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SCREENING DE FÁRMACOS POR DRIED SPOTS E *FAST*-GC–MS APLICADOS EM AMOSTRAS DE EMERGÊNCIA TOXICOLÓGICA

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Dissertação submetida ao Programa de Pós-Graduação em Ciências da Saúde da Universidade Federal de Ciências da Saúde de Porto Alegre como requisito para a obtenção do grau de Mestre.

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RESUMO

A observação de intoxicação envolvendo antidepressivos e antipsicóticos são recorrentes nos serviços de saúde gerando uma demanda para o desenvolvimento de técnicas analíticas para lidar com emergências relacionadas à toxicologia clínica. As abordagens baseadas na extração absortiva em papel, amplamente conhecida como *Dried Spots* (DS), permite a rápida detecção e identificação de substâncias tóxicas em amostras de material seco, tornando-a uma ferramenta valiosa na gestão de emergências e intoxicações. Assim, o objetivo dessetrabalho foi o desenvolvimento de uma metodologia analítica para o *screening* de medicamentos em amostras de plasma utilizando o sistema de extração de DS confeccionado em placas de 24 poços e com subsequente detecção do material extraído por cromatografia gasosa rápida acoplada a espectrometria de massa (*fast*-GC-MS). A otimização do preparo da amostra foi realizada através de planejamentosexperimentais multivariados para a obtenção da melhor resposta extrativa da amitriptilina, clorpromazina, desipramina, desvenlafaxina, fluoxetina, imipramina, norfluoxetina, nortriptilina, prometazina, venlafaxina e levomepromazina. O método foi validado de acordo com parâmetros estabelecidos pelo guia da Agência Europeia de Medicamentos que incluem, o limite inferior de quantificação (LLOQ), seletividade, linearidade, precisão, exatidão, integridade da diluição, *carryover* e estabilidade. A validação foi bem sucedida, o LLOQ variou de 20 a 60 ng/mL, a linearidade teve $r^2 > 0,99$ para todos os analitos, a precisão intra-dia variou de 0,12% a 17,7%, a precisãointer-dia de 1,1% a 18,1% e a exatidão de 87,8% a 112,2%. O método proposto foi aplicado em 102 amostras de plasma humano provenientes do Centro de InformaçõesToxicológicas do Rio Grande do Sul, com positividade de 90,2%, sendo amitriptilina, fluoxetina e clorpromazina as substâncias com maior frequência. Com os resultados obtidos, o *Dried Plasma Spot* associado a placa de 24 poços e juntamente com *fast*-GC-MS, apresentou vantagens como um método de baixo custo, fácil de implementar e rápido, sendo eficaz, principalmente, para implementação no contexto de análises de emergência.

Palavras chave: Toxicologia clínica; *Dried Plasma Spot*; *fast*-GC-MS; antidepressivos e antipscicóticos

ABSTRACT

The observation of poisoning cases involving antidepressants and antipsychotics are recurrent in health services, generating a demand for the development of analytical techniques to deal with emergencies related to clinical toxicology. Approaches based on paper absorptive extraction, widely known as Dried Spots (DS), allow the rapid detection and identification of toxic substances in dry material samples, making it a valuable tool in the management of emergencies and poisonings. Thus, the objective of this work was the development of an analytical methodology for the screening of drugs in plasma samples using the DS extraction system made in 24-well plates and with subsequent detection of the extracted material by fast gas chromatography coupled to mass spectrometry (*fast*-GC–MS). The optimization of sample preparation was performed through multivariate experimental designs to obtain the best extractive response of amitriptyline, chlorpromazine, desipramine, desvenlafaxine, fluoxetine, imipramine, norfluoxetine, nortriptyline, promethazine, venlafaxine and levomepromazine. The method was validated according to parameters established by the European Medicines Agency guide, which include the lower limit of quantification (LLOQ), selectivity, linearity, precision, accuracy, dilution integrity, carryover and stability. Validation was successful conducted with LLOQ ranged from 20 to 60 ng/mL, linearity with $r^2 > 0.99$ for all analytes, intra-day precision ranged from 0.12% to 17.7%, precision inter-day from 1.1% to 18.1% and accuracy ranged from 87.8% to 112.2%. The proposed method was applied to 102 human plasma samples obtained from the Toxicological Information Center of Rio Grande do Sul, with a positivity of 90.2%, with amitriptyline, fluoxetine and chlorpromazine being the most frequent substances. With the results obtained, the Dried Plasma Spot associated with a 24-well plate and together with *fast*-GC-MS, presented advantages as a low-cost method, easy to implement and fast, being effective, mainly, for implementation in the context of analyzes of emergency.

Keywords: Clinical toxicology; Dried Plasma Spot; *fast*-GC-MS; antidepressants and antipsychotics

LISTA DE FIGURAS

LISTA DE ABREVIATURAS

OMS	Organização Mundial da Saúde
ADT	Antidepressivos tricíclicos
CIATox	Centros de Informação e Assistência Toxicológicas
CIT	Centro de Informação Toxicológica
CIT/RS	Centro de Informação Toxicológica do Rio Grande do Sul
DBS	*Dried Blood Spot*
DPS	*Dried Plasma Spots*
DS	*Dried Spots*
GC-MS	Cromatografia gasosa acoplada a espectrometria de massas
IMAO	Inibidores da monoaminoxidase
ISRS	Inibidores seletivos da recaptação de serotonina
LC-MS/MS	Cromatografia líquida acoplada a espectrometria de massas sequencial
LLE	Extração líquido-líquido
SINAN	Sistema de Informação de Agravos de Notificação
SINITOX	Sistema Nacional de Informações Tóxico-Farmacológicas
SNC	Sistema Nervoso Central
SPE	Extração por fase sólida

SUMÁRIO

1. REFERENCIAL TEÓRICO

Uma intoxicação se define pela interação entre um agente tóxico e um organismo, produzindo efeitos nocivos, bem como, manifestações clínicas e síndromes tóxicas (EATON; GILBERT, 2012). Essa interação é decorrente da exposição do organismo a diversas substâncias químicas, como produtos químicos industriais, medicamentos,drogas, toxinas, entre outras, ocorrendo de forma acidental ou intencional. A gravidade dos efeitos da intoxicação pode variar de acordo com a dose,via de exposição, tempo de exposição, idade, saúde e outros fatores individuais.

As intoxicações são consideradas um grave problema de saúde pública e uma importante causa de mortalidade, correspondendo a uma significativa parcela dos atendimentos de serviços de saúde (OMS, 2023; SANTOS *et al*., 2021). Estima-se, que, apenas em 2019, ocorreram cerca de 84 mil fatalidades em decorrência de intoxicações. De modo geral, as diferenças de gênero e faixa etária na taxa de mortalidade por intoxicações são bem conhecidas e documentadas. Dados sugerem que os homens são mais propensos a se envolver em comportamentos de risco apresentando uma taxa de mortalidade 67% superior as mulheres. Adicionalmente, os jovens são mais propensos a se envolver em comportamentos de risco, como o uso de drogas e álcool. Além disso, esse grupo de risco pode não estar ciente dos perigos associados ao uso de certas substâncias, o que os torna mais vulneráveis à agentes tóxicos. Paralelamente, idosos podem apresentar comorbidades, como a diminuição da função renal ou hepática, que podem alterar a capacidade do corpo de biotransformar e excretar certos medicamentos ou substâncias tóxicas, tornando-os mais vulneráveis a intoxicações acidentais (OMS, 2022; SANTOS *et al*., 2021; ELLER et al., 2020; NISTOR et al., 2017)

Considerando o cenário global das intoxicações, é possível verificar que os medicamentos se destacam, sendo o grupo de agente tóxico mais frequente neste tipo de situação desde os últimos 30 anos (SILVA e OLIVEIRA, 2018). O Sistema de Informação de Agravos de Notificação (SINAN) registrou, em 2021, aproximadamente 148 mil casos de intoxicação, dentre eles, a classe de substâncias com maior frequência foi a de medicamentos, com 82.199 casos, seguindo por drogas de abuso, com 18.077 casos (MINISTÉRIO DA SAÚDE, 2023). Esses dados vão de acordo com os registros do Sistema Nacional de Informações Tóxico-Farmacológicas (SINITOX), noqual, o grupo dos medicamentos foi responsável por quase 27% das

intoxicações (FIOCRUZ, 2020). Contudo, apesar desses números serem preocupantes, eles não representam o cenário real. Estima-se que esses dados sejam ainda maiores devidoa subnotificação de casos e a dificuldade no diagnóstico de intoxicações (MAGALHÃES, CALDAS, 2019).

Existem diversos fatores que podem estar associados ao elevado número de intoxicações por medicamentos no Brasil. O país é um dos maiores mercados consumidores de medicamentos, e até 35% das vendas estão ligadas à prática da automedicação (SERENO, SILVA, SILVA, 2020). De acordo com dados apresentados por Oliveira e colaboradores, a porcentagem de brasileiros que se automedicam chega a 77% da população aumentado a probabilidade da ocorrência do uso indevido, abuso e sobredosagem (OLIVEIRA *et al.*, 2020). Adicionalmente, a sobredose intencional de medicamentos é comumente observada nas tentativas de suicídio. (FINKELSTEIN*et al.*, 2015; SPILLER, APPANA, BROCK, 2010; ANJOS *et al.*, 2021). Ao analisar o banco de dados do SINAN, é possível identificar 228.646 registros de intoxicação humana, como em casos de tentativa de suicídio em todos os estados do país, no período entre 2020 e 2022. Desse total, 183.703 registros (80,3%) estão associados ao uso de medicamentos (MINISTÉRIO DA SAÚDE, 2023).

É importante mencionar que os casos de intoxicação devem ser tratados com atenção e prioridade, pois o prognóstico do paciente pode ser influenciado pela rápida identificação da exposição e pelo tratamento administrado nas primeiras horas da admissão de emergência (OLIVEIRA *et al*., 2020). Ressalta-se, que, apesar de algumas intoxicações apresentarem manifestações clínicas leves, todo paciente intoxicado deve ser tratado como uma intoxicação potencialmente grave para que sequelas sejam evitadas (OLSON, 2014). No mais, em um ambiente clínico e emergencial, o acesso ao histórico do paciente pode não estar disponível, sendo difícil a obtenção de informações como substâncias envolvidas na exposição, dose, tempo e via de exposição (CANTILENA, 2012). Portanto, o diagnóstico dos agentes envolvidos na intoxicação representa uma etapa crucial para o prognóstico do paciente. Além disso, é imprescindível considerar as estatísticas e informações fornecidas por sistemas ou serviços especializados em toxicologia clínica, como o SINAN, SINITOX e os Centros de Informação e Assistência Toxicológicas (CIATox ou CIT). O conhecimento dos índices e prevalências das intoxicações é fundamental para a implementação de novas metodologias e análises, ampliando o escopo de

resultados. Dessa forma, os laboratórios podem fornecer resultados mais assertivos aos pacientes e à equipe médica envolvida, além de prestar assessoria especializada.

Diante esse cenário, torna-se importante o desenvolvimento de estratégias e manobras clínicas visando o prognóstico positivo para o paciente. O manejo e tratamento da intoxicação são diretamente influenciados pelo diagnóstico correto, para o qual são utilizadas abordagens como exame clínico para verificar os sinais vitais e identificar possíveis síndromes tóxicas, contudo, a identificação das síndromes tem sido dificultada pelas manifestações inespecíficas e poliuso de substâncias (AMARAL e HERNANDEZ, 2014; UGES, 2011). Deste modo, as análises toxicológicas são imprescindíveis para o diagnóstico correto, além de auxiliar na escolha adequada de tratamento e monitorar o processo de detoxificação, contribuindo para o prognóstico positivo, evitando medidas terapêuticas desnecessárias ao paciente (DOS SANTOS; VIANNA; SEBBEN, 2022; OLSON, OHRA, 2018).

1.1 Panorama das intoxicações por antidepressivos e antipsicóticos

As classes de medicamentos mais frequentemente associadas aos casos de intoxicação humana incluem antidepressivos, antipsicóticos, benzodiazepínicos e o acetaminofeno (DOS SANTOS *et al.,* 2023; CIT/RS, 2022). Em especial, neste trabalho, serão abordados os medicamentos das classes dos antidepressivos e antipsicóticos.

Os antidepressivos e antipsicóticos são prescritos para o tratamento de transtornos mentais, o qual pode ser definido como uma perturbação clinicamente significativa na cognição, na regulação emocional ou no comportamento de um indivíduo. Os transtornos mais comuns são a depressão, ansiedade, bipolaridade e esquizofrenia. Estima-se que uma a cada oito pessoas no mundo sofre com algum tipo de transtorno mental, totalizando 970 milhões de indivíduos (OMS, 2022).

Tanto os antidepressivos quanto os antipsicóticos são classificados como fármacos psicotrópicos e estão entre os mais prescritos no mundo. Além disso, o uso desses medicamentos vem aumentando progressivamente a cada ano (PRADO *et al.*, 2017). O fácil acesso promove o seu uso em casos de intoxicação, tanto

intencional, como no caso de tentativa de suicídio, quanto acidental (MATHIAS, GUIDONI, GIROTTO, 2019).

O Sistema Nacional de Dados de Intoxicação dos Estados Unidos da América reportou, em 2022, 136 mil relatos de intoxicação por antidepressivos, representando a quarta categoria de substância com maior frequência de casos e oitavo lugar quanto a fatalidade. Os antipsicóticos registraram 7 mil relatos, ocupando o primeiro lugar na categoria de substâncias com maiores casos de fatalidades (GUMMIN *et al*., 2021). Concomitantemente, é comprovado que a depressão e o transtorno de bipolaridade são fortes fatores de risco para o suicídio, sendo agravado quando não realizado o tratamento adequado (TOO *et al*., 2018; NOCK *et al*., 2009; CRUMP *et al.*, 2013). Na Inglaterra, os antidepressivos são os medicamentos mais comuns ingeridos na tentativa de suicídio e que representam 20% dos casos fatais (HAWTON *et al*., 2010; WELLS *et al*, 2009; BERGEN *et al.*, 2010). Estima-se que o uso de antidepressivos alcance 1,85 milhão de pessoas por ano na Suécia. Além disso, os Inibidores Seletivos de Recaptação de Serotonina (ISRS) representam 76% do consumo desses medicamentos. O Departamento de Química Forense do Conselho Nacional de Medicina Legal da Suécia investigou 14.857 casos de suicídio e com base nas análises toxicológicas, os antidepressivos tiveram positividade de 20% e dentre eles, os ISRS foram responsáveis por 48% (ISACSSON, HOLMGREN, AHLNER, 2005).

O perfil de intoxicação observado é semelhante ao relatado pelo Centro de Informação Toxicológica do Rio Grande do Sul (CIT/RS). No ano de 2021, ocorreram 25 mil casos de intoxicação, sendo que 16.438 desses casos foram causados por medicamentos, tornando-se o grupo com maior frequência de ocorrência. Os antidepressivos foram a classe de medicamento mais frequentemente, registrando 3.223 casos, enquanto a classe de antipsicóticos ocupou o quarto lugar com 1.487 casos registrados (CIT/RS, 2022).

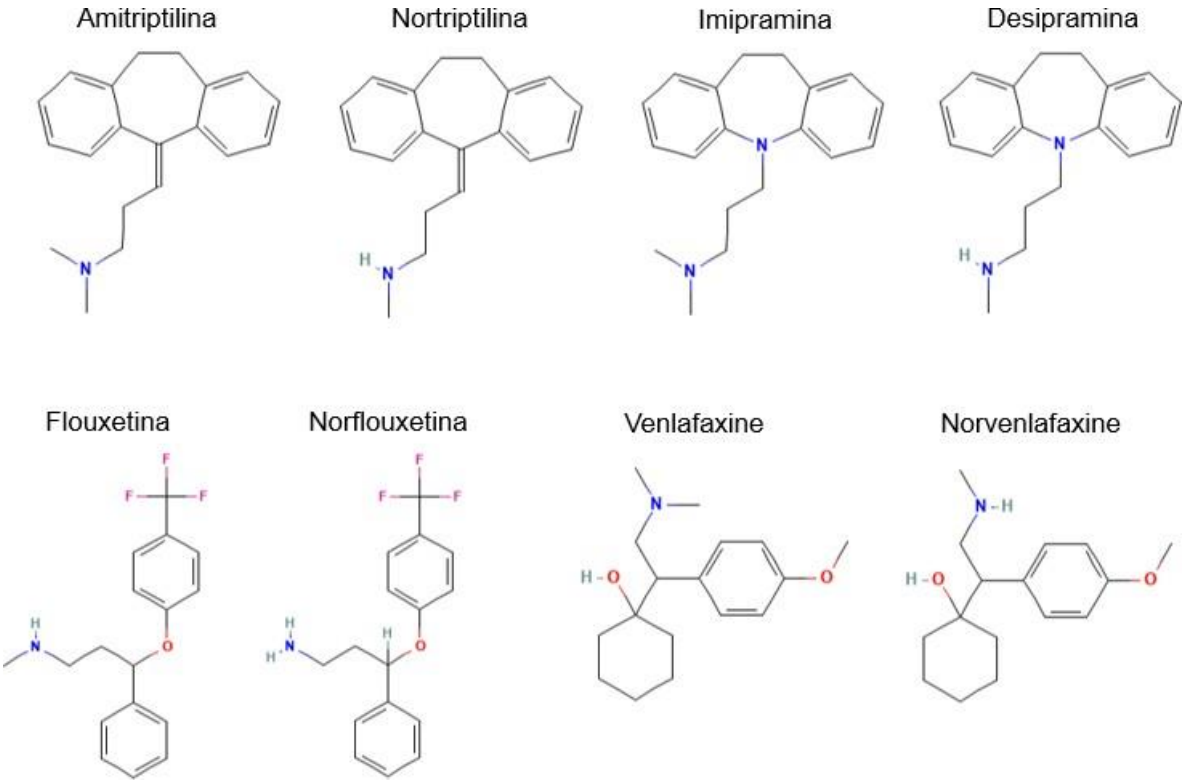
Diante do exposto, a preocupação em relação aos casos de intoxicação por antidepressivos e antipsicóticos no Brasil se torna uma questão de saúde pública que demanda atenção. É essencial, portanto, que sejam realizadas análises toxicológicas para auxiliar na identificação do agente tóxico, permitindo um diagnóstico preciso e um prognóstico favorável para o paciente. Além disso, é importante ressaltar que os dados obtidos por essas análises são fundamentais para conscientizar a população sobre o uso correto e seguro de medicamentos, bem como para promover

intervenções educacionais e regulatórias que visem reduzir a incidência de intoxicações.

1.2.1 Antidepressivos

Os medicamentos antidepressivos são empregados no tratamento de diversos transtornos mentais, tais como depressão, mudanças de humor, transtorno bipolar e síndrome do pânico, entre outros. Existem diversas classes de antidepressivos, que podem ser classificados de acordo com seu mecanismo de ação ou sua estrutura química (SPIELMANS *et al.*, 2013). Além dos ISRS, anteriormente mencionados, as principais classes desses medicamentos incluem os antidepressivos tricíclicos (ADT), os inibidores da monoaminoxidase (IMAO), os atípicos e os de nova geração. Em especial, neste trabalho, serão abordadas as classes dos ADT e ISRS, os quais possuem as estruturas ilustradas na **Figura 1**.

Figura 1. Estrutura química dos antidepressivos



Fonte: National Centeí foí Biotechnology Information, 2023.

Os ADTs são classificados dessa forma devido à presença comum de três anéis de benzeno presente na sua estrutura química. O seu mecanismo de ação consiste em dois transportadores de recaptação e três proteínas receptoras: inibindo os transportadores pré-sinápticos de recaptação de norepinefrina; inibição de transportadores pré-sinápticos de recaptação de serotonina; bloqueio dos receptores pós-sinápticos adrenérgicos α_1 e α_2; bloqueio de receptores muscarínicos pós-sinápticos; e bloqueando os receptores H_1 da histamina pós-sináptica (HILLHOUSE; PORTER, 2015). O bloqueio da recaptação de serotonina e noradrenalina nos terminais pré-sinápticos, leva ao aumento da concentração desses neurotransmissores na fenda sináptica, contribuindo para seu efeito terapêutico. O antagonismo dos receptores adrenérgicos, muscarínicos e histaminérgicos favorece os efeitos colaterais, como, tontura, comprometimento da memória e sonolência, respectivamente. Embora os ADTs tenham eficácia similar aos ISRSs, é comum haver mais relatos de efeitos adversos após o seu consumo pois apresentam um menor limiar para superdosagem (MORACZEWSKI e AEDMA, 2022; HILLHOUSE; PORTER, 2015).

A afinidade dos receptores pelos ADTs é influenciada pela sua estrutura química. ADTs que possuem anéis ligados a aminas terciárias, como amitriptilina, clomipramina, doxepina, imipramina e trimipramina, tendem a favorecer o bloqueio da recaptação de serotonina. Por outro lado, ADTs que possuem ligações à aminas secundárias, como desipramina, nortriptilina e protriptilina, apresentam maior bloqueio da recaptação de norepinefrina (MORACZEWSKI e AEDMA, 2022; HILLHOUSE; PORTER, 2015).

