UNIVERSIDADE D
COIMBRA

Elsa Mariana Veiga Morgado

Relatórios de Estágio e Monografia intitulada "Pharmacokinetic Monitorization of Monoclonal Antibodies: The example of Infliximab in Inflammatory Bowel Diseases" referentes à Unidade Curricular "Estágio", sob a orientação do Dr. Paulo Jorge da Silva Monteiro, da Doutora Lígia Maria de Sousa Ferreira e da Professora Doutora Ana Cristina Bairrada Fortuna, apresentados à Faculdade de Farmácia da Universidade de Coimbra, para apreciação na prestação de provas públicas de Mestrado Integrado em Ciências Farmacêuticas.

Setembro de 2021

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Setembro 2021

Eu, Elsa Mariana Veiga Morgado, estudante do Mestrado Integrado em Ciências Farmacêuticas, com o n.º 2016250086, declaro assumir toda a responsabilidade pelo conteúdo do Documento Relatórios de Estágio e Monografia intitulada "Pharmacokinetic Monitorization of Monoclonal Antibodies: The example of Infliximab in Inflammatory Bowel Diseases" apresentados à Faculdade de Farmácia da Universidade de Coimbra, no âmbito da unidade de Estágio Curricular.

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Coimbra, 02 de setembro de 2021.

Elsa Mariana Veiga Morgado

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Parte I

Relatório de Estágio em Farmácia Comunitária

Farmácia
São José

Sob orientação do Dr. Paulo Monteiro

Resumo

Este relatório, tendo por base uma análise SWOT e apresentação de três casos clínicos, pretende explanar e analisar o Estágio Curricular realizado na Farmácia São José em Coimbra, com início a 11 de janeiro de 2021 e término a 30 de abril de 2021.

Palavras-chave: Relatório, Estágio Curricular, Farmácia São José.

Abstract

This report, based on a SWOT analysis and presentation of clinical cases, intends to explain and analyze the Curricular Internship carried out at the São José Pharmacy in Coimbra, starting on January 11th, 2021 and ending on April 30th, 2021.

Keywords: Report, Curricular Internship, São José Pharmacy.

Abreviaturas

COVID-19 - *Coronavirus Disease 2019*

FSJ - Farmácia São José

MICF - Mestrado Integrado em Ciências Farmacêuticas

MNSRM - Medicamentos Não Sujeitos a Receita Médica

MSRM - Medicamentos Sujeitos a Receita Médica

PVF - Preço de Venda à Farmácia

PVP - Preço de Venda ao Público

SARS-CoV-2 - *Severe Acute Respiratory Syndrome Coronavirus 2*

SWOT - *Strengths, Weaknesses, Opportunities and Threats*

1. Introdução

O farmacêutico é um agente da saúde pública que preza pela promoção da saúde, informação e uso racional do medicamento, exercendo as suas funções com inteira autonomia técnica e científica[1,2].

De acordo com o Artº 44º, n.º 2 da Diretiva 2013/55/UE, do Parlamento Europeu e do Conselho de 20 de novembro de 2013, e com o postulado no plano de estudos do Mestrado Integrado em Ciências Farmacêuticas (MICF), a conclusão e obtenção do grau de Mestre em Ciências Farmacêuticas implica uma formação de, pelo menos, cinco anos, com obrigatoriedade de estágio em Farmácia Comunitária[3]. Posto isto, ciente da importância do Estágio Curricular na consolidação dos conhecimentos teóricos adquiridos, bem como na capacitação e munição de *skills* para a prática da profissão, realizei o Estágio na área de Farmácia Comunitária, com duração de quatro meses (670 horas), na Farmácia São José (FSJ), sob a orientação do Diretor Técnico, Dr. Paulo Monteiro.

2. Farmácia São José

A FSJ situa-se na Avenida Calouste Gulbenkian, em Coimbra, próxima de várias instituições e entidades de Saúde Públicas e Privadas, sendo a sua localização uma mais-valia para a prestação de cuidados, informações e cedência de medicamentos aos utentes que a estes centros recorrem.

O horário de funcionamento da FSJ é das 8h30min às 21h de segunda a sábado, tendo ainda dias de serviço permanente (24 horas). A Farmácia está dividida em dois pisos. O piso inferior conta com dez balcões de atendimento e dois gabinetes que auxiliam na prestação de serviços farmacêuticos, como a medição da pressão arterial, colesterol, administração de injetáveis ou consultas. O piso superior está afeto às funções de gestão e aprovisionamento, conta com um robot, zona de receção de encomendas, laboratório e sala de reuniões. A equipa é constituída por onze farmacêuticos, quatro técnicos de farmácia e três técnicos auxiliares. A excelência e profissionalismo da equipa, a modernidade das instalações, aliadas à vasta variedade de produtos fazem desta farmácia uma referência na cidade de Coimbra.

3. Análise SWOT

O presente relatório tem como intuito a realização de uma análise SWOT (*Strengths, Weaknesses, Opportunities and Threats*) **(Tabela 1)**, tendo em conta uma dimensão interna (Pontos Fortes e Fracos) e uma dimensão externa (Oportunidades e Ameaças).

Tabela 1 - Resumo dos pontos abordados na análise SWOT.

SWOT	
Pontos Fortes • Localização • Organização Interna • Sentido Pedagógico • Leque alargado de Produtos • Produção de Medicamentos Manipulados	**Pontos Fracos** • Conhecimentos insuficientes relativos a nomes comerciais e Medicamentos Sujeitos a Receita Médica • Insegurança em Receitas Manuais e Regimes Especiais de Comparticipação • Lacunas científicas sentidas
Oportunidades • Literacia em Saúde • Formações	**Ameaças** • Medicamentos esgotados • Relutância perante os Medicamentos Genéricos • Locais de venda de Medicamentos Não Sujeitos a Receita Médica

3.1. Pontos Fortes

3.1.1. Localização

A proximidade geográfica da FSJ ao Centro Hospitalar e Universitário de Coimbra, ao Instituto Português de Oncologia de Coimbra Francisco Gentil, à Maternidade Bissaya Barreto, ao Centro de Saúde de Celas, a consultórios privados, a Estabelecimentos de Ensino, bem como, de uma significativa zona residencial, conduz a uma ampla heterogeneidade de utentes e patologias, o que impõe uma necessidade de *stock* e sortido de medicamentos sujeitos a receita médica (MSRM), de medicamentos não sujeitos a receita médica (MNSRM) e outros produtos de saúde muito variados. A diversidade de situações estimula constantemente o sentido de aprendizagem.

3.1.2. Organização Interna

A FSJ pauta-se por uma organização interna coesa e consistente. Tendo em conta que todo o meu estágio decorreu durante a pandemia COVID-19 (*Coronavirus Disease 2019*) e mais especificamente, no período que se revelou como sendo dos mais críticos para o país, a FSJ desenvolveu um plano de contingência, cumprindo todas as regras legais estipuladas pelo Governo. Deste modo, a equipa estava dividida em dois turnos, os quais não se cruzavam. Havia protocolos de entrada e saída da Farmácia, bem como, regras de higiene e de troca de

máscara por períodos regulares. Dentro da Farmácia as tarefas e responsabilidades eram divididas pelos colaboradores, de modo a obter o melhor rendimento, contudo, sempre que necessário o espírito de equipa sobressaía para colmatar qualquer necessidade. A organização interna por turnos, que leva inevitavelmente a um menor número de funcionários nas horas de maior movimento, permitiu-me, desde cedo, ajudar ativamente quer no *back-office*, quer no atendimento, com supervisão e ajuda, sempre que necessário. Esta participação ativa contribuiu fortemente para a minha aprendizagem e evolução no que concerne ao sistema Sifarma®, para o entendimento do funcionamento da farmácia, para a forma como me devo dirigir e expressar com o utente e para a interligação de conhecimentos teóricos. Posto isto, a adaptação à FSJ revelou-se bastante fácil, dada toda a organização, simpatia e acolhimento por parte da equipa.

3.1.3. Sentido Pedagógico

O meu interesse pessoal em absorver toda a experiência, tirando dela o máximo de conhecimento técnico e científico, aliado ao ótimo sentido pedagógico e ensinamentos da equipa, culminaram numa interação muito profícua, onde vi todas as minhas dúvidas esclarecidas de forma exímia. A equipa procurou transmitir todos os conhecimentos que julgou importantes para a minha formação, o que complementou aqueles que adquiri durante os cinco anos do MICF. Ressalvo os fortes conhecimentos transmitidos relativos a uma boa gestão da Farmácia, onde foi explorado o nosso sentido crítico relativamente às mais vastas situações, como *stocks* mínimos e máximos, prazos de validade, gestão de devoluções, gestão de campanhas, bem como o pleno entendimento de uma fatura, com todos os seus componentes, preço de venda ao público (PVP), preço de venda à farmácia (PVF), descontos, etc. Relativamente ao atendimento, ressalvo a simpatia e gosto pela explicação de todos os segmentos de cosmética, exaltando as diferenças e semelhanças entre as linhas das diferentes marcas, bem como a explicação de todos os lineares como higiene oral, puericultura, suplementos ou dispositivos médicos. Aprendi ainda pela observação prática qual a postura do farmacêutico ao balcão, a comunicação mais correta e empática e a saber ouvir e escutar o utente. Estas explicações foram cruciais para que me sentisse mais confiante no atendimento e aconselhamento ao utente.

3.1.4. Leque alargado de Produtos

Um dos pontos fortes da FSJ, que representa também um dos mais profícuos na minha formação, foi o facto de possuir um vasto leque de produtos, marcas, laboratórios e referências. Esta abrangência contribuiu imensamente para o aumento do meu conhecimento sobre os mais variados produtos, permitindo-me contactar com quase todos os produtos

passíveis de serem vendidos numa farmácia. A FSJ é conhecida pelos utentes como a Farmácia que tem sempre tudo.

3.1.5. Produção de Medicamentos Manipulados

Embora a indústria Farmacêutica cubra grande parte das necessidades medicamentosas da população, o facto é que existem determinadas preparações para as quais não é rentável a produção em massa, do ponto de vista farmacoeconómico. A Farmácia Comunitária e o Farmacêutico vêm, deste modo, colmatar os nichos não ocupados pela indústria Farmacêutica, permitindo o ajuste e personalização da terapêutica a cada doente, a alternativa a casos de intolerância a excipientes específicos, a associação de substâncias não disponíveis no mercado, entre outros. Posto isto, e tendo em conta que o farmacêutico é o profissional mais habilitado e especializado na preparação de medicamentos manipulados, considero este serviço uma mais-valia para a valorização da profissão.

Dada a adjacência da FSJ a vários hospitais, clínicas e consultórios e, tendo em conta que nem todas as farmácias têm capacidade para assegurar este serviço, a FSJ conta com a preparação de inúmeros medicamentos manipulados. Tive a oportunidade de observar e ajudar na preparação de cápsulas de Minoxidil, xarope de Captopril, pomada de Hidroquinona, Dexaval® (cr) e Ketrel® (cr), entre outros. A FSJ recorre ao *Software Soft Galeno*® no auxílio da ficha de preparação do manipulado, que inclui as matérias-primas e quantidades a usar, o modo de preparação, os aparelhos necessários, o controlo de qualidade e o cálculo do PVP. O *software* automaticamente atualiza o *stock* dos produtos utilizados. As especialidades médicas para as quais se observava uma maior afluência a este serviço é a dermatologia e pediatria.

Relativamente à reconstituição de preparações extemporâneas, observei e também preparei o medicamento Menopur® (Menotropina), estimulante da ovulação e gonadotropinas para o tratamento da infertilidade.

Ressalvo a importância da Unidade Curricular de Farmácia Galénica para o pleno entendimento da teoria e prática inerente à produção de medicamentos manipulados.

3.2. Pontos Fracos

3.2.1. Conhecimentos insuficientes relativos a nomes comerciais e Medicamentos Sujeitos a Receita Médica

Um dos pontos menos positivos e a serem melhorados na minha *performance* foram os conhecimentos relativos a nomes comerciais e a necessidade ou não de receita médica para a

dispensa de medicamentos. Dada a diversidade e panóplia sempre crescente de novas moléculas e nomes comerciais, revelei alguma dificuldade, especialmente na questão de associar os nomes comerciais ao princípio ativo. Relativamente à necessidade ou não de receita médica, percebi também que estas questões não são imutáveis, havendo alteração deste estatuto. Sempre que sentia dificuldade recorria à equipa que prontamente me ajudava e elucidava, e o sistema informático Sifarma® era também útil para ver algumas dúvidas respondidas.

3.2.2. Insegurança em Receitas Manuais e Regimes Especiais de Comparticipação

Embora na Unidade Curricular de Farmacologia Geral tenhamos aprendido a interpretar uma receita médica manual e a entender as suas exigências, senti inicialmente bastante insegurança na cedência de medicamentos neste registo, precisando sempre de confirmar a medicação com um colaborador da farmácia. Este modelo de receitas exige bastante atenção na identificação do nome do medicamento e dosagem e na conferência da vinheta do prescritor, assinatura, identificação da exceção, validade, número de embalagens prescritas e assinatura do médico.

Os regimes excecionais de comparticipação, como a majoração do escalão de comparticipação implicam o cumprimento de condições específicas no ato da prescrição, como a patologia, a especialidade clínica do médico prescritor e a inclusão de menções à regulamentação do regime especial[4].

Deste modo, senti algumas inseguranças na validação de receitas médicas manuais associadas a estes regimes. Por exemplo, sucedeu-me atender um utente que trazia uma receita com a menção da portaria correta para a sua condição de saúde e para o medicamento que tomava, contudo não a poderia aceitar pelo facto de o médico prescritor não ter a especialidade médica exigida. Nestes casos, o encargo financeiro do doente é superior, ficando o Farmacêutico impotente de alterar a situação.

3.2.3. Lacunas científicas sentidas

Revelei alguma dificuldade no aconselhamento de situações muito específicas como afeções de pele, pelo facto de não ter experiência na observação destas situações. Com o avançar do estágio, fui observando o atendimento da equipa e percebendo algumas situações.

Outra das lacunas que experienciei, prendeu-se com os produtos de uso oftálmico, pois não obstante os princípios ativos tenham sido abordados em várias Unidades

Curriculares, não houve nenhuma que focasse especificamente a área de oftalmologia, que requer cuidados específicos.

Relativamente à venda de produtos de uso veterinário também senti necessidade de recorrer à ajuda de outros colaboradores. Embora a Unidade Curricular de Preparações de Uso Veterinário nos transmita muitos conhecimentos relativamente a esta área, senti que é um mercado muito vasto, revelando-se um ponto fraco na minha prestação do estágio.

3.3. Oportunidades

3.3.1. Literacia em Saúde

O farmacêutico comunitário tem um papel fundamental no aumento e disseminação da literacia em saúde na população. Este papel revelou-se fundamental no auxílio à gestão da pandemia COVID-19. Assim, tive oportunidade de transmitir informação fidedigna aos utentes, desde modos de transmissão, períodos de incubação, sintomatologia, medidas de prevenção e proteção e importância da vacinação. Dada a imensa informação passada pelos meios de comunicação social, vários utentes recorriam ao farmacêutico para ver as suas dúvidas esclarecidas. Devido às várias notícias que iam saindo sobre possíveis medicamentos que poderiam auxiliar no tratamento e profilaxia da COVID-19, foram vários os utentes que se dirigiram à farmácia para solicitar Ivermectina, um medicamento antiparasitário. Coube-me a mim e à equipa explicar que não havia consenso científico nem evidências suficientes que apoiassem a sua utilização, no âmbito da infeção causada pelo *severe acute respiratory syndrome coronavirus 2* (SARS-CoV-2)[5].

