facts and main beliefs in present tense.

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<i>References</i>	Complete and correct format, well organized	Beside the point, Incomplete	Wrong format and structuring



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