Os ADTs são rapidamente absorvidos pelo trato gastrointestinal e sofrem metabolismo de primeira passagem. Adicionalmente, se ligam fortemente à albumina plasmática, atingindo até 95% em concentrações terapêuticas, e aos tecidos extravasculares, indicando grandes volumes de distribuição, resultando em uma meia-vida prolongada, em torno de 24 horas, com exceção da amitriptilina que apresenta uma meia-vida de 46 horas (KERR; McGUFFIE; WILKIE, 2001). A biotransformação ocorre principalmente no fígado, por enzimas do citocromo P450 (CYP450). A via pode variar dependendo do tipo de antidepressivo tricíclico, mas em geral, ocorre por desmetilação, hidroxilação e subsequente glucuronidação. Os TCAs terciários, como a amitriptilina e imipramina, são biotranformados em seus produtos

de aminas secundárias, como nortriptilina e desipramina, respectivamente, por meio da desmetilação mediada pela enzima CYP2C19. Os TCAs secundários, como a nortriptilina e a desipramina, são hidroxilados em seus produtos hidroxilados por enzimas, respectivamente. Os produtos resultantes são então conjugados com ácido glicurônico e excretados na urina. Os produtos ativos geralmente apresentam uma menor atividade inibitória da recaptação de serotonina e norepinefrina do que os compostos precursores (D'EMPAIRE *et al.*, 2011; ALCHAKEE *et al.*, 2022). As concentrações terapêuticas dos antidepressivos tricíclicos variam de 0,02 a 0,5 µg/mLe a concentração plasmática tóxica de 0,3 a 1 µg/mL (SCHULZ *et al.*, 2020).

Os ISRS atuam na inibição da recaptação da serotonina no terminal axônio pré-sináptico, aumentando a quantidade de serotonina na fenda sináptica e estimulando os receptores pós-sinápticos por um período prolongado. Este mecanismo específico, por não obter efeitos sobre outros neurotransmissores, apresenta uma menor incidência de efeitos colaterais quando comparado às outras classes de antidepressivos. Por este motivo, os ISRS são opções de primeira linha para o uso terapêutico (CHU e WADHWA, 2022). Assim como os ADT os ISRS apresentam uma rápida absorção e uma alta biodisponibilidade, com ligação de proteínas plasmáticas variando entre 56 - 98%. O processo de biotransformação dos ISRS envolve principalmente as enzimas hepáticas do citocromo P450, com o CYP2D6 e o CYP3A4 sendo as principais enzimas envolvidas na biotransformação de vários compostos dessa classe de medicamentos. Adicionalmente, é importante mencionar que a fluoxetina é um ISRS que possui uma inibição significativa da enzima CYP2D6, o que pode levar a interações medicamentosas importantes uma vez que esta enzima é responsável pela biotransformação de muitos medicamentos, incluindo alguns antidepressivos tricíclicos, antipsicóticos, beta-bloqueadores e opioides (HERNANDEZ, RODRIGUES, TORRES, 2017).

Embora os ISRSs tenham um perfil farmacológico geralmente seguro e uma boa tolerabilidade, o uso excessivo desses medicamentos pode levar à síndrome serotoninérgica, especialmente quando combinado com outros antidepressivos ou drogas de abuso. A síndrome serotoninérgica é caracterizada por uma variedade de sintomas, incluindo agitação, taquicardia, hipertensão, hipertermia, hiperreflexia, tremores, midríase e náusea (HERNANDEZ, RODRIGUES, TORRES, 2017). Embora não exista um tratamento específico para a síndrome serotoninérgica, é importante

interromper imediatamente o uso do medicamento que está causando a condição. Além disso, o tratamento de suporte é essencial, o que pode incluir hidratação intravenosa, monitoramento dos sinais vitais e controle dos sintomas específicos. Os benzodiazepínicos são frequentemente usados para tratar a agitação, pois ajudam a reduzir a atividade excessiva do sistema nervoso central. No entanto, em casos graves, outros medicamentos podem ser necessários para controlar a pressão arterial elevada e a hipertermia.(CHU e WADHWA, 2022).

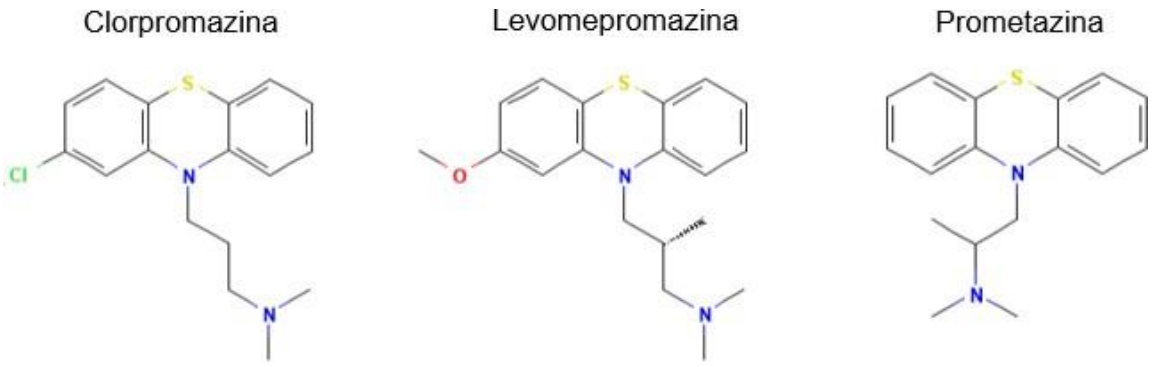
1.2.2 Antipsicóticos

Os antipsicóticos são prescritos para o tratamento de alucinações e delírios em pacientes com distúrbios neuropsiquiátricos, como, esquizofrenia e transtorno bipolar. A descoberta da clorpromazina, classificada como uma fenotiazina, representou um marco importante na história da psicofarmacologia e da psiquiatria, uma vez que foi o primeiro antipsicótico a ser desenvolvido com comprovada eficácia clínica (MELTZER, 2013; GOLLAPUDI e RADHAKRISHNAN, 2011).

O bloqueio pós-sináptico dos receptores dopaminérgicos D_2 está relacionado à melhora dos sintomas psicóticos e cognitivos. No entanto, episódios extrapiramidais podem ser comuns devido ao bloqueio destes receptores no núcleo estriado cerebral (MIYAKE, MIYAMOTO, JARSKOG, 2012).

Com a descoberta da clorpromazina e o avanço da química medicinal, novos fármacos foram sintetizados e introduzidos na prática clínica, como a prometazina e levomepromazina. As moléculas desses compostos são apresentadas na **Figura 2**.

Figura 2. Estrutura química dos antipsicóticos



Fonte: National Centeí foí Biotechnology Information, 2023.

A levomepromazina é utilizada, principalmente, como sedativa para transtornos psicóticos, além de apresentar eficácia em broncodilatação, analgésico pós-operatório e controle de dor e náusea. As ações terapêuticas da levomepromazina como um antipsicótico são decorrentes da sua capacidade de bloqueio dos receptores de dopamina, especificamente nos receptores D_2, apresentando maior afinidade à ligação dos receptores do que a clorpromazina e clozapina (GREEN *et al.*, 2004; HIGASHIMA *et al*., 2004; DEVAPRIAM *et al*., 2008).

Dentre outros fármacos derivados dos fenotiazínicos, inclui-se a prometazina. Apesar da prometazina ser classificada como um anti-histamínico e ser prescrita para controle e tratamento de condições alérgicas, ela é prescrita como coadjuvante no tratamento psiquiátrico, devido a suas características para sedação, enjoo, náuseas e vômitos (BJM, 2003; SOUTHARD e AL KHALILI, 2022).

A capacidade da prometazina de atravessar a barreira hematoencefálica e agir no SNC resulta nas suas propriedades antidopaminérgicas, anti-histamínicas e anticolinérgicas. Seu mecanismo de ação ocorre devido ao bloqueio dos receptores H_1 da histamina, mediando assim a vigília e a atenção. Adicionalmente, atua também, como antagonista direto nos receptores mesolímbicos de dopamina e receptores alfa-adrenérgicos no cérebro, evitando náuseas e vômito (OHLOW; MOOSMANN, 2011; MOREIRA; GUIMARÃES, 2007).

As concentrações terapêuticas dos antipsicóticos em questão, variam entre 0,02 a 0,1 µg/mL e as concentrações plasmáticas tóxicas são de 0,4 µg/mL para levomepromazina e 1 µg/mL para clorpromazina e prometazina. No entanto, efeitos adversos e intoxicações por antipsicóticos podem ser verificados ainda na dose terapêutica. Os sinais e sintomas clínicos da intoxicação incluem principalmente depressão do SNC, síndrome extrapiramidal e taquicardia (SCHULZ *et al.*, 2020; OLSON, 2014; THANACOODY, 2020).

1.3 Métodos analíticos para determinação de substâncias psicoativas

Nas análises toxicológicas de emergência geralmente são empregados testes rápidos de triagem, como a imunocromatografia para detecção de substâncias psicoativas. Contudo, essas técnicas apresentam pouca detectabilidade para diferentes classes de compostos, ou seja, há a possibilidade de não ocorrer a

detecção do agente tóxico específico e, ainda, é comum a ocorrência de resultados falso-positivos, devido à sua baixa especificidade (BRAHM *et al.*, 2010; MASTERNAK; PADAŁA; KARAKUŁA-JUCHNOWICZ, 2021; SEGEL; BALKRISHNAN; HIRTH, 2017; SAITMAN, PARK, FITZGERALD, 2014).

Além disso, os resultados positivos ou inconclusivos em imunoensaios necessitam, obrigatoriamente, de análise confirmatória por metodologias robustas, sendo recomendado a utilização de técnicas com elevada seletividade como a cromatografia líquida acoplada à espectrometria de massas sequencial (LC-MS/MS) ou a cromatografia gasosa acoplada a espectrometria de massas (GC-MS). Estudos apontam para uma substituição gradual dos testes de imunocromatografia pelo *screening* utilizando técnicas de LC-MS/MS ou GC-MS, uma vez que ambas as metodologias permitem diferenciar compostos com estrutura química similar ou em baixas concentrações (RITTNER *et al*., 2001; ADAMOWICZ, KAŁA, 2010; EICHHORST *et al.*, 2009; STRANO-ROSSI *et al.*, 2010). Apesar do avanço na área instrumental a análise de amostras biológicas *in natura* não é compatível com essas técnicas, sendo necessário o emprego de etapas de extração que promovem a purificação e subsequente concentração do xenobiótico da matriz (LUIZ OENNING *et al*., 2020; NIU *et al*., 2018).

Técnicas de extração clássicas, como extração por fase sólida (SPE, do inglês: *solid-phase extraction*) ou extração líquido-líquido (LLE, do inglês: *liquid–liquid extraction*) demandam tempo, elevado volume de amostra e solventes tóxicos, além de serem por muitas vezes caras e não compatíveis com os princípios da química verde (SAJID; PŁOTKA-WASYLKA, 2022; NOMURA *et al*., 2013; CHENG *et al*., 2019). A partir desse cenário, novas metodologias foram desenvolvidas a fim de se adequar às novas demandas da toxicologia e da química verde, como o caso da técnica de *Dried Spots* (DS).

A técnica de DS foi originada nos anos 60 sendo incialmente aplicada para a triagem neonatal de distúrbios metabólicos hereditários e, atualmente, vem sendo amplamente aprimorada para o monitoramento clínico e toxicológico de diversas substâncias (DÉGLON *et al*., 2010; LI *et al*., 2020; DA SILVA *et al*., 2020; ANTUNES; CHARÃO; LINDEN, 2016). O DS baseia-se na adição de um pequeno volume de amostra em um papel filtro específico, após a absorção da amostra pelo papel, a extração do analito é promovida pela solubilização do material em diferentes

solventes orgânicos (WAGNER *et al*., 2014; JAIN *et al.*, 2017; SCRIBEL *et al*., 2019; CHRISTIAN *et al.*, 2020).

Atualmente, existe uma ampla diversidade de matrizes biológicas compatíveis com o DS, como por exemplo, sangue total, plasma, urina e fluido oral, denominando-se *Dried Blood Spots*, *Dried Plasma Spots*, *Dried Urine Spots* e *Dried Oral Fluid Spots*, respectivamente (DEMIREV, 2012; LI *et al.*, 2020; PABLO; BREAUD; CLARKE, 2020; HAN *et al*., 2021). Paralelamente, a compatibilidade com amostras postmortem foi verificada por LAUER *et al*. que utilizaram alíquotas de utilizam sangue total periférico, urina, bile, humor vítreo, pericárdio e líquido cefalorraquidiano aplicando no papel filtro (LAUER *et al*., 2013).

Uma das suas principais vantagens do DS é a coleta amostral facilitada. As amostras de *Dried Blood Spot* (DBS) podem ser obtidas através de perfurações com lanceta em um dos dedos da mão, aplicando em um card específico. Dessa maneira, é dispensado treinamento, profissionais habilitados e ambiente esterilizado pois a manipulação do material biológico no local de coleta é mínima, além de ser considerada uma coleta segura, menos invasiva e com poucos riscos quando comparada à coleta de punção venosa ou coleta de urina assistida, por exemplo (LI, LEE, 2014; ANTELO-DOMÍNGUEZ *et al.*, 2013).

O transporte sem monitoramento de temperatura é possível devido à alta estabilidade dos analitos no papel seco. FENG *et al*., 2019 relatam que venlafaxina e a desvenlafaxina pode ser estável por três meses. DÉGLON *et al.,* analisaram fluoxetina e norfluoxetina por 30 dias e constataram que ambas apresentaram boa estabilidade e não obtiveram diferença significativa entre as temperaturas, confirmando que o DBS pode ser armazenado em ambiente temperatura por um período relativamente longo. Adicionalmenete, outros estudos apontam para a estabilidade prolongada de diversas substâncias, não havendo a necessidade do controle de temperatura durante a estocagem (WAGNER *et al*., 2014; PALMER, COOPER, DUNN, 2019; MORETTI *et al*., 2019).

Uma desvantagem importante do método de bioamostragem por DBS é a necessidade de ajustes e correções nos valores de hematócrito para garantir resultados precisos. Amostras de sangue com baixas concentrações de hemoglobina e consequentemente, hematócritos baixos, se espalham mais rapidamente do que

aquelas com concentrações normais ou altas de hemoglobina, levando à resultados equivocados (ADAM *et al*., 2000; LI, LEE, 2014).

Dessa maneira, a utilização de *Dried Plasma Spots* (DPS) possui as mesmas vantagens do DBS, além de dispensar a necessidade de cálculos de correção de hematócritos (LI *et al*., 2020; DA SILVA *et al*., 2020). Concomitantemente, a escolha do DPS está de acordo com o contexto clínico, na qual, a análise do plasma é mais indicada, pois revela a disponibilidade do agente tóxico para distribuição ou reabsorção, permitindo assim a correta correlação com sinais e sintomas e o direcionamento do paciente para o tratamento mais adequado (POKLIS, 2012; CANTILENA, 2012).

O DS necessita de uma metodologia analítica sensível e específica para a identificação dos compostos. Deste modo, a GC-MS apresenta uma boa sensibilidade e especificidade, algo fundamental para a detecção de fármacos em amostras de DS com baixa concentração, principalmente para aplicações na toxicologia clínica (ANTUNES, CHARÃO, LINDEN, 2016; MARDER *et al.*, 2020).

A cromatografia gasosa (GC) convencional pode ser definida como um método físico-químico de separação onde os constituintes da amostra são vaporizados em um injetor operando com alta temperatura e eluídos de acordo com sua interação com uma fase estacionária ou com uma fase móvel, portanto, assume-se que a volatilidade da molécula seja o principal requisito para separação dos compostos alvo. A eluição é feita por um fluxo de fase móvel gasosa inerte que não apresenta uma interação com as moléculas de interesse, sendo sua única função transportar o analito através da fase estacionária, composta por uma coluna cromatográfica dentro de um forno de aquecimento (SKOOG *et al*., 2014). Assim, os componentes da amostra são eluídos conforme a temperatura do forno e interação com a coluna, sendo então conduzidos para o detector, emitindo um sinal elétrico que é registrado graficamente (PENTEADO, MAGALHÃES, MASINI, 2008).

O acoplamento do GC com a espectrometria de massas (MS, do inglês: *mass spectrometry*) une as vantagens de ambas as metodologias e podem ser descritas pela elevada eficiência na separação química dos compostos, obtenção de informação estrutural da molécula, massa molar e alta seletividade e especificidade, possibilitando a identificação de diferentes substâncias (CHIARADIA, COLLINS, JARDIM, 2008). Em GC-MS, devido ao uso constante de uma energia de 70 eV, a

fragmentação das moléculas será linear, como uma “impressão digital”, permitindo a comparação de espectros com bibliotecas de referência mundial possibilitando a identificação de compostos desconhecidos pelo analista. A presença de bibliotecas cromatográficas, é uma das principais vantagens no GC-MS frente a um LC-MS, o qual, só é possível a identificação do composto perante a aquisição de padrões analíticos.

A análise de antidepressivos e antipsicóticos em GC-MS já está bem descrita na literatura (CARAMELO *et al*., 2019; WINECKER, 2009; PIETRACCI *et al*., 2012; FENG *et al*., 2019; ROSADO *et al*., 2017; SOARES *et a*l., 2021; CHEN *et al*., 2017; GUNNAR *et al.*, 2004; DA FONSECA *et al*., 2013; LUIZ OENNING *et al*., 2020). No entanto, a duração da análise cromatográfica pode ser uma desvantagem, apresentando em média um tempo total entre 20 - 35 minutos para uma única análise.

Dessa maneira, uma vertente do GC-MS, o *fast*-GC-MS, é uma alternativa para otimizar o tempo necessário para a análise cromatográfica. A troca de uma coluna convencional, normalmente com comprimento de 30 m (metros) e diâmetro interno de 0,32 mm (milímetro), para uma coluna menor, apresentando comprimento de 10 a 20 m e diâmetro de 0,10 a 0,25 mm, permite reduzir os tempos de execução cromatográfica através de ajuste do forno gradiente de temperaturas, fluxo do gás e pressão, aumentando, desse modo, o número de pratos teóricos (STRANO-ROSSI *et al*., 2010; MAŠTOVSKÁ, LEHOTAY, 2003; CAGLIERO *et al*., 2022).

Em geral, os métodos de *fast*-GC-MS são construídos com um tempo de análise inferior a 10 minutos. DÉGLON *et al.* analisou quatro antidepressivos e RANA, URALETS, ROSS nove, em ambas as corridas cromatográficas apresentaram menos de seis minutos (DÉGLON *et al*., 2010; RANA; URALETS; ROSS, 2008). A vantagem de acelerar a o tempo de análise cromatográfico utilizando a *fast*-GC-MS, é a possibilidade de aumentar o alcance das análises laboratoriais e a rapidez com que se conclui o resultado de uma amostra (DÖMÖTÖROVÁ, MATISOVÁ, 2008). Essa vantagem é extremamente valiosa, especialmente no cenário de emergência toxicológica clínica.

2. OBJETIVOS

2.1. Objetivo Geral

Desenvolver de uma metodologia analítica para o *screening* demedicamentos em amostras de plasma utilizando o sistema de extração de DS confeccionado em placas de 24 poços e com subsequente detecção do material extraído por cromatografia gasosa rápida acoplada a espectrometria de massa (fast-GC-MS).

2.2 Objetivos Específicos

- Otimizar dos parâmetros da técnica de *Dried Plasma Spots*;
- Desenvolver o método cromatográfico e espectrométrico;
- Validar a metodologia conforme guia de validação bioanalítica;
- Avaliar a estabilidade dos analitos no papel seco;
- Analisar amostras de emergência provindas do Centro de Informação Toxicológica do Rio Grande do Sul (CIT/RS).

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4. ARTIGO CIENTÍFICO

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A new approach with dried plasma spots: a rapid determination of antidepressants and antipsychotics by fast-GC-MS

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Abstract

The increasing prevalence of poisoning cases related to antidepressants and antipsychotics has raised concerns. To address this issue, a new adaptation of the dried plasma spot technique was developed using a 24-well plate and fast gas chromatography-mass spectrometry. The method involves the optimization of extraction variables and sample preparation and was successfully validated. The limits of quantitation ranged from 20 to 60 ng/mL, and accuracy ranged from 87.8% to 112.2%. The technique was applied to 102 human plasma samples from a Toxicological Information Center, with positivity of 90.2%. The method provides a cheap, easy-to-implement, and fast approach, making it ideal for toxicological emergency laboratories and promoting valuable support for healthcare professionals managing poisoning cases involving antidepressants and antipsychotics.

Keywords: antidepressants, antipsychotics, dried plasma spot, fast gas chromatography-mass spectrometry.

Introduction

According to the World Health Organization, over 970 million individuals worldwide who are affected by mental illness. This condition includes a broad range of disorders that affect an individual's thinking, mood, and behavior, such as anxiety, depression, bipolar disorder, schizophrenia, and substance use disorders [1]. While psychotherapy is a primary form of treatment for mental illness, medication may also be prescribed in some cases to manage symptoms such as depression, anxiety, or psychosis. In this scenario, antidepressants and antipsychotics are commonly used medications with a broad range of different pharmacologic effects [2,3].