3.3.2. Formações

O início do meu Estágio Curricular em Farmácia Comunitária coincidiu com a implementação de um novo Sifarma®. Assim, prontamente me foram cedidas formações e plataformas exploratórias deste novo sistema operativo, que se revelaram fundamentais no desenvolver de todas as atividades dentro da Farmácia.

Durante os quatro meses de estágio, tive a oportunidade de assistir a inúmeras formações desde MNSRM, a suplementos alimentares e cosméticos. A oferta formativa que chegava à Farmácia era enorme por parte de diversos laboratórios e, desde logo, a equipa fomentou o nosso envolvimento em todas as formações que, dado o período pandémico, se realizavam todas virtualmente. Considero que estas formações são de máxima importância e pertinência, quer na introdução de novos produtos, quer em revisões e explicações globais dos segmentos das marcas. Neste último cenário, ressalvo os *webinars* dinamizados pelos

laboratórios Uriage e Filorga, que providenciaram formações a explicar todas as gamas, e inclusive casos clínicos, que me ajudaram a entender o objetivo de cada produto e a facilitar o cruzamento de linhas para alcançar, da melhor forma, todas as necessidades do utente.

Enumero então algumas das formações que frequentei: Produtos medicalizados Uriage, Cuidados cosméticos Uriage, Darphin (linha intral), Webinar FAMA VI – "Contraceção de Emergência - Casos Clínicos", dinamizado pela Gedeon Richter Portugal e BioActivo® Q10 Forte, promovido pela Pharma Nord. Por fim, foi-me também possibilitado a inscrição na plataforma Academia Cosmética Ativa *online*, onde pude realizar diversas formações sobre todas os produtos e gamas do grupo, bem como uma academia de vendas e diversos *webinars*.

3.4. Ameaças

3.4.1. Medicamentos esgotados

Uma realidade com que me deparei durante o meu estágio foi a de medicamentos esgotados. Grande parte das situações eram fáceis de resolver, optando por um medicamento equivalente, mas de outro laboratório. Contudo, esta questão nem sempre era bem compreendida pelo utente. Outras situações mais críticas prendiam-se com medicamentos para os quais não havia alternativa. Um exemplo de um medicamento imprescindível para os doentes com esquizofrenia era o Xeplion®. Este medicamento estava disponível pelo projeto Via Verde do medicamento, o qual consiste numa via excecional de aquisição dos medicamentos abrangidos, que pode ser ativada, com base numa receita médica válida, quando a Farmácia não tem *stock* do medicamento pretendido[6]. Contudo, mesmo utilizando esta via excecional, havia dificuldades na aquisição do medicamento, sendo necessário estar constantemente a fazer o pedido.

3.4.2. Relutância perante os Medicamentos Genéricos

O papel do farmacêutico é informar e aumentar a literacia em saúde dos utentes, contudo a falta de conhecimento, leva ainda muitos utentes a descredibilizar os Medicamentos Genéricos e a terem bastante relutância na sua compra. Nestas situações, a equipa prontamente explicava que os Medicamentos Genéricos são equivalentes aos de marca (mesma substância ativa, forma farmacêutica, dosagem e indicações terapêuticas), cumprindo as normas rígidas e especificações de qualidade exigidas pela Agência Europeia do Medicamento e a entidade regulamentar nacional, o INFARMED. A venda de Medicamentos Genéricos afigura-se como uma oportunidade de ganhos financeiros quer para a farmácia, quer para o utente.

3.4.3. Locais de venda de Medicamentos Não Sujeitos a Receita Médica

O crescente aumento de locais de venda de MNSRM constituiu uma concorrência forte para as farmácias, tendo em conta que as grandes superfícies conseguem margens e preços mais apelativos. Contudo, a Farmácia e o Farmacêutico têm como vantagem o forte conhecimento científico sobre os produtos, o que permite a diferenciação do atendimento.

Atualmente, assistimos a um progressivo aumento de *sites* na *internet* passíveis de venderem estes produtos. O que por um lado representa e acompanha o atual desenvolvimento tecnológico e preferências de um consumidor cada vez mais digital pode, por outro lado, ameaçar os locais de vendas físicos. Na minha opinião, têm que ser providenciados mecanismos que permitam prevalecer o aconselhamento farmacêutico, mesmo perante estas novas plataformas.

4. Casos Clínicos

Caso Clínico 1

Uma utente do sexo feminino, com cerca de 60 anos, dirige-se à farmácia e pede um creme para as hemorroidas. Apresento como sugestões, Faktu® pomada retal à base de cloridrato de cinchocaína (anestésico) e policresuleno (anti-séptico) e o Procto-Glyvenol® creme retal, uma associação de tribenosido (anti-inflamatório) e lidocaína (anestésico).

Contudo, refiro que as pomadas/cremes têm apenas um efeito anti-inflamatório e anestésico, sendo mais indicado a toma de Daflon® ou a associação de ambos. Daflon® é um medicamento venotrópico que vai diminuir a distensibilidade venosa e a estase venosa, diminuindo o risco de prolapso e fissura.

No decorrer da conversa, tento perceber se a utente sofre de obstipação ou fezes duras, que podem agravar o quadro de hemorroidas, aconselhando, nesse caso um laxante osmótico, como o Laevolac® xarope, e referindo as medidas não farmacológicas como a ingestão de fibras e fluídos.

Sugeri ainda lavar com água fria ou aplicar gelo em situações agudas. A utente agradeceu o aconselhamento e levou os medicamentos supracitados.

Caso Clínico 2

Uma utente do sexo feminino, com 23 anos, dirige-se a farmácia pedindo algo para parar a diarreia, referindo-se ao Imodium®. De imediato questionei se tinha outros sintomas associados, nomeadamente febre, vómitos, diarreia com sangue ou muco, para despistar um

possível cenário de gastroenterite, no qual seria contraindicado a cedência de Imodium® (loperamida). A utente negou ter outros sintomas associados, doenças ou medicação recorrente que pudesse causar um quadro de diarreia, referindo apenas tomar a pílula anticoncecional.

Como medidas não farmacológicas, recomendei a reposição de fluídos e eletrólitos (água) e aconselhei evitar a administração de fibras e leite ou produtos lácteos até 24 horas após a cura da diarreia.

Como medidas farmacológicas, sugeri Bi-Oral Suero® Solução Oral Morango para a manutenção do equilíbrio hidroeletrolítico, o qual contém ainda probióticos que ajudam ao equilíbrio da flora bacteriana do trato gastrointestinal. Relativamente à cedência de loperamida e, tendo em conta que a etiologia da diarreia não era conhecida, tomei a liberdade de apresentar à utente um novo dispositivo médico – *Lenodiar Adult*, dos Laboratórios Aboca. Este produto reduz as descargas de diarreia, devido a um complexo molecular vegetal de taninos e polifenóis - Actipan-P. Este complexo normaliza a consistência das fezes, e diminuiu a inflamação da mucosa intestinal, uma vez que forma uma película protetora com efeito barreira que limita o contacto da parede do intestino com microrganismos e agentes irritantes e tem uma ação antioxidante, diminuindo a irritação da mucosa[7]. É recomenda a toma de 2 cápsulas 2-3 vezes ao dia.

A utente aceitou a sugestão e levou os produtos mencionados. Por fim, alertei que caso tivesse um aumento da diarreia, febre ou fezes com sangue, que deveria consultar imediatamente o médico.

Caso Clínico 3

Uma utente do sexo feminino, com 30 anos, dirige-se à farmácia com uma dermatite de contacto alérgica na zona da axila, potencialmente motivada pelo uso de desodorizante. A utente refere ainda bastante prurido e comichão. A senhora traz uma embalagem de um corticosteroide tópico de potência elevada, o qual é sujeito a receita médica. Explicando a impossibilidade da cedência do medicamento, apresento como alternativa um outro corticosteroide tópico, a hidrocortisona, que é um medicamento não sujeito a receita médica de venda exclusiva em farmácia. Tendo presente o protocolo de dispensa[8], certifiquei-me que a utente cumpria os requisitos necessários para a dispensa, apresentava uma dermatite ligeira a moderada, não tinha comorbilidades, nem medicação concomitante e não estava grávida nem a amamentar. Sugeri a utilização de Pandermil® creme, 2 vezes por dia, em camadas finas na área afetada, com duração máxima de 7 dias, após os quais se não existissem melhoras, deveria

consultar um médico. Embora seja uma utilização de curta duração e um corticoide tópico de baixa potência, aconselhei a diminuir a posologia para uma aplicação 1 vez por dia, assim que notasse melhoras e, posteriormente, aplicar dia sim dia não, para fazer o desmame da hidrocortisona de forma correta.

Como a utente referia bastante prurido, expliquei que o mesmo pode ser consequência de uma secura extrema e que, por isso, era fundamental hidratar a zona afetada, devendo proceder à hidratação em associação com o corticoide tópico. A utente agradeceu o aconselhamento e levou os produtos mencionados.

5. Conclusão

Na minha opinião, o Estágio Curricular em Farmácia Comunitária é extremamente importante, pois o estagiário tem a oportunidade de fazer o cruzamento teórico das várias Unidades Curriculares lecionadas no MICF, adquirir novas competências técnicas e científicas e sentir o peso da responsabilidade que a profissão acarreta. Poder ajudar o outro é, sem dúvida, algo muito gratificante, e as Farmácias Comunitárias permitem uma maior proximidade ao utente, estando ao nosso alcance, a cada atendimento, fazer a diferença na vida daquela pessoa.

Estou muito grata à FSJ e a toda a equipa por todos os ensinamentos transmitidos, por estimularem a minha autonomia e por, sem dúvida, me terem preparado para ingressar no mercado de trabalho.

Por fim, e fazendo uma análise crítica do setor, sinto que o farmacêutico comunitário poderia ter ainda um papel mais preponderante na saúde dos portugueses. O farmacêutico é o profissional mais habilitado a fazer revisão da medicação, acompanhamento farmacoterapêutico, e ainda ajudar na gestão e controlo de doenças crónicas, como hipertensão arterial, Diabetes *mellitus*, entre outros. Embora muitas farmácias já tenham estes serviços, considero que há ainda um longo caminho para a sua disseminação e plena aceitação. Para tal, seria fundamental trabalhar em rede com o Serviço Nacional de Saúde, tendo por exemplo acesso ao perfil clínico do doente, para que o farmacêutico pudesse trabalhar de forma ainda mais estreita com o médico e outros profissionais de saúde. Na minha opinião, muitas consultas de acompanhamento da hipertensão arterial e diabetes poderiam ser feitas na farmácia, em articulação com o médico de família, o que ajudaria a diminuir a carga dos Centros de Saúde.

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Parte II

Relatório de Estágio em Indústria Farmacêutica

Departamento Médico e Científico

Sob orientação da Doutora Lígia Ferreira

Resumo

Este relatório, tendo por base uma análise SWOT, pretende explanar e analisar o Estágio Curricular realizado na Consultora Farmacêutica Owlpharma - Consulting, Lda. (Owlpharma), com início a 3 de maio de 2021 e término a 30 de julho de 2021.

Palavras-chave: Relatório, Estágio Curricular, Owlpharma.

Abstract

This report, based on a SWOT analysis intends to explain and analyze the Curricular Internship carried out at the Owlpharma - Consulting, Lda., starting on May 3rd, 2021 and ending on Jully 30th, 2021.

Keywords: Report, Curricular Internship, Owlpharma.

Abreviaturas

COVID-19 - *Coronavirus Disease 2019*

CSI - *Core Safety Information*

CTD - *Common Technical Document*

FI - Folheto Informativo

MICF - Mestrado Integrado em Ciências Farmacêuticas

PSURs - *Periodic Safety Update Reports*

QRD - *Quality Review of Documents*

RCM - Resumo das Características do Medicamento

SWOT - *Strengths, Weaknesses, Opportunities and Threats*

1. Introdução

A Faculdade de Farmácia da Universidade de Coimbra propicia aos seus estudantes a possibilidade de realizarem o Estágio Curricular na área de Indústria Farmacêutica para além do Estágio em Farmácia Comunitária, sendo, por isso, um elemento distintivo que capacita os seus alunos para uma melhor adaptação ao mundo profissional futuro. Ciente desta oportunidade, optei por estagiar na Owlpharma - Consulting, Lda. (Owlpharma), no Departamento Médico e Científico, sob orientação da Doutora Lígia Ferreira, durante o período de três meses (3 de maio de 2021 a 30 de julho de 2021).

Em consequência da pandemia COVID-19 (*Coronavirus Disease 2019)* e, dada a criticidade da situação epidemiológica vivida em Portugal, a obrigatoriedade de teletrabalho vigorou durante este período de estágio. Para tal foi me cedido um computador, com ligação à rede da empresa.

O presente relatório visa analisar e explanar a minha prestação e experiência no Estágio Curricular desenvolvido na Consultora Farmacêutica Owlpharma, por meio de uma análise SWOT (*Strengths, Weaknesses, Opportunities and Threats*).

2. Owlpharma

A Owlpharma, empresa de Consultoria Farmacêutica, criada em 2013, iniciou o seu percurso com sede no Instituto Pedro Nunes em Coimbra. Atualmente, tem escritórios na Avenida da Guarda Inglesa, em Coimbra e na Rua Padre Américo, em Lisboa.

A Owlpharma tem quatro áreas de atuação: Garantia de Qualidade, Assuntos Regulamentares, Assuntos Médicos e Científicos e Farmacovigilância, acompanhando, deste modo, todo o ciclo de vida do medicamento. Com uma equipa altamente qualificada, jovem e dinâmica, conta com mais de 40 colaboradores entre os quais 38 farmacêuticos. Embora criada apenas em 2013, a evolução e crescimento da Owlpharma são notórios, acumulando prémios e consolidando-se como uma referência a nível nacional e internacional.

3. Análise SWOT

O presente relatório tem como intuito a realização de uma análise SWOT **(Tabela 1)**. Deste modo irei enumerar e fundamentar os Pontos Fortes (*Strenghs*), Pontos Fracos (*Weaknesses*), Oportunidades (*Opportunities*) e Ameaças (*Threats*), procurando refletir sobre as atividades desenvolvidas, competências adquiridas e as dificuldades sentidas no decurso dos 3 meses de estágio na Owlpharma.

Tabela I - Resumo dos pontos abordados na análise SWOT.

SWOT	
Pontos Fortes • Integração na equipa • Formações Internas • Variedade de tarefas realizadas • Apoio constante	**Pontos Fracos** • Inseguranças iniciais • Escasso conhecimento de ferramentas avançadas de *Microsoft Excel* • Não contacto com o Departamento de Farmacovigilância
Oportunidades • Contacto com o Departamento de Assuntos Regulamentares • Desenvolvimento de competências • Contacto com a realidade do setor	**Ameaças** • Pandemia COVID-19

3.1. Pontos Fortes

3.1.1. Integração na equipa

A integração na Owlpharma revelou-se bastante fácil, dado todo o esquema organizacional, simpatia e acolhimento da equipa. No primeiro dia, os diretores de departamento e diretores gerais fizeram questão de se dirigirem aos escritórios para receberem os estagiários. A apresentação foi iniciada com a história, evolução e organização da Owlpharma levadas a cabo pelos diretores desta, culminando com formações iniciais de algumas das áreas de atuação da mesma. Foram ministradas formações referentes a bases iniciais de Assuntos Regulamentares, introdução à Farmacovigilância e elaboração de textos científicos. Algumas das formações eram seguidas de uma breve avaliação de aferição dos conhecimentos. Estas formações revelaram-se muito profícuas e pertinentes para o pleno entendimento das tarefas subsequentes.