The main indication of antidepressants is to alleviate symptoms associated with depression and anxiety [4]. Different types of antidepressants act on different neurotransmitters to modulate mood and emotional states. Antipsychotics, on the other hand, are prescribed to manage symptoms of psychotic disorders such as schizophrenia and bipolar disorder [5]. However, both antidepressants and antipsychotics can produce a range of side effects, including nausea, dizziness, sexual dysfunction, weight gain, insomnia, metabolic disturbances, extrapyramidal symptoms, sedation, and cognitive impairments, among other side effects [6-9]. Globally, cases of intoxication are increasingly occurring due to the high number of prescriptions for these drugs.

In 2022, the National Intoxication Data System of the U.S. reported 136,000 cases of intoxication caused by antidepressants, which ranked fourth in terms of frequency of cases and eighth in terms of fatality. Meanwhile, antipsychotics registered 7,000 cases and ranked first in terms of substances with the highest cases of fatalities [10]. Furthermore, a study analyzing cases reported to U.S. poison control centers from 2000 to 2014 showed that the rates of exposure to antidepressants increased by 64%, and the rates of exposure to antipsychotics increased by 63% over the analyzed period [11]. Similar reports show that there has been an increase in the number

of cases of antidepressants and antipsychotic intoxication in Australia, Asia, and Europe [12,13]. In Brazil, an increase in the number of cases of poisoning by antidepressants and antipsychotics has been reported in recent years. Such occurrences represent a serious threat to public health, underscoring the need for more comprehensive measures to mitigate the risk of exposure to medication-related harm [14].

Intoxication cases generally require careful attention, as patients may present with non-specific clinical signs and symptoms. Obtaining information about the toxic agent can also be challenging, making an accurate diagnosis difficult. Therefore, analyzing these substances is crucial for the adequate management and treatment of intoxicated patients, particularly within the first few hours of hospital admission [15,16]. Both liquid chromatography (LC) and gas chromatography (GC) coupled with mass spectrometry (MS) are proposed as efficient analytical methodologies for the determination of toxic agents. However, a sample preparation step is mandatory before the analysis itself.

In this context, the dried spot (DS) biosampling approach is widely recommended due to its pre-analytical simplicity and numerous advantages. These include a small volume requirement, increased stability of numerous analytes, and easy storage and transportation [17-20]. This method presents several advantages over traditional sampling techniques, as it is less invasive and requires minimal preparation.

The DS technique is a sample collection approach that involves the deposition of a small amount of a biological sample onto a specialized filter paper. As the paper absorbs the sample, it dries quickly, creating a dried spot that contains a considerable amount of biological components, including proteins, cells, and metabolites. These components become trapped, and the resulting dried spot can be easily stored and transported. For further analysis, a small portion of the dried spot can be extracted and reconstituted in a desorption solution. The DS technique can be used to collect various biological matrices, such as blood, urine, and saliva for toxicology screening [21-

24].

The use of GC for analyzing antidepressants and antipsychotics is already well described in the literature [25-29]. Nevertheless, it is notable that the duration of the analytical run, which usually ranges from 20 to 35 minutes, can be a disadvantage. Therefore, fast GC methodologies can be applied to achieve rapid analysis of numerous compounds, reducing the time and, consequently, the costs when compared to traditional GC methodologies. These methods typically utilize narrow-bore columns and higher carrier gas flow rates to decrease analysis time while maintaining adequate resolution and sensitivity [30,31]. As a result, fast GC can be a useful tool for high-throughput analysis in a variety of applications.

Taking into account this increased demand, the aim of this work was to develop a rapid, straightforward, and efficient method for the determination of antidepressants and antipsychotics in dried plasma samples (DPS) by fast GC coupled with mass spectrometry (fast-GC-MS), to provide a rapid diagnosis in intoxication cases that occurred in Rio Grande do Sul state, Brazil.

Materials & Methods

Material and reagents

Acetonitrile and methanol (LC-MS grade) were purchased from Merck® (Darmstadt, Germany). Analytical standards of amitriptyline, chlorpromazine, desipramine, fluoxetine, imipramine, nortriptyline, promethazine, venlafaxine, levomepromazine, and ketamine-d_4 (1 mg/mL) were acquired from Cerilliant® (Round Rock, TX, USA). Norfluoxetine and desvenlafaxine (1 mg/mL) were acquired from LGC® (Teddington, London, UK). The qualitative filter paper, grade 1, 125 mm diameter obtained from Whatman® (Westborough, USA).

Preparation of solutions

A working solution with all analytes was prepared in methanol, at a concentration of 10

μg/mL. Solutions were stored in borosilicate vials at − 20 °C when not in use. Fortified solutions employed for optimization and validation procedures were obtained by adding the working solution to blank plasma samples. All blank plasma samples were collected from the researchers themselves and previously tested for searched analytes. The internal standard solution of ketamine-d_4 was prepared at 500 ng/mL in methanol and acetonitrile in a proportion 1:1 (v/v).

Instrumental and chromatographic conditions

The chromatographic analysis was performed in a GC-MS-QP 2010 Plus gas chromatograph coupled to mass spectrometry (Shimadzu, Kyoto, Japan) and autosampler model AOC-20i (Shimadzu, Kyoto, Japan). Chromatographic separation was carried out in a Rxi fused silica capillary column model RH-Rix-5MS (20 m × 0.18 mm × 0.18 μm). Ultrapure helium 5.5 was used as a mobile phase, with a constant flow of 1.52 mL min^{-1}. The injector temperature was set at 250 °C. The initial oven temperature was set at 180 °C, holding for 0.50 min, increasing to 200 °C at 60 °C/min, and held for 0.3 min, then increased to 300 °C at 50 °C/min, and holding for 1.20 min, totalizing 4.3 min of chromatographic run. The injection was performed using split mode in the proportion of 1:5. The pressure was at 297 kPa and the temperature of the ion source was set at 200 °C and the interface at 200 °C. The mass spectrometer was operated in electron ionization (EI) mode at 70 eV. The acquisition was performed in scan mode. **Table 1** shows the respective *m/z* of identification and quantification ions and retention time for each analyte. Data were extracted using GCMSsolution software (Shimadzu, Kyoto, Japan).

Experimental Procedure for dried plasma spots on a 24-Well Plate

The DPS extraction was performed as follows: Grade 1 qualitative filter paper with a diameter of 125 mm was cut into 15 mm diameter disks using a circular paper cutter. The disks were washed with an acetone solution for decontamination and dried completely. Two 15 mm

disks were transferred to each well of a polypropylene 24-well plate, resulting in a total of 36 mg of paper as an absorbent layer. An aliquot of 70 µL of plasma was transferred to each well and left to completely dry at room temperature for 30 min. The analytes were desorbed by adding a 320 µL mixture of methanol and acetonitrile in a 1:1 (v/v) ratio, containing an internal standard (ketamine-d_4; 500 ng/mL). The plate was placed on an orbital shaker at 200 rpm for 10 minutes. Afterward, a 200 µL aliquot of the extraction solvent mixture was collected and dried under nitrogen. The residue was reconstituted with 50 µL of methanol and 2 µL of the resulting solution was injected into the analytical system.

Optimization of the methodology proposed

Optimizing the variables in an extraction methodology can enhance its performance and analytical response using statistical models based on multivariate experimental designs [31]. In order to optimize the extraction, a simplex-centroid design using a ternary mixture of methanol, isopropanol, and acetonitrile was performed to find the best desorption solvent. Subsequently, Doehlert design was used to optimize two variables, one at three different levels and the other at five different levels. In total, the experiment was conducted to test the sample volumes (70, 100, 120, 140, and 170 µL) and desorption solvent volume (180, 250, and 320 µL).

Validation parameters

The method validation was based on The European Medicines Agency's (EMA) guideline on bioanalytical method validation [32]. The parameters evaluated were the low limit of quantification (LLOQ), selectivity endogenous and exogenous, linearity, carryover, dilution integrity, precision, accuracy, and stability.

The LLOQ was determined through six replicates and calculated relative standard

deviation (RSD) which provided satisfactory precision of ≤ 20%. To assess the selectivity endogenous of the method, plasma samples from six different individuals were tested to source any interference. In the selectivity study, samples spiked with common drugs used in poisoning cases and/or hospital administration protocols were used. The analytes investigated were cocaine and its metabolites, haloperidol, sertraline, citalopram, lidocaine, codeine, tramadol, and dipyrone, each at a concentration of 10 µg/mL. Linearity was assessed over five runs for each of the six concentrations, and heteroscedasticity was evaluated before applying weighting factors to adjust linearity. Carry-over was investigated after the upper calibrator, using three runs of blank samples. Accuracy and precision (within-run and between-run) were evaluated with five replicates of each quality control (low, medium, and high) and LLOQ over three different runs. The percent target for both criteria was set at < 20% RSD for the LLOQ and <15% RSD for the QC. Dilution integrity was evaluated by analyzing blank plasma samples spiked with analytes at concentrations of 5000 ng/mL. The samples were diluted in a 1:10 (v/v) ratio with ultrapure water and analyzed using the obtained regression equation. The acceptance criteria were set at <15% RSD.

Stability was monitored for 60 days under two scenarios, i.e., stored at room temperature and in a refrigerator. Each variable was analyzed in triplicate using quality controls on days 0, 1, 7, 15, 30, and 60.

Authentic samples

The proposed methodology was carried out in 102 samples obtained from suspected cases of antidepressant and antipsychotic poisoning attended by the Toxicological Information Center of Rio Grande do Sul (CIT/RS). In addition, data from patients were collected, such as gender, age, the circumstance of poisoning, exposure site, evolution, poisoning assessment, and seasons when the poisoning occurred. This study was approved by the Ethical Committee for Human Studies of the Federal University of Health Sciences of Porto Alegre, Brazil, and by the Ethical

Committee for Human Studies of the School of Public Health, from the Health State Department of Rio Grande do Sul, Brazil (CAAE 31992920.3.0000.5345).

Results and discussion

Optimization evaluation of the variables

The choice of solvents for the optimization step was based on the interaction with analytes and the compatibility with the plastic material of the 24-well polypropylene plate. The triangular surface obtained from the optimization (**Figure 1**) demonstrates in red the best response being a mixture of methanol and acetonitrile 1:1 (v/v) with a coefficient of determination (r^2) of 0.9962. After obtaining the optimization results for the desorption solvent, was conducted the optimization of the sample and absorptive solvent volumes (**Figure 2)**. The response surface graph demonstrates that the best interaction was using 70 μL of sample and 320 μL of extractor solvent (r^2 = 0.8504).

Method validation

After optimizing the method was fully validated. LLOQ values were determined in 20 ng/mL for venlafaxine, imipramine, and amitriptyline, 50 ng/mL for chlorpromazine, levomepromazine, and promethazine, 60 ng/mL for fluoxetine, norfluoxetine, desvenlafaxine, nortriptyline, and desipramine. For linearity, heteroscedasticity was calculated, and weighting factors were applied to all analytes. The determination coefficients were higher than 0.995, demonstrating good linearity. The values of linear range, weighting factors, and determination coefficients are informed in **Table 1**.

No interference was detected in the retention time of the analytes in the exogenous and endogenous selectivity assay. In addition, carryover did not observe in blank samples after the upper limit of quantification.

The precision essay was represented by the RSD, the within-run precision ranged from 0.12% to 17.7% and the between-run precision ranged from 1.1% to 18.1%. The accuracy was estimated by relative error (RE), which varied from 87.8% to 112.2%. In both assays, the coefficient of variation (CV) obtained was in accordance with recommended by the guideline, which is a CV % of up to 20% for LLOQ and up to 15% for QCs (**Table 2)**.

In dilution integrity, the results were demonstrated acceptably. Precision within-run precision ranged from 0.7% to 12.3% and between-run precision ranged from 6.8% to 14.2%, accuracy varied from 95.7% to 105.1%.

For stability data analysis, the geometric mean of the analytes was compared to the zero time analysis and reported in percentage. The analytes showed satisfactory stability for all analytes for 30 days in both storage assays with CV < 15%. The results were according to those reported in the literature [34-36]. In addition, fluoxetine, amitriptyline, venlafaxine, and desvenlafaxine demonstrated stability for up to 60 days. One of the advantages of dried spot stability is that its storage becomes easier and requires less physical space when compared to the storage of raw material, such as, for example, urine and whole blood [37]. Storage of absorbent paper was further facilitated by using the 24-well plate.

The chromatograms obtained by fast-GC-MS from a plasma sample spiked with the substances in low-quality control concentration were illustrated in **Figure 3**.

Application of the proposed method in samples

In all, 102 samples were analyzed, totalizing 92 samples positive for at least one substance, having a positivity of 90.2% (**Table 3)**.

Of the 102 samples, 47 were above the therapeutic concentration and six samples reached the toxic dose [38], amitriptyline (n = 4), promethazine (n = 1), and fluoxetine (n = 1). The most common antidepressant was amitriptyline and fluoxetine, with 19 positive samples each, while

among the class of antipsychotics, it was chlorpromazine (n = 24).

Positive samples for two analytes represented about 22.8% (n = 21), samples positives for three substances at 3.2% (n = 3) and there was one positive for five substances, which are amitriptyline, desipramine, fluoxetine, and nortriptyline. It is interesting to mention that all samples that tested positive for nortriptyline were positive for amitriptyline, only the samples above fluoxetine therapeutic concentration were positive for norfluoxetine, and one sample was positive for desvenlafaxine and venlafaxine.

It is possible to verify that there were no major differences in terms of gender and the most common age group was teenagers and young adults (**Table 4)**. The place of exposure was mostly at home (n = 63, 61.8%). The principal circumstance was a suicide attempt (42.2%) followed by an individual accident (10.8%). The assessment of intoxication was divided into mild and moderate, but fortunately, the evolution of the cases had a positive prognosis with a cure rate of 79.4%. Additionally, it notices a pattern in the seasons, winter had 50% more cases registered than summer.

Conclusion

This study developed a fast, easy, inexpensive, and efficient extraction using dried plasma spot by fast gas chromatography-mass spectrometry (fast CG-MS). The 24-well plate proved to be a new alternative for the dried plasma spot technique, being easy to use and adding the technique of fast GC-MS, the optimization of time is most advantageous, enabling increased sample productivity. Validation was successful, proving reproducibility and stability on dry paper. The applicability of the method in 102 samples had a positivity of 90.2% showing the importance of its implementation in the routine of clinical emergency toxicology.

Author Contributions

Giovanna Cristiano de Gouveia: Conceptualization, Methodology, Validation, Writing - review & editing; **Bruno Pereira dos Santos**: Validation, Writing - review & editing; **Gabriela Ramos Borges**: Methodology, Validation, Writing - review & editing; **Viviane Cristina Sebben**: Conceptualization, Writing - review & editing; **Sarah Eller**: Conceptualization, Supervision, Writing - review & editing; **Tiago Franco de Oliveira**: Conceptualization, Funding acquisition, Project administration, Writing - review & editing.

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Conflict of interest

There are no financial or other relations that could lead to a conflict of interest.

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Figures Captions

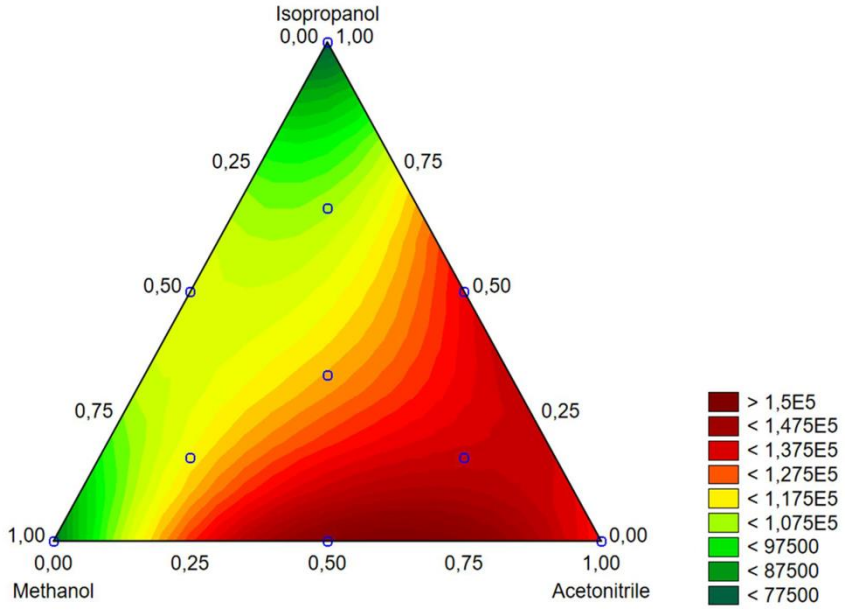

Figure 1. The triangular surface obtained from a simplex-centroid design for the optimization of extraction solvent.

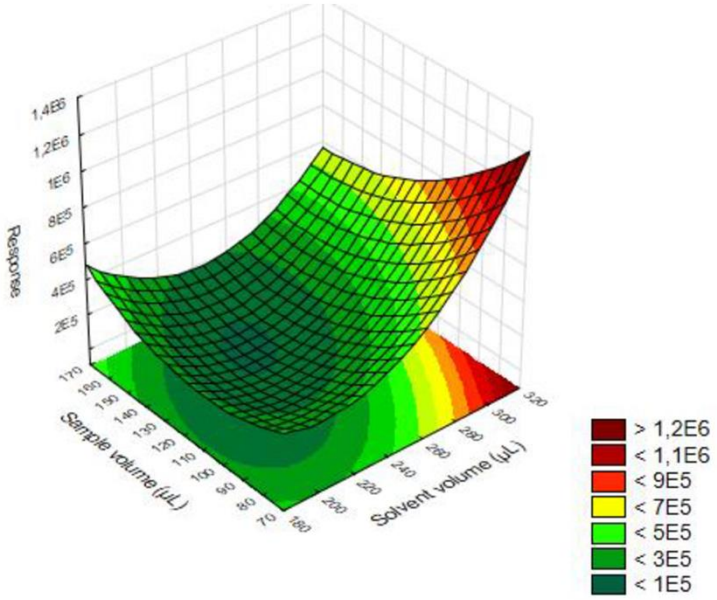

Figure 2. Response surface obtained from Doehlert design for the optimization of the volumes of extraction solvent and sample.

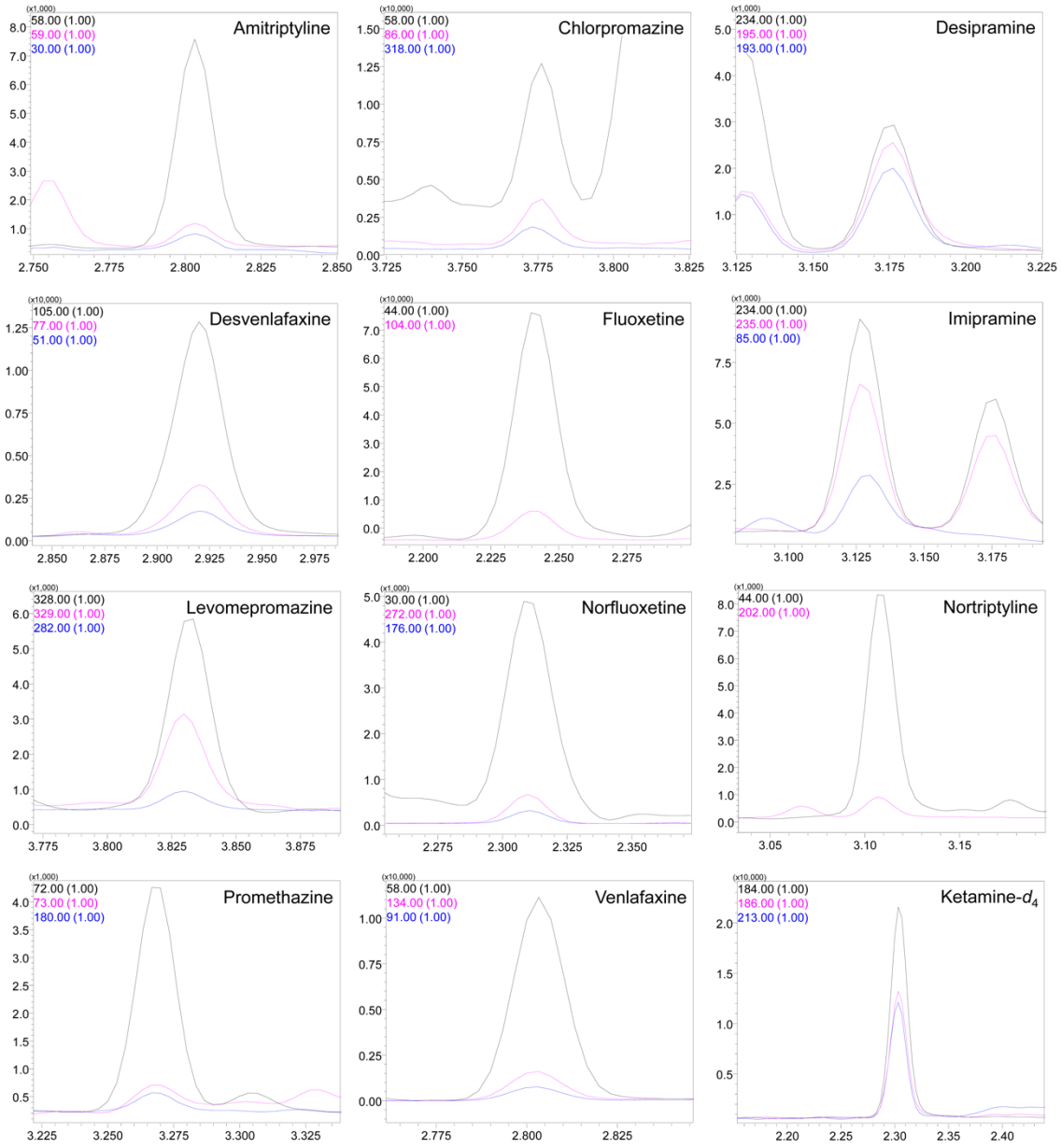

Figure 3. Chromatogram obtained by fast GC-MS from sample spiked at low-quality control concentration analytes and the internal standard (ketamine-d_4) at 150 ng/mL.

Table 1. The values of linear range, weighting factors, determination coefficients, identification and quantification ions and retention time for each analyte.

Analyte	Linearity range (ng/mL)	Weight factor	r^2	Retention Time	Quantification ion (*m/z*)	Identification ions (*m/z*)
Amitriptyline	20-900	1/y½	0.996	2.80	58	59, 30
Chlorpromazine	50-800	1/x½	0.999	3.77	58	86, 318
Desipramine	60-900	$1/x^2$	0.995	3.17	234	195, 193
Desvenlafaxine	60-900	1/x½	0.998	2.92	105	77, 51
Fluoxetine	60-900	$1/x^2$	0.997	2.24	44	104
Imipramine	20-900	1/y½	0.998	3.12	234	235, 85
Levomepromazine	50-800	1/x½	0.995	3.82	328	329, 282
Norfluoxetine	60-900	1/y½	0.998	2.30	30	272, 176
Nortriptyline	60-900	$1/x^2$	0.995	3.10	44	202
Promethazine	50-800	$1/x^2$	0.999	3.26	72	73, 180
Venlafaxine	20-900	$1/x^2$	0.997	2.80	58	134, 91
Ketamine-d_4 (IS)	-	-	-	2.30	184	186, 213

r^2 (determination coefficient).

Table 2. Accuracy and precision data.

Analyte	Quality control (ng/mL)	Precision (RSD%)[1]		Accuracy (%)
		Within-run	Between-run	
Fluoxetine	60	10.8	15.1	98.6
	150	11.5	1.8	98.2
	450	4.7	3.8	100.0
	740	5.3	1.2	95.7
Norfluoxetine	60	16.6	16.4	103.5
	150	0.1	12.1	94.1
	450	6.2	12.4	95.7
	740	7.5	7.0	102.0
Venlafaxine	20	17.7	18.1	98.9
	60	9.6	9.6	104.3
	450	9.1	7.8	95.0
	740	8.0	2.5	101.7
Desvenlafaxine	60	12.3	15.1	107.6
	150	7.4	9.2	97.3
	450	8.3	9.7	98.0
	740	11.9	3.0	102.9
Amitriptyline	20	9.8	17.1	100.3
	60	14.6	9.7	103.2
	450	6.7	1.4	104.2
	740	6.1	4.2	98.8
Nortriptyline	60	13.3	2.9	105.0
	150	11.9	1.3	90.4
	450	9.1	12.0	105.5
	740	9.5	5.5	108.9
Imipramine	20	17.6	4.2	96.3
	60	5.9	6.5	108.4
	450	7.0	1.1	92.1
	740	7.6	2.1	102.6
Desipramine	60	12.4	7.0	91.7
	150	9.1	6.6	96.6
	450	11.6	5.8	98.5
	740	7.5	9.4	102.6
Promethazine	50	13.4	14.7	90.7
	150	6.1	8.6	100.8
	400	6.9	6.5	99.6
	650	7.4	10.2	97.6
Chlorpromazine	50	12.8	15.5	108.0
	150	7.8	9.1	112.2
	400	13.5	13.0	106.0
	650	11.0	12.1	109.5
Levomepromazine	50	13.0	16.5	96.0
	150	9.0	8.6	87.8
	400	10.3	10.4	98.8
	650	7.3	9.3	102.3

[1]RSD (Relative standard deviation).

Table 3. Frequencies of positivity of the analyzed samples and their respective concentration ranges

Analyte	n	%	Concentration Range			Above of therapeutic concentration	%
Amitriptyline	19	18.6	120	-	758	9	47.4
Chlorpromazine	22	21.6	54	-	483	20	90.9
Desipramine	1	1.0	304			1	100.0
Desvenlafaxine	8	7.8	82	-	696	4	50.0
Fluoxetine	19	18.6	70	-	1305	6	31.6
Imipramine	1	1.0	29			0	0.0
Levomepromazine	-	-	-			-	-
Norfluoxetine	6	5.9	95	-	411	-	-
Nortriptyline	13	12.7	67	-	345	3	23.1
Promethazine	6	5.9	63	-	1074	4	66.7
Venlafaxine	1	1.0	70			0	0.0
Negatives	35	34.3	-			-	-
Total	**102***	**-**		**-**		**47**	**46.0**

* There are samples positive for more than one substance, thus, the total of the positives for each substance is more than the total of samples analyzed (n=102)

Table 4. Characterization of intoxication cases in which toxicological analysis was performed.

Variable	n	%	Variable	n	%
Total	**102**	**100**	**Age group**		
			< 1 year	2	2.0
Gender			1 - 4 years	2	2.0
Female	52	51.0	5 - 9 years	8	7.8
Male	45	44.1	10 - 14 years	11	10.8
Unknown	5	4.9	15 - 19	20	19.6
			20 - 29	16	15.7
Exposure site			30 - 39	15	14.7
Home	63	61.8	40 - 49	7	6.9
Public space	2	2.0	50 - 59	9	8.8
External ambient	1	1.0	60 - 69	5	4.9
Unknown	36	35.3	70 - 79	1	1.0
			> 80	1	1.0
Evolution			Unknown	5	4.9
Cure	81	79.4			
Death	1	1.0	**Poisoning assessment**		
Unknown	20	19.6	Mild	33	32.4
			Moderate	36	35.3
Circumstance			Severe	13	12.7
Suicide attempt	43	42.2	Fatal	1	1.0
Individual accident	11	10.8	Differential diagnosis	9	8.8
Abuse	5	4.9	Not toxic	10	9.8
Violence, mistreatment or homicide attempt	2	2.0	**Seasons**		
			Winter	37	36.3
Administration error	1	1.0	Autumn	26	25.5
Unknown	40	39.2	Spring	20	19.6
			Summer	19	18.6

5. CONCLUSÃO

Neste trabalho, foi possível desenvolver uma metodologia de DPS para determinação de amitriptilina, nortriptilina, fluoxetina, norfuoxetina, imipramina, desipramina, venlafaxina, desvenlafaxina, clorpromazina, prometazina e levomepromazina em plasma utilizando *fast*-GC-MS. A validação do método foi realizada seguindo os critérios estabelecidos pelo guia EMEA 2011-*Guideline on bioanalytical method validation*, provando reprodutibilidade, confiabilidade e estabilidade no papel seco. Após a completa validação o método foi aplicado com sucesso a 102 amostras de emergência toxicológica do Centro de Informação Toxicológica do Rio Grande do Sul, obtendo-se uma positividade de 90,2 % nos casos analisados. A técnica de bioamostragem se mostrou fácil, barata e eficiente, principalmente associada a placa de 24 poços, aumentando a produtividade de análises e facilidade para armazenar o DPS, além de se utilizar volumes baixos de amostra (70 µL) e solvente (320 µL). A utilização do *fast*-GC-MS trouxe a vantagem de rapidez, provando ser uma boa alternativa para reduzir o tempo de análise. Essas vantagens são importantes no contexto da toxicologia clínica emergencial, promovendo resultados rápidos visando um diagnóstico correto e prognóstico positivo para o paciente.

6. ANEXOS

6.1 ANEXO I – Parecer do Comitê de Ética em Pesquisa

UNIVERSIDADE FEDERAL DE CIÊNCIAS DA SAÚDE DE PORTO ALEGRE

PARECER CONSUBSTANCIADO DO CEP

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: Desenvolvimento de metodologias por espectrometria de massas para toxicologia de emergência

Pesquisador: TIAGO FRANCO DE OLIVEIRA

Área Temática:

Versão: 3

CAAE: 31992920.3.0000.5345

Instituição Proponente: Universidade Federal de Ciências da Saúde de Porto Alegre

Patrocinador Principal: Financiamento Próprio

DADOS DO PARECER

Número do Parecer: 4.328.757

Apresentação do Projeto:

Resumo:

Ao longo dos últimos anos, os casos de intoxicações têm aumentado no Brasil e no mundo, estando estes principalmente relacionados com exposição a fármacos e drogas de abuso. Assim, é de extrema importância que seja realizada a determinação dos toxicantes para que se proceda o melhor tratamento e manejo correto do paciente. Na toxicologia clínica, a espectrometria de massas (MS) é a metodologia de escolha para a determinação de xenobióticos em matrizes biológicas, já que a partir dela é possível gerar resultados exatos e precisos. A MS é uma técnica analítica de identificação molecular baseada na análise da relação massa carga (m/z) do composto, tendo como resultado a produção de um espectro de massas. Diante do exposto, esta proposta tem como objetivo desenvolver métodos multi-analito para a determinação de fármacos, drogas de abuso e seus produtos de biotransformação por utilizando a espectrometria de massas como técnica analítica, de forma a gerar resultados confiáveis, rápidos e exatos, podendo assim auxiliar no diagnóstico das intoxicações. Os métodos a serem desenvolvidos serão aplicados em amostras provenientes de casos atendidos pelo Centro de Informação Toxicológica do Rio Grande do Sul (CIT/RS). A partir da validação analítica, será possível disponibilizar as técnicas desenvolvidas aos serviços de avaliação e controle de intoxicações do Brasil. Ademais, os resultados obtidos através das análises das amostras contribuirão como uma base de dados para o desenvolvimento de novas políticas de toxicovigilância no Rio Grande do Sul e Brasil.

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Bairro: Sarmento **CEP:** 90.050-170
UF: RS **Município:** PORTO ALEGRE
Telefone: (51)3303-8804 **E-mail:** cep@ufcspa.edu.br

Objetivo da Pesquisa:

Objetivo Primário:

Desenvolver metodologias analíticas para diagnóstico laboratorial de intoxicações utilizando a espectrometria de massas. A partir disso, aplicar os métodos em amostras oriundas de casos atendidos pelo Centro de Informação Toxicológica do Rio Grande do Sul.

Objetivo Secundário:

Desenvolver de métodos para determinação de fármacos e drogas de abuso em sangue e urina por espectrometria de massas; Desenvolver e validar métodos cromatográficos e espectrométricos; Proceder a síntese de novos preparos de amostras; Aplicar o método em amostras de sangue
e urina de casos reais cedidas pelo Centro de Informação Toxicológica do Rio Grande do Sul (CIT/RS).

Avaliação dos Riscos e Benefícios:

Riscos:

O risco à sua saúde dos participantes voluntários da pesquisa será mínimo, uma vez que a coleta de 3 mL de sangue periférico e 10 mL de urina já foi realizada durante o atendimento do caso pelo CIT/RS, na unidade de saúde onde o paciente fora atendido para envio ao laboratório do CIT/RS. A coleta de sangue pode ter alguns efeitos indesejados, como hematomas e dor no local da picada, assim como a coleta de urina pode gerar
constrangimento ao paciente. No entanto, pelo projeto usar amostras de descarte, não será solicitada/realizada nenhuma coleta com pacientes. Além disso, um possível risco será a quebra acidental o sigilo, por meio da utilização dos dados de atendimentos. Para evitar isso, serão tomadas medidas para que essa quebra não venha a ocorrer de forma alguma, tais como identificação das amostras e casos com código numérico próprio,diferente do já existente na rotina do CIT/RS. Nenhum dos pesquisadores terá acesso ao nome dos pacientes ou quaisquer outra informação não citada no projeto. Todas informações serão guardadas em arquivos do tipo Excel ou Word com login e senha, tendo acesso a estes arquivos somente os pesquisadores integrantes deste projeto de pesquisa. Além disso, uma lista mestra com os códigos numéricos das amostras e relação com a numeração própria do CIT/RS será feita e mantida exclusivamente em guarda com a responsável e chefe de divisão do CIT/RS, MSc. Viviane Cristina Sebben.

Benefícios:

Serão desenvolvidas metodologias analíticas para o diagnóstico de intoxicações por fármacos e drogas de abuso. Além disso, serão implementados métodos na rotina do Núcleo de Análise Laboratorial do Centro de Informação Toxicológica do Rio Grande do Sul, assim como poderão ser disponibilizados por outros centros de informação e assistência toxicológica. Por fim, os resultados obtidos através das análises das amostras contribuirão como uma base de dados para o desenvolvimento de novas políticas de toxicovigilância no Rio Grande do Sul e Brasil.

Comentários e Considerações sobre a Pesquisa:

A pesquisa possui relevância acadêmica e a parceria UFCSPA/CIT, otimizando recursos financeiros e colaborando para formação de recursos humanos atentos às necessidades que rião atender Indivíduos atendidos pelo Centro de Informação Toxicológica do Rio Grande do Sul (CIT/RS) realizaram o uso de pelo menos uma das substâncias (fármacos e drogas de abuso) elencadas no estudo, tendo assim oportunidade de utilização de amostras, antes descartadas, para a confirmações dos resultados da metodologia analítica instrumental em desenvolviento nessa pesquisa.

Considerações sobre os Termos de apresentação obrigatória:

Todos os termos de apresentação obrigatória apresentados foram revisados e encontram-se adequados. A solicitação de dispensa do TCLE foi devidamente justificada através da carta-resposta, onde foi dado todo o esclarecimento sobre as amostras, que já chegam coletadas ao CIT, não havendo possibilidade de contato com o participante; contudo há risco de quebra de sigilo e por isso a equipe adotará código numérico próprio, diferente do já existente na rotina do CIT/RS.Nenhum dos pesquisadores terá acesso ao nome dos pacientes ou quaisquer outra informação não citada no projeto. Todas informações serão guardadas em arquivos do tipo Excel ou Word com login e senha, tendo acesso a estes arquivos somente os pesquisadores integrantes deste projeto de pesquisa. Além disso, uma lista mestra com os códigos numéricos das amostras e relação com a numeração própria do CIT/RS será feita e mantida exclusivamente em guarda com a responsável e chefe de divisão do CIT/RS, MSc. Viviane Cristina Sebben. As amostras durante 3 meses são guardadas como contra-prova. Nenhuma amostra será cedida pelo CIT/RS antes deste período. Após este período, todas amostras são consideradas de descarte, mas não são necessariamente descartadas. O NAL-CIT/RS mantém parte das amostras para uso interno, como uso em controle interno de qualidade, uso em validações de metodologias analíticas, síntese de controles positivos, dentre outros, possibilitando o uso de amostras de casos de 2018 e 2019. Com a quantidade de amostras já guardadas e a média de análises de emergência realizadas por ano, espera-se atingir no mínimo o número de 400 amostras, tendo assim garantia de se alcançar

esse valor.

Recomendações:

Não se aplica.

Conclusões ou Pendências e Lista de Inadequações:

Não se aplica.

Considerações Finais a critério do CEP:

De acordo com o parecer do Relator.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PB_INFORMAÇÕES_BÁSICAS_DO_PROJETO_1521315.pdf	15/09/2020 18:31:38		Aceito
Projeto Detalhado / Brochura Investigador	Projeto_corrigido.pdf	15/09/2020 18:31:10	TIAGO FRANCO DE OLIVEIRA	Aceito
Folha de Rosto	folhaDeRosto.pdf	15/09/2020 18:30:40	TIAGO FRANCO DE OLIVEIRA	Aceito
Outros	Carta_resposta.pdf	15/09/2020 18:30:09	TIAGO FRANCO DE OLIVEIRA	Aceito
Outros	TCUD_corrigido.pdf	15/09/2020 18:29:35	TIAGO FRANCO DE OLIVEIRA	Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	Dispensa_de_TCLE_corrigido.pdf	15/09/2020 18:29:08	TIAGO FRANCO DE OLIVEIRA	Aceito
Outros	Termo_de_anuencia_Central_Analitica.pdf	13/07/2020 10:58:41	BRUNO PEREIRA DOS SANTOS	Aceito
Outros	Declaracao_de_evidente_interesse_publico_e_SUS.pdf	29/04/2020 17:24:59	BRUNO PEREIRA DOS SANTOS	Aceito
Outros	Termo_de_autorizacao_institucional.pdf	29/04/2020 17:23:31	BRUNO PEREIRA DOS SANTOS	Aceito
Outros	Termo_de_compromisso_entrega_relatorio_semestral_e_final.pdf	29/04/2020 17:22:41	BRUNO PEREIRA DOS SANTOS	Aceito
Outros	Termo_de_anuencia_do_local.pdf	29/04/2020 17:19:14	BRUNO PEREIRA DOS SANTOS	Aceito

Situação do Parecer:
Aprovado

Necessita Apreciação da CONEP:
Não

PORTO ALEGRE, 08 de Outubro de 2020

Assinado por:
Fernanda Bordignon Nunes
(Coordenador(a))

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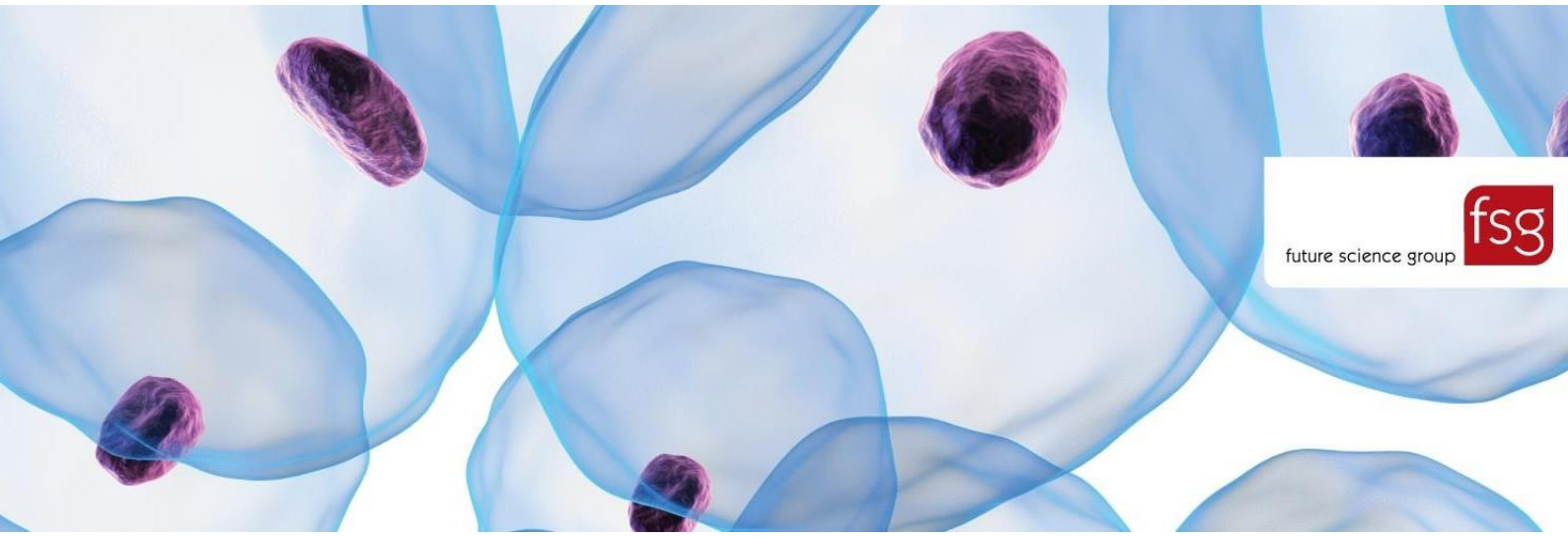
6.2 ANEXO II – Normas de Publicação da Revista



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This document outlines how to prepare articles for submission. We recommend you read these guidelines in full before submitting your article. A pre-submission enquiry to the Journal Editor is also strongly encouraged before submission.

These guidelines apply to the following journals:

- *Bioanalysis*
- *Future Drug Discovery*
- *Future Medicinal Chemistry*
- *Future Science OA*
- *International Journal of Pharmacokinetics*
- *Pharmaceutical Patent Analyst*
- *Therapeutic Delivery*

Separate Author Guidelines for the journal *BioTechniques* can be found on the website here.

Table of Contents

Journal information

Aims and scope information can be found on the individual journal webpages linked below, along with information regarding Editorial Board members and indexing.