3.1.2. Formações Internas

As formações internas desenvolvidas pelos colaboradores da Owlpharma eram uma constante, o que, para mim, se afigurou como uma mais-valia, para aprofundar os meus conhecimentos e reavivar certos conteúdos lecionados durante o Mestrado Integrado em Ciências Farmacêuticas (MICF). São exemplos, a formação em *Core Safety Information* (CSI), Alterações de Segurança, Pesquisa de Literatura, Boas Práticas de Farmacovigilância, aplicação

de Testes de Legibilidade e *Quality Management System*. Previamente à realização de uma tarefa, era prática comum receber formação para o pleno entendimento da mesma e das suas especificidades inerentes.

Uma vez que a empresa tem como lema a formação contínua e a excelência formativa dos seus colaboradores, todos estes são motivados e encorajados a realizarem formações em áreas do saber que considerem que acrescentam valor ao seu trabalho, havendo posteriormente uma partilha de conhecimentos. Esta filosofia denota um grande sentido pedagógico e valorização constante de conhecimentos e formação.

3.1.3. Variedade de tarefas realizadas

A Doutora Lígia Ferreira, como tutora e orientadora de estágio, procurou sempre proporcionar-me uma experiência abrangente, para que pudesse ter contacto com diferentes tarefas.

O Departamento Médico e Científico é responsável pela escrita e elaboração da parte não clínica (módulos 2.4, 2.6 e 4) e parte clínica (módulos 2.5, 2.7 e 5) do *Common Techinical Document* (CTD), pela elaboração e revisão científica do Resumo das Características do Medicamento (RCM) e do Folheto Informativo (FI), testes de legibilidade, revisão de materiais promocionais, preparação de resumos, pósteres e apresentações, presença e relatórios em *Advisory Boards* e, ainda relativamente à toxicologia, a redação de relatórios toxicológicos, relatórios de avaliação de risco ambiental, relatórios de avaliação de risco de impurezas, entre outros. O departamento é também responsável pela elaboração de documentos de farmacovigilância, nomeadamente *Periodic Safety Update Reports (*PSURs) e *Risk Management Plans*.

Deste vasto leque de tarefas e âmbito do Departamento Médico e Científico, realizei vários PSURs. Os PSURs apresentam uma análise crítica da relação benefício-risco do produto em questão, tendo em consideração informações de segurança novas ou emergentes. Novas informações de segurança podem determinar investigações sobre um determinado efeito adverso ou interação e despoletar ações com vista a proteger a saúde pública, como por exemplo, a atualização das informações fornecidas aos profissionais de saúde e doentes[1].

Realizei também um quadro resumo de todos os estudos clínicos utilizados no módulo 5 do CTD (5.2 *Tubular listing of all clinical studies*) e ainda os módulos 4.3 e 5.4, que dizem respeito às referências bibliográficas. Auxiliei em trabalhos de tradução de FI, em testes de legibilidade e ainda num relatório de *Advisor Board*.

Os testes de legibilidade têm como objetivo provar que o FI é legível e compreendido pela população em geral. A população alvo de estudo tem de ter mais de dezoito anos, não

ter respondido a um questionário nos últimos seis meses, não tomar o medicamento em questão e não ser profissional de saúde. A amostra deve primar pela heterogeneidade de idades, géneros e habilitações literárias, de modo a ser o mais representativa possível. Os questionários devem ter entre doze a quinze perguntas, nos quais o participante deve responder com base na informação constante no FI. É necessária uma ronda piloto (4 a 10 participantes) para avaliar a compreensibilidade das questões e fazer as alterações necessárias, seguida de duas rondas com dez participantes cada. Dos vinte participantes, dezoito (90 %) têm de encontrar a informação e destes, 90 %, dezasseis participantes têm de a compreender.

Por fim, contribuí para a sistematização de informação científica que possa suportar uma nova posologia de um medicamento, com vista numa maior *compliance* pelo utente.

Todas estas tarefas contribuíram fortemente para enriquecer o meu conhecimento e entendimento deste setor.

3.1.4. Apoio constante

Ainda que o estágio se tenha realizado totalmente de forma remota, o apoio, colaboração e proximidade foram constantes. Fazendo recurso de plataformas digitais, a equipa ia comunicando, fazendo a passagem das tarefas e a sua explicação. Sempre que sentia necessidade ou dúvidas recorria a essa plataforma, onde, prontamente, me elucidavam quer por mensagem, quer por meio de videochamada. Ressalvo, ainda, as reuniões semanais do departamento, que permitiam a divisão de tarefas e a comunicação entre todo o departamento. Durante o período de estágio, a equipa depositou sempre bastante confiança e autonomia no estagiário, o que considero bastante positivo. Sendo o estágio realizado remotamente, o *feedback,* após a conclusão de uma tarefa, afigurava-se como um elemento crucial e motivador para o prosseguimento do trabalho.

3.2. Pontos Fracos

3.2.1. Inseguranças iniciais

Apesar de ter os conhecimentos base fornecidos pelo MICF, inicialmente senti alguma insegurança perante o preenchimento dos documentos, por ser algo novo que nunca tinha contactado. A área regulamentar é alicerçada por uma legislação e burocracia muito rígidas, com uma variedade de *Guidelines* e documentos oficiais que devem ser totalmente compreendidos, para o pleno entendimento das funções. Um dos pontos menos positivos da minha prestação e que devo colmatar, é o investimento na familiarização com os documentos supracitados. Contudo, as formações e apoio da equipa foram fundamentais para ter as ferramentas necessárias para a execução das tarefas.

3.2.2. Escasso conhecimento de ferramentas avançadas de *Microsoft Excel*

Um dos pontos menos positivos da minha *performance* foram os escassos conhecimentos das ferramentas avançadas de *Microsoft Excel*. Ciente da importância desta ferramenta de trabalho é, sem dúvida, uma lacuna que ambiciono colmatar, com posteriores formações.

3.2.3. Não contacto com o Departamento de Farmacovigilância

Devido ao volume de trabalho do Departamento Médico e Científico e às necessidades do Departamento de Farmacovigilância não surgiu oportunidade de realizar tarefas neste setor. No entanto, tive oportunidade de assistir a algumas formações do Departamento de Farmacovigilância e preparei vários documentos de farmacovigilância, como os PSURs.

3.3. Oportunidades

3.3.1. Contacto com o Departamento de Assuntos Regulamentares

O contacto com o Departamento de Assuntos Regulamentares configurou uma oportunidade para aumentar os meus conhecimentos nesta área do saber. Neste tive possibilidade de realizar a verificação de vários CSI e uma harmonização. Os CSI contêm toda a informação de segurança base de uma substância ativa ou combinação de substâncias ativas que deve constar no RCM. O objetivo é comparar os dois documentos (CSI e RCM) e certificar que toda a informação do CSI aplicável ao produto se encontra no RCM.

A harmonização visa comparar a informação do RCM e FI do Medicamento de Referência com o RCM e FI do Medicamento Genérico e ainda certificar que são seguidas as regras e orientações do *Template Quality Review of Documents* (QRD).

3.3.2. Desenvolvimento de competências

Para além da aplicação dos conhecimentos teóricos em contexto real, durante os meus três meses de estágio desenvolvi capacidades linguísticas, informáticas e métodos de trabalho.

A maioria dos documentos são redigidos na língua inglesa, o que me permitiu diariamente contactar com esta língua e aprofundar as minhas competências linguísticas, especialmente em vocabulário técnico-científico.

3.3.3. Contacto com a realidade do setor

Sendo a Owlpharma uma Consultora Farmacêutica que trabalha com vários clientes, permitiu-me contactar com uma ampla diversidade de grupos farmacoterapêuticos de

medicamentos, com indústrias farmacêuticas nacionais e internacionais e, por isso, ter uma noção da abrangência do mercado farmacêutico.

3.4. Ameaças

3.4.1. Pandemia COVID-19

Como já referido, o Estágio Curricular na Owlpharma decorreu exclusivamente em regime teletrabalho, o que retira a oportunidade de experienciar o dia-a-dia num escritório de uma Consultora Farmacêutica. Considero ter desenvolvido uma maior autonomia em consequência desta situação e ressalvo, mais uma vez, a forte comunicação entre todo o departamento, com recurso a videochamadas e grupos de conversação.

4. Conclusão

A possibilidade de executar um Estágio Curricular na área da Indústria Farmacêutica constitui uma mais-valia para os estudantes da Faculdade de Farmácia da Universidade de Coimbra, o que, indubitavelmente, os prepara melhor para o mercado de trabalho futuro.

As Unidades Curriculares de Assuntos Regulamentares do Medicamento e Farmacovigilância e Farmacoepidemiologia são cruciais para o entendimento desta área e setor, permitindo que o aluno ingresse no plano de estágio com as aptidões e conhecimentos essenciais. Contudo, dado o tempo limitado para as lecionar, é impossível cursar todos os conhecimentos e, sem dúvida, que o Estágio Curricular é uma mais-valia para absorver conhecimentos novos, interligar bases teóricas e permitir a sua execução prática.

O balanço que faço do estágio desenvolvido na Owlpharma é muito positivo, tendo sido uma experiência muito enriquecedora, que complementou e enriqueceu o meu plano curricular. Durante os três meses de estágio, tive oportunidade de percecionar e ter uma noção global da área de atuação de uma consultora farmacêutica, bem como, adquirir competências científicas, informáticas e linguísticas.

Sem dúvida que a área de atuação do Farmacêutico é muito vasta, graças a um plano curricular abrangente. Contudo, o rigor, a qualidade, a segurança e a eficácia são sempre pilares basilares que movem a conduta do Farmacêutico, o qual centra toda a sua atuação no doente, contribuindo assim para a promoção da saúde, informação e uso racional do medicamento.

Um muito obrigado a toda a equipa Owlpharma.

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Parte III

Monografia

"Pharmacokinetic Monitorization of Monoclonal Antibodies: The example of Infliximab in Inflammatory Bowel Diseases"

FACULDADE DE FARMÁCIA
UNIVERSIDADE D
COIMBRA

Sob orientação da Professora
Doutora Ana Cristina Bairrada Fortuna

Resumo

Os anticorpos monoclonais (mAbs) revolucionaram o tratamento das doenças inflamatórias intestinais. Ainda assim, existe uma significativa variabilidade farmacocinética na resposta a estes fármacos, levando a uma percentagem considerável de não respondedores primários e secundários. A perda de resposta aos mAbs pode ser explicada pelas baixas concentrações séricas de fármaco, formação de anticorpos contra o fármaco, entre outras causas, que, em conjunto, serão explanadas nesta monografia. Este trabalho inicia-se com a compreensão da farmacocinética dos mAbs de forma a identificar os fatores que a determinam. Neste contexto, será ainda salientada a importância da monitorização terapêutica dos mAbs e analisada a relevância da monitorização terapêutica reativa e proativa de mAbs, focando particularmente o Infliximab no tratamento de doenças inflamatórias intestinais.

Palavras-chave: Monitorização Terapêutica de Fármacos, Anticorpos Monoclonais, Infliximab, Doenças Inflamatórias Intestinais, Doença de Crohn, Colite Ulcerosa, Anticorpos Anti-Fármaco, Otimização, Limiar Terapêutico.

Abstract

Monoclonal antibodies (mAbs) have revolutionized the management of inflammatory bowel diseases (IBD). However, there is a significant variability in the pharmacokinetics and response to mAbs, leading to a considerable percentage of primary or secondary non-responders. The loss of response to mAbs can be linked to low drug serum concentration, formation of anti-drug antibodies (ADAS), and other causes, explained in this monography. Firstly, a comprehensive review of mAbs pharmacokinetics will be herein addressed to identify the underlying factors. In this context, the importance of therapeutic drug monitoring (TDM) of mAbs will be herein emphasized and the relevance of reactive and proactive TDM of mAbs will be also analysed, focusing particularly the Infliximab on IBD.

Keywords: Therapeutic Drug Monitoring, Monoclonal antibodies, Infliximab, Inflammatory Bowel Diseases, Crohn's disease, Ulcerative Colitis, Anti-Drug Antibodies, Optimization, Therapeutics threshold.

Abbreviations

ADAS - Anti-Drug Antibodies

ADCC - Antibody Dependent Cellular Cytotoxicity

AGA - American Gastroenterological Association

ATI - Antibodies to Infliximab

BMI - Body Mass Index

BSA - Body Surface Area

CD - Crohn's Disease

CDAI - Crohn's Disease Activity Index

C_H - Constant domains of Heavy chains

CL - Clearance

C_L - Constant domains of Light chains

CRP - C - Reactive protein

ECCO - European Crohn's and Colitis Organisation

ELISA - Solid Phase Enzyme linked Immunosorbent Assay

ESPGHAN - European Society for Paediatric Gastroenterology Hepatology and Nutrition

Fab - Fragment Antigen Binding

Fc - Fragment Crystallizable

FcRn - Neonatal Fc receptor

FcγR - Fc-gamma-receptor

HBI - Harvey Bradshaw Index

HMSA - Homogenous Mobility Shift Assay

HPLC - High-Pressure Liquid Chromatography

i.m. - Intramuscular

i.v. - Intravenous

IBD - Inflammatory Bowel Diseases

IFX - Infliximab

IgG - Immunoglobulin G

mAbs - Monoclonal Antibodies

mTNF - Membrane-bound TNF

POC - Point of Care Testing

QALY - Quality-Adjusted Life-Year

RIA - Radio-Immunoassay

s.c. - Subcutaneous

sTNF - Soluble Tumour Necrosis Factor

TC - Trough Concentrations

TDM - Therapeutic Drug Monitoring

Tmax - Time of the Maximum Concentration

TMDD - Target-Mediated Drug Disposition

TNF - Tumour Necrosis Factor

TNFR1 - Tumour Necrosis Factor Receptor 1

TNFR2 - Tumour Necrosis Factor Receptor 2

TNF-α - Tumour Necrosis Factor Alpha

UC - Ulcerative Colitis

Vd - Volume of Distribution

V_H - Variable domains of Heavy chains

V_L - Variable domain of Light chains

VNTRs - Variable Number of Tandem Repeats

1. Introduction

Monoclonal antibodies (mAbs) are 150-kD glycoproteins based on the structure of physiological Immunoglobulin G (IgG). The mAbs are composed by four polypeptide chains: two heavy chains (50 kDa), and two light chains (25 kDa), helded by disulfide bonds (**Figure 1**)[1,2].

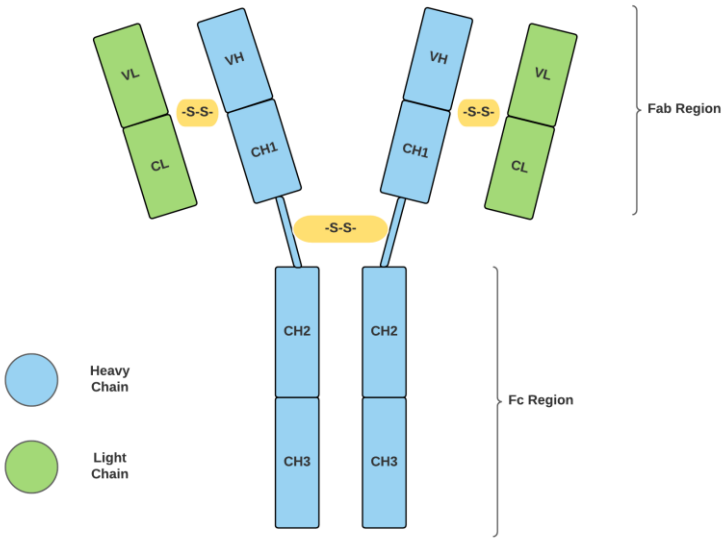

Figure 1 - Structure of mAbs. CH, Constant domains of Heavy chains; CL, Constant domains of Light chains; Fab, Fragment Antigen Binding; Fc, Fragment Crystallizable; -S-S, Dissulfide Bonds; VH, Variable domains of Heavy chains; VL, Variable domains of Light chains.

The mAb structure has constant domains of heavy chains (C_H) and constant domains of light chains (C_L). In the same way, there are variable domains of heavy chains (V_H) and variable domains of light chains (V_L) (**Figure 1**)[2].

V_H and V_L change according to the target antigen and constitute the antigen binding-site. V_H, V_L and the C_{H1} domains form the fragment antigen binding (Fab). C_{H2} and C_{H3} constitute the antibody's fragment crystallizable (Fc) region that can bind to a diversity of cell surface receptors, such as the Fc-gamma-receptor (*FcγR*), the neonatal Fc receptor (*FcRn*) and the components of the complement system[2].

IgG1, IgG2, IgG3, and IgG4 are four subclasses of IgG that can all neutralize target antigens. IgG1 and IgG3 are often powerful effector mechanism triggers, while IgG2 and IgG4 elicit more modest responses. The IgG1 subclass comprises the majority of the available therapeutic mAbs[2].

According to the therapeutic indications, the two major classes of mAbs are the anti-inflammatory mAbs for autoimmune diseases [such as rheumatoid arthritis, spondylarthropathies, psoriatic arthritis and inflammatory bowel diseases (IBD)] and the

antitumor mAbs (solid tumours and haematological malignancies). However, there are mAbs that treat clinical dysfunctions like heterozygous familial hypercholesterolemia, transplant rejection, paroxysmal nocturnal haemoglobinuria and severe asthma. This emphasizes the potential of mAbs which, over the last few decades, have improved the treatment of several diseases[1].

Despite the proved efficacy of this pharmacotherapeutic class, an initial response to mAbs rates varies between 50–90 % and, in most patients, the initial response is lost throughout time, culminating in disease progression. The mechanism for losing therapeutic activity can be explained through therapeutic drug monitoring (TDM). TDM of mAbs guides a more effective individual dose to achieve disease remission and it has been considered as "the next dimension in personalized targeted mAb therapy"[1].

Since this work will focus on the importance of TDM of mAbs, their pharmacokinetics will be firstly discussed in the next section.

2. Pharmacokinetics of mAbs

2.1. Absorption

The oral bioavailability of mAbs is not appreciable due to their large molecular weight, low membrane permeability and limited stability in the presence of gastrointestinal proteases. Consequently, the most frequent route of administration of mAbs is intravenous (i.v.) infusion, followed by subcutaneous (s.c.) and intramuscular (i.m.) injections[2].

The s.c. administration of mAbs implies an absorption process from the injection site to the lymphatic system, draining, then, into the systemic circulation. This significantly depends on the convective transport of mAbs. With increased molecular weight, the percentage of mAbs recovery in lymphatic *versus* blood vessels increases. The flow in lymphatic vessels is very slow in comparison to the bloodstream within, therefore the absorption of mAbs into the systemic circulation is also slow, in case of s.c. and i.m. administrations. It is observed a gradual and slow increase in plasmatic concentration (1.7–13.5 days) and delayed time of the maximum concentration (Tmax) (6–8 days). Tmax is significantly influenced by lymphatic flow rate[2].

In s.c. administration, the bioavailability varies within the range of 52–80 % and mAbs can suffer presystemic elimination. Presystemic elimination is considered a blend of the effect of soluble peptidase activity in the interstitial space, endocytosis with lysosomal degradation and interaction with phagocytic immune cells in the lymph nodes. For instance, s.c.

bioavailability of rituximab was found to be inversely proportional to the dose, probably due to the saturation of the *FcRn* that protects proteins from lysosomal degradation at the absorption site and the lymphatic vessels[2].

Absorption depends on the characteristics of the injection site (e.g. pressure gradient in the interstitial space, catabolic capacity, blood flow at the injection site, *FcRn* affinity), and of the patient (e.g. body weight, gender, age, activity level, disease state, respiratory rate and blood pressure). For example, hypodermis thickness improves with body weight, reduces with age, and differs between gender. Meanwhile, in the case of 2 hours of exercise, the flow of lymph rises by 83 %, which can have a considerable impact on the serum concentrations of mAbs[2].

The appearance of more convenient delivery systems makes outpatient prefer the s.c. administration[3]. Adalimumab is an example of an anti-Tumour Necrosis Factor (TNF) agent with s.c. administration. The pre-filled syringe allows patients to do the administration at home[4].

2.2. Distribution

After attaining the bloodstream, the mAbs are distributed mainly in the central compartment, with its volume of 2-3 L, similar to plasma total water[2]. The total volume of distribution (Vd) is relatively small (5–10 L). This results from their difficulty to cross the cell membranes, as a consequence of their large molecular weight and hydrophilicity/polarity[5].

The extent of mAbs distribution relies on the extravasation process, the antibody binding, and clearance (CL) from the tissue (like intracellular uptake and degradation). mAbs extravasation, i.e., transition from vascular space to the interstitial tissue space, can transpire via three processes: convective transport (the main mechanism), transcytosis through vascular epithelial cells via the *FcRn,* and passive diffusion (which does not play a significant role). Transcytosis process may be an important extravasation route for mAbs when convection transport is restricted. In numerous studies, IgG transport has been demonstrated to be bidirectional, in both the basolateral to apical and apical to basolateral directions, which can be demonstrated by *FcRn* mecanism[2].

After extravasation, antibody distribution depends on its diffusion, convection, and affinity to target antigens. If there is no target antigen to bind to mAbs, or the target is in the plasma, it is likely that the distribution of mAbs should be limited. On the other hand, if mAbs have a target in the tissue compartment, then a greater Vd is expected[2].

2.3. Elimination

Antibodies are eliminated by metabolism (catabolism) or excretion. mAbs elimination occurs principally via intracellular catabolism by lysosomal degradation to amino acids. Except in pathologic situations, mAbs are not excreted in the urine, due to its high molecular weight. Biliary elimination accounts for only a minor portion of mAb elimination[2].

There are two different catabolic pathways: 1) *FcRn* - nonspecific elimination by interaction between the Fc region of the antibody and the Fc receptor and 2) target-mediated drug disposition (TMDD) through specific interaction among the Fab region of the antibody and its specific target epitope. TMDD leads to endocytosis internalization of mAbs into a vesicle and subsequent lysosomal degradation[2,5].

The elimination rate by TMDD is determined by the target receptor's expression (which is usually limited), the affinity of mAbs for their target, mAbs dose, the rate of mAbs internalization, and the rate of catabolism inside the target cell. Due to the high specificity and affinity binding of mAbs for its target, TMDD is a major route of elimination, particularly at low doses and concentrations. In case of higher doses, the elimination of mAbs by TMDD is frequently saturated due to the limited target receptors, with a poor influence in the total CL of mAbs[2]. In this situation the *FcRn* elimination becomes dominant[6].TMDD exhibit a dose-dependent elimination.

In general, mAbs targeting cell-surface receptors have nonlinear CL that is influenced by antigen expression, while mAbs targeting soluble antigens have dose-proportional behaviour with linear CL[5]. TMDD have a nonlinear pharmacokinetic, because it is more frequent to target membrane receptors[6].

The binding of Fc domain to *FcγR*, expressed on numerous immune cells, such as monocytes, macrophages, myeloid progenitor cells, and dendritic cells, is a receptor-mediated endocytosis of mAbs. The endocytosis of the complex is followed by intracellular catabolism[2].

The pinocytosis does not differentiate which proteins are degraded, consequently, a protective mechanism for IgG is needed to preserve their concentrations in the plasma. Protected elimination by *FcRn*, which is also known as the Brambell receptor, is a common pathway shared by endogenous IgG and therapeutic mAbs. *FcRn* has a weak affinity for IgG at physiologic pH, but, when the endosome is acidified, *FcRn* affinity increases. Once bound, the *FcRn-IgG* complex will be returned to the cell surface and, when physiologic pH is reached, the IgG molecule will be released from the binding. Proteins that are not linked to *FcRn* in the endosomes are degraded (**Figure 2**). *FcRn*-mediated recycling protects roughly two-thirds of

IgG molecules, involving therapeutic mAbs. Nonetheless, there is a limit to the *FcRn* recycling pathway. At high concentrations of IgG, either exogenously as at high dose i.v. immunoglobulin therapy or endogenously, as in multiple myeloma, the *FcRn* recycling process will be saturated, resulting in an increased CL of IgG and a shorter half-life. It is expected hypogammaglobulinemia to reduce CL and lengthen the half-life of therapeutic mAbs[2].

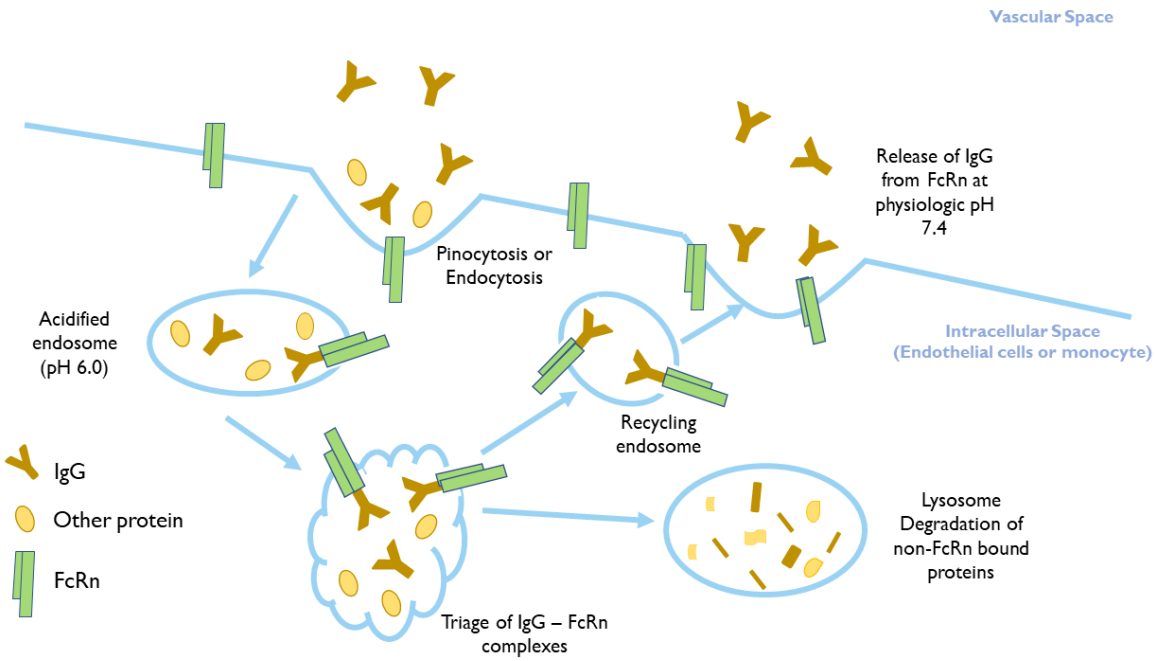

Figure 2 - Scheme of *FcRn* salvage pathway[2]. *FcRn*, Neonatal Fc receptor; IgG, Immunoglobulin G.

The binding of mAbs to the *FcRn* in endothelial cells is essential for the long half-life and low CL rate of mAbs. The humanized and fully human mAbs have improved *FcRn* affinity with subsequently longer half-lives[2].

An increase of mAb CL, which can be due to numerous factors, can entail trough concentrations (TC) to be below the minimum effective concentration[1].

3. Factors influencing mAbs' Pharmacokinetics

The interpatient variability regarding the pharmacokinetics of mAbs can involve immunogenicity factors, like the presence of anti-drug antibodies (ADAS), the amount of antigenic target burden, concomitant drugs, serum concentration of mAbs, patients body weight and body surface area (BSA), gender, serum albumin, alkaline phosphatase levels, C - reactive protein (CRP) and genetic variants. Generally, mAbs have a Vd approximately to the circulating plasma volume and a half-life of 3–4 weeks. However, all these factors can change Vd and half-life time with a huge impact on the efficacy of treatment (**Table I**). These factors will be explained and discussed below[1].

Table I - Factors that affects pharmacokinetics and respective impact.

Factors affecting Pharmacokinetics	Impact on Pharmacokinetics
Large target burden	↑ Cl
ADAS	↑Cl
↑IgG (*FcRn* saturation)	↑Cl (↓$T_{1/2}$)
Immunosuppressive	↑[mAb] ↓ ADAS
Low serum albumin	↑Cl
↑ Alkaline phosphatase	↑Cl
↑ CRP	↑Cl
↑ Metastatic sites	↑Cl
↑ BMI	↑Cl

ADAS, Anti-Drug Antibodies; BMI, Body Mass Index; CL, Clearance; FcRn, Neonatal Fc receptor; [mAb], Monoclonal Antibody Concentration; $T_{1/2,}$ Half-life time; ↓, Decrease; ↑, Increase.

3.1. Immunogenicity

Immunogenicity refers to the ability of a particular substance to induce an immunological response[2].

Patients who receive therapeutic mAbs (exogenous proteins) can develop an immune response that results in the formation of ADAS - endogenous antiglobulin against mAbs. The immunogenicity of mAbs depends on the structure and murine content of mAbs, immune status of the patient, concomitant use of immunosuppressive drugs, dose and duration of therapy, and the route of administration of mAbs[2].

Immunogenicity decreases as the fraction of human mAbs increases, however, even fully human mAbs may have immunogenicity. Low doses of mAbs have been found to elicit a stronger immune response than higher doses of the same mAbs. The chance of eliciting an immunological response grows as the period of treatment lengthens[2].

When compared to i.m. or i.v. administration, s.c. administration has a higher likelihood of triggering an immunological response, possibly due to the formation of aggregates at the injection site[1,2].

The formation of ADAS is usually a polyclonal response, with interindividual differences[2]. ADAS can increase CL, decrease mAbs serum levels, and affect the safety profile of mAbs, due to hypersensitivity reactions. mAbs-ADAS complex is inactive and may impair efficacy by reducing free mAbs. The complex accelerates mAbs CL because it triggers lysosomal degradation in the same manner as the target's mediated elimination[5].

The ADAS can be neutralizing antibodies or non-neutralizing antibodies. Neutralizing ADAS bind to mAbs active sites and, at low quantities, may not show a clinical effect. However, at higher quantities, there is a strong possibility to decrease clinical efficacy of the drug. Therefore, the level of neutralization is determined by the titter of ADAS. Non-neutralizing ADAS have no effect on the antigen-binding ability of mAbs[2]. However, ADAS formation can be transient (disappear with time) or persistent[7].