Journal	Medline-indexed	Impact Factor (2021)	Open Access*	Accelerated publication option?
Bioanalysis	Yes	2.695	Optional	Yes
Future Drug Discovery			**Full open access**	Yes
Future Medicinal Chemistry	Yes	4.767	Optional	Yes
Future Science OA	PMC-indexed		**Full open access**	Yes
International Journal of Pharmacokinetics			**Full open access**	Yes
Pharmaceutical Patent Analyst	Yes		Optional	Yes
Therapeutic Delivery	Yes		Optional	Yes

*Optional: This journal is a subscription journal; however, authors can opt for Open Access publication if they wish (or it is a requirement of their funding); Full open access: This journal is a fully gold open access title.
PMC: PubMed Central.

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More information and a proposal form can be found on our website here: https://www.future-science.com/special-issues.

Article types considered

	Bioanalysis	*Future Drug Discovery*	*Future Medicinal Chemistry*	*Future Science OA*	*Int. J. Pharmacokinetics*	*Pharmaceutical Patent Analyst*	*Therapeutic Delivery*
Review-based articles:							
Review	✓	✓	✓	✓	✓	✓	✓
Systematic Review	×	✓	✓	✓	✓	×	✓
Perspective	✓	✓	✓	✓	✓	✓	✓
Special Report	✓	✓	✓	✓	✓	✓	✓
Regulatory	✓	×	×	×	×	×	✓
Technology Evaluation	✓	×	×	×	×	×	×
Patent Review	×	×	✓	×	×	✓	×
Patenting Trends	×	×	×	×	×	✓	×
Case History	×	✓	×	×	×	×	×
Original research articles:							
Research Article	✓	✓	✓	✓	✓	×	✓
Preliminary Communication	✓	✓	✓	✓	✓	×	✓
Short Communication	✓	✓	✓	✓	✓	×	✓
Study Protocol/Trial Design	×	✓	×	✓	×	×	×
Trial Results	×	✓	×	✓	×	×	×
Bioanalytical Challenge	✓	×	×	×	×	×	×
Methodology	✓	✓	✓	✓	✓	×	✓
Meta-Analysis	✓	✓	✓	✓	✓	×	✓
Case Study/Report/Series	×	✓	×	✓	×	×	×
Added-value articles:							
White Paper	✓	✓	✓	✓	✓	✓	✓
Editorial	✓	✓	✓	✓	✓	✓	✓
Commentary	✓	✓	✓	✓	✓	✓	✓
Conference Report	✓	✓	✓	✓	✓	✓	✓
Company/Institutional Profile	×	✓	×	✓	✓	×	×
Patent Spotlight	×	×	×	×	×	✓	×
Hot Topic	×	×	×	×	×	✓	×
Letter to the Editor	✓	✓	✓	✓	✓	✓	✓

At-a-glance article formatting checklist

	Word count	Abstract	Keywords	Future Perspective	Exec. Summary	Summary Points	Ref. limit	Figures/Tables permitted?
Review-based articles:								
Review	4000–8000	✓	✓	✓	✓	×	~80	✓
Systematic Review	4000–8000	✓ (structured)	✓	×	×	✓	As appropriate	✓
Perspective	4000–8000	✓	✓	✓	✓	×	~80	✓
Special Report	3000–5000	✓	✓	✓	✓	×	~50	✓
Regulatory	2000–3000	✓	✓	×	×	✓	~50	✓
Technology Evaluation	2000–3000	✓	✓	✓	✓	×	~50	✓
Patent Review	4000–10,000	✓	✓	✓	✓	×	~80	✓
Patenting Trends	4000–10,000	✓	✓	✓	✓	×	~150	✓
Case History	8000	✓	✓	✓	✓	×	~80	✓
Original research articles:								
Research Article	5000–8000	✓ (structured)	✓	×	×	✓	~80	✓
Preliminary/Short Communication	2000–4000	✓ (structured)	✓	×	×	✓	~50	✓
Study Protocol/Trial Design	5000	✓ (structured)	✓	×	×	✓	~50	✓
Trial Results	5000	✓ (structured)	✓	×	×	✓	~50	✓
Bioanalytical Challenge	2000–3000	✓ (structured)	✓	×	×	✓	~50	
Methodology	3000–5000	✓	✓	×	×	✓	~50	✓
Meta-Analysis	4000–8000	✓ (structured)	✓	×	×	✓	~80	✓
Case Study/Report/Series	1500–3000	✓	✓	×	×	✓	~50	✓
Added-value articles:								
White Paper	1500–3000	✓	✓	×	×	✓	~50	✓
Editorial	1500	×	✓	×	×	×	20	×
Commentary	1500–3000	×	✓	×	×	×	20	×
Conference Report	1500	✓	✓	×	×	×	20	×
Company/Institutional Profile	2000	✓	✓	×	×	✓	20	One of each max.
Patent Spotlight	1500–3000	✓	✓	✓	✓	×	~50	✓
Hot Topic	1000–1500	×	✓	×	×	×	20	×

Letter to the Editor	1500	×	✓	×	×	×	20	×

[1]All Future Science Group journals also encourage the inclusion of a **plain language abstract** and a **tweetable abstract** where appropriate. Further information on these sections are included below.

[2]For article types where Figures & Tables are permitted, unless specified differently above, we have no strict limit on the number included (we prefer these to be included as needed to represent the data appropriately); however, in some cases where there is a high number of figures/tables (generally >8 Figures/Tables combined) the Editor will recommend that some are included as online-only supplementary materials.

[3]For certain journals, including *Bioanalysis*, this limit can be exceeded. Please consult the Journal Editor if your article does not fit within these limits.

Article templates

In the first instance, Future Science Group journals are happy to consider articles that have not been fully formatted to the journal style. However, should you wish to format your article in advance of submission, the following templates are designed to help you do so, and are available on our website: https://www.future-science.com/authorguide/preparingyourarticle

- **Title page template**
- **Article body template**

Please note, articles will need to be formatted to the journal style prior to final acceptance.

Search engine optimization

Why are search engines important?

One of the most common ways for readers to find an article is using a search engine, such as Google, Google Scholar or Bing. Therefore, it is important to write your article with a few points in mind, to help interested readers find your work.

How can I help my article be discovered?

- Include key phrases that represent your research in the abstract. Think about what you might search for when looking for articles yourself and include this.
- Make sure the most important/relevant key phrase is also in the article title whilst ensuring the content has a natural flow.
- Choose appropriate keywords that reflect the content of your work – where different words are commonly used to describe the same thing (e.g., a full term and an abbreviation), include both.
- Aim to be as concise as possible in the abstract (within the journals' word limit of 120 words or fewer).
- Provide a tweetable abstract when you submit your article (for more information see Article sections).

Article types

Future Science Group publishes a range of article types, descriptions of which are outlined below. Authors are encouraged to consult the 'at-a-glance formatting checklist' for details on word counts and other formatting requirements.

The information below gives an overview of the requirements for each article type published by Future Science Group. However, authors should consult the International Committee of Medical Journal Editors (ICMJE) "*Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals*" (https://www.icmje.org/recommendations/), in particular the section on "*Preparing a Manuscript for Submission to a Medical Journal*" prior to submitting to a Future Science Group journal, for more detailed information.

Review-based articles

Review

Reviews aim to highlight recent significant advances in research, ongoing challenges and unmet needs; authors should be concise and critical in their appraisal of the subject matter and strive for clarity. The focus should be on key, defining developments rather than providing a comprehensive literature survey. Reviews should provide balanced coverage of the field and not focus predominantly on the author's own research. Authors are encouraged to include their own perspective on current trends and future directions, particularly in the 'Future perspective' section. Review articles undergo external peer review.

Future Science Group journals consider both Narrative and Systematic Reviews. For more information on Systematic Reviews, see below.

Word limit: 4000–8000 words (excluding abstract, executive summary, references and figure/table legends)
Reference limit: ~80
Required sections (for a more detailed description see Article sections):

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Body of article
- Future perspective
- Executive summary
- References
- Reference annotations
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Figures/Tables: should be submitted as separate files (see guidelines below)

Systematic Review

Systematic Reviews should systematically gather, appraise and synthesize evidence around a specific question. Systematic Review articles undergo external peer review.

Authors of Systematic Reviews **must** provide a supporting Cover Letter on submission briefly detailing:

- Relevance to the journal's audience
- Objectives of the review (including the question the review intends to address)
- How the review advances understanding of the field

Systematic Reviews should be conducted following the recommendations of PRISMA (http://www.prisma-statement.org/). A summary of required sections in provided below, but further information on these should be taken from the PRISMA checklist. In addition, a completed PRISMA checklist should be provided as Supplementary Materials on submission of the article.

Word limit: 4000–8000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: As appropriate based on literature search
Required sections (for a more detailed description see Article sections):

- Title (maximum 120 characters) – should identify the work as a Systematic Review
- Author(s) names & affiliations
- Structured abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Introduction: including review rationale and the question being addressed (with reference to participants, interventions, comparisons, outcomes and study design [PICOS])
- Methods
- Results
- Discussion
- Conclusions
- Summary points
- References
- Reference annotations
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Supplementary materials: A completed PRISMA checklist (http://www.prisma-statement.org/) should be included for publication alongside the Systematic Review
- Figures/Tables: should be submitted as separate files (see guidelines below)

Perspective

Perspectives have the same basic structure and length as Review articles; however, they should be more speculative and forward-looking, even visionary. They offer the author the opportunity to present criticism, address controversy or provide a personal angle on a significant issue. Authors of Perspectives are encouraged to be opinionated, with all positions concisely and clearly argued and referenced. Perspective articles undergo external peer review; however, reviewers will be briefed to review these articles for quality and relevance of argument only. They will not necessarily be expected to agree with the author's position.

Word limit: 4000–8000 words (excluding abstract, executive summary, references and figure/table legends)
Reference limit: ~80
Required sections: As for Review articles (see above)

Special Report

Special Reports are short narrative review-style articles that highlight a particular niche area, be it a specific emerging field, novel hypotheses or method. Articles are categorized as Special Reports at the discretion of the Editorial team. Special Report articles undergo external peer review.

Word limit: 3000–5000 words (excluding abstract, executive summary, references and figure/table legends)
Reference limit: ~50
Required sections: As for Review articles (see above)

Regulatory

Regulatory articles highlight important regulatory issues, and provide background information, issues and advice for implementing and following the regulations.

Word limit: 2000–3000 words (excluding keywords, abstract and references).
Reference limit: ~50
Required sections: As for Review articles (see above)

Technology Evaluation

The aim of a Technology Evaluation is to provide the objective assessments of the technical performance of analytical instruments in development or clinical use to help inform bioanalytical scientists. They should provide an overview of a new experimental or computational technological device or procedure. The significance and potential implications of the developed technology or application must be explicit.

Word limit: 2000–3000 words (excluding abstract, executive summary, references and figure/table legends)
Reference limit: ~50
Required sections: As for Review articles (see above)

Patent Review

Patent Reviews should provide an objective and concise appraisal of a selection of patents in a chosen area. Discussions should be placed within the context of the relevant R&D landscape. Authors are expected to offer a commentary on the significance, potential and essential content of the patents under discussion. The patents reviewed should be from a variety of companies/assignees, and should be timely (i.e. ideally granted within the past 1-4 years). The majority of the references cited in the article should be taken from the patent literature. Patent Reviews should provide balanced coverage of the field and not focus predominantly on the author's own research. Where controversial or novel ideas are presented, opposing viewpoints should be briefly mentioned and appraised. Authors are encouraged to include their own perspective on current trends and future directions.

Word limit: 4000–10,000 words (excluding abstract, keywords, executive summary and references).
Reference limit: ~80
Required sections: As for Review articles (see above)

Patenting Trends

Patenting Trends provide analyses of patenting activity within selected parameters; e.g., geographical regions, sectors, compound/drug classes, therapeutic areas. The intention is to survey the broader patent landscape rather than the scientific content of individual patents.

Word limit: 4000–10,000 words (excluding abstract, keywords, executive summary and references).
Reference limit: ~150
Required sections: As for Review articles (see above)

Case History

Case History articles take readers through the history of a drug, from conception to current status. They also speculate on what's next for the drug, in terms of potential for new indications, and discuss what went well/did not go well, competition, controversies and so on.

Word limit: 8000 words (excluding abstract, keywords, executive summary and references).
Reference limit: ~80
Required sections: As for Review articles (see above)

Drug/Device/Vaccine Evaluations, Clinical Trial Protocols & Clinical Trial Evaluations

Separate author guidelines for the submission of these article types are available. Please contact the Drug Evaluations Commissioning Editor to request a copy.

Original research articles

General information for original articles

Authors are advised to consult the **Methods Reporting Checklist for Authors**, available here. In addition, the **EQUATOR Network** provides contains a comprehensive searchable database of reporting guidelines and also links to other resources relevant to research reporting.

Cover letter: Authors **must** provide a supporting Cover Letter on submission briefly detailing:

- Relevance to the journal's audience
- Where the novelty in the study lies
- How the study advances understanding of the field
- Direct and potential implications of the findings

Please note: All journals will consider studies presenting positive, negative or inconclusive data.

Peer review: All types of original article undergo external peer review.

Experimental details & data: Where a novel experimental procedure has been employed full details must be provided, such that a skilled scientist would be able to reproduce the results presented. The synthesis of all new compounds must be described in detail. Details of routine or previously reported experimental procedures should be provided via references only. Experimental procedures and/or data running to more than two Word document pages should be placed in a supplementary file.

Reporting of sex & gender information: Authors are encouraged to consult the SAGER Guidelines to ensure the accurate reporting of sex and gender information in study design, data analysis, results and interpretations of findings.

Data & materials sharing: From 1st July 2018, the ICMJE requires that all manuscripts that report the results of clinical trials must contain a data sharing statement, as described on their website here: http://www.icmje.org/recommendations/browse/publishing-and-editorial-issues/clinical-trial-registration.html. Please see the Author Disclosure Form (available on our website here) for further information. For other types of original research, authors should be able to provide additional relevant original data underpinning their research, if requested by the Editor or reviewers.

Data deposition: Authors are encouraged to make underlying data/biological materials available upon reasonable request, where this is possible. We also encourage the deposition of data/materials to a discipline-specific, community-recognized repository where one exists, or a generalist repository if no suitable specific resource is available, in order to aid in the future replication of methods or the completion of follow-on studies. Repositories can be found via sites such as re3data.org. Where data have been deposited in a public repository, authors should state at the end of the abstract the dataset name, repository name and number.

Protocols: Where relevant, we recommend that authors submit concise, reproducible protocols to protocols.io, and include the link as a reference in the article. An example of such a protocol can be found here: http://bit.ly/2rPyrkL.

Clinical trial reporting: For authors presenting the results of clinical trials, the guidelines recommended by CONSORT and GPP3 should be followed. In addition, where available the clinical trial registration number should be included at the end of the abstract, and on the first mention of the trial in the main body of text. Unregistered clinical trials should be declared as such, and the reason for nonregistration should be provided. Mention of other trials should also include the relevant registration number, where available.

Secondary outcomes, exploratory analyses, and *post hoc* analyses should be clearly identified as such; these may be included in the primary publication or published separately, in which case they should clearly reference the primary publication and should not be published before it.

Diagnostic accuracy studies: Where a diagnostic accuracy study has been carried out, authors should follow the recommendations of STARD (http://www.equator-network.org/reporting-guidelines/stard/).

Observational studies: Where observational research has been carried out, authors should follow the recommendations of STROBE (https://www.strobe-statement.org/).

Required sections (for a more detailed description of these sections see Article sections):

- **Title** (maximum 120 characters)
- **Author(s) names & affiliations**
- **Structured abstract** (maximum 120 words)
- **Plain language summary** (optional; maximum 250 words)
- **Tweetable abstract** (optional; ~200 characters)
- **Keywords** (5–10)
- **Introduction**
  - Should only cite directly pertinent references
  - Should not include data of conclusions from the work being reported
- **Patients & methods/Materials & methods/Experimental**
  - Where an organization was paid or otherwise contracted to help conduct the research (e.g., data collection and management), this should be detailed
  - Should include information indicating that the research was approved or exempted from the need for review by the responsible review committee (institutional or national). Where no formal ethics committee is available, a statement indicating that the research was conducted according to the principles of the Declaration of Helsinki should be included
  - Information on the selection and description of participants should define how authors measured race or ethnicity and justify their relevance
  - Should include information on where the reagents were sourced, where relevant
- **Results**
  - Numeric results should be given not only as derivatives (e.g., percentages) but also as the absolute numbers from which the derivatives were calculated
  - Statistical significance of results should be specified, if any
- **Discussion**
  - Authors should distinguish between clinical and statistical significance, and avoid making statements on economic benefits and costs unless the manuscript includes the appropriate economic data and analyses
  - Authors should avoid claiming priority or alluding to work that has not been completed
- **Conclusions**
- **Summary points**
- **References**
- **Reference annotations**
- **Author contributions:** brief summary of the contribution of each individual meeting the criteria to be listed as an author on the manuscript
- **Acknowledgements:** author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors

- **Disclosures:** to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- **Ethical conduct of research statement**
- **Data sharing statement** (for studies reporting the original results of a clinical trial or the secondary analysis of clinical trial data only)
- **Figures/Tables:** There are no strict limits on the number allowed (we prefer these to be included as needed to represent the data appropriately); however, in some cases where there is a high number of figures/tables (generally >8 Figures/Tables combined) the Editor will recommend that some are included as online-only supplementary materials
    - Should be submitted as separate files (see guidelines below)

Several types of original article are accepted, as detailed below:

Research Article

Research Articles should present novel work that makes a significant impact within the scope of the journal, and which represents an important advancement in knowledge or understanding. Routine or incremental work is not suitable for full Research Articles. Research should be reported succinctly; the inclusion of detailed background discussion is to be avoided. Supporting data or further experimental details can be submitted as Supplementary Information. Articles submitted as Research Articles may be recommended to be considered as a Preliminary/Short Communication at the discretion of the Journal Editor.

Word limit: 5000–8000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: ~80

Preliminary Communication

A type of brief report, Preliminary Communication articles are intended for short reports of studies that present promising improvements or developments on existing areas of research. The significance and potential implications of the developments must be explicit.

Word limit: 2000–4000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: ~50

Short Communication

A type of brief report, Short Communication articles are short articles that build on a previously published study, document partial research results from an ongoing study, or discuss results from studies limited in scope.

Word limit: 2000–4000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: ~50

Study Protocol and Trial Design Articles

Study protocols or trial design articles can cover proposed or ongoing research. For protocols of registered trials, the last line of the abstract should include the trial registry and the unique identifying number.

Word limit: 5000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: ~50

Trial Results

Trial result articles describe and discuss either positive or negative results from trials. For negative studies, authors should highlight and discuss any Exceptional Responders. For protocols of registered trials, the last line of the abstract should include the trial registry and the unique identifying number.

Word limit: 5000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: ~50

Bioanalytical Challenge

Bioanalytical Challenge articles are a type of original research article that provide insights and solutions to specific laboratory issues. The challenge should be presented clearly in the introduction. The articles are focused on the practical laboratory aspects, and authors are encouraged to describe failures as well as successes.

Word limit: 2000–3000 words (excluding abstract, keywords, executive summary and references).
Reference limit: ~50
Required sections: (for a more detailed description see Article sections)

- Title
- Author(s) names & affiliations
- Structured abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords
- Introduction
    - Should only cite directly pertinent references
    - Should not include data of conclusions from the work being reported
- Materials & methods/Experimental
    - Where an organization was paid or otherwise contracted to help conduct the research (e.g., data collection and management), this should be detailed
    - Should include information indicating that the research was approved or exempted from the need for review by the responsible review committee (institutional or national). Where no formal ethics committee is available, a statement indicating that the research was conducted according to the principles of the Declaration of Helsinki should be included
    - Information on the selection and description of participants should define how authors measured race or ethnicity and justify their relevance
- Results & Discussion
    - Numeric results should be given not only as derivatives (e.g. percentages) but also as the absolute numbers from which the derivatives were calculated
    - Statistical significance of results should be specified, if any
    - Authors should avoid claiming priority or alluding to work that has not been completed
- Conclusions and practical tips
- Future perspective
- Summary Points
- References
- Reference annotations
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Ethical conduct of research
- Figures/Tables: should be submitted as separate files (see guidelines below)

Methodology

Methodology articles should provide an overview of a novel study method, test or procedure. The method described may be either completely novel or may offer a demonstrable improvement on an existing method. The significance and potential implications of the developments must be explicit.

Word limit: 3000–5000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: ~50
Required sections: Article sections may vary depending on the type of method being presented. At a minimum, the article should include (for a more detailed description see Article sections):

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Introduction
- Methods
    - This section will vary depending on the type of work being presented; however, in general, this section will include a general description of the method and its validation. Enough detail should be provided for the independent recreation of the method. Appropriate subheadings should be included.
- Discussion and conclusions
- Summary points
- References
- Reference annotations
- Author contributions: brief summary of the contribution of each individual meeting the criteria to be listed as an author on the manuscript
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Ethical conduct of research statement
- Figures/Tables: should be submitted as separate files (see guidelines below)

Meta-Analysis

Meta-Analyses use statistical methods to combine data from multiple, systematically selected studies. Meta-Analyses undergo external peer review.

Authors of Meta-Analyses **must** provide a supporting Cover Letter on submission briefly detailing:

- Relevance to the journal's audience
- Objectives of the analysis (including the question the analysis intends to address)
- How the analysis advances understanding of the field

Meta-Analyses should be conducted following the recommendations of PRISMA (https://prisma-statement.org/). A summary of required sections is provided below, but further information on these should be taken from the PRISMA checklist. **In addition, a completed PRISMA checklist should be provided as Supplementary Materials on submission of the article.**

Word limit: 4000–8000 words (excluding abstract, summary points, references and figure/table legends)
Reference limit: As appropriate based on literature search
Required sections (for a more detailed description see Article sections):

- Title (maximum 120 characters) – should identify the work as a Meta-Analysis
- Author(s) names & affiliations
- Structured abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Introduction: including review rationale and the question being addressed (with reference to participants, interventions, comparisons, outcomes and study design [PICOS])
- Methods
- Results
- Discussion
- Conclusions
- Summary points
- References
- Reference annotations
- Author contributions: brief summary of the contribution of each individual meeting the criteria to be listed as an author on the manuscript
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Supplementary materials: A completed PRISMA checklist (https://prisma-statement.org/) should be included for publication alongside the Systematic Review
- Figures/Tables: should be submitted as separate files (see guidelines below)

Case Study/Case Report/Case Series

Case Studies/Reports/Series present a notable medical case or series of related cases of interest and aim to further the reader's understanding of the issues relating to such situations. Case study/report/series articles undergo external peer review.

Word limit: 1500–3000 words
Reference limit: ~50
Required sections (for a more detailed description see Article sections):

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Body of the article. A suggested structure could be:
    - Presentation of case – setting and patient details/history
    - Initial diagnosis/assessment
    - Treatment/management
    - Outcome and implications
- Discussion/conclusion
- Summary points
- References
- Reference annotations
- Author contributions: brief summary of the contribution of each individual meeting the criteria to be listed as an author on the manuscript
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Ethical conduct of research statement
- Figures/Tables: should be submitted as separate files (see guidelines below)

Plain Language Summary of Publication articles

Separate author guidelines for the submission of these article types are available. Please contact us to request a copy.

For more information on plain language options in Future Science Group journals, please see below.

Added-value articles

All Future Science Group journals consider a range of shorter, added-value article types. The purpose of these articles is to provide readers with commentary, opinion and correspondence on topics of relevance to the journal scope. These articles are often invited by the Editor; however unsolicited article proposals are also welcome.

White Paper

White Papers are authoritative reports that bring together the opinions and current thinking of leading stakeholders or recognized experts. They may offer recommendations, outline proposals and aim to set out current 'consensuses' related to an issue. The issue under discussion should be of immediate importance to the advancement of the field. White Papers may undergo external peer review and will be accepted at the discretion of the Editor.

Word limit: 1500–3000 words (**Please note:** For certain journals, including *Bioanalysis*, this limit can be exceeded. Please consult the Journal Editor if your article does not fit within these limits).
Reference limit: ~50
Required sections (for a more detailed description see Article sections):

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Body of the article
- Discussion/conclusion
- Summary points
- References: approximately 50 references or fewer
- Reference annotations
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information

Editorial

Editorials are short articles that provide an insight into, or snapshot of issues of topical importance to the journal's target audience or researchers and other professionals. The intention is that the article should offer an expert perspective on a topic of recent interest. More detailed discussions can take the form of Commentary articles. Invited Editorial articles undergo internal review; unsolicited Editorials will undergo external peer review at the Editor's discretion.

Word limit: 1500 words maximum (excluding keywords and references).
Reference limit: ~20
Required sections (for a more detailed description see Article sections):

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Plain language summary (optional; maximum 250 words)
- **Please note:** No main abstract is required in Editorials/Commentaries
- Keywords (5–10)
- Body of article
- References: A maximum of 20 references are permitted
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- **Please note:** No figures, tables or boxes are permitted

Commentary

Commentaries are short articles that are similar to Editorials yet provide a more detailed discussion of a topic. Invited Commentary articles undergo internal review; unsolicited Commentaries will undergo external peer review at the Editor's discretion.

Word limit: 1500–3000 words (excluding keywords and references).
Reference limit: ~20
Required sections: As for Editorial

Conference Report

Conference Reports summarize the most important research presented at a recent relevant meeting or event. It is not usually feasible to attempt comprehensive coverage of the conference; authors should therefore focus on those presentations that are most topical, interesting or thought-provoking. Invited Conference Report articles undergo internal review; unsolicited Conference Reports will undergo external peer review at the Editor's discretion.

Word limit: 1500 words maximum (excluding abstract, conference details and references).
Reference limit: ~20
Required sections:

- Title (maximum 120 characters)
- Conference details (title, date, location)
- Abstract/overview of meeting (120 words maximum)
- Body of article
- References: a maximum of 20 references are permitted
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- **Please note:** No figures, tables or boxes are permitted in Conference Reports

Company/Institutional Profile

Company Profiles and Institutional Profiles allow representatives from a particular organization to describe the work currently being carried out by their team, relevant to the field of the journal in question. This can include pharmaceutical and biotechnology companies, or academic institutes/departments and government organizations.

These reports are intended to provide an insight into the history and strategy of a company/institution, and profile its capabilities, advanced technologies and future potential. Individuals are invited to write a Company or Institutional Profile at the Editor's discretion, and the contents of the piece undergo internal review.

Word limit: 2000 words
Reference limit: ~20
Required sections (for a more detailed description of these sections see Article sections):

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Abstract (maximum 120 words)
- Keywords (5–10)
- Introduction: brief factual account of the history and strategy of the company including background information e.g., the year the company was founded, number of employees etc.
- Body of article
- Summary points
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- References: a maximum of 20 references are permitted
- Figures/tables: if necessary, only **one of each** is permitted, which should be submitted as separate files

Patent Spotlight

Patent Spotlights are review-style articles that focus on a single patent, or collection of closely related patents. They offer the author the opportunity to put cutting-edge inventions into context, and to discuss the potential impact of the patented compounds or healthcare products among the wider drug discovery community. Authors of Patent Spotlights are encouraged to be opinionated and forward-thinking, and are able to offer a unique perspective on the future of their field of research in light of an innovative pharmaceutical invention. Authors are welcome to choose to discuss their own work, or that of colleagues or companies working within their field of expertise. Patent Spotlight articles undergo external peer review.

Word limit: 1500–3000 words (excluding abstract, keywords, executive summary and references).
Reference limit: 50
Required sections:

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Abstract (maximum 120 words)
- Plain language summary (optional; maximum 250 words)
- Tweetable abstract (optional; ~200 characters)
- Keywords (5–10)
- Body of article
- Future perspective
- Executive summary
- References: limit of 50 references
- Reference annotations
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information
- Figures/Tables: should be submitted as separate files (see guidelines below)

Hot Topic

Hot Topic articles provide an objective and factual overview of a specific issue or topic of debate – be it a legal concept, landmark case, or policy. They should contain the following: a concise overview of a topic and its history if appropriate; discussion (e.g. of recent developments, current opinions, ongoing debate, etc.); future outlook (e.g. short summary of likely developments or future challenges).

Word limit: 1000–1500 words (excluding keywords and references).
Reference limit: ~20
Required sections:

- Title (maximum 120 characters)
- Author(s) names & affiliations
- Tweetable abstract (optional; ~200 characters)
- **Please note:** No main abstract is required in Hot Topic articles
- Keywords (5–10)
- Body of article
- References
- Acknowledgements: author acknowledgements, plus, where relevant, details of individuals who contributed to the article, but who did not fulfill the criteria to be listed as authors
- Disclosures: to include funding information, financial and/or conflict-of-interest disclosures, disclosure of any writing assistance (and the funding source for this), and any other relevant information

- **Please note:** No figures, tables or boxes are permitted in Hot Topic articles

Letter to the Editor

Readers may submit Letters to the Editor, commenting on an article published in the journal.

Word limit: 1500 words

Inclusion of Letters to the Editor in the journal is at the discretion of the Editor, and they may undergo external review. All Letters to the Editor will be sent to the author of the original article, who will have 28 days to provide a **Letter in Reply** to be published alongside the Letter.

Article sections

The following list provides notes on the key article sections; authors should consult the 'At-a-glance formatting checklist' to determine which sections are required for their submission.

Title

Concisely and clearly conveys the scope/novelty of the article including any key words or phrases people might use to search on the topic; not more than **120 characters**. Should not include abbreviations if possible, and should avoid redundant language such as "A study of…".

Author(s) names & affiliations

Including full name, postal address, phone and fax numbers, and e-mail address. Note: we can only list one corresponding author. Where available, authors should also add their ORCID iD during the manuscript submission process. For more information on ORCID, see below. Where patient authors are included, **an affiliation of 'Patient author' should be included** (alongside any additional affiliation desired), to facilitate discoverability on indexing services such as PubMed.

Guidance on author sequence:

Author sequence is at the authors' discretion; however, Future Science Group journals suggest following the recommendations in GPP3 Appendix Table 2 (https://www.ismpp.org/gpp3), whereby authors are listed either in order of the level of their contribution, or alphabetically. The corresponding author should always be indicated.

Guidance on a change of affiliation during writing:

Where an author has changed their affiliation prior to the publication of an article, the affiliation should reflect where the major part of the work was completed. Current affiliation and contact information should be listed in an acknowledgement.

Authorship criteria:

Future Science Group follows the recommendations of the ICMJE as regards authorship – authorship should be based on the following four criteria:

1. Substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work; AND
2. Drafting the work or revising it critically for important intellectual content; AND
3. Final approval of the version to be published; AND
4. Agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Contributors who do not fulfill all four criteria should be listed in the acknowledgements section.

Future Science Group is supportive of diverse authorship groups and collaboration. We support the ICMJE recommendations that **individuals who meet the first authorship criterion should be given the opportunity to meet the other criteria wherever possible**, and encourage collaboration and co-authorship with colleagues in the locations where the research is conducted.

Fair accreditation of authorship:

Future Science Group Editors will endeavor to identify papers during the submission process where authorship/contributorship has not been appropriately designated, or that might fall into the category of 'helicopter science', and raise the matter with the submitting author accordingly. In addition, we encourage readers who have concerns on this issue to contact the Journal Editor (via the 'Email us' link on each journal homepage).

Patient authorship:
Future Science Group is supportive of the inclusion of patients in all stages of research, including in the authorship of papers. Patient authors can include:

- **A person who lives with or is affected by a disease or condition** (i.e., a broad definition of patient that includes those with lived conditions or receiving health or social care, caregivers, family members and members of patient advocacy groups who represent them)
- **A person who provides unique and valuable input from the patient perspective to the publication**.
- **A person who meets all the criteria required for authorship, as listed above.** Authors are encouraged to refer to this tool, which highlights how each of the four criteria above can be interpreted from the patient author perspective.

Further useful information for patient authors can be found in the **WeCan training module on "Patients in Publications"**.

Group authorship:
When a group name is included as an author (e.g., the XYZ Study Group), the respective group member names should be listed in the acknowledgements section. In relevant Medline/PubMed-indexed journals, these individuals are acknowledged as contributors to the article. The submitting author/agent should therefore ensure that group member names are included in full, are spelled correctly, and appear in the order they wish them to be listed on Medline/PubMed. More guidance from Medline can be found here: https://www.nlm.nih.gov/bsd/policy/authorship.html.

Changes to authorship:
Should a change to authorship be required either before or after article publication, this should be brought to the attention of the Journal Editor. This will then be investigated, and corrections made if deemed appropriate by the Editor and with the agreement of all authors involved (including those being added/removed).

Abstract
Not more than **120 words**; no references should be cited in the abstract. The abstract should highlight the importance of the field under discussion within the journal's scope, and clearly define the parameters of the article.

Structured abstract (for article types where this is specified e.g., research articles)
Not more than **120 words**, broken down into Aims, Patients & Methods/Materials & Methods, Results and Conclusions. For authors presenting the results of clinical trials, the guidelines recommended by CONSORT should be followed when writing the abstract, and the clinical trial registration number included at the end of the abstract, where available.

Data deposition: where data have been deposited in a public repository, authors should state at the end of the abstract the data set name, repository name and number.

Lay abstract

Plain Language Summary (within article)
Plain Language Summaries (PLS) within an article are a short, text-only summary of the article with any technical jargon removed. PLS should be of a similar length to a regular abstract or shorter (no more than 250 words) and are featured within an article alongside the main abstract (and on

PubMed, for journals that are indexed there). PLS are peer reviewed, and wherever possible should be submitted at the same time as the manuscript.

We recommend structuring the PLS as a series of questions, such as:

- What is this article about?
- What were the results?
- What do the results of the study mean?

Example:

What is this summary about?: Sodium oxybate is a medicine for narcolepsy symptoms. It contains a high level of sodium. Should people taking sodium oxybate and their doctors worry about the sodium increasing their risk of heart or cardiovascular problems? This is a summary of an article that reviewed 20 years of published data to answer that question.

What were the results?: We found that sodium oxybate was not linked to cardiovascular risks, such as heart attacks or strokes.

What do the results mean?: This suggests that the sodium in sodium oxybate may not add cardiovascular risk for people with narcolepsy. People currently taking sodium oxybate should talk to their doctor to ask if they need to be concerned about the sodium in their medicine. People who take sodium oxybate are unlikely to need to change their sodium oxybate medicine because of the sodium.

For links to some useful resources when writing PLS, see below.

Tweetable abstract

A tweetable abstract is a short summary of your article used to share it on Twitter, helping a wider audience discover your work. Tweetable abstracts are included in the article publication and can be shared directly on Twitter via the **'Click to Tweet'** feature on the article page. Authors are encouraged to provide a tweetable abstract during submission summarizing the key findings from the article and including any relevant hashtags. Tweets can be up to a maximum of 240 characters, however we recommend ~200 characters for a tweetable abstract (shorter tweets generally receive better engagement).

Please note: The tweetable abstract field is compulsory on our submission sites; if you do not wish to include one, please type "Not applicable" in this section instead.

Keywords

Up to ten keywords (minimum of three), including therapeutic area, mechanism(s) of action etc., plus names of drugs and compounds mentioned in the text.

Body of the article

Article content should be arranged under relevant headings and subheadings to assist the reader.

Future perspective

The author is challenged to include speculative viewpoint on how the field will have evolved 5–10 years from the point at which the article was written.

Executive summary

Not more than 600 words. Bulleted summary points that illustrate the main conclusions made throughout the article. Where appropriate, relevant headings that correspond to those in the manuscript should be inserted.

Example:

Executive summary

Background
- To investigate PK/PD relations and the feasibility to implement therapeutic drug monitoring a LC-MS/MS method to quantify vemurafenib is dried blood spots (DBS) samples was developed.

Experimental
- Whatman FTA DMPK-A cards were used for sample collection.
- This DBS method was validated according to the FDA and the latest EMA guidelines on method validation.

Results & discussion
- DBS samples can be collected by finger prick, since the effect of blood volume and blood spreadability was within the acceptance criteria.
- The DBS concentrations of vemurafenib in DBS samples collected in the outpatient clinic were all within the validated range of the developed assay.

Conclusion
- The assay is considered suitable to quantify vemurafenib in DBS samples.

Summary points (for article types where this is specified e.g., Research Article)

8–10 bullet point sentences highlighting the key points of the article.

Example:

Summary points

- Previously, the real-world observational GioTag study indicated that sequential use of afatinib and osimertinib warranted further assessment as a treatment strategy in patients with *EGFR* mutation-positive non-small-cell lung cancer; however, overall survival (OS) data were immature.
- In this updated analysis, median OS was 41.3 months (90% CI: 36.8–46.3) and 2-year OS rate was 80%.
- In patients with an *EGFR* Del19 mutation at the onset of treatment with afatinib, median OS was 45.7 months (90% CI: 45.3–51.5) and 2-year OS rate was 82%.
- Overall, the median time on EGFR-TKI treatment was 28.1 months (90% CI: 26.8–30.3).
- Median time on osimertinib treatment was 15.6 months (90% CI: 13.8–17.1) indicating that substantial clinical benefit with osimertinib can be achieved in a second-line setting following afatinib.
- These data, along with high rate of accrual of T790M in patients treated with afatinib, especially in patients with Del19-positive disease, indicate that sequential afatinib followed by osimertinib is potentially a feasible therapeutic strategy.
- Prospective data are required to evaluate the OS of patients treated with different EGFR-TKIs, and sequential regimens, in patients with *EGFR* mutation-positive non-small-cell lung cancer.

Author contributions

Author contributions (for article types where this is specified e.g., Research Article)

Brief summary of the contribution of each individual meeting the criteria to be listed as an author on the manuscript. For example: “Author X was responsible for study conception and design; authors X and Y were responsible for acquisition of data; authors X, Y and Z were responsible for data analysis, and drafting and revision of the manuscript.”

Acknowledgements

Author acknowledgements, plus, where relevant, details of individuals who contributed to the article, such as study group members, or those who contributed but who did not fulfill the criteria to be listed as authors.

Disclosures

For more information, see Disclosures. Should include:

Financial and conflict of interest disclosures (or lack of), including:

- Disclosure of financial support for the current work
- Author conflict of interest disclosures
- Writing assistance disclosure, along with any sources of funding for such assistance

Ethical conduct of research disclosure (where relevant)

Data sharing statement (where relevant)

For an example financial and conflict of interest disclosure, see here.
For an example ethical disclosure, see here.
For example data sharing statements, see here.

ORCID

Future Science Group is pleased to be a Member Organization of ORCID, the Open Researcher and Contributor ID, underlining our commitment to transparency and discoverability for our authors: https://www.future-science.com/orcid

Please note: ORCID resources in many languages (including Spanish, Portuguese, Chinese and Arabic, amongst others) can be found here: https://info.orcid.org/outreach-resources/

What is ORCID?

ORCID provides researchers with a unique identifier – an ORCID iD – plus a mechanism for linking their research outputs and activities to their ORCID iD.

An ORCID iD is a unique and persistent digital identifier that ensures your work is correctly associated with you, regardless of whether your name is similar to (or the same as!) another individual, or if your name changes.

ORCID is integrated into many systems used by publishers, funders, institutions and other research-related services.

Why register?

Your ORCID iD:

- Distinguishes you and ensures your research outputs and activities are correctly attributed to you
- Reliably and easily connects you with your contributions and affiliations
- Reduces form-filling: you enter data once, have it reused often
- Improves recognition and discoverability for you and your research outputs
- Is interoperable: it works with many institutions, funders, and publishers
- Is persistent: you can use it throughout your research career

Watch "Why ORCID?" to learn more: https://vimeo.com/237730655

Connect your ORCID iD to Future Science Group

Future Science Group's submission system, ScholarOne, supports ORCID, giving you the option to include an ORCID iD when you submit a manuscript to us.

If you already have an ORCID iD, simply associate this when creating your account on ScholarOne. Alternatively, if you have yet to register for an ORCID iD, click the appropriate link to create one:

By providing an ORCID iD during the submission process, it can then be incorporated by the journal into your accepted article's metadata, ensuring your work is appropriately attributed to you and that your ORCID record is updated accordingly.

Your ORCID iD will also be published within the article so that readers can link to your public ORCID profile, and from there to your other work.

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Submitting authors/agents and the Journal Editor are unable to add ORCID iDs to a co-author's account – only the individual themselves can do so. However, all authors are provided with information on how to do this during the manuscript submission process, and if added prior to article acceptance their ORCID iD will also be published within the article.

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Disclosures

The following provides further information on financial, COI, ethical and data sharing disclosures that should be included in all relevant publications. Authors should also complete and submit the Author Disclosure form (only one author needs to complete the form on behalf of all co-authors for the manuscript).

Financial & competing interests disclosure

Disclosing any information about financial support for the current work, as well as the interests of the author(s) that could influence how readers receive and understand the work. This includes information related to:

- **The work under consideration for publication** – detailing any resources received directly or indirectly (via your institution) to enable the completion of the work (with a timeframe **from the initial conception of the work, to the present**) – such as grants. This includes funding for any **writing assistance** that has been used in the creation of the manuscript, which should be stated along with the sources of funding for such assistance.
- **Relevant financial activities outside the submitted work** – disclosing interactions (e.g., personal, academic or financial relationships) with any entity that could be considered broadly relevant to the work, that could be perceived to influence, or that gives the appearance of potentially influencing, the submitted work. Authors should disclose any such interactions that have occurred for a period of **36 months prior to the submission**.
- **Intellectual property**
- **Any other relationships not covered above** that could be perceived by readers to have influenced, or give the appearance of potentially influencing, the work.

For further detail, authors should refer to the **Author Disclosure Form** (available here), which should also be completed and submitted alongside their manuscript submission.

These requirements are based on the ICMJE Conflict of Interest policies (http://www.icmje.org/conflicts-of-interest/).

Example financial & competing interests disclosure:

"This work was supported by a grant from FUNDING BODY (grant no.: XYZ12345). AUTHOR 1 has received consultancy fees from COMPANY A and COMPANY B. AUTHOR 2 has received speaker fees from COMPANY C, has been an advisory board member for COMPANY D, and owns stock in COMPANY E. Author 3 holds a patent for XXX (patent number: XXX). The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

Medical writing and editorial support were provided by WRITER of MEDICAL COMMUNICATIONS COMPANY, and were funded by COMPANY A."

Ethical conduct of research

For studies involving data relating to human or animal experimental investigations, authors should obtain appropriate institutional review board approval and state this within the article (for those investigators who do not have formal ethics review committees, the principles outlined in the Declaration of Helsinki should be followed, and this should be stated accordingly).