Taking Infliximab (IFX) as an example, antibodies to Infliximab (ATI) have been detected in up to 17 % of IBD patients on maintenance IFX[7]. These patients have a higher risk of suffering an infusion and hypersensitivity reactions, like anaphylaxis[6,7]. ATI can occur in a variety of isotypes (IgG, IgE, and IgM) and are directed towards distinct IFX epitopes. ATI that binds to the IFX idiotype directly limit drug activity by impeding the binding of IFX to TNF (neutralizing antibodies). However, antiallotype antibodies can be directed against epitopes on the Fab domain other than the idiotype, which probably will not inhibit the activity of IFX. As low TC can be an indication of ATI formation, ATI measuring is required. When ATI are positive, it is critical to follow up subsequent samples to exclude persistent ATI. In patients with persistent or sustained ATI, discontinuing IFX is the best clinical option, and it is also the most cost-effective[7].

3.2. Amount of antigenic target burden

Patients with most active disease have higher antigen expression and low mAbs serum concentrations. For example, in inflammatory diseases, high levels of circulating tumour necrosis factor alpha (TNF-α) and CRP are linked with more aggressive disease and consequently increased CL of IFX. In the ATLAS study, the proportion of tissue TNF-α to anti-TNF-α mAbs was raised in tissue with moderate to severe inflammation, indicating that there is insufficient anti-TNF-α mAbs to bind to TNF-α in these tissues[1].

In patients with HER2-positive metastatic breast cancer, high levels of circulating extracellular domain of HER2 result in increased CL of trastuzumab. The number of metastatic was identified as the most influential covariate associated with trastuzumab CL, with a 22 % faster CL in patients with four or more metastatic sites, compared with those with fewer metastases. The higher CL results in lower exposure to trastuzumab at steady state, compromising trastuzumab efficacy[1].

On the other hand, patients with a large tumour burden had a CL value of 0.249 L/day, compared to 0.199 L/day in patients with tumour burdens below the median, according to a pharmacokinetic analysis of a bevacizumab population[5].

This proves that, in cases of active diseases, the standard dose may be insufficient[1].

3.3. Concomitant drugs

An example of drug-drug interaction is the coadministration of immunosuppressive drugs (azathioprine, mercaptopurine, and methotrexate) with anti-TNF-α agents. These immunosuppressive drugs raise the serum concentrations of mAbs and reduce the development of ADAS. IFX levels were lower in patients who did not receive immunosuppressive therapy. The mechanism by which immunosuppressive agents increase the anti-TNF-α antibodies exposure is not yet determined. However, they are likely to act by reducing the formation of ADAS and/or diminishing the amount of target burden, because immunosuppressive drugs reduce TNF-α levels and inflammation, potentially limiting the disease and target-mediated CL of anti-inflammatory mAbs[1,5].

3.4. Concentration of serum IgG

FcRn recycles mAbs, and, consequently, increases their half-life time. If the concentration of mAbs or endogenous proteins enlarges, *FcRn* becomes saturated, and both the recycling of mAbs and their half-life time decreases[5].

3.5. Body weight and body surface area

The most common relevant factors identified in investigations on mAbs population pharmacokinetics are body weight and BSA, both strongly related to the Vd[1].

To minimize variability of mAbs exposure between patients, most mAbs are dosed considering patient body weight or BSA. When the dose of mAbs is linearly adjusted for body weight, slim or obese patients may be under or overdosed, respectively, because circulating plasma volume is not linearly associated with body weight[1].

Higher body mass index (BMI) has been found to appreciably increase loss of response to anti-TNF therapy. The mechanism underlying this association is not completely understood, but there are studies that found an increased CL in patients who had higher BMI (>30 kg/m^2) compared with non-obese patients[8].

These studies seem to indicate that the higher the Vd of the mAbs the higher the CL, probably because of a higher probability to suffer endocytosis with subsequent degradation.

3.6. Gender

Gender is a covariate for CL and Vd for several mAbs, even after body size correction. In several studies with panitumumab, rituximab, bevacizumab, and IFX, females usually have 23–39 % slower CL and 14–22 % smaller Vd in comparison to males. Nonetheless, there are other studies, where no body size-independent effect of gender on pharmacokinetics of rituximab or IFX was found. The clinical relevance of gender is still unclear and gender-adjusted dosing presently is not suggested for any mAbs[1]. More studies are required in this field.

3.7. Biochemical Data: Serum albumin, alkaline phosphatase and C-reactive protein levels

Albumin and alkaline phosphatase serum levels have also been recognized as covariates of mAbs CL. Studies suggest that the levels of albuminemia are inversely proportional to CL and indicate a positive relationship between alkaline phosphatase and CL[1].

For instance, bevacizumab CL in patients with low albuminemia is 19 % faster, while those with high alkaline phosphatase is 23 % faster. Regarding IFX, patients with low serum albumin have shown a 19.1 % faster CL. The underlying mechanisms by which low serum albumin and high alkaline phosphatase increase mAbs CL are unknown. However, there are two theories regarding this subject: due to disease severity (protein catabolism) or as a consequence of *FcRn* impairment with an increased IgG CL[1].

Hypoalbuminemia is a marker of cachexia and high protein turnover derived to chronic systemic inflammatory diseases. The endogenous catabolism for albumin is highly connected with the catabolic turnover of IgG. Thus, hypoalbuminemia leads to an in increased catabolic degradation of mAbs, which results in increased CL and decreased serum concentrations of mAbs[2].

CRP levels are considered an indirect marker of TNF-α concentration and correlate positively with mAbs CL. However, CRP is a relatively unspecific indicator of systemic inflammation and is controlled by a variety of factors[2]. In patients with Crohn's disease (CD), higher serum CRP concentration is related with increased IFX elimination[5].

3.8. Genetic Variants

The genetic polymorphisms can affect the pharmacokinetics of mAbs, more specifically distribution and elimination.

A genetic variant in the *FCGRT* gene affects the expression of one of the protein components of the heterodimeric *FcRn*. A 37-base pair variable number of tandem repeats (VNTRs) polymorphism in the FCGRT promoter region alters the degree of *FcRn* expression. VNTR3/VNTR3, the most common genotype, expresses 1.66-fold more *FcRn* transcript, when compared to the VNTR3/VNTR2 genotype. Therefore, patients with IBD who were heterozygous for VNTR3 had 14 % lower IFX exposure, probably resulted of a reduction of *FcRn* protection from lysosomal degradation[2].

The clinical response for trastuzumab, cetuximab, rituximab and IFX are correlated with a genetic polymorphism in the gene encoding for *Fcγ*R IIIa (exchange of valine (V) to phenylalanine (F) at position 158). The amino acid exchange impacts the affinity of IgG1 to the *Fcγ*R. V allele provides an increased binding affinity and improved mediation of Antibody Dependent Cellular Cytotoxicity (ADCC). As a result, V/V genotype patients had a higher rate of objective response and a longer progression-free survival. For IFX, it was suggested that F/F genotype reduces CL[2].

If ADCC is a primary elimination pathway, Fc*γ*R polymorphisms may affect mAb disposal. However, if ADCC is not a relevant CL process, it may have no relevant impact[2].

4. Importance of Therapeutic Drug Monitoring of Monoclonal Antibodies

The purpose of TDM is to individually adjust the drug dose according to the drug concentration of each patient. TDM allows to achieve maximal clinical efficacy and minimize adverse effects[5].

One or more of the following characteristics are required for TDM: serum concentration and response correlation; no direct clinical assessment of the drug's efficacy or toxicity; pharmacokinetics interpatient variation; small therapeutic window; flexibility in dosing; and accessibility of a standardized and validated method for serum concentration measurement[1,5].

In the particular case of mAbs, there is no traditional small therapeutic window because most mAbs do not have a maximum tolerated dose. Nonetheless, mAbs have an exposure-response relationship, no direct clinical measurement of drug effect, pharmacokinetic variability, which make mAbs good candidates for TDM[1].

For instance, TDM of IFX and adalimumab in IBD is sustained and supported by a consistently strong exposure-response relation and several patients do not achieve TC. TDM predicts which patients would benefit from dose intensification, drug switch or surgery[9]. IBD

patients have chronic intestinal inflammation that has pro-neoplastic effects, but the risk to develop dysplasia or cancer can be lowered with a correct and improved medical therapy that induces mucosal repair. So, TDM of IFX has an important role, guaranteeing the effectiveness of the therapy[10].

For TDM to be argued in guidelines, we need to have some information related to mAbs. Firstly, the exposure-response relation and target range of TC and ADAS must be defined. Secondly, the ideal timing (induction phase, maintenance phase or at loss of response) and the frequency of mAbs TDM must be established. In addition, it is important to know which patients will benefit from TDM (all patients or a subgroup of patients). Thirdly, prospective randomized trials are required to prove that TDM is more cost-effective than standard care. Lastly, a low cost, easy and rapid validation test to measure concentrations of mAbs and ADAS. Treatment algorithms should be developed and optimized (**Annex I**)[1].

5. Economic View

Considering the latest *Infarmed* report about hospital consumption, 2 of the 3 drugs that had the greatest increase in financial costs were mAbs in oncology. Among the several therapeutic classes consuming the highest costs, immunomodulators revealed the greatest financial charges for the Portuguese National Health Service and the greatest weight in the market[11].

A study investigated whether the TDM was more cost-effective than an empiric dose-escalation strategy, in patients with CD that loss response to IFX therapy. Two cohorts of patients were compared for both approaches over a 1-year period. The incremental cost-effectiveness ratio of the empiric strategy was defined as cost per quality-adjusted life-year (QALY) gained, in comparison with the TDM strategy. The results showed that the TDM approach produced equivalent QALYs when compared with the empiric strategy (0.801 *versus* 0.800, respectively), but was less expensive ($31,870 versus $37,266, respectively). Similar rates of remission (63 % versus 66 %) and response (28 % versus 26 %) were found. The TDM strategy was related to a higher percentage of surgeries (48 % versus 34 %) and lower use of high dose of IFX (41 % versus 54 %). These results support the hypothesis that TDM is the most cost-effective alternative because the same therapeutic effect was attained with lower doses[12].

Another study proved that individualised therapy, TDM, is more cost-effective than dose intensification in patients with CD, whose response to anti-TNF treatment was lost. In a randomised controlled trial, it was investigated the cost-effectiveness of TDM following an

algorithm (**Table 2**) to identify the causes for therapeutic failure in comparison of empiric dose escalation. Patients (n=69) with secondary IFX failure were randomly assigned to either empiric IFX dose intensification (5 mg/kg every 4 weeks) or TDM based on IFX and ADAS concentrations using the projected algorithm (**Table 2**). The cost of treating patients according to the algorithm was €6038, compared to €9178 for empiric IFX dose intensification. However, illness control was similar, as measured by response rates of 58 % and 53 %, respectively[13].

Table 2 - Treatment algorithm proposed for patients with CD with secondary loss of response to IFX[13].

	Detectable ADAS	**Undetectable ADAS**
Subtherapeutic IFX <0.5 µg/mL	**Group 1** Insufficient IFX bioavailability due to induced immunogenicity of IFX. ↓ Change to different Anti – TNF: Adalimumab 80 mg s.c. at inclusion followed by 40 mg s.c. every other week; dose intensification allowed.	**Group 2** Insufficient IFX bioavailability due to non-immune mediated pharmacokinetics. ↓ Intensify IFX treatment: IFX 5 mg/kg i.v. every 4 weeks.
Therapeutic IFX ≥0.5 µg/ mL	**Group 4** Consider (A) Pharmacodynamics (B) Non-functional ADAS (C) False positive test ↓ Repeat IFX and ADAS analyses and handle accordingly. If unchanged results, then act as in group 3.	**Group 3** Pharmacodynamics: anti-TNF-α is ineffective due to non-TNF driven disease. ↓ Anti-TNF is discontinued. Review of clinical condition at discretion of the investigator: - If relapse of CD use drugs with other target (immunosuppressive, glucocorticosteroids, and/or other biologics). Consider surgery, if appropriate.

ADAS, Anti-Drug Antibodies; IFX, Infliximab; i.v., Intravenous; s.c., Subcutaneous

These studies proved that the TDM in consequence of secondary IFX failure, using an algorithm based on IFX and ADAS, significantly reduces treatment costs per patient, when compared with empiric IFX dose escalation.

6. Data analysis / Analytic Methods

TDM requires standardized and validated methods for the measurement of mAbs and ADAS concentrations. Regarding mAbs concentrations, it is important to recognize if the measurement is for free or total (free, soluble target bound and ADAS bound) mAbs concentration[5]. Traditionally, TDM of biological drugs is based on TC, needing sample collection just before the next drug's administration[9].

For i.v administered mAbs, the two-compartment model is the most used. However, for s.c administration, a one-compartment model is frequently used due to slow absorption[1].

The solid phase enzyme-linked immunosorbent assay (ELISA), the radio-immunoassay (RIA) and the high-pressure liquid chromatography (HPLC)-based homogenous mobility shift assay (HMSA) are the analytical methods most used by laboratory quantification services[9].

ELISA is the most frequently used to quantify mAbs and ADAS concentrations because it is commercially available, user friendly and easily submitted to automatization. Drug concentrations are expressed in mg/L, consequently, it can be directly compared among different analytical methods[9].

On the other hand, ADAS values cannot be directly compared between different methods, because these use ADAS as a calibrator and the titters are expressed in relative units (e.g. ng/mL equivalents of the calibrator antibody). To make dose optimization algorithms assay-independent, classifications like "low," "intermediate," and "high" ADAS have been applied. To allow inter-laboratory harmonization of ADAS concentrations, universal calibrators for quantifying ADAS should be implemented[9].

To capture and detect ADAS by ELISA, two binding sites must be accessible. The bivalency is weakened if ADAS are already bound to the medication in serum; the bridge between the capture and detection antibodies cannot be established, and the ADAS cannot be measured. As a result, only free ADAS fraction excess may be quantified. Kopylov *et al.* developed an ELISA that uses an antihuman λ-chain antibody for detection, which does not require a free binding site for detection in ADAS[9].

However, ELISA assay is the most used method in TDM, even though the results take a long time to be available and it requires trained personnel. In opposition, rapid tests based on lateral flow take 20 to 30 minutes to generate the results and can be done by non-trained lab personnel[9,14].

In a cohort of IBD patients, a study compared a rapid test for quantitative assessment of IFX concentration (Quantum Blue® Infliximab) to one ELISA assay. Accordingly, rapid test and ELISA assays measured comparable IFX levels. Both assays were accurate in detecting undetectable, low, and adequate levels, although the rapid test had lower specificity for supratherapeutic IFX levels[14].

Point of care testing (POC) is another lateral flow analytical device that shows great promise in advancing TDM and allows drug dosage optimization based on real-time pharmacokinetic data. This is especially useful in situations where intra-patient pharmacokinetic variability is high, such as severe UC. CE-marked POC lateral flow immunoassays are available to measure IFX, ATI and adalimumab in serum[9].

7. TNF-α in Inflammatory Bowel Diseases

TNF is a powerful pro-inflammatory cytokine, mainly produced by immune cells as a result of infections or tissue damage. However, chronic inflammation is a consequence of a dysregulation of TNF. There are two forms of TNF, membrane-bound TNF (mTNF) and soluble TNF (sTNF) and two types of receptors, TNF receptor 1 (TNFR1) expressed in nearly every cell, and TNF receptor 2 (TNFR2) expressed in immune cells, mostly in regulatory T cells. TNFR1 is correlated to inflammation signalling pathways and tissue degeneration while TNFR2 is associated with immune modulation and tissue regeneration. sTNF and mTNF, can both activate TNFR1, however TNFR2 activation only occurs with mTNF binding[15,16].

IBD, CD and Ulcerative Colitis (UC) are chronic gastrointestinal conditions with increased levels of pro-inflammatory cytokines that result of a dysregulated immune response of human organism to the intestinal microbial flora. Scientific evidence suggests that normal intestinal bacteria trigger an abnormal immune reaction in people with a genetic predisposition[17].

CD can affect any part of the digestive tract, whereas UC almost always affects only the large intestine. Symptoms of IBD vary depending on affected part of the intestine. However, patients with CD usually have chronic diarrhoea and abdominal pain while patients with UC have intermittent episodes of abdominal pain and bloody diarrhoea. Persistent diarrhoea can lead to weight loss, malnutrition, and affects the intestinal absorption process and Vd[17].

Although there is no cure for IBD, many medications, including aminosalicylates, corticosteroids, immunomodulatory medications, biological agents, and antibiotics, can reduce inflammation and alleviate IBD symptoms[17].

Biologic therapies selectively block the inflammatory cascade, like agents targeting TNF-α. Biological drugs are now a crucial component of IBD treatment[9,18].

IFX administration in CD patients is often monitored, and therefore it will be focused as an example of the importance of monitoring mAbs in the following sections.

7.1. Infliximab

IFX is a chimeric monoclonal IgG1 antibody against TNF-α, indicated for the treatment of CD and UC. IFX binds with high affinity to soluble and transmembrane forms of TNF-α (**Figure 3**) but not to lymphotoxin α (TNFß)[19].

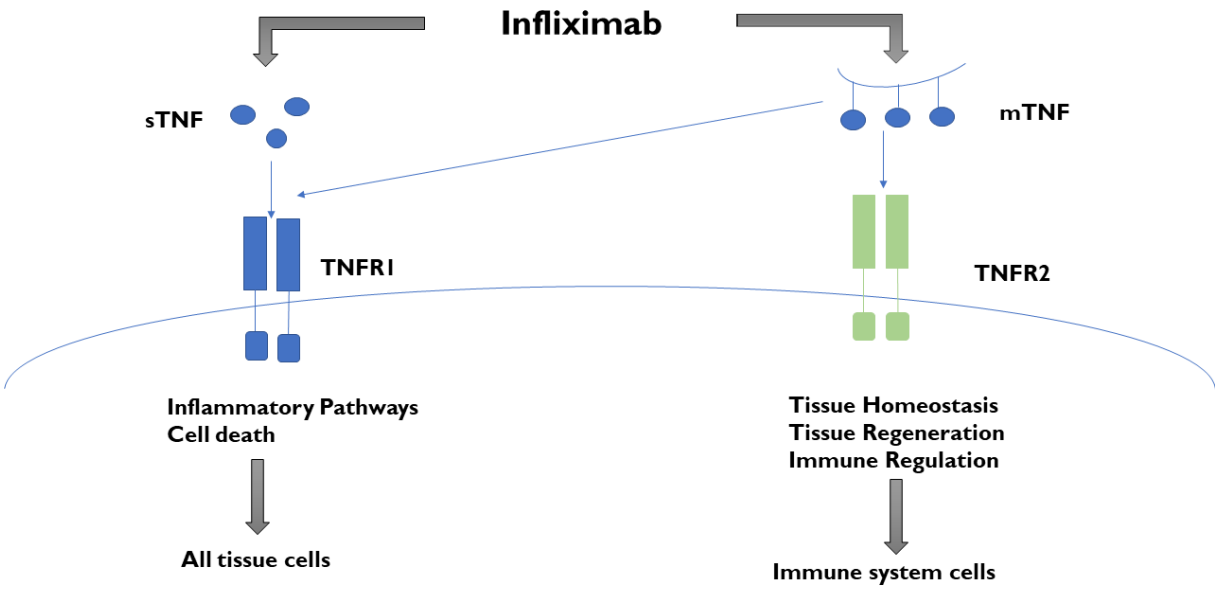

Figure 3 - Representation of IFX mechanism of action currently approved[15]. mTNF, Membrane-bound Tumour Necrosis Factor; sTNF, Soluble Tumour Necrosis Factor; TNFR1, Tumour Necrosis Factor receptor 1; TNFR2, Tumour Necrosis Factor receptor 2.

The overall mechanisms of IFX are still in debate, however, it is suggested that neutralization, outside-to-inside signalling, ADCC, and apoptosis of inflammatory immune cells are some of them **(Figure 4)**[20].

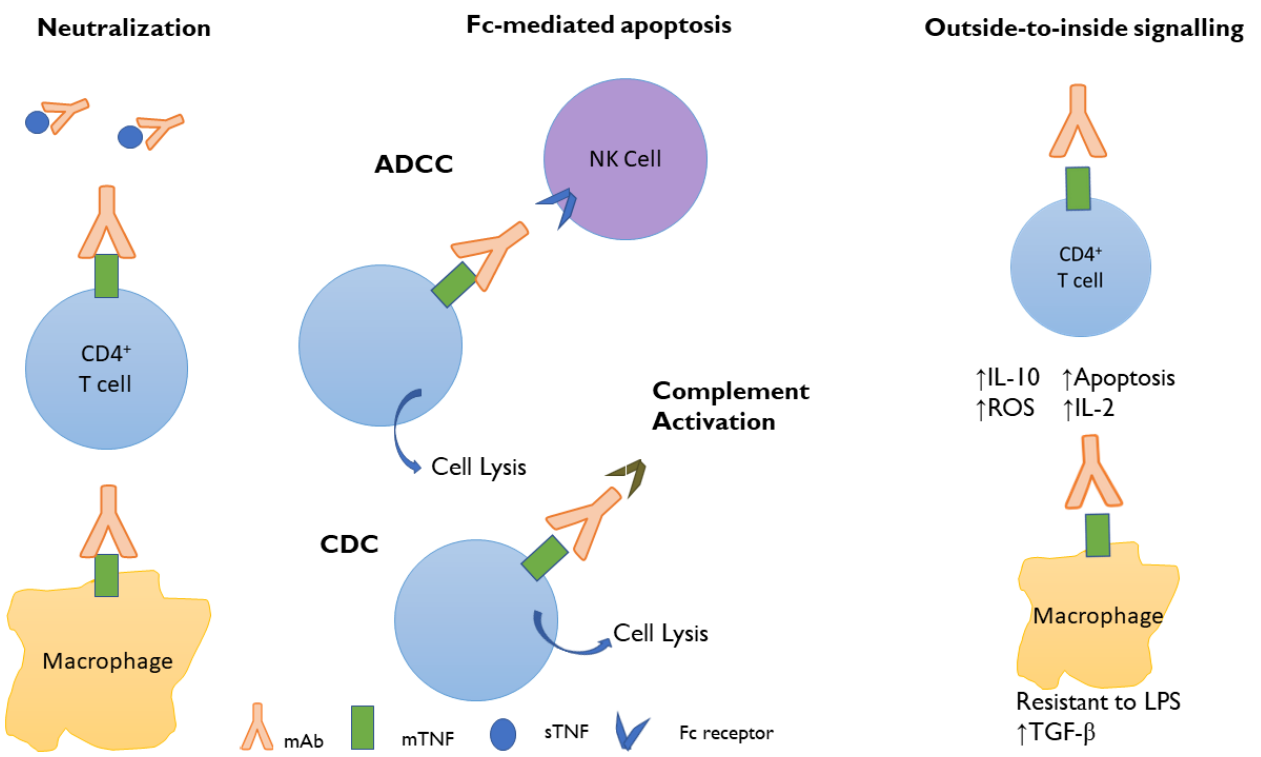

Figure 4 - Mechanism of action proposed for IFX. ADCC, Antibody Dependent Cellular cytotoxicity; CDC, Complement-Dependent Cytotoxicity; LPS, Lipopolysaccharide; mTNF, Membrane-bound Tumour Necrosis Factor; NK, Natural killer; ROS, Reactive Oxygen Species; sTNF, Soluble Tumour Necrosis Factor.

Posology of IFX is 5 mg/kg administered as an i.v. infusion and the regimen includes an induction phase (i.v. at weeks 0, 2, and 6) followed by maintenance treatment (i.v. every 8 weeks) in responder patients[19].

After i.v. administration, an acute infusion reaction is described as an adverse effect that occurs within 1 hour after infusion. In opposition, delayed hypersensitivity reaction is characterized as the occurrence of myalgia, arthralgia, fever, or rash, and occurs 1–14 days after infusion[21].

Complete response in CD patients may be defined as the cessation of diarrhoea and abdominal cramping, as well as the cessation of fistula drainage and the closure of all draining fistulas. Partial response can be characterized as a reduction in the quantity of diarrhoea and abdominal cramping, as well as a reduction in the drainage, size, or number of fistulas. Non-responders exhibit outcomes that do not fit one of the aforementioned characteristics[21].

In UC patients, complete response may be defined as absence of diarrhoea, haematochezia, and abdominal cramping whereas partial response was defined as a decrease in the quantity of diarrhoea, haematochezia, and abdominal cramping[21].

Based on clinical indices, response and remission terms have commonly defined, with recourse to Crohn's Disease activity Index (CDAI) and Harvey Bradshaw Index (HBI)[8].

In CDAI (**Annex 2**), the definition of initial clinical response is a reduction in more than 70 points or 100 points from baseline; 25 % or more decrease in the total score, or a combination of both. Secondary loss of response is characterized as a CDAI increase of more than 70 points from the pre-induction score linked with a total score of greater than 175; an increase in CDAI of ≥35 % from the baseline score, or instead the need to introduce a new treatment for active CD. HBI (**Annex 3**) classify initial response as a reduction in score of ≥2 for the period of or after induction phase and clinical remission as a score of <5[8].

CDAI has been used in clinical trials, but it has some limitations, such as the parameters used to define remission. CDAI is difficult to calculate in clinical practice, requires the patient's daily data, is biased towards diarrhoea (which is often caused by factors other than inflammation), is ineffective in patients with stomas, and has not been validated for use following surgery. The HBI, on the other hand, is simple to calculate and measure, and less subject to confounding variables based on clinical criteria alone, but diarrhoea yet is severely weighted[22].

Approximately one-third of IBD patients receiving anti-TNF therapy are primary non-responders, which means that the lack of response occurs during the induction phase (8–12 weeks) and the secondary loss of response occurs in up to 40 % of patients during the first year of therapy[23]. However, some studies point to a percentage of 60 % where patients experienced secondary loss of response[24].

TDM is a potential technique for optimizing IBD therapies. The significantly interindividual variability may include how patients metabolize the drug (pharmacokinetics), the magnitude and duration of response to therapy (pharmacodynamics), immunogenicity, disease characteristics (phenotype, location, severity) and treatment strategy[8,23].

It is important to distinguish the reactive TDM, which occurs in case of loss of response or intolerance to mAbs, and the proactive TDM, which occurs when drug dose is adjusted to a target drug concentration to prevent relapse[9].

7.2. Reactive Therapeutic Drug Monitoring

The aim of reactive TDM is identify the reasons underlying the loss or no response to a biological therapy and facilitate therapeutic decisions, like increase drug, add immunomodulator, switch in class or switch out of class. Reactive TDM is more cost-effective

and is associated with better endoscopic outcomes when compared to empiric IFX dose optimization[9,12,18].

In 40 % of patients with CD, there is a loss of response to anti-TNF therapy. As already aforementioned, loss of response can result from mechanisms such as: low drug serum concentrations, development of ADAS, activation of cytokine pathways other than TNF, and non-inflammatory disease processes such as irritable bowel syndrome or bacterial overgrowth (**Annex 4**)[23].

Patients who are on maintenance IFX medication and have detectable TCs have higher rates of clinical remission, reduced serum CRP levels, and improved endoscopic rates. Subtherapeutic concentrations of IFX are associated with increased disease activity in CD. In addition, ATI has been linked to an increased risk of infusion reactions and lower IFX concentrations[21].

Empiric escalation of therapy is a blind approach, which can add significant additional costs and delay more effective therapy, especially if repeated attempts of empiric dose escalation are made. Furthermore, further drug exposure in individuals with immune-mediated pharmacokinetic failure (ADAS) is probable to result in acute or delayed hypersensitivity reactions. Excessive drug exposure could theoretically increase the likelihood of drug-related adverse events including serious infections, albeit this has not yet been proven[23].

The use of a priori diagnostic tests to target treatment based on the most likely mechanistic cause for lack of response is an alternative to the empiric approach[12].

7.2.1. Clinical Studies of Reactive Therapeutic Drug Monitoring

Reactive TDM is supported by clinical studies which are described below.

The concentrations of ATI and IFX were tested in 155 participants mainly due to loss of IFX response (49 %), partial response after starting IFX (22 %), and potential autoimmune/delayed hypersensitivity reaction (10 %). ATI was found in 35 patients (23 %) and a change to another anti-TNF treatment was related with a complete or partial response in 92 % of positive ATI patients (**Table 3**), while dose escalation had a response of 17 %. The presence of ATI clearly evidences that immunogenicity to IFX has been developed, and further treatment would result in a decreased clinical response or possible infusion reactions, so the best action is to change to another-TNF agent. Dose escalation (possible by increasing the dose or decreasing the interval between doses) was linked with complete or partial clinical response in 86 % of patients with subtherapeutic IFX concentrations. Patients with clinical

symptoms and therapeutic IFX concentrations were kept on the same dose 76 % of the time, and endoscopic/radiographic evaluation revealed no evidence of active inflammation 62 % of the time[21].

Reduced infusion reactions, greater clinical remission, lower CRP, and endoscopic healing are all linked to a 4-week post-infusion IFX concentration of >12 µg/mL and detectable TCs. So, if the patient has therapeutic concentrations and active disease, the best option is change to a different mechanism of action, a non-anti-TNF agent[21].

It is also crucial to follow-up the patient to access if the approach was correct[21].

In conclusion, this study demonstrated that ATI and IFX concentrations influence treatment decisions in 73 % of patients, and that TDM is a valuable complement to clinical and endoscopic/radiological evaluation[21].

Table 3 shows a TDM algorithm to elucidate the therapeutic decisions in case of loss of response to IFX. This model takes into account ADAS and TC. Since the ADAS are, usually, a consequence of subtherapeutic concentrations, it is my opinion that ADAS data needs to be linked with concentrations of IFX instead of being considered an isolated variable.

Table 3 - Scheme of therapeutic decisions in patients with clinical symptoms[21].

Positive ADAS	Change to another anti-TNF agent	If disease persist, change to a different mechanism of action – non-anti-TNF agent
Therapeutic IFX concentration (>12 µg/mL at 4 weeks or detectable TC)	Active disease on endoscopy/ radiology	Change to a different mechanism of action - non-anti-TNF agent
	Inactive disease on endoscopy/radiology	Investigate for alternate etiology of symptoms
Subtherapeutic IFX concentration (<12 µg/mL at 4 weeks or undetectable TC)	Increase dose or frequency (↓interval)	If disease persist, change to different anti-TNF agent
	Change to different anti-TNF agent	If disease persist, change to a different mechanism of action - non-anti-TNF agent

ADAS, Anti-Drug Antibodies; IFX, Infliximab; TC, Through concentration; TNF, Tumour Necrosis Factor; ↓, decrease.

As mentioned earlier, reactive TDM has the potential to identify the mechanism(s) (pharmacodynamic, pharmacokinetic or immunogenic) underlying the loss of response[9].

Mechanistic failure or pharmacodynamic loss of response occurs when patients have adequate drug exposure but no response to therapy. This probably results from the fact that the disease is not being developed by the antigen that the drug targets[23]. Regarding this scenario, switching to a drug of another class, with a different mechanism of action, may be the optimal strategy (**Table 4**).

Patients may suffer from pharmacokinetic failure because of insufficient drug exposure due to increased drug CL. In this situation, dose increase may result in therapeutic response.

If ADAS are detected, especially at high titters, it means that drug CL has risen due to immunogenic mechanism. In this case, switching to a drug from the same group but with a different chemical structure is expected to be the best pharmacologic option. If ADAS are not detected, CL is increased by non-immune processes, like high inflammatory burden, leading to rapid drug utilization and/or excessive elimination due the faecal loss (indicated by higher CRP, faecal calprotectin, and/or low albumin)[23].

Table 4 - Overview of causes for loss of response and the corresponding therapeutic decision[9].

Drug concentration	ADAS	Possible Cause for loss of response	Intervention
High	Not relevant to measure	***Pharmacodynamic***	Swich to another class
Adequate	Not relevant to measure	***Pharmacokinetic*** (May require higher concentrations)	Dose intensification If response not recovered → swich to another class
Low	Intermediate or absent	***Pharmacokinetic*** Check compliance	Increase drug concentrations (↓ interval, ↑dose or + immunosuppressive)
Absent	Intermediate or absent	***Pharmacokinetic*** Check compliance	Increase drug concentrations (↓ interval, ↑dose or + immunosuppressive) or consider re-induction
	High	***Immunogenicity***	Switch within class or to another class

ADAS, Anti-Drug Antibodies; ↑, increase; ↓, decrease; +, Add

7.2.2. Guidelines regarding Reactive Therapeutic Drug Monitoring

Current guidelines regarding reactive TDM are very conservative in their recommendations.

The European Crohn's and Colitis Organisation (ECCO) reports that there is insufficient evidence to recommend for or against reactive TDM to improve clinical outcomes. ECCO refers that in terms of clinical outcomes, there was no meaningful difference. Nonetheless even after considering the utilization of biosimilar drugs, reactive TDM shows to have economic benefits[25].

In opposition, the American Gastroenterological Association (AGA) proposes reactive TDM to guide treatment switches in adults with active IBD treated with anti-TNF agents[26].

The British Society of Gastroenterology also recommends that treatment options for failure of initial anti-TNF therapy may be based on clinical context and serum concentrations of drug and ADAS. In addition, they advise that patients with secondary loss of response to anti-TNF therapy may have serum drug and ADAS concentrations measured to inform appropriate changes in treatment[22].

European Society for Paediatric Gastroenterology Hepatology and Nutrition (ECCO-ESPGHAN) Guideline supports TDM to guide therapy modifications in patients with active CD who are being treated with anti-TNF medications, rather than empirically escalating dose or switching therapy[27].

In spite of several international guidelines appeal for reactive TDM, Casteele *et. al.* enumerates factors for its uncertain benefit: firstly, the included trials that had a high risk of bias due to poor protocol adherence and heterogeneous and mostly retrospectively established optimum trough and ADAS thresholds. Secondly, to start intervention and to quantify intervention success, most studies relied on clinical disease activity, with or without biochemical or endoscopic evidence of disease activity. At least, in people with CD, clinical disease activity has a small connection with endoscopic disease activity, surgical risk, and disease-related consequences. Thirdly, there are missed treatment opportunities because of sub optimally thresholds for drug concentration and ADAS[23].

Overall, the risk-benefit reveals that reactive TDM may be advantageous when compared with empiric drug increment[23].

7.3. Proactive Therapeutic Drug Monitoring

The aim of proactive TDM is targeting drug concentrations in the optimal therapeutic range, at specific time-points, to improve response rates and prevent secondary loss of response and the development of ADAS (low drug concentrations can predict immunogenicity)[18].

It can be used to better guide biologic de-escalation and withdrawal in remission patients, as well as when reintroducing anti-TNF medication after a drug holiday and assessing patient compliance with biologic therapy. To prevent relapse, TDM is required after de-escalation to keep TC>2.4 μg/mL (**Figure 5**)[18].

An observational study revealed that proactive TDM of IFX was independently associated with fewer surgeries [odds ratio (OR): 0.36; 95 % confidence interval (CI): 0.13–0.95; p=0.039], higher rates of mucosal healing (OR: 3.26; 95 %CI: 1.68–6.31; p<0.001) and significantly reduced unfavourable outcome (like surgery, hospitalization, treatment failure or no mucosal healing) (OR: 0.358; 95 %CI: 0.188-0.683; p=0.002), compared to conventional management[9].

In a larger retrospective multi-center trial, proactive TDM was linked to fewer treatment failures, IBD-related hospitalizations, IBD-related surgeries, ATI, and infusion reactions. Furthermore, proactive TDM was found to be helpful even after reactive TDM, since it resulted in fewer treatment failures and hospitalizations[9].

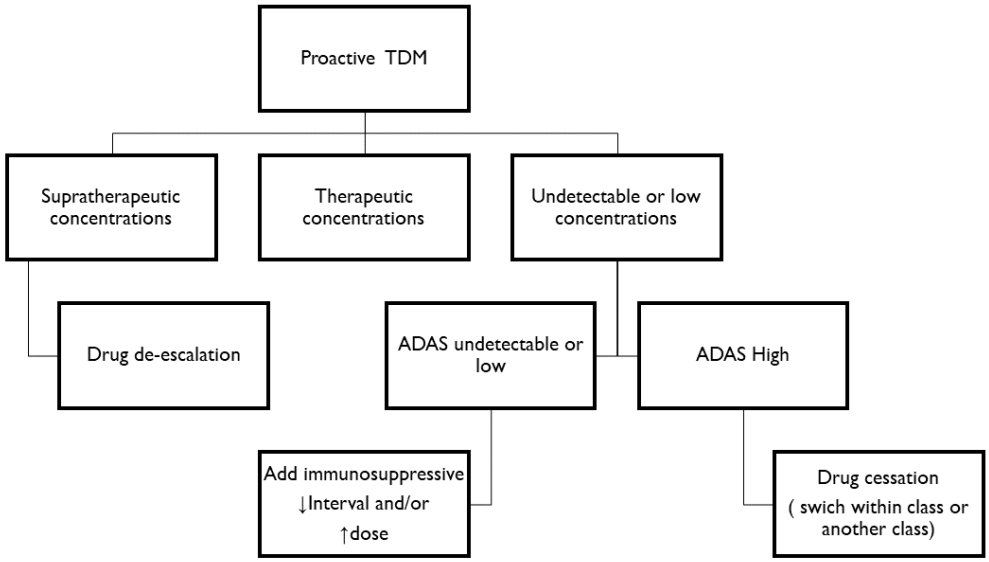

Figure 5 - Summary of Proactive TDM[18]. ADAS, Anti-Drug Antibodies; TDM, Therapeutic Drug Monitoring; ↓, decrease; ↑, increase.

7.3.1. Clinical Studies of Proactive Therapeutic Drug Monitoring

Casteele *et al.* performed a 1-year randomized controlled trial, involving 263 adults (178 with CD and 85 with UC) with stable responses to maintenance IFX therapy, like full responders or partial responders. Patients with ATI>8 μg/mL and patients with higher

concentrations of IFX due to secondary loss of response were ineligible. The aim of TAXIT study was to determine if dosing based on proactive TDM increases remission rates and if continuous proactive TDM is superior to clinically based dosing, in patients taking IFX for maintaining remission[24].

Firstly, all patients were dose optimized to achieve an IFX TC within the interval of 3–7 μg/mL (optimization phase) following the TAXIT algorithm (**Figure 6**)[24].

Briefly, in supra-optimal concentrations patients, the dose was reduced to 5 mg/kg (if on 10 mg/kg). Following that, the interval between infusions was increased by 2 weeks each time (to a maximum interval of 12 weeks). Regarding suboptimal concentrations patients, the interval between infusions was reduced 2 weeks each time (to a minimum interval of 4 weeks). Following that, the dose was increased to a maximum of 10 mg/kg[24].

Patients who achieved an IFX TC within the optimal interval were then randomly assigned (1:1) to groups that received IFX dosing based on their clinical symptoms and CRP, or continued dose based on TC (maintenance phase). In this last group, each IFX TC was assessed for each patient, and the dosage regimen was modified for the next infusion based on the TAXIT algorithm to keep patients between 3–7 μg/mL[24].

Within the 263 tested patients, 115 had an optimal TC of IFX (3–7 μg/mL), 76 presented TC values lower than 3 μg/mL, and 91 % of those, 69 patients, achieved the goal after dose escalation. This leads to a higher proportion of remission in CD patients taking IFX, after dose escalation and a decrease in the concentration of CRP, but it is not consistently observed in UC patients[24].

After dose reduction, 93 % of 72 patients with TCs>7 g/mL achieved the goal TC, resulting in a reduction of 28 % in drug cost[24].

The primary end point was clinical (HBI≤4 for CD and partial Mayo score [PMS]≤2 with no individual subscore>1 for UC) and biological ([CRP]<5 mg/L) remission at year 1 after optimization[24].

Results show that remission (primary endpoint) was reached by 66 % of patients whose dose was based on clinical symptoms and CRP and 69 % of patients whose dosing was based on TC. Disease relapsed occurred in 21 patients (17 %) who were given clinically based dosing and 9 patients (7 %) were TDM was performed (p=0.018)[24].

IFX TC between 3–7 μg/mL, lead to more efficient drug use, and probably is a good threshold. These results indicate that maintaining an adequate exposure led to better short-

term clinical outcomes and can reduce the risk of loss of response. After dose optimization, the authors concluded that concentration-based on TC was not superior to clinically based dosing for establishing remission after 1 year. However, it was linked with fewer flares throughout treatment[24].

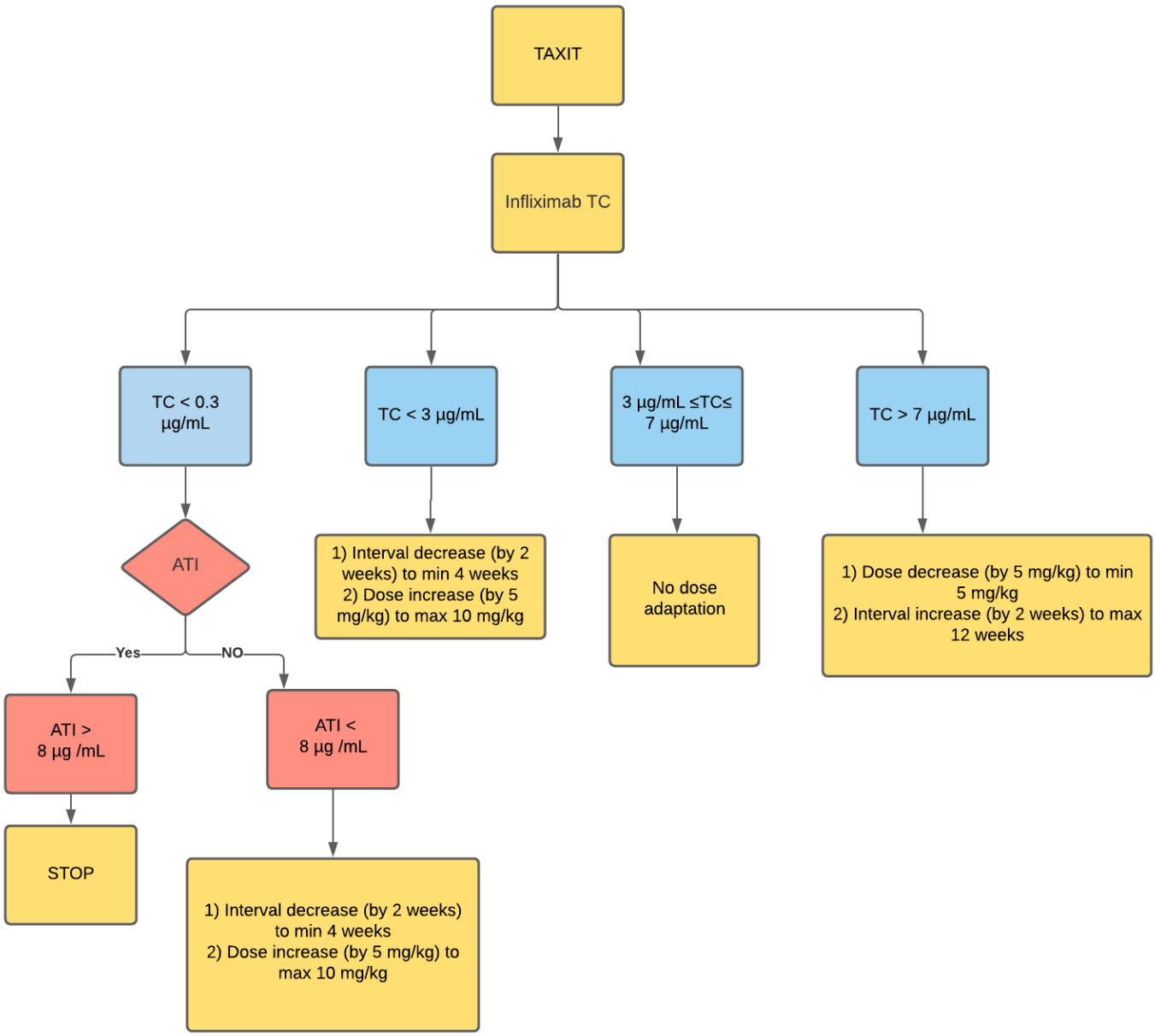

Figure 6 - Scheme of TAXIT algorithm[24]. ATI, Antibodies to Infliximab

TAXIT trial failed its primary end points, however it was handicapped by methodological imperfections. For example, only patients in remission were randomized, and they were exposed to a dose optimization intervention first[28]. In my opinion, the optimization phase is already a proactive TDM approach that proved the increases in remission rates.

Considering that these diseases are very oscitant, and that the inflammation is triggered by numerous factors, it is important to measure concentrations at specific time points to make sure that patients achieve IFX TC between 3–7 μg/mL.

Another clinical study is TAILORIX, a prospective, randomized trial performed to determine if TDM to sustain serum levels of IFX above 3 μg/mL generates higher rates of clinical and endoscopic remission when comparing with patients with a dose based on symptoms only[29].

The methodology consisted of a double-blind trial enrolling 122 biologic-naïve adult patients with active CD and taking IFX in association with an immunosuppressant. CDAI >220 with signs of active inflammation (CRP>5 mg/L and/or faecal calprotectin >250 μg/g), visible ulcers at ileocolonoscopic were assessed to confirm disease activity[29].

All patients received 5 mg/kg of IFX i.v. at weeks 0, 2, and 6. Patients were randomized to one of three IFX maintenance groups (DIS1, DIS2, Control) at week 14 of treatment, with IFX administered each 8 weeks from week 14 to week 54. In DIS1 group dose was increased (2 maximum) in 2.5 mg/kg increments based on clinical symptoms and biomarkers and/or serum IFX concentrations; in DIS2 group dose was increased from 5 mg/kg (maximally 1 time) to maximum dose of 10 mg/kg based on the same criteria; finally, in the Control group, dose was increased to maximally 10 mg/kg based only on clinical symptoms. CD activity index scores, biomarkers, and IFX serum concentrations were measured at baseline and at weeks 2, 4, 6, 12, and 14 of therapy, and subsequently every 4 weeks thereafter up to week 54[29].

In conclusion, in this prospective randomized trial, dose increase of IFX based on a mixture of symptoms, biomarkers and/or serum drug concentrations, was not superior to dose increase based on symptoms[29].

The high rate of dosage escalation in the Control group was the most obvious reason for the divergent outcome in TAILORIX. Patients could only raise their IFX dose based on symptoms, which may have pushed patients and physicians to over-report and inflate symptoms, leading to dose escalation[29].

Furthermore, TDM was only utilized from week 14 onwards, which may have resulted in ‘missed chances' in the early stages of the treatment. The IFX doses provided were calculated based on the TC, which was measured before the previous infusion and the result was delayed for several days due to shipment to a central lab. As a result, an 8-week wait may be too long for some individuals to maintain their treatment response. Another explanation

was that out of 43 dose escalation chances based on CDAI in the DIS groups, 53 % were avoided per protocol since the biomarkers were not increased. While, in the Control group, normal CRP and/or faecal calprotectin were found in 60 % of the CDAI based dose escalation occurrences[29].

In my opinion, it is safer to increase the IFX dose to 10 mg/kg when we are certain that the patient needs it (based on IFX concentrations and ATI), instead of increasing the dose based only on symptoms. If the patient has ATI, it can suffer serious adverse events. I think that we need to have into consideration all the factors: symptoms, biomarkers, IFX concentrations and ATI and endoscopy/radiology exams. It is essential to have all the information required to make the best decision for every patient.

7.3.2. Therapeutic Drug Monitoring in induction phase

The role of TDM during the induction phase of anti-TNF therapy is yet poorly understood, and the therapeutic drug window is mostly uncertain. The induction phase is critical because patients often present a higher inflammatory burden, with elevated CRP, reduced albumin, and drug loss through stool, resulting in higher drug CL and lower concentrations, prompting to the ADAS formation. Early optimization of anti-TNF therapy would tremendously help these active patients by preventing some of the primary nonresponse (related to low drug concentrations) and by leading to better short and long-term outcomes. Recent data shows that higher serum anti-TNF drug concentrations during induction phase are correlated with better therapeutic outcomes in IBD[30].

7.3.3. Guidelines regarding Proactive Therapeutic Drug Monitoring

AGA Institute reports that, in patients with quiescent IBD treated with anti-TNF agents, the benefit of routine proactive TDM over no therapeutic monitoring is uncertain[26].

Moreover, according to ECCO guidelines, there is presently insufficient evidence to recommend for or against the use of proactive TDM to enhance clinical outcomes in CD patients in clinical remission under anti-TNF agent[25].

However, the British Society of Gastroenterology recommends that all IBD patients should be revised 2–4 weeks after finishing induction phase of anti-TNF therapy to measure response and optimise maintenance dosing based on clinical response, serum drug and ADAS concentrations, blood inflammatory markers, faecal biomarkers, or endoscopy[22].

ECCO-ESPGHAN Guideline supports early proactive TDM in patients on anti-TNF drugs[27].

7.4. Therapeutic threshold of anti-TNF-α

Is the therapeutic threshold identical for all patients and for the different phases of the treatment?

Therapeutic TC and ADAS thresholds are not clearly defined. The therapeutic TC depends greatly on the intervention phase (induction *versus* maintenance), the treatment objective (clinical remission *versus* endoscopic remission), and the stage of disease activity (severe active *versus* mild active disease; luminal *versus* perianal disease)[23]. However, in **Table 5** are proposed target concentrations of IFX to achieve favourable therapeutic outcomes, taking into account the intervention phase and the weeks of treatment.

Various studies show that patients with IBD who have a TC of <3 μg/mL are more likely to lose efficacy and develop ADAS[31]. The equation derived from linear regression indicates that the TC quantified at 8 weeks can be used to make an estimation of IFX CL using the following equation[31]:

$$CL\ (mL/h) = -1.076 * serum\ IFX\ TC + 17.11$$

The results suggest that an IFX TC>3 μg/mL may represent an IFX CL rate of >13.882 mL/h. Due to drug accumulation, the TC of IFX measured after a single dose may be lower than the TC obtained after numerous IFX doses. In patients with IBD, prospective studies can be utilized to generate standard curves from serum IFX TC and determine IFX CL[31].

In a cross-sectional study, Yarur *et al.* found that individuals with healing fistulas had considerably higher median serum IFX levels than those with active fistulas [15.8 versus 4.4 μg/mL, respectively ($p<0.0001$)]. They confirmed that patients with fistulizing CD benefit from higher TC (≥10.1 μg/mL) than those usually recommended for luminal CD (3–7 μg/mL) **(Table 6)**. So, it was concluded that there is a significant correlation between serum IFX levels and rates of fistula healing[32].

Therefore, probably, there is not a universal optimal cut-off for IFX serum concentrations. Presumably, different concentrations are required to achieve different outcomes, such as higher concentrations for endoscopic and fistula healing comparing with for symptomatic remission[28,32].

Table 5 and **Table 6** are important tools to follow-up patients in induction and maintenance phase and help identify the concentrations range that can be applied to different treatment objective.

Table 5 - Proposed IFX target concentrations for achieving favourable therapeutic outcomes in IBD[9].

Infliximab	
Time Point	**Drug Concentrations (µg/mL)**
Induction (week 2)	≥20-25
Induction (week 6)	≥10-15
Post – Induction (week 14)	≥3-7
Maintenance	≥3-7

Table 6 - IFX concentration threshold associated with clinical remission/response or mucosa healing[33].

Infliximab		
Time Point	**Drug Concentrations for Clinical Response/Remission (µg/mL)**	**Drug Concentrations for Mucosa Healing (µg/mL)**
Induction (week 2)	≥20	≥25
Induction (week 6)	≥10	N/A
Post – Induction (week 14)	≥3	≥7
Maintenance	≥3	≥7

N/A – not available

More research is necessary to confirm the exact concentration thresholds of clinical and endoscopic remission.

8. Conclusions

There are several factors that affect pharmacokinetics of mAbs, including target burden, the formation of ADAS, the amount of IgG, concomitant administered drugs, levels of albumin, alkaline phosphatase and CRP, and others. These factors can explain, at least in part, the variability of response against biological drugs. Therefore, TDM has a crucial role, with the potential to guide to a more individual dose, which increases efficacy and safety.

The causes of nonresponse related to drug factors, can be due do pharmacokinetic, pharmacodynamic or immunogenicity mechanisms. It is crucial to determine the mechanism who can have a response in accordance with the cause.

Immunogenicity mechanisms are associated with ADAS production, which can be a consequence of low drug concentrations. It is very important to measure ADAS concentrations before dose increase in order to prevent infusion reactions and side effects. In case of high and persistent ADAS, it is safer and more effective to switch within class to a different chemical structure or change to another class. Pharmacokinetic failure can be modified through increased doses and pharmacodynamic, by changing to an out of class agent.

TDM is associated with better therapeutic outcomes, giving more quality of life to patients. In short, reactive TDM is an approach to the management of patients with loss of response to biologics while proactive TDM allows to achieve therapeutic concentrations and prevents loss of response. Both are cost-effective and provide a more rational and safe approach to achieve disease remission[18].

Regarding IFX in IBD, there are several studies that demonstrate the relevance of a TDM approach with less treatment failure, higher rates of remission and mucosal healing, fewer surgeries, lower risks for ATI and for serious infusion reactions[9].

In my opinion, to make the best decision for every patient the following factors should be taken into account: symptoms, biomarkers, concentrations of IFX and ATI and endoscopy/radiology exams. However, further research studies and clinical trials are required to achieve the full confidence in TDM of biological therapies.

Individualized mAbs dose guidance by TDM has the potential to be effective and helpful to optimize the treatment in additional disease indications like oncology. For that, adequate exposure needs to be defined[24].

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10. Annexes

Annex 1 – Scheme of TDM to managing secondary loss of response[12]

Testing	ADAS	Drug concentration	CT Enterography/ Colonoscopy	Next step	Presumed mechanism
Yes	Present	-	-	Swich to different anti-TNF (adalimumab) **1.1**	ADAS
	Absent	Therapeutic	Moderate inflammation	Swich to different class **1.2**	Mechanistic override TNF (Pharmacodynamic)
			Mild/min inflammation	Continue dose (5 mg/kg) **1.3**	Non-inflammatory causes
		Subtherapeutic	-	Increase dose (10 mg/kg) **1.4**	Low drug concentration
No	-	-	-	Increase dose (10 mg/kg) **1.4**	Unknown (low drug concentration addressed first)

1.1 Swich to different Anti-TNF (adalimumab)	Survive	Initial response	Sustained response		Remission
					Response
			Not sustained response – increase dose	Sustained response	Remission
					Response
				Not sustained response	Switch to different class (surgery) **1.2**
		Not initial response			Switch to different class (surgery) **1.2**

1.2 Swich to different class (surgery)	Sustained response		Remission
			Response
	Not sustained response – Restart biologic	Sustained Response	Remission
			Response
		No sustained response	Active disease

1.3 Continue IFX 5 mg/kg	Sustained response	Min/mild inflammation with symptoms
	No sustained response	Swich to different class (surgery) **1.2**

1.4 Increase dose IFX to 10 mg/kg	Initial response	Sustained response	Remission
			Response
		No sustained response	Swich to different anti-TNF (adalimumab) **1.2**
	No initial response		Swich to different anti-TNF (adalimumab) **1.2**

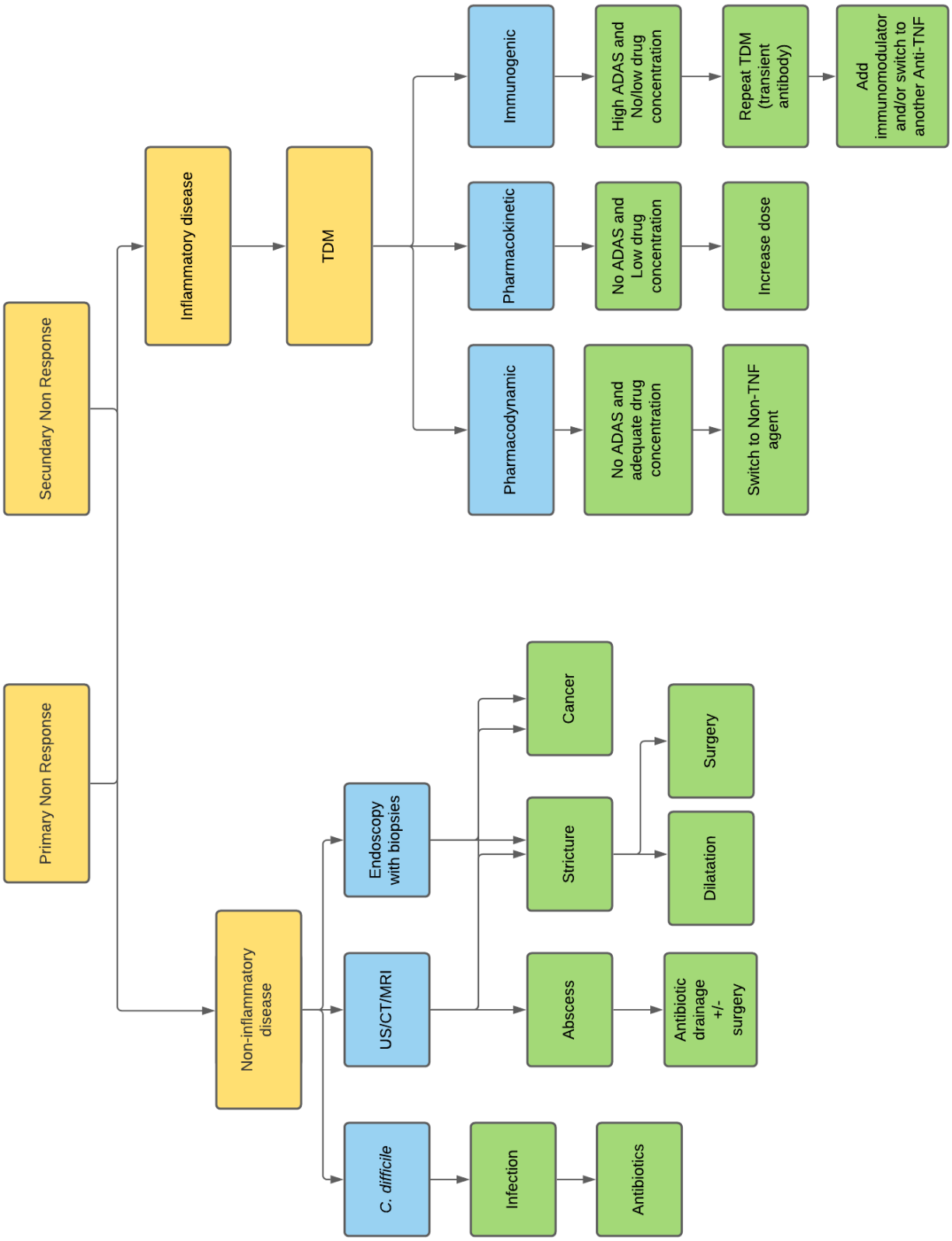
Annex 2 - Crohn's Disease Activity Index[34, 35]

Variable	Quantity	Multiple	Total
Number of liquid or soft stools per day		2	
Abdominal pain (0=none, 1=mild, 2=moderate, 3=severe)		5	
General well-being (0=well, 1=slightly under par, 2=poor, 3=very poor, 4=terrible)		7	
Number of complications: arthralgias, iritis, erythema nodosum, pyoderma gangrenosa, aphthous ulcerations, anal fissure, anal fistula, anal abscess, fever >37° past week, intestinal obstruction		20	
Diphenoxylate or Loperamide for diarrhea (no=0, yes=1,)		30	
Abdominal mass (no=0, questionable=2, yes=5)		10	
Deviation from normal hematocrit (N=42 for female, 47 for male)		6	
% Deviation from baseline weight		1	
Total CDAI score			
CDAI score: < 150 = Symptomatic remission; 150-220 = Mild; 220-450 = Moderate > 450 = severe			

Annex 3 - Harvey Bradshaw Index[35,36]

Variable	Quantity
Number of liquid or soft stools per day	
Abdominal pain (0 = none, 1 = mild, 2 = moderate, 3 = severe)	
General well-being (0 = very well, 1 = slightly below par, 2 = poor, 3 = very poor, 4 = terrible)	
Number of complications: arthralgias, uveitis, erythema nodosum, pyoderma gangrenosa, aphthous ulcers, anal fissure, anal fistula, anal abscess.	
Abdominal mass (0 = none, 1 = dubious, 2 = definite, 3 = definite and tender)	
<5 = Remission; 5-7 = Mild disease; 8-16 = Moderate disease; >16 = Severe disease	

Annex 4 - Clinical steps to the management of primary nonresponse and secondary loss of response[8]



CT, Computed tomography; MRI, Magnetic resonance imaging; US, Ultrasonography.