In addition, for investigations involving human subjects, authors should obtain informed consent from the participants involved and include an explanation of how this was obtained in the manuscript.

Example ethical disclosure:

"The authors state that they have obtained institutional review board approval from INSTITUTION for the research described. In addition, they have obtained verbal and written informed consent from the patients for the inclusion of their medical and treatment history within this work."

Data sharing statement

For studies reporting the original results of a clinical trial or the secondary analysis of clinical trial data, authors should include a data sharing statement, as described on the ICMJE website: http://www.icmje.org/recommendations/browse/publishing-and-editorial-issues/clinical-trial-registration.html

Authors are asked to specify whether their manuscript reports **either** the original results of a clinical trial, **or** the secondary analysis of clinical trial data that have been shared with them.

Original results of a clinical trial

For the reporting of original results, authors will be asked to complete the following table (found in the Author Disclosure Form), which will form the basis of the data sharing statement:

Will individual, de-identified participant data be available (including data dictionaries)?	
What data in particular will be shared?	
What other documents will be available, if any (e.g., study protocol, statistical analysis plan, etc.)?	
When will data be available (start and end dates)?	
By what access criteria will data be shared? To include: - With whom? - For what types of analyses? - By what mechanism?	

Examples:

"The authors certify that this manuscript reports original clinical trial data. The data will not be made publicly available."

"The authors certify that this manuscript reports original clinical trial data. Individual, de-identified participant data that underlie the results reported in this article (text, tables, figures, and appendices) are available from the corresponding author following publication, including the clinical study report and study protocol."

"The authors certify that this manuscript reports original clinical trial data. Data reported in this manuscript are available within the article or posted publicly at www.clinicaltrials.gov, according to the required timelines. Additional data from the study (e.g., study protocol) are available upon reasonable request."

Secondary analysis of shared clinical trial data

For the reporting of secondary analyses of clinical trial data that have been shared with the authors, a statement to this effect must be included, including the source of the data.

Example:

"The authors certify that this manuscript reports the secondary analysis of clinical trial data that have been shared with them, and that the use of this shared data is in accordance with the terms (if any) agreed upon their receipt. The source of this data is: *****."

References

Key points

- Authors should focus on recent papers and papers older than 5 years should not be included except for an over-riding purpose.
- Primary literature references, and any patents or websites, should be numerically listed in the reference section in the order that they occur in the text (including any references that only appear in figures/tables/boxes).
- Websites should only be cited where necessary and a peer-reviewed source is unavailable (authors should be aware that websites can subsequently become obsolete). Where included, a title and full web address should be provided, along with the date the site was accessed by the author(s).
- Preprints should only be cited where necessary and a peer-reviewed source is unavailable (authors should check in the final iteration of their article whether a peer-reviewed source has become available during article processing/review process, and replace the citation as necessary). Where included, the authors, title and full web address should be provided, along with the date the preprint was accessed by the author(s).
- Information from manuscripts submitted but not accepted should be cited in the text as "unpublished observations" with written permission from the source.
- Avoid citing a "personal communication" unless it provides essential information not available from a public source, in which case the name of the person and date of communication should be cited in the text, with written permission from the source.
- References should be denoted numerically and in sequence in the text, using Arabic numerals placed in square brackets, e.g., [12].
- Quote first six authors' names. If there are more than six, then quote first three *et al*.
- Reference annotations: 6–8 references should be highlighted that are of particular significance to the subject under review as "* of interest" or "** of considerable interest". Each of the chosen references should be annotated with a brief sentence explaining why the reference is considered to be of interest/particular interest.

  Example:

  18 Timmerman P, White S, Cobb Z, de Vries R, Thomas E, van Baar B. Update of the EBF recommendation for the use of DBS in regulated bioanalysis integrating the conclusions from the EBF DBS-microsampling consortium. *Bioanalysis* 5(17), 2129–2136 (2013).

  •• **This article sets a standard for dried blood spot method validation.**

- Any references that are cited in figures/tables/boxes that do not appear in the text should also be numerically listed in the reference section in the order that the figure/table/box appears in the text.
- The Future Science Group EndNote style can be downloaded from our website at: https://www.future-science.com/authorguide/preparingyourarticle
- The Future Science Group Citation Style Language (.csl) for use with Zotero and other CSL 1.0.1-compatible software such as Mendeley and Papers can be found here: https://www.zotero.org/styles?q=Future%20Science%20Group

Journal articles

- Author's names should appear without full stops in their initials
- List up to six authors' names. If there are more than six authors, then quote the first three only followed by *et al*.

- A full stop follows authors' names
- Article title given in full
- Journal name should be in italics and abbreviated to standard format, including full stops
- Volume number, with the issue number in brackets (if available), followed by comma
- Page number range separated by a hyphen with no spaces, followed by the year in brackets, and then a full stop
- ***Example:***
    - Fluhler E, Hayes R, Garofolo F *et al.* 2014 White Paper on recent issues in bioanalysis: a full immersion in bioanalysis (Part 1 – small molecules by LCMS). *Bioanalysis* 6(22), 3039–3049 (2014).

Book chapters

- Author's names should appear without full stops in their initials
- List up to six authors' names. If there are more than six authors, then quote the first three only followed by *et al*.
- A full stop follows authors' names
- Chapter title given in full and followed by full stop
- Book title preceded by 'In:'
- If a particular edition is referenced, put in brackets e.g. (2nd Edition)
- List Editors as for authors, followed by (Ed.) for single editor or (Eds) for multiple
- Publisher name followed by town/state and country they are based in
- Page number range separated by a hyphen with no spaces, followed by the year in brackets, and then a full stop
- ***Example:***
    - Han J, Borchers CH. Chemical derivatization to enhance metabolomic analysis by LC/ESI–MS. In: *Advanced LC-MS Applications in Metabolomics.* Borchers CH, Han J, Parker C (Eds). Future Science Group, London, UK, 6–21 (2015).

Meeting abstract

- Author/speaker names should appear without full stops in their initials
- List up to six author/speaker names. If there are more than six, then quote the first three only followed by *et al*.
- A full stop follows author/speaker names
- Title of presentation followed by full stop
- Note either 'Presented at:' or 'Proceedings of'
- Town/state, country and date in full, followed by a full stop.
- ***Example:***
    - Staack R. Protein quantification by LBA or LC-MS: key criteria for the definition of the bioanalytical strategy. Presented at: *7th European Bioanalysis Forum Open Meeting*. Barcelona, Spain, 19–21 November 2014.

Websites

- If it is the homepage of a company or organization
    - List the name of the company or organization follow by a full stop
    - Include the URL of the website
    - Do not include 'http://' unless the address does not include 'www'
    - Include the date accessed
- If it is a particular webpage/document
    - Reference as much information as possible about the article (title, authors, date etc.)
    - Include the URL of the website
    - Do not include 'http://' unless the address does not include 'www'

  - Include the date accessed
- ***Example (organization homepage):***
  - US Food and Drug Administration. www.fda.gov (Accessed 30 July 2015)

Guidance articles

- Title of guidance followed by full stop
- Organization name followed by town/state and country they are based in
- Year of publication in brackets, followed by a full stop
- ***Examples:***
  - *Guidance for Industry Bioanalytical Method Validation. US Department of Health and Human Services, US FDA,* Rockville, MD, USA (2001).
  - *Guideline on bioanalytical method validation.* European Medicines Agency, London, UK (2011).

Preprints

- Author's names should appear without full stops in their initials
- List up to six authors' names. If there are more than six authors, then quote the first three only followed by *et al*.
- A full stop follows authors' names
- Preprint title given in full
- Include the URL of the preprint
- Include the date accessed
- ***Example:*** Hamilton DG, Page MJ, Finch S, Everitt S, Fidler F. How often do cancer researchers make their data and code available and what factors are associated with sharing? Available at: https://www.medrxiv.org/content/10.1101/2022.03.10.22272231v1 (Accessed: 16 March 2022).

Reference annotations

Papers or of particular interest should be identified using one or two asterisk symbols:

- * = of interest
- ** = of considerable interest

Each of the chosen references should be annotated with a brief sentence explaining why the reference is considered to be of interest/particular interest.

Example:

59. Cardillo TM, Sharkey RM, Rossi DL, Arrojo R, Mostafa AA, Goldenberg DM. Synthetic lethality exploitation by an anti-Trop-2-SN-38 antibody–drug conjugate, IMMU-132, plus PARP inhibitors in *BRCA1/2*-wild-type triple-negative breast cancer. *Clin. Cancer Res.* 23(13), 3405–3415 (2017).

•• **This preclinical study demonstrated antitumor responses of an anti-Trop2 antibody–drug conjugate in both mouse and monkey models.**

Making the most of your article

We encourage authors to enhance their article with various digital features to help readers discover and learn about their research. More information is available here. With in-house graphics and video teams, we can offer a range of services to assist you in the preparation of all digital enhancements. If you are interested in including any digital enhancements with your article, please contact Joanne Walker at any stage.

All digital features undergo peer review. With the exception of graphical abstracts, digital features are published alongside the article as supplementary materials and can be accessed via a thumbnail on the article page. All digital features can be accessed free of charge. Authors retain the copyright of any digital feature they submit to us.

When submitted with the original submission, there are no costs to publish a digital feature. Costs are incurred if we are required to create or edit the feature.

Digital features (apart from graphical abstracts) can be added post-publication. In this instance, a fee of $500 is charged to take into account the additional editorial and publication processing time and costs.

Features are also posted on Figshare, allowing them to be cited independently (for more information in Figshare, see below).

Plain Language Summaries

Plain Language Summaries (PLS) provide a summary of an article in non-technical, jargon-free language that is understandable to non-specialist audiences. These are valuable to a range of readers, including patients, patient advocates, the general public, non-specialist clinicians, research scientists, decision-makers and a range of professionals in the healthcare community.

Future Science Group journals offer several options for authors who wish to publish a PLS, details of which can be found below, along with links to external useful resources.

Plain Language Summary (within article)

PLS within an article are a short, text-only summary of the article with any technical jargon removed. PLS should be of a similar length to a regular abstract or shorter (no more than 250 words) and are featured within an article alongside the main abstract (and on PubMed, for journals that are indexed there). PLS are peer reviewed, and wherever possible should be submitted at the same time as the manuscript.

Plain Language Summary (alongside article)

Longer form PLS, providing a more detailed summary of the paper, can also be submitted as a supplementary file alongside an article submission. Any format will be considered, including written, video and audio format.

For authors wishing to feature a PLS alongside their article, we offer a writing and development service. If you are interested in learning more, please contact us.

Standalone Plain Language Summary of Publication articles

Plain Language Summary of Publication articles (PLSPs) can be published in all Future Science Group journals. These are standalone articles published in the journal with their own unique DOI and are

thus fully citable. They are plain language, visually enriched articles that provide a summary of a key publication, from a FSG journal or elsewhere.

PLSPs are written by authors of the original publication, ideally with a patient as co-author (although this is not mandatory). Additional authors not involved with the original publication can be included in the PLSP; however, they must meet the authorship criteria stipulated by the ICMJE. Following submission of the PLSP to the appropriate Future Science Group journal, prior to styling into our PLSP template, it will be externally peer reviewed for readability and understanding by suitable individuals selected by the Journal Editor on the basis of experience and expertise.

Separate guidelines are available for the preparation of standalone PLSPs. If you are interested in submitting a standalone PLSLP, please contact us to discuss the next steps.

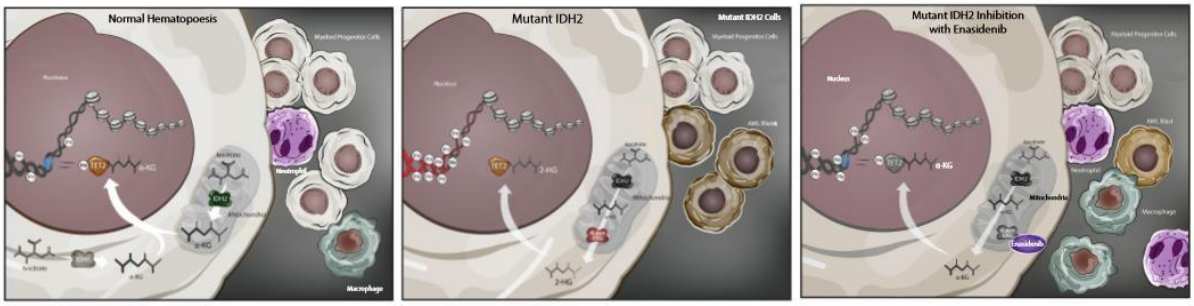
Click here to see all of our Plain Language Summaries.

Links to external useful resources

- Future Science Group' website on plain language summaries: https://www.plainlanguagesummaries.com/
- Multi-Regional Clinical Trials Center (MRCTC) Clinical Research Glossary listing research words and their meanings: https://mrctcenter.org/clinical-research-glossary/
- EMA medical terms simplifier
- Medical Terms in Lay Language (from the University of Iowa)
- Flesch-Kincaid Readability Test
- Plain Language Summaries (PLS) of Publications Toolkit
    - From Envision Pharma Group, co-created with support from Patient Focused Medicines Development (PFMD)
- Recommendations of the EU expert group on clinical trials on "Summaries of Clinical Trial Results for Laypersons": https://ec.europa.eu/health/sites/health/files/files/eudralex/vol-10/2017_01_26_summaries_of_ct_results_for_laypersons.pdf
- PFMD resources, including:
    - Blog post: "Plain language summaries: how patients are helping bring the fruits of research to the public"
    - Blog post: "Big pharma, small pharma - the biotech industry is embracing Plain Language Summaries"
    - How to Guide: "Plain language summaries (PLS) of peer-reviewed publications and conference presentations: practical 'How-To' Guide for multi-stakeholder co-creation"
- Open Pharma resources
    - Article published September 2021 in Current Medical Research and Opinion: "Open Pharma recommendations for plain language summaries of peer-reviewed medical journal publications"
    - What's in a name? A crib sheet for plain language documents
- ISMPP endorsed and sponsored article published November 2021 in Current Medical Research and Opinion "Plain language summaries of publications of company-sponsored medical research: what key questions do we need to address?"

Graphical abstract

All Future Science Group journals encourage the use of graphical abstracts; a concise, visual summary of the main findings of the article, helping readers to quickly understand the findings of the paper and its relevance to them.

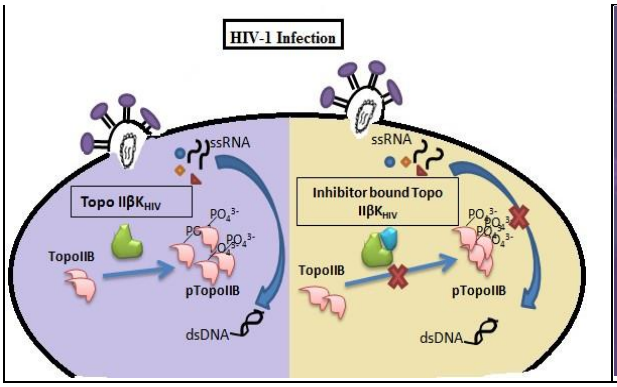


Graphical abstracts are made freely accessible to all readers and feature prominently on the article webpage alongside the main abstract. They will also be used by the Journal Editors to promote articles to audiences via social media.

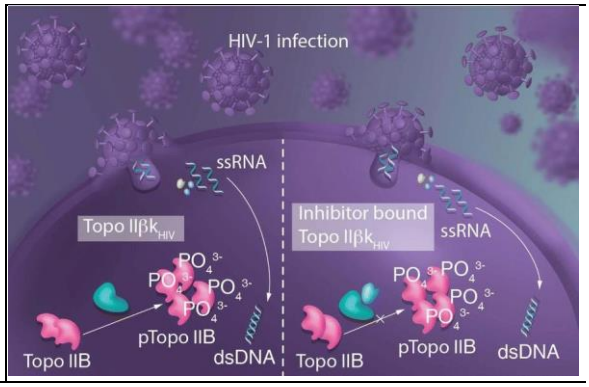
Graphical abstracts are peer reviewed alongside the article and should be submitted with the first draft. However, this does not need to be the final version – we are happy to accept a rough sketch or equivalent that will resemble the final version. The final version can then be created whilst the draft is being reviewed and finalized based on the reviewers' feedback.

Submission requirements: Authors should provide a single image or split panels in one image, ideally using font HELVETICA; size 8 points. Files should be supplied as a .jpg, .pdf or .tif file. If required, we can provide a range of design support services, from polishing an existing figure to completely creating the graphical abstract from a hand-drawn figure.

Before



After



Click here to see all of our Graphical Abstracts.

Infographics

Infographics go beyond the graphical abstract and provide a more in-depth, at-a-glance overview of the information presented in the article. An example is available at https://www.futuremedicine.com/doi/10.2217/fon-2017-0646. Infographics will appear at the end of the article PDF and online alongside the article. These will also be used by the Editor when sharing

details of the article via social media (for more information on our social media activities please see the 'Post-publication tools' section).

Submission requirements: Please supply as a PDF and Illustrator file (if possible). The font size should be minimum 9pt (clearly legible).

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Videos

With more and more content being consumed in video format, we offer a number of options to help authors summarize their scientific research and journal articles through engaging videos.

Videos abstracts are short, 2–3-minute videos that provide the reader with a summary of the paper. Typically produced by the author, these can be published alongside articles free of charge. Video abstracts are featured alongside articles as well as on our **YouTube channel** (where they are tagged with keywords, a short description and a link to the original publication). For an example video abstract, please visit https://www.futuremedicine.com/doi/10.2217/fon-2017-0636. Download our guide to preparing a video abstract (available on our website here) or check out our handy video.

Animated videos provide a ~90 second overview of a research paper where the article is converted into a bespoke animation with an engaging voiceover script. Videos are featured alongside the journal publication and also hosted on the *Video Journal of Biomedicine* with their own citable DOI and the video transcript and metadata. All animated videos are published free to access, even if you have not chosen our open access option.

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- If you are interested in any of our options to feature a video alongside your article, please email Joanne Walker in the first instance.

Figures, tables, boxes & supplementary materials (including videos)

Summary figures, tables and boxes are very useful, and we encourage their use in certain article types (see above section on Article types for details on which articles can include figures/tables/boxes). The author should include illustrations to condense and illustrate the information they wish to convey. Commentary that augments an article and could be viewed as 'stand-alone' should be included in a separate box. An example would be a summary of a particular trial or trial series, a case study summary or a series of terms explained. _Article_types_1

Figures, tables and boxes should be numbered consecutively according to the order in which they have been first cited in the text.

Figure/table/box guidelines

- **File submission:** All figures, tables and boxes should be **submitted as separate files**, not within the main manuscript document.
    - It is acceptable to include e.g., one table file with multiple tables included, but this should be a separate document to the main text file.
- **File format:** Figures, tables and boxes should be submitted in an **editable format** where possible. Figures that can be included without editing (e.g., photos, imaging data, etc.) can be submitted as raster files (.jpg, .png or .tif). Other figures (e.g., graph/bar charts or complex illustrations) should ideally be provided as vector files (.ai, .eps or .svg) if possible, otherwise as a .jpg, .pdf or .tif. Tables/boxes should be provided as e.g., Microsoft Word or Microsoft Excel files, and must be editable. If you are uncertain whether the format of your files is appropriate, please check with the Journal Editor.
- **Resolution:** Figure resolution should be as high as possible, ideally 300 dpi or higher for a .jpg. Images that are blurry or illegible in any way will not be accepted.
- **Font:** If possible, please use Helvetica 8pt.
- **Abbreviations:** All abbreviations used within Figures/tables/boxes should be defined in the legend (even if previously defined in the body of the manuscript).
- **Photomicrograph:** Please ensure that **scale bars** are included in figures where appropriate (e.g., photomicrographs). Symbols, arrows or letters used in photomicrographs should contrast with the background. Please explain internal scale and identify the method of staining in photomicrographs.
- **Editing of figures:** Future Science Group journals apply the Council of Science Editors recommendations for digital images, specifically:
    - No specific feature within an image may be enhanced, obscured, moved, removed, or introduced.
    - Adjustments of brightness, contrast, or color balance are acceptable if they are applied to the whole image and as long as they do not obscure, eliminate, or misrepresent any information present in the original.
    - The grouping of images from different parts of the same gel, or from different gels, fields, or exposures must be made explicit by the arrangement of the figure (e.g., dividing lines) and in the text of the figure legend.
    - If the original data cannot be produced by an author when asked to provide it, acceptance of the manuscript may be revoked.

  For further information, please see the 'Scientific misconduct & retraction' section on our website here.

Future Science Group is able to offer a number of design services to authors, from polishing an existing figure to creating one from scratch (subject to fees). If you would be interested in learning more about this service, please contact Joanne Walker.

Chemical structures

If possible, please submit structures drawn in ISISDraw or ChemDraw format. However, chemical structures can be redrawn in-house. Please use the following conventions:

- Always indicate stereochemistry where necessary – use the wedge and hash bond convention for chiral centers and mark cis/trans bonds as such.
- Draw small peptides (up to five amino acids) in full; use amino acid abbreviations (Gly, Val, Leu, etc.) for larger peptides.
- Refer to each structure with a number in the text; submit a separate file (e.g., not pasted throughout the text) containing these numbered structures in the original chemical drawing package that you used.

Color figure charge

Future Science Group has a charge for the printing of color figures (e.g., each color figure) in the print issue of the journal. We have no page charges, unlike some other publishers, and aim to keep our color charge to a minimum.

This charge does not apply to the online (including PDF) version of articles, where all figures appear in color at no charge.

Our charges for color figures can be found here.

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  - not more than three figures from works published by a single publisher for an article, and not more than three figures from works published by a single publisher for a book chapter (and in total not more than thirty figures from a single publisher for re-publication in a book, including a multi-volume book with different authors per chapter).
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What should I submit to the journal to show permission has been obtained?

Please send us copies of letters or forms granting you permission for the use of copyrighted material so that we can see that any special requirements with regard to wording and placement of credits are fulfilled. Keep the originals for your files. If payment is required for use of the figure, this should be covered by the author.

Supplementary materials

All Future Science Group journals conduct double-anonymized peer review. Authors should ensure any supplementary materials files are **anonymized** (including the file names) to allow anonymity of the authors.

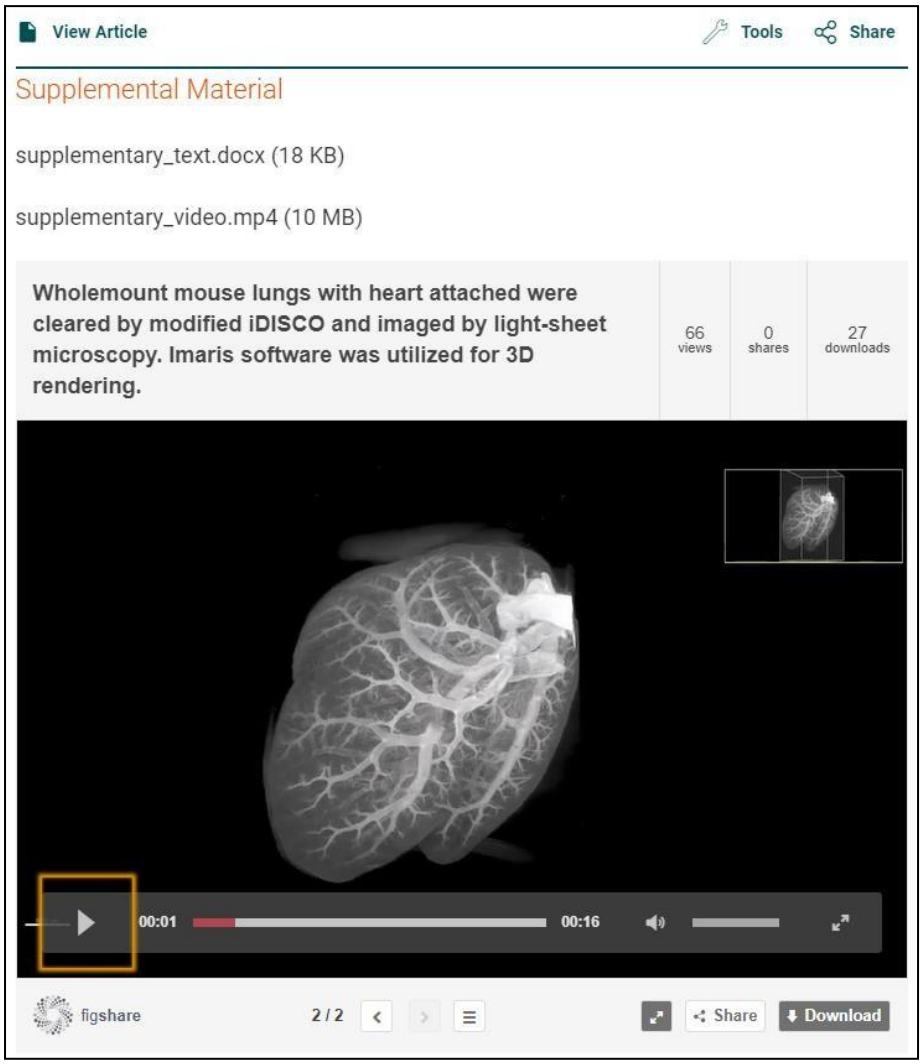
File size: Total files uploaded to the journal site cannot exceed 100MB. If you have files exceeding this limit, or have any difficulty uploading your files, please contact the Journal Editor for assistance.

Figure, tables and boxes larger than one A4 page will be included as online-only supplementary information. At the Editor's discretion data or experimental details can also be included.

Supplementary materials on Figshare

Figshare is a repository where users can make all of their research outputs, in any file format, available in a citable, shareable and discoverable manner. Future Science Group partners with Figshare to make all our supplementary files freely available and citable in their own right. Supplementary files feature both within the article on the journal site and on the Future Science Group Figshare portal.

To view an example, please see the following article's supplementary files on the journal site, and on the Figshare platform.



Supplementary videos

Our articles can be supported by videos online, including mechanism of action videos, videos of procedures, etc.

Videos are freely available to all readers and featured alongside the article abstract as supplemental files within the Details section (as in the example below). Videos are shared via social media (across Twitter, LinkedIn and Facebook) and can also be made available via the Future Science Group YouTube channel.

The copyright for any videos provided by the author will remain with the author. The author will grant the journal a license to publish the video, in accordance with the terms of the Copyright Assignment Form.

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Measurements of length, height, weight and volume should be reported in metric units (meter, kilogram or liter) or their decimal multiples.

Temperatures should be in degrees Celsius.

Centrifuge speeds should be given in g rather than rpm.

Any other units should be reported using the International System of Units (SI) where possible.

Statistics

Describe statistical methods with enough detail to enable a knowledgeable reader with access to the original data to judge its appropriateness for the study and to verify the reported results.

When possible, appropriate indicators of measurement error or uncertainty (such as confidence intervals or error bars) should be included.

Please define any statistical terms, abbreviations and symbols used.

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First draft submission should be made via our online submission system. Please ensure that you have prepared a complete text minus the title page, acknowledgements, and any running headers with author names, to allow anonymized review, and have not compromised your anonymity in any uploaded Supporting Information files or additional files for review. Please also have a separate title page prepared containing authors details, acknowledgements and disclosures.

File size: Total files uploaded to the journal site cannot exceed 100MB. If you have files exceeding this limit, or have any difficulty uploading your files, please contact the Journal Editor for assistance.

To help with the speed of processing of an article, authors should ensure that their article has been edited for language and grammar by a fluent English speaker prior to submission.

Submitting agents

Any third party (such as a medical writer or assistant) can submit via ScholarOne Manuscripts as a Submitting Agent. A "Guide to Article Submission for Submitting Agents" is available here.

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Future Science Group journals have different publishing options, depending on the title you are publishing in. Our subscription journals are 'hybrid' – articles are published behind a paywall as standard (accessible to journal subscribers and those who choose to pay a one-off fee to access the article), but authors also have the option to pay a fee to publish their article open access (making them freely available for all readers to access). Other journals are fully open access, with all articles requiring the payment of the open access fee on acceptance for publication. To find out what options are available for your chosen journal, see the table of information above. For more information on Open Access publication, see below.

Transfer of copyright

For authors publishing via the standard, non-Open Access route, copyright of the article is transferred to the journal. However, the author retains certain rights to re-use the content in future. For further details on this, please read the details in the Copyright Assignment Form, available here.

Required forms

These documents should be completed and submitted alongside article submissions as appropriate.

Compulsory:

Author Disclosure Forms – required for **all** submissions. These are available on our website.
PLEASE NOTE: Only one disclosure form is required per submission. One author (e.g., the submitting or corresponding author) should complete the form on behalf of all co-authors.
EITHER: Copyright Assignment Form – required if the open access option is not being taken. For articles where copyright is to be assigned to the journal, please complete the Copyright Assignment Form available on our website.
OR: Open Access Form – required if the open access option is being taken. For authors wishing to publish their article open access, a copyright form is not required. Please see visit the Open Access page for details on this option, and for the forms required.

PLEASE NOTE: Either a copyright form or an open access form will be required for an article to be published, which **must include a signature** (either signed by hand and scanned or including a valid e-signature, e.g., see here).

Optional:

Accelerated Publication Form – required if the accelerated publication option is being taken. For authors opting to use the Accelerated Publication service, please complete the form available on our website. For more information on this option, see below.

Initial quality review

Once the manuscript has been received in-house, it will undergo initial internal quality review by the Journal Editor. In all cases this will include checking:

- The article is within scope for the journal and relevant to its audience.
- The novelty of the work, including screening with plagiarism detection software.
- The clarity of presentation.
- That, for original research submissions, appropriate institutional review board approval has been obtained and is described within the article; and for investigations involving human subjects, informed consent has been obtained from the participants involved and an explanation of how this was obtained is included in the manuscript.

Each journal will also have specific criteria the Journal Editor will assess against to decide which articles will proceed to peer review. These vary by journal, but include criteria such as topicality, study sample size, etc.

Peer review

Articles deemed suitable for consideration following the above quality review will then proceed to external double-anonymized peer review (dependent on article type – for more details, see our Editorial policies on 'External peer review'). Please note, double-anonymized peer review cannot be guaranteed where a paper has been previously posted to a preprint server.

This usually takes around 4 weeks, although an Accelerated Publication option is also available. Please provide a list of suitable peer reviewers with your initial submission.

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After peer review is complete, time is allowed for any revisions (suggested by the referees/Editor) to be made. This period is approximately 2 weeks, but this subject to the nature of the revisions.

In-house production

Accepted manuscripts will undergo production in-house. This will involve type-setting, copy-editing, proof-reading and re-drawing of any graphics. Authors will receive proofs of their article for approval and sign off.

Please note that once the author receives the copy of their article for approval, our production department will need to hear from them within a tight deadline to ensure the issue is published on schedule. If you believe you may be away and unable to check the galley proofs at any point, please let the Journal Editor know.

Production process:

- Manuscript accepted by Journal Editor, and sent to the Production team
- Manuscript is typeset, figures/tables formatted, and house styles applied

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- **Author receives an email with information on how to access their article on the PXE platform (please be vigilant in case the email goes into your junk email folder). They are asked to:**
    - Answer any queries highlighted by the Production Editor
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- XML files of the final article are produced
- Article is published online, in the journal's 'Ahead of print' section
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Subscription journal green & gold open access options

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Authors who wish to use a green route to open access should refer to our self-archiving policy. Future Science Group journals conform to most green open access mandates. Authors who are unsure of the route, should contact our open access helpdesk via email.

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Should you wish to opt for gold open access publication within an FSG hybrid journal, you can do so by paying an article processing charge (APC). See the 'Article processing charges' section for current information on APCs for the various journals. The forms that you will need to complete can be found in the 'Required forms' section.

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If you have a query regarding our Open Access Options please contact Roshaine Wijayatunga, Head of Open Access Publishing.

Access tokens

What is an access token?

An access token allows a single user to access a certain amount of content an indefinite number of times. Most commonly, this is a single article – but can be any set of content, up to and including access to all of our content. Please note that access tokens are only available to authors for non-commercial purposes. For authors or third parties wishing to host an article on a company website, please contact us at reprints@future-science-group.com.

Why use an access token?

An access token offers a cost-effective and time-efficient way of offering access to a targeted group of people. It allows an author to share their work with their colleagues, peers and friends effortlessly by providing them a link to directly access the article, increasing the visibility of the article and its readership.

How does it work?

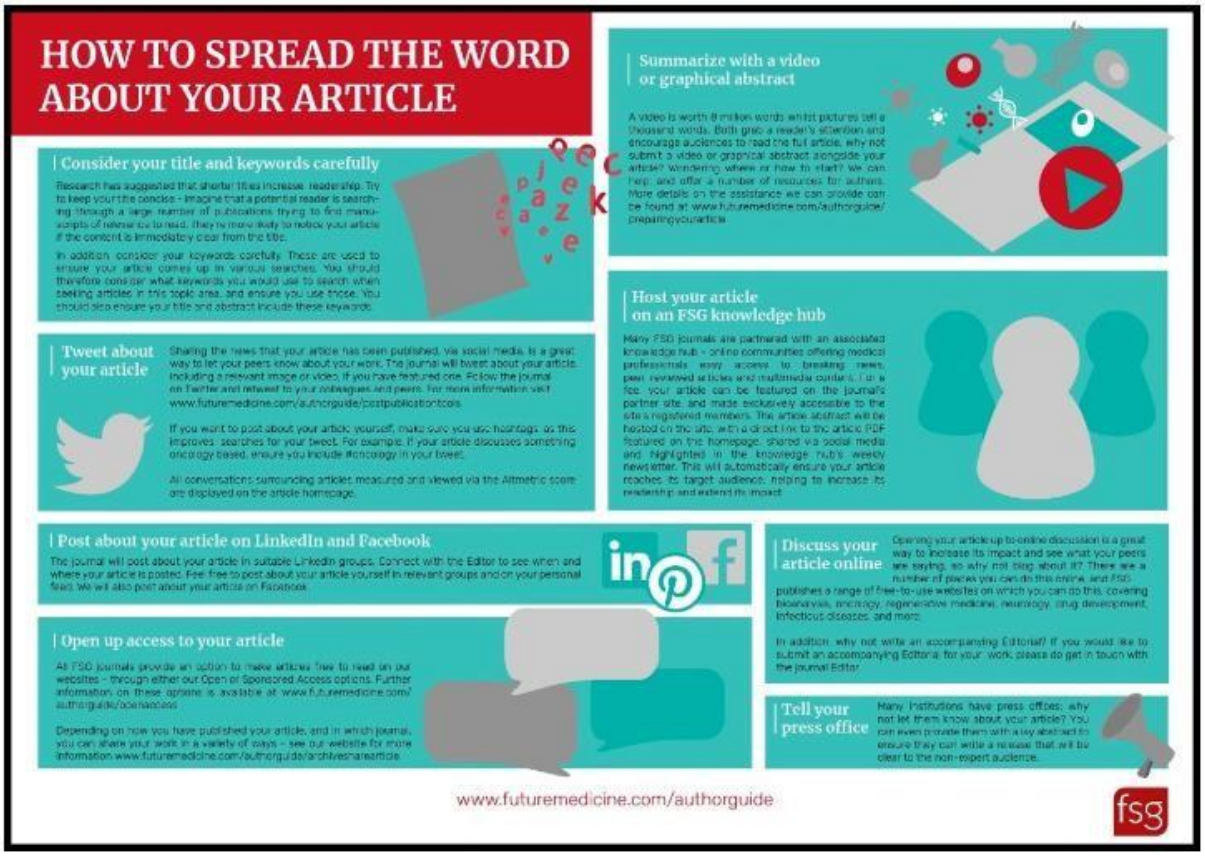
- An author should request access tokens directly through a staff member of Future Science Group, stating the article which they wish to purchase the tokens for.
- Once payment has been processed, the access tokens are immediately available for activation. The author will receive an automated e-mail that contains the details of how to share the access tokens with their colleagues, peers and friends - including suggested text which they can use as the base for any e-mail they might send.
- Each user who wishes to access the content must be provided with the activation link (contained in the e-mail the author receives) - in order to access the content, they must simply click on the link, then register (or login, if already a registered user) and the content will be available to them.
- There is no time limit within which the tokens must be activated, so there is no pressure on the author to ensure the content is accessed immediately.
- Once all the access tokens have been used, additional bundles of 50 can be purchased and the author can continue to distribute his content in the same way as described above.

Costs

Access tokens can be purchased in bundles of 50 at a time at a cost of **$150 per 50 tokens**, which is very cost effective compared with a single user purchasing a single pay-per-view article. Therefore, an author can share his work at a cost of only $3 per person – with no expiry date.

Post-publication tools

There are many ways to help increase the reach of your article; see the information below, and check out our infographic on "How to spread the word about your article" here:



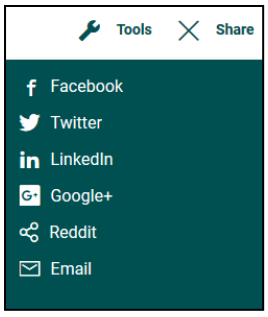
Social media

Future Science Group will share published articles via our social media channels, and encourage authors to suggest hashtags or accounts to @mention on our Author Disclosure form.

Authors are also encouraged to provide a **tweetable abstract** for the Journal Editor to use, summarizing the key message of the article (for more information see Article sections).

Sharing your own work

Sharing the news that your article has been published, via social media, is a great way to let your peers know about your work. Twitter, Facebook and LinkedIn are all great places to spread the word. All Future Science Group articles include sharing links at the top of the page, making it easy for you to create posts for your various accounts:



Twitter

Tips for creating a tweet about your article:

- Include a link to your article! Consider using a URL shortener, such as Bitly, to shorten your article URL.
- Include an image – tweets that include images attract far more engagement than those that don't. We are happy for authors to include images of any figures, or perhaps another image you think reflects the content of your article well
    - Be sure to check any image you use is in the public domain
- Use appropriate hashtags (utilize http://hashtagify.me/ for information on hashtags) – by marking out a key word or phrase by preceding it with a hashtag (such as #AlzheimersDisease), your tweet will be searchable and discoverable by other users. Many people follow certain hashtags regularly, so do have a look to find out what the key terms are for your subject area.
- @mention your co-authors, institution, funders etc. – most universities and some individual departments have their own Twitter accounts, and by mentioning their username (such as @futuresciencegp), they will be notified of your tweet in the Mentions section of their account. Once you have their attention, they may click to read your article or share your tweet with their followers! The tweet will also be visible to anyone who follows you, as normal.
    - Beware of the difference between @mention and @reply – an @reply places the username at the front of your tweet, and is generally used to reply to another user's tweet or to send them a specific message. By placing the username at the start of your tweet (e.g., with no text before it), the tweet will only be visible to the user you've replied to, or anyone who follows both you and the other user. So if you want your tweet to be widely seen, include the username within your tweet but not at the beginning.
- @mention the journal – all Future Science Group journals have a Twitter account (see below), and we'll be sure to re-tweet you if you mention us!
- Ask your co-authors to re-tweet your message, to spread the word further to their networks. You can also encourage people to re-tweet in your tweet itself!

Facebook

You may think that Facebook is just for personal use, but it can be a great tool to spread the word about your article. Post information about your article on your own profile, add depth to your information along with a related image, and a link to the article. Don't forget to copy in @futuresciencegroup so we can 'like' and share it as well.

LinkedIn

Future Science Group and many of the individual journals have their own LinkedIn groups. We encourage authors to join these groups, and post about their article (or any other topics they think would be of interest to the group members).

Journal social media sites

Journal	Twitter	LinkedIn group
Bioanalysis	https://twitter.com/@fsgbio	Bioanalysis – the Journal, the Zone https://www.linkedin.com/groups/2819540
BioTechniques	https://twitter.com/@MyBioTechniques	https://www.linkedin.com/company/biotechniques/
Future Drug Discovery	https://twitter.com/fsdrugdiscovery	Future Science Group https://www.linkedin.com/groups/2105775
Future Medicinal Chemistry	https://twitter.com/@fsgfmc	Future Medicinal Chemistry https://www.linkedin.com/groups/2822505
Future Science OA	https://twitter.com/@fsgfsoa	Future Science: Open Access https://www.linkedin.com/groups/8575275
Int. J. Pharmacokinetics	https://twitter.com/@fsgipk	Future Science Group https://www.linkedin.com/groups/2105775
Pharmaceutical Patent Analyst	https://twitter.com/@fsgppa	Pharmaceutical Patent Analyst https://www.linkedin.com/groups/3973811
Therapeutic Delivery	https://twitter.com/@fsgtde	Therapeutic Delivery https://www.linkedin.com/groups/3479647

Press releases

Issuing a press release describing a new publication can help the article reach a wider potential audience and leads to increased articles views. We encourage authors to consider a press release to describe their article. If you are interested in this option, please let us know. We can provide assistance writing and distributing the press release as required.

Sharing on an FSG specialist website

Many Future Science Group journals are partnered with an associated specialist website; an online community offering medical professionals easy access to breaking news, peer-reviewed articles and multimedia content. For a fee, your article can be featured on the journal's partner site, and made exclusively accessible to site's registered members. The article abstract will be hosted on the site, with a direct link to the article PDF featured on the homepage, shared via social media and highlighted in the website's weekly newsletter. This will automatically ensure your article reaches its target audience, helping to increase its readership and extend its impact. If you are interested in finding out more about this option, as well as other options for reaching your target audience via our digital sites, please contact Joanne Walker for further information.

Article metrics

Various article metrics are available on an article's page on our website, including download numbers and citations, and information from Altmetric and Dimensions.

Altmetric

All Future Science Group articles are tracked by Altmetric, with each article receiving an Altmetric score reflecting the quantity and reach of the attention it has received. Click on this score on each article page to find out more about how much and where an article is being talked about! For more information on Altmetric, go to https://www.altmetric.com.

Dimensions

Information from the Dimensions platform can be viewed alongside articles, including citation information. Click on the Dimensions badge on the article page to find out more information.

Editorial policies

The Future Science Group cross-journal Editorial Policies can be viewed and downloaded from our website